

GEM4 - AUGUST 2006

8/10/06

EXPERIMENTAL TECHNIQUES

1. OPTICAL TWEEZERS - DR. MATTHEW J. LANG (MIT)

FROM MOLECULAR MECHANICS TO CELLULAR MACHINERY
FORCES ON OPTICAL TRAPS

- SCATTER \Rightarrow PUSH BEAD OUT OF FOCAL POINT

- GRADIENT \Rightarrow PUSH BEAD TOWARDS FOCAL POINT

STABLE TRAP: $F_{\text{SCATTER}} = F_{\text{GRADIENT}}$

TRAPPING LASERS \Rightarrow NEAR INFRARED (LESS DAMAGING TO
BIOLOGICAL SPECIMENS)

CALIBRATIONS \Rightarrow CHARACTERIZE TRAP STIFFNESS

$$F_{\text{TRAP}} = K \Delta x$$

K = TRAP STIFFNESS (HOOKEAN)

Δx = DISPLACEMENT OF PARTICLE FROM CENTER OF TRAP

2. ATOMIC FORCE MICROSCOPY (AFM) AND MOLECULAR FORCE MICROSCOPY - DR. KRYSZTYN VAN ULJET (MIT)

AFM \Rightarrow "SEEING BY FEELING"

\hookrightarrow CANTILEVER WITH PROBING TIP \Rightarrow MEASURES

VERTICAL DISPLACEMENTS / FORCES

★ FEEDBACK LOOP TO APPLY CONSTANT FORCE WHILE
IMAGING. (FINE STRUCTURE)

HIGH RESOLUTION DUE TO ERROR BETWEEN CONSECUTIVE
POINT SCANS

FOR FORCE SPECTROSCOPY (MOLECULAR) THE AFM CAN BE USE

TO "POKE" THE SPECIMEN AND THEN RETRACT THE PROBE AND LOOK FOR THE HYSTERESIS TO CHARACTERIZE THE TIP-SPECIMEN INTERACTION.

THIS APPROACH CAN BE USED TO PROBE SINGLE MOLECULAR INTERACTIONS \Rightarrow DIFFICULTY IN OBTAINING THE RIGHT CHEMISTRY

3. MICRORHEOLOGY OF COMPLEX FLUIDS - DR. PETER SO (MIT)

- ELASTIC COMPONENT (G')
 - LOSS COMPONENT (G'')

$G^*(\omega) = G'(\omega) + iG''(\omega)$
 COMPLEX SHEAR MODULUS IS FREQUENCY DEPENDENT

(ACTUAL DEFORMATION)

- ACTIVE METHOD \rightarrow MAGNETIC MICRORHEOMETER
 - PASSIVE METHOD \rightarrow SINGLE PARTICLE TRACKING
 (THERMAL FORCES) \rightarrow MULTIPLE PARTICLE TRACKING
- } HIGH SPEED

MAGNETIC MICRORHEOMETER

\rightarrow GENERATES MAGNETIC FIELD TO ATTRACT FERRO-MAGNETIC PARTICLE

\rightarrow FORCE DEPENDS ON FIELD GRADIENT AND PARTICLE VOLUME

\rightarrow BANDWIDTH LIMITED BY INDUCTANCE (\sim KHz)

PROBLEMS:

\rightarrow SIZE OF PARTICLE

\rightarrow BEAD ROLLING ON SURFACE

SINGLE PARTICLE TRACKING

THERMAL FORCES: LANGEVIN EQUATION

$$m\ddot{u} = \underbrace{f(t)}_{\text{FORCE}} + \underbrace{\int_0^t \gamma(t-t') \dot{u}(t') dt'}_{\text{MEMORY FUNCTION}}$$

ACCELERATION

WHAT NEEDS TO BE MEASURED IS THE MEAN SQUARE DISPLACEMENT (MSD)

$$\langle \Delta R^2(\tau) \rangle = \langle (\vec{v}(\tau) - \vec{v}(t))^2 \rangle$$

$\vec{v}(t)$ COMES FROM POSITION TRACKING OF PARTICLE
 - RESOLUTION DEPENDS ON AMOUNT OF PHOTONS REACHING THE DETECTOR.

TWO- AND MULTIPLE PARTICLE TRACKING

LOOKS AT CORRELATED MOTION BETWEEN PARTICLES

→ DOES NOT DEPEND ON PARTICLE SIZE BUT ON DISTANCE BETWEEN PARTICLES

→ MEASURES GLOBAL PROPERTIES INSTEAD OF LOCAL PROPERTIES

→ DRAWBACK: USE OF WIDE FIELD CAMERA INSTEAD OF PHOTODIODE ⇒ LOWER TIME RESOLUTION

4. MICROSCOPY - DR. PETER SU (MIT)

SEE SLIDES FOR RAY TRACING EXAMPLES AND MICROSCOPE MODEL.

DIFFRACTION AND INTERFERENCE DEVIATES IDEAL OPTICS INTO "REAL" OPTICS

POINT SPREAD FUNCTION (PSF) ⇒ IMAGE OF AN IDEAL POINT

$$FWHM \sim \frac{\lambda}{2} \quad \leftarrow \text{LIGHT WAVELENGTH}$$

IN REAL IMAGING WHAT WE SEE IS THE CONVOLUTION OF THE OBJECT WITH THE PSF ⇒ OBJECT LOOKS "FUZZY"

CONVOLUTION THEOREM: THE CONVOLUTION IN SPATIAL DOMAIN IS EQUIVALENT TO THE MULTIPLICATION IN THE FREQUENCY DOMAIN (FOURIER SPACE)

OTF \Rightarrow OPTICAL TRANSFER FUNCTION

\hookrightarrow LOSS OF HIGH FREQUENCY COMPONENTS

HOW TO FIX THIS PROBLEM??

\rightarrow DECONVOLUTION

$$\hat{I}(\vec{k}) = \tilde{O}(\vec{k}) \cdot \text{OTF}(\vec{k})$$

DECONVOLUTION

$$\downarrow$$

$$\tilde{O}(\vec{k}) = \hat{I}(\vec{k}) \cdot \text{OTF}(\vec{k})^{-1}$$

\mathcal{F} = FOURIER TRANSFORM

\mathcal{F}^{-1} = INVERSE \mathcal{F}

$$\downarrow$$

$$O(\vec{r}) = \mathcal{F}^{-1}[\tilde{O}(\vec{k})]$$

CONFOCAL MICROSCOPY

ONLY THE LIGHT IN THE FOCAL PLANE IS ACQUIRED, SO YOU CAN HAVE DEPTH DISCRIMINATION AND GENERATE 3D IMAGES.

MULTIPHOTON MICROSCOPY

- ONLY IMAGES AT HIGH PHOTON FLUX \Rightarrow FOCAL POINT

\hookrightarrow 3D RESOLUTION

- USE IR \Rightarrow LESS DAMAGING TO BIOLOGICAL SPECIMENS

- LESS PHOTO TOXICITY

5. MICROPIPETTE ASPIRATION & MICROFLUIDICS

DR. C.T. LIM (NATIONAL UNIVERSITY OF SINGAPORE)

MALARIA \Rightarrow STIFFENS THE CELL

USE μ -PIPETTE AND μ -FLUIDICS TO STUDY MECHANICAL PROPERTIES OF INFECTED RED BLOOD CELLS (RBC)

DUAL PIPETTE ASSAY \Rightarrow APPLY LOW FORCES TO SEPARATE

ADHERENT CELLS OR PROTEIN COATED LIPOSOMES

MICROFLUIDICS \Rightarrow SMALL SAMPLE VOLUME

LOOK AT CELLS DEFORMING WHILE PASSING THROUGH A NARROW CHANNEL ($\sim 2\mu\text{m}$)

6. MEMS BASED TOOLS -

LAST TWO LECTURES MISSING!!