UNIVERSITÉ LAVAL

Optical Detection of DNA Based on Cationic Polythiophenes

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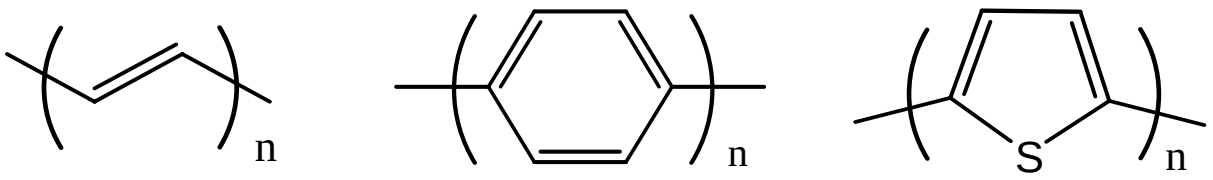
Département de Chimie

Chaire de Recherche du Canada

Polymères Électroactifs et Photoactifs

CONJUGATED POLYMERS



- **ELECTRICAL PROPERTIES**

Semiconductors to conductors

- **OPTICAL PROPERTIES**

Absorption and emission in the UV-visible range

- **STABILITY, MECHANICAL PROPERTIES**

APPLICATIONS

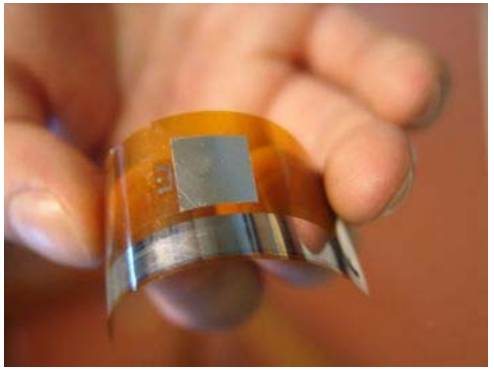
Conductors

Electroluminescent diodes

Transistors

Photovoltaic cells

Sensors



POLY(2,7-CARBAZOLE)S

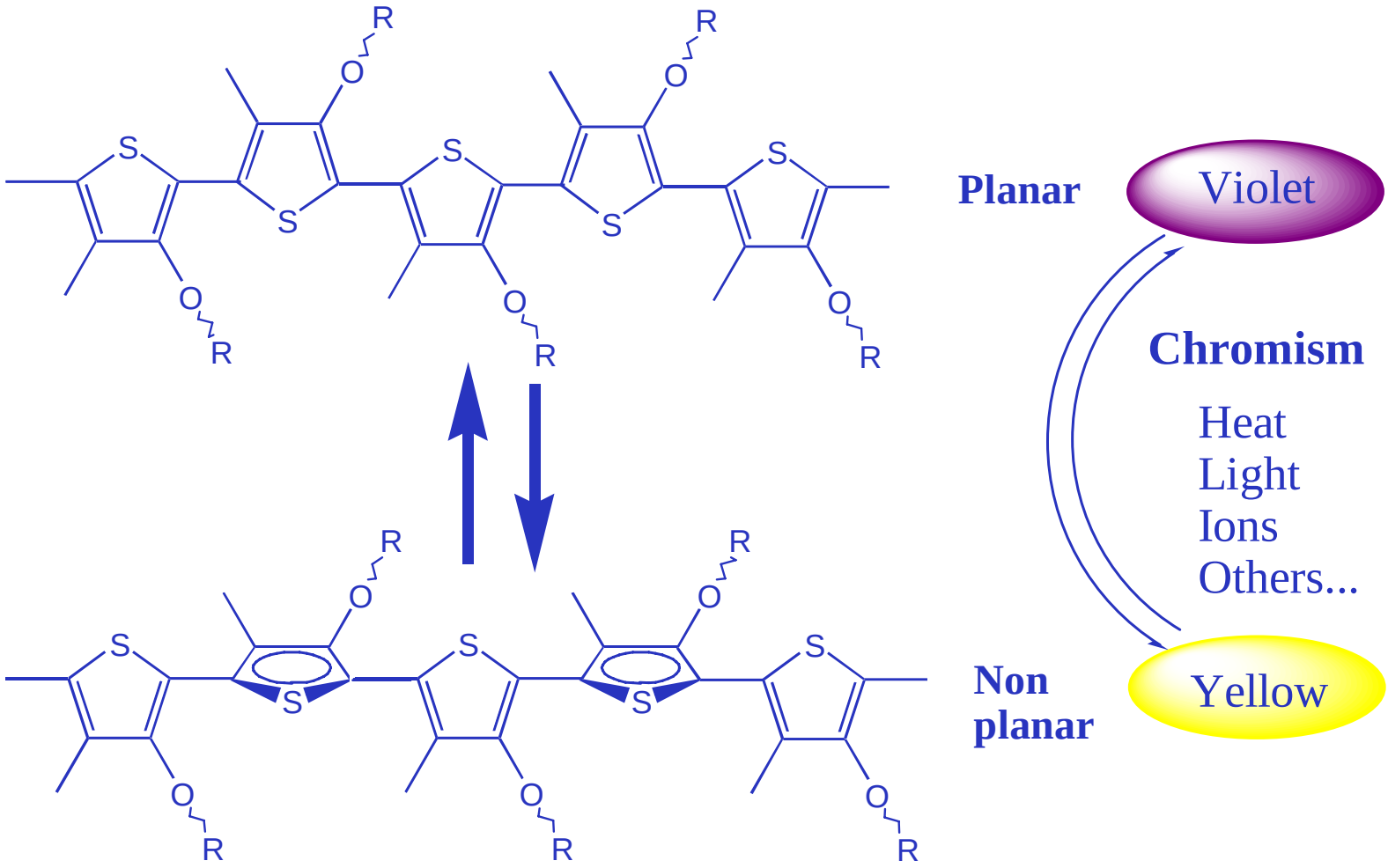
Co-Monomers

Polymers

410 nm
(3.0 eV)

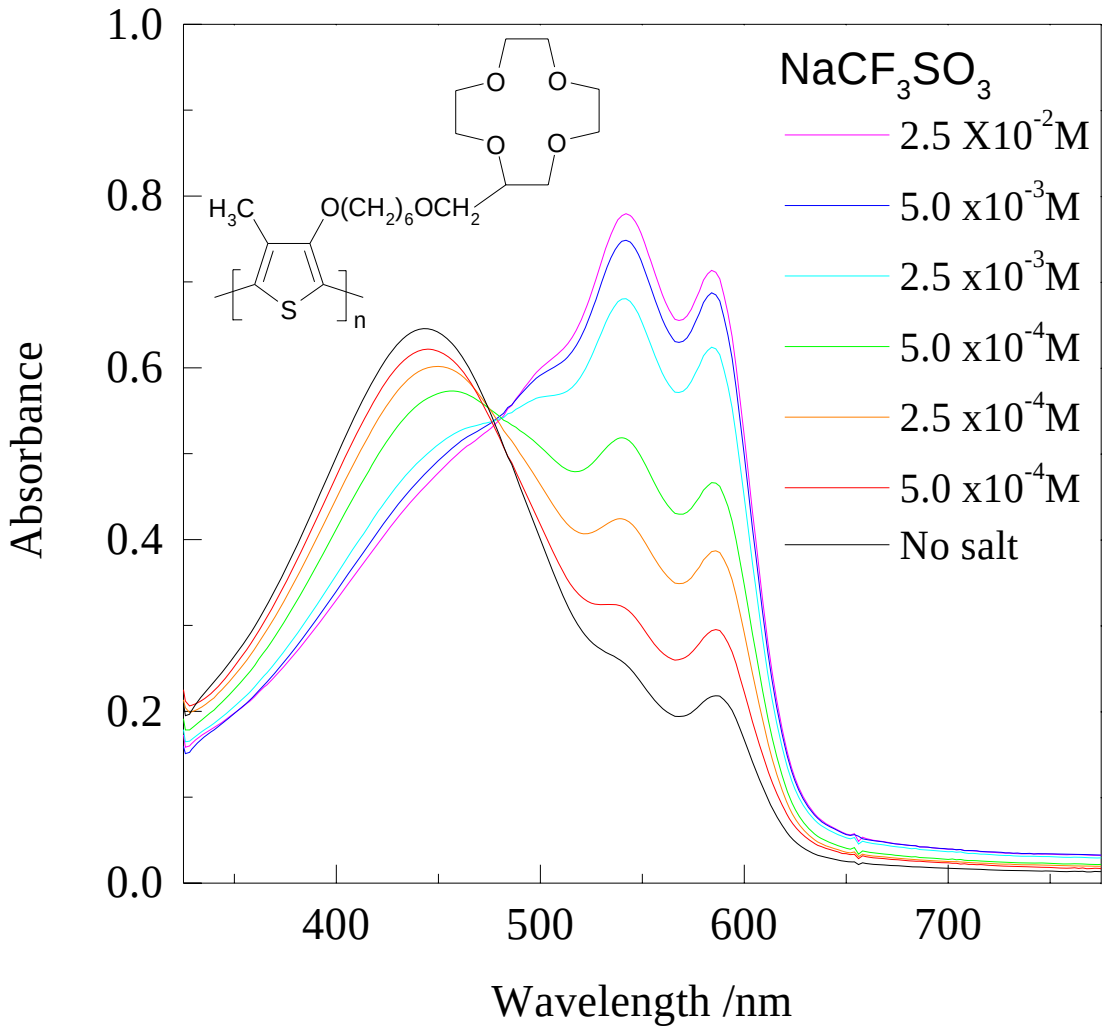
1100 nm
(1.1 eV)

OPTICAL PROPERTIES

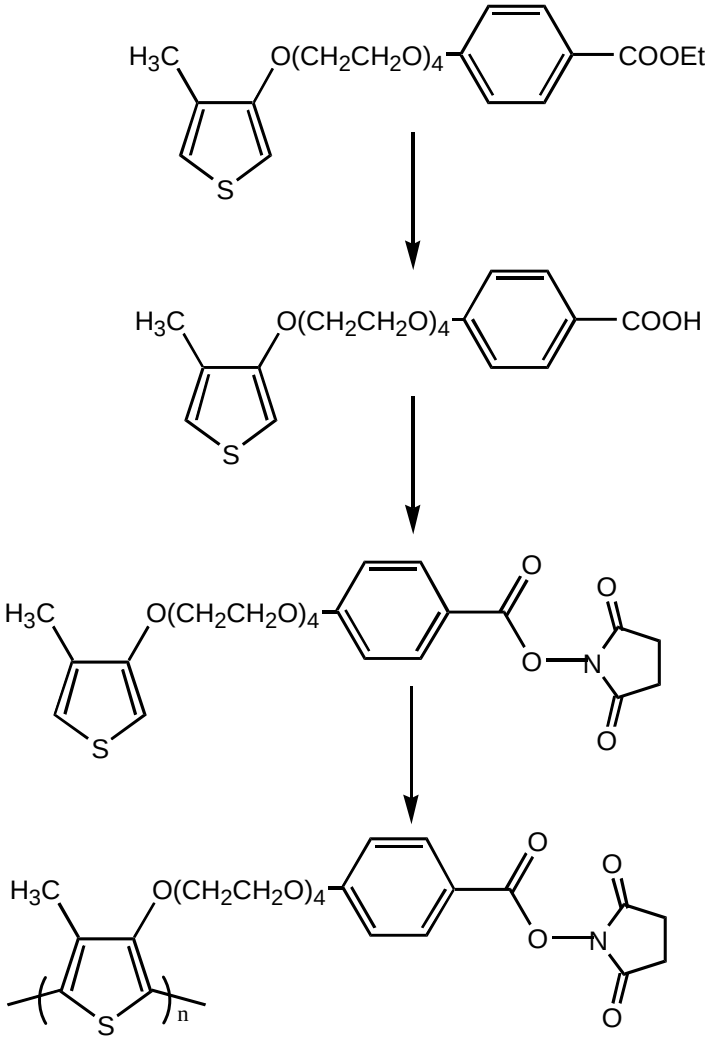


Adv. Mater., 11, 1491 (1999)

