



## W.B.C. Diluting Fluid

R016

WBC diluting fluid is used for performing the WBC (Leucocyte) count.

### Composition\*\*

#### Ingredients

Glacial acetic acid	2.00 ml
Gentian Violet (1% w/v)	1.00 gm
Distilled water	97.00 ml
Final pH ( at 25°C)	2.2±0.2

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

- 1) Draw EDTA anticoagulated blood to 0.5 mark in the capillary end of WBC pipette.
- 2) Carefully, wipe excess blood outside the pipette by using cotton.
- 3) Draw diluting fluid up to 11 mark.
- 4) Mix the contents in pipette and after 5 minutes by discarding few drops, fill the counting chamber and allow the cells to settle for 2-3 minutes.
- 5) Focus on 1 of the "W" marked areas (each having 16 small squares) by turning objective to low power. 10X
- 6) Count cells in all 4 "W" marked corner squares.

### Principle And Interpretation

WBC diluting fluid is used for performing the WBC (Leucocyte) count. Glacial acetic acid lyses the red cells. Gentian violet slightly stains the nuclei of the leucocytes. The blood specimen is diluted 1:20 in a WBC pipette with the diluting fluid and the cells are counted under low power of the microscope by using a counting chamber. The number of cells in undiluted blood is reported per cumm ( $\mu$ l) of whole blood.

### Quality Control

#### Appearance

Purple coloured, clear solution.

#### Clarity

Clear with no insoluble particles.

#### pH

2.00-2.40

#### Calculation

Number of WBCs/cumm( $\mu$ l) of whole blood = No. of WBCs counted X Dilution / Area counted X Depth of fluid  
where, Dilution=20; Area counted=  $4 \times 1 \text{ sq. mm} = 4 \text{ sq. mm}$ ; Depth = 0.1 mm (constant)

No. of leucocytes / cu mm ( $\mu$ l) of whole blood = No. of cells counted X 20 / 4 X 0.1  
= No. of cells counted X 50

### Storage and Shelf Life

Store below 30°C in tightly closed container and away from bright light. Use before expiry date on label.

### Reference

- 1) Text book of Medical Laboratory Technology; Praful B. Godkar

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#### Disclaimer :



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