Ethanol detection with Langmuir-Blodgett films containing alcohol dehydrogenase immobilized in phospholipid matrices

A. C. Perinotto[1], L. Caseli[1,2], V. Zucolotto[1], T. Viitala.[3], O. N. Oliveira Jr[1]
[1]*Instituto de Física de São Carlos, Universidade de São Paulo,Campus São Carlos, SP, Brazil*
[2]*Universidade Federal de São Paulo, Campus Diadema, SP, Brazil*
[3] *KSV Instruments, Helsinki, Finland*

The selective determination of ethanol molecules in aqueous solutions is important for several areas including medicine, food industry and forensic science [1,2]. Moreover, the immobilization of enzymes in organized two-dimensional matrices allows for biotechnological applications such as in the building of biosensors [3]. The aim of this study was to investigate the adsorption of alcohol dehydrogenase (ADH) on Langmuir and Langmuir-Blodgett (LB) films, with the latter deposited onto Au-interdigitated electrodes (IDEs) and used in biosensors to detect alcohol. The Langmuir and LB films were fabricated with dimystoyl phosphatidic acid (DMPA), ADH and DMPA+ADH. The adsorption kinetics for ADH at the air-water interface and at a pre-formed DMPA Langmuir film was monitored by surface pressure-time curves and PM-IRRAS spectroscopy, from which we observed that ADH is surface active with or without DMPA. The formation of LB films onto Au-substrates was monitored with a quartz crystal microbalance, while fluorescence spectroscopy was used to study adsorption onto quartz substrates. The detection of ethanol was carried out with electrical capacitance measurements, whose data were treated with Principal Components Analysis (PCA). It was possible to distinguish ethanol in buffer solutions diluted from 10^{-3} v/v down to 10^{-8} v/v, and to distinguish ethanol from ascorbic acid or uric acid at 0.2 mM. It has been therefore demonstrated the suitability of using phospholipid LB films as matrix for ADH, which can be used in sensing ethanol.

Keywords: Langmuir, Langmuir-Blodgett, alcohol dehydrogenase, biosensor.

Work supported by CNPq and FAPESP.

[1] J. K. Park, H. J. Yee, K. S. Lee, W. Y. Lee, M. C. Shin, T. H. Kim, S. R. Kim, Anal. Chim. Acta **390**, 83 (1999).
[2] G. G. Guilbault, G. Palleschi, G. Lubrano, Biosens. Bioelectron. **10**, 379 (1995)
[3] L. Caseli, F. N. Crespilho, T. M. Nobre, M. E. D. Zaniquelli, V. Zucolotto, O. N. Oliveira Jr., Journal of Colloid and Interface Science **319**, 100 (2008)

e-mail angelo@ifsc.usp.br; Grupo de Polímeros Bernhard Gross – IFSC-USP – Av. Trabalhador Sãocarlense, 400, 13560-970 – São Carlos-SP, Brazil.