FORMATION AND DETECTION OF ARTIFICIAL LIPID BILAYERS ON OPTICAL FIBRES

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INTRODUCTION

The understanding of cellular membranes and transport of biological species through them is of crucial importance in life sciences. In practice, **artificial lipid bilayers**, mimicking cellular membranes, are created to reach the aforementioned goals. A convenient way to produce them is the **Droplet Interface Bilayer** (DIB) technique [1], in which two droplets, each of them surrounded by a monolayer of phospholipids, are approached closed enough. In our device, **optical fibres** are used as a support for those droplets [2] and facilitate **fluorescence measurements**. Our set up will be validated using a fluorescent **Sodium Indicator** which is sensitive to the transport of Sodium ions across the artificial bilayer. We first check the influence of Sodium ions concentration on the fluorescence intensity in cuvettes and on our device made of optical fibres. Then, we form artificial lipid bilayers using two fibres configurations to determine which one is the most relevant for membranes stability.

METHODS

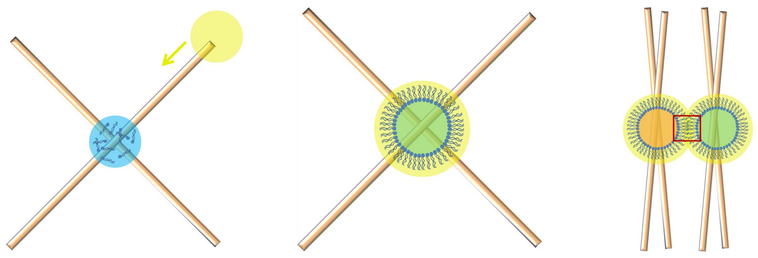


Fig. 1 Sketches of the method used to create DIB on optical fibres.

Fig. 1 shows the method used to create DIB :

- An aqueous droplet with phospholipids is immobilized at the node between two fibres.
- A droplet of oil encapsulate the water droplet.

  → Formation of a lipid monolayer.
- Two sets of fibres with lipid monolayer are approached close enough.

  → **Formation of an artificial lipid bilayer.**

OPTICAL DEVICE

Our device is represented in Fig. 2. It allows us to perform **fluorescent measurements** on the droplets with the optical fibres.

Synchronous detection is used to increase the sensibility.

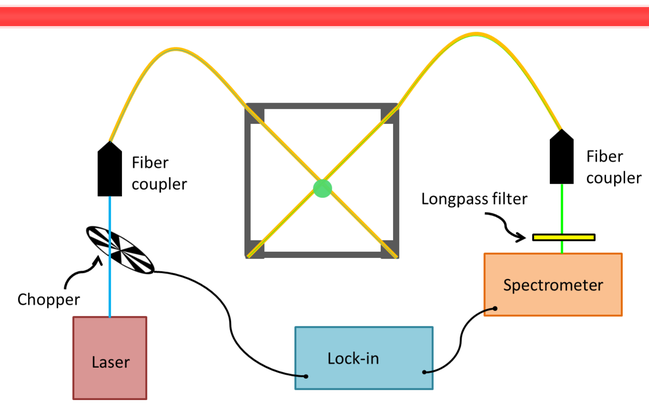


Fig. 2 Optical device use for fluorescence measurements with optical fibres.

Detection of the DIB formation is shown in Fig. 3 :

- Sodium ions and a pore forming protein are in one aqueous droplet.
- Fluorescent **sodium indicator**, Sodium Green [3] (see Fig. 4), is in the other droplet.
- Transport of Sodium through the membrane indicates the formation of the lipid bilayer. It is detected by fluorescent measurement via the optical fibres.

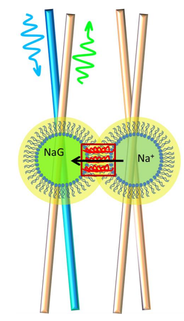


Fig. 3 Detection of the formation of the membrane using a sodium probe and a pore forming protein.

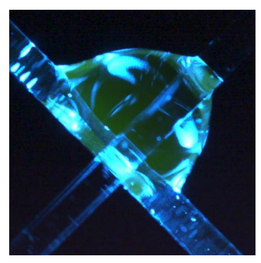
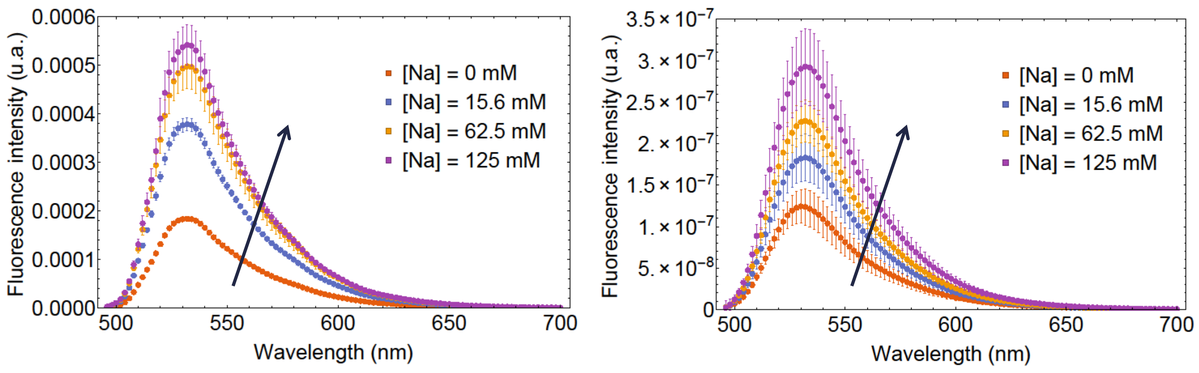


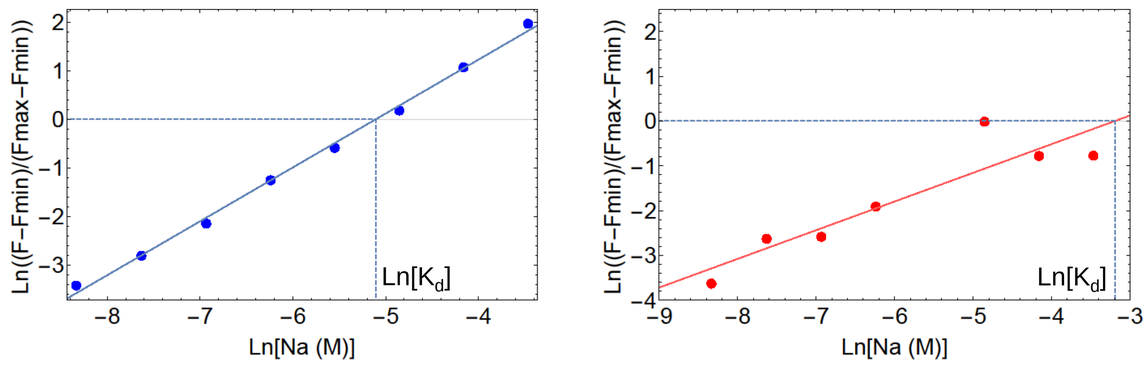
Fig. 4 A droplet of Sodium Green at the node between two optical fibres.

RESULTS

Fluorescence intensity of **Sodium Green** as a function of **[Na⁺]** in cuvettes (left) and on fibres (right)



Dissociation constant (K_d) of Sodium Green for cuvettes (left) and for fibres (right)

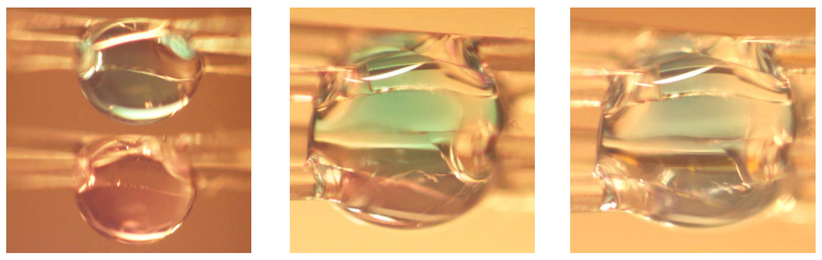


Cuvettes : K_d = 6 mM.

Fibres : K_d = 40 mM.

Results on fibres are similar but less accurate than in cuvettes.

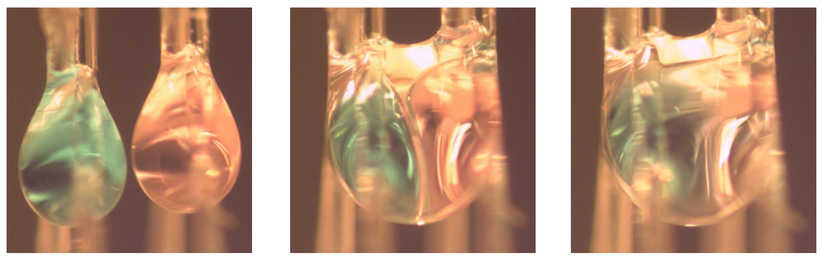
Formation of DIB on **horizontal** fibres vs on **vertical** fibres



t = 0 t = 30 min t = 7h15

Fig. 5 DIB on horizontal fibres.

Lifetime = 7h00



t = 0 t = 5 min t = 1h30

Fig. 6 DIB on vertical fibres.

Lifetime = 1h20

Droplet interface bilayers are created with both configurations but **last longer on horizontal fibres.**

CONCLUSION AND FUTURE WORKS

- A new device for artificial lipid bilayers fabrication that allows fluorescent measurements is presented.
- With an horizontal fibres configuration, the Droplet Interface Bilayers last several hours.
- With our device made of optical fibres, the variation of Sodium Green fluorescence with Na^+ concentration is similar to the one obtain in cuvettes.

In the future :

- We will confirm the formation of the DIB with the detection of the transport of Na^+ using Sodium Green.
- We will study the evaporation of the droplets which could affect fluorescence measurements and DIB lifetime.

REFERENCES

[1] K. Funakoshi, H. Suzuki and S. Takeuchi, Anal. Chem., 78, 8169-8174 (2006)

[2] M. Lismont, N. Vandewalle, B. Joris, L. Dreesen, Appl. Phys. Lett. 150, 133701-133705 (2014)

[3] Molecular Probes, *Sodium Green* (2001)

ACKNOWLEDGMENTS

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