IONOCHROMISM

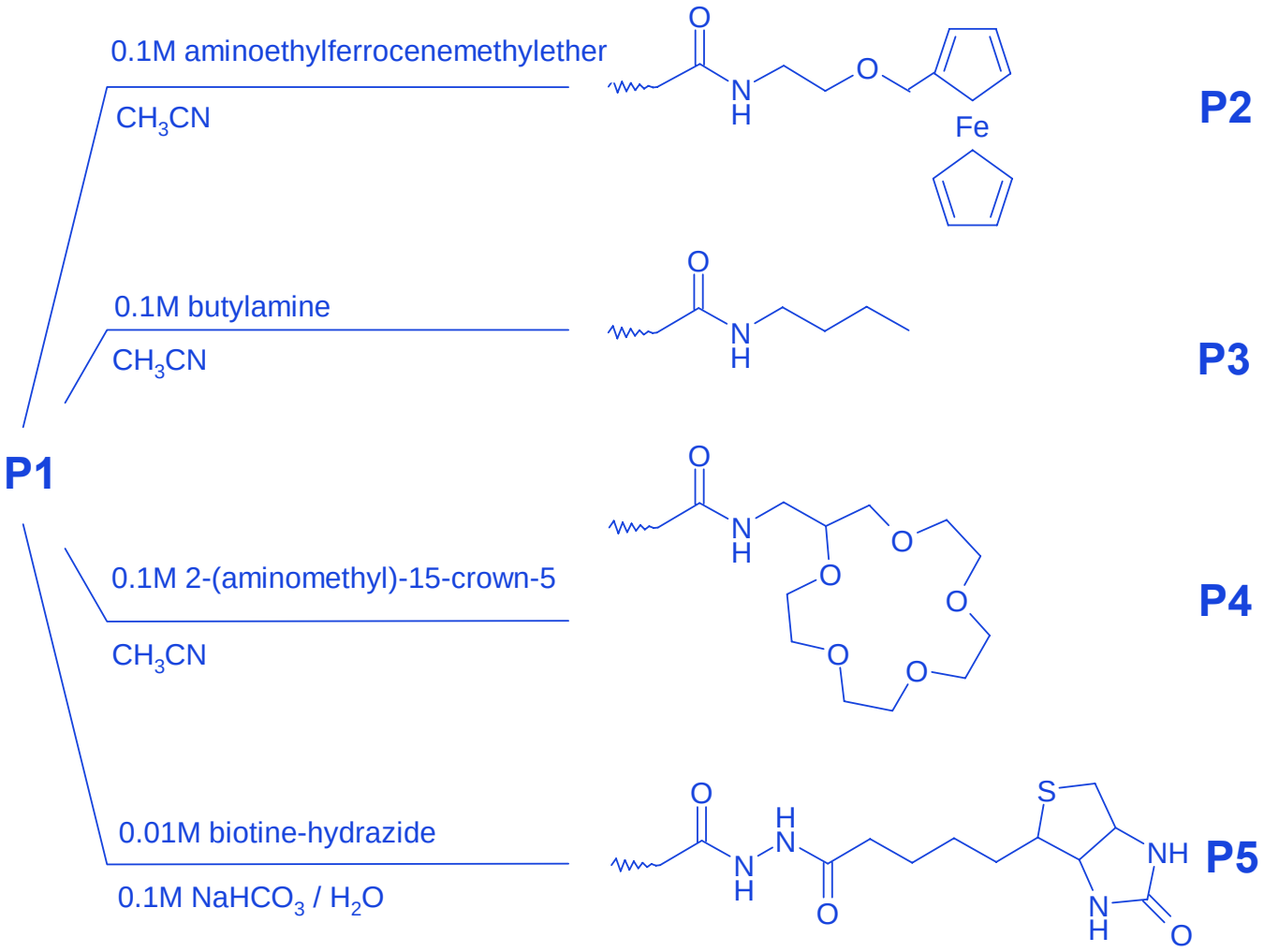


PRE-POLYMER APPROACH

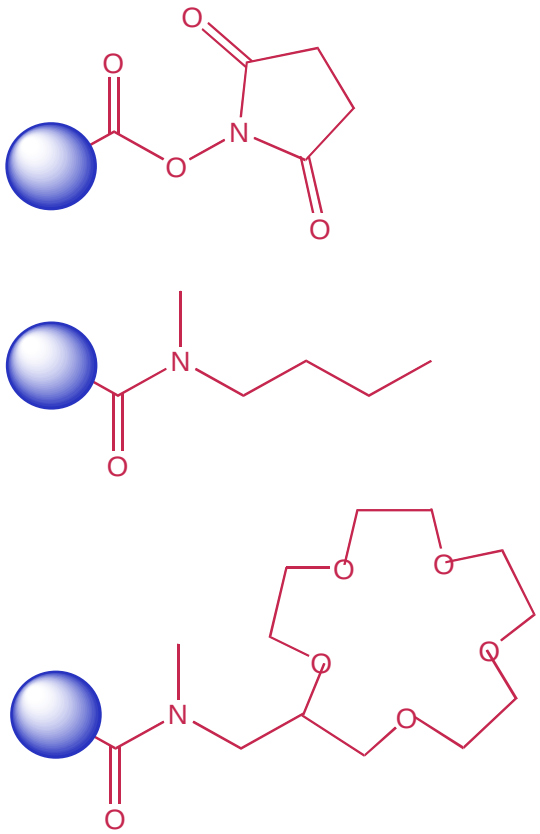


P1

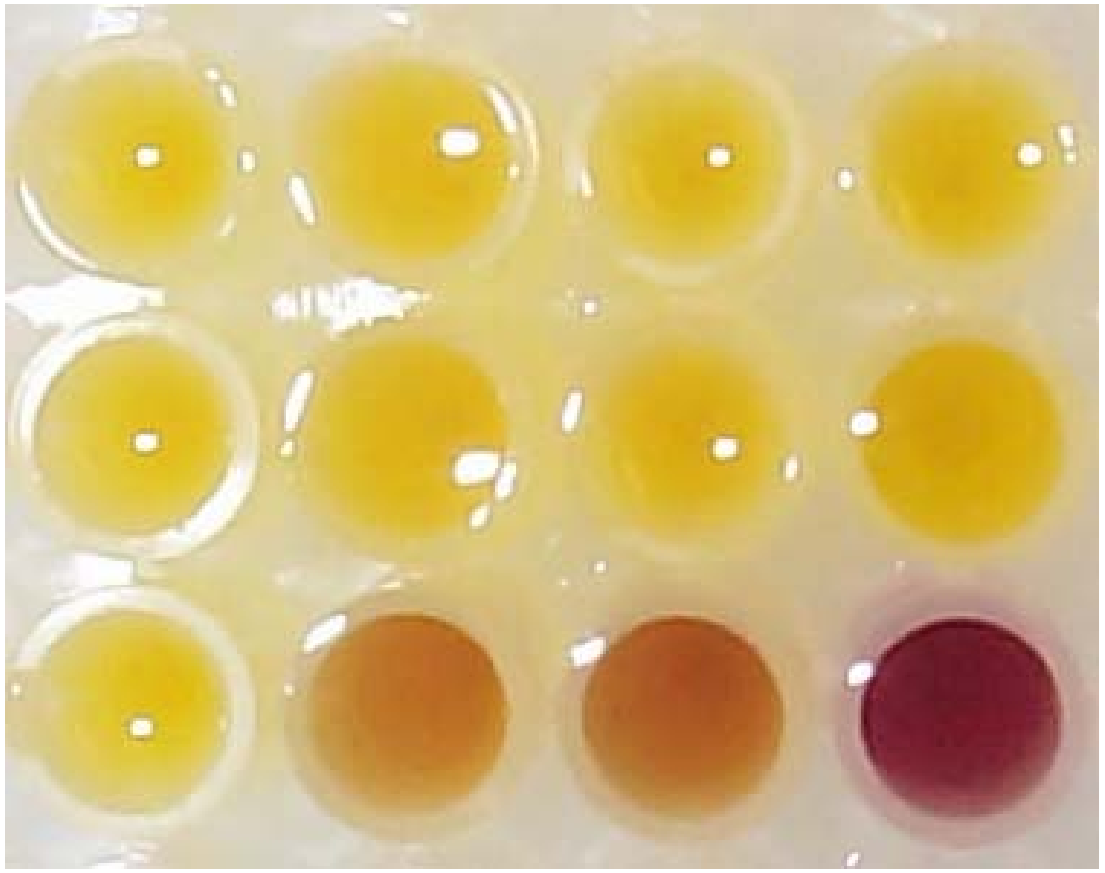
Post-Functionalization



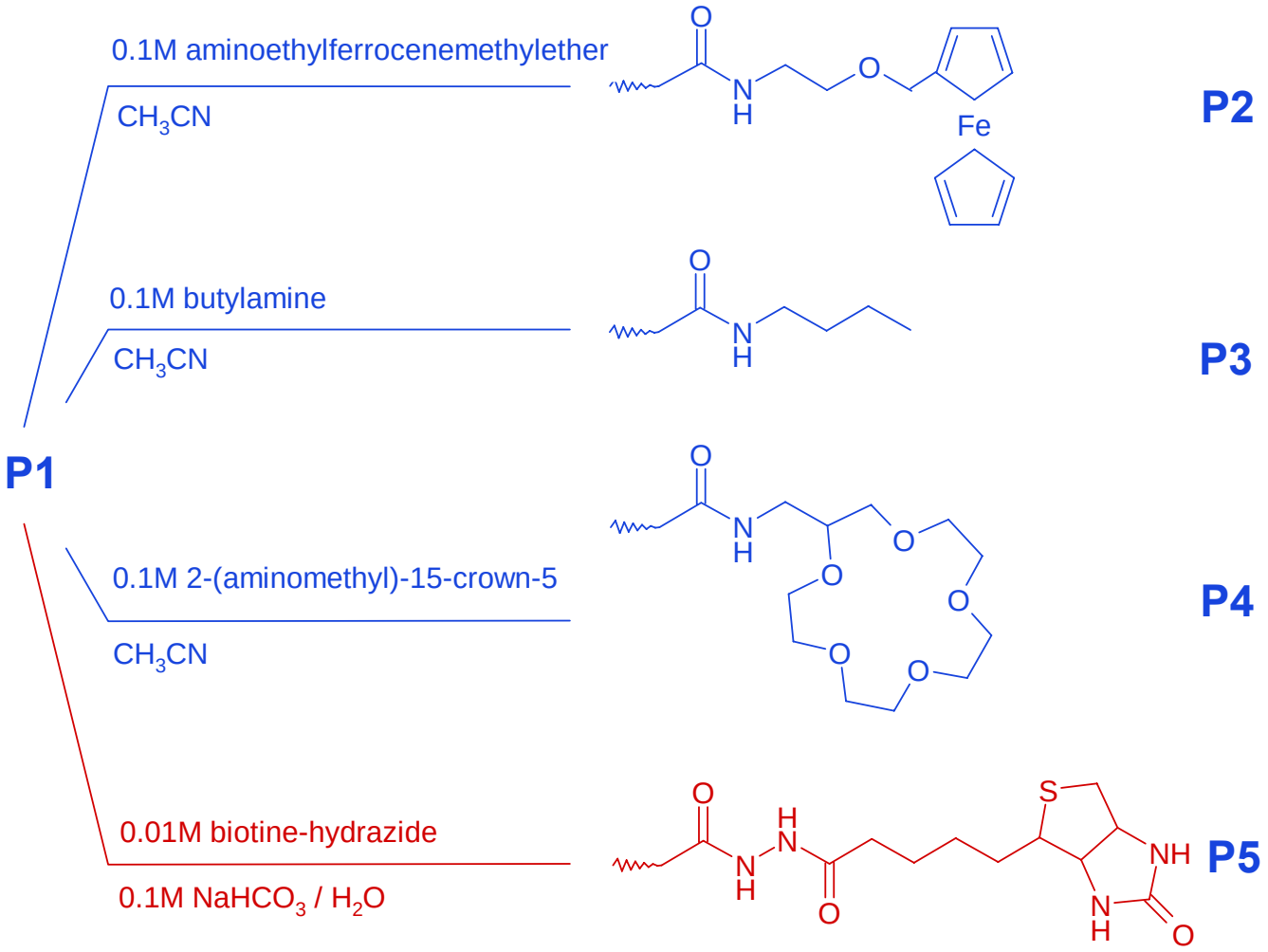
IONOCHROMISM



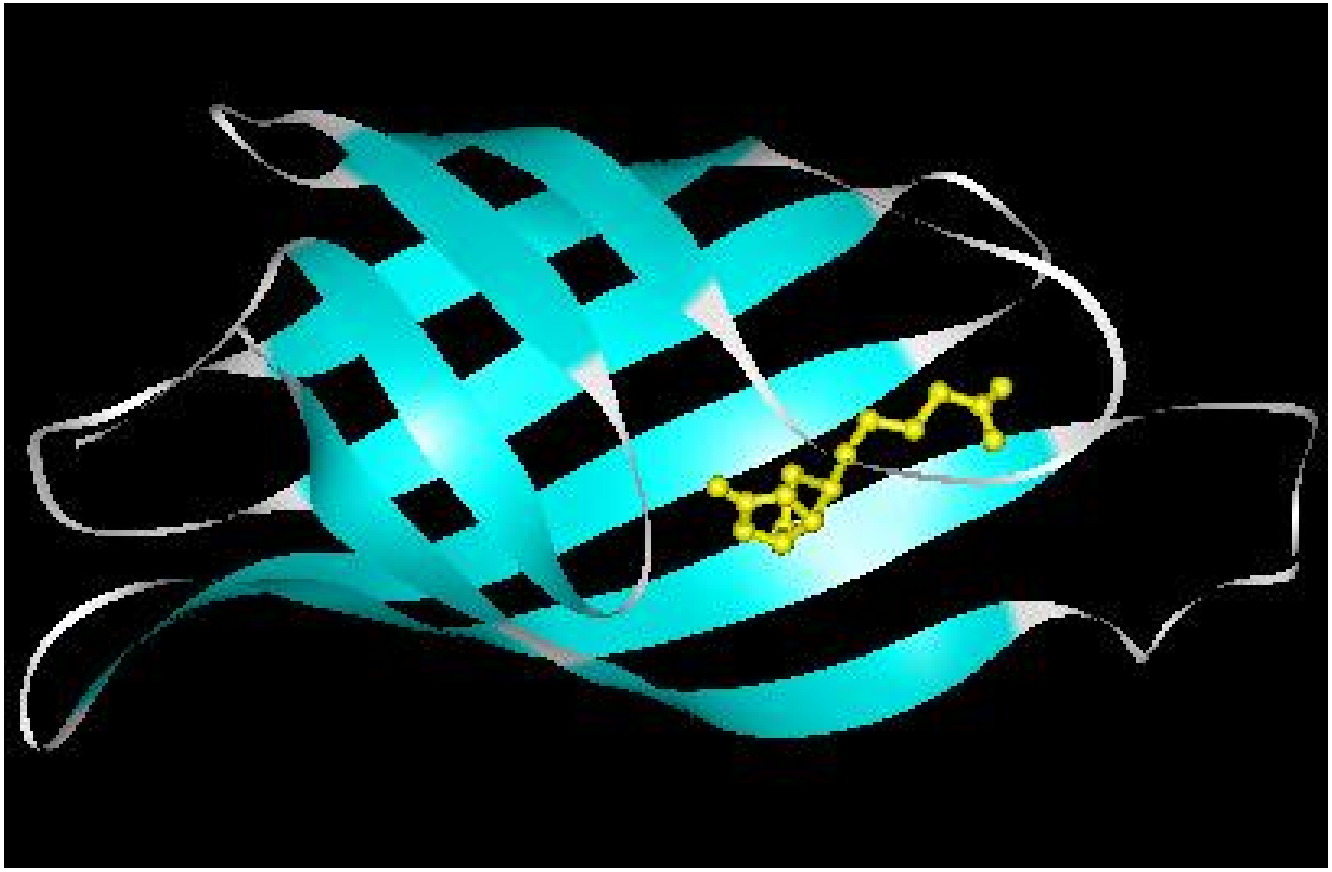
Solvent $LiCF_3SO_3$ $NaCF_3SO_3$ KCF_3SO_3



Post-Functionalization

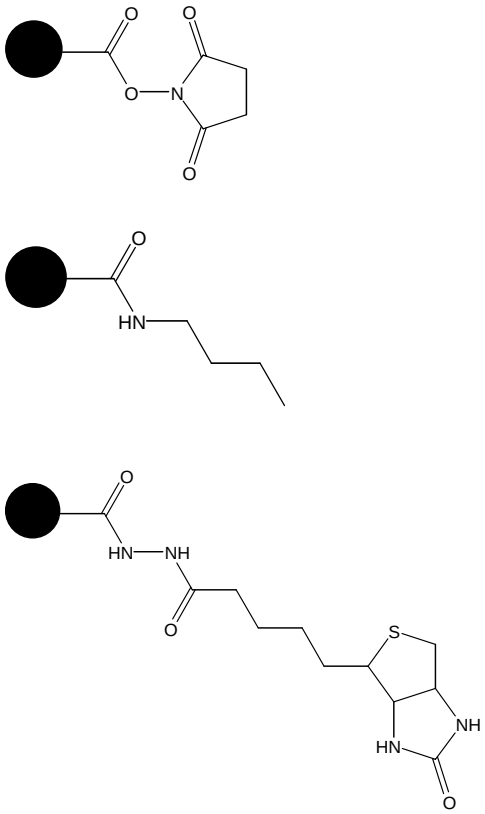


BIOCHROMISM



$K_a \sim 10^{15}\ M^{-1}$ Avidin 65,500 Da

BIOCHROMISM

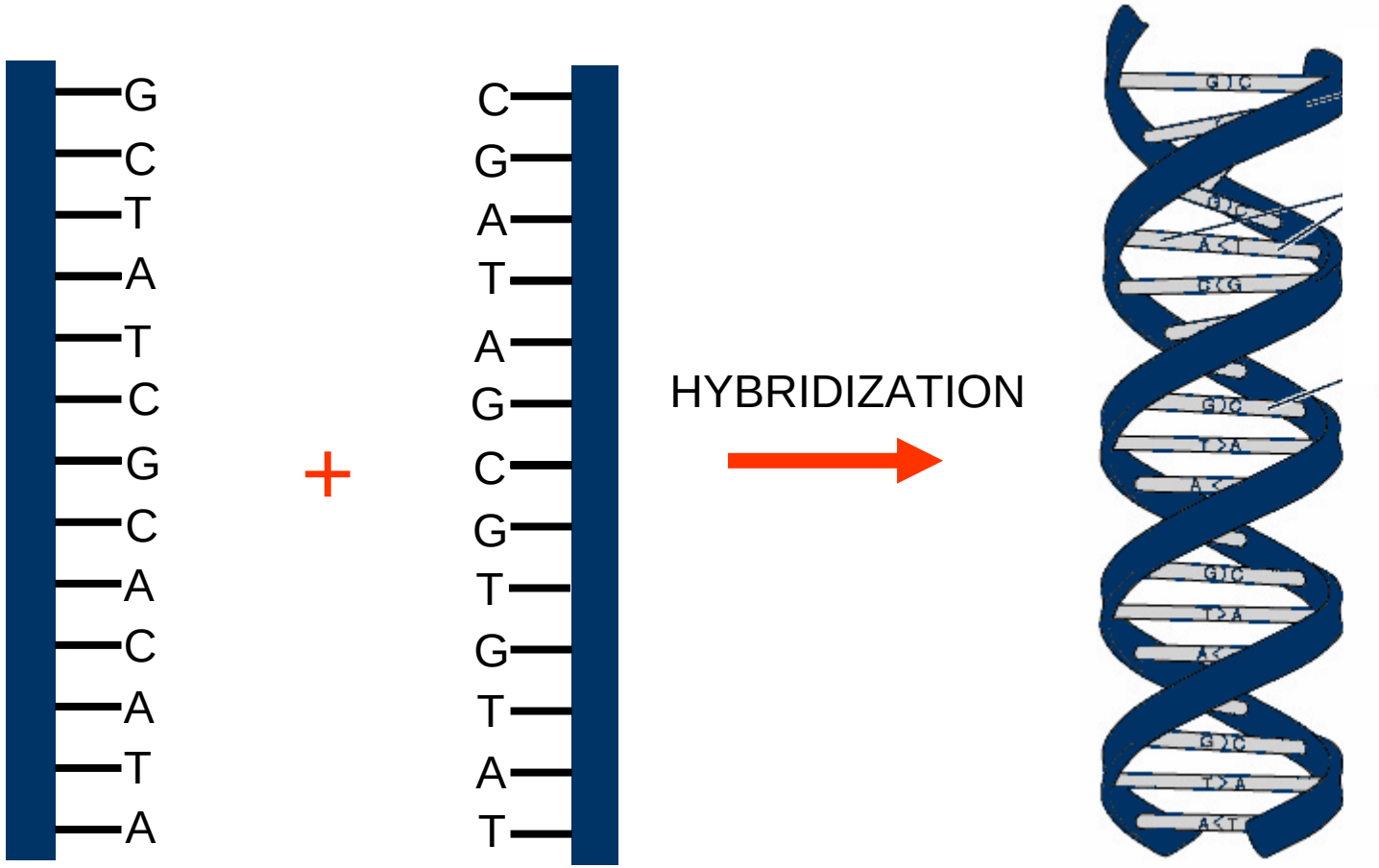


Solvent +Avidin

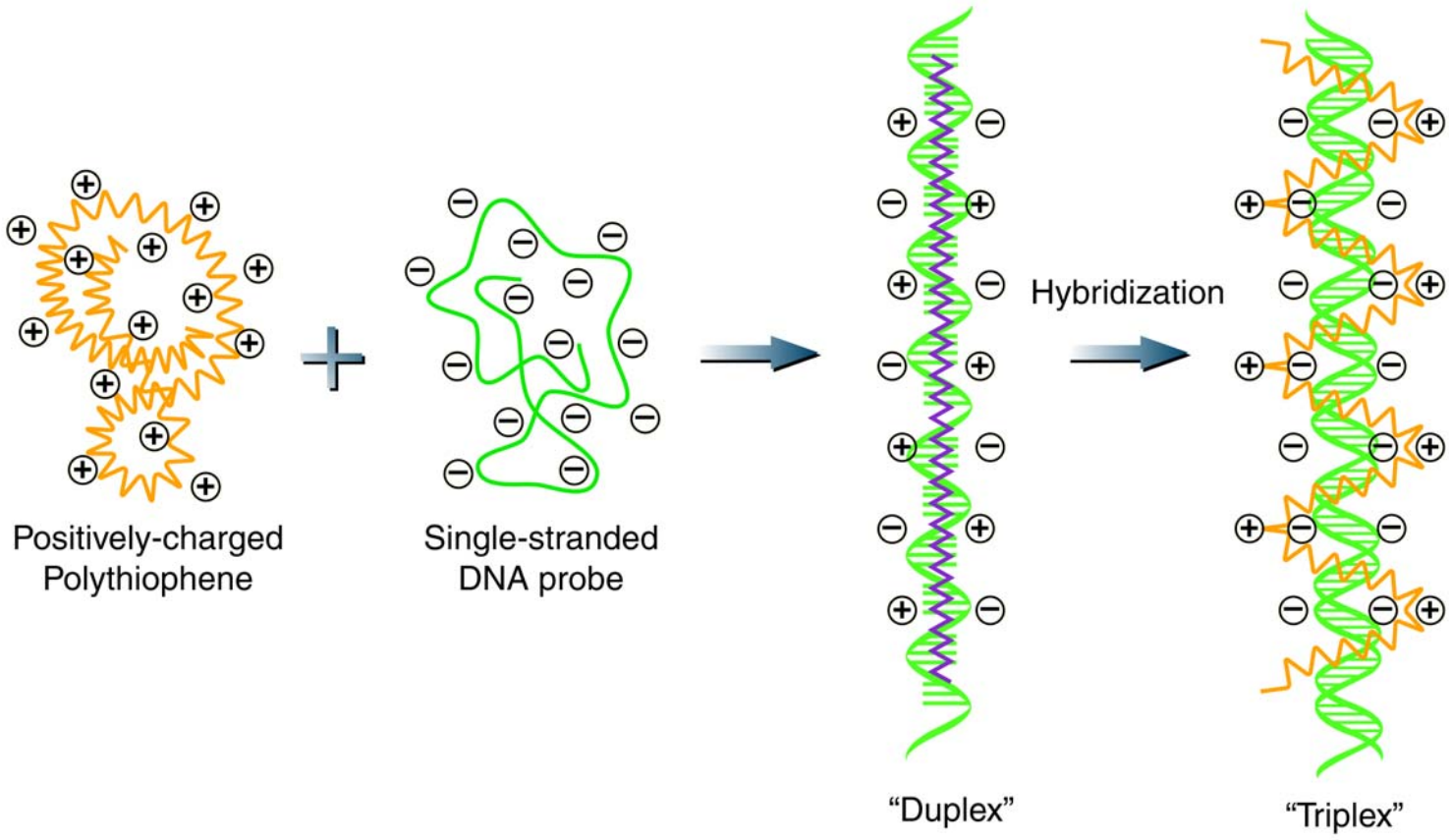
S. Bernier, S. Garreau, M. Béra-Abérem, M. Leclerc, JACS, 124, 12463 (2002)

OLIGONUCLEOTIDES

Probe : Sequence of nucleic acids able to recognize a complementary strand



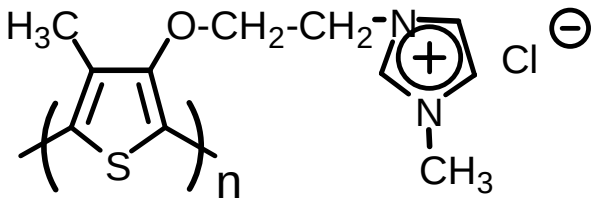
DNA-CHROMISM

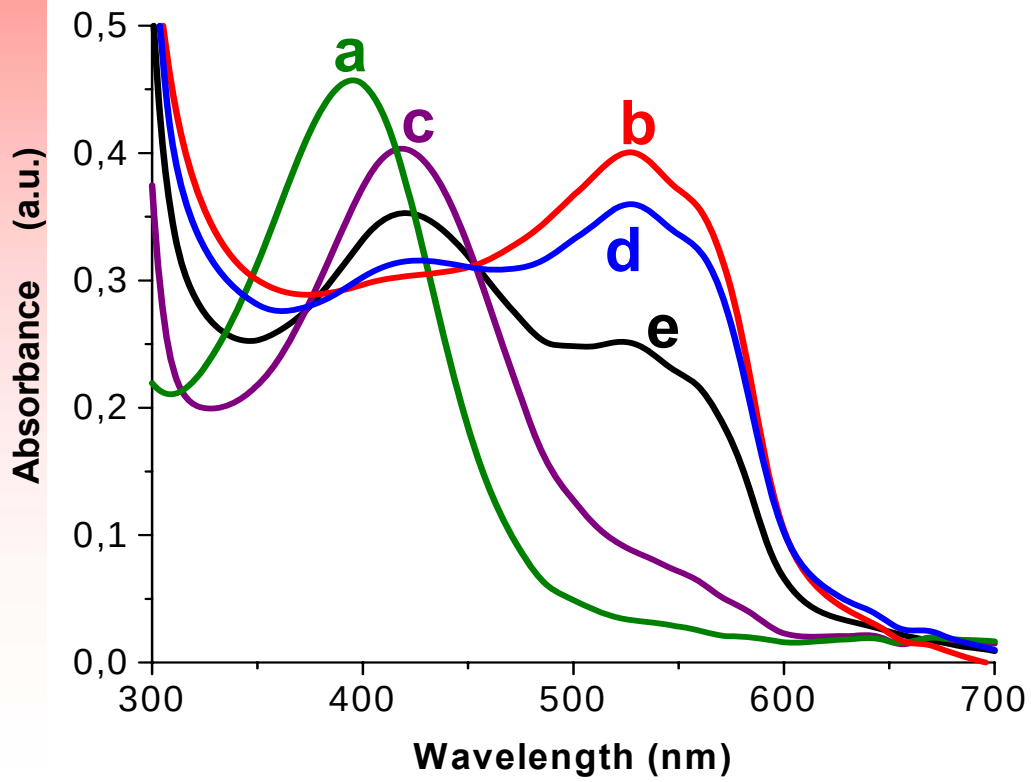


Acc. Chem. Res., 41, 168 (2008)

COLORIMETRY

a- Poly at 55°C
b- Poly / X1
c- Poly /X1 /Y1
d- Poly /X1 /Y2
e- Poly /X1 / Y3





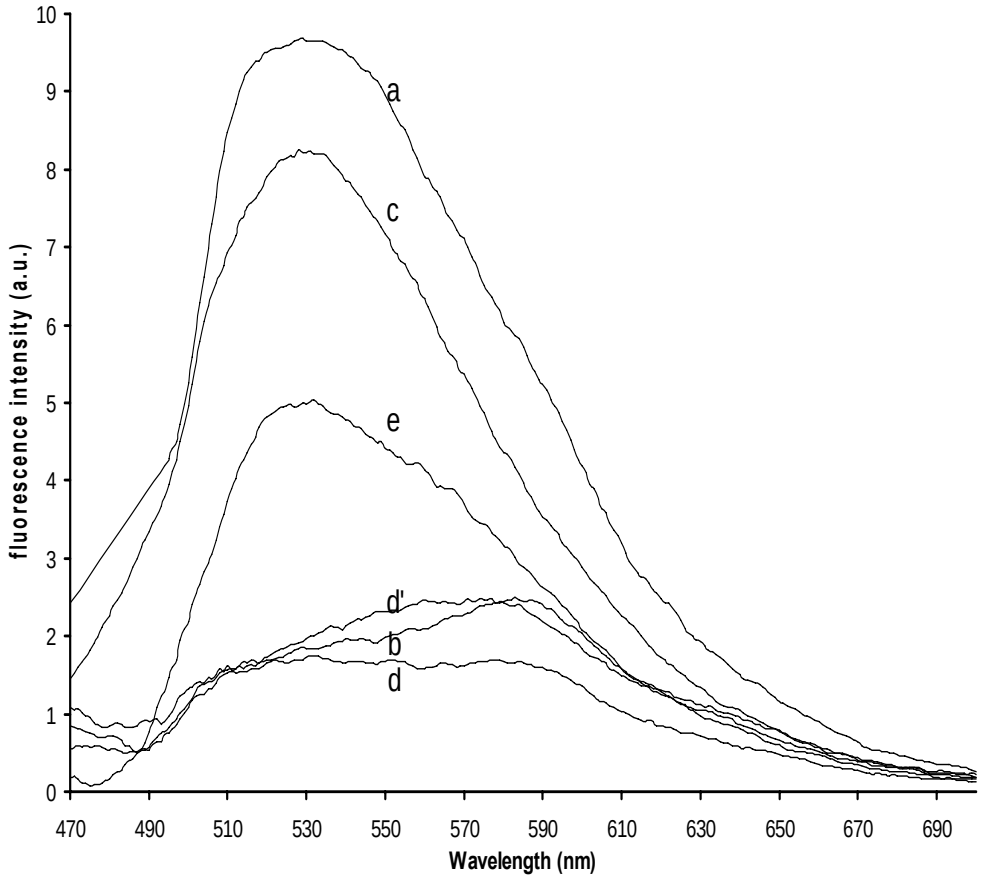
Specific oligonucleotide of *Candida Albicans*
X1 5' CATGATTGAACCATCCACCA 3'
Y1 3' GTACTAACTTGGTAGGTGGT 5'

Specific oligonucleotide of *Candida Dubliniensis*
X2 5' CATGATTGAAGCTTCCACCA 3'
Y2 3' GTACTAACTTCGAAGGTGGT 5'
Y3 3' GTACTAACTTCGTAGGTGGT 5'

