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Electrochemical biosensors: Towards point-of-care cancer diagnostics

Joseph Wang *

Department of Chemical & Materials Engineering and Chemistry and Biodesign Institute, Arizona State University, Tempe, AZ 85287-5801, USA

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Abstract

Wide-scale point-of-care diagnostic systems hold great promise for early detection of cancer at a curable stage of the disease. This review discusses the prospects and challenges of electrochemical biosensors for next-generation cancer diagnostics. Electrochemical biosensors have played an important significant role in the transition towards point-of-care diagnostic devices. Such electrical devices are extremely useful for delivering the diagnostic information in a fast, simple, and low cost fashion in connection to compact (hand-held) analyzers. Modern electrochemical bioaffinity sensors, such as DNA- or immunosensors, offer remarkable sensitivity essential for early cancer detection. The coupling of electrochemical devices with nanoscale materials offers a unique multiplexing capability for simultaneous measurements of multiple cancer markers. The attractive properties of electrochemical devices are extremely promising for improving the efficiency of cancer diagnostics and therapy monitoring. With further development and resources, such portable devices are expected to speed up the diagnosis of cancer, making analytical results available at patient bedside or physician office within few minutes.

Keywords: Biosensors; Cancer; Electrochemical detection; Genetic testing; Immunosensors; Tumor markers; Nanomaterials

1. Introduction

1.1. Towards point-of-care cancer diagnostics

Early diagnosis of cancer is crucial for the successful treatment of the disease. Highly sensitive methods are urgently needed for measuring cancer diagnosis markers present at ultra-low levels during early stages of the disease. Such methods should facilitate early detection and an adequate selection of the treatment of diseases and should lead to increased patient survival rates. Existing diagnostic tests (e.g., ELISA) are not sensitive enough and detect proteins at levels corresponding to advanced stages of the disease. Smaller, faster, and cheaper (one-step) devices are highly desired for replacing time-consuming laboratory-analyses. Making analytical results available at patient bedside within few minutes will greatly improve the monitoring of cancer progress and patient therapy.

Advances in molecular biology have led to a much understanding of potential biomarkers that can be used for cancer diagnosis. The realization of point-of-care cancer diagnostics thus requires proper attention to the major challenge of multi-target detection. Arrays of biosensors, detecting protein signature patterns or multiple DNA mutations, can be used to help screening and guide treatment.

Innovative biosensor strategies would allow cancer testing to be performed more rapidly, inexpensively, and reliably in a decentralized setting. In this review article I will discuss the use of electrochemical biosensors for decentralized clinical testing and the prospects and challenges of using such devices for point-of-care cancer diagnostics.

2. Why electrochemical biosensors?

Over the past three decades we have witnessed a tremendous amount of activity in the area of biosensors. Biosensors are small devices employing biochemical molecular recognition properties as the basis for a selective analysis. The major processes involved in any biosensor system are analyte recognition, signal transduction, and readout. Due to their specificity, speed, portability, and low cost, biosensors offer exciting opportunities for numerous decentralized clinical applications, ranging from 'alternative-site' testing (e.g., physician's office),

* Tel.: +1 480 727 0399; fax: +1 480 727 0412.
E-mail address: joseph.wang@asu.edu.

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emergency-room screening, bedside monitoring, or home self testing.

Electrochemical devices have traditionally received the major share of the attention in biosensor development (Turner et al., 1986; Wang, 2000a). Such devices produce a simple, inexpensive and yet accurate and sensitive platform for patient diagnosis. The name electrochemical biosensor is applied to a molecular sensing device which intimately couples a biological recognition element to an electrode transducer. The purpose of the transducer is to convert the biological recognition event into a useful electrical signal. Amperometric and potentiometric transducers are most commonly used in conjunction with electrochemical biosensors. In potentiometric devices, the analytical information is obtained by converting the biorecognition process into a potential signal in connection to the use of ion-selective electrodes (ISE). Amperometric biosensors operate by applying a constant potential and monitoring the current associated with the reduction or oxidation of an electroactive species involved in the recognition process. An amperometric biosensor may be more attractive because of its high sensitivity and wide linear range. Elegant research on new sensing concepts, coupled with numerous technological innovations, has opened the door to widespread clinical applications of amperometric devices (Wang, 1999). The high sensitivity, specificity, simplicity, and inherent miniaturization of modern electrical bioassays permit them to rival the most advanced optical protocols. Such miniaturization allows packing of numerous microscopic electrode transducers onto a small footprint of a biochip device, and hence the design of high-density arrays.

