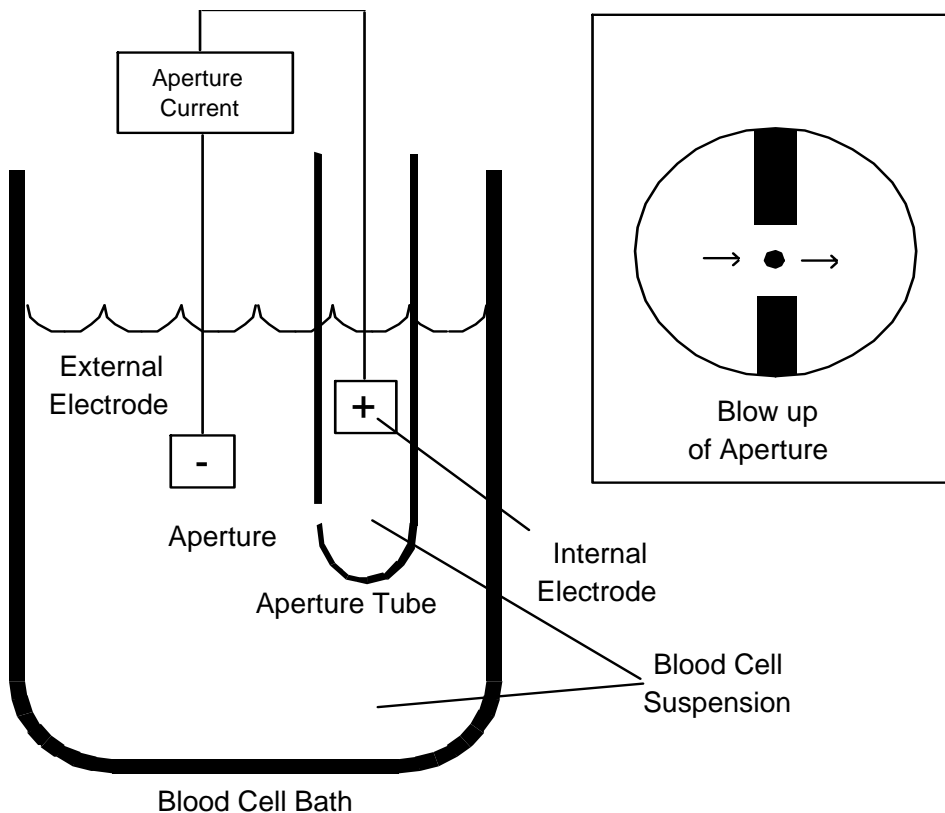




HEMATOLOGY ANALYZERS

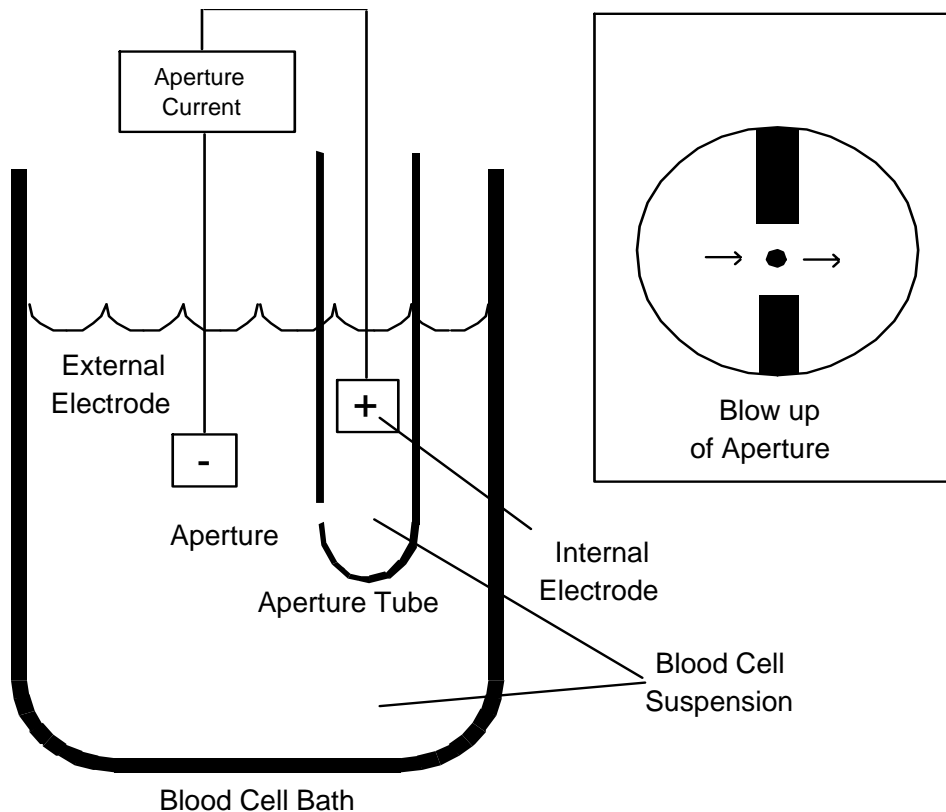
- During this presentation I will familiarize you with the various operating principles, components and functions of a Hematology Analyzer.
- During this presentation I will go over:
 - The general sequence of events involved in counting RBCs and WBCs.
 - I will describe the Coulter Method of counting cells
 - And I will cover differential analysis (or, how we count individual types of WBCs).

The Coulter Method



- ▶ A small opening, called an *aperture*, is between two electrodes in an electrolytic solution containing a whole blood sample.
- ▶ The electrodes on either side of this aperture allow an electrical current to flow through the aperture.

