

# Applied Single-Molecule Biophysics Group



## T-COSSA LAB

Center for Interdisciplinary Nanophysics, Department of Physics, University of Ottawa

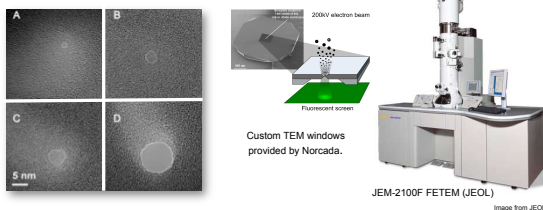
### I. Mission Statement

Our research is dedicated to the development of novel techniques and methods to manipulate and characterize single-molecules using nanofluidic devices, to unravel the basic physics governing the behaviour of biological molecules in nanoconfined geometries, and ultimately to translate these discoveries into new tools for the life sciences.

While our research efforts are driven by the exploration of unique detection and actuation modalities, the manipulation and characterization of individual molecules within nanoscale fluidic environments (i) represents an important and largely uncharted regime in which interesting new physics can dominate and (ii) enables new quantitative measurements, with exceptional sensitivity and speed, on the structure and behavior of biological molecules.

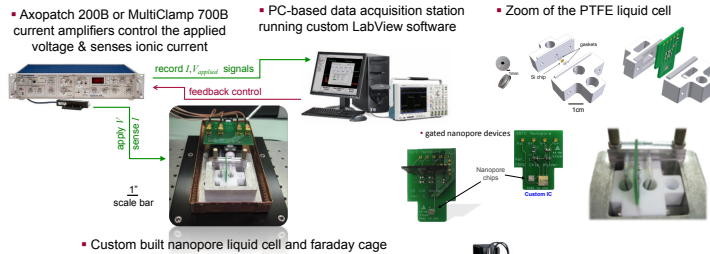
### II. Solid-State Nanopores

We fabricate pores with single-nanometre precision by exposing a 30 nm thick free-standing SiNx membrane, to a tightly focused high-energy electron beam with the help of a 200 kV field emission TEM. Prolonged irradiation of the SiNx membrane with an electron beam focused to a spot of a few nanometres leads to the formation of a hole via sputtering.



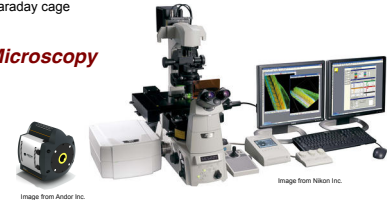
### III. Tools and Measurement Techniques

#### Low-noise ionic current recording



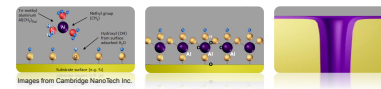
#### Single-Molecule Fluorescence Microscopy

Fluorescently labeled molecules can be tracked at high speed in nanofluidic channels and nanopores by employing a Nikon Ti-E microscope equipped with a high magnification objective - 60x Oil 1.27 NA, and 100x Oil 1.45NA - and Andor iXon3 eMCCD camera.



#### Atomic Layer Deposition (ALD) System

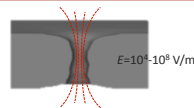
An Atomic Layer Deposition (ALD) system is used to fabricate multilayered membranes and fine tune pore sizes and surface properties with atomic control.



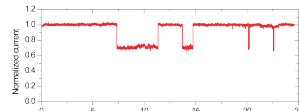
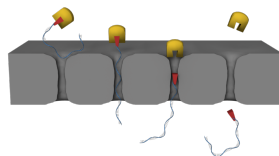
### IV. Research Projects

#### Nanopore Force Spectroscopy

Characterization of intermolecular interactions is critical to understand the molecular processes that govern biology. Nanopores offers a high-throughput approach to Single-Molecule Force Spectroscopy, with applications in areas such as pharmaceutical research (screening of drug-receptor interactions), and clinical diagnostics (identification of cancer markers via antibody-ligand interactions, or genotyping).



- Use electric field to pull on charged molecules. Force range ~ 0.1 - 10^2 pN
- No intricate single-molecule manipulations
- Automated measurements
- No Surface Chemistry

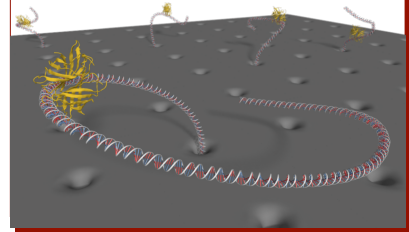


The electric field inside the nanopore is used to apply a force on an individual bond. The ionic current is monitored to measure the distribution of dissociation times of the molecular complex.

Following Kramer's theory, we analyze the relationship between dissociation timescale and applied potential:

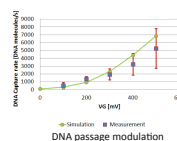
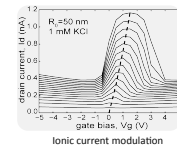
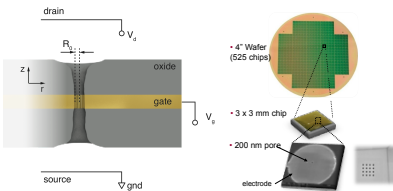
$$\langle \tau \rangle = \tau_0 \exp \left[ \frac{E_{\text{bind}} - \text{Force}(V) \Delta x}{k_B T} \right]$$

Proteins bound to dsDNA molecules. Each molecular complex is threaded through a sub-5 nm solid-state nanopore to unravel DNA-protein interactions or the local protein structure for gene regulation studies.

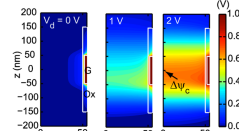


#### NanoFluidic Transistor

We use field-effect gated nanopore devices to study long-range de-screening effects under high electric fields. When immersed in a solution, the applied bias at the gate is used to modulate the passage of DNA molecules flowing through the channel (hence the transistor analogy).



A molecule's ionic screening cloud can be distorted due to the relaxation effect under high electrical field and strong electro-diffusion.



The critical field strength is:  $E_c \sim k_B T / q \lambda_D \sim 10^7 \text{ V/m}$  or  $10 \text{ mV/nm}$  (for physiological conditions)  
Long-range electrostatic interactions  $> 10^2$ 's of Debye length

#### Nanopore Publications:

Jetha N.; Feehan C.; Wiggan M.; Tabard-Cossa V.; and Marziali A.; Biophysical Journal 100, 2974-2980 (2011)  
Liu Y.; Huber D.; Tabard-Cossa V.; Dutton R. W.; Applied Physics Letters 97, 143109 (2010)  
Tabard-Cossa V.; Wiggan M.; Trivedi D.; Jetha N.; Dwyer J. R.; Marziali A.; ACS Nano 3 (10), 3009-3014 (2009)  
Branton D.; Deamer W. D.; Marziali A.; Tabard-Cossa V. et al.; Nature Biotechnology 26, 1146-1153 (2008)  
Wiggan M.; Tropini C.; Tabard-Cossa V.; Jetha N.; and Marziali A.; Biophysical Journal 95, S317-S323 (2008)  
Tabard-Cossa V.; Trivedi D.; Wiggan M.; Jetha N.; Marziali A.; Nanotechnology 18, 305505 (2007)

#### Collaborators:

- uOttawa (Michel Godin)
- UBC (Andre Marziali)
- Stanford (Roger Howe, Bob Dutton) & Stanford Genome Technology Center (Ron Davis)

#### Funding:

