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Colloidal chitin-soaked nanoporous carbon nanotube thin films as biocompatible immobilization matrix of amperometric enzyme biosensors with long life-time

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Abstract
Well-working amperometric enzyme biosensors are ideal tools in the (bio-) analytical chemistry sector as they offer the superb selectivity of biological recognition elements together with the simplicity and sensitivity of modern electrochemical equipment.1 Key factor for successful electrochemical enzyme biosensor manufacture is a gentle but leak-protected biocatalyst fixation ('immobilization') on the electrode surface that translates substrate conversion into signal.
Here, a carbon nanotube (CNT)/chitin electrode layer is introduced as nanoporous composite enzyme immobilization matrix, for glucose oxidase (GOx) as model protein and platinum or gold disk electrodes as physicochemical transducers. Motivation behind an integration of the marine biopolymer chitin into the desired biosensor architecture was a gain of matrix biocompatibility and, as a consequence, a good GOx survival and related analytical response stability. The target biosensor design was actually realized via simple drop/dry coating steps, namely via serial load of the electrode disks with µL droplets of (1) a water suspension of CNT, (2) a water solution of GOx and (3) an aqueous colloidal chitin suspension, with solvent evaporation allowed after droplet placement. Optionally, a thin epoxy-based polymeric top-coat was placed as extra barrier against enzyme loss; it was obtained via application of commercial cathodic electrodeposition paint (EDP).
Repeated conventional amperometric calibration runs confirmed for the CNT/GOx/Chitin/EDP glucose biosensors a linear range that extended competitively wide up to a few tens of mM and, with sensors in between trials stored in phosphate buffersolutions of pH 7.0and at 4℃, signal stability was observedfor periods up to weeks. Successful with close to ideal recovery rates werequantitative assessments of spiked model samples and continuous biosensor use in a flow-based three-electrode electrochemical cell with scheduled on-line glucose calibration measurements during uninterrupted flow cell operation. Apparently, electrode surface-immobilized GOx entities were in the CNT/chitin environment kept healthily in place and thus able to maintain their pronounced bio-catalytic activity for long.Possible was as a resultan electrochemical biosensing service on an extended time scale for glucose quantifications, with adequate analytical figures of merit for analyte quantifications.

Keywords: Biosensors,amperometry, carbon nanotubes, chitin, enzyme immobilization

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