Flow-cytometrie

Měřící princip

Měření rozptylu světla na buňkách a biologických částicích (nefelometrie)

Měření fluorescence obarvené DNA a RNA

Měření impedance procházejících buněk a biologických částic (elektrochemická metoda)

Měření absorbance hemoglobinu (spektrofotometrie)

Měřící zařízení

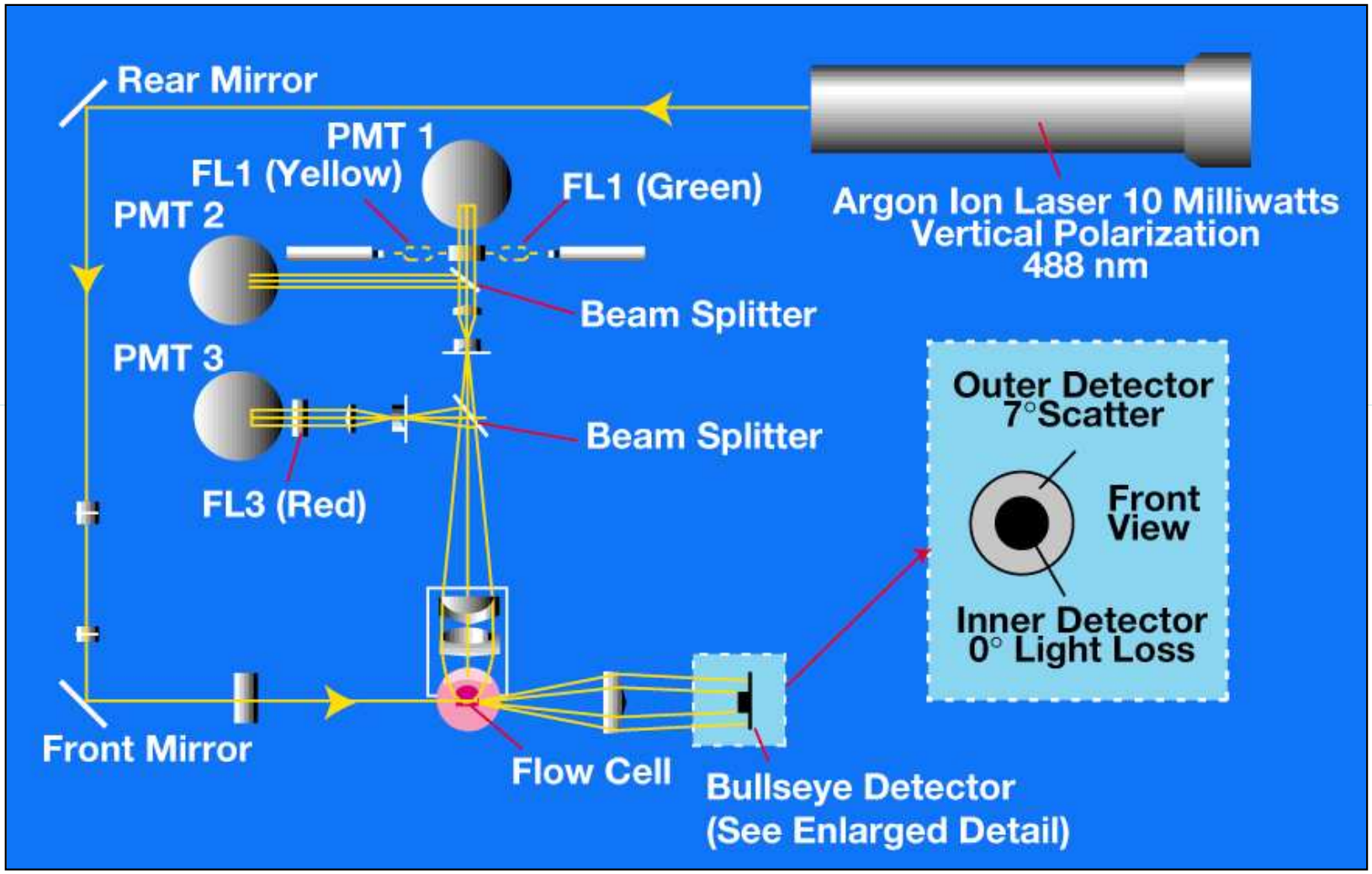
Optický systém

Průtoková optická cela (leukocyty, erytrocyty, trombocyty, retikulocyty)

