

isolated from nitrate-grown cells of *Desulfovibrio desulfuricans* (Dd) ATCC 27774 catalyses the six-electron reduction of nitrite to ammonia. Previous electrochemical studies demonstrated that a simple electrocatalytic mechanism can be applied to this system. Its substrate specificity, availability and stability under ambient conditions makes this enzymatic system a promising candidate for use in a biosensor device. An electrochemical study of gel-immobilized Dd NiR on a glassy carbon electrode revealed both enzymatic activity and amperometric response to nitrite. In this study it was observed that the catalytic current density is a function of the nitrite concentration in solution and follows a characteristic Michaelis-Menten-type substrate dependence. Such a biosensor device (NiR-electrode) bears the option to be used for analytical determination of nitrite in complex media.

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China - Novel biosensor system

In ENZYME MICROB. TECHNOL. (17/5 (413-417)1995) S. Ya, D. Leu, K. Ge, K. Chen and L. Nice of Hunan University report on "A novel glutamine biosensor system based on a conductance-surface acoustic wave frequency response".

A new type of glutamine biosensor system based on a conductance-surface acoustic wave (SAW) frequency response, in which a SAW resonator oscillating at 61 MHz and a biosensor involving kidney cortex tissue or *Escherichia coliform* and a pair of parallel electrodes were used, was developed for the determination of glutamine. Glutaminase from porcine kidney or *Escherichia coliform* was used as a biocatalyst of the hydrolysis reaction of glutamine. There was an excellent linear relationship between the frequency shift response and the glutamine concentration in the range of 6.8×10^{-4} M to 6.8×10^{-3} M. The SAW frequency responses obtained from the proposed biosensor system were compared with the

conductance response. Some other factors such as pH and selectivity are also discussed.

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Ireland - Biosensor from *Thermus aquaticus*

In ENZYME MICROB. TECHNOL. (17/5 (472-476) 1995) D. Compagnone, C.J. McNeil, D. Athey, C. Di Ilio and G.G. Guilbault of University College Cork report on "An amperometric NADH biosensor based on NADH oxidase from *Thermus aquaticus*".

A biosensor for the determination of the reduced cofactor nicotinamide adenine dinucleotide (NADH) has been developed using the enzyme NADH oxidase from *Thermus aquaticus* immobilized on an Immobilon AV membrane. A hydrogen peroxide electrode was used as the detection system. The NADH electrode showed a monotonous response with near linearity in the 5×10^{-7} to 2×10^{-5} M range, with a detection limit of 2×10^{-7} M. Analytical parameters such as pH, response time and lifetime were characterized. The probe showed no change in sensitivity between pH 4.5 and 9.5 and retained 70% of its initial activity after 50 days of storage in buffer. A method for the measurement of lactic dehydrogenase (LDH) activity was developed using the biosensor. The response was linear in the 10-1000 U l⁻¹ range. The addition of NADH and LDH to serum gave recovery values between 94 and 107%. The determination of LDH in two control sera and in three serum samples was performed with a standard spectrophotometric procedure and the biosensor. The results correlated well.

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China - Human urea and pumpkin seeds!

In ANAL. CHIM. ACTA (307/1 (61-69)1995) D. Liu, K. Ge, K. Chen, L. Nie and S. Yao of Hunan University report on "Clinical analysis of urea in human blood by coupling a surface acoustic wave sensor with urease extracted from pumpkin seeds".

A surface acoustic wave (SAW) urease sensor system was developed in which a SAW resonator

oscillating at 61 MHz and a pair of parallel electrodes were utilized, by coupling with urease extracted from pumpkin seeds. The Michaelis constant and the maximum reaction rate of urease were estimated as 2.08×10^{-3} mol/l and 8.85 kHz/min, respectively, at pH 7.5 and 25.0°C. The analytical characteristics of the urease sensor system, including influences of pH, temperature on frequency response, calibration curves, reproducibility, selectivity over enzymatic interferences were determined. The recovery of the sensor system ranged from 96 to 105% and the experimental detection limit of urea was 0.50 µg/ml (i.e., 8.3×10^{-6} M, S/N = 3). The SAW urease sensor system possesses the characteristics of good reproducibility and specificity. It has been successfully applied to the analysis of urea in human blood samples. The assay results are consistent with the results tested by conventional colorimetry and also support clinical diagnosis.

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USA - 'Its the real thing' biosensor

In ANAL. BIOCHEM. (227/1 (216-224) 1995) P.J. Devine, N.A. Anis, J. Wright, S. Kim, A.T. Eldefrawi and M.E. Eldefrawi of Univ. of Maryland School of Medicine report on "A fiber-optic cocaine biosensor".

A fiber-optic biosensor was developed for detection of cocaine, its metabolites, and other coca alkaloids, using a monoclonal antibody (mAb) against a derivatized benzoylecgonine (BE). The mAb was immobilized noncovalently on quartz fibres and a flow fluorometer was used to detect changes in evanescent wave fluorescence. A fluorescein (FL) conjugate of BE bound to the mAb specifically in a saturable manner and with high affinity ($K(d) = 7.6$ nM). Cocaine or other test compounds competed with FL-BE for binding to the mAb in a concentration-dependent manner, thereby reducing the initial rate or steady-state fluorescence. Addition of cocaine to the flow buffer after reaching steady-state fluorescence enhanced the dissociation of bound FL-BE, and cocaine

removal allowed fiber regeneration for multiple measurements. The detection limits for cocaine, cocaethylene, norcocaine, and BE were 5, 5, 29, and 30 ng/ml, respectively, but for ecgonine it was 4600 ng/ml and for methylecgonine it was 2000 ng/ml. Tropacocaine was detected at 10 ng/ml, but atropine was detected at 2900 ng/ml. The biosensor discriminated by 833-fold between cocaine and its stereoisomer pseudococaine. Structural features necessary for high-affinity recognition by this mAb are benzoate and 3β configuration, both of which are found in BE, cocaine, norcocaine, and cocaethylene.

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USA - Immobilization techniques for biosensor design

In TRENDS BIOTECHNOL. (13/5 (178-185)1995) W.H. Scouten, J.H.T. Luong and R.S. Brown of Utah State University report on "Enzyme or protein immobilization techniques for applications in biosensor design".

New generations of biosensors are emerging that are based on novel and promising transducers such as miniature, reagentless-mediated electrodes, field-effect transistors, piezoelectric and optical devices. Reagentless-mediated biosensors can be constructed by co-immobilizing both enzymes and mediators onto a miniaturized electrode using electropolymerization, thus improving the sensitivity and speed of the response. Even more promising is the development of electrochemical sensors, in which electron transfer is made directly from a redox enzyme to an electrode surface via molecular wires. While this has only been reported, so far, for a specific enzyme entrapped in N-methylpyrrole under defined circumstances, the development of new oriented immobilization techniques, coupled with progress in protein engineering, may make direct electron transfer the rule rather than the exception.

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