

# DNATOOL

Conception de multiblocs hybrides polymère-ADN et développement d'outils microfluidique pour la caractérisation en régime très dilué

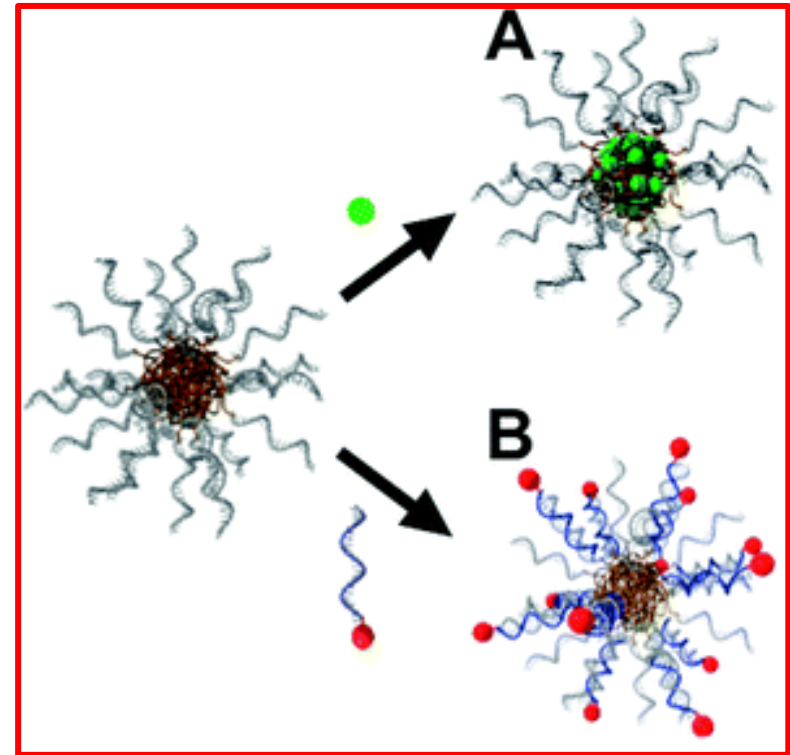
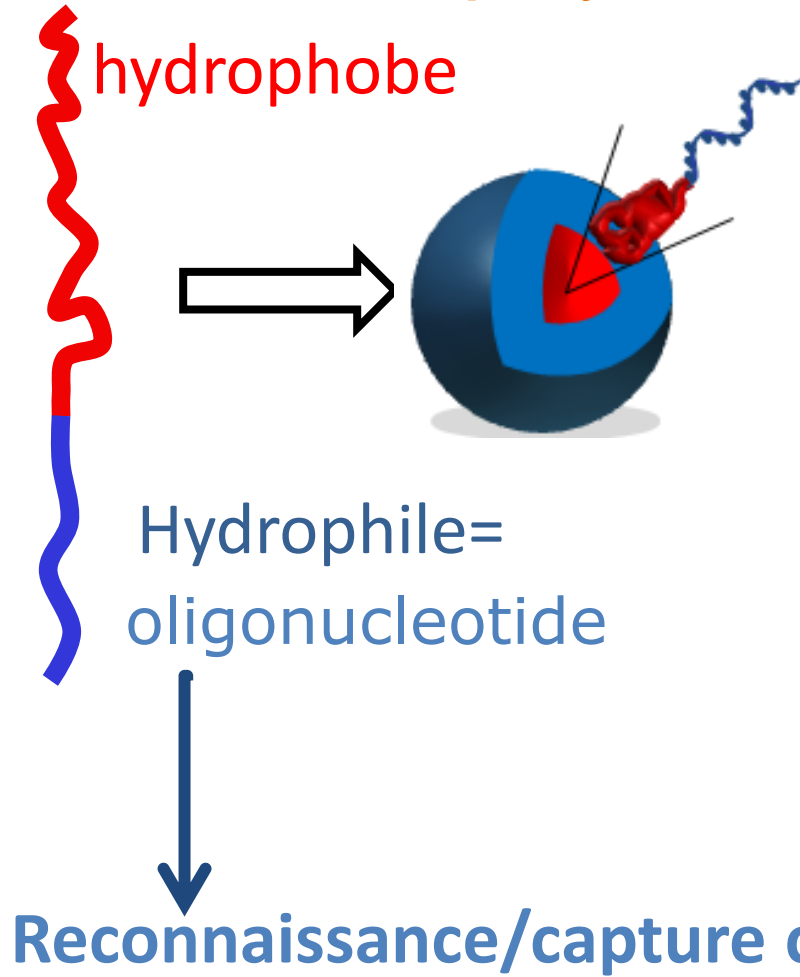
F. GAUFFRE

Institut des Sciences Chimiques de  
Rennes

Journées Nationales Nanosciences et  
Nanotechnologies 2012

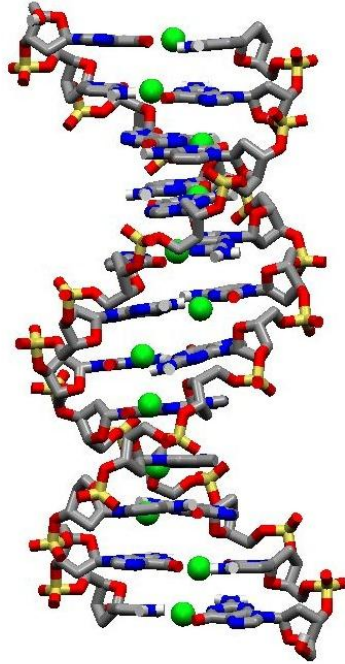


# copolymères hybrides



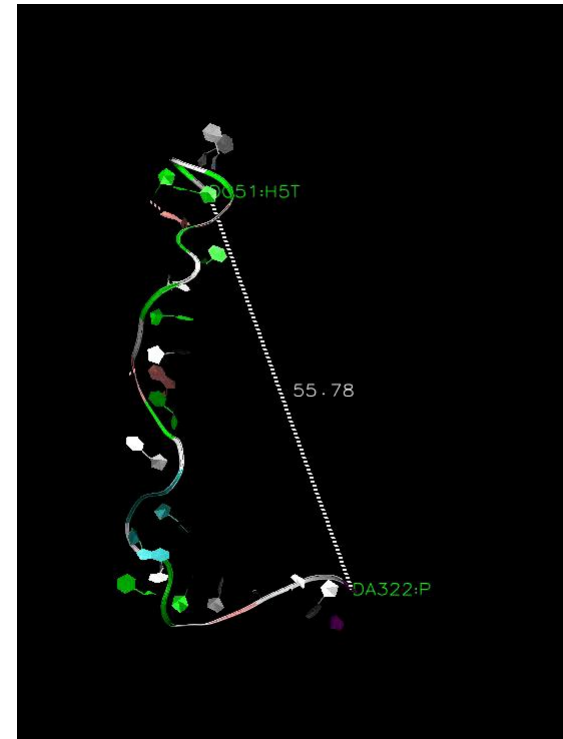
*Herrmann, Angewandte, 2006*

# Technologie ADN pour l'assemblage



Oligonucleotides

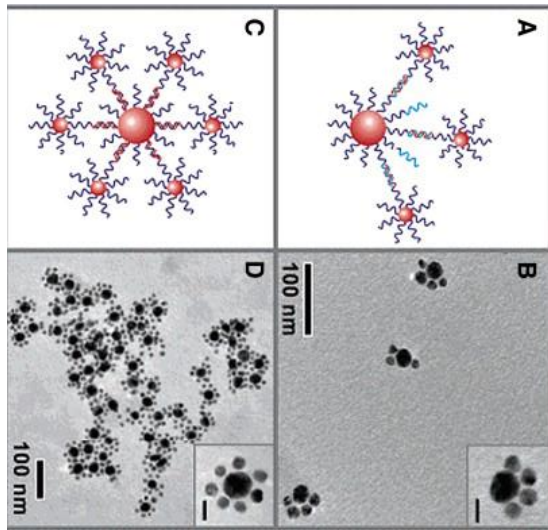
- Reconnaissance spécifique
- Reversibilité en température