FLUORESCENCE

Violet → Yellow

Non- fluorescent → fluorescent



a) Poly

b) Poly / X1

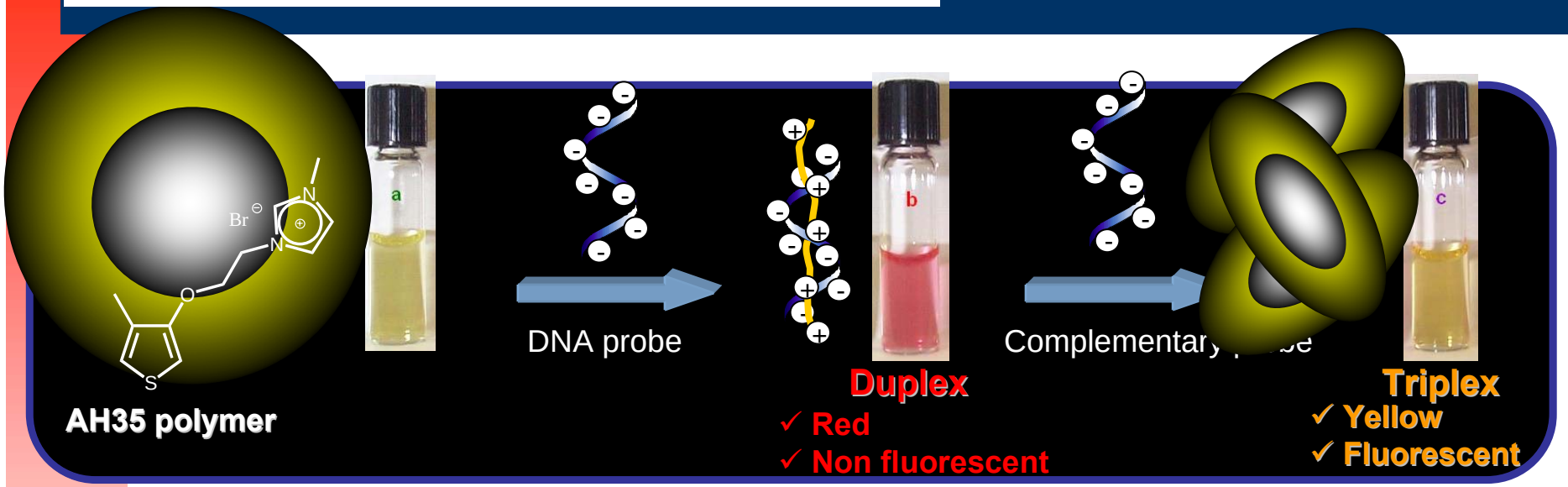
c) Poly / X1 / Y1

d) Poly / X1 /Y2

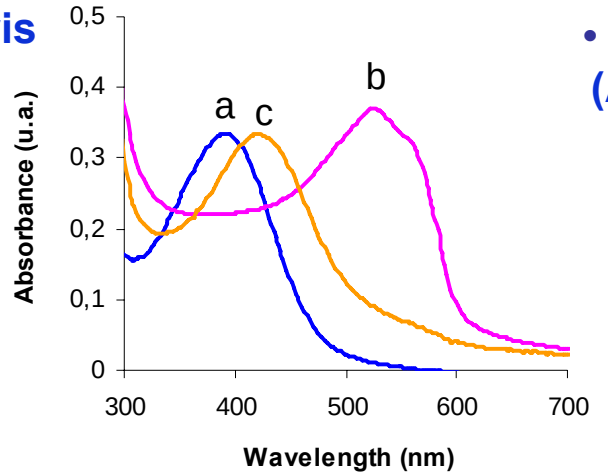
d') Poly / X1 /100Y2

e) Poly / X1 / Y3

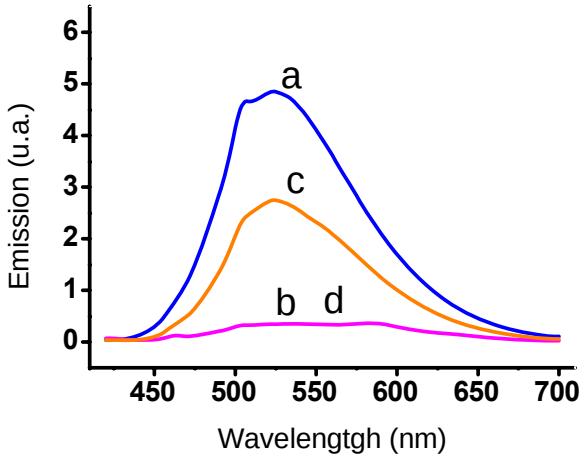
“Classical” detection method



• **UV-vis**

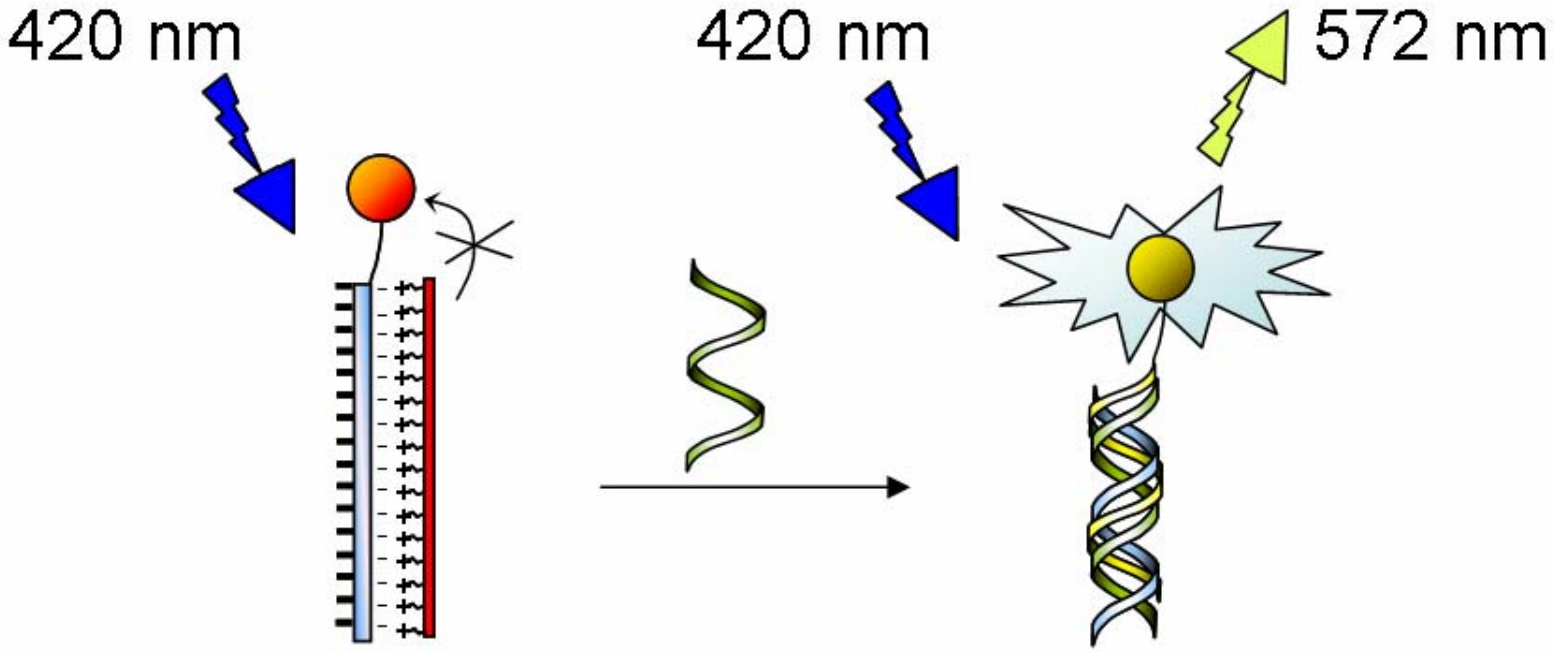


• **Fluorescence (λ_{exc} = 400 nm)**



Ho, H. A. et al *Angew.Chem., Int. Ed.* **2002**, 41, 1548.

FRET



PCR-Free Detection of Anthrax

CENTRE DE RECHERCHE EN INFECTIOLOGIE

DEFENCE R&D DÉFENSE

Microbiology
DNA Preparation

CERMA centre de recherche sur les matériaux avancés

Polythiophene-Based
Biosensor Detection

National Research Council Canada

Microfluidic Cartridges
Nanofabrication

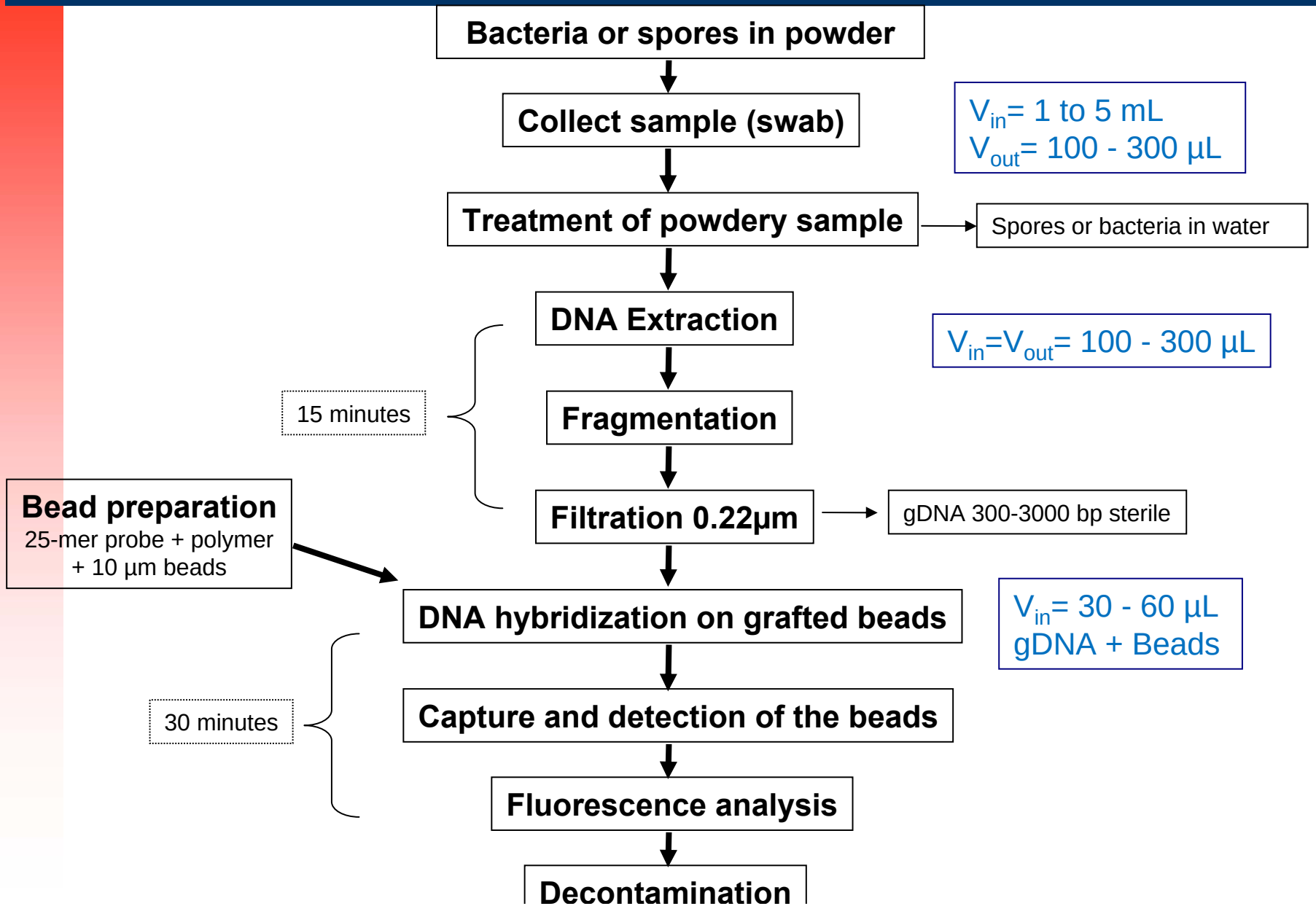
COPL Centre d'optique, photonique et laser

Optics
Engineering

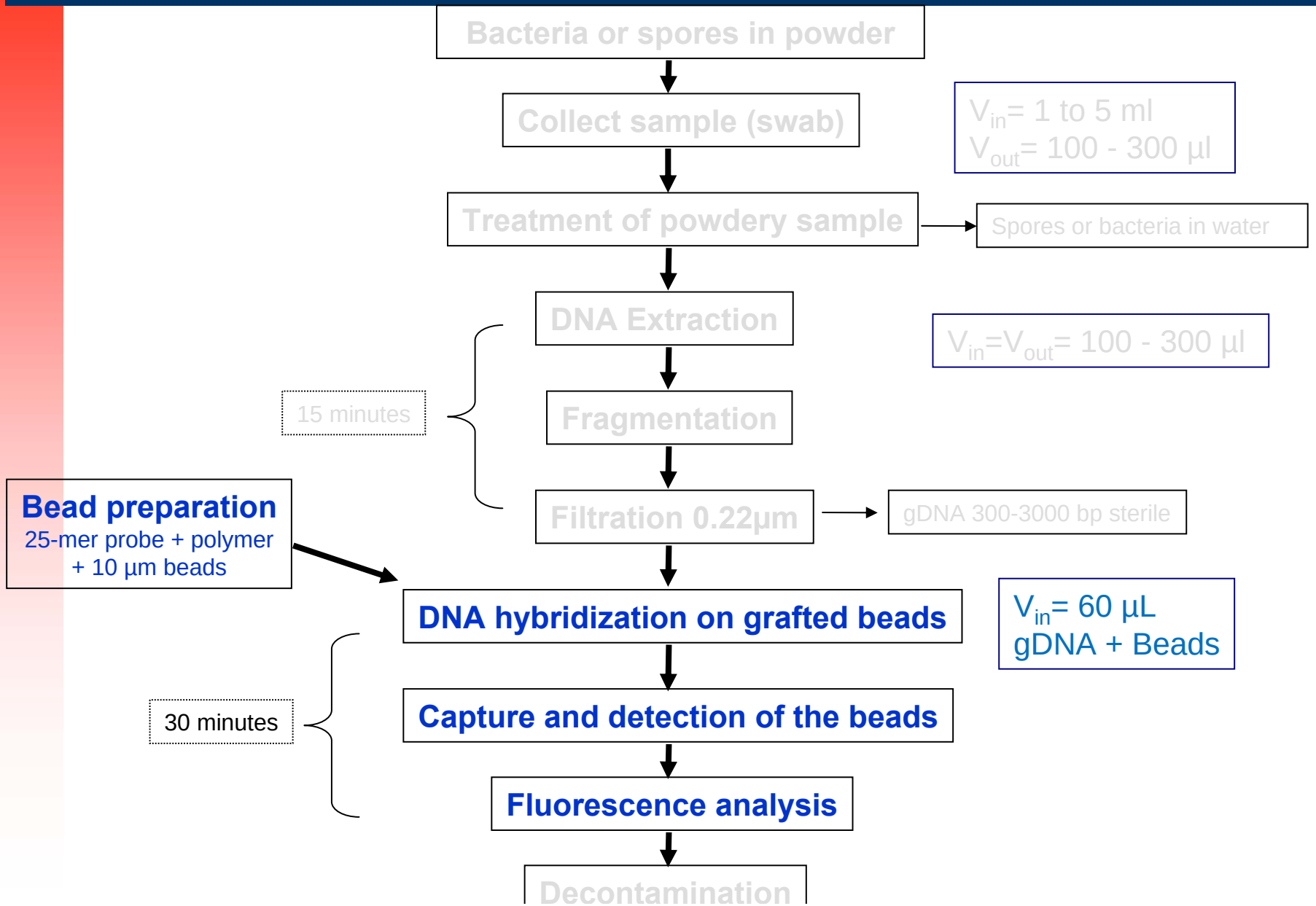
Gendarmerie royale du Canada | Royal Canadian Mounted Police

