Integrated capacitive biosensors based on filamentous phages

D. Mercier [a], R. Santoro [a], G. Deschuyteneer [c], I. Sbille [a], X. Tang [b], L. Moreno [b], D. Flandre [b], P. Soumillion [c], and J. Proost [a]

Université catholique de Louvain, 1348 Louvain-la-Neuve, Belgium **joris.proost@uclouvain.be**

[a] *Division of Materials and Process Engineering (IMAP/iMMC) – Place Sainte Barbe 2*

[b] *Information and Communication Technologies, Electronics and Applied Mathematics (ICTEAM) – Place du Levant 3*

[c] *Groupe de Biochimie et Génétique Moléculaire Bactérienne (BGMB/ISV) – Place Croix du Sud 5*

Abstract

• In this work, we report on the succesfull selection and identification of peptide motives that exhibit a specific affinity to anodic alumina surfaces when multivalently displayed on a filamentous phage.

• It was also demonstrated that, for a selected phage clone, a chemical functionalisation (biotinylation) of the bacteriophage does not deteriorate its specific affinity to anodic alumina. Such biotinylated bacteriophages, after being immobilised onto an alumina surface, also allow for the quantitative detection of streptavidine using an ELISA protocol.

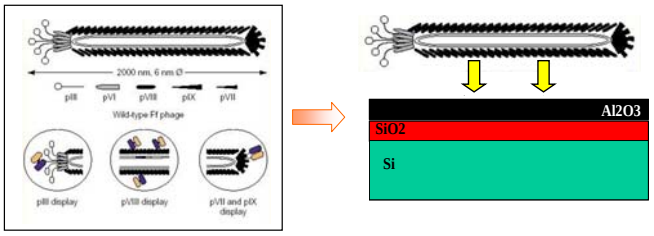
• When immobilising selected filamentous phages on interdigitated Al/Al_2O_3 electrodes, a characteristic change of the capacitance signal with frequency is observed.

• These results are believed to pave the way for shifting the surface design of integrated biosensing devices from traditional, chemically modified synthetic surfaces towards molecular linkers based on genetically engineered polypeptides.

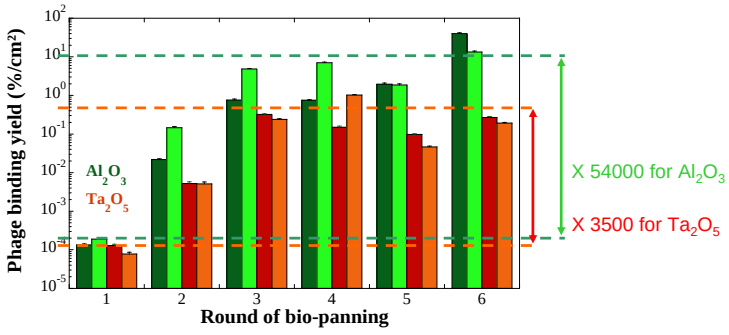
Substrates and phages

• 150 nm thick dense Al_2O_3[1] and Ta_2O_5 obtained by anodising of Al and Ta thin films

• Filamentous M13 phage library[2] with octapeptide pVIII-display[3]



Results : (1) Biopanning rounds



(2) Identification of selected peptide sequences

Amino acid sequences								Phage binding yield* Al_2O_3 10^{-2} %/cm²	Blank 10^{-2} %	ratio $\frac{Al_2O_3}{blank}$	Phage binding yield Ta_2O_5 10^{-2} %/cm²	Blank 10^{-2} %	ratio $\frac{Ta_2O_5}{blank}$
D	P	S	K	P	G	S	S	5.02	0.03	193	0.44	0.03	17
E	P	S	K	A	A	G	T	142.15	0.10	1457	17.50	0.10	176
E	N	T	P	R	G	V	Q	109.46	0.12	921	2.00	0.12	17