Komůrka k měření impedance (el. odporu) erytrocyty, trombocyty

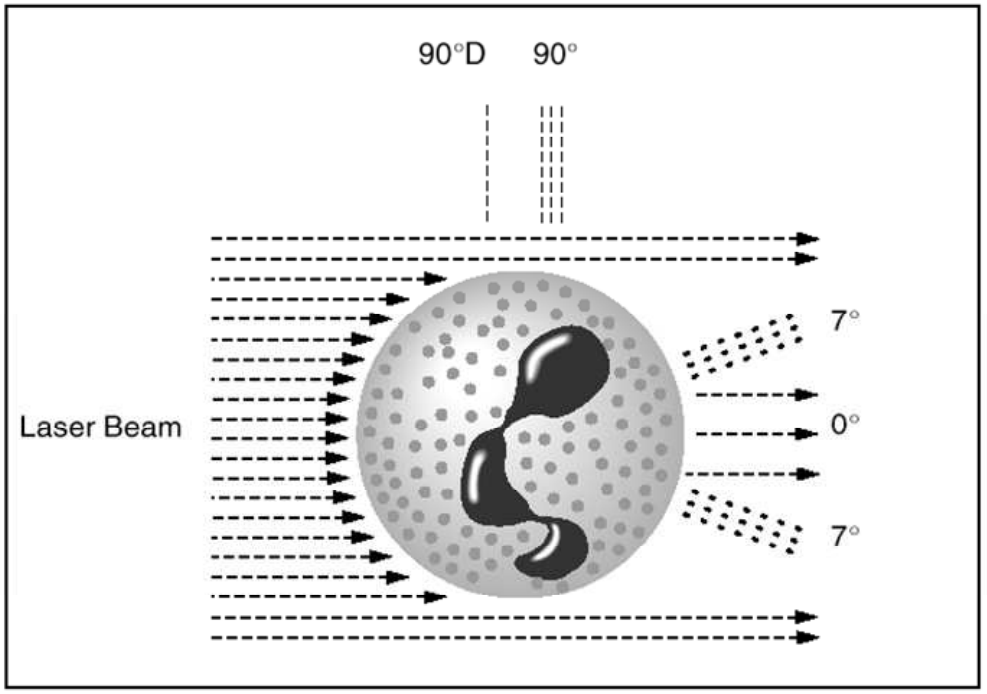
Průtoková spektrofotometrická cela (hemoglobin)

Optical Bench



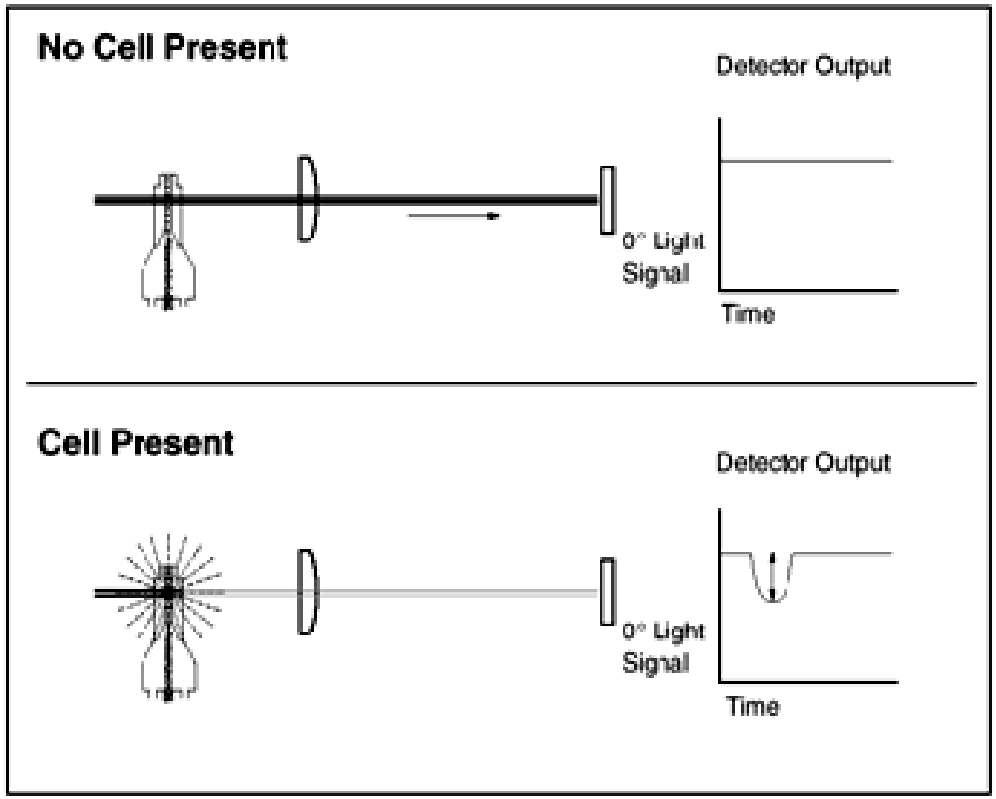
Multi **A**ngle **P**olarised **S**catter **S**eparation (**MAPSS**)