First Responders

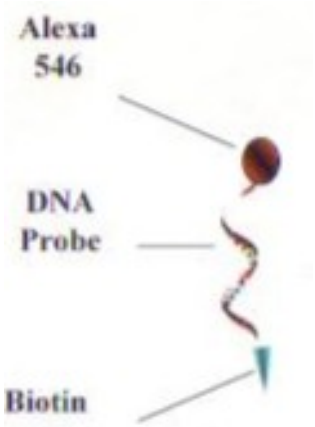
General Scheme



General Scheme



Bead Preparation

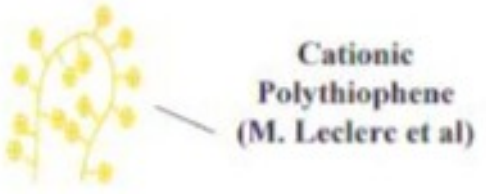


+

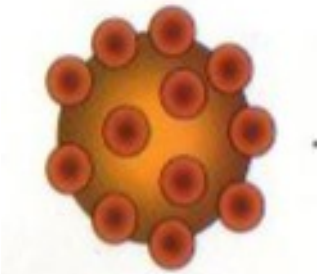
10-micron beads with Streptavidin coating

→ **Probe-Grafted Beads**

Probe-Grafted Beads +

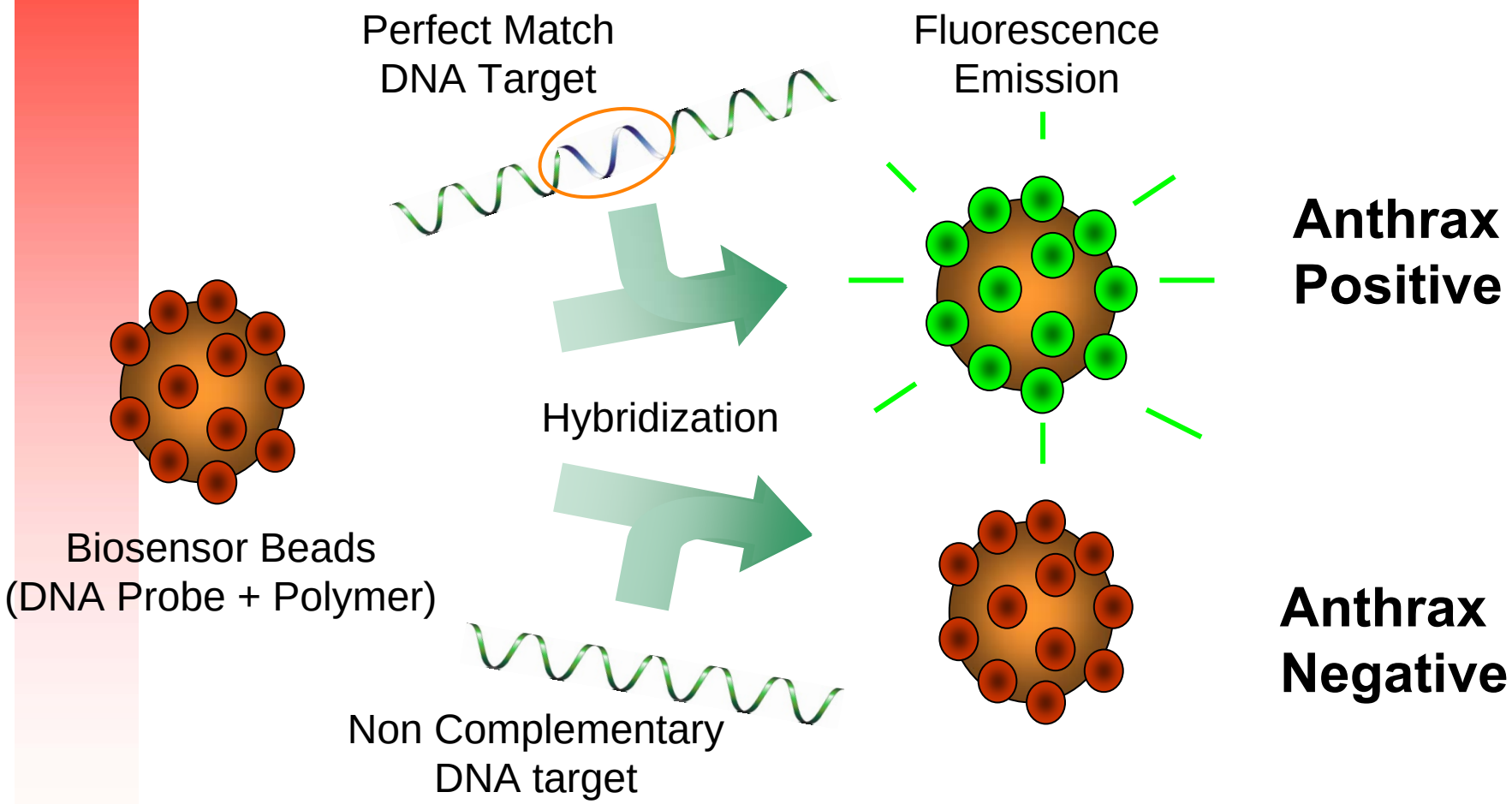


→

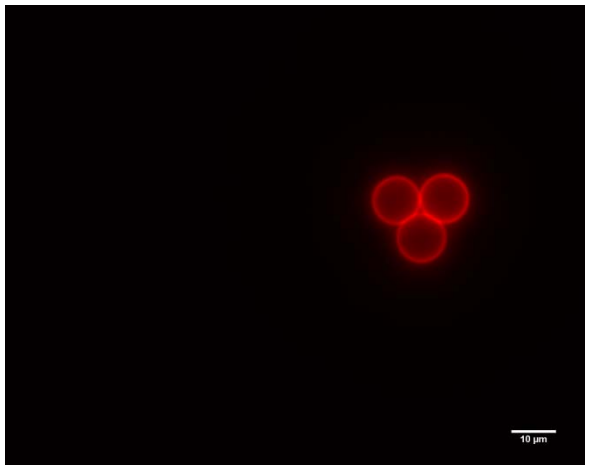


Biosensor Beads (DNA Probe + Polymer)

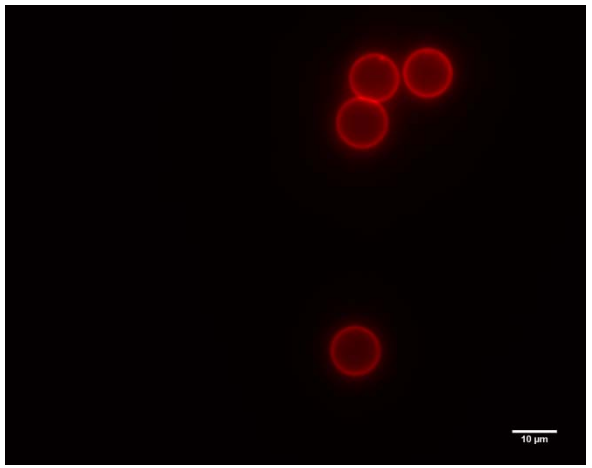
DNA Target Recognition with Biosensor Beads