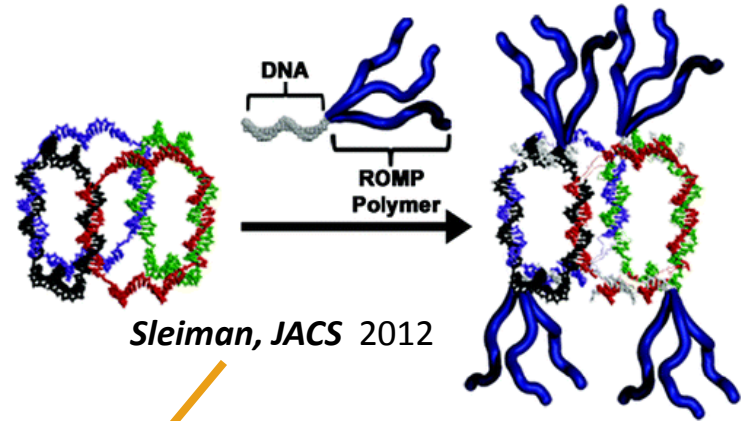
M.-N. Brut, LAAS

- Synthétiques, commerciaux

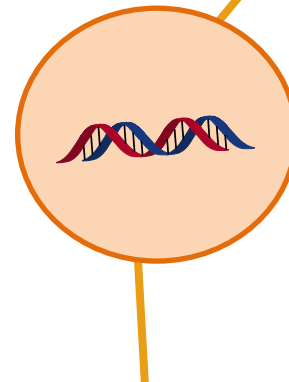
# Technologie ADN pour l'assemblage



Mirkin JACS, 2006



Sleiman, JACS 2012

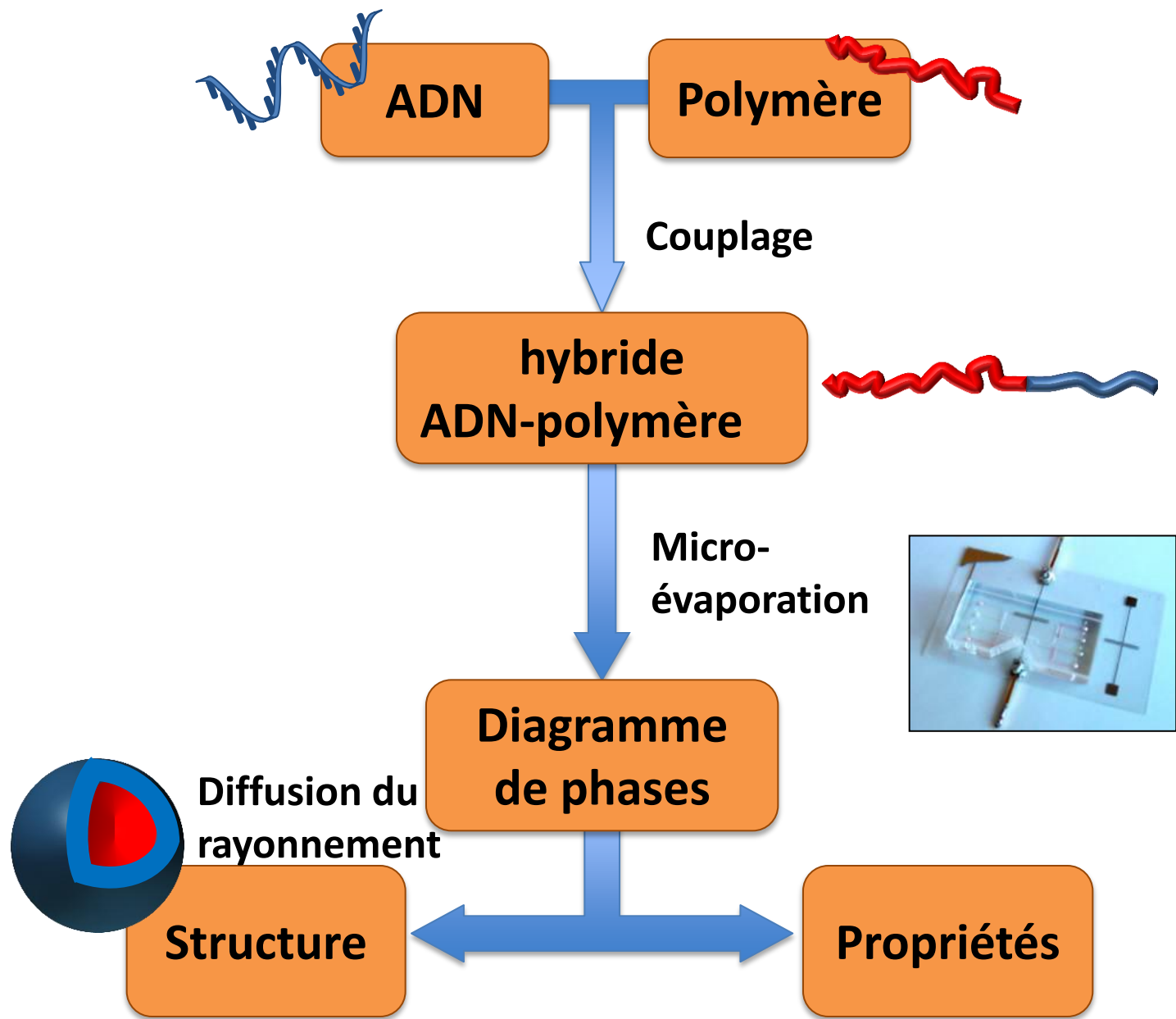


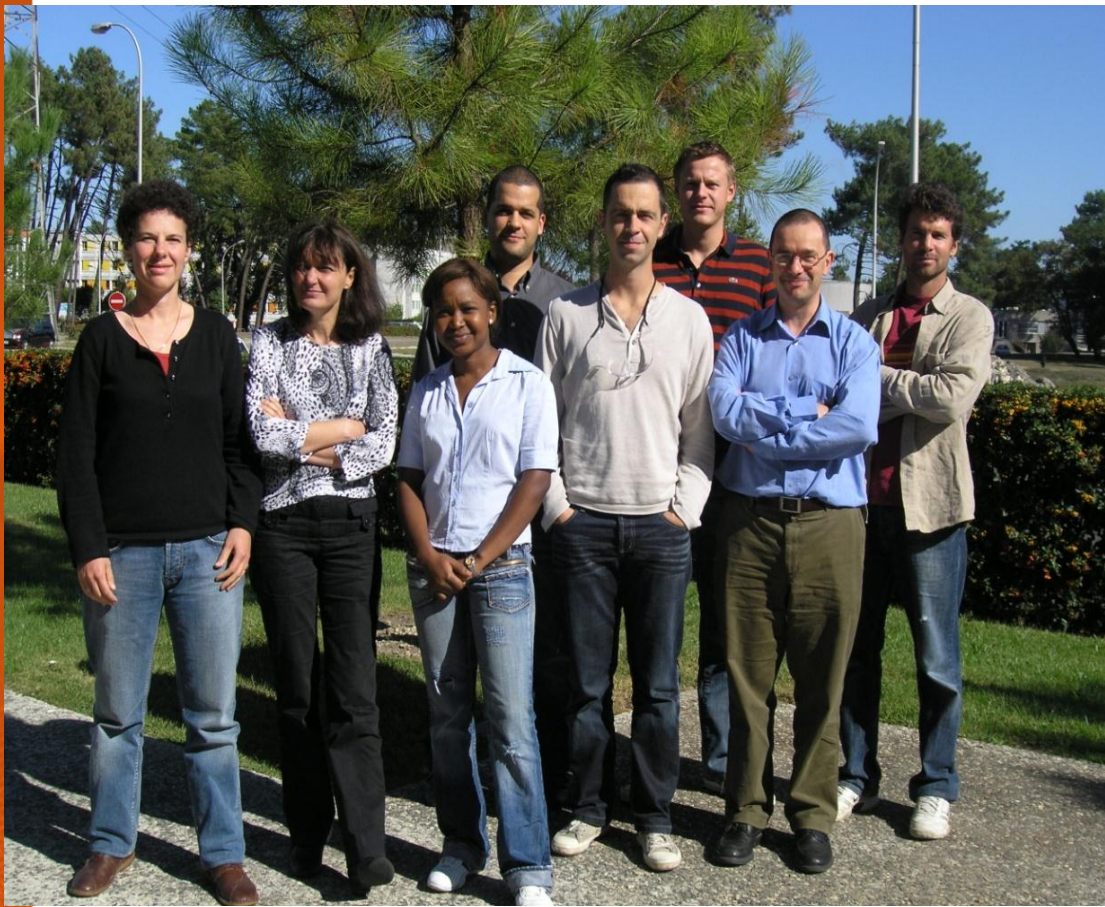
Rothmund, P. Nature 2006

Our main objective in this project is to master the **elaboration of nano-objects based on the self-aggregation, in solution, of block copolymers containing a single oligonucleotide (ODN)**. These hybrid objects have many potential applications in biology and therapeutics: tools for the screening of pathological genes, reagents for antisense strategy (RNA<sub>m</sub> coding for tyrosine kinase receptors, the protein raf, the telomerase may also be interesting targets), genes transfer, agents for the identification of new DNA binding proteins...In colloidal assemblies, the self-assembled nano-structures correspond to the state of thermodynamical equilibrium, so that the objects form spontaneously. In addition, such dispersed systems present a large specific area available for interactions. These two features can be advantageously used to produce colloidal sensors, that are sensitive and easy to process in comparison with solid-supported sensors. The large and very promising field of investigations opened by the self-assembled DNA-“tools” requires however the perfect control and knowledge of those nano-objects. This is why **this four years long project will be focused only on their elaboration and characterization.**

Our project will be implemented in three well-defined and complementary steps:

- 1) The **synthesis** of block copolymers containing various ODN.
- 2) The use of **microfluidic** devices to construct the phase diagram of our ODN-based block copolymers in aqueous solutions. This will allow us to select the most interesting polymer systems depending on the type of nano-object.
- 3) Full **characterization** of the structures and stability of the previously selected structure, keeping present in mind their potential applications.





**LOF (Bordeaux):**

J. Leng

J. Dehmoune

**IMRCP (Toulouse):**

F. Gauffre

C. Mingotaud

R. Mayap-Talom

G. Fuks

L. Kaps

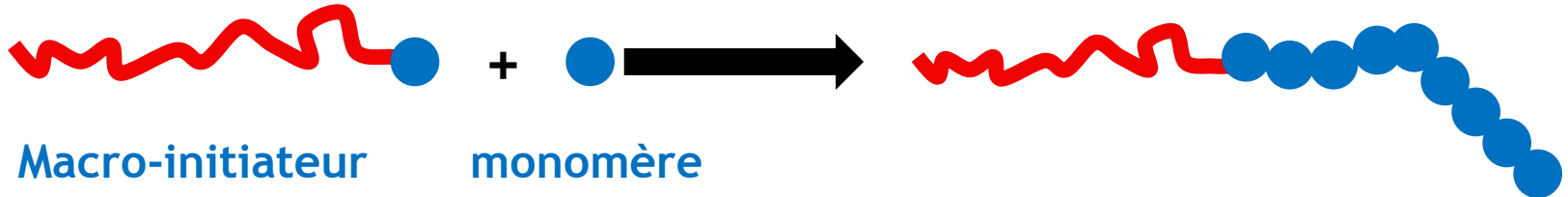
P. Vicendo

**LCVN (Montpellier):**

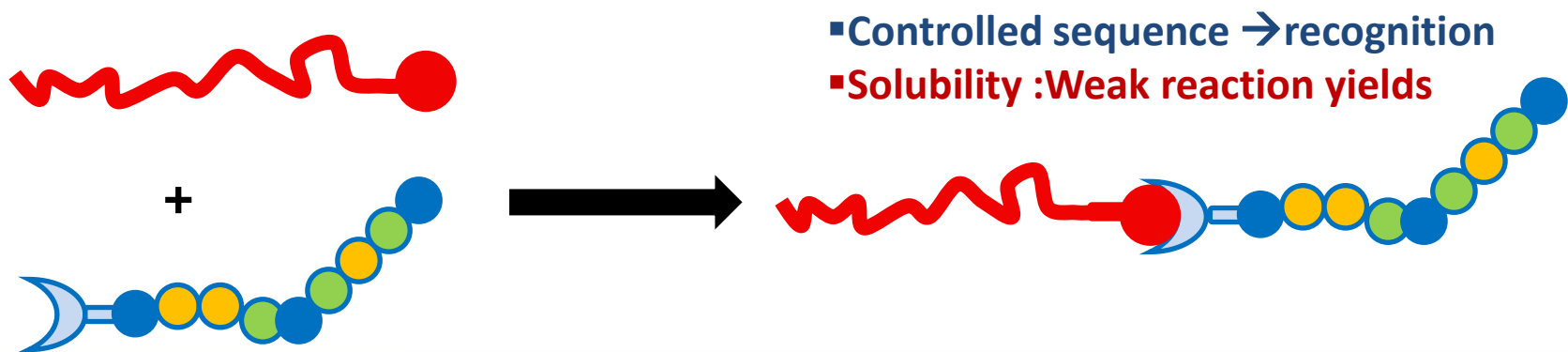
J. Oberdisse

SYNTHÈSE

➤ Divergente



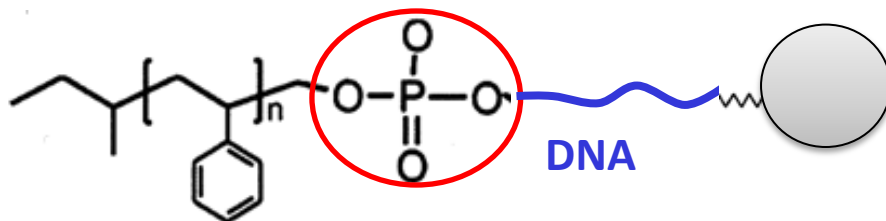
➤ Convergente



➤ Convergent synthesis: choice of the linker?

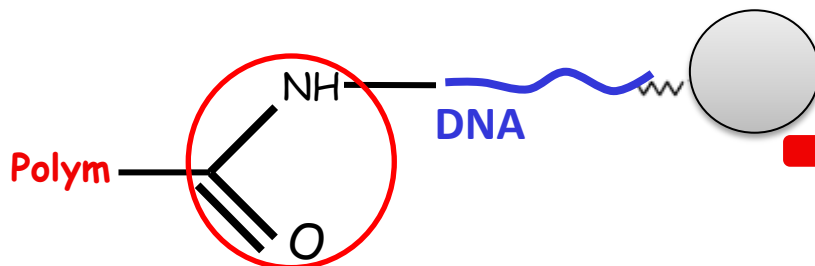
**DNA** -OH; NH<sub>2</sub>; Click

→ Phosphodiester linkage



**Polymer** -OH HO- **DNA**

→ Peptide linkage



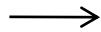
**Polymer** -COOH H<sub>2</sub>N- **DNA**

## Purification and characterization

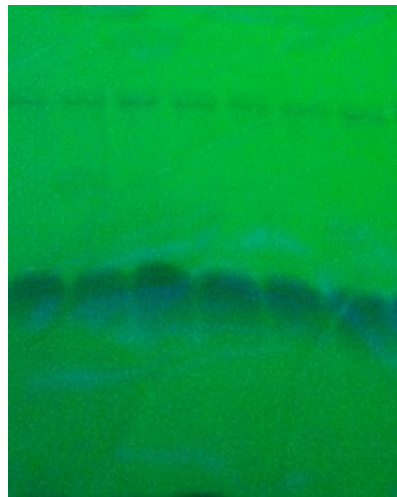
- limited amount of materials
- biochemical techniques

## ▪ Maldi-TOF

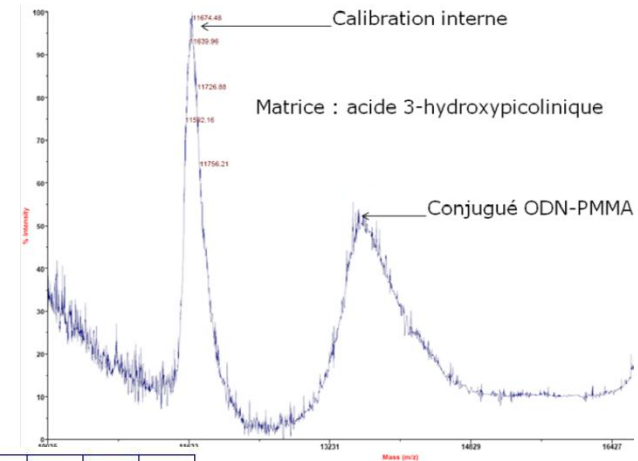
Conjugates



Free DNA



Gel electrophoresis





$1000 < M_n < 12000 \text{ g.mol}^{-1}$

5'-CCTCGCTCTGCTAATCCTGTTA-3' (12-22 bases)

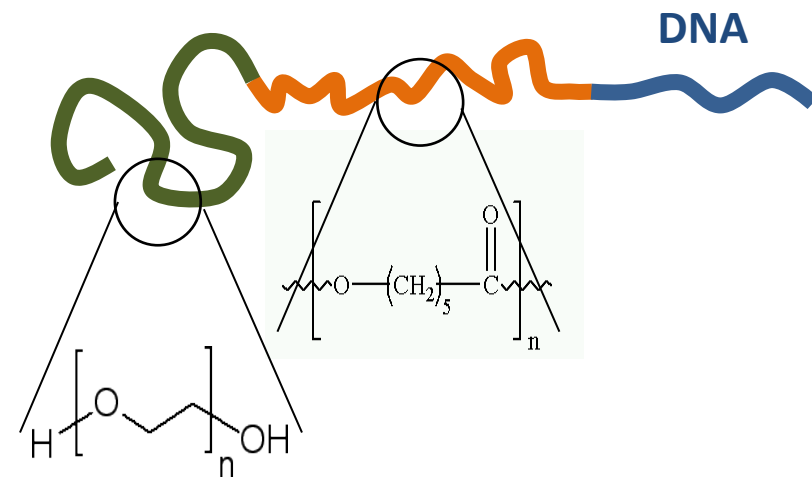
$3000 < M_n < 7000 \text{ g.mol}^{-1}$

DNA Sequence : no hair pin

### ▪ Diblocks

Poly(xxx)		T <sub>g</sub>
isoprene		-60°C
caprolactone		60°C
dimethylsiloxane		-90°C
methylmetacrylate		+120°C

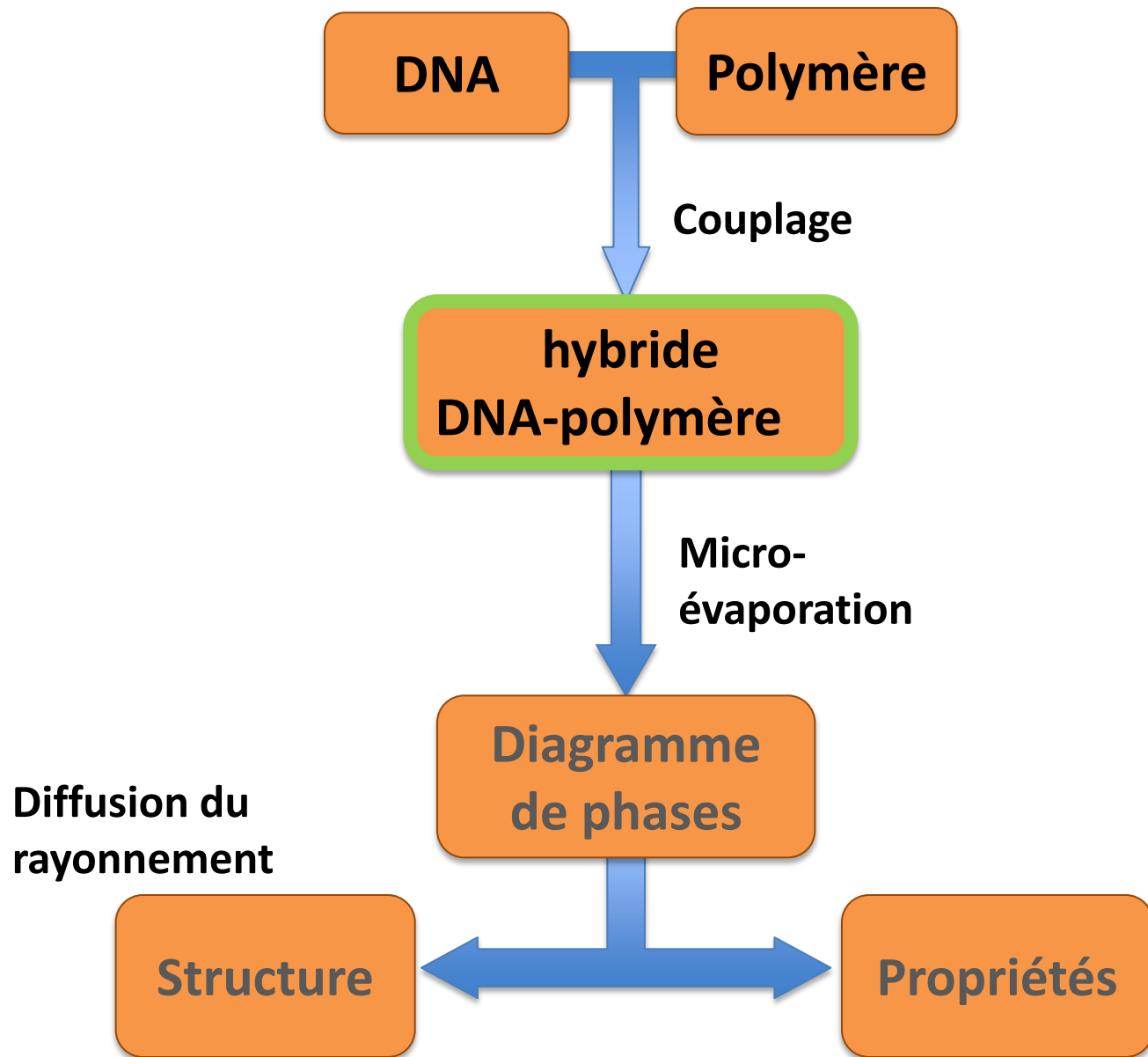
### ▪ Triblocks



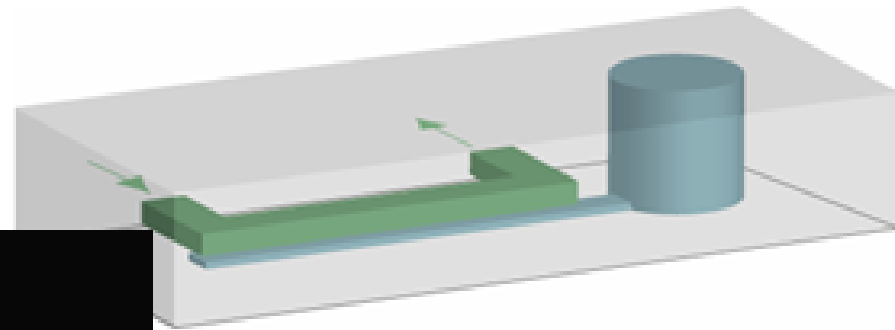
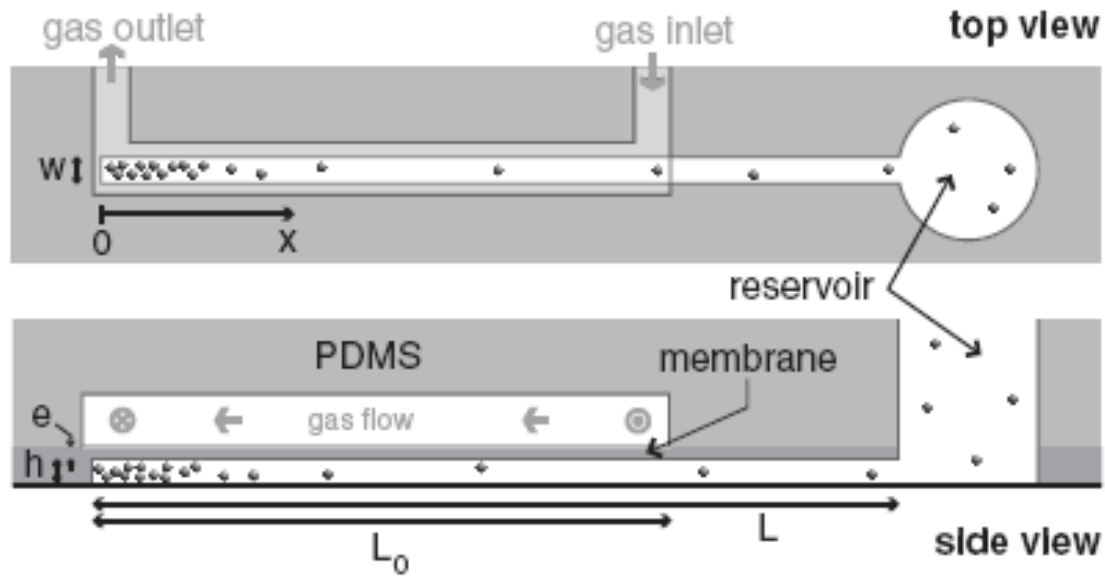
Poly(ethylene oxide)-b-Poly(caprolactone)-DNA

•5 nouveaux diblock hybrides: biodegradables; vitreux...

•1 triblock « pegylé »

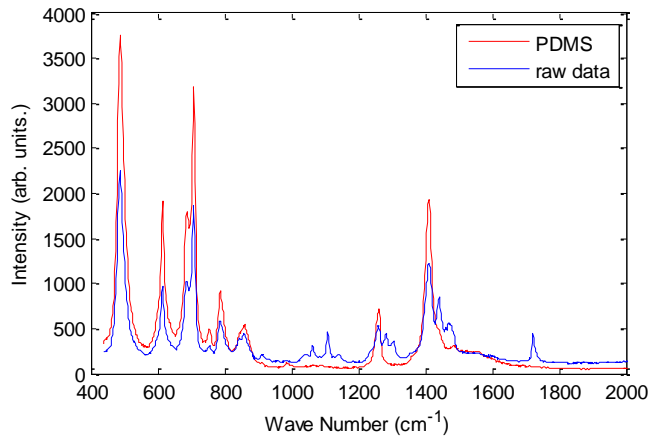


M I C R O E V A P O R A T I O N

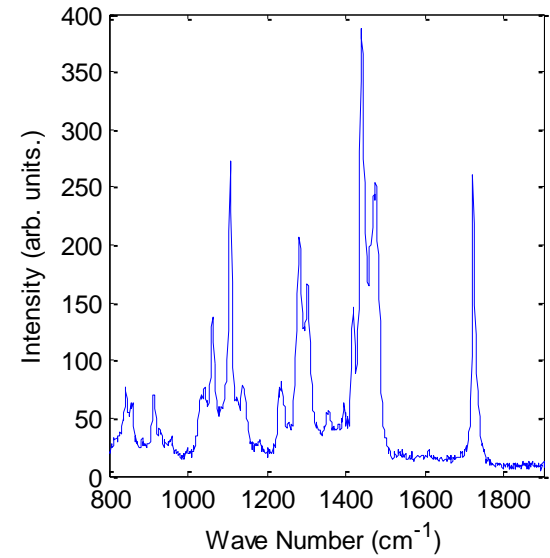


↑ isotropic    ↑ lamellar    ↑ cubic    ↑ hexagonal     $100\ \mu\text{m}$

# - Spectroscopie Raman



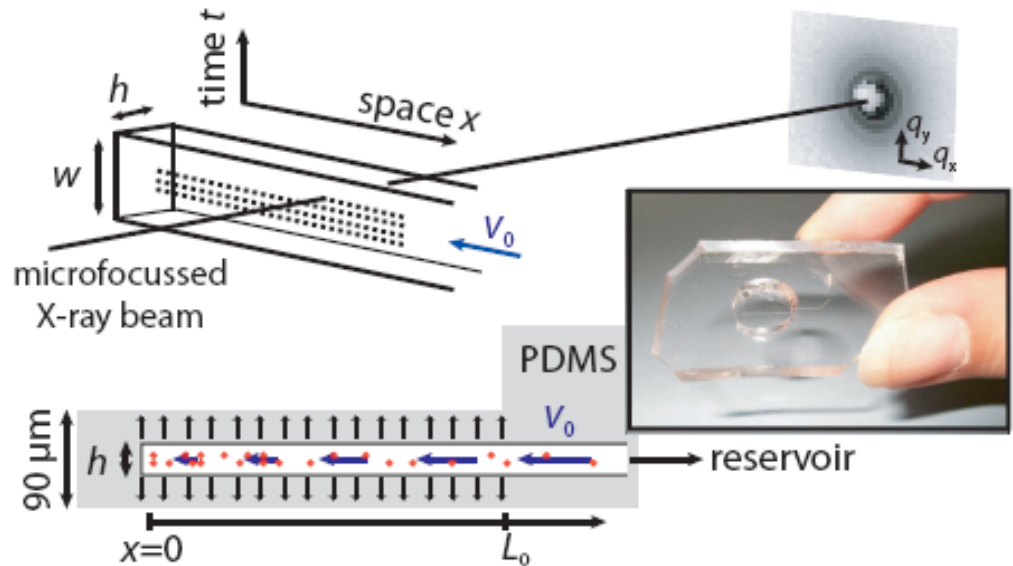
→  
Soustraction du  
signal PDMS



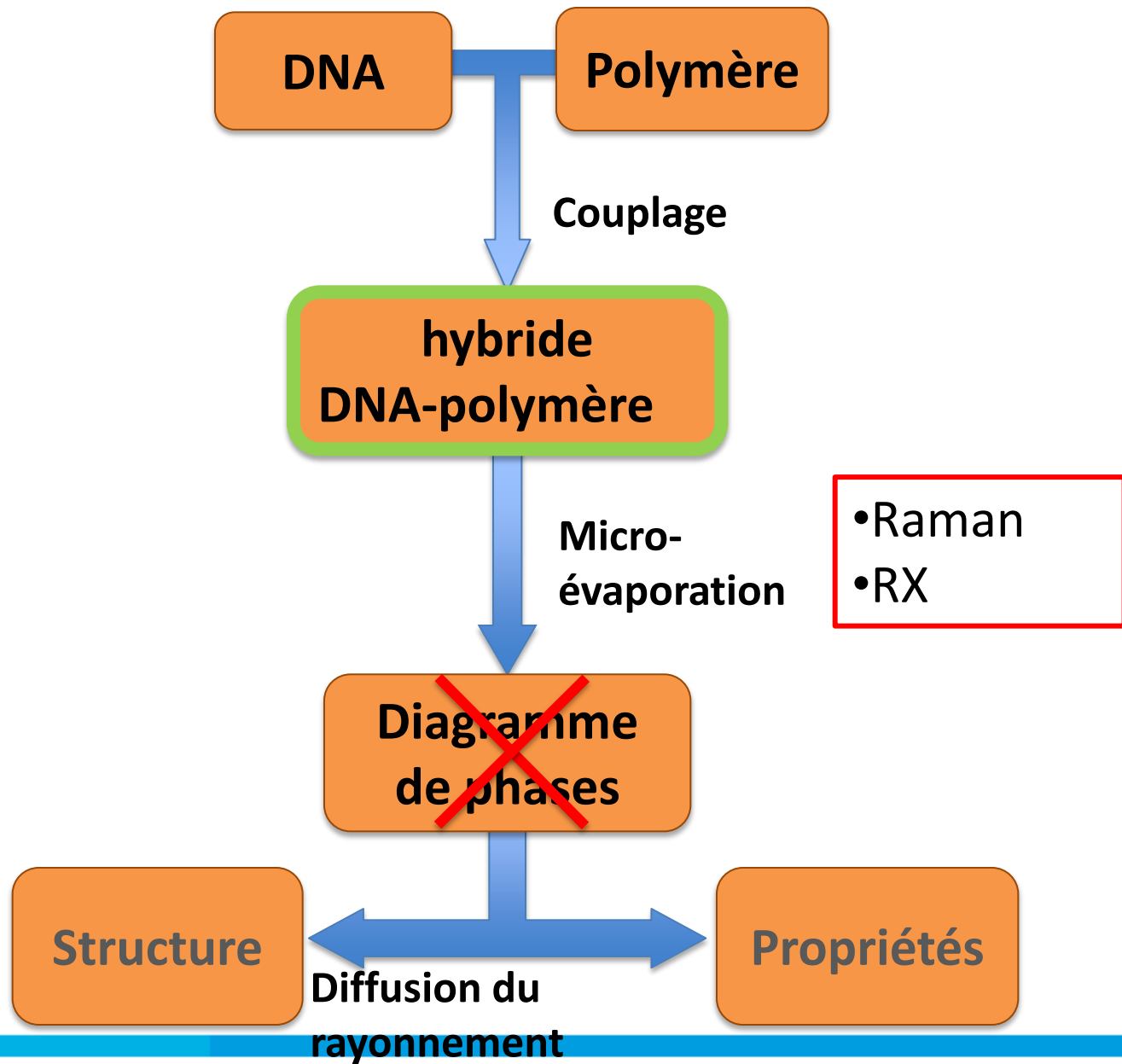
# - SAXS



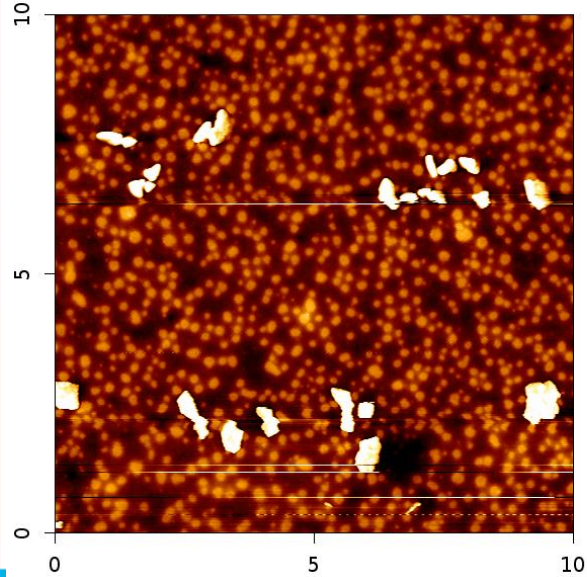
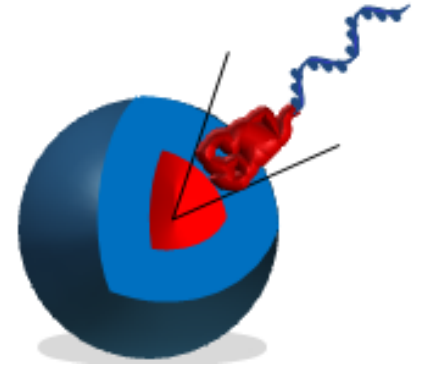
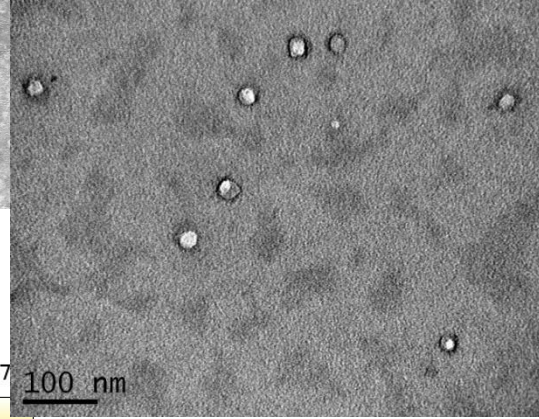
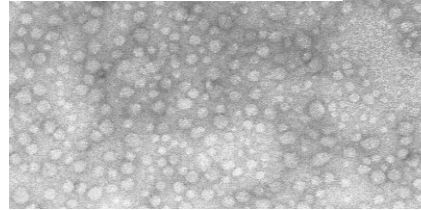
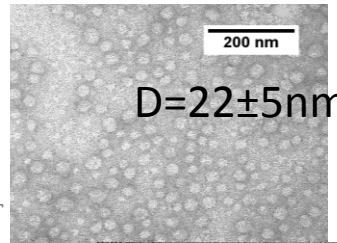
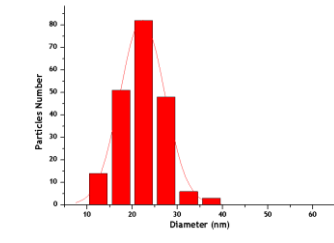
Microfocussed beamline  
for on-chip time & space  
resolved measurements



- Implémentation de mesures RAMAN et SAXS sur le micro-évaporateur
- Zone d'intérêt trop diluée



## Caractérisation/surface



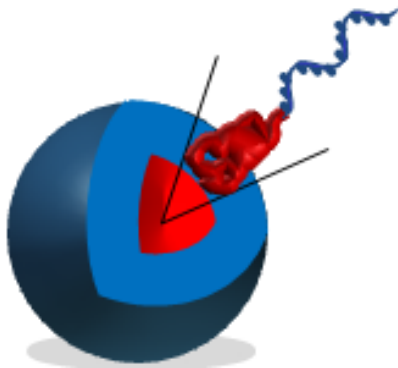
PMMA(6300)-DNA(6800)<sub>22</sub>

Formation de micelles →  
comportement atypique ADN

- Formation de micelles avec tous les polymères (blocs ADN plus court pour vésicules)

# Caractérisation en solution

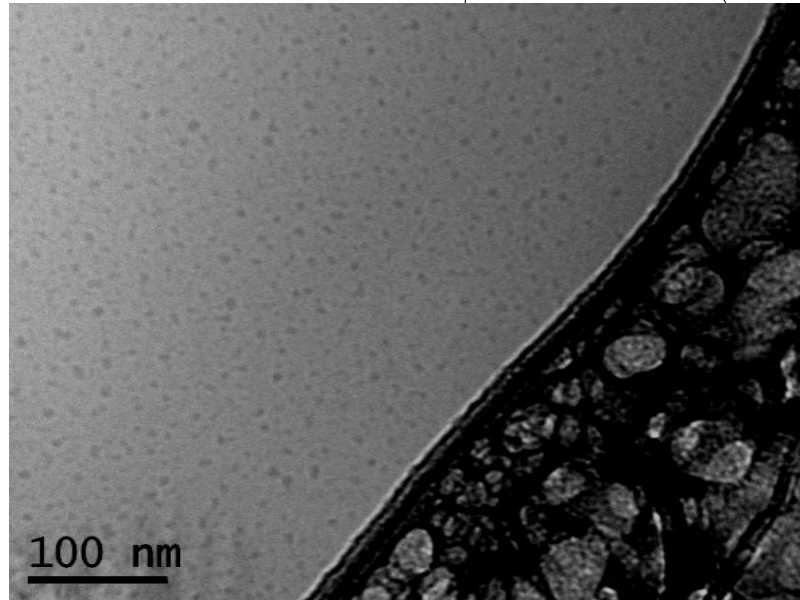
PMMA(6300)-DNA(6800)<sub>22</sub>



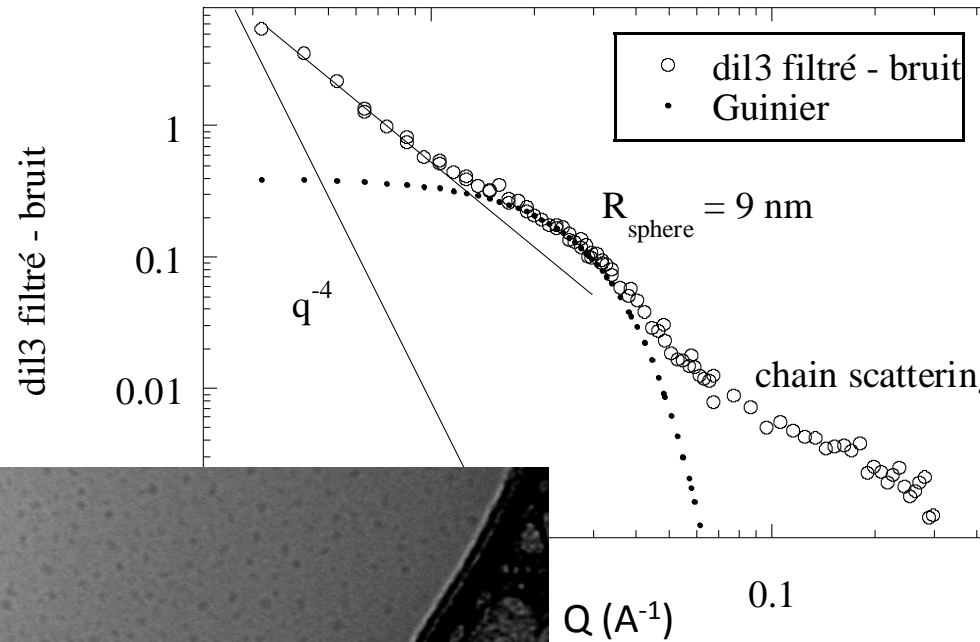
$N_{agg} \sim 30$

$D_{core} \sim 8 \text{ nm}$

$D \sim 30 \text{ nm}$



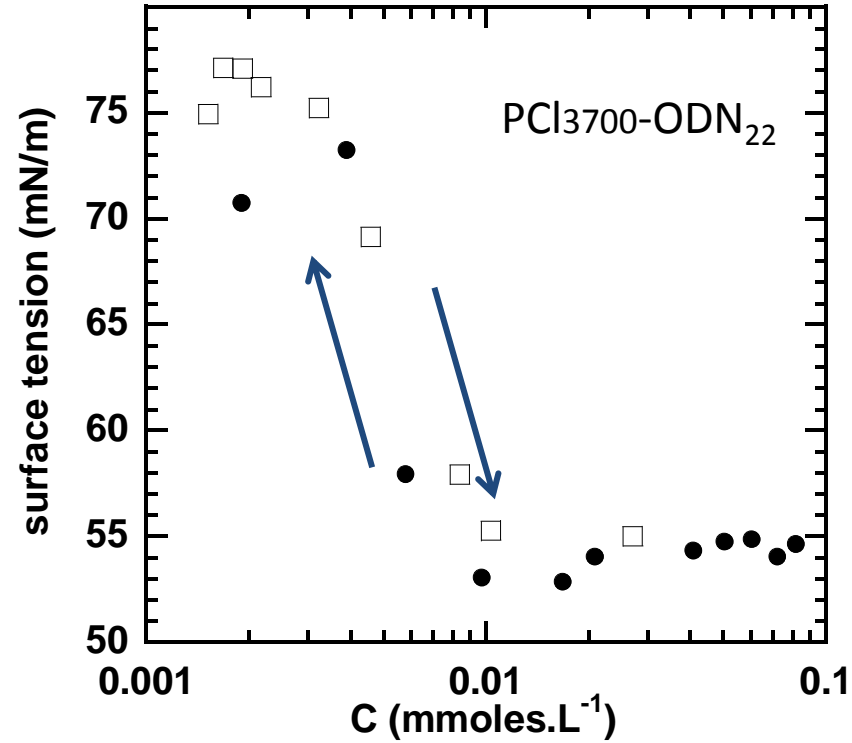
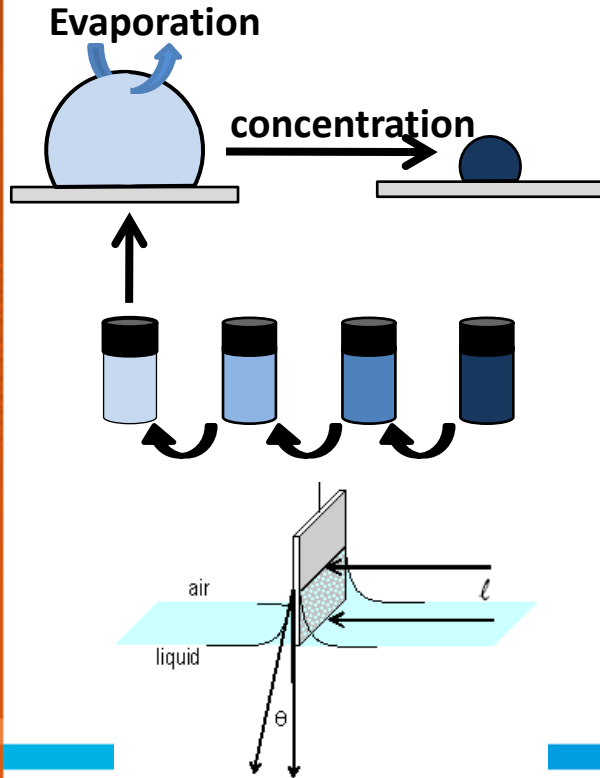
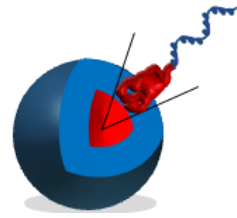
PEOPCLODN\_dil3\_filtre



