BIOSENSORS BASED ON BIO-NANOSTRUCTURES IMMOBILIZED ON SILICON CHIPS.

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Biochips, devices with biological macromolecules located in spatially-defined areas, play an increasingly important role in the molecular and medical biosciences. Fluorescence is the dominant method for array interrogation but requires a label to be added to one of the components, bulky and expensive instrumentation and sophisticated software for data interpretation. Recently attention has been focused on electronic and electrochemical sensors based on self-assembled molecules on surfaces, because they present the sensitivity of fluorescence-based systems but being cheaper, more compact and potentially allowing near instant diagnostics at the point of use [1]. One of the most studied systems as biosensor is the self-assembled functionalized monolayer of alkanethiols on gold [2]. It is clear, however, that deserve importance to construct ordered molecular layers on semiconductor surfaces, mainly on Si, because the wide range of possible applications in conjunction with the advanced silicon technology.

In this context, we have been working in an easy and widely applicable method for the attachment of different nano-objects [3] (nucleic-acids, enzymes, proteins, nanoparticles…) to semiconducting silicon wafers. The chemistry involves the reaction of Si-H layer with a bifunctional hydrocarbon containing different alkene chains (alkylation reaction). Since the reaction is irreversible, via the stable Si-C bond [4], and it is not influenced by the functional groups at the other end of the molecule, the resulting monolayer presents an uniform composition where both components are well mixed, despite being of different chain length and tail-group. Moreover the results reveal that the resulting molecular films present a composition that is representative of the composition of the solution. Therefore, by a simple dilution method it is possible to control the mean spacing between reactive groups on the surface and this is an useful method for nanoscale surface design [5].

When alkene and aldehyde terminated chains are mixed, it is possible to specifically localise proteins or nucleic acids, such as the PNA (Peptide Nucleic Acid), to defined surface positions, with little non-specific adsorption at non-reactive sites. Proteins or the peptidic bonds in the PNA structure are immobilized by reaction of surface lysine residues (common to almost every protein) with the aldehyde terminated group of the monolayer, but not on the methyl terminated group.

This protocol opens the way for using semiconducting silicon surfaces for the production of protein or PNA biochips, since all the proteins and the peptide nucleic acids immobilised appear to remain in their functional state. Therefore, it is possible to detect protein-protein interactions, protein-small molecule or PNA_DNA interactions on the surface electronically. This feature is critical in allowing electrochemical/electronic methods to be exploited in

protein or nucleic acids analysis [6]. Such chips would be expected to be of tremendous utility for comparative proteomics and in molecular medicine, drug discovery and diagnostics or even for the detection of mutations of DNA.

References:

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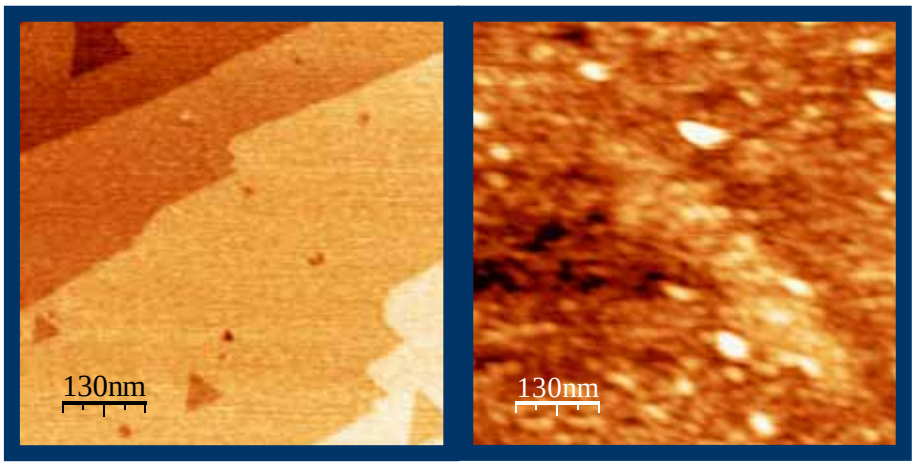

Figure 1: A) AFM images in air of a mixed monolayers 1:1 $C_{11}/C_{10}CHO$, on Si(111). B) Same surface after the immobilization of PNA molecules.