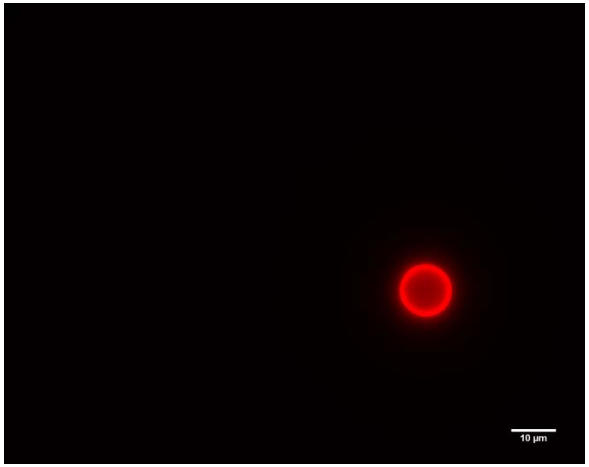


Fluorescence Microscope Imaging
Duplex versus Triplex



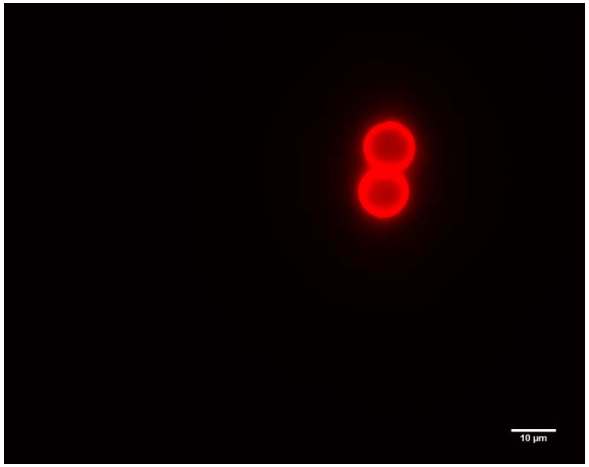
Duplex



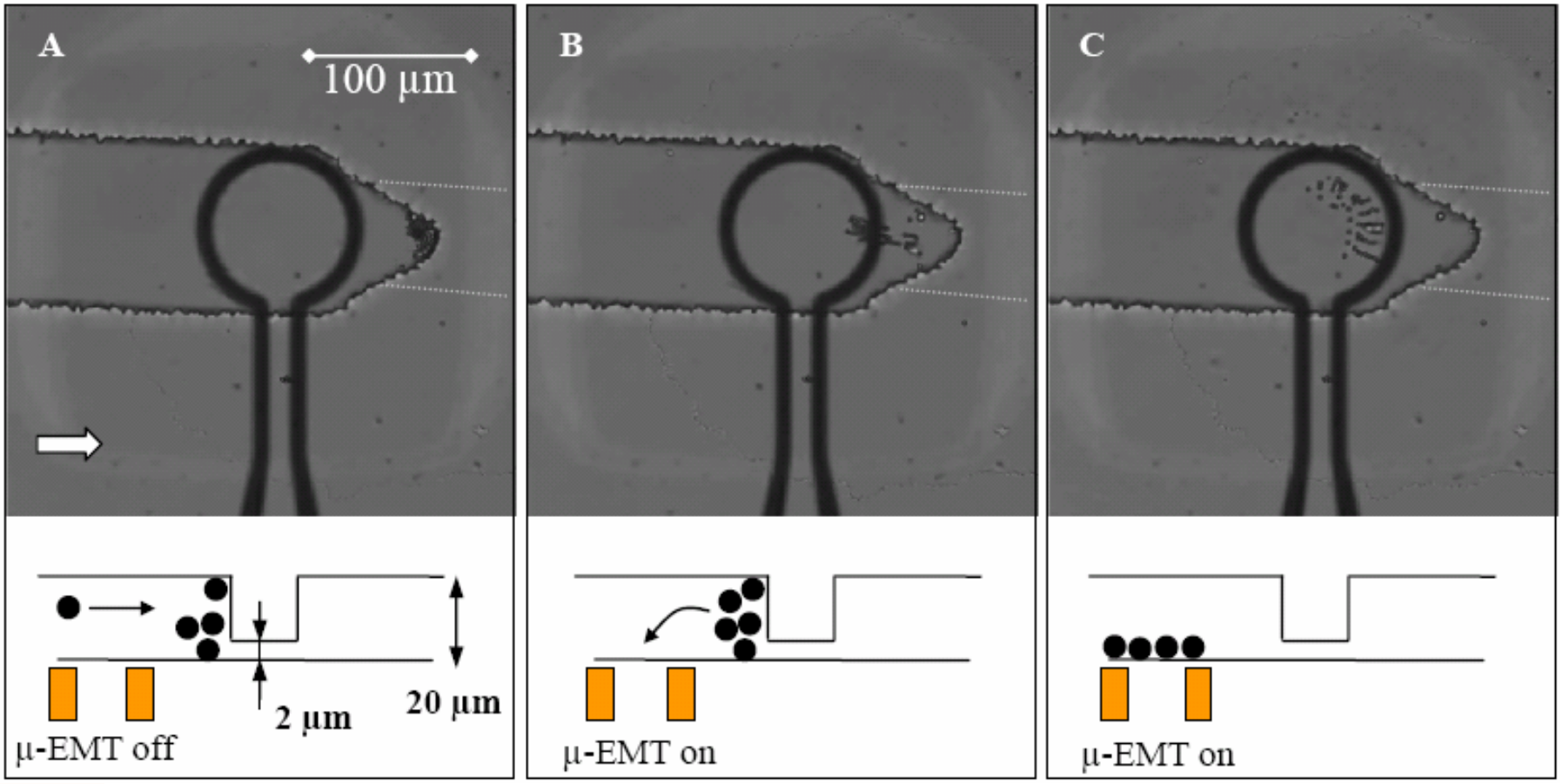


Triplex

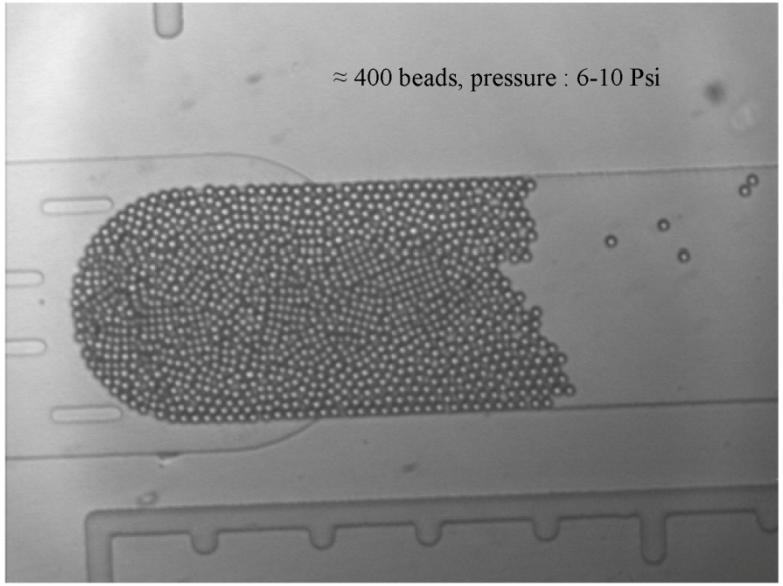
~ 1 million positive targets



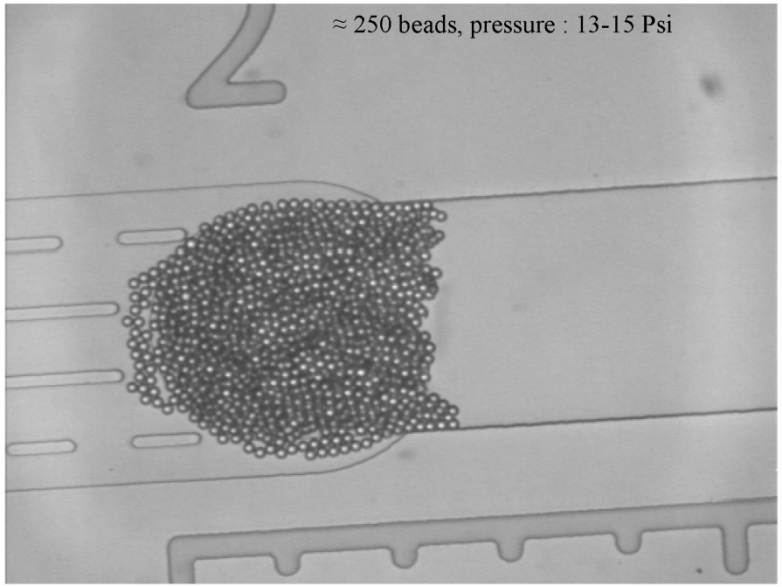
Microfluidic device / filtration



Pressure Effects on Bead Bed



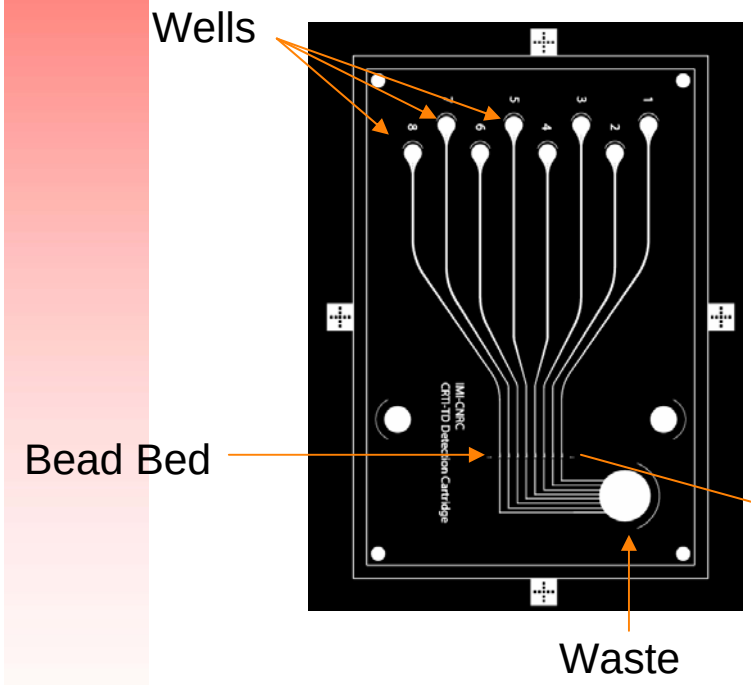
~ 400 beads @ 6-10 psi

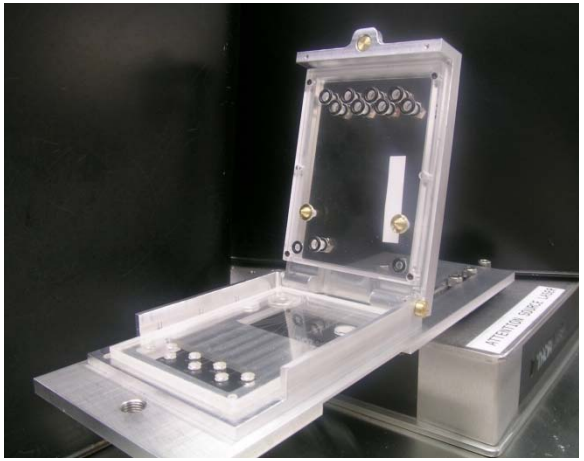


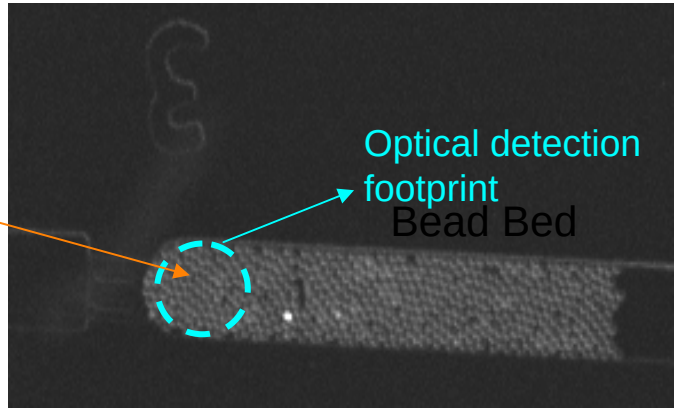
~ 250 beads @ 13-15 psi

Biodetection

Fluorescence Analysis





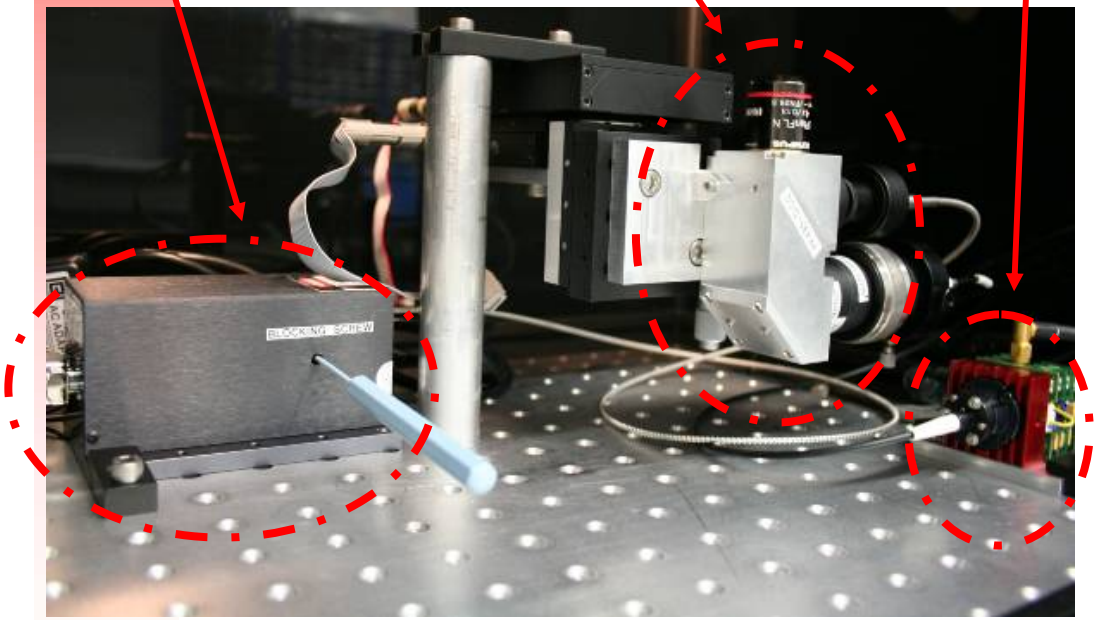


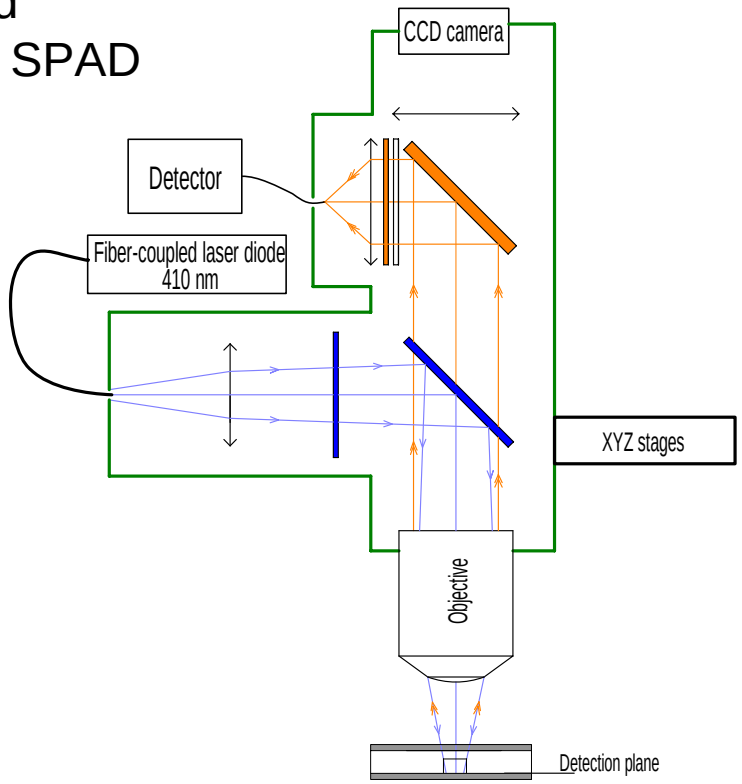
Integrated Optics for Laser-Induced Fluorescence

Compact fiber-coupled laser

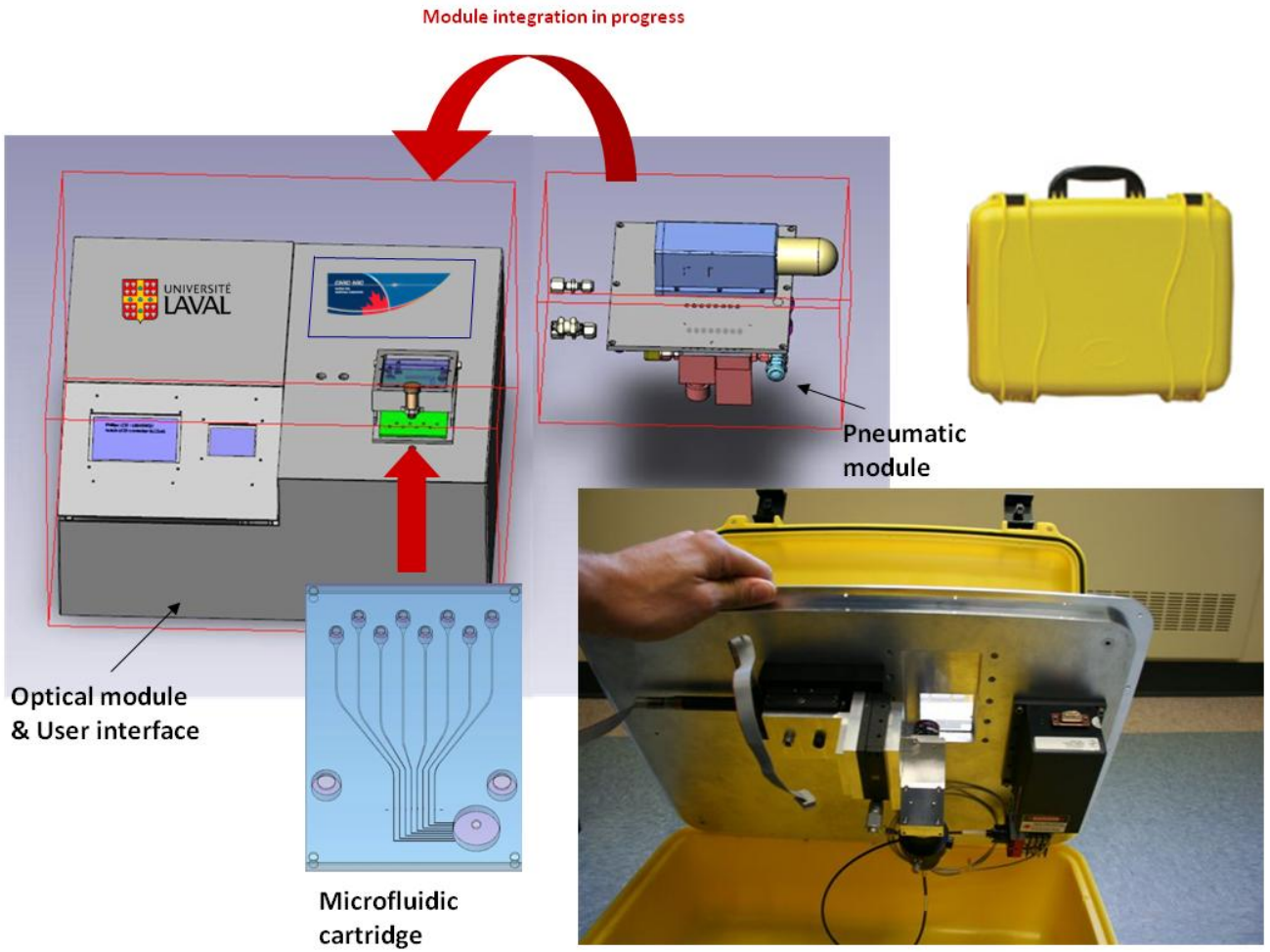
Detection head with integrated optical components

