White blood cell (WBC) count

Introduction and principle: The white blood cell count denotes the number of white blood cells per unit volume of whole blood.

- Normal WBC count range from 4000 11000 cells/ mm³ this count varies with age.
- WBC count is useful to indicate infections or may be employed to follow the progress of certain diseases.
- White blood cells in the circulation are not white in the sense that a sheet of white paper is white, but in the sense that they are transparent and not colored.
- White cells are fewer in number than red cells.
- White blood cells are counted in a similar manner to red cells, using a haemocytometer.

Methods:

- 1- Manual method.
- 2- Electronic cell counting

Manual white cell count material and instruments

- 1. Anticoagulated whole blood.
- 2. Turk's diluting fluid composed of :
 - Glacial acetic acid, 3 ml → to haemolyse RBCs.
 - Aqueous gention violet $(1\% \text{ w/v}) 1 \text{ ml} \Rightarrow$ to color the nuclei of WBC
 - Distilled water up to 100 ml.
- 3. WBC pipette. Which is composed of a stem, mixing chamber, white bead inside the mixing chamber, aspiration tube (rubber sucking tube)
- 4. Haemocytometer (Neubauer's counting chamber) with a cover slip.
- 5. Microscope.
- 6. Lancet.
- 7. Cotton

Procedure:

- Obtain a drop of blood in the same manner as in RBC count. Draw blood up to the mark 0.5 using WBC pipette.
- Remove blood from outside of the pipette with a clean gauze.
- Aspirate diluting fluid up to mark 11. The dilution is 1:20.
- Gently rotate the pipette horizontally with your hand to ensure a proper amount of mixing for 3 minutes.
- After mixing discard the first four drops of the mixture.
- Fill the counting chamber with diluted blood by holding the pipette at 45° with the slide and allow the mixture to seep under the cover slip, the filled chamber should be allowed to stand for a minute prior to counting.
- Count the WBCs using the low power 10 x objectives.

• Count all WBCs in four large corner squares and add the result together to obtain the total number of cells counted. In counting the cells that touch the outside lines of the large square, count only those that touch the left and lower outside margin. The WBCs look like black dots.

PRECAUTIONS: Are the same as for RBC count.

Calculation:

Count the number (N) of cells in 4 large squares located at the four corners of the chamber. The size 4 large squares in which "N" numbers of cells are found is: $1 \times 1 \times 1 / 10 \times 4 = 4 / 10 \text{ mm}^3$

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Where 1 mm is the sideline of the large square, 1/10 mm is the depth of the counting chamber between coverslip and the ruling, 4 is the number of large squares used to count.

Therefore the total numbers of cells in 1 mm^3 are = N x 10/4 (diluted sample).

The actual total number of cells before dilution should be:

N x 10/4 x 20 = **N x 50**

Medical applications

Increased number of WBCs indicates that there is **leukocytosis** which could be physiological or pathological such as infection.

• Physiological Conditions

1. Age:

a. Newborn will always have an increased count. <u>a newborn</u> has a high white blood cell count, ranging from 9,000 to 30,000 leukocytes / mm^3 . This number falls to adult levels within two weeks.

b. Childhood, pregnancy and delivery shows increased count.

c. There is no significant change in old age compared to adult values.

2. Females during pregnancy and parturition.

3. Stress like severe exercise, severe pain, and excitation.

4. Diurnal variation: WBC count may vary from hour-to-hour (highest count in evening and lowest count in morning).

5. Digestive leukocytosis (after digestion).

6. Injection of adrenaline.

7. After removal of spleen (spleenectomy)

• Pathological Conditions

1. Acute pyogenic infections (e.g. pneumonia, appendicitis and tonsillitis).

2. Leukemia (abnormal increase with immature cells) count may go up to 1,00,000 to 3,00,000 per cu mm.

3. Acute hemorrhage.

4. Tissue damage resulting from burns, operations, myocardial infarction.

- 5. Malignant neoplasms.
- 6. Metabolic disorder (e.g. gout, diabetic acidosis).

Leucopenia is a condition of decreased number of WBCs. Leukopenia occurs when the WBC falls below 4,000 / mm³. Patients with severe leukopenia should be protected from anything that interrupts skin integrity, placing them at risk for an infection that they do not have enough white blood cells to fight. For example, leukopenic patients should not have intramuscular injections, rectal temperatures or enemas. A WBC of less than 500 / mm³ places the patient at risk for a fatal infection.

Physiological Leucopenia: Rare but sometimes due to:

- 1. Exposure to cold
- 2. Aspirin.
- 3. The WBC count tends to be lower in the morning and higher in the late afternoon.

Pathological Leucopenia

1. Infection → Typhoid, viral, or overwhelming bacterial infections (It is not uncommon for the elderly to fail to develop leukocytosis as a response to infection).

- 2. Starvation \rightarrow Malnutrition.
- 3. Viral infections → Measles, chickenpox, influenza and rubella
- 4. Drugs → Antimetabolites, antimicrobials (Sulfonamides, chloramphenicol)

5. Hematological disorders → Aplastic anemia, pernicious anemia, irradiation.WBC counts are age-related. It is not uncommon for the elderly to fail to develop leukocytosis as a response to infection.