* Phage input : 1.5 10^{12}

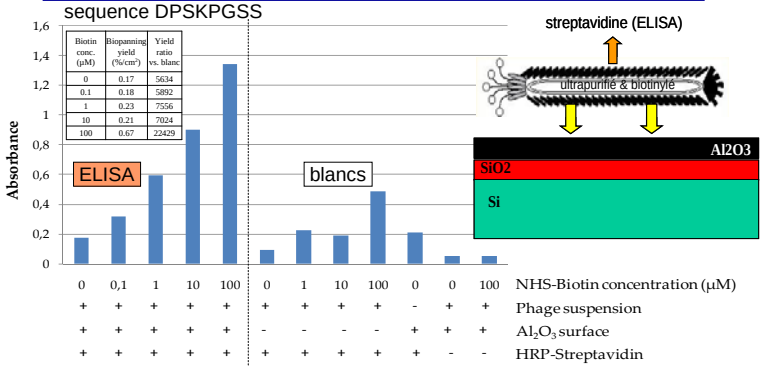
⇒ Specific affinity more important towards Al_2O_3 (PZC ≅ 8-9) than Ta_2O_5 (PZC ≅ -0.7)

E	P	P	N	K	P	G	T	0.48	0.20	2.4
D	P	A	A	R	T	Q	V	11.68	8	1.5
E	P	V	Q	K	A	G	S	65.58	34.22	1.9
E	P	I	K	G	G	G	S	110.12	33.75	3.3

Sequences with much less affinity to Al_2O_3

polar charged +
polar charged -
polar non charged (with OH group)
non polar

(3) Phage biotinylation and streptavidine recognition

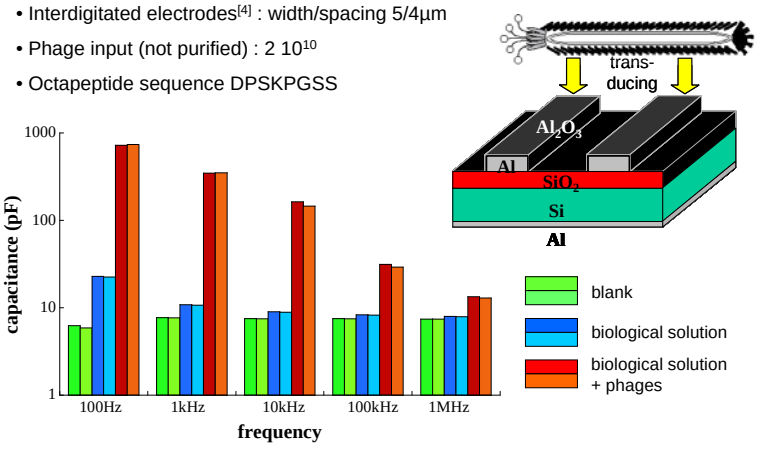


(4) Capacitance measurements on Al_2O_3 electrodes

• Interdigitated electrodes[4] : width/spacing 5/4µm

• Phage input (not purified) : 2 10^{10}

• Octapeptide sequence DPSKPGSS



References

[1] Van Overmeere Q., Nysten B., Proost J., Appl. Phys. Lett. 94 (2009) 074103.
[2] Kriplani U., Kay B.K., Current Opinion in Biotechnology 16 (2005) 470.
[3] Petrenko V.A., Sraith G.P., Gong X., Quinn T., Protein Engineering 9 (1996) 667.
[4] Moreno-Hagelsieb L., Lobert P.E., Pampin R., Bourgeois D., Remacle J., Flandre D., Sensors and Actuators B 98 (2004) 269.

Acknowledgements :
Financial support of the Région Wallonne (Programme d'Excellence **NANOTIC**) is gratefully acknowledged.

Valorisation potential

• use concept of specific phage affinity to (anodic) metallic oxides for bio (-sensing) applications

• J. Proost, R. Santoro, G. Deschuyteneer, P. Soumillion, D. Flandre, "*Genetically modified bacteriophage, biosensor containing same, and method of use*", International Patent Application No. PCT/EP2010/057279.