INTRACELLULAR RHEOLOGY OF RED BLOOD CELLS

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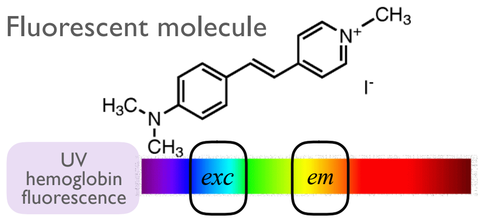
Molecular rotors

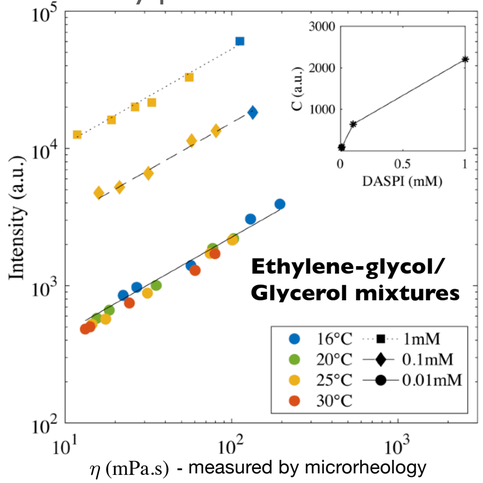
➤ DASPI Molecular rotor

Fluorescent molecule



- DASPI spectral properties are decoupled from those of hemoglobin

➤ Viscosity probe in model fluids



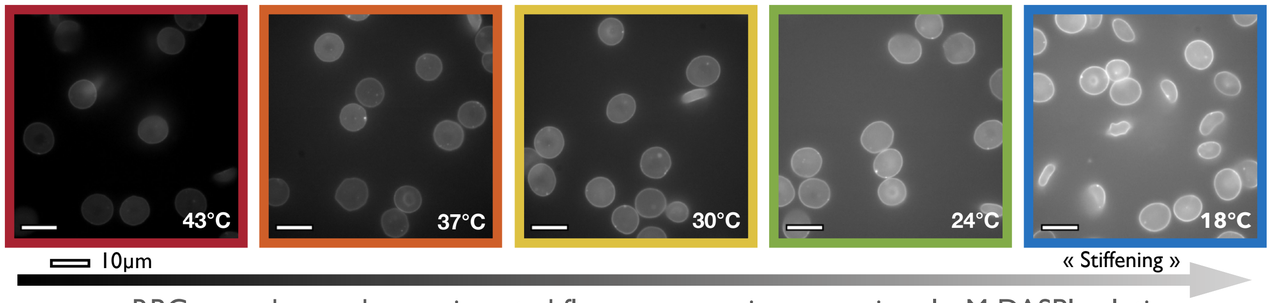
- Förster-Hoffman relation[1]

$I = C \cdot \eta^x$ with exponent $x = 0.7$

- DASPI signal depends little on temperature

« Stiffening » of RBCs

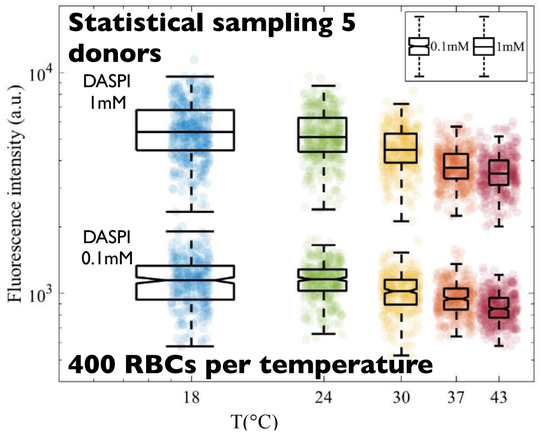
➤ Temperature-stiffened RBCs : membrane rigidity and cytosol viscosity decrease with temperature[2,3]

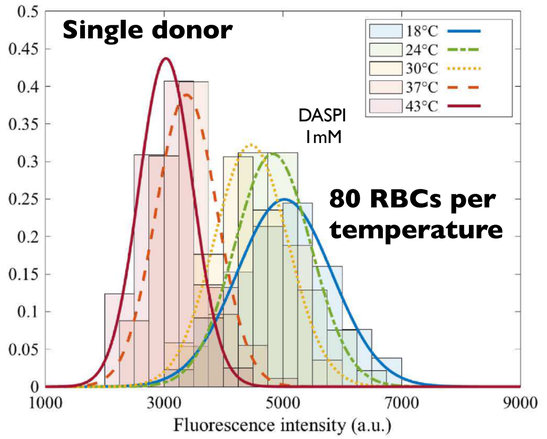


RBCs are observed on an inverted fluorescence microscope in a 1mM DASPI solution
Temperature is controlled by an objective heater through the immersion oil in contact with the sample

- Cells fluorescence increases with decreasing temperature i.e with stiffening
- DASPI penetrates homogeneously into RBCs (confocal microscopy images not shown)

➤ Fluorescence intensity and RBC stiffness

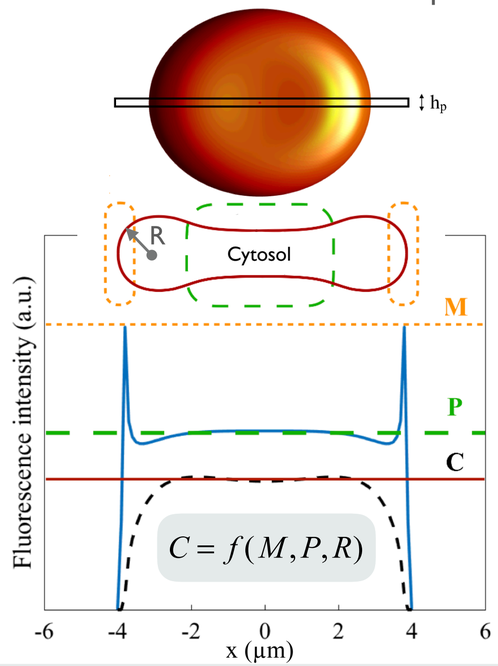




- The decrease in intensity with temperature reflects the decrease in cell stiffness
- The technique allows to evaluate the stiffness of individual cells (1dot = 1 RBC) and the stiffness heterogeneities within a sample (width of the intensity distribution)

Intracellular rheology

The signal is composed of membrane and cytosol contributions. By working on intensity profiles, we propose a geometric model of RBC for contribution separation



Our RBC model provides (not shown) :

- The separation of membrane and cytosol contributions
- Cytosol fluorescence consistent with independent measurements of hemoglobin viscosity with temperature

DASPI: intracellular nanorheology probe

A. Briole, T. Podgorski, and B. Abou, Molecular Rotors as intracellular probes of Red Blood Cell stiffness, *submitted to Soft Matter, 2021*