Fiber-coupled Multielement SPAD

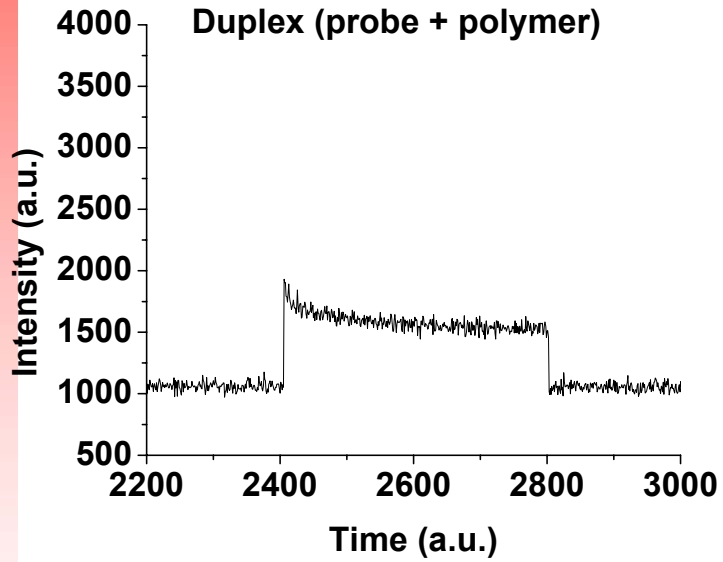


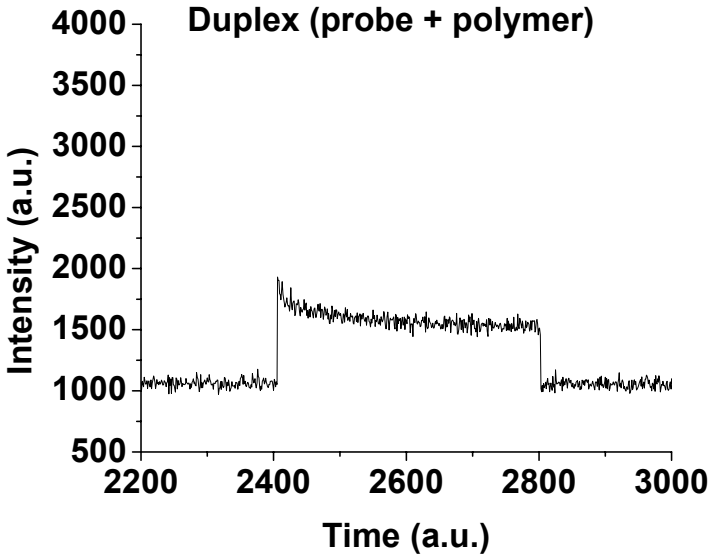


Technological Integration

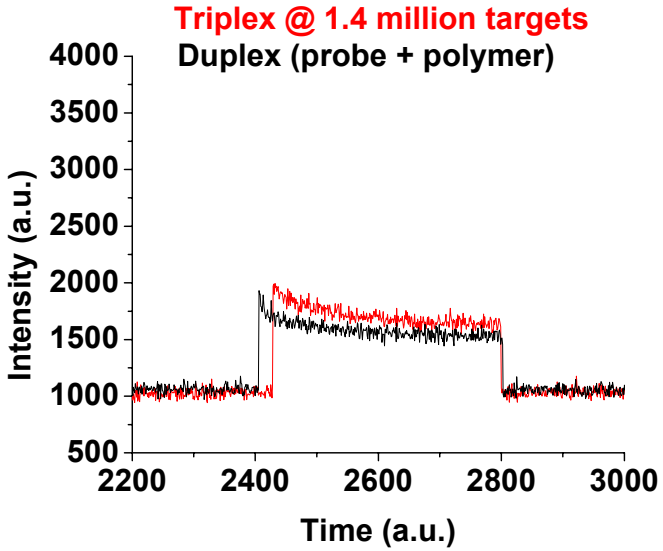


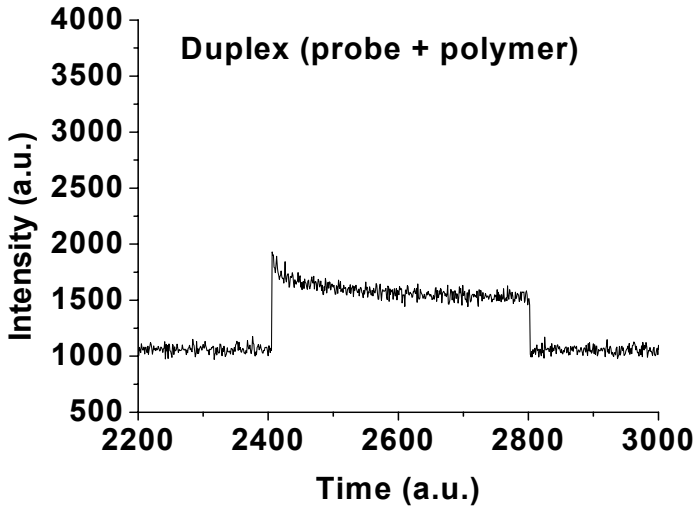
Duplex Beads Without Targets
(Excitation at 405 nm and emission at 575 nm)





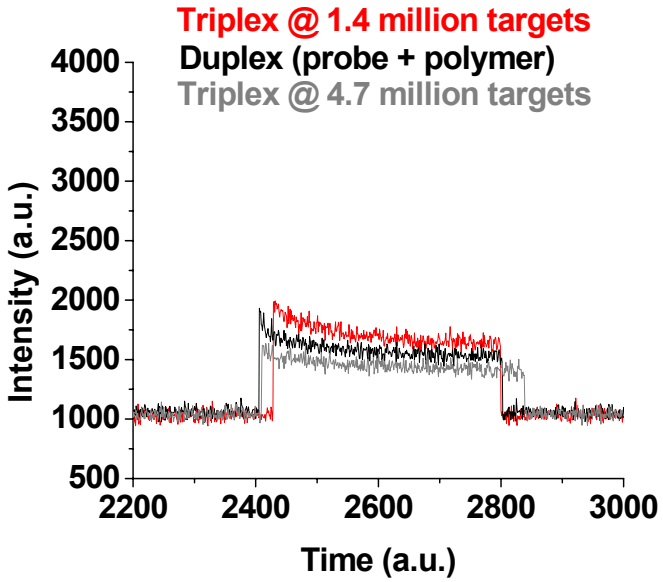
Duplex Beads vs Triplex Beads
(Excitation at 405 nm and emission at 575 nm)

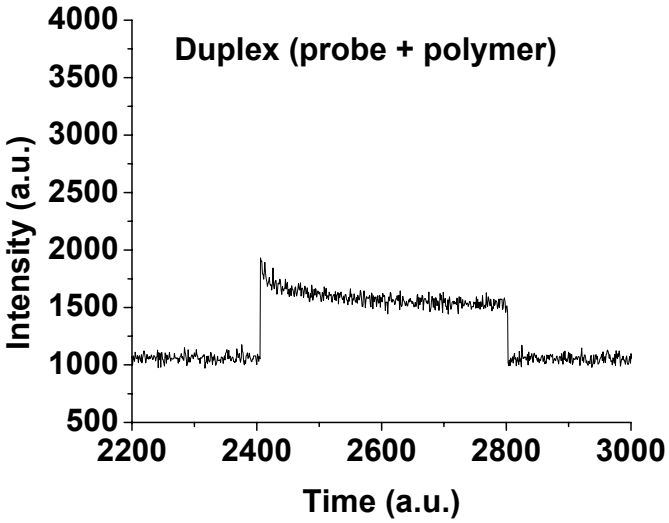




NEGATIVE TARGETS

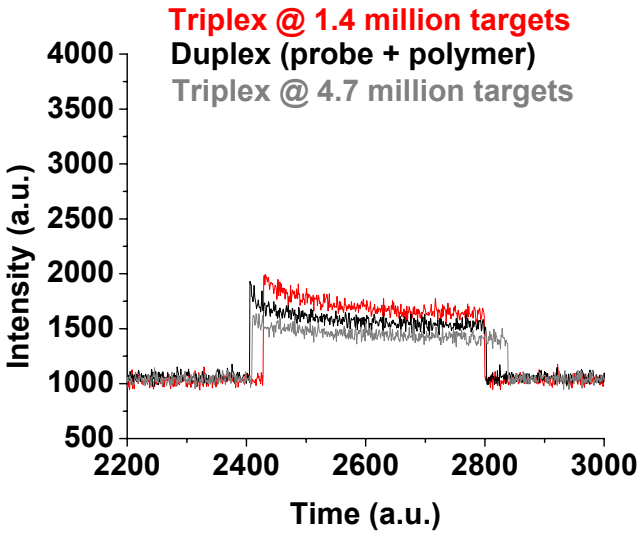
Duplex Beads vs Triplex Beads
(Excitation at 405 nm and emission at 575 nm)



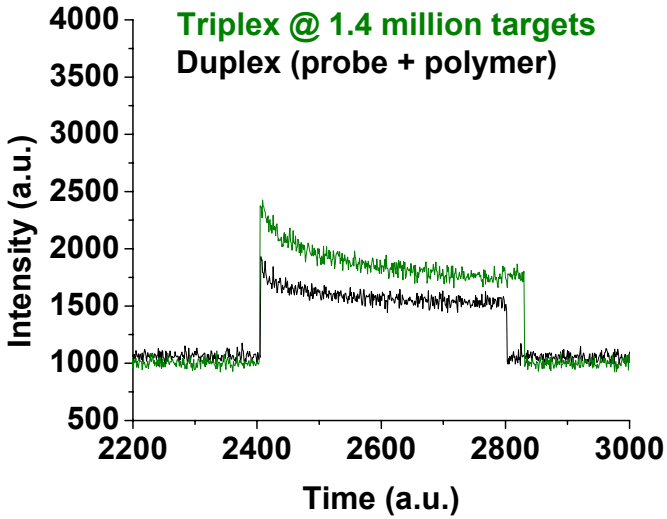


NEGATIVE TARGETS

Duplex Beads vs Triplex Beads
(Excitation at 405 nm and emission at 575 nm)

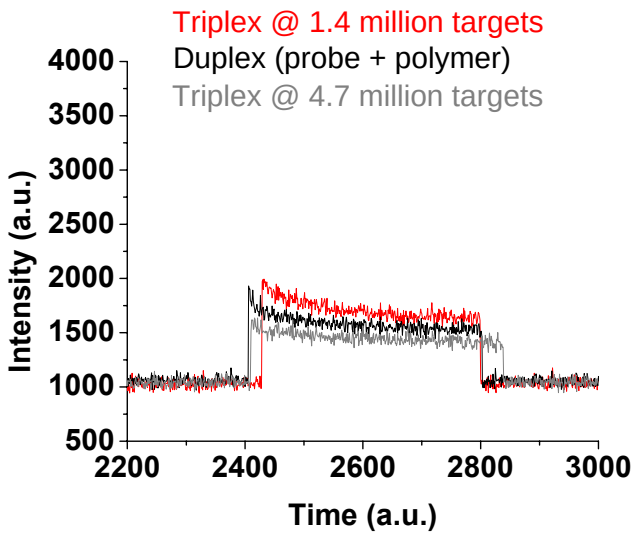


NEGATIVE TARGETS

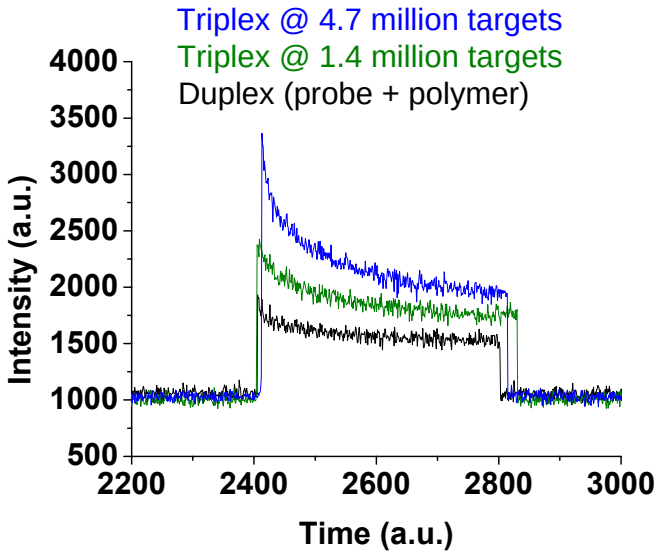


POSITIVE TARGETS

Duplex Beads vs Triplex Beads
(Excitation at 405 nm and emission at 575 nm)