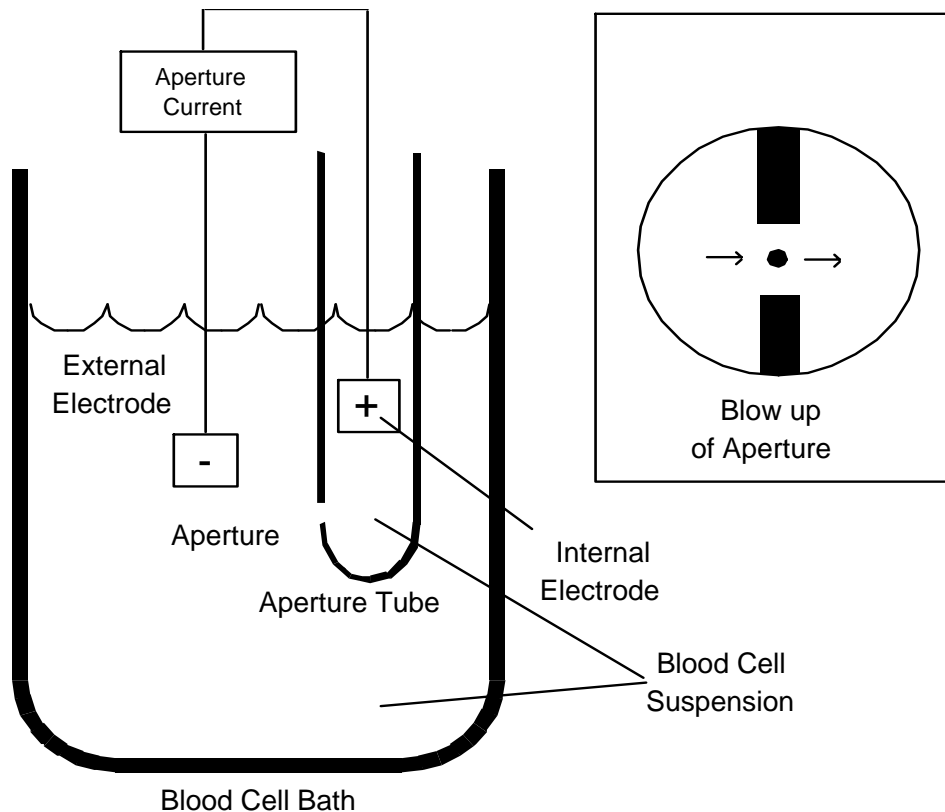
The Coulter Method



Cells have higher electrical resistance than the electrolytic solution.

- *When a cell passes through the aperture, it acts as a variable resistor changing the amount of current flow between the electrodes.*
- *The generates an electrical pulse which is recorded as 1 cell.*
- *Summing the pulses gives us the number of cells counted.*
- *The size of the pulse is proportional to the size of the cell . . . the bigger the cell, the bigger the pulse.*

The Coulter Method



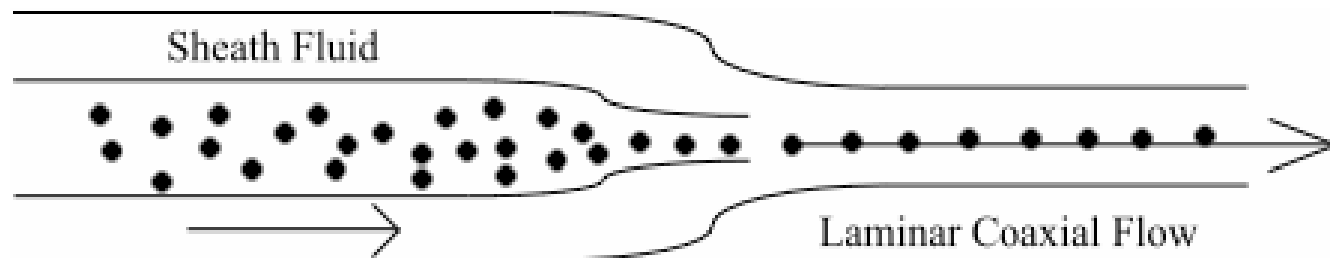
- ▶ *The amount of electrolytic suspension drawn through the aperture is precisely controlled to allow the system to count and size particles for an exact reproducible volume.*
- ▶ *Several thousand particles per second are individually counted and sized with great accuracy.*



Flow Cytometry (Laser Diffraction Method)

- *Cytometry*
 - *The measurement of the physical and chemical characteristics of cells*
- *Flow cytometry*
 - *The technique where such measurements are made as the cells pass in a fluid stream through a measuring point surrounded by an array of detectors.*

Flow Cytometry (Laser Diffraction Method)



- ▶ *Assuming laminar (non-turbulent) flow of liquid through a tube, referred to as sheath fluid, viscous drag at the outer boundary of the tube will result in a higher flow velocity nearer the center of the tube.*
- ▶ *The **Bernoulli Effect** associates such changes in velocity with inverse changes in pressure such that any particle in the fluid will move towards and remain in the center.*



Flow Cytometry (Laser Diffraction Method)

- *To maintain this focusing, the flow of the stream requires a velocity of several meters per second.*
 - *At this speed, a typical cell would travel its own diameter in a few microseconds.*
 - *This means we need a system capable of rapid analysis; this is done with a highly focused laser beam for excitation and sensitive photo-multiplier tubes (PMTs) for detection.*



Flow Cytometry (Laser Diffraction Method)

- The laser beams are usually focused to a spot between 10 and 60 microns in diameter.
- As a cell passes through the laser beam, several physical processes take place:
 - *Absorption*
 - *Diffraction*
 - *Refraction*
 - *Reflection*
- *The magnitudes and patterns of these pulses are sorted electronically.*



Flow Cytometry (Laser Diffraction Method)