- Structures des micelles caractérisées
- Présence d'aggrégats en solution → effet « collant » de l'ADN

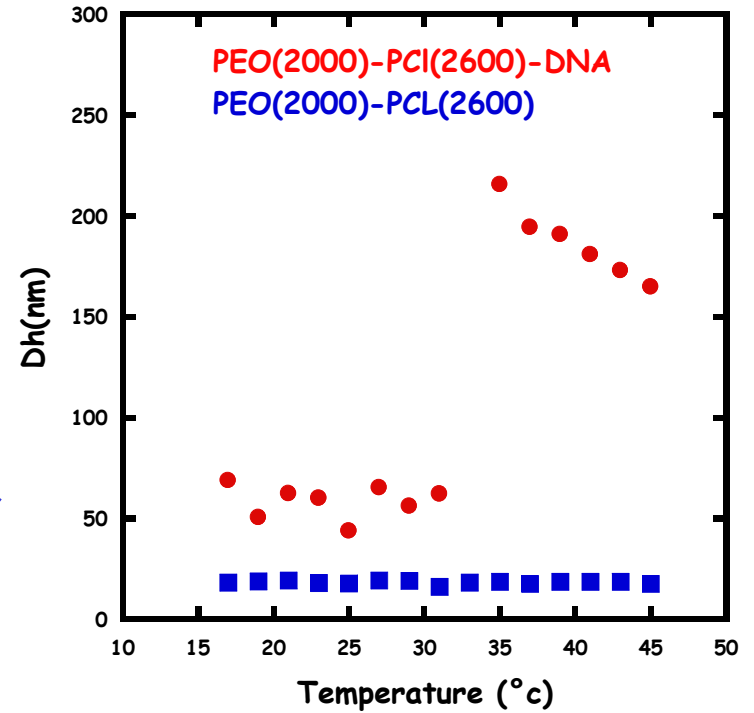
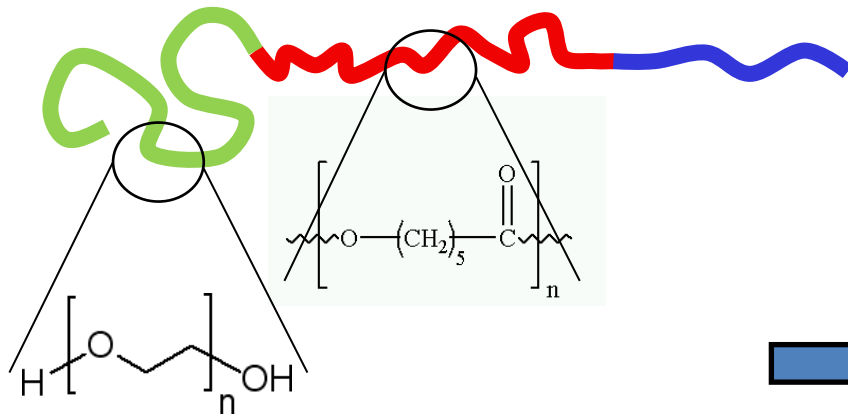
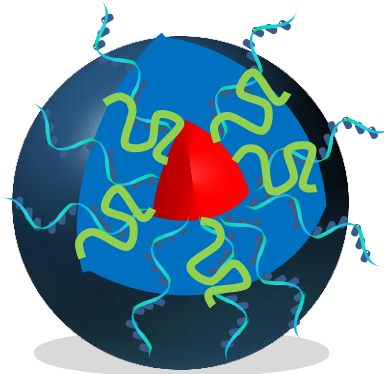
# Reversibilité

Unimers  $\rightleftharpoons$  Micelles

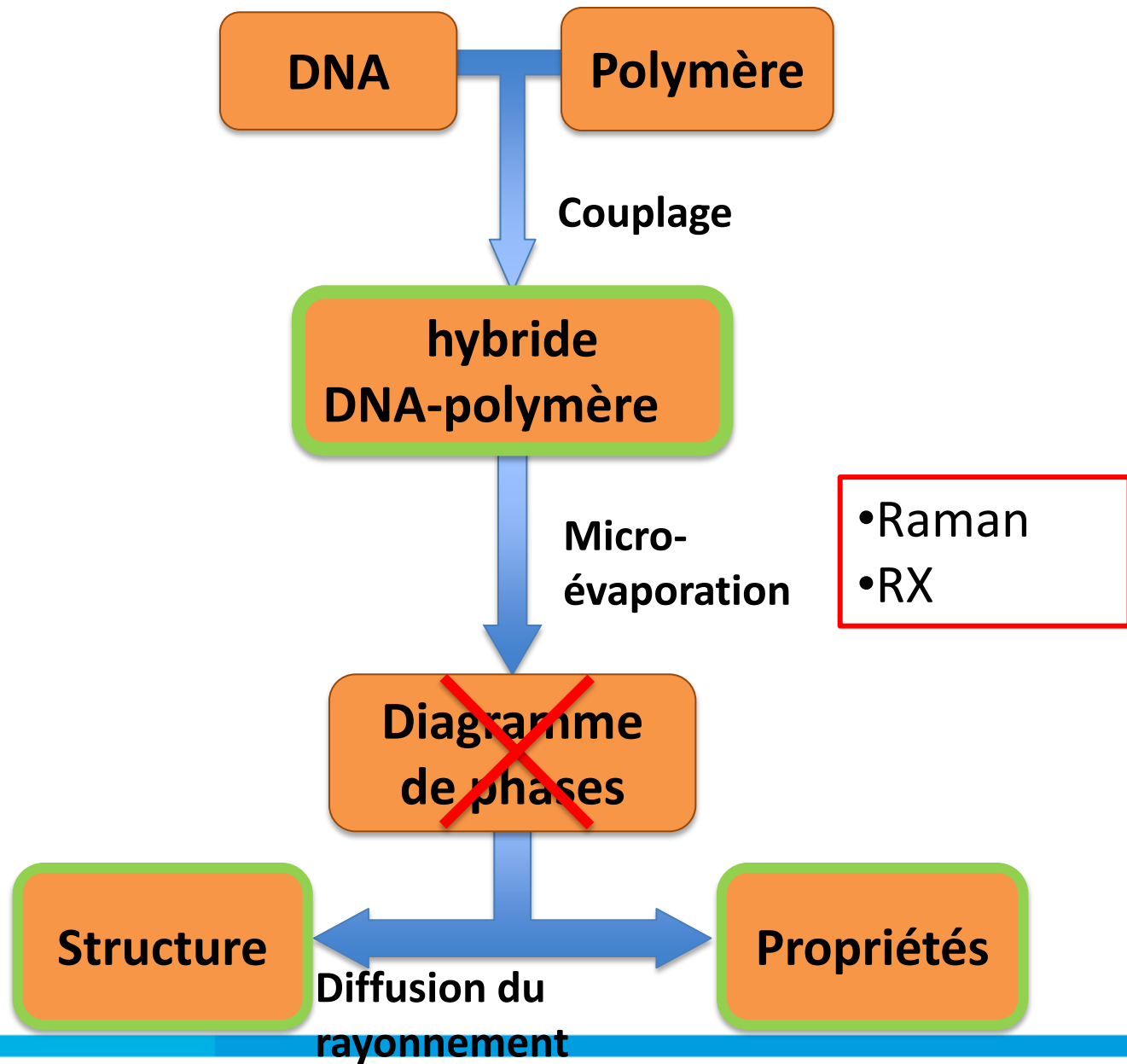


• Transition réversible avec tous les polymères → particularité ADN

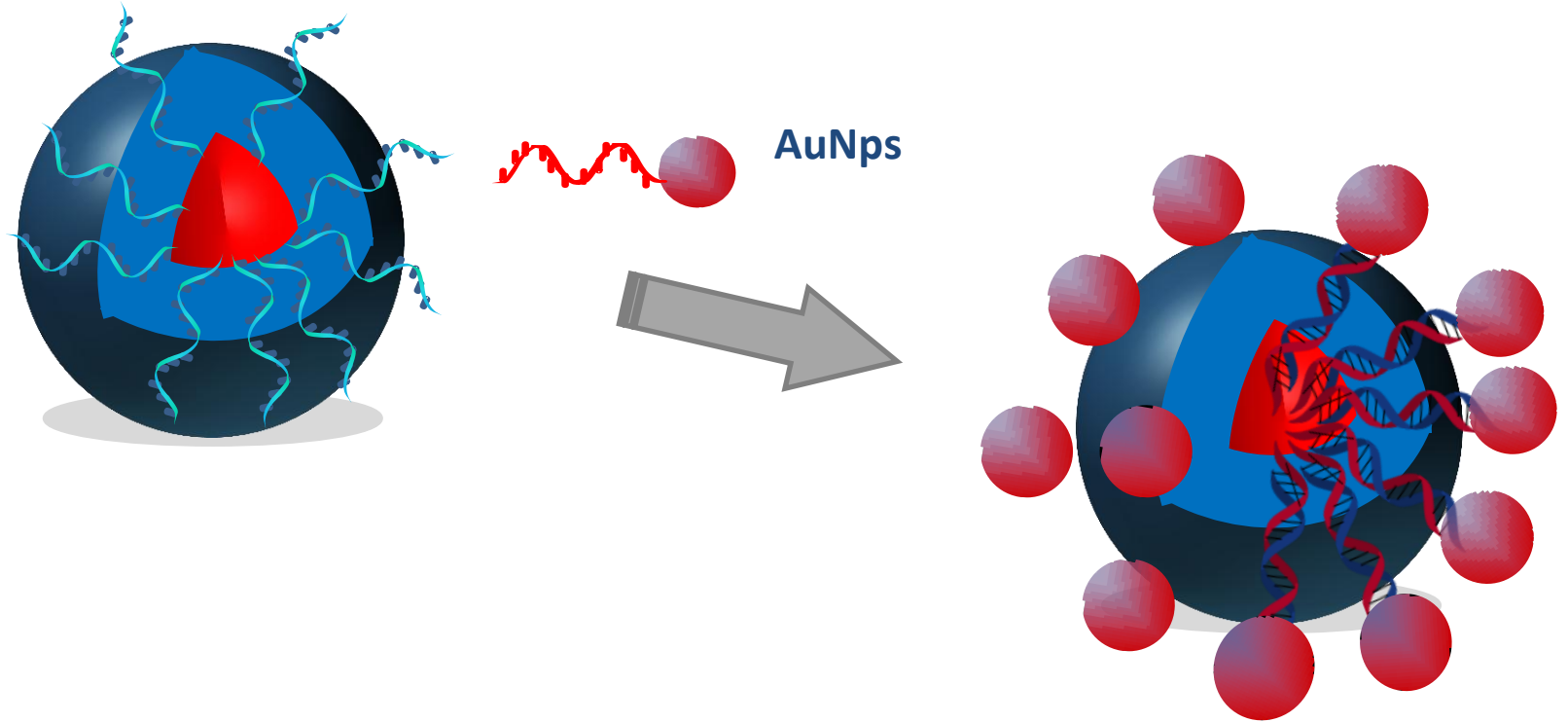
# Comportement thermo responsif



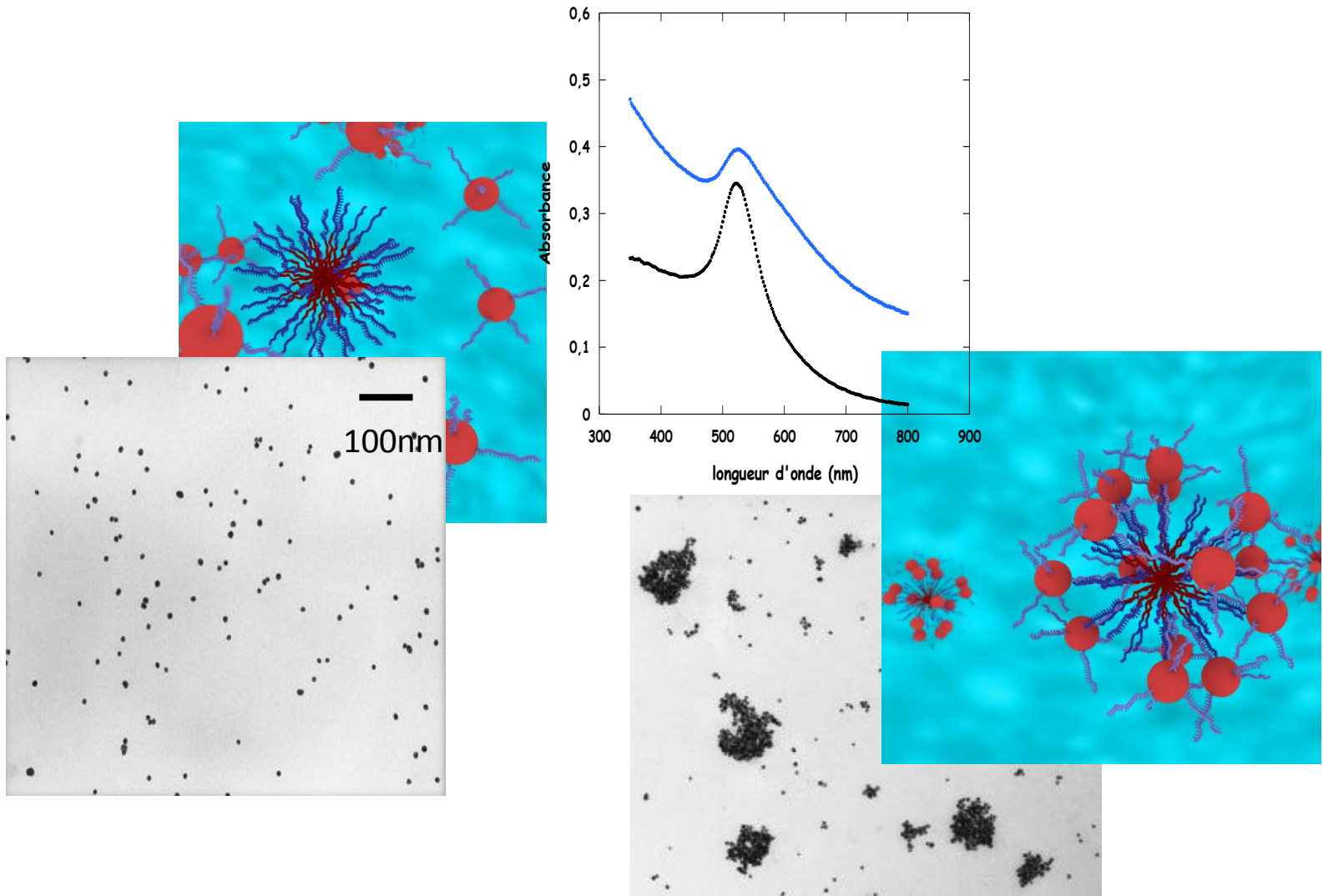
➔ Thermo-responsive behavior



# Perspective 1: assemblages

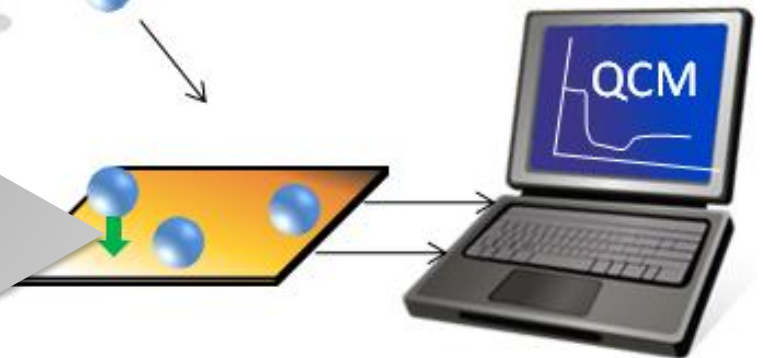
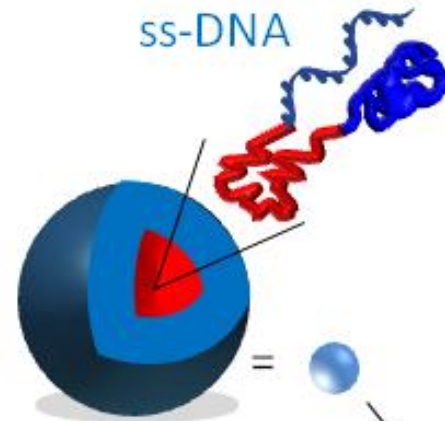


