

***The Complete Blood Cell Count (CBC)***

***CBC – Part 2***

***WBC Differential Count and Morphology***

***A Clinical Pathology 201 Study Module***

***by***

**Carolyn Sue Walters, MHS, MT(ASCP)**

**Department of Pathology**

**School of Medicine**

**Louisiana State University Health Sciences Center**

**New Orleans, Louisiana**

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CSW  
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2001

# C. Sue Walters, MHS, MT(ASCP)



**Associate Professor  
Department of Pathology  
LSU Health Sciences Center  
New Orleans, LA**

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# *The CBC*

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# ***Special Acknowledgment***

**Special thanks is given to Ms. Angela Foley, MS, MT(ASCP), Department of Clinical Laboratory Sciences, LSUHSC School of Allied Health in New Orleans, LA for the use of some of her images of blood cells and for her assistance in the art of creating image files.**

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# *The CBC*

This is the second module of a 4-part study exercise regarding the CBC. The 4 parts are entitled:

- CBC – Part 1      The hemogram
- CBC – Part 2      WBC differential & blood morphology
- CBC – Part 3      RBC morphology & platelet estimate
- CBC – Part 4      Post-test

The review of hematopoiesis and blood cell morphology (i.e., Hematology Atlas located on the LSUHSC server) is recommended as a prerequisite for the CBC exercise.

[click here to continue](#)

# Feedback

**Feedback as to the quality and usefulness of this exercise is solicited and suggestions for improvement are welcomed. Please forward your remarks by E-mail [cwalte@lsuhsc.edu](mailto:cwalte@lsuhsc.edu)**

**or via US MAIL:**

**C. Sue Walters, MHS, MT(ASCP)  
LSUHSC Department of Pathology  
1901 Perdido Street  
New Orleans, LA 70112**

**[click here to continue](#)**

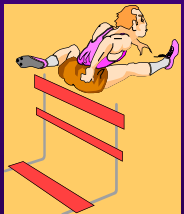
# Directions

The directions for navigating through the exercise are given on the next 3 pages. They are the same as those used in the other modules of this 5-part exercise. Click on:



to visit the directions before continuing with the exercise.

or



to go directly to the Main Menu.

# Directions, continued

The following directional icons are provided throughout the exercise for your convenience. You can click on:

in the upper left hand corner of every page to return to the previous page

in the upper right corner of the page to return to the Main Menu selection.

[click here to continue](#)



# Directions, continued

You can click on:

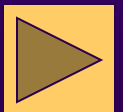


in the lower right corner of the page to continue.



Quit

in the lower right corner of the Main Menu page to Quit (i.e., end the exercise).



## Directions, continued

“Hot points” (symbols, words, phrases) have been inserted on the pages as navigational tools and can be identified by their “gold” color. If it’s “gold”, click on it to move to the next text/data entry. Also, sounds have been added in a few places for emphasis.

Caution, failure to follow the structured order of the “hot points” may result in confusion. If you use the mouse without placing the cursor directly on the “hot point” , you may skip over vital information.

Remember, if it’s **gold**, click on it. Try it!

# Special Comments

This exercise has numerous **images**. You may note that, when a page contains images, there may be a rather long delay before you regain control of the cursor. Please be patient. I think you will find the images are worth the wait.

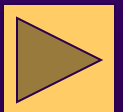
## **NOTE:**

Some animation and/or interactive **affects** may be lost if you attempt to replay a page by returning to the previous page and then advancing to that page again.

Now, click on the **gold** to begin.

# ***CBC - Part 2***

## **The Complete Blood Cell Count (CBC) Part 2 WBC Differential Count & Morphology**



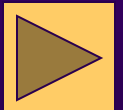
# MAIN MENU

- Introduction & Review of WBC Morphology
- Manual differential WBC count
- Total WBC count vs. differential WBC count
- Relative % vs. absolute number
- Review: mature & immature WBC; non-neoplastic WBC alterations; neoplastic WBC alterations



Quit

# ***Introduction & Review of WBC Morphology***



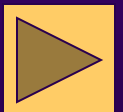
# WBC distribution & morphology

Evaluation of the **distribution and morphology** of white blood cells is one of the most valuable procedures used in examination of the blood. The information obtained may furnish the diagnosis, serve as a guide to therapy and as an indicator of harmful effects of radiotherapy and chemotherapy.



## continued

**A study of white blood cell morphology and distribution of blood cells is an essential part of the clinical description of practically every disease. A normal number and distribution of cells in the blood are so important as physiologic constants that some authorities say the absence of disease cannot be determined until this information is available.**

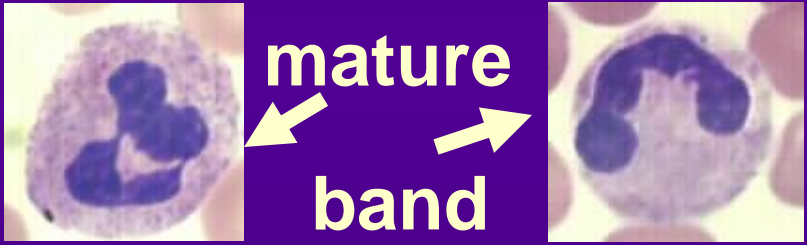




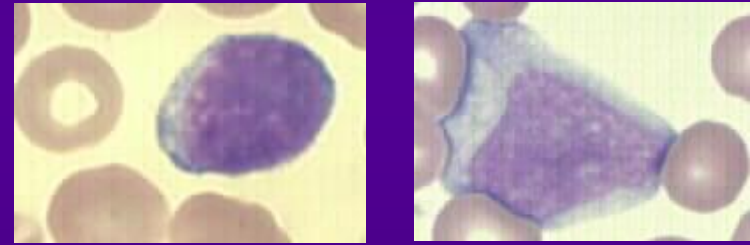
# What WBC are present in normal peripheral blood?

In normal peripheral blood, there are three basic types of leukocytes (illustrated below).

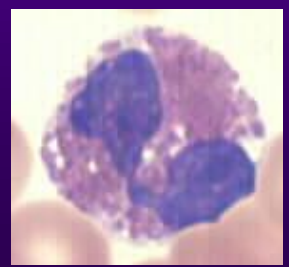
## 1. granulocytes neutrophils



## 2. lymphocytes normal atypical (few)



## eosinophils



## basophils



## 3. monocytes



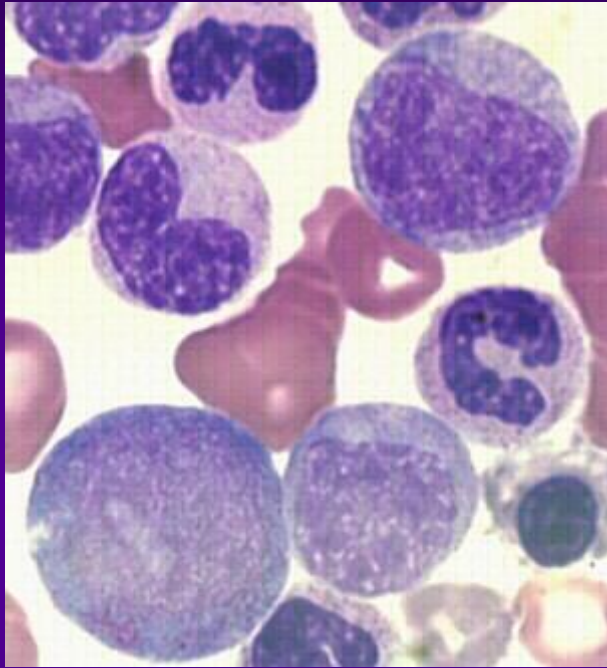
## Are white blood cell abnormalities associated with disease?

An abnormal number and/or distribution of leukocytes (WBC) may be seen in disease. Immature WBC and/or WBC with abnormal alterations may also be seen. The immature and abnormal cells are distinguishable from normal cells by their **morphologic** characteristics.

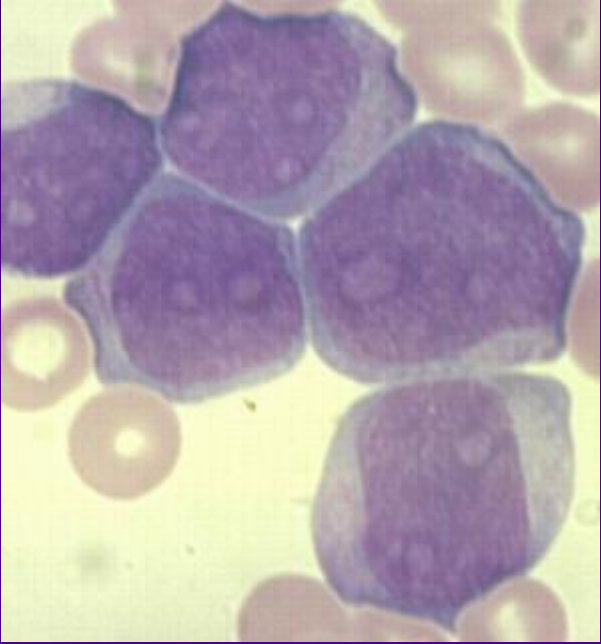
Recognition and identification of these abnormalities play a major role in the diagnosis and treatment of true blood diseases and numerous other pathologic processes.

Review immature and abnormal WBC **or** **continue**  
**w/o review**

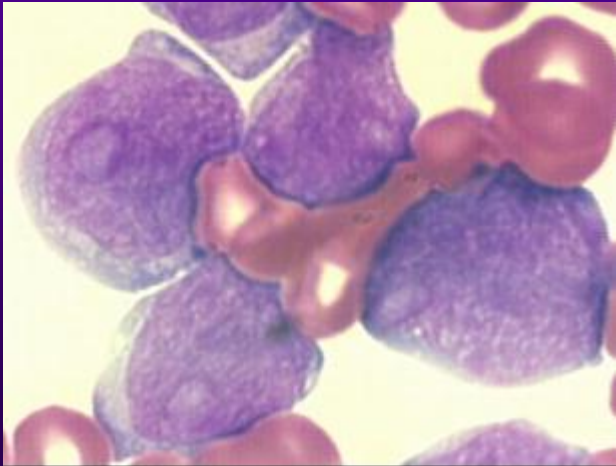
# Examples of immature WBC:



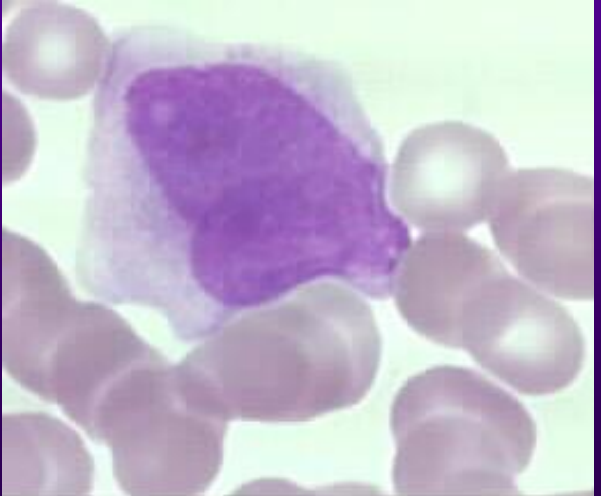
granulocytes  
(various stages)



myeloblasts



lymphoblasts



monoblasts



# Examples of granulocytes in various stages of maturation:

1 late myeloblast or early promyelocyte

2 late promyelocyte or early myelocyte

3 myelocyte

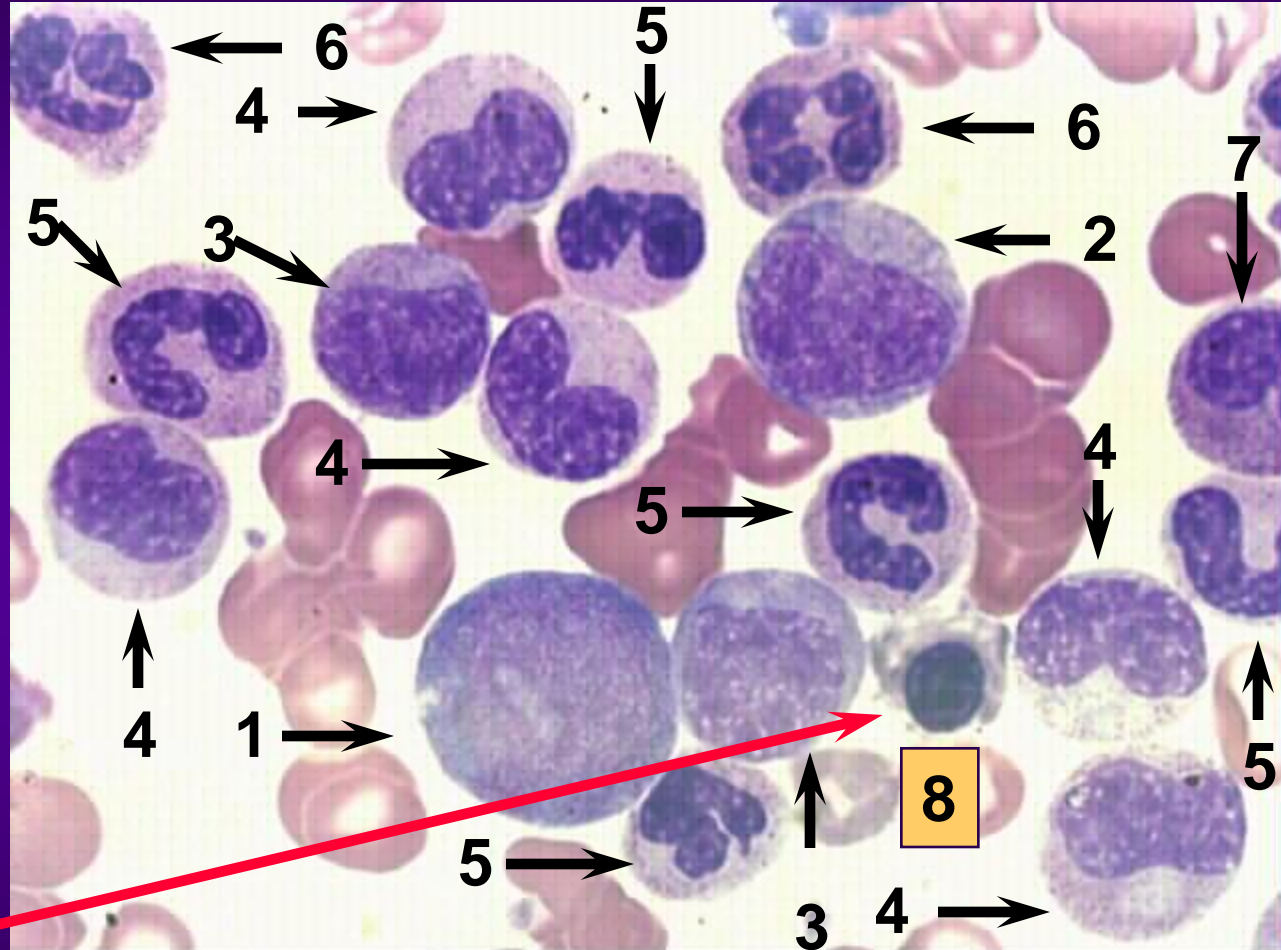
4 metamyelocyte

5 band neutrophil

6 mature segmented neutrophil (PMN)

7 eosinophil

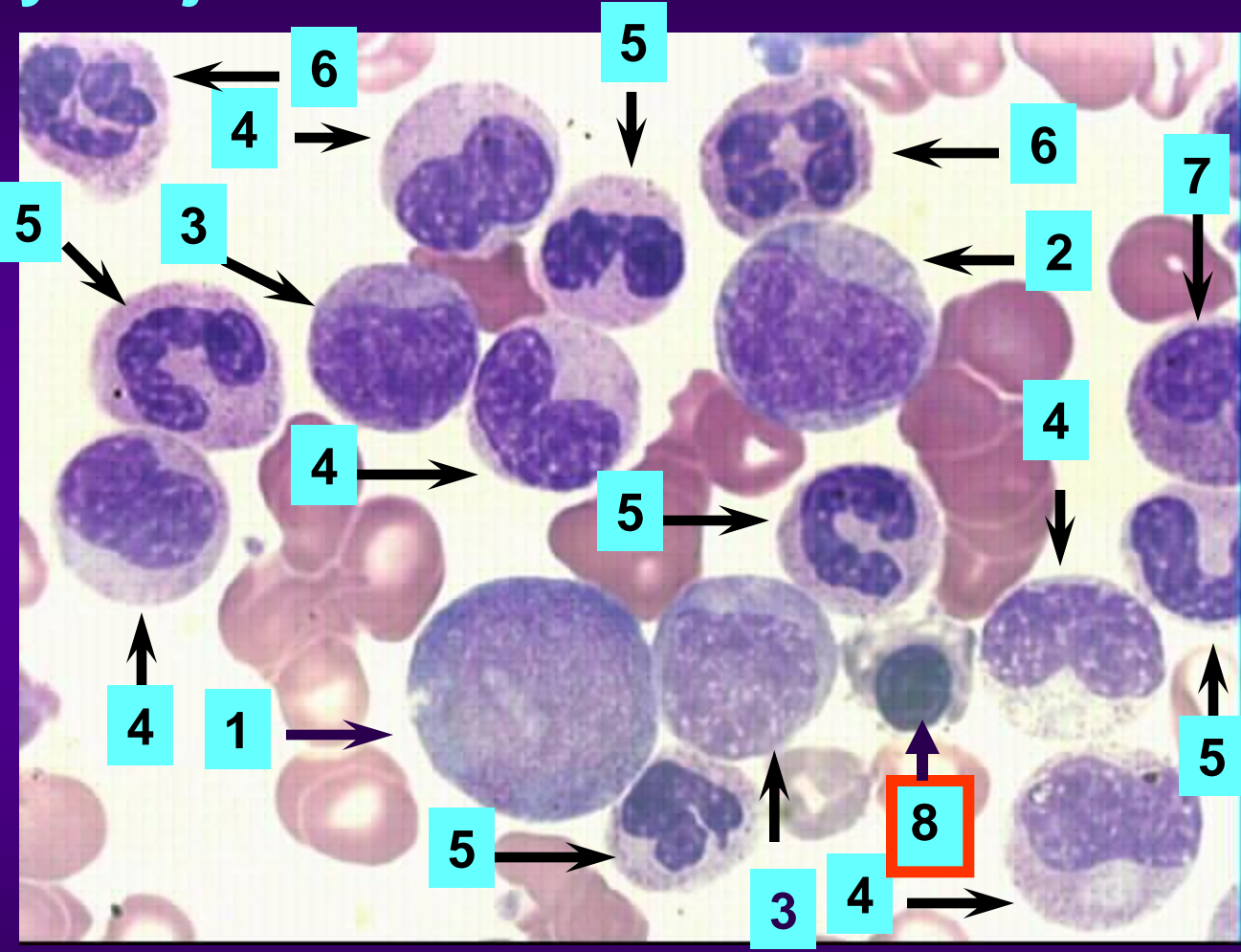
8 **Whoa!** That's not a WBC. It's a nucleated RBC but will also be included in the total WBC count.





# Now, Can you Identify the stages of granulocytes just illustrated?

- 1 late myeloblast or early promyelocyte
- 2 late promyelocyte or early myelocyte
- 3 myelocyte
- 4 metamyelocyte
- 5 band neutrophil
- 6 mature segmented neutrophil (PMN)
- 7 eosinophil
- 8 NRBC

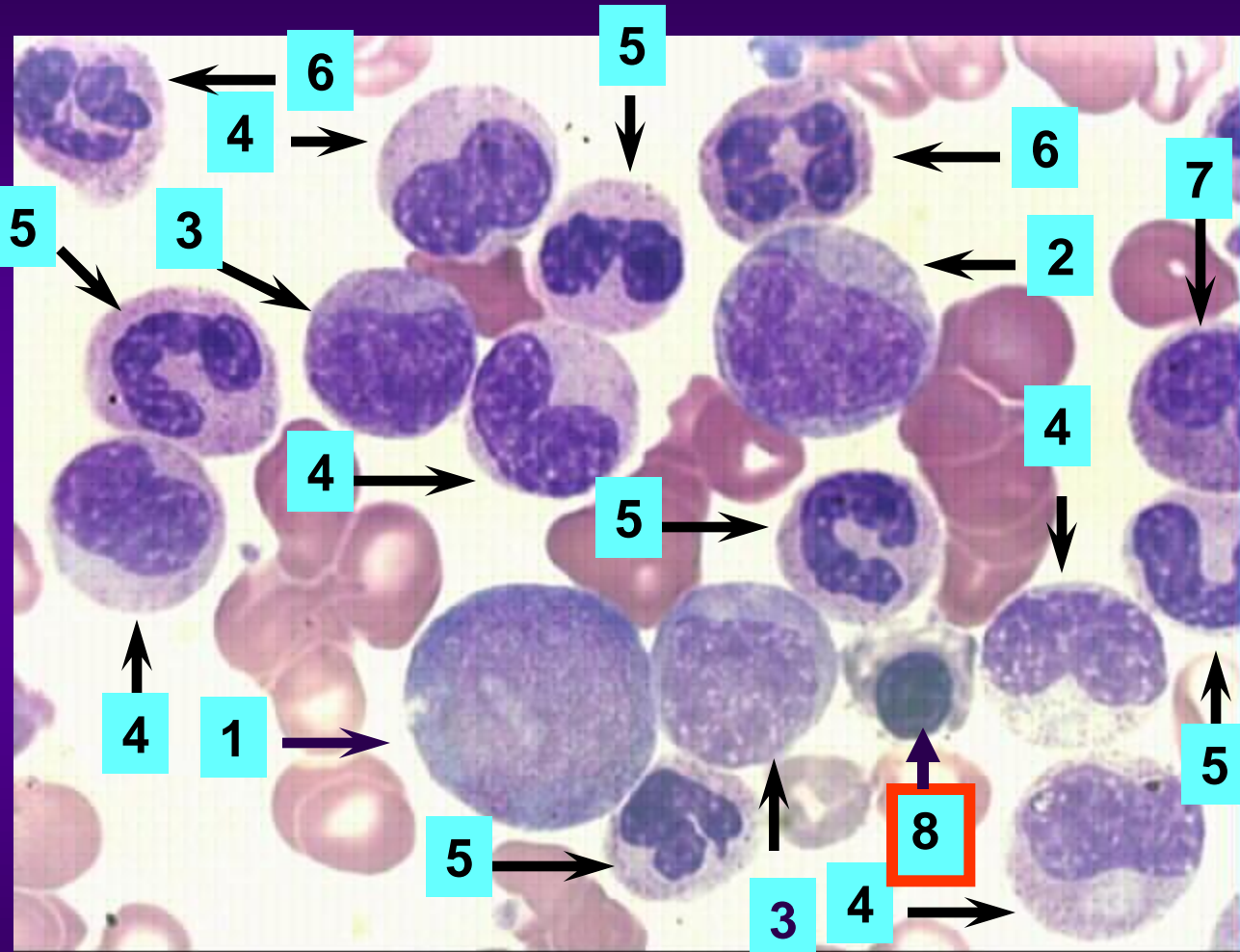


# Overview of the stages of granulocytes just illustrated:

menu

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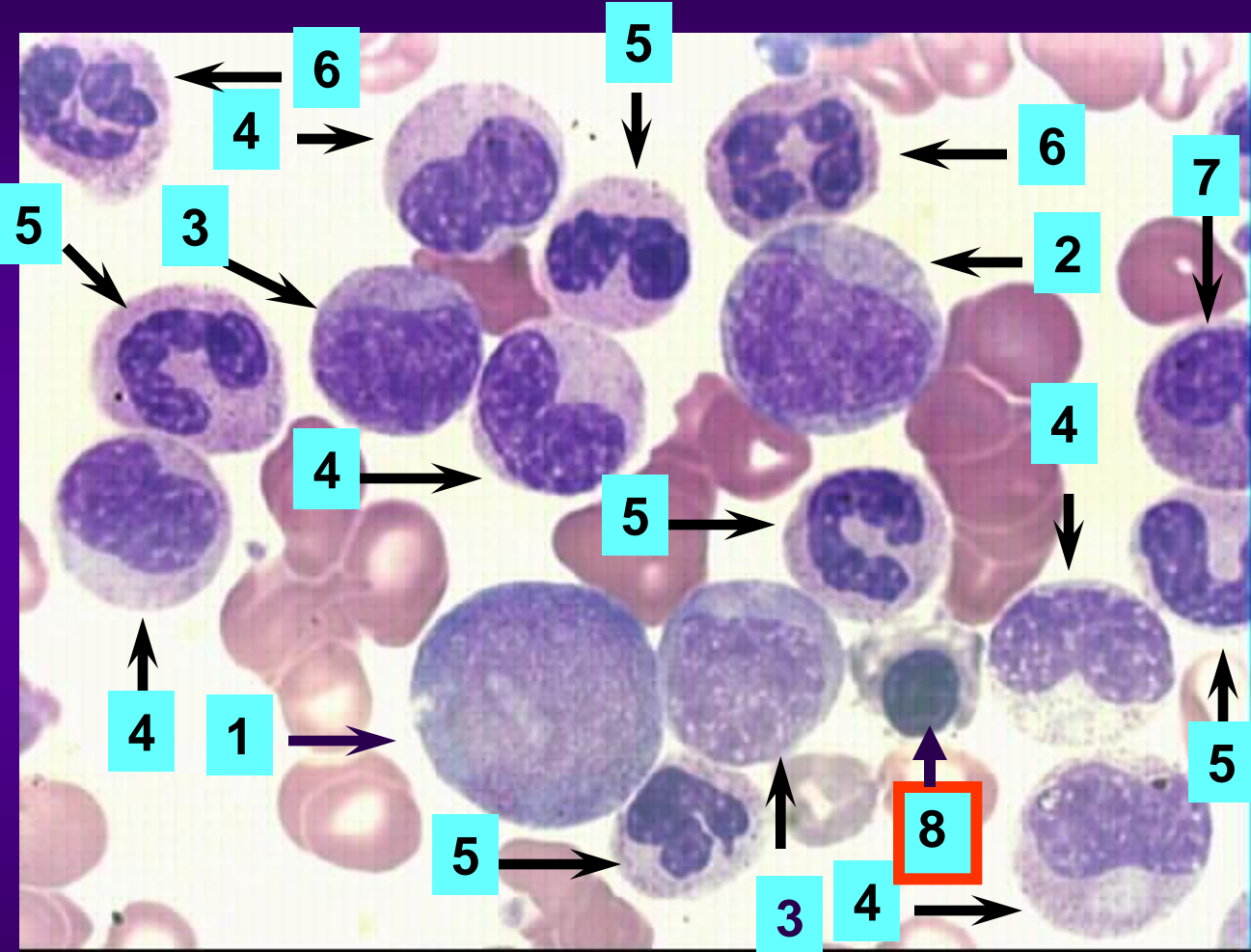
- 1 late myeloblast or early promyelocyte
- 2 late promyelocyte or early myelocyte
- 3 myelocyte
- 4 metamyelocyte
- 5 band neutrophil
- 6 mature segmented neutrophil (PMN)
- 7 eosinophil
- 8 NRBC



review granulocytes again or continue

# Another look at the granulocytes just illustrated:

- 1 late myeloblast or early promyelocyte
- 2 late promyelocyte or early myelocyte
- 3 myelocyte
- 4 metamyelocyte
- 5 band neutrophil
- 6 mature segmented neutrophil (PMN)
- 7 eosinophil
- 8 NRBC

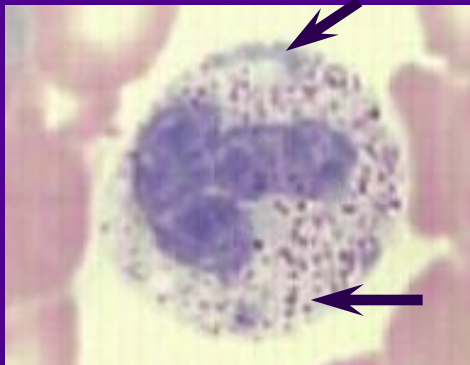




# Examples of WBC with acquired non-neoplastic alterations:

## neutrophils

in bacterial infections



with  
Dohle bodies  
and/or  
toxic granulation

in megaloblastic anemias



with nuclear  
hypersegmentation (ie, > 5  
lobes)

## lymphocytes

in viral infections



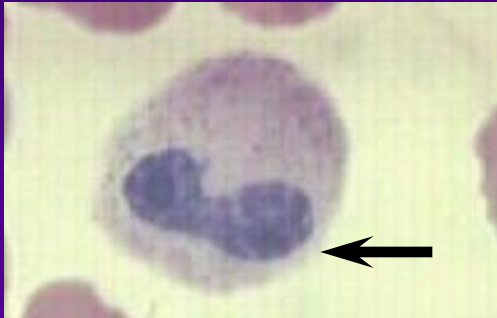
with reactive  
(atypical) changes

inherited disorders



# Examples of WBC with inherited non-neoplastic alterations:

## Pelger-Huet Anomaly



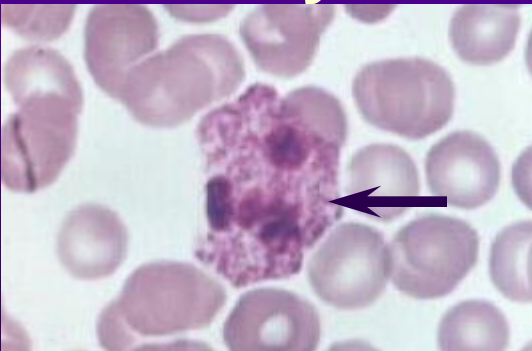
hyposegmented nuclei

## May-Hegglin Anomaly



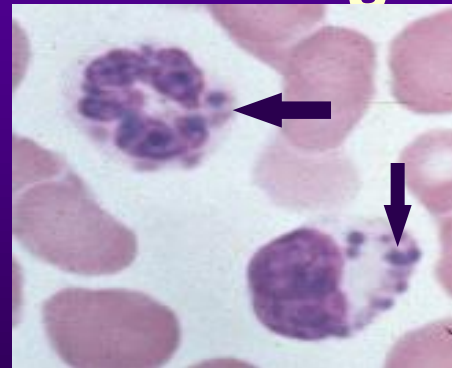
cytoplasmic blue bodies

## Alder-Reilly Anomaly



cytoplasmic black granules

## Chediak-Higashi Syndrome

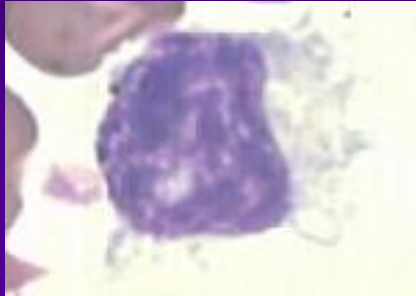


cytoplasmic large  
black granules



# Examples of neoplastic WBC alterations:

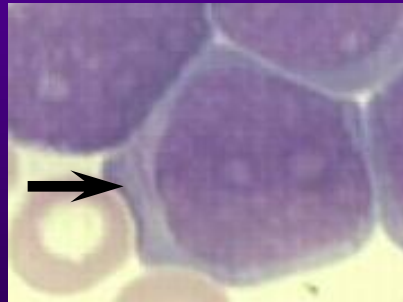
## hairy cell leukemia



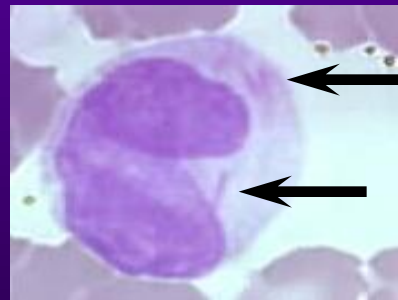
&

## hairy cell lymphocytes

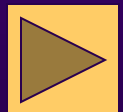
## acute myelocytic leukemias



myeloblasts



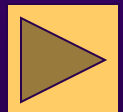
w/ Auer rod(s)



## What can be learned from a differential WBC count?

Because the total WBC count does not differentiate WBCs as to cell lines, a differential WBC count (“diff”) is performed to provide information regarding the frequency distribution of WBCs and to identify increases or decreases when they occur in one or more of the cell lines.

A morphologic study of the various blood cells (i.e., WBC, RBC, & platelets) is made during the differentiation process to detect and identify atypical and/or abnormal cells.



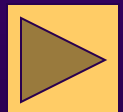
## What methods can be used for the differential WBC count ?

The differential WBC count and blood cell study may be performed by one of several methods, e.g.:

- A manual microscopic examination of a Wright's (or Wright's Giemsa) stained peripheral blood smear.

**OR**

- An automated multi-channel instrument (in this exercise, it is a Coulter STK-S 5-part differential), which is discussed in the automated CBC section.



# End of Introduction

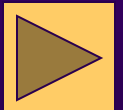
This concludes the Introduction to the Differential WBC Count. Select one of the following:

Go to Manual Differential WBC Count, the next section, to continue with the exercise as designed.

OR

Return to the Main Menu and make an alternate selection.

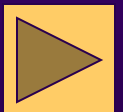
# ***Manual Differential WBC Count***



## What kind of blood smear is used ?

**A peripheral blood film (made from a drop of blood from an EDTA anticoagulated tube or skin puncture) is stained with Wright (or Wright-Giemsa) stain and microscopically examined using scanning (10x), high-dry (40x), and oil (100x) objectives.**

**Refer to the Clinical Pathology 202 Course Manual for the complete procedure.**



## What studies will be performed on the stained blood smear?

**A complete differential WBC count (“diff”) which includes identification of mature and (if any) immature WBC and description of WBC morphologic alterations.**

- complete description of RBC morphology which includes (if any) a description of RBC morphologic alterations and, if present, the number of NRBC per 100 WBC.**
- platelet estimate of adequacy which includes (if any) abnormal morphologic alterations, distribution, and clumps.**

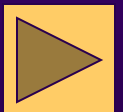




## continued:

**This section is limited to the differential count and morphology of white blood cells.**

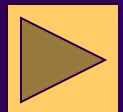
**RBC morphology and platelet estimate discussions are addressed later in other sections.**



## How are the WBC identified and classified?

Typical nuclear and cytoplasmic morphologic features provide a means by which WBC can be identified and classified as to:

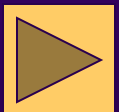
- **cell line** (i.e., granulocytes [neutrophils, eosinophils, or basophils], lymphocytes, or monocytes)
- **naturity** (i.e., mature cell or specific immature stage of development).
- **abnormal morphology** (i.e., nuclear or cytoplasmic alterations)



## How are the WBC differentiated and enumerated?

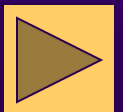
**At least 100 WBC** are counted and a tabulation is made as to the number of each leukocytic cell type included in the count.

The 100 cell count provides the *RELATIVE* number (or percent) of each white blood cell type present in the peripheral blood.



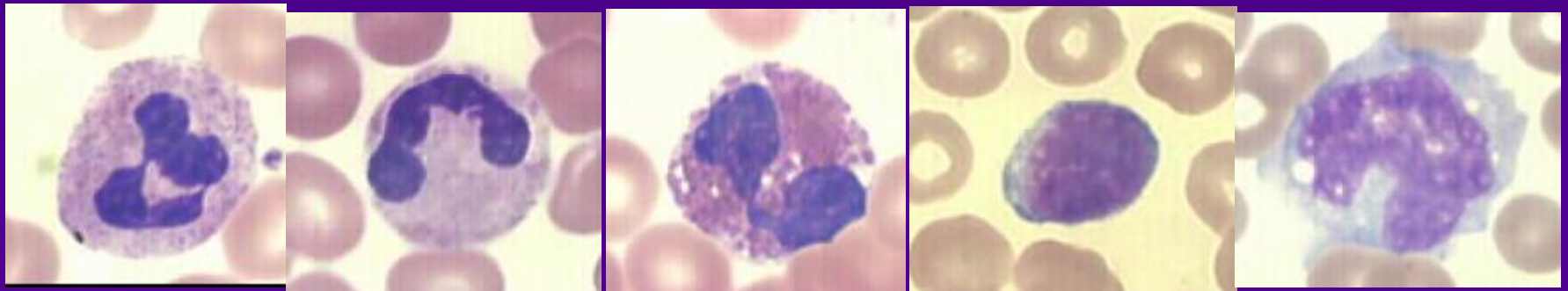
**How can you determine whether there is an increase or decrease of one or more of the cell lines?**

**Increases or decreases in a white blood cell line (or type) can then be determined by comparing the number obtained on the differential count with established reference ranges.**



# Reference Range (Relative %)

Reference ranges (values considered to be normal) for differential WBC counts may vary among laboratories, but are usually about:



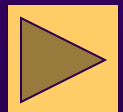
**segmented  
neutrophils  
50-70%**

**band  
neutrophils  
0- 5%**

**eosinophils  
1-5%**

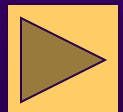
**lymphocytes  
20-40%**

**monocytes  
1-6%**



# What terminology is used to indicate an increased or decreased number of a specific white blood cell line?

<u>Cell Line</u>	<u>Increased:</u>	<u>Decreased:</u>
<b>Neutrophils</b>	Neutrophilia	Neutropenia
<b>Lymphocytes</b>	Lymphocytosis	Lymphocytopenia
<b>Monocytes</b>	Monocytosis	Monocytopenia
<b>Eosinophils</b>	Eosinophilia	Eosinopenia
<b>Basophils</b>	Basophilia	Basopenia



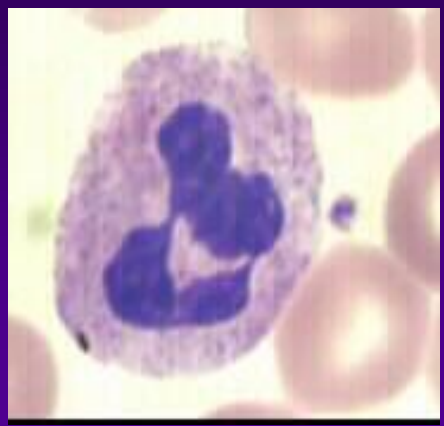
# Review Morphology?

**Review WBC morphology**

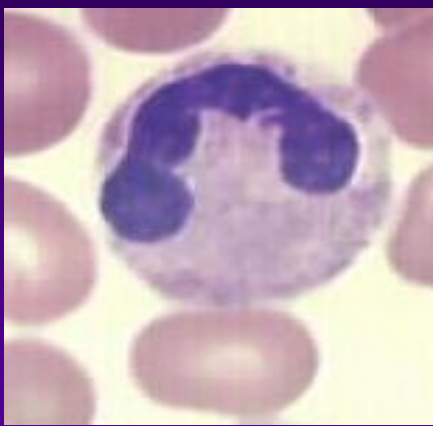
**or**

**continue without review**

# WBC in Normal Blood (Adults):



**PMN**



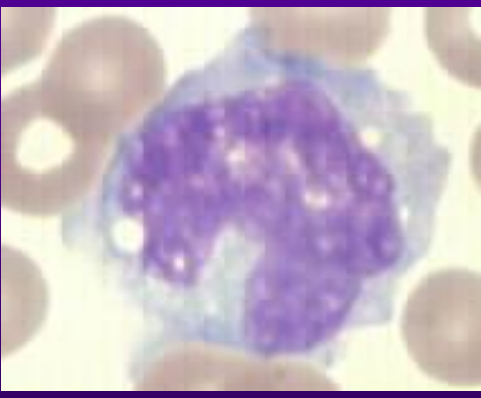
**band  
(neutrophils)**



**eosinophil**



**basophil**



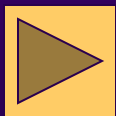
**monocyte**



**lymphocyte**

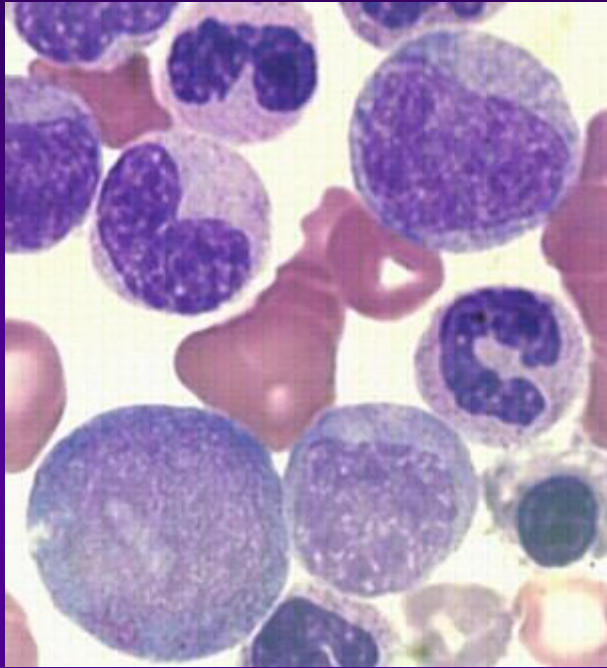


**ATL (few)  
(atypical lymphocyte)**

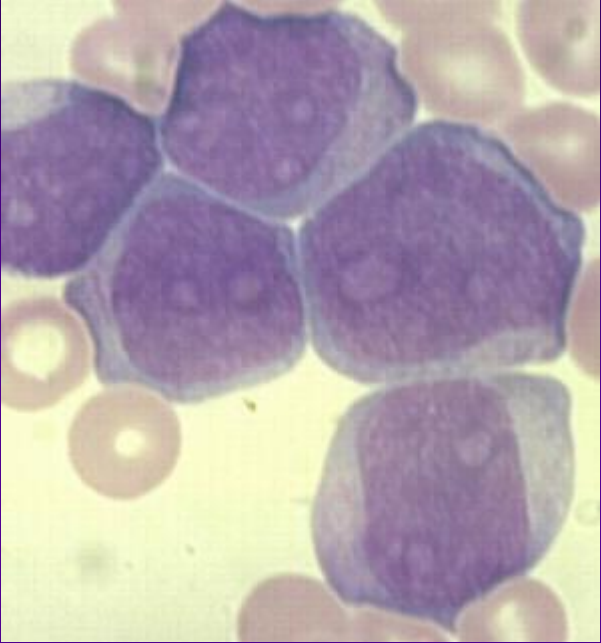




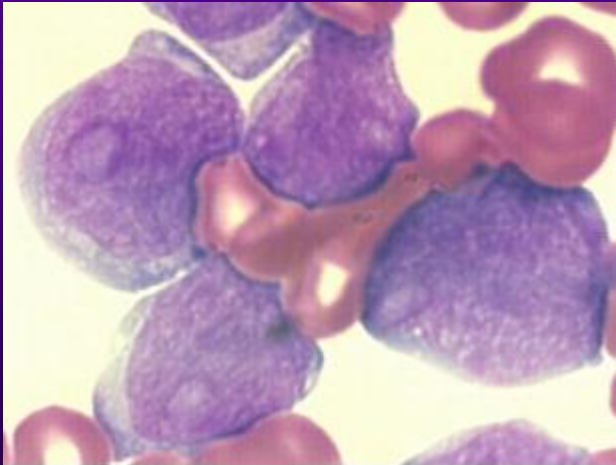
# Examples of immature WBC:



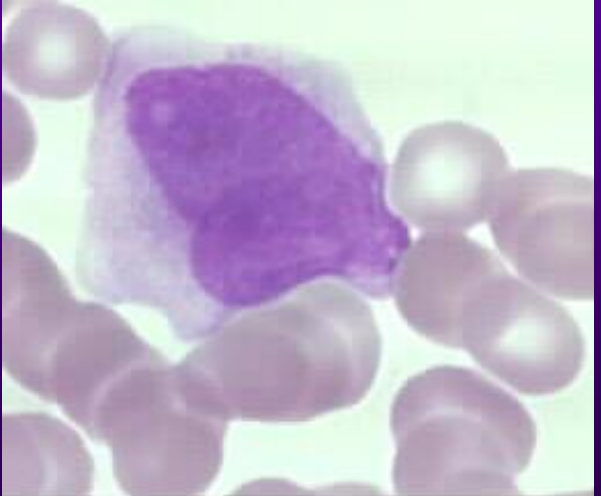
**granulocytes  
(various stages)**



**myeloblasts**



**lymphoblasts**

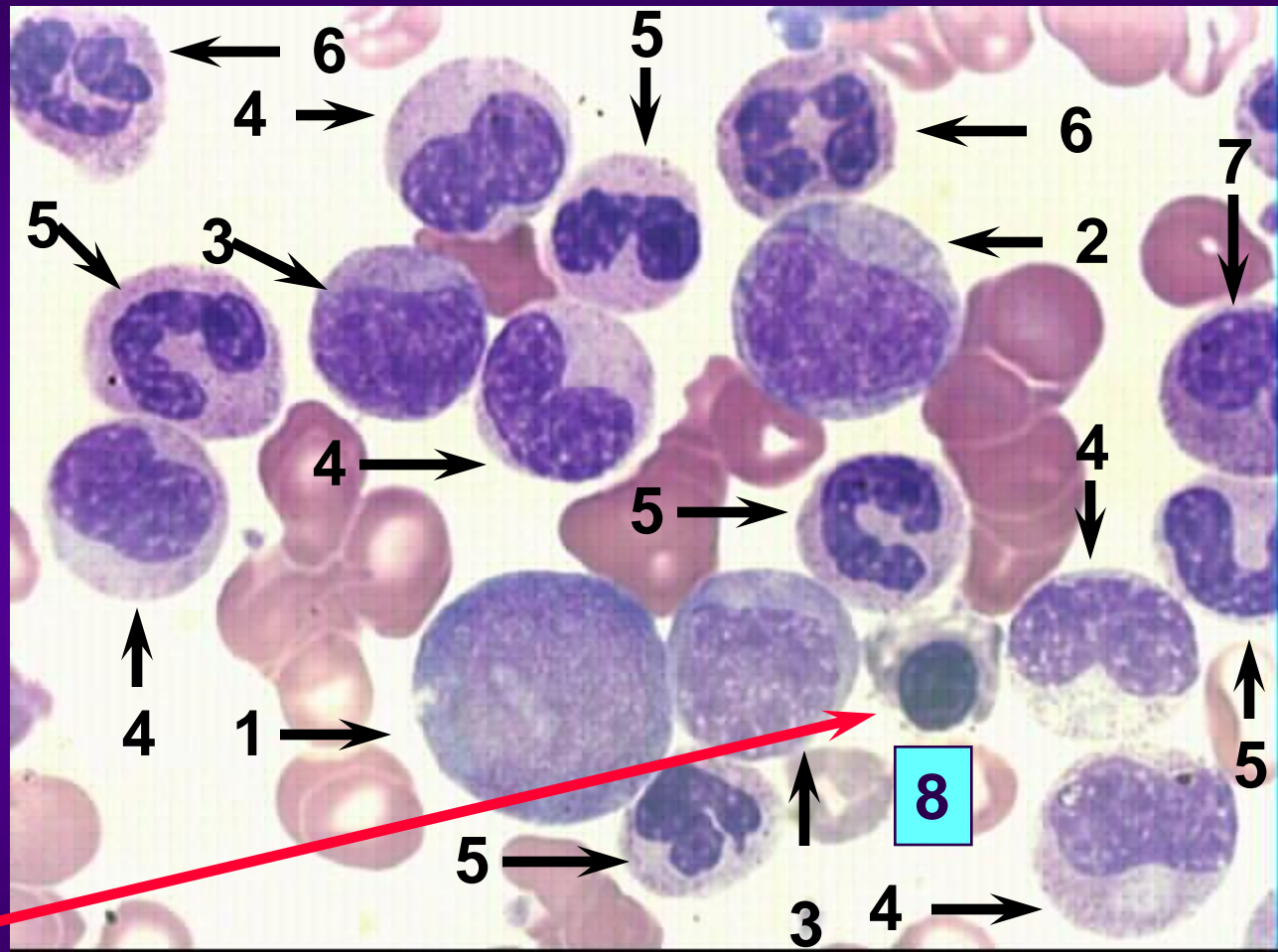


**monoblasts**



# Examples of granulocytes in various stages of maturation:

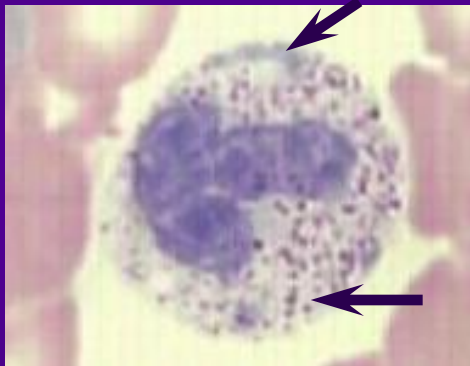
- 1 late myeloblast or early promyelocyte
- 2 late promyelocyte or early myelocyte
- 3 myelocyte
- 4 metamyelocyte
- 5 band neutrophil
- 6 mature segmented neutrophil (PMN)
- 7 eosinophil
- 8 It's a nucleated RBC but will also be included in the WBC count.



# Examples of WBC with acquired non-neoplastic alterations:

## neutrophils

in bacterial infections



with  
Dohle bodies  
and/or  
toxic granulation

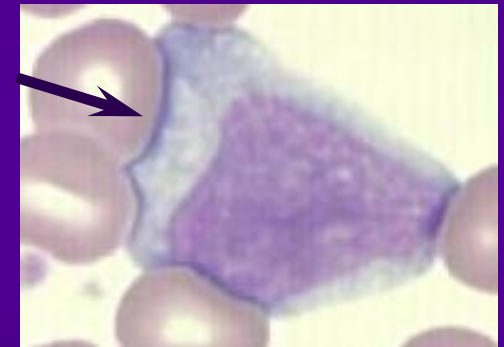
in megaloblastic anemias



with nuclear  
hypersegmentation (ie, > 5  
lobes)

## lymphocytes

in viral infections

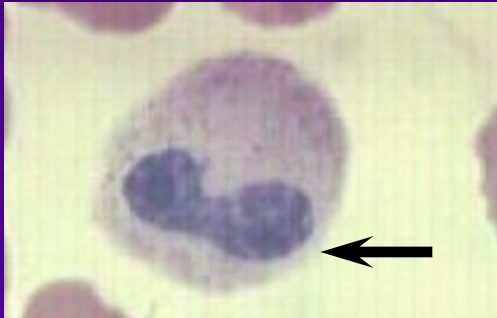


with reactive  
(atypical) changes



# Examples of WBC with inherited non-neoplastic alterations:

## Pelger-Huet Anomaly



hyposegmented nuclei

## May-Hegglin Anomaly



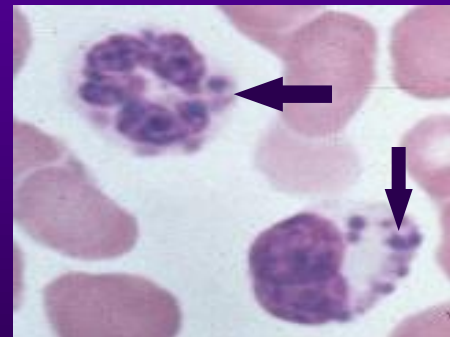
cytoplasmic blue bodies

## Alder-Reilly Anomaly



cytoplasmic black granules

## Chediak-Higashi Syndrome



cytoplasmic large black granules





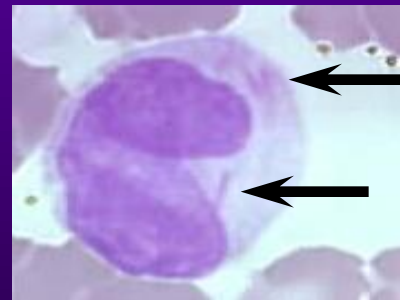
# Examples of neoplastic WBC alterations:

## hairy cell leukemia



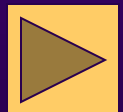
## hairy cell lymphocytes

## acute myelocytic leukemias



myeloblasts

w/ Auer rod(s)



# Examples of nucleated RBC in various stages of maturation.

**proerythroblast**

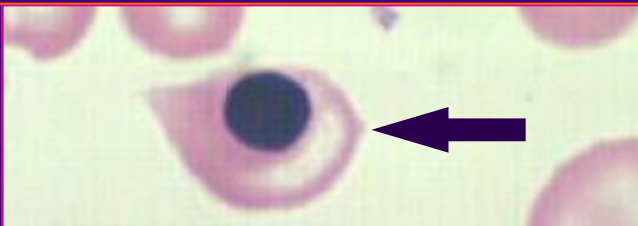
(earliest form w/ nucleoli)



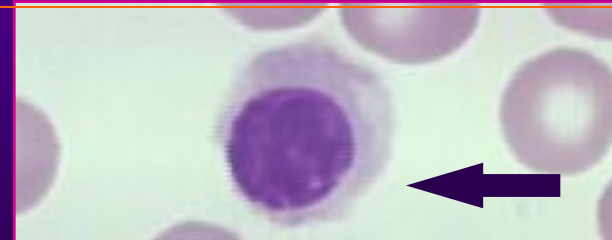
**basophilic erythroblast**  
(ill-defined or absent nucleoli)



**orthochromatophilic erythroblast**  
(last stage before extrusion of nucleus)



**polychromatophilic erythroblast**  
(cytoplasmic evidence of HGB)



# End of Manual Differential WBC Count & Morphology

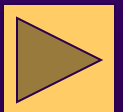
This concludes the Manual WBC Differential Count and Morphology section. Select one of the following:

Go to Total WBC Count vs. Differential WBC Count, the next section, to continue with the exercise as designed.

OR

Return to the Main Menu and make an alternate selection.

***Total WBC Count***  
***vs.***  
***Differential WBC Count***

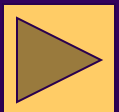




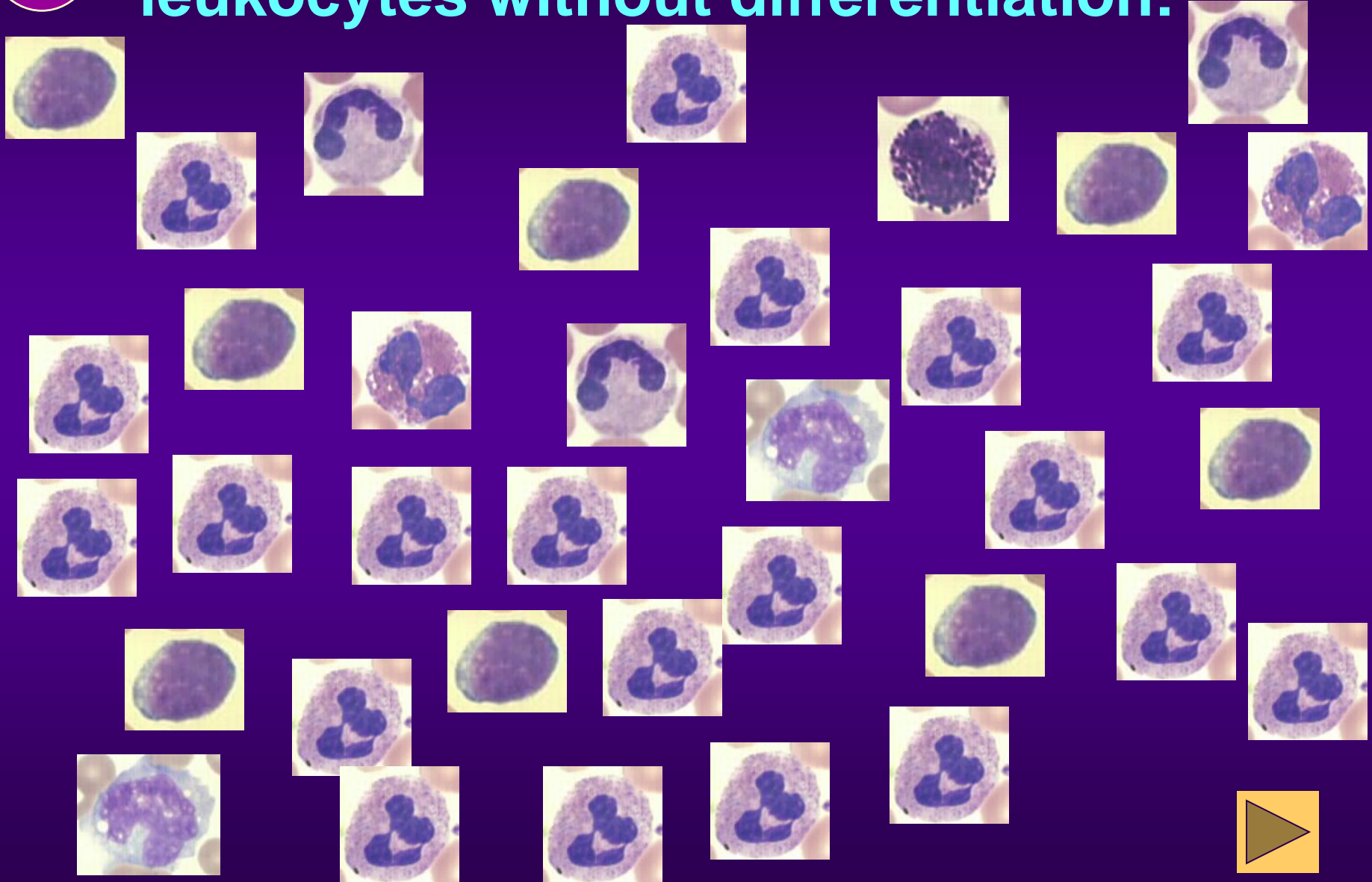
## How does the differential WBC count differ from the total WBC count?

The total WBC count reflects the total number of all leukocytes in circulation but does not differentiate leukocytes as to their various cell lines (e.g., neutrophils, lymphocytes), stage of maturity, (e.g., band, metamyelocyte), or abnormalities when present (e.g., toxic granulation, hypersegmented nuclei).

A differential WBC count must be performed to provide that information.



# Total WBC count = total number of leukocytes without differentiation:



# “Diff” - differentiates and enumerates WBC according to cell type, e.g.:



neutrophils, mature

lymphocytes

monocytes

eosinophils

basophils

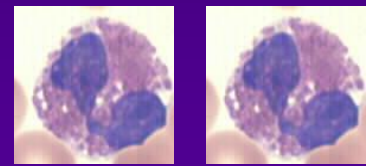


# “Diff” - provides frequency distribution (relative % and absolute number/ $\mu$ L, e.g. total WBC = 10,000/ $\mu$ L):

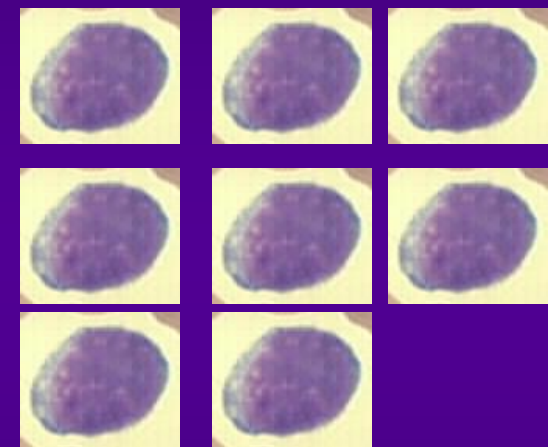
**neutrophils, mature**  
**65%**                      **6500/ $\mu$ L**



**eosinophils**  
**3%**    **300/ $\mu$ L**



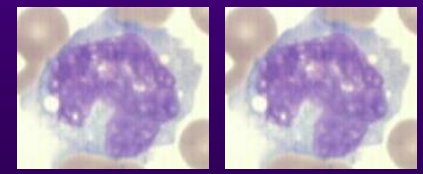
**lymphocytes**  
**28%**    **2800/ $\mu$ L**



**basophils**  
**1%**    **100/ $\mu$ L**

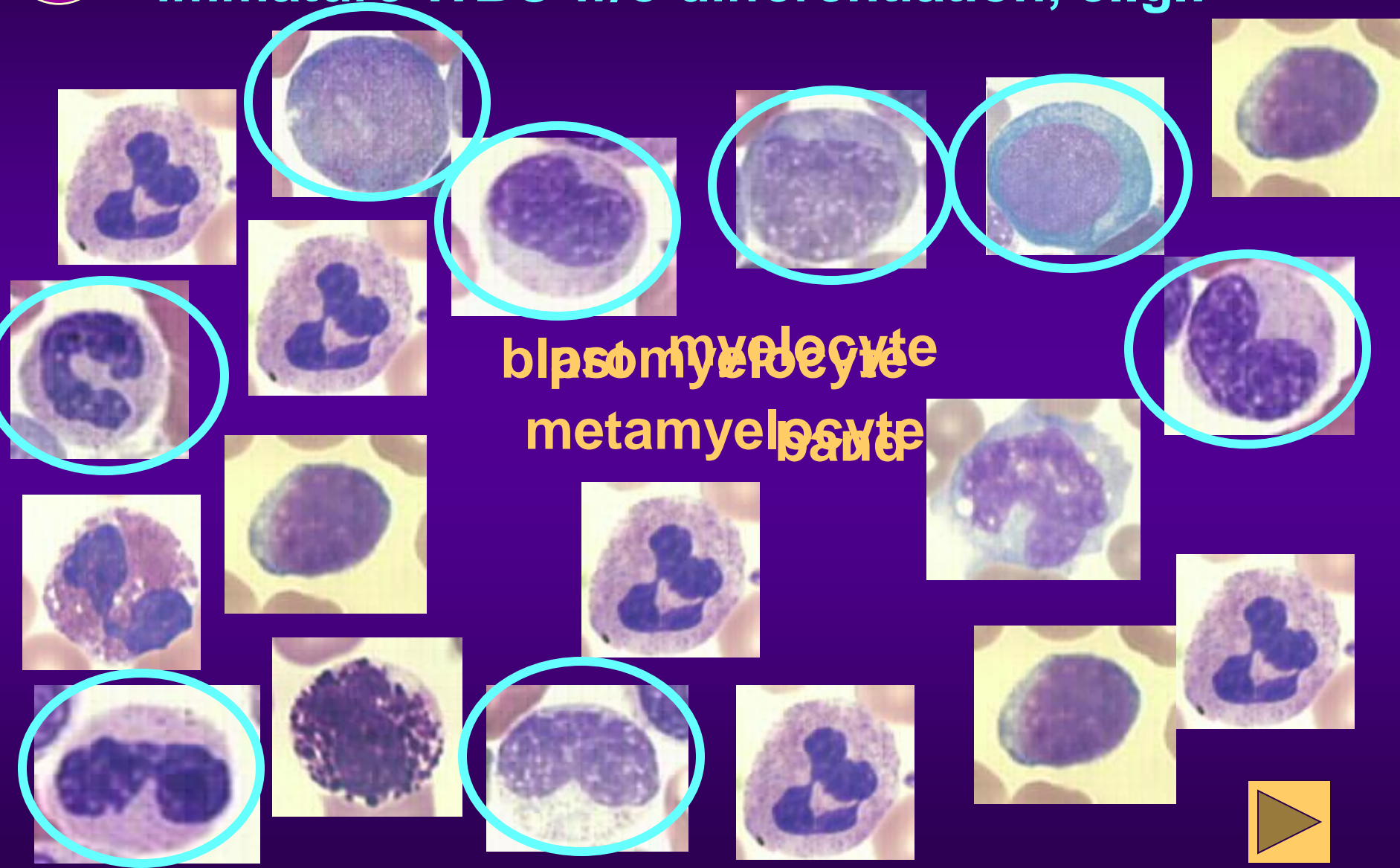


**monocytes**  
**3%**    **300/ $\mu$ L**





# Total WBC count - mature and (if present) immature WBC w/o differentiation, e.g.:

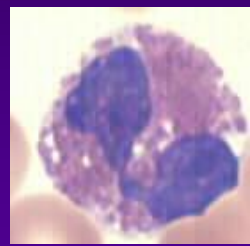


blast  
monocyte  
metamyelocyte  
band



# “Diff” - differentiates and enumerates mature and (if present) immature WBC, e.g.:

## mature



neutrophil

eosinophil

basophil

lymphocyte

monocyte

## immature

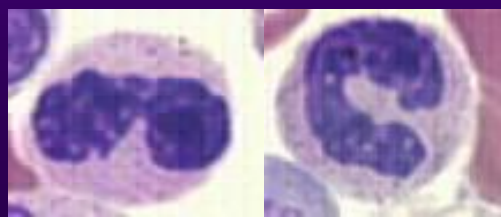


blast

promyelocyte

myelocyte

metamyelocyte



neutrophilic band



“Diff” - provides frequency distribution (relative % and absolute number e.g., total WBC=50,000/ $\mu$ L):

neutrophil, mature  
50% 25000/ $\mu$ L



neutrophil, bands  
15% 7500/ $\mu$ L



metamyelocyte  
12% 6000/ $\mu$ L



myelocyte  
7% 3500/ $\mu$ L



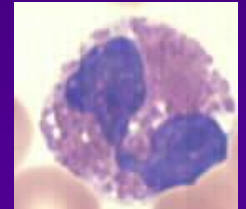
promyelocyte,  
2% 1000/ $\mu$ L



blast  
1% 500/ $\mu$ L



eosinophil  
5% 2500/ $\mu$ L



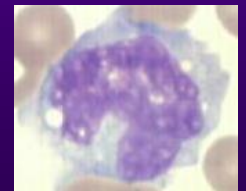
basophil  
3% 1500/ $\mu$ L



lymphocyte  
5% 2500/ $\mu$ L

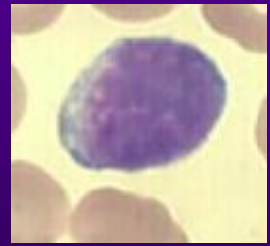
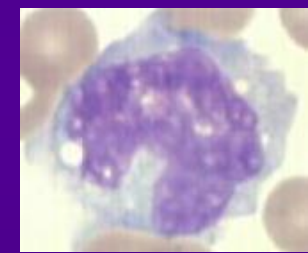
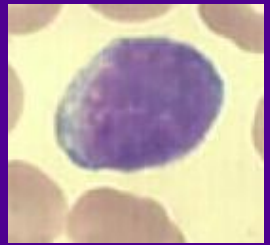
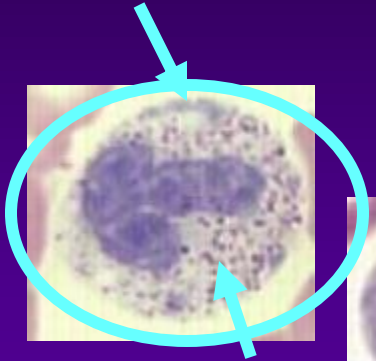


monocyte  
0% 0/ $\mu$ L



# Total WBC count - normal and abnormal WBC without differentiation, e.g.:

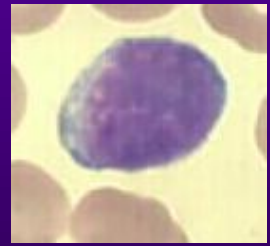
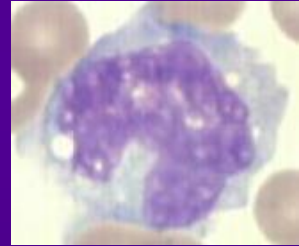
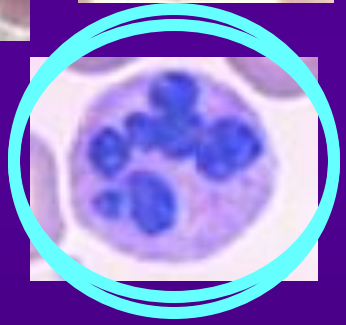
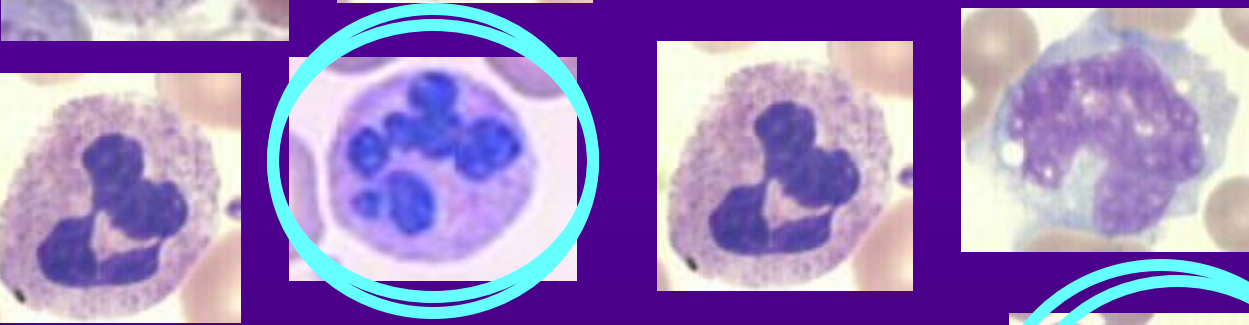
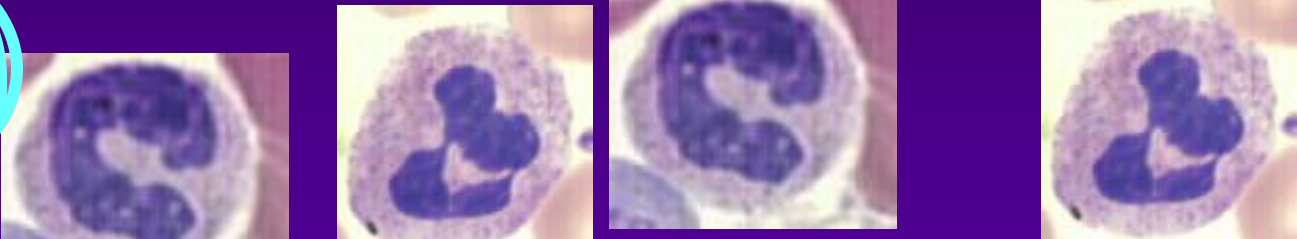
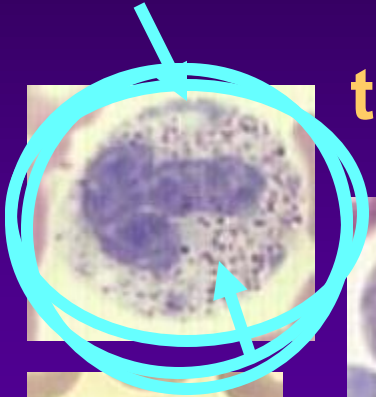
## acquired WBC alterations





# “Diff” - differentiates abnormal WBC with acquired alterations, e.g.:

toxic granulation hypersegmented neutrophils  
Döhle bodies

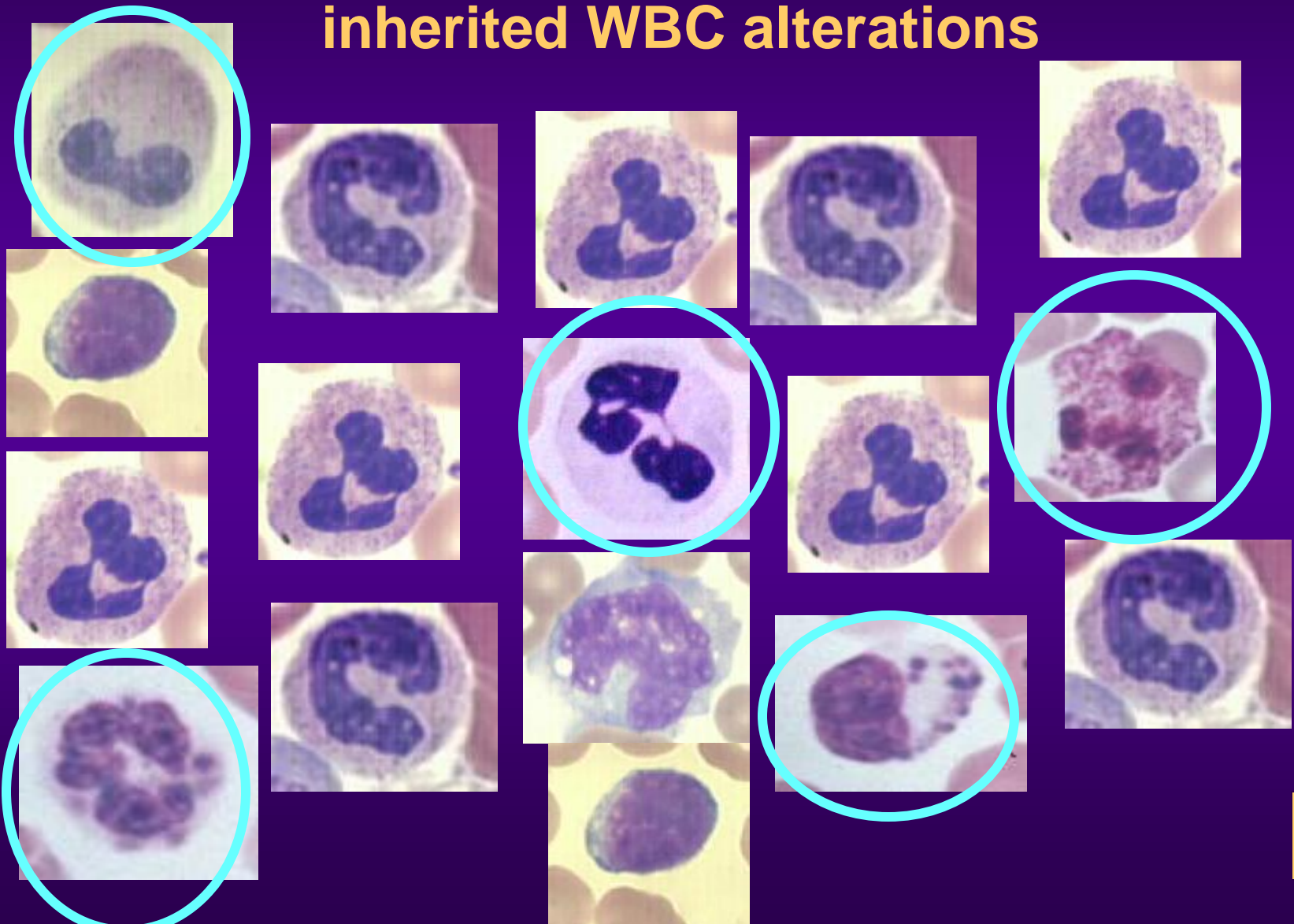


reactive/atypical lymphocytes



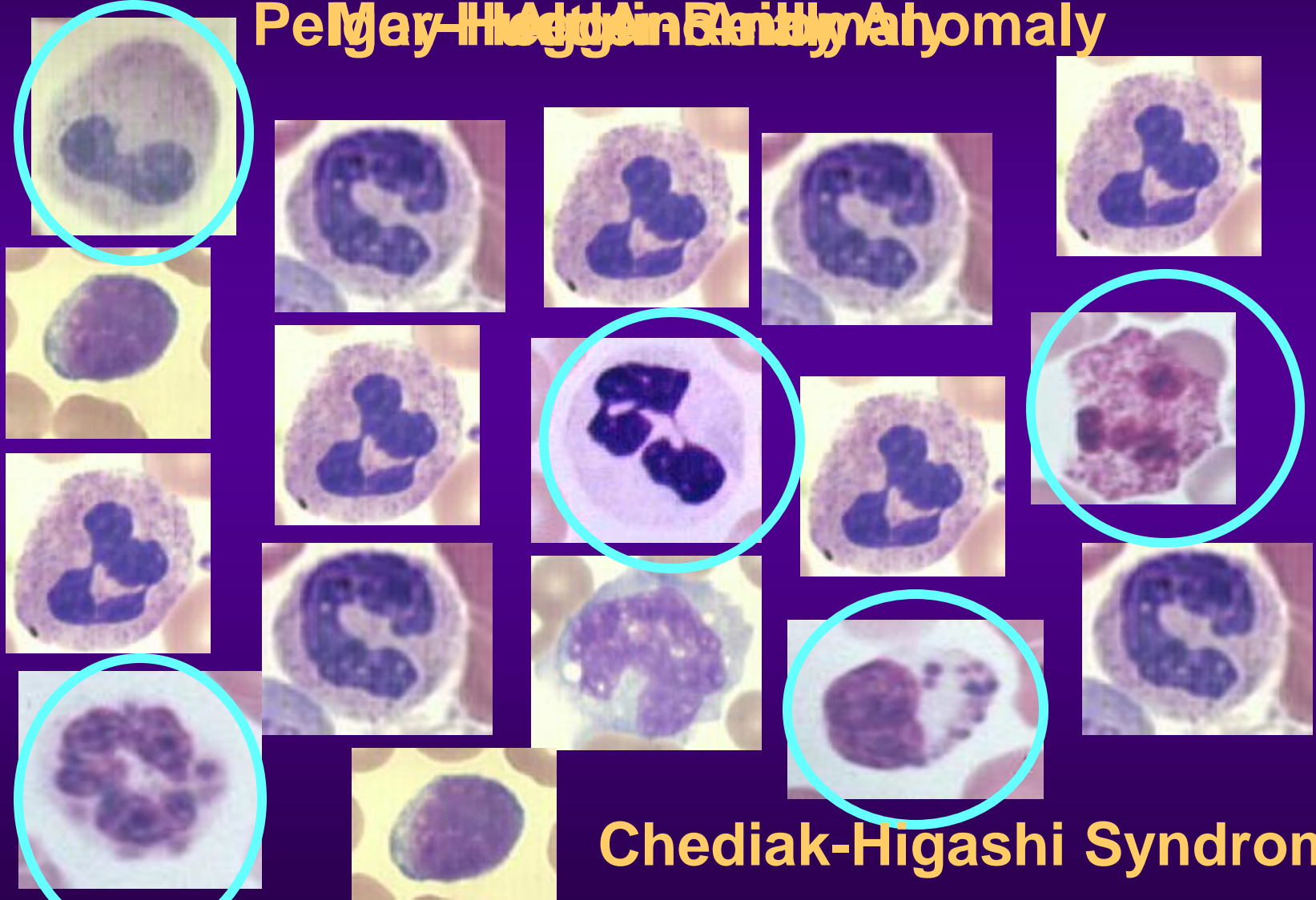
# Total WBC count - normal and abnormal WBC without differentiation, e.g.:

## inherited WBC alterations



# “Diff” - differentiates abnormal WBC with inherited alterations, e.g.:

## Pelger-Huet anomaly

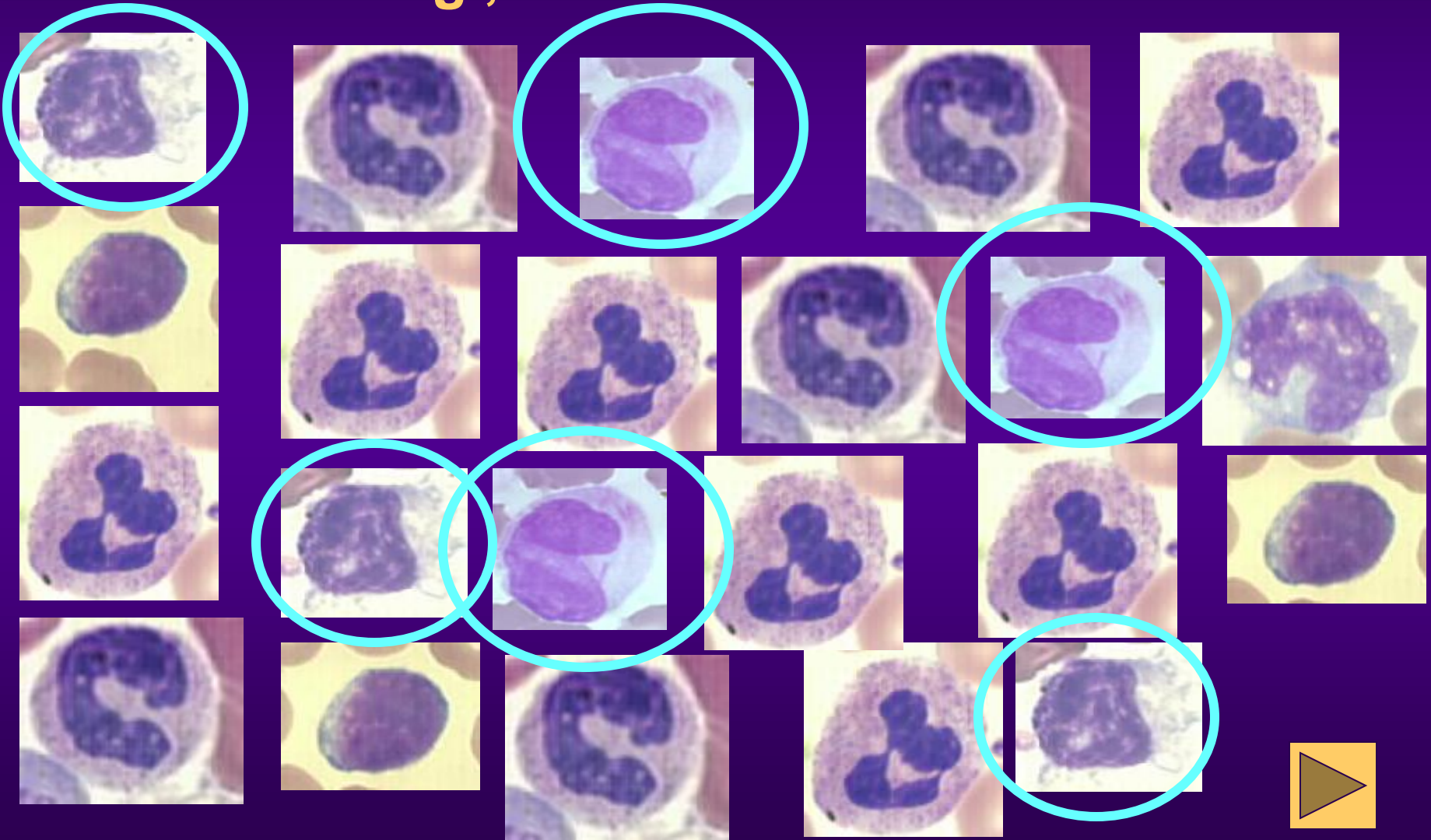


## Chediak-Higashi Syndrome



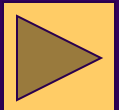
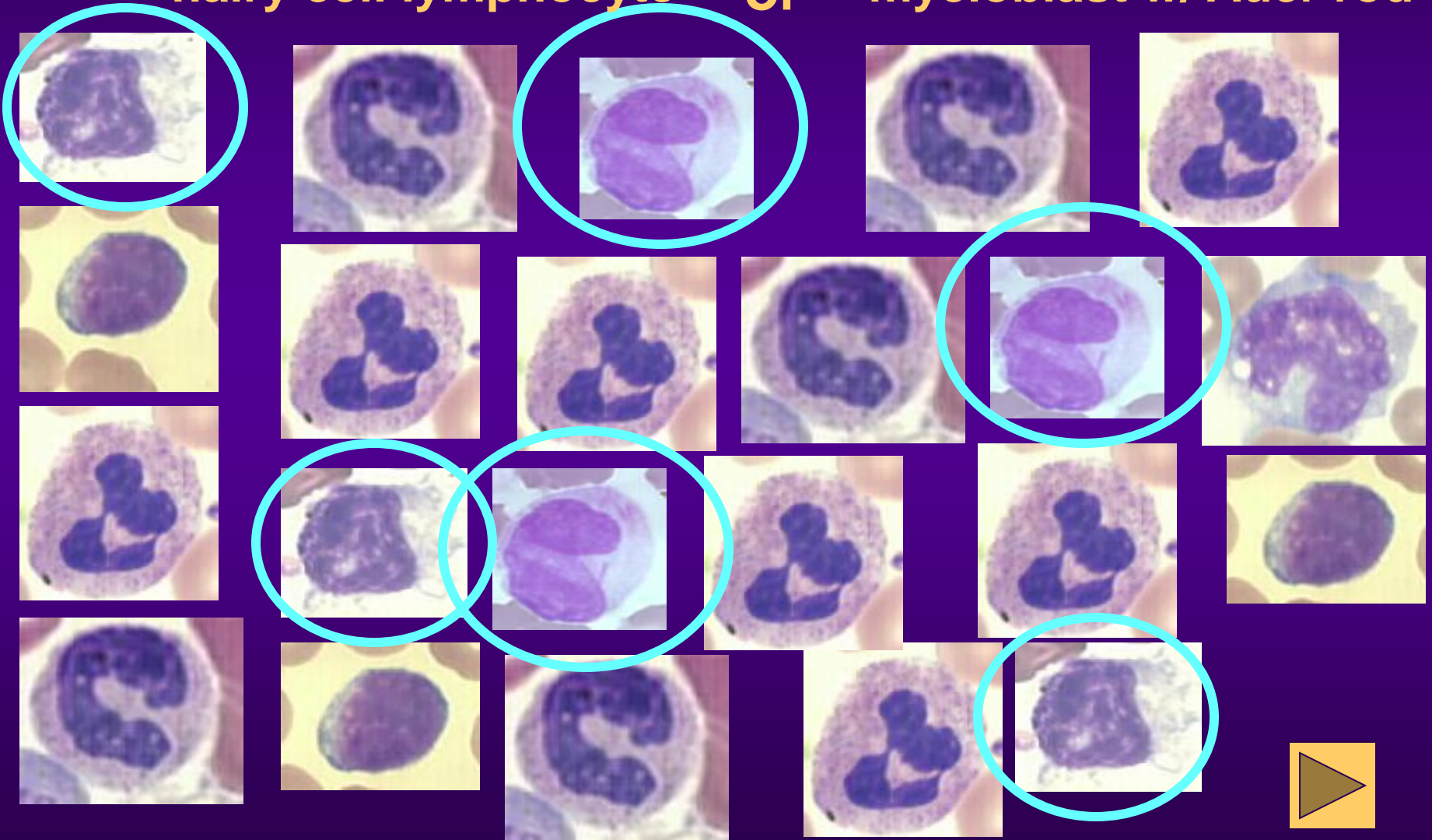


# Total WBC count - normal and (if present) neoplastic WBC w/o differentiation: e.g., leukemic alterations



# “Diff” - differentiates WBC with neoplastic alterations, e.g.:

hairy cell lymphocyte or myeloblast w/ Auer rod



# End of Total vs. Differential WBC Counts

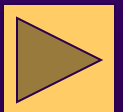
This concludes the Introduction to the Differential WBC Count. Select one of the following:

Go to Relative % vs. Absolute Number, the next section, to continue with the exercise as designed.

OR

Return to the Main Menu and make an alternate selection.

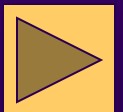
# ***Relative % vs. Absolute Number***



## What is meant by the relative % and absolute number of WBC ?

**Relative % is based on the differential count of 100 white blood cells and reflects the per cent of each cell type present in circulation.**

**If the total number of white cells in circulating blood is known and the relative per cent of each white cell type is known, then the absolute number of each cell type per  $\mu\text{L}$  of blood can be calculated.**

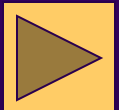




# continued:

**For example: given a patient with a total WBC count of 8,000/ $\mu$ L and the differential WBC count shown below (i.e., the number observed for each cell type in the 100 white cell count):**

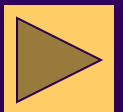
<b>Segmented neutrophils</b>	<b>60%</b>
<b>Band neutrophils</b>	<b>5%</b>
<b>Lymphocytes</b>	<b>30%</b>
<b>Monocytes</b>	<b>2%</b>
<b>Eosinophils</b>	<b>2%</b>
<b>Basophils</b>	<b>1%</b>




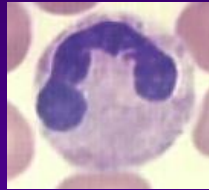
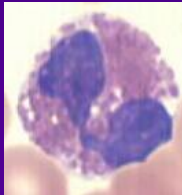


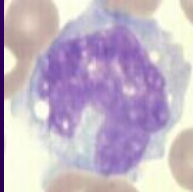
# continued:

Then the absolute number of each cell type/ $\mu\text{L}$  can be calculated by multiplying the per cent of each cell type by the total number of WBC/ $\mu\text{L}$ .

Segmented neutrophils	60%	x 8,000 = 4,800
Band neutrophils	5%	x 8,000 = 400
Lymphocytes	30%	x 8,000 = 2,400
Monocytes	2%	x 8,000 = 160
Eosinophils	2%	x 8,000 = 160
Basophils	<u>1%</u>	x 8,000 = 80
Total = 100%		= 8,000



# What are the relative reference ranges for leukocytes?

<u>Cell Type</u>	<u>Reference Ranges (Relative %)</u>		
<b>Neutrophils</b>	<b>40-72 (PMN)</b>		
			<b>0-5 (bands)</b>
<b>Eosinophils</b>	<b>0-6</b>		
<b>Basophils</b>	<b>0-0.2</b>		
<b>Lymphocytes</b>	<b>24- 45</b>		
<b>Monocytes</b>	<b>0.4 - 10</b>		



Do the relative values always indicate which cell line is ↓ or ↑ ?

**If the total WBC count is “normal” (i.e., within the established reference range), the relative values are a good reflection of the number of each cell type present, including increases or decreases.**

**However, if the total WBC count is abnormal (i.e., increased or decreased), the relative percentage must be converted to an absolute number of each cell type present in order to determine which cell line is involved.**

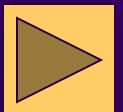


## How is the relative % converted to an absolute number ?

For each white blood cell line, multiply the percentage of cells counted (i.e., during the 100 cell count) by the total white blood cell count to obtain the absolute number for the cell line.

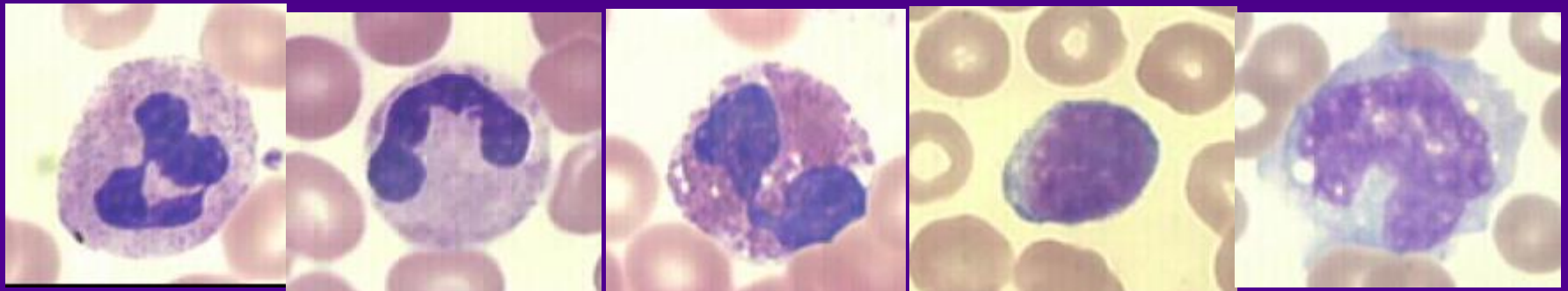
**Total WBC/ $\mu$ L x relative % = absolute no./ $\mu$ L**

**For example:**



Given a patient whose total WBC is  $8,000/\mu\text{L}$ ,

and the **relative distribution** of leukocytes on the peripheral blood smear is as shown below:



segmented  
neutrophils  
65%

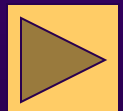
band  
neutrophils  
5%

eosinophils  
2%

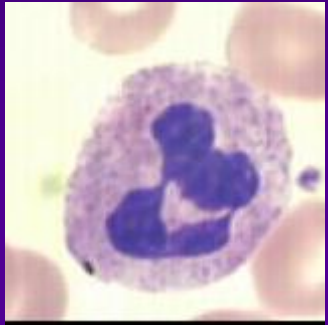

lymphocytes  
25%

monocytes  
3%

How are the **absolute numbers** determined?



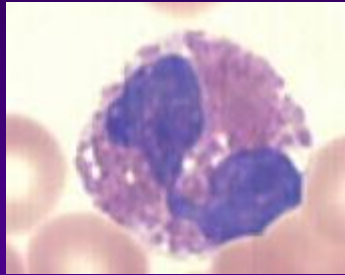
# How are the absolute numbers calculated (total WBC = 8,000/ $\mu$ L)?

<u>Cell Type</u>	<u>Relative %</u>	<u>Absolute No.</u>
	65 (8,000 x 0.65)	5,200/ $\mu$ L
<b>Neutrophil (mature segmented)</b>		
	5 (8,000 x 0.05)	400/ $\mu$ L
<b>Neutrophil (band)</b>		



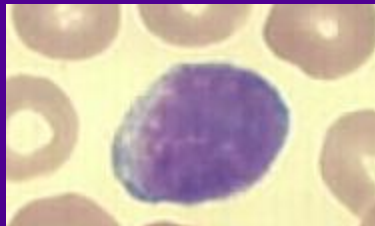
continued:

Cell Type                      Relative %                      Absolute No.



2                      (8,000 x 0.02)                      160/μL

Eosinophil



25                      (8,000 x 0.25)                      2,000/μL

Lymphocyte



3                      (8,000 x 0.03)                      240/μL

Monocyte





# Total WBC 8,000/ $\mu$ L x Relative %:

<u>Cell Type</u>	<u>Relative %</u>	<u>Absolute No.</u>
Neutrophil (mature segmented)	65	5,200/ $\mu$ L
Neutrophil (band)	5	400/ $\mu$ L
Lymphocyte	2	160/ $\mu$ L
Monocyte	25	2,000/ $\mu$ L
Eosinophil	3	240/ $\mu$ L
<hr/>		<hr/>
	<b>Total 100</b>	<b>8,000/<math>\mu</math>L</b>

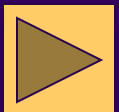


# What are the absolute reference ranges for leukocytes?

The total WBC count reference range for adults is 4,500-11,000/ $\mu$ L at Charity & University Hospitals, but may vary slightly among laboratories:

## Reference Ranges

CELL TYPE	RELATIVE %	ABSOLUTE NO./ $\mu$ L
Segmented neutrophils	42 - 72	1800 - 8000
Neutrophilic bands	0 - 5	0 - 550
Eosinophils	1 - 6	45 - 550
Basophils	0 - 0.2	0 - 200
Lymphocytes	24 - 45	1100 - 5000
Monocytes	0.4 - 10	200 - 1100

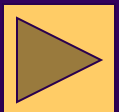


**In this case, did the relative per cents reflect the absolute numbers ?**

**The total WBC count was within the reference range.**

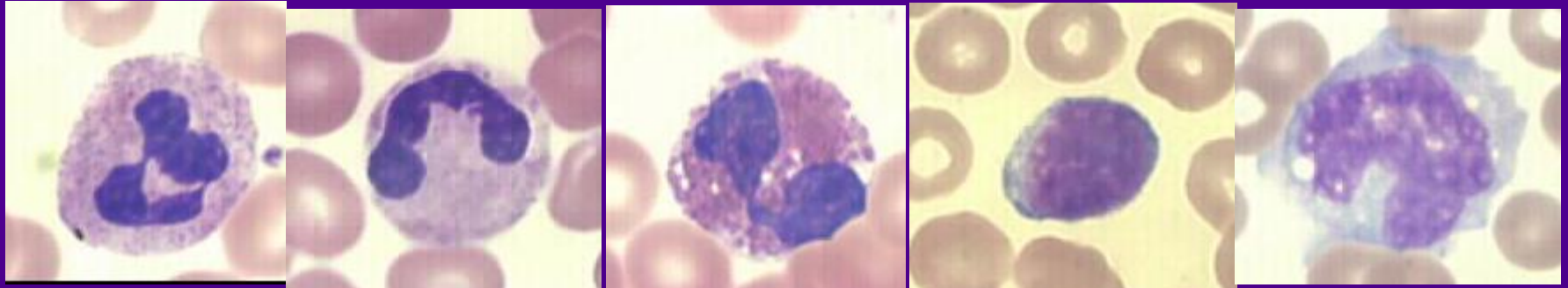
**The relative per cent for each cell type was within reference range.**

**Therefore, because both the total and relative frequencies are within reference ranges, the relative % is a reflection of the absolute numbers in terms of normal or abnormal.**



However, given a patient whose total WBC is 15,000/ $\mu$ L,

with a relative distribution of leukocytes on the peripheral blood smear the same as the previous patient:



segmented  
neutrophils  
65%

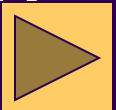
band  
neutrophils  
5%

eosinophils  
2%

lymphocytes  
25%

monocytes  
3%

Is the interpretation the same for both patients?



**What are the calculated absolute numbers in this case (i.e., total WBC = 15,000/ $\mu$ L)?**

**15000 x 0.65 = 9750 segmented neutrophils**

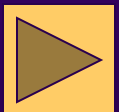
**15000 x 0.05 = 750 band neutrophils**

**15000 x 0.02 = 300 eosinophils**

**15000 x 0.25 = 3750 lymphocytes**

**15000 x 0.03 = 450 monocytes**

**The relative per cents are normal, but how do these absolute numbers **compare** with the established reference ranges?**







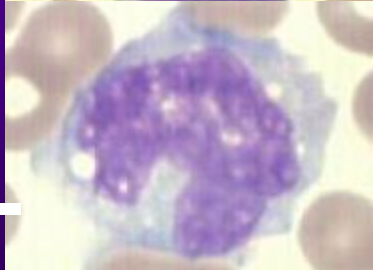
# Are the absolute numbers still within the reference ranges?

## Absolute No./ $\mu$ L

<u>Cell Type</u>	<u>Patient</u>	<u>Reference Ranges</u>
Neutrophils (mature)	9750 H	1800 - 8000
Neutrophils (bands)	750 H	0 - 550
Eosinophils	300	0 - 600
Lymphocytes	3750	1100 - 5000
Monocytes	450	200 - 1100



# Comparison of relative % and absolute numbers:

<u>Cell Type</u>	<u>Relative %</u>		<u>Absolute No.</u>
Neutrophil (mature segmented)	65		9,750/ $\mu$ L
Neutrophil (band)	5		750/ $\mu$ L
Eosinophil	2		300/ $\mu$ L
Lymphocyte	25		3,750/ $\mu$ L
Monocyte	3		450/ $\mu$ L
	<hr/>		<hr/>
	<b>Total 100</b>		<b>15,000/<math>\mu</math>L</b>





# Do the relative and absolute values have the same interpretation in this case?

The **relative percentages** for all cell types for this patient were within the reference range (i.e., normal).

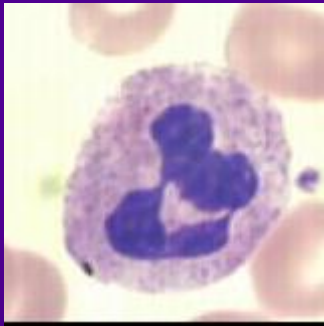
However, in this case, there is neutrophilia when converted to **absolute numbers** based on a total WBC count of  $15,000/\mu\text{L}$  because:

- ❖ segmented (mature) neutrophils =  $9,750/\mu\text{L}$  which exceeds the reference range ( $1800 - 8000/\mu\text{L}$ )
- ❖ band neutrophils =  $750/\mu\text{L}$  which also exceeds the reference range ( $0 - 550/\mu\text{L}$ ).



Given another patient whose total  
WBC is  $15,000/\mu\text{L}$ ,

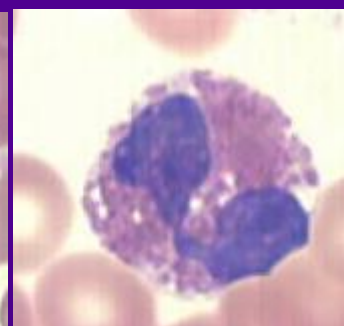
and a relative distribution of  
leukocytes on the peripheral blood  
smear as shown below:



segmented  
neutrophils  
75%



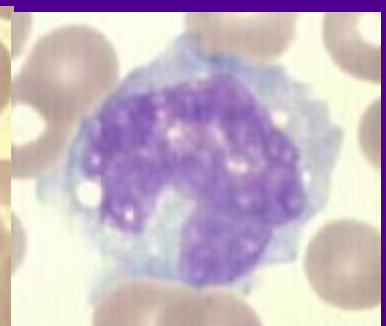
band  
neutrophils  
5%



eosinophils  
2%



lymphocytes  
15%



monocytes  
3%

How are these data interpreted?



# How are the absolute numbers calculated (total WBC = 15,000/ $\mu$ L)?

<u>Cell Type</u>	<u>Relative %</u>	<u>Absolute No.</u>	
	75	(15,000 x 0.75)	11,250/ $\mu$ L
<b>Neutrophil (mature segmented)</b>			
	5	(15,000 x 0.05)	750/ $\mu$ L
<b>Neutrophil (band)</b>			



Cell Type   Relative %   Absolute No.

		2	(15,000 x 0.02)	300/μL
----------------------------------------------------------------------------------	-----------------------------------------------------------------------------------	---	-----------------	--------

**Eosinophil**

		15	(15,000 x 0.15)	2,250/μL
----------------------------------------------------------------------------------	-----------------------------------------------------------------------------------	----	-----------------	----------

**Lymphocyte**

		3	(15,000 x 0.03)	450/μL
------------------------------------------------------------------------------------	-------------------------------------------------------------------------------------	---	-----------------	--------

**Monocyte**



# Total WBC 15,000/ $\mu$ L x Relative %:

<u>Cell Type</u>	<u>Relative %</u>		<u>Absolute No.</u>
Neutrophil (mature segmented)	75	H	H 11,250/ $\mu$ L
Neutrophil (band)	5	N	H 750/ $\mu$ L
Lymphocyte	2	N	N 300/ $\mu$ L
Monocyte	15	L	N 2,250/ $\mu$ L
Eosinophil	3	N	N 450/ $\mu$ L
<hr/>			<hr/>
	<b>Total 100</b>		<b>15,000/<math>\mu</math>L</b>



## Do the relative and absolute values have the same interpretation?

According to the **relative percentages**, this patient had increased neutrophils (neutrophilia) and decreased lymphocytes (lymphocytopenia).

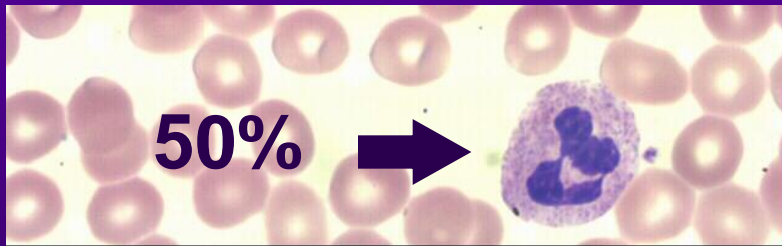
When converted to **absolute numbers** based on a total WBC count of  $15,000/\mu\text{L}$ ,

- there is indeed neutrophilia ( $11,250/\mu\text{L}$ ) which **exceeds** the reference range ( $11 - 5000/\mu\text{L}$ )
- but the number of lymphocytes (i.e.,  $2,250/\mu\text{L}$ ) is **within** the reference range ( $1000-5000/\mu\text{L}$ ).

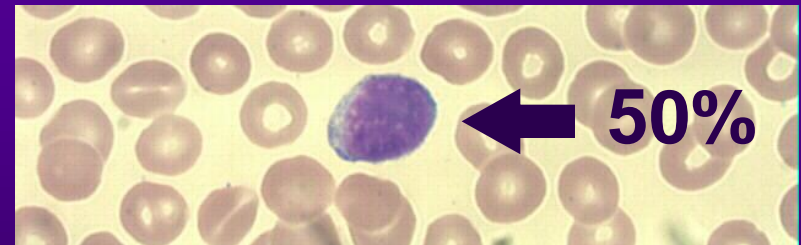


## What if the patient's total WBC is decreased? For example...

If a patient's total WBC count is  $2,500/\mu\text{L}$  and the following **relative distribution** is found on the differential WBC count, what interpretation can be made regarding increased or decreased cell lines?



neutrophils



lymphocytes

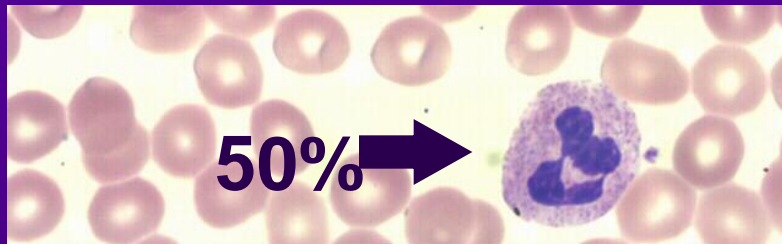




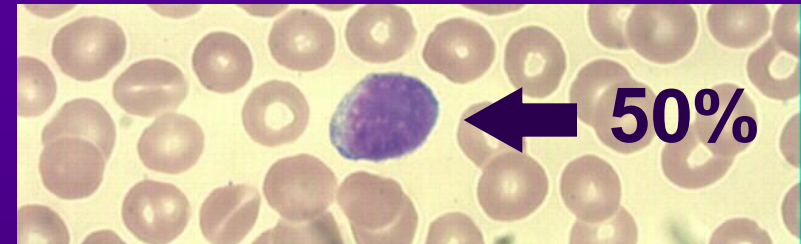
## How are these data interpreted?

According to the relative reference ranges (i.e., neutrophils 40-72% and lymphocytes 24-45%), the relative values for:

- Neutrophils (50%) were within “normal” range.
- Lymphocytes (50%) were increased.



neutrophils

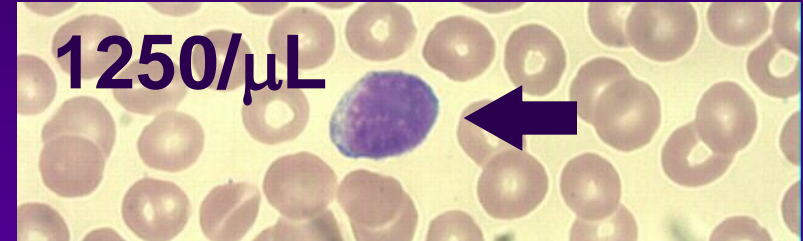
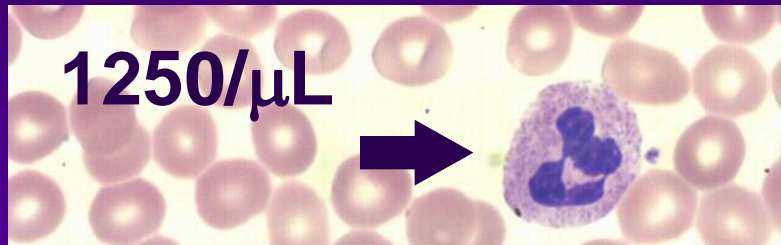


lymphocytes

- Does this patient have lymphocytosis?

# What was your evaluation?

When evaluated with the total WBC count of  $2500/\mu\text{L}$ , the absolute numbers are:



neutrophils

lymphocytes

If you responded yes (i.e., lymphocytosis), you were wrong. The patient has neutropenia. Although the relative % of PMN is “normal”, the absolute number ( $1250/\mu\text{L}$ ) is less than the lower limit of “normal”  $1800/\mu\text{L}$ ).

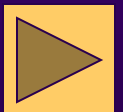
Also, even though the relative % for lymphocytes is increased, the absolute number ( $1250/\mu\text{L}$ ) is within “normal” range ( $1100 - 5000/\mu\text{L}$ ).



## Summary of relative % vs. absolute number:

An increase or decrease in a cell line is determined by the **absolute number** of that type of cell in circulating peripheral blood. To determine the absolute number, the total WBC count and frequency distribution must be determined.

Relative % is reliable as an indicator of normal only if both the percent and the total number of WBC are within normal.



## End of Relative % vs. Absolute Number

This concludes the Introduction to the Differential WBC Count. Select one of the following:

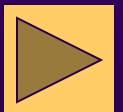
Go to Review of White Blood Cell Morphology, the next section, to continue with the exercise as designed.

OR

Return to the Main Menu and make an alternate selection.



# ***Review of WBC Morphology***



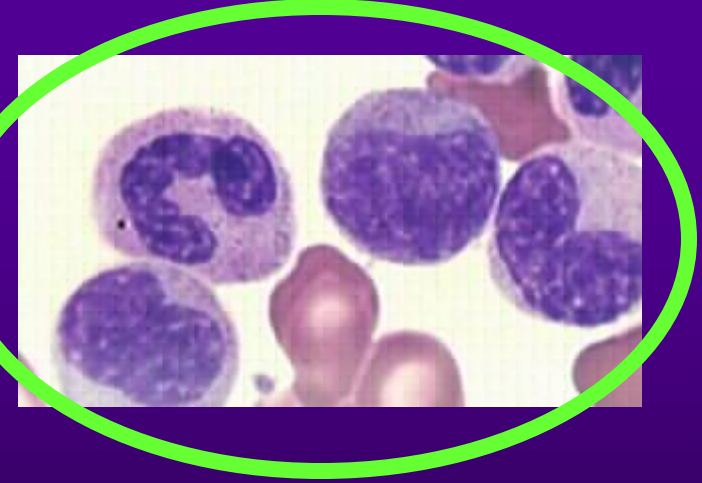
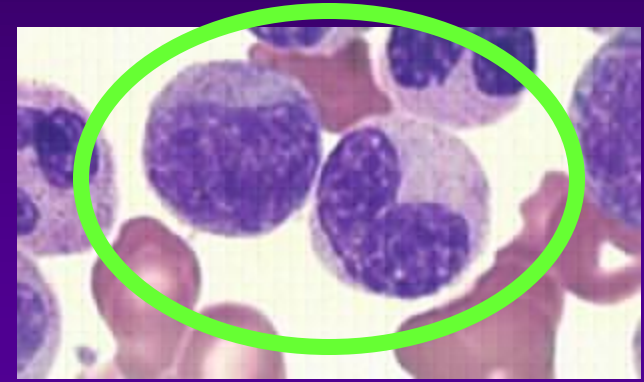
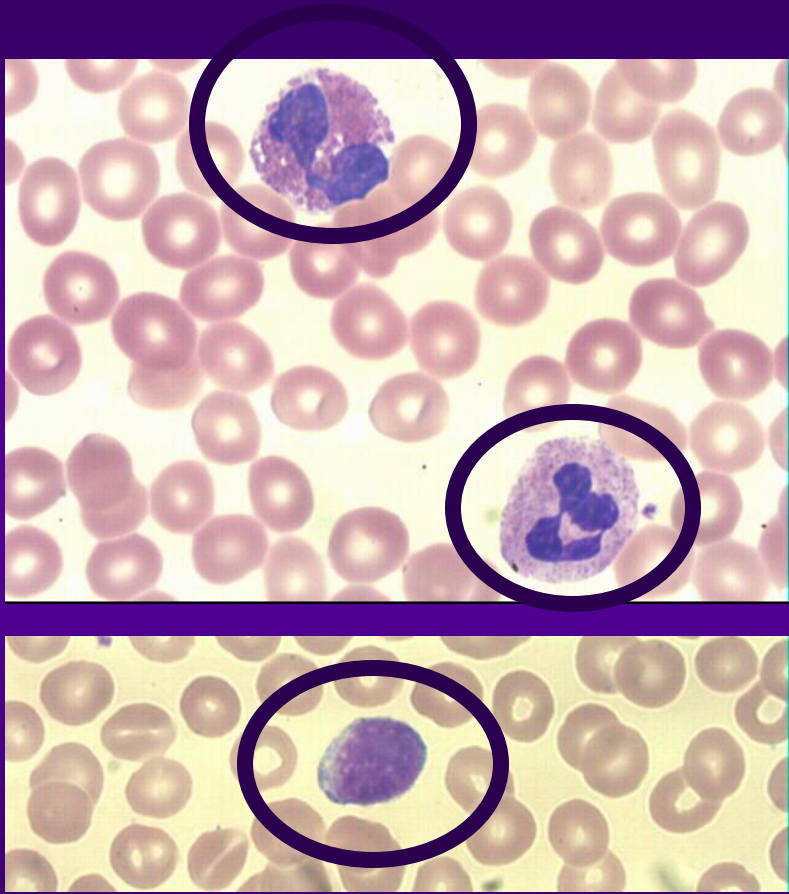
# What other study of WBC is included in a “diff”?

**In addition to the differential WBC count, the leukocytes are examined as to maturity and morphology.**

**Are immature cells present? Do the cells show abnormal acquired or inherited morphologic alterations?**



# E.g., are the WBC mature or immature?

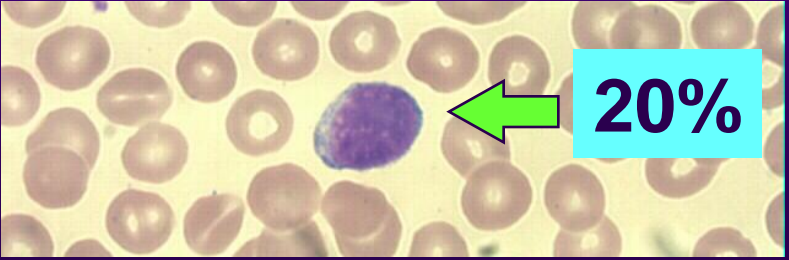
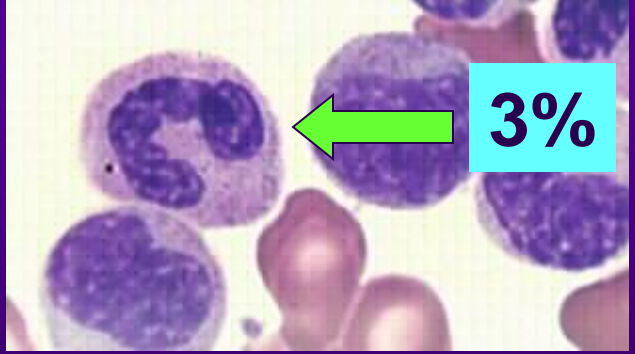
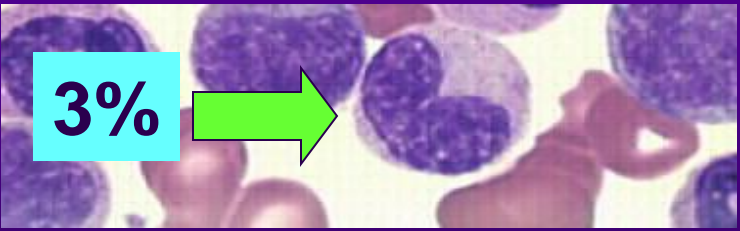
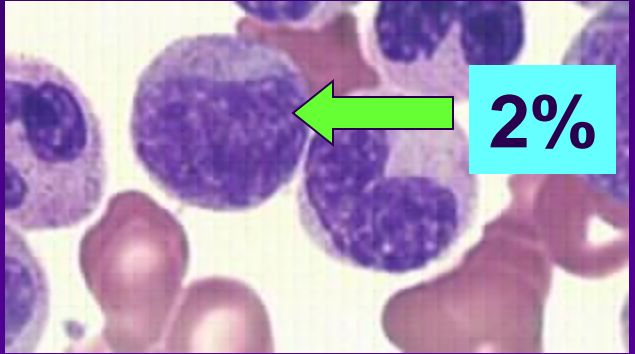
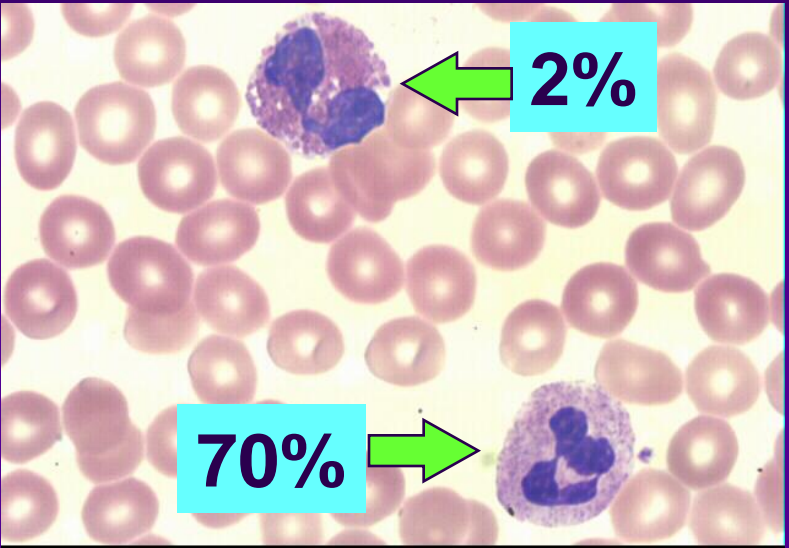


mature cells      or      immature neutrophils





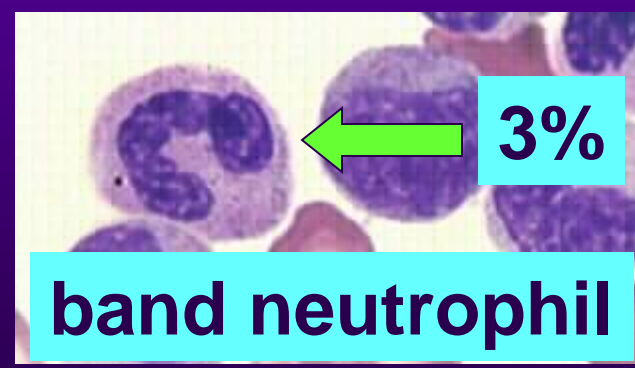
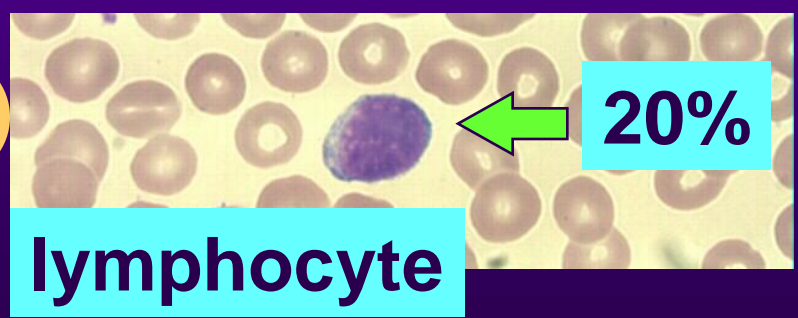
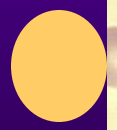
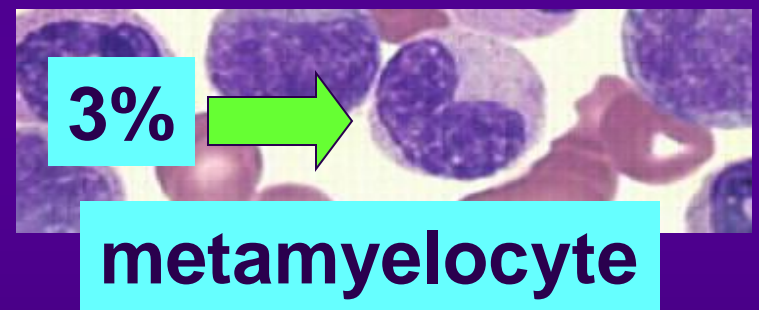
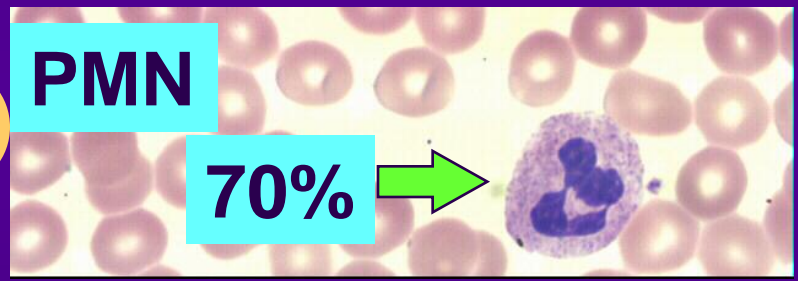
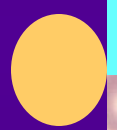
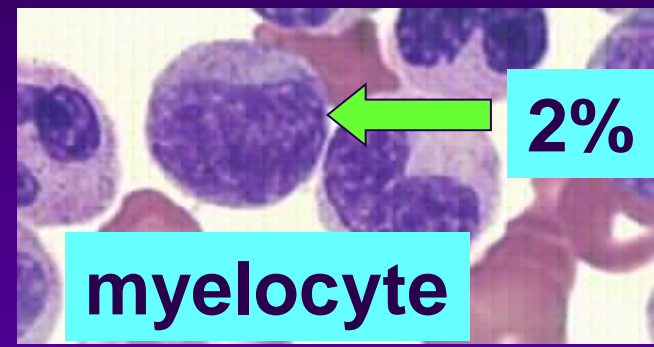
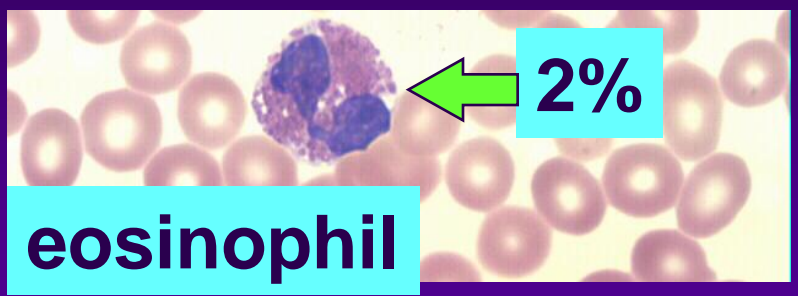
For example, if the mature and immature cells shown in the example "diff" were present in the percentage shown below:



# What leukocytic cells can be identified?

## mature

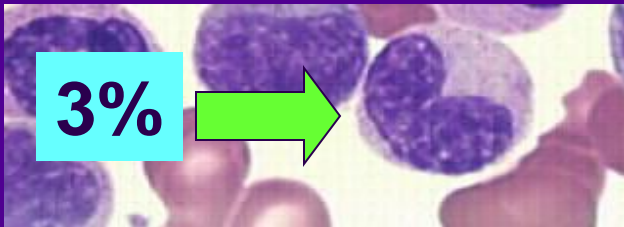
## immature



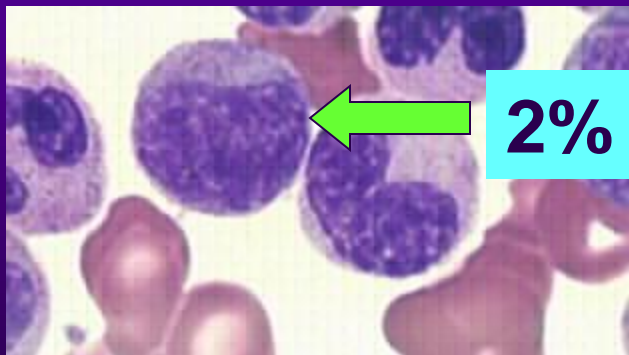
# Are the immature cells shown in the example ever seen in normal blood?



band neutrophil (normally may be present in blood up to about 5%)



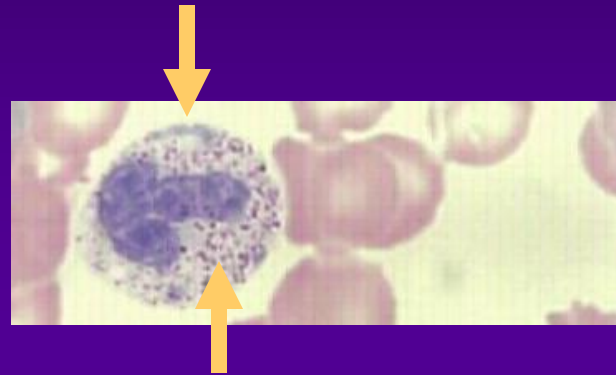
neutrophilic metamyelocyte (are not normally present in blood)



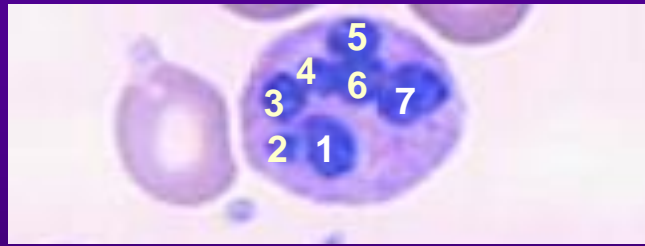
neutrophilic myelocyte (are not normally present in blood)



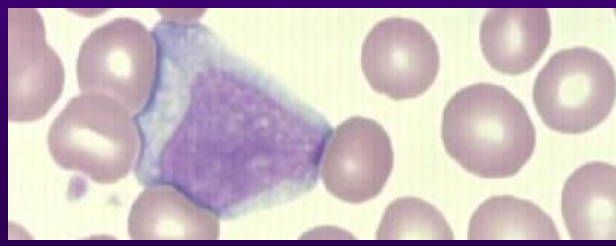
# Examples of acquired non-neoplastic alterations that can be differentiated on a "diff", e.g.?



neutrophil with Dohle bodies  
and toxic granulation



hypersegmented neutrophil

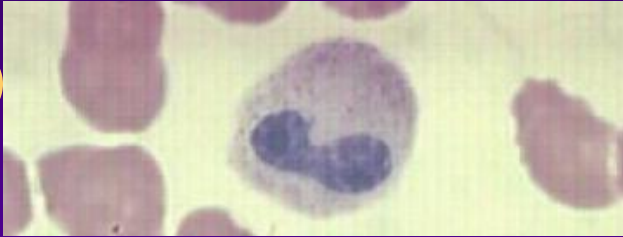


atypical/reactive lymphocyte





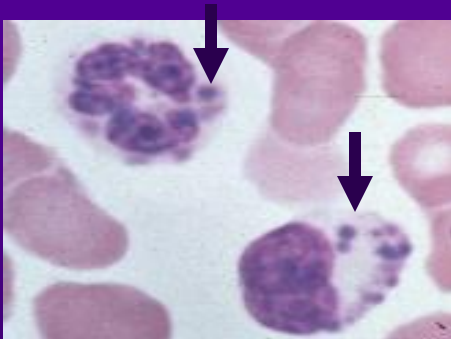
# Examples of inherited abnormal non-neoplastic alterations of neutrophils that can be differentiated on a "diff".



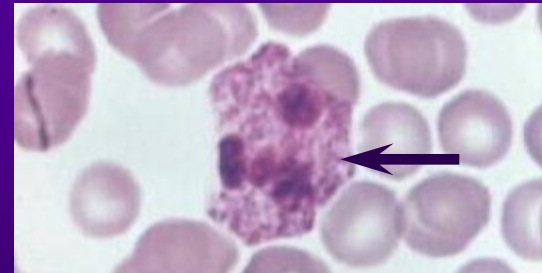
hyposegmented neutrophil in Pelger-Huet anomaly



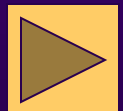
cytoplasmic blue bodies in May-Hegglin anomaly



cytoplasmic large black granules in Chediak-Higashi syndrome



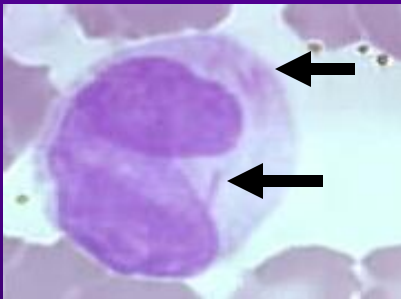
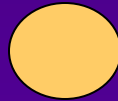
cytoplasmic black granules in Alder-Reilly anomaly



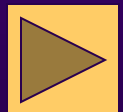
## Examples of neoplastic WBC alterations that can be differentiated on a “diff”:



**hairy cell lymphocytes seen in hairy cell leukemia**



**Auer rods seen in some variants of acute myelocytic leukemias**



# Are cell types other than WBC differentiated and enumerated as part of the “diff”?

Although NRBC are not included in the 100 white blood cell count, identification and enumeration of the number of NRBC/100 WBC is a part of the information provided by a “diff”. For example, the number of



proerythroblast



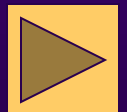
basophilic  
erythroblast



orthochromatophilic  
erythroblast



polychromatophilic  
erythroblast





## ***End of CBC - 2***

**This concludes the CBC – Part 2 segment of the study module, “The Complete Blood Cell Count (CBC)”.**

**Click on Main Menu to review a section.**

**OR**

**Click on  to quit the exercise.**

# ***THE END***

**Additional modules of the CBC are presented in:**

**CBC – Part 1      The hemogram**

**CBC - Part 3      RBC morphology & platelet estimate**

**CBC – Part 4      Post-test**

**Review of “Hematology Atlas”, a module on normal and abnormal blood cell morphology and maturation is recommended as a prerequisite for the CBC modules.**

**quit**