0° Light Loss:	SIZE
7° Scatter:	COMPLEXITY
90° Scatter:	LOBULARITY
90° D Scatter:	GRANULARITY



0^0 Light Loss

- 0^0 Light Loss is proportional to cell SIZE
- Full light intensity = no cells in the laser beam
- Decreased light intensity = cells in the laser beam
- Detected by the inner element of the Bull's Eye Detector

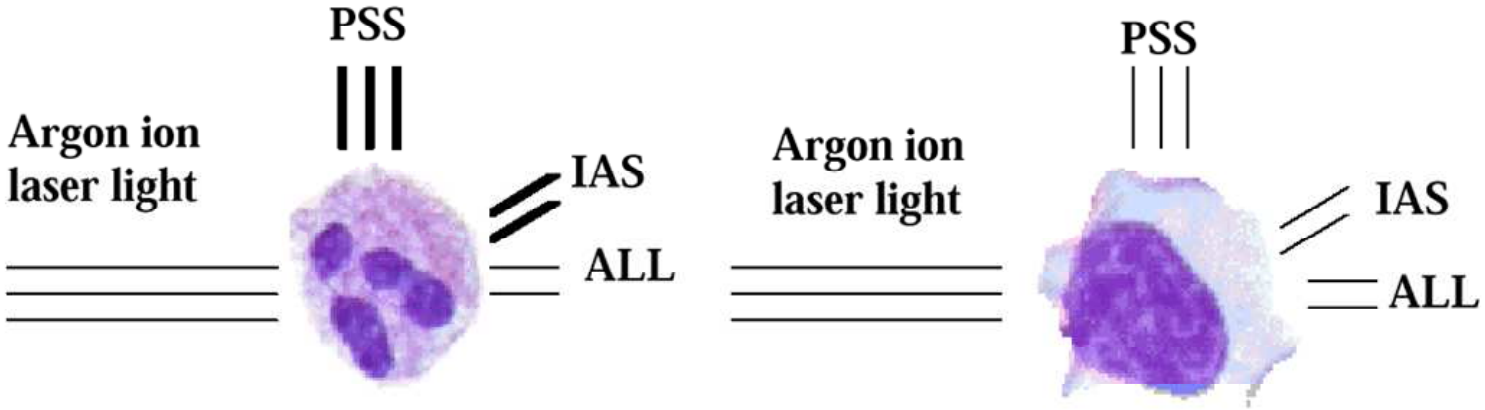


7° Light Scatter

- ✓ A cell with a complex internal structure produces a larger 7^{o} Light scatter signal than a cell of low complexity
- ✓ 7° Light Scatter can be used to measure cellular complexity
- ✓ Greater cell complexity = greater low angle scatter
- ✓ Detected by the outer element of the Bull's Eye Detector

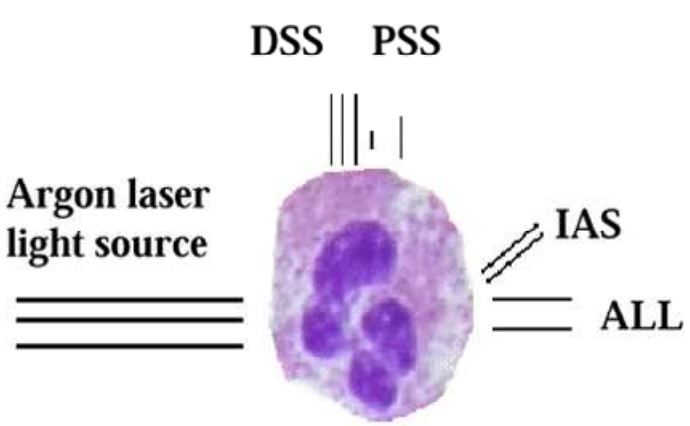
90° Light Scatter

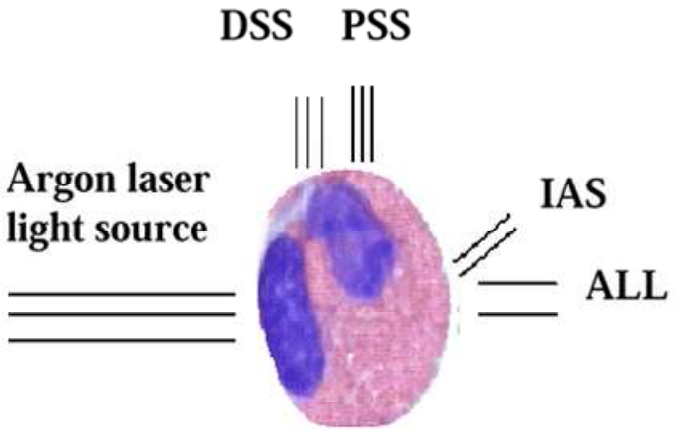
- ✓ 90° Light Scatter is used as a measurement of the lobularity of a cell's nucleus
- ✓ 90° Light Scatter is a function of NUCLEAR LOBULARITY
- ✓ Greater lobularity = greater 90° Light Scatter
- ✓ 90° Light Scatter is detected by a photomultiplier tube (PMT2)



90° **D**epolarized Light Scatter

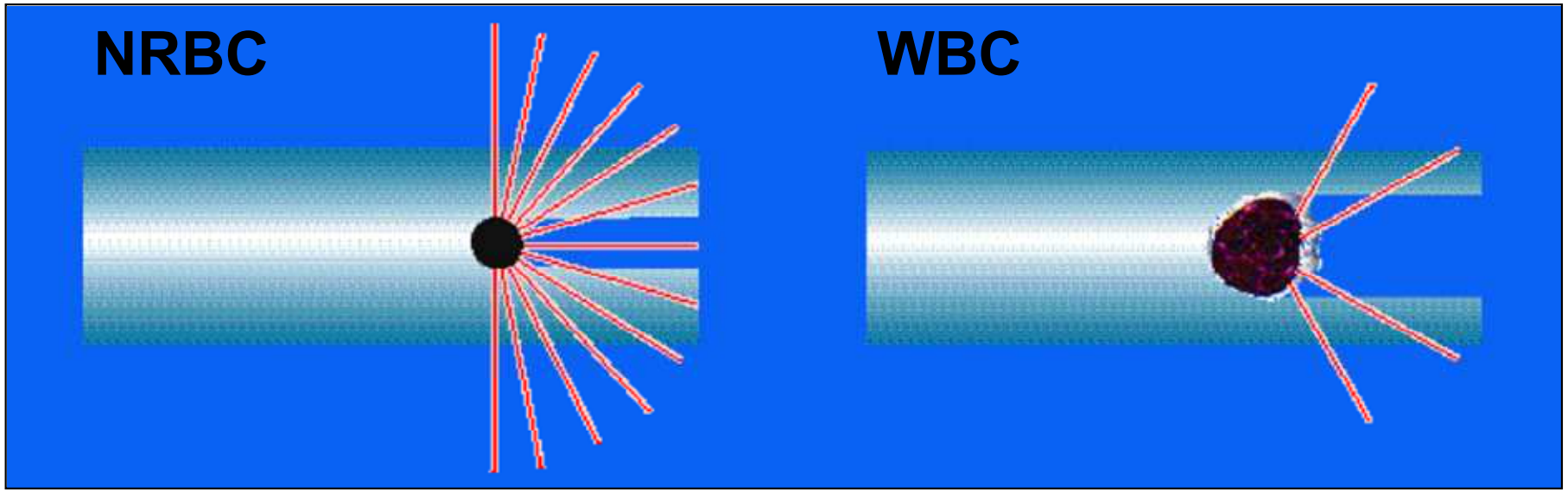
- ✓ 90° **D**epolarized Light Scatter can be used to differentiate eosinophils from neutrophils
- ✓ Granules found in eosinophils, depolarize light during the course of scattering it
- ✓ 90° D is detected by a photomulitiplier tube (PMT1)





Fluorescence

Red fluorescence is a measurement of the amount of DNA stained by the dye.
Axial light loss (0º) measures cell size.



WBC Differential

- ✓ Differential Analysis by MAPPS
- ✓ Cell Populations are colour coded
- ✓ Flags to alert of abnormalities