# Perspective 1: assemblages



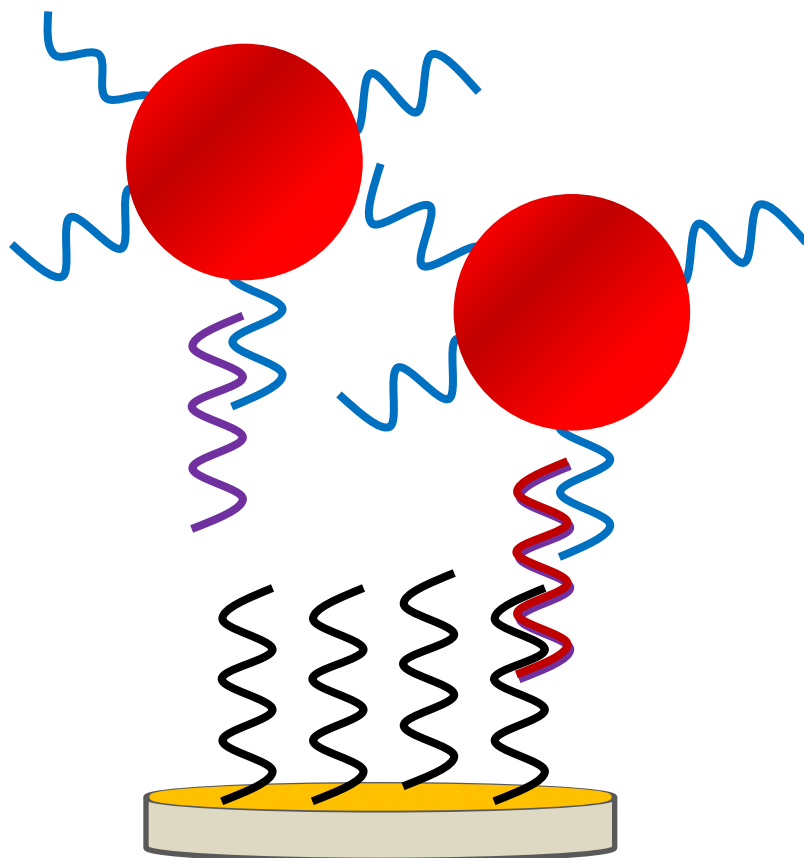
# Perspective 2: capteurs

## Aptamer



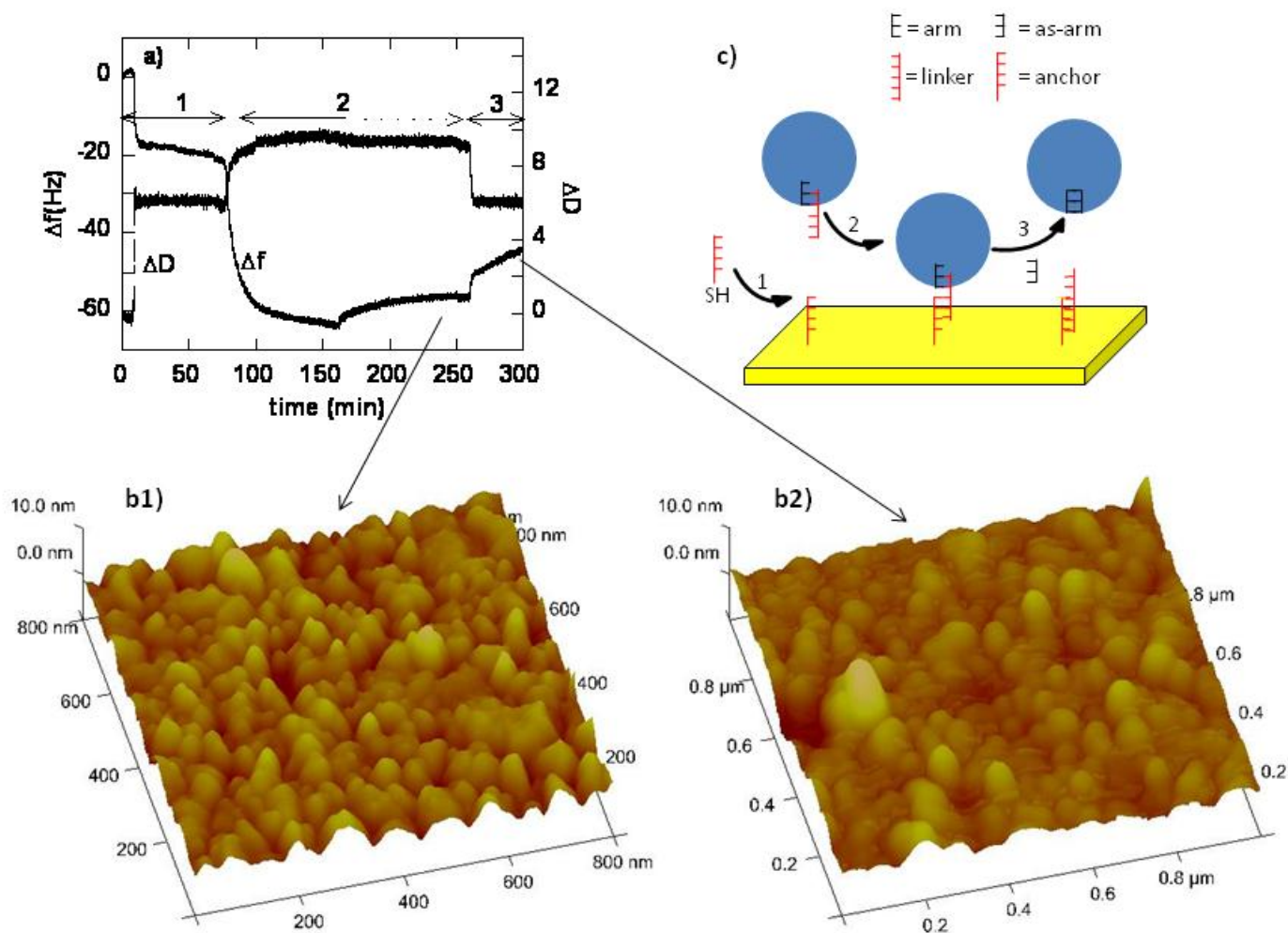
<http://www.molgen.mpg.de/~nabt/background.html>

## Perspective 2: capteurs



Mayap Talom, R. et al. *Chemistry*, (2011)

# Perspective 2: capteurs



Mayap Talom, R. et al. *Chemistry*, (2011)

synthèse

- Synthèse d'une bibliothèque de diblocs +1 triblock pegylé

Microfluidique

- Implémentation Raman
- RX

caractérisation

- Structure (size), Stabilité
- Propriétés spécifique bloc ADN

ADN-techno

- détection acoustique
- Assemblages nanoparticules d'or

Projet FAMOUS

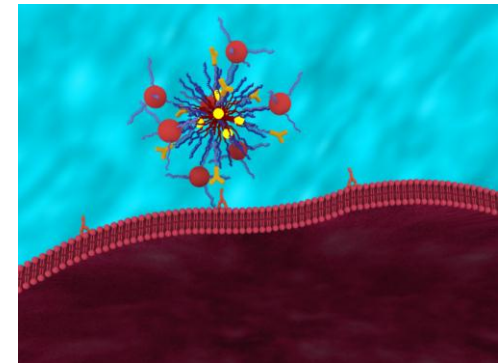
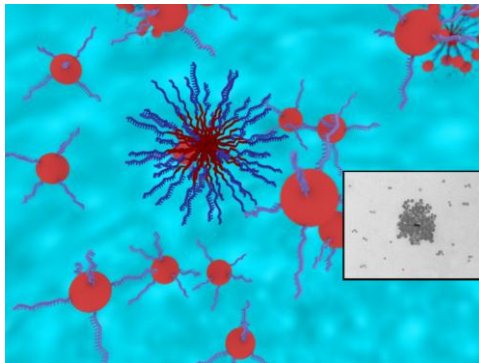
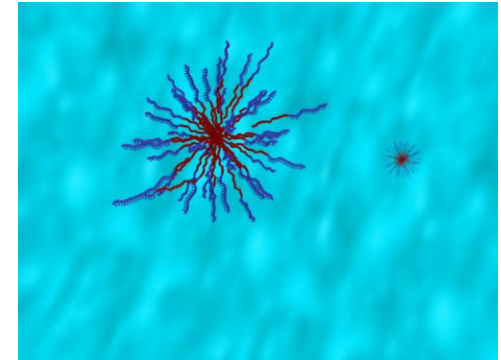
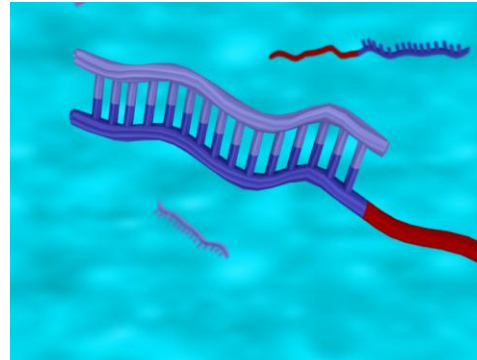
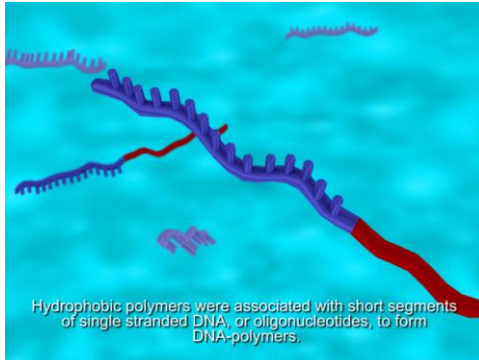
- Développement d'un capteur à base d'apta micelles pour la détection d'acides biliaires

# Production Scientifique

- 9 Publications/ +15 communications orales dont 3 invitées
  1. *Investigation of the reversibility of the unimer-to-aggregate transition in block copolymers by surface tension-measurements* **JCIS**, 387,180-186, **2012**
  2. *Confined drying of a complex fluid drop: phase diagram, activity, and mutual diffusion coefficient* **SOFT MATTER** , 8, 5923-5932, **2012**
  3. *Biohybrid Block Copolymers: toward Functional Micelles and Vesicles* **Chem. Soc. Rev.**, **2011**, 40
  4. *DNA–Polymer Micelles as Nanoparticles with Recognition Ability* **Chemistry - A European Journal**, 10.1002/chem.201101561, **2011**
  5. *Application of microevaporators to dynamic exploration of the phase diagram* **J. App. Phys.** 107, 084905, **2010**
  6. *Time-resolved microfocused small angle X-ray scattering investigation of the microfluidic concentration of charged nanoparticles* **European Physical Journal**, 34, 58, **2011**
  7. *Micro-evaporators with accumulators for the screening of aqueous phase diagrams* **App. Phys. Lett.** 95, 033108, **2009**
  8. *Microfluidics for kinetic inspection of phase diagrams* **C.R. Chimie**, 12, 258, **2009**
  9. *Microfluidic crystallization* **Lab Chip**, 9, 24, **2009**

# Production Scientifique

- Thèse de R. Mayap Talom soutenue en oct 2011.
- Réalisation film animation « grand public » (Gdr or nano)



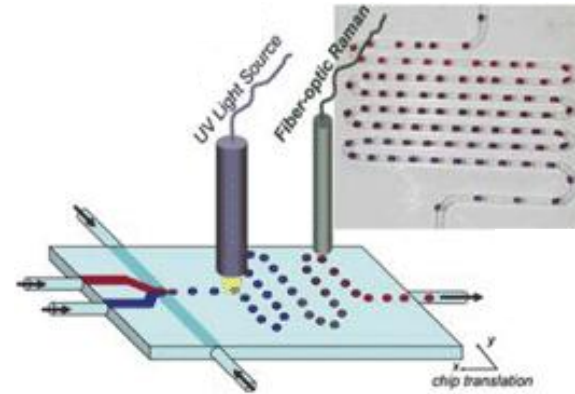
Présenté à Poitiers, Exposition temporaire : L'or, de l'alchimie aux nanotechnologies, 27 février - 27 mai **2012**, Poitiers.

<http://blogperso.univ-rennes1.fr/fabienne.gauffre/index.php/>

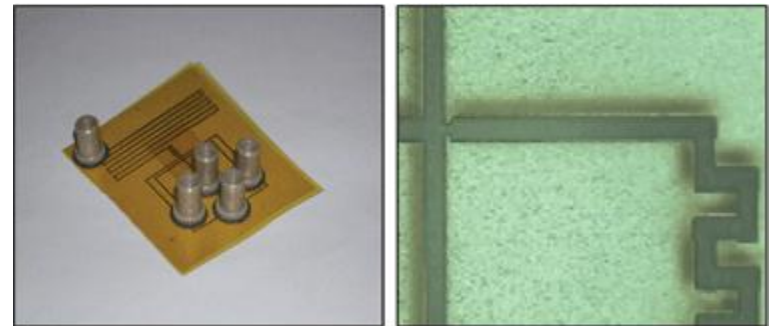
# Méthodes d'analyse

*in situ* au format micro

- › Fluorescence / optique / lumière
- › Spectroscopies UV / Vis / IR / Raman  
µscope Raman confocal : volume sondé  $\frac{1}{4} \mu\text{m}^3$



- › rayons x  
nécessite des matériaux compatibles  
Ray Barrett (ESRF), Jean-Baptiste Salmon (LOF)



- › RMN embarquée