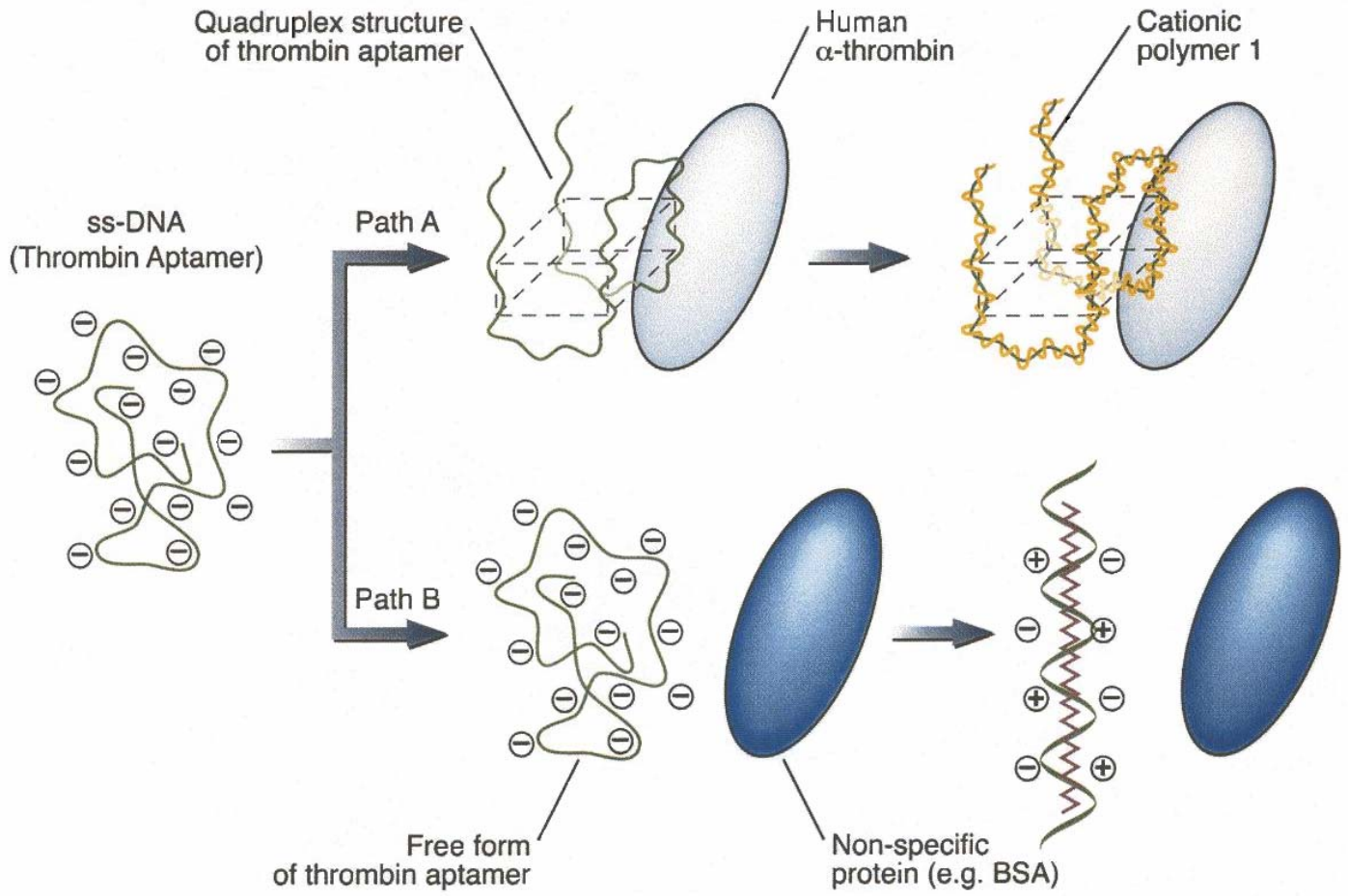


NEGATIVE TARGETS



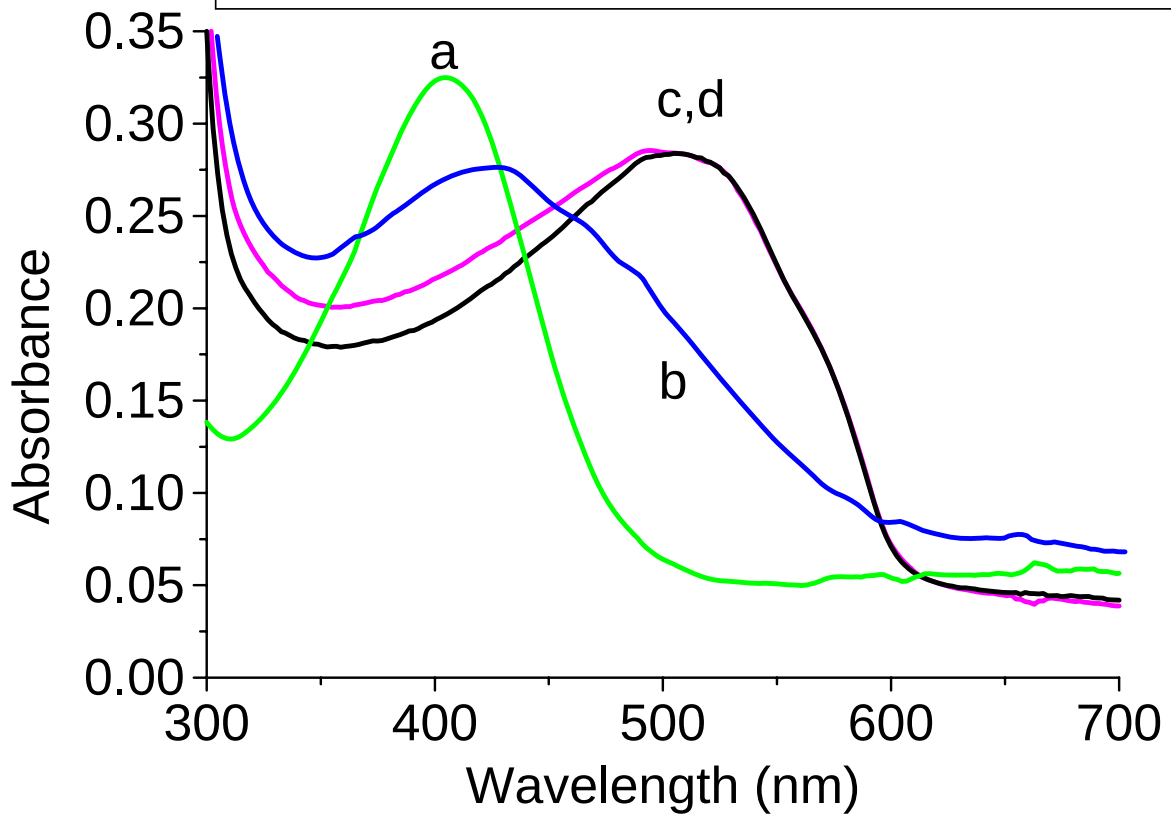
POSITIVE TARGETS

APTAMERS


Quadruplex structure
of thrombin aptamer
Human
α-thrombin
Cationic
polymer 1
ss-DNA
(Thrombin Aptamer)
Path A
Path B
Free form
of thrombin aptamer
Non-specific
protein (e.g. BSA)

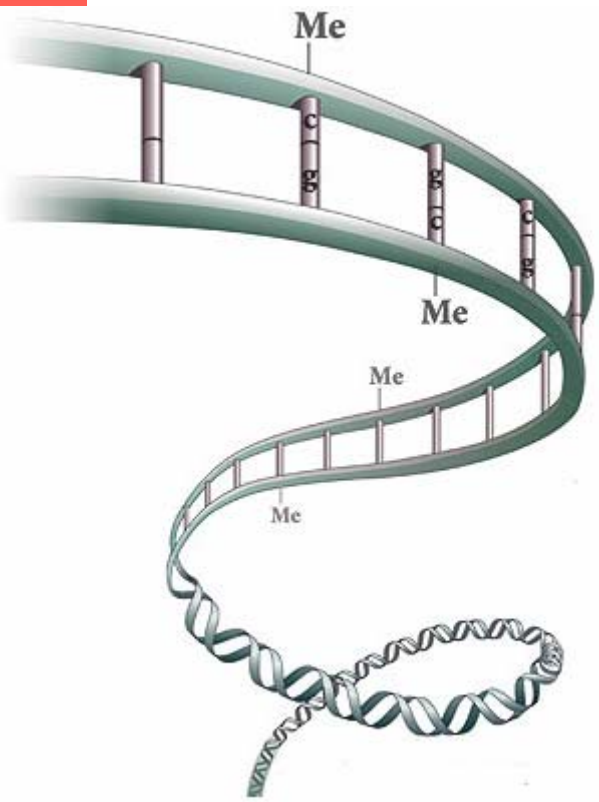
APTAMERS

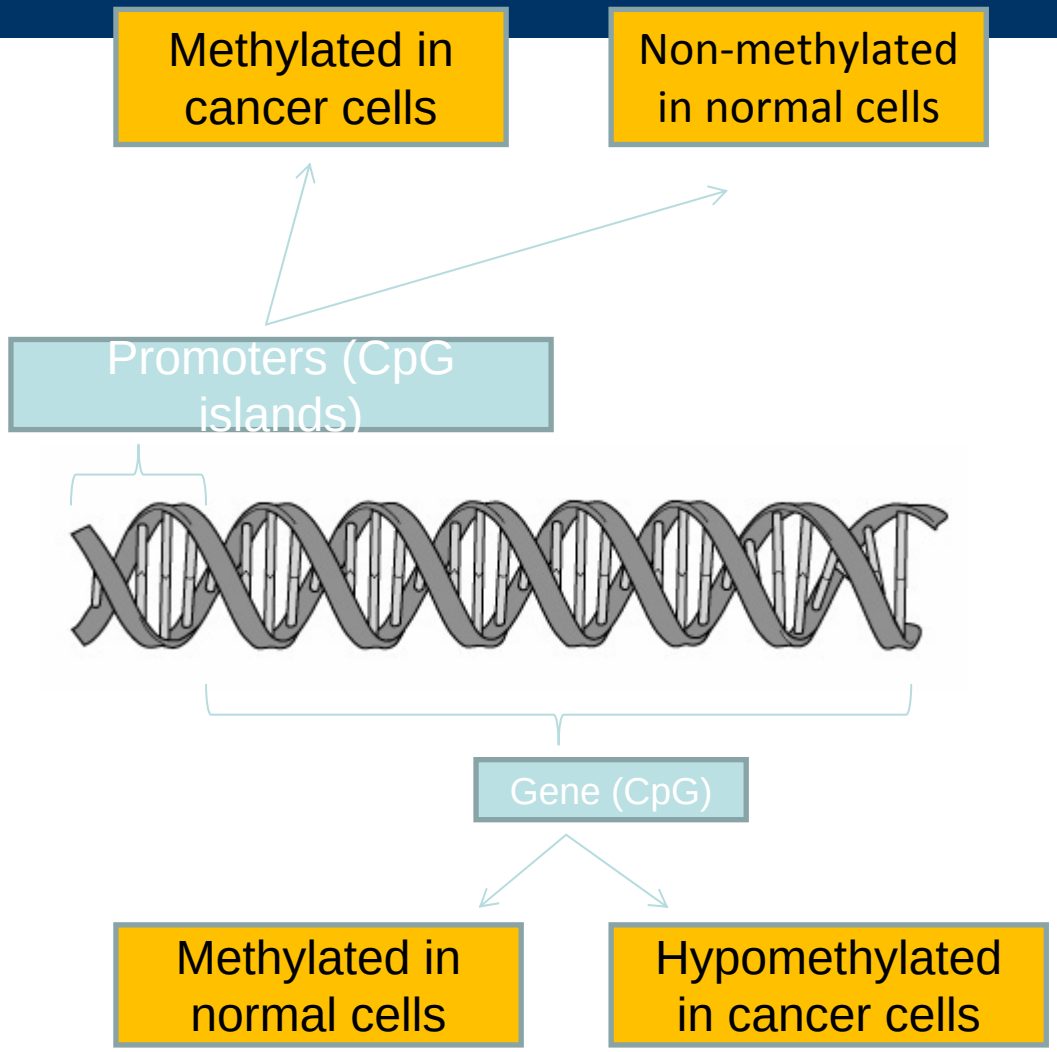
a) polymer 1 in water
b) Complex(1/1/1): Human thrombin/Specific thrombin DNA (X1)/poly 1
c) Mixture: Human thrombin/Non-specific thrombin DNA (X2)/poly 1
d) Mixture: BSA/Specific thrombin DNA (X1)/poly 1



Methylated DNA

Cancer diagnostics





[1] Strichman-Almasshanu, L. Z. et *al*. A genome-wide screen for normally methylated human CpG islands that can identify novel imprented genes. *Genome Research.* **12**, 543-554 (2002)

[2] Feinberg, A. P. et *al.* The history of cancer epigenetics. *Nature Cancer.* **4**, 143-153 (2004)

Methylated DNA

5’- CAT GAT CGA ACG ATC GAC CA – 3’

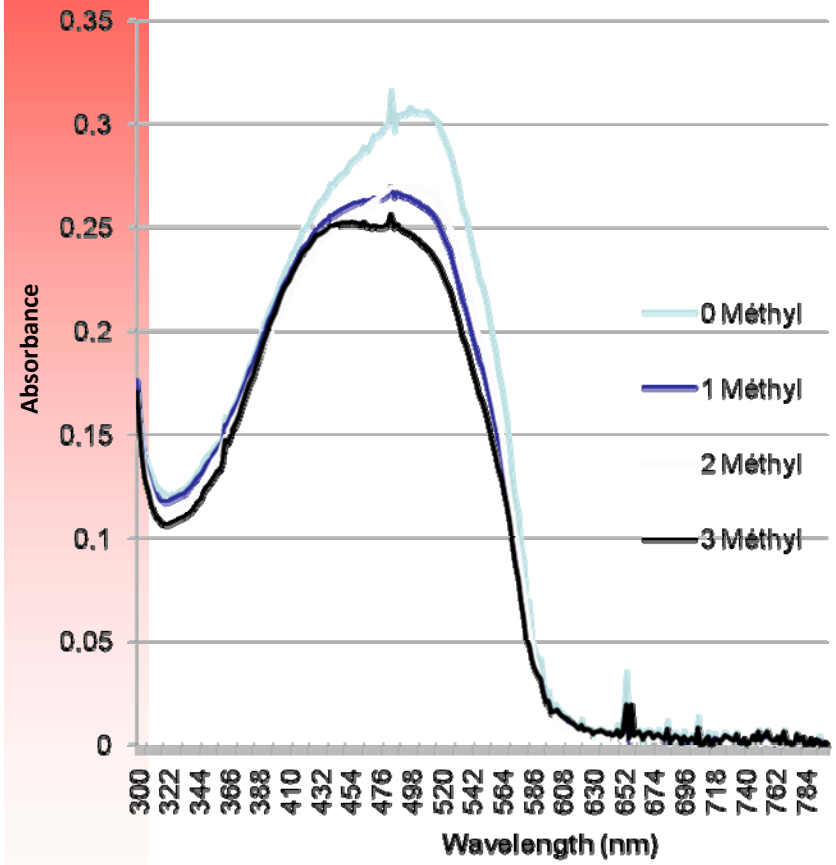


Figure 1. UV-Vis absorption spectra corresponding to the different methylated duplexes at 50°C after 30 minutes.

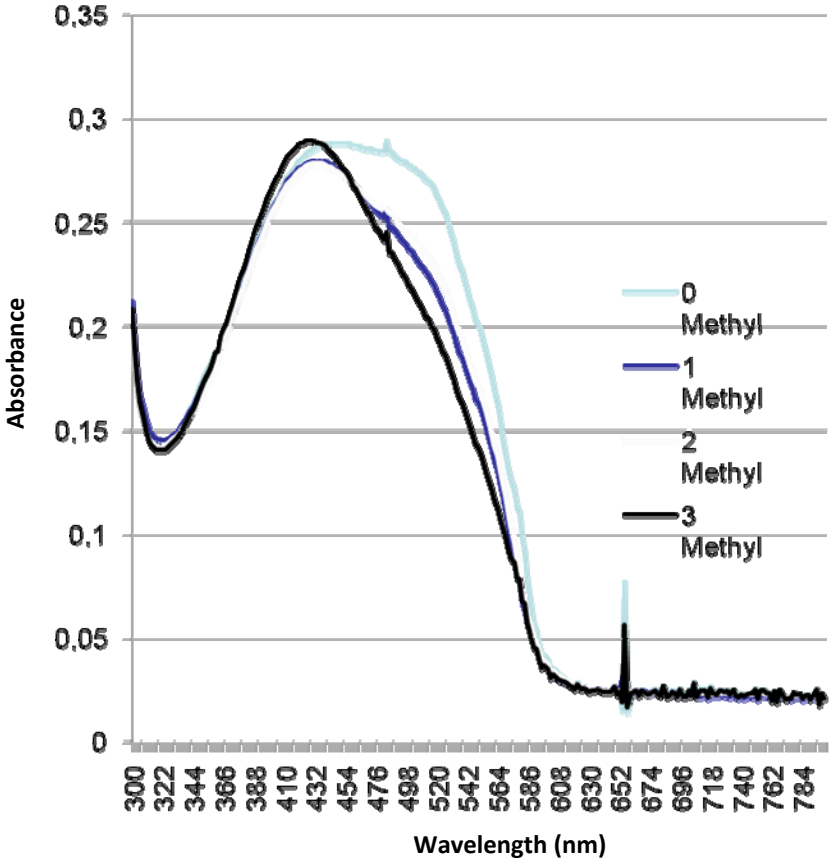


Figure 2. UV-Vis absorption spectra corresponding to different methylated duplexes at 65°C after 3 minutes.

$$$

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- NSERC (DISCOVERY, STRATEGIC)
- CANADIAN FOUNDATION for INNOVATION
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