2.1. Electrochemical biosensors for decentralized clinical testing

Electrochemical biosensors have played a major role in the move towards simplified testing, including home-use devices. Indeed, easy-to-use self-testing glucose strips, based on screen-printed enzyme electrodes, coupled to pocket-size amperometric meters, have dominated the $5 billion/year diabetes monitoring market over the past two decades (Newman and Turner, 2005). Such disposable enzyme electrodes generate the analytical information within 5–10 s in connection to 0.5–10 μL fingerstick blood samples. Hand-held battery-operated clinical analyzers, combining different amperometric biosensors and potentiometric ISE (on a single disposable cartridge) have been shown extremely useful for rapid point-of-care measurements of multiple electrolytes and metabolites (Erickson and Wilding, 1993). Hand-held electrochemical devices, with accuracy similar to that of benchtop analyzers, have been developed for bedside blood gas monitoring (Wahr et al., 1996).

While cancer-related assays are more complex than home self testing of glucose, modern electrochemical bioaffinity sensors, such as DNA- or immunosensors, have recently demonstrated great potential for monitoring cancer-related protein markers and DNA mutations. Although still at the basic research stage, it is envisioned that further development should transfer these electronic assays into small test cartridges. Representative examples of these capabilities and opportunities are discussed in the following sections.

3. Electrochemical bioaffinity assays for nucleic acids and proteins

3.1. Electronic detection of DNA hybridization and DNA damage

Wide-scale genetic testing requires the development of fast-responding, highly sensitive and specific, low-cost user-friendly miniaturized analytical devices. Traditional methods for detecting DNA hybridization are too slow and labor intensive. Biosensors offer a promising alternative for simpler, faster, and cheaper DNA assays. Hybridization biosensors commonly rely on the attachment of a single-stranded (ss) oligonucleotide probe onto a transducer surface to recognize – via base pairing – its complimentary target sequence (Wang, 2000b).

Electrochemical devices have received considerable attention in the development of sequence-specific DNA hybridization biosensors (Mikkelsen, 1996; Gooding, 2002). Such devices offer elegant routes for interfacing, at the molecular level, the DNA recognition and signal-transduction events. Electrochemical DNA hybridization biosensors rely on the conversion of the DNA base-pair recognition event into a useful electrical signal (Fig. 1). Such hybridization event is commonly detected via the increased current signal of an electroactive indicator (that preferentially binds to the DNA duplex), in connection to the use of enzyme- or redox labels, or from other hybridization-induced changes in electrochemical parameters (e.g., conductivity or capacitance). Careful consideration must be given to the surface chemistry and related surface-blocking chemistries, as needed for controlling the binding efficiency and for minimizing non-specific adsorption.

Specific mutations in the p53 gene were measured by coupling a highly specific peptide nucleic acid (PNA) recognition layer with chronopotentiometric electronic transduction (Wang et al., 1997). Gao's group reported recently on the ultrasensitive detection of cancer marker genes in mRNA extracted from

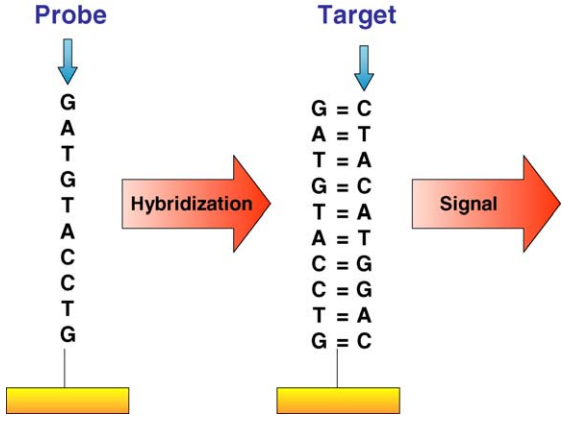

Fig. 1. Steps involved in the detection of a specific DNA sequence using an electrochemical DNA hybridization biosensor [reproduced from Gooding (2002) with permission].

human breast tissues (without a RT-PCR step), based on the catalytic oxidation of the guanine nucleobase by a redox threading intercalator (Tansil et al., 2005). Willner's team developed an amplified detection of telomerase activity, extracted from 1000 HeLa cancer cells, in connection to hybridization of a biotin-labeled oligonucleotide to the telomere repeat units and subsequent binding of avidin-alkaline phosphatase, along with chronopotentiometric signal transduction (Parlov et al., 2004). Chronopotentiometric transduction was also employed for label-free (guanine-based) electrical detection of known mutations related to the breast-cancer genes BRCA1 and BRCA2 (Wang and Kawde, 2000). An amplified guanine signal, obtained by using the electrocatalytic action of a $Ru(bpy)_3^{+2}$ redox mediator, was also employed for the detection of gene expression in mRNA samples from breast tumor samples (Armistead and Thorp, 2002).

Electrochemical sensors have also shown to be extremely useful for detecting small damage to DNA induced by various chemical agents, enzymatic digestion, or ionizing radiation (Fojta, 2002). Such electrical detection of DNA damage reflects the fact that the electrochemical response of DNA is strongly dependent on the DNA structure. In addition to monitoring changes in the intrinsic faradaic (e.g., guanine oxidation) or tensammetric signals of DNA, such studies examine the response of electroactive damaging agents (e.g., carcinogens) interacting with DNA in connection to the use of DNA-modified electrodes. Similar electrodes can be used for the biosensing of anticancer drugs in body fluids (Brett et al., 1996).

3.2. Electrochemical immunoassays and protein arrays

Abnormal concentrations of certain proteins can indicate the presence of various cancers. For the past two decades Heineman's group has developed highly sensitive enzyme electrochemical immunoassays (Ronkainen-Matsuno et al., 2002). Such protocols rely on labeling of the antibody (or antigen) with an enzyme which acts on a substrate and generate an electroactive product that can be detected amperometrically. Enzyme immunosensors can employ competitive or sandwich modes of operation. In addition to enzyme labels, it is possible to use metal markers and redox tags for electronic transduction of antigen–antibody interactions.

The development of electrochemical immunosensors for measuring tumor markers was recently reviewed in this journal (Lin and Ju, 2005). The first electrochemical immunosensor for a cancer marker was developed in the late 1970s (Aizawa et al., 1979). The device involved a competitive assay of hCG (human chorionic gonadotropin) in connection to a catalase label and amperometric monitoring of the enzymatic reaction. Meyerhoff's group reported on a novel sandwich electrochemical enzyme immunoassay for the simultaneous measurement of the PSA (Prostate-Specific Antigen) and hCG protein markers in undiluted whole blood (Meyerhoff et al., 1995). A gold-coated microporous membrane facilitated direct assays, without separation or washing steps. Such simplification holds great promise for the creation of portable point-of-care test systems. Ju's group reported on a reagentless amperometric immunosensor based on direct electrochemistry of the enzyme label horseradish peroxidase for measuring low levels of carcinoma antigen 125 (Dai et al., 2003). Jin's group described a capillary electrophoretic enzyme non-competitive immunoassay with an electrochemical detection for measuring the CA125 tumor marker (He et al., 2003). Microfluidic devices, carrying out such capillary-electrophoretic enzyme immunoassays with amperometric detection on microchip platforms (Wang et al., 2001a), should be extremely attractive for future cancer diagnostics. The integration of additional (sample processing/manipulations) steps onto these 'Lab-on-Chip' devices should facilitate assays of complex biological fluids.

It is well recognized now that simultaneous measurements of combination of tumor markers (i.e., distinct protein patterns) can play a major role in the diagnosis of cancer (Weisner, 2004). Protein signature patterns, reflecting different stages of the disease, can be readily detected using immunosensor chips based on multiple antibody-functionalized transducers. Electrode arrays, having multiple individually addressable electrochemical transducers, can be readily fabricated to meet the requirements of multi-analyte protein arrays. The use of a single enzyme label with such arrays requires proper spatial separation of the individual transducers to eliminate cross talk problems due to diffusion of the enzymatically generated electroactive species from one electrode to the neighboring one. For example, Karube's group designed an electrochemical protein array consisting of 36 platinum working electrodes, with different capture antibodies, and operated in the sandwich enzyme immunoassay mode (Kojima et al., 2003). As indicated in Fig. 2, for the detection of AFP (α-1-fetoprotein) and β_2MG (β_2-microglobulin), no apparent cross reactivity was observed using interelectrode distances of 700 μm. Wilson (2005) described recently a dual-electrode enzyme immunosensor for simultaneous amperometric measurements of the two tumor markers carcinoembryonic antigen (CEA) and α-fetoprotein (AFP). High sensitivity down to the 1 ng/mL level was reported, along with the absence of cross-talk effects. Nucleic acid ligands, known as aptamers, also offer great promise for electrochemical sensing of proteins. Label-free detection of aptamer–protein interactions has been documented recently in connection to electrochemical impedance spectroscopic transduction of the binding event (Rodriguez et al., 2005).

One-dimensional (1D) nanostructures, such as semiconductor- or conducting-polymer nanowires (NW), are extremely attractive for designing high-density protein arrays. Because of their high surface-to-volume ratio and novel electron transport properties, their electronic conductance is strongly influenced by minor surface perturbations (e.g., binding of biomolecules), hence indicating great promise for label-free real-time protein detection. Due to the extreme smallness of these nanomaterials, it is possible to pack a large number of antibody-functionalized NW onto a remarkably small footprint of an array device (Melosh et al., 2003). Such 1D materials thus offer the prospect of massive redundancy in nanosensor arrays and suggest great promise for assays of multiple disease markers in ultrasmall sample volumes. Several studies already indicated the potential of

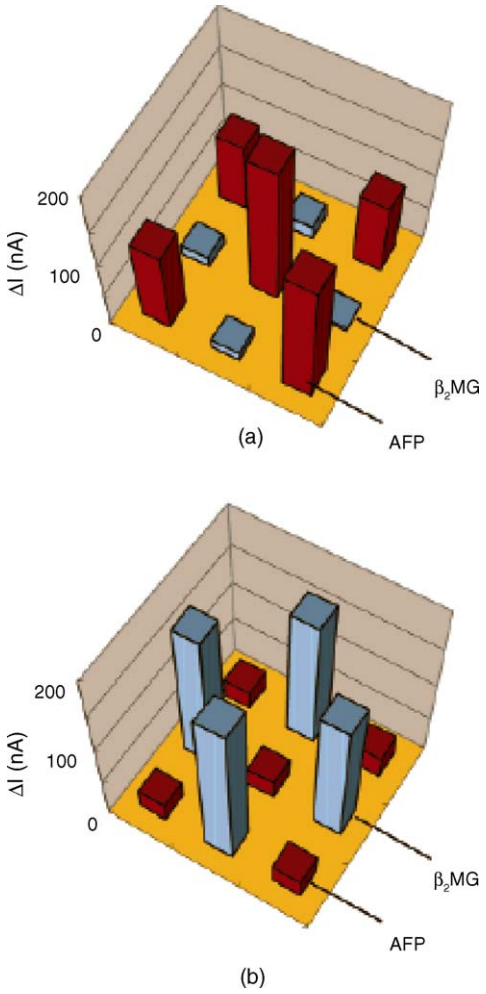

Fig. 2. Multianalyte immunosensing using a multielectrode antibody array. Antibodies for AFP and β_2MG were immobilized at the sites in red and blue and the current signals reflect the oxidation of hydrogen-peroxide product of the enzyme tag [reproduced with from Kojima et al. (2003) with permission].

functionalized NW for highly sensitive real-time biodetection. For example, Lieber's laboratory demonstrated highly sensitive protocols for monitoring DNA hybridization (Hahm and Lieber, 2004) or single viruses (Patolsky et al., 2004) in connection to p-type silicon NW (SiNW) functionalized with PNA probes or antibodies for influenza, respectively. Discrete conductance changes, characteristic of the binding event, were observed at extremely low target concentrations (e.g., Fig. 3). A very recent contribution from the same laboratory demonstrated the use of an antibody-functionalized silicon nanowire sensor array for the multiplexed label-free real-time monitoring of cancer markers in undiluted serum samples (Zheng et al., 2005). The biosensing utility of other 1D nanomaterials, such as conducting polymer NW or carbon-nanotubes, has also been illustrated (Ramanathan et al., 2005; Katz and Willner, 2004).

4. Nanoparticle-based bioelectronic assays

As indicated in the previous section, the emergence of nanotechnology has opened new horizons for electrochemical biosensors (Wang, 2005). In addition to nanowires, it is possible to use nanoparticles tags for designing electrical bioaffinity assays with remarkable sensitivity and multiplexing capability. The rapid progress of nanomaterial-based electrochemical biosensors suggests the major impact they may have upon cancer diagnostics in the near future. The ultrahigh sensitivity of such nanoparticle-based electrochemical sensing protocols opens up the possibility of detecting cancer markers that cannot be measured by conventional methods and could lead to an early detection of the disease.

Stripping voltammetry has been particularly useful for detecting metal nanoparticle tags, owing to its 'built-in' accumulation (electrodeposition) step that leads to a remarkable sensitivity (Wang, 1985). About five years ago, two groups (Dequaire et al., 2000; Wang et al., 2001b) demonstrated the use of gold nanoparticle tracers for stripping-based electrochemical

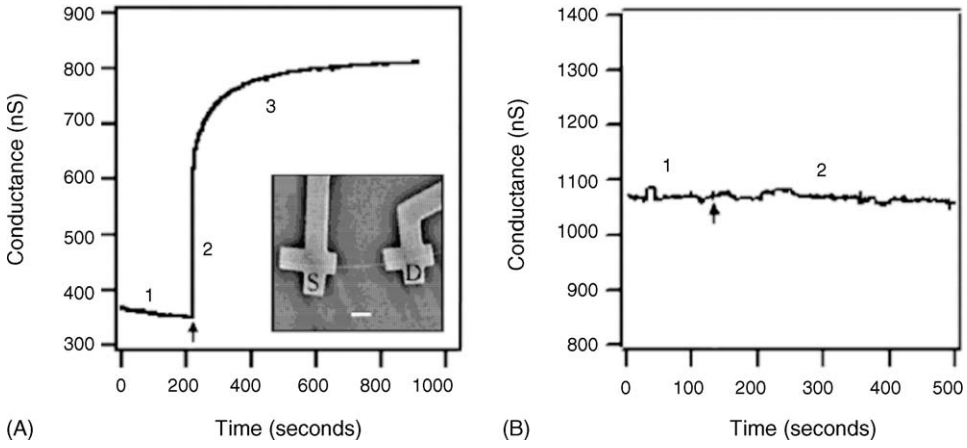

Fig. 3. DNA hybridization measurements at SiNW functionalized with PNA probes. (A) Real-time conductance response to 60 fM WT DNA sample. The arrow marks the point in time when the sample was added. The inset shows a SEM image of a typical SiNW device with source (S) and a drain (D) indicated; scale bar is 1 μm. (B) Time dependent conductance in DNA-free solution; the arrow indicates the point in time when a new solution sample was added [reproduced with from Hahm and Lieber (2004) with permission].

detection of DNA hybridization and antibody–antigen interactions. Such protocols relied on capturing the gold nanoparticles to the hybridized target or captured antigen, followed by their dissolution and electrochemical stripping measurement of the metal tracer. A triple-amplification bioassay, coupling polymeric carrier-sphere amplifying units (loaded with numerous gold nanoparticles tags) with the preconcentration feature of the electrochemical stripping detection and a catalytic electroless enlargement of the multiple gold-particle tags was also demonstrated (Kawde and Wang, 2004).

Adramatic signal amplification can be achieved by linking the biorecognition units to polymeric microbeads carrying a large number of redox tracers. For example, ultrasensitive electrical DNA detection was reported on the basis of polystyrene beads impregnated with the ferrocenecarboxaldehyde redox marker (Wang et al., 2003a). An amplified electrochemical sandwich immunoassay based on a polyelectrolyte-coated ferrocene microcrystal was also reported recently (Mak et al., 2005). Capturing of the antibody-conjugated ferrocene microcystal was followed by release of a large amount of the redox marker through the capsule wall (by a releasing agent), and led to a highly sensitive amperometric biodetection. The successful clinical realization of these ultrasensitive bioelectronic detection schemes requires proper attention to non-specific adsorption issues that commonly control the detectability of bioaffinity assays.

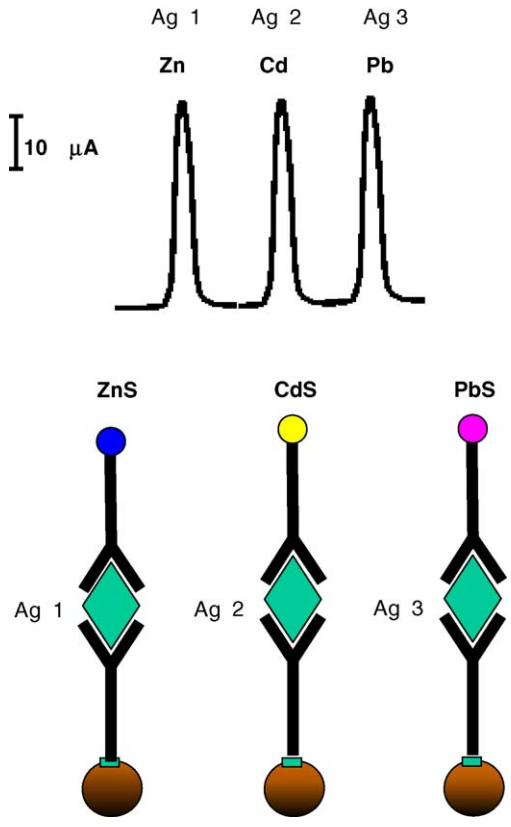

Fig. 4. Simultaneous monitoring of multiple proteins in connection to different inorganic nanocrystal tags and electrochemical stripping transduction [reproduced with from Liu et al. (2004) with permission].

Nanoparticle-induced changes in the conductivity across a microelectrode gap were exploited for highly sensitive electronic detection of DNA hybridization (Park et al., 2002). The capture of the nanoparticle-tagged DNA targets by probes immobilized in the gap between the two closely spaced microelectrodes, and a subsequent silver precipitation, resulted in a conductive metal layer across the gap, and led to a measurable conductivity signal. Such hybridization-induced conductivity signals, offered low detection limits down to the 0.5 pM level.

Inorganic nanocrystals offer an electrodiverse population of electrical tags as needed for multiplexed clinical testing. We demonstrated the use of different inorganic-nanocrystal tracers for a multi-target electronic detection of proteins (Liu et al., 2004) or DNA (Wang et al., 2003b). Four encoding nanoparticles (cadmium sulfide, zinc sulfide, copper sulfide, and lead sulfide) were thus used to differentiate the signals of four proteins or DNA targets in connection with sandwich immunoassay and hybridization assay, respectively, along with stripping voltammetry of the corresponding metals. Each binding event thus yielded a distinct voltammetric peak, whose size and position reflected the level and identity, respectively, of the corresponding antigen or DNA target (Fig. 4). The concept can be readily scaled up and multiplexed by using a parallel high-throughput automated microwell operation, with each microcavity capable of carrying out multiple measurements.

5. Conclusions and future prospects

Elegant research on new sensing concepts has opened the door to a widespread clinical applications of electrochemical devices. Such devices are extremely useful for delivering the diagnostic information in a fast, simple, and low cost fashion, and are thus uniquely qualified for meeting the demands of point-of-care cancer screening. The high sensitivity of modern electrochemical bioaffinity assays should facilitate early detection and treatment of diseases and should lead to increased patient survival rates. The attractive properties of electrochemical devices are thus extremely promising for improving the efficiency of diagnostic testing and therapy monitoring, and for point-of-care cancer testing, in general. The main challenge is to bring electrochemical techniques to the patient's side for use by non-laboratory personnel without compromising accuracy and reliability. The realization of decentralized electronic testing of cancer would thus require additional extensive developmental work. Special attention should be given to non-specific adsorption issues that commonly control the detection limits of electrochemical bioaffinity assays. It is expected that the creativity of electrochemists and material scientists, coupled with proper resources, will revolutionize cancer diagnostics in a manner analogous to their current leading role in diabetes monitoring. Disposable cartridges, containing electrode strips (coated with numerous receptors) along with related sample processing, could thus offer early screening of cancer in a point-of-care setting, by measuring abnormalities in protein profiles within few minutes. Accordingly, there is no doubt that electrochemical biosensors will become a powerful tool for cancer diagnostics in the near future.

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References

Armistead, P.M., Thorp, H.H., 2002. Electrochemical detection of gene expression in tumor samples: overexpression of rak nuclear tyrosine kinase. Bioconjugate Chem., 172–176.

Aizawa, M., Moriaka, A., Suzuki, S., Nagamura, Y., 1979. Enzyme immunosensor III amperometric determination of human choriogonadotropin by membrane bound antibody. Anal. Biochem. 94, 22–28.

Brett, A.M., Serrano, S., Macedo, T., Raimundo, D., Marquez, M., LaScalea, M., 1996. Electrochemical determination of carboplatin in serum using a DNA-modified glassy carbon electrode. Electroanalysis 8, 992–995.

Dai, Z., Yan, F., Chen, J., Ju, H., 2003. Reagentless amperometric immunosensors based on direct electrocehmistry of HRP for determination of carcinoma antigen. Anal. Chem. 75, 125.

Dequaire, M., Degrand, C., Limoges, B., 2000. An electrochemical metalloimmunoassay based on colloidal gold label. Anal. Chem. 72, 5521.

Erickson, K., Wilding, P., 1993. Evaluation of a novel point-of-care system, the i-STAT portable clinical analyzer. Clin. Chem. 39, 283–288.

Fojta, M., 2002. Electrochemical sensors for DNA interactions and damage. Electroanalysis 14, 1449–1463.

Gooding, J.J., 2002. Electrochemical DNA hybridization biosensors. Electroanalysis 14, 1149–1156.

Hahm, J., Lieber, C., 2004. Direct ultrasensitive electrical detection of DNA and DNA sequence variations using nanowire nanosensors. Nano Lett. 4, 51–55.

He, Z., Gao, N., Jin, W., 2003. Determination of tumor marker CA125 by capillary electrophoretic enzyme immunoassay with electrochemical detection. Anal. Chim. Acta 497, 75–81.

Kawde, A., Wang, J., 2004. Amplified electrical transduction of DNA hybridization based on polymeric beads loaded with multiple gold nanoparticle tags. Electroanalysis 16, 101–105.

Katz, E., Willner, I., 2004. Biomolecule-functionalized carbon nanotubes: applications in nanobioelectronics. Chem. Phys. Chem. 5, 1085–1091.

Kojima, K., Hiratsuka, A., Suzuki, H., Yano, K., Ikebukuro, K., Karube, I., 2003. Electrochemical protein chip with arrayed immunosensors with antibodies immobilized in a plasma-polymerized film. Anal. Chem. 75, 1116–1122.

Lin, J., Ju, H., 2005. Electrochemical and chemiluminescent immunosensors for tumor markers. Biosens. Bioelectron. 20, 1461–1470.

Liu, G., Wang, J., Kim, J., Jan, M.R., 2004. Electrochemical coding for multiplexed immunoassays of proteins. Anal. Chem. 76, 7126–7130.

Mak, W.C., Cheung, K.Y., Trau, D., Warsinke, A., Scheller, F., Rennenberg, R., 2005. Electrochemical bioassay utilizing encapsulated electrochemical active microcrystal biolabels. Anal. Chem. 77, 2835–2841.

Melosh, N., Boukai, A., Diana, F., Gerardot, B., Badolato, A., Petroff, P., Heath, J., 2003. Ultrahigh density nanowire lattices and circuits. Science 300, 11–15.

Meyerhoff, M.E., Duan, C., Meusel, M., 1995. Novel non-spearation sandwich-type electrocehmical enzyme immunoassay system for detecting marker proteins in undiluted blood. Clin. Chem. 41, 1378–1384.

Mikkelsen, S.R., 1996. Electrochemical biosensors for DNA sequence detection. Electroanalysis 8, 15–22.

Newman, J., Turner, A.P.F., 2005. Home blood glucose biosensors: a commercial perspective. Biosens. Bioelectron. 20, 2435–2453.

Park, S., Taton, T.A., Mirkin, C.A., 2002. Array-based electrical detection of DNA with nanoparticle probes. Science 295, 1503–1506.

Parlov, V., Willner, I., Dishon, A., Kotler, M., 2004. Amplified detection of tolemerase activity using electrochemical and quartz microbalance measurements. Biosens. Bioelectron. 20, 1011–1021.

Patolsky, F., Zheng, G., Hayden, O., Lakamyali, M., Zhuang, X., Lieber, C.M., 2004. Electrical detection of single viruses. Proc. Natl. Acad. Sci. U.S.A. 101, 14017–14022.

Ramanathan, K., Bangar, M.A., Yun, M., Chen, W., Myung, N.V., Mulchandani, A., 2005. Bioaffinity sensing using biologically functionalized conducting polymer nanowire. J. Am. Chem. Soc. 127, 496–497.

Rodriguez, M., Kawde, N., Wang, J., 2005. Aptamer biosensor for label-free impedance spectroscopy detection of proteins based on recognition-induced switching of surface charge. Chem. Commun., 4267.

Ronkainen-Matsuno, N., Thomas, J., Halsall, A., Heineman, W.R., 2002. Electrochemical immunoassays moving into the fastlane. Trends Anal. Chem. 21, 213–219.

Tansil, N.C., Xie, F., Xie, H., Gao, Z., 2005. An ultrasensitive nucleic acid biosensor based on the catalytic oxidation of guanine by a novel redox threading intercalator. Chem. Commun., 1064–1066.

Turner, A.P., Karube, I., Wilson, G., 1986. Biosensors: fundamentals and applications. Oxford Science Publications, Oxford.

Wahr, J., Lau, W., Tremper, K., Hallock, L., Smith, K., 1996. Accuracy and precision of a new, portable, handheld blood gas analyzer, the IRMA(R). J. Clin. Monit. 12, 317–324.

Wang, J., 1985. Stripping Analysis. VCH, New York.

Wang, J., Rivas, G., Cai, X., Chicharro, M., Beleiter, A., Mowat, M., Palecek, E., Nielsen, P., 1997. Detection of point mutation in the p53 gene using a peptide nucleic acid biosensor. Anal. Chim. Acta 344, 111–118.

Wang, J., 1999. Amperometric biosensors for clinical and therapeutic drug monitoring: a review. J. Pharm. Biomed. Anal. 19, 53–74.

Wang, J., 2000a. Analytical Electrochemistry. Wiley, New York.

Wang, J., 2000b. DNA biosensors and gene chips. Nucl. Acids Res. 28, 3011–3016.

Wang, J., Kawde, A., 2000. Pencil-based renewable biosensor for label-free electrochemical detection of DNA hybridization'. