T e c h n i c a l   N o t e s

Electrochemical sensor for nucleic acids revelation

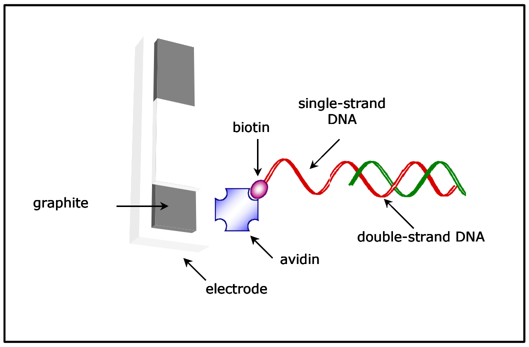

Fig. 1. Schematic diagram of DNA immobilization and hybridization on graphite electrode

Several mutations responsible for numerous inherited human disorders are now known, therefore the specific detection and analysis of DNA sequences and the study of gene polymorphism play more and more a fundamental role in rapid diagnosis of genetic diseases.
Electrochemical detection of DNA hybridization usually involves both monitoring the increased electrochemical response of a redox-active indicator, that recognizes the DNA duplex (indirect method) or probing hybridization-induced changes in the intrinsic signal of nucleic acid (direct method).

Technobiochip's Electrochemical DNA Sensor enables the monitoring of adsorbed DNA species at carbon electrodes using the Potentiometric Stripping Analysis (PSA). The procedure involves the use of an electroactive indicator, daunomycin hydrocloride, which intercalates the double stranded DNA. The single-stranded DNA is immobilized on the graphite electrode surface using the avidin-biotin high affinity interaction. This procedure involves the formation of an avidin layer onto the graphite electrode surface and then the binding of biotinylated DNA (Fig. 1).

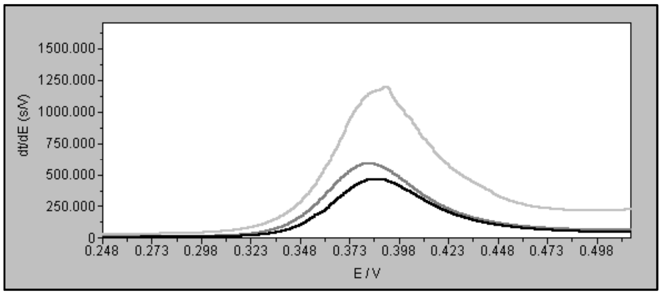

Fig. 2. Sensor response by PSA The black line shows the peak after the probe immobilization. The other lines show the same measurements after: a) interaction with a non-complementary sequence (gray line) and b) hybridization with a complementary sequence (light gray line).

During the stripping process the potential is recorded and processed. The response is plotted as dt/dE vs E and a peak-shaped pattern is obtained. If the target sequence is present in the sample, a hybrid will occur on the electrode surface, but the daunomycin-hybrid complex will appear with a higher peak (Fig. 2).

Technobiochip's Electrochemical sensor has been used for Neuroblastoma genetic mutations detection, the most common solid extracranial cancer in children.

DNA hybridization-based methods, usually start with PCR or other biochemical amplification, whilst our DNA sensor can detect DNA mutations without any previous manipulation step.
This new biosensor strategy has numerous advantages compared with traditional assays for the diagnostic screening, and in particular:

- short time-consuming;
- savings in consumption of reagents, probes and especially samples;
- multi-array features to detect simulteously several mutations;
- label-free detection;
- low-cost assay.

Papers

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Pighini M., Piras L., Fenu S., Longo L., Tonini G.P., Cocco M. (2000). Development of DNA-sensors for the detection of genetic mutations associated with the human cancer presence. *Sensors and microsystems.* Di Natale C., D'Amico A., Dario P.(ed.), p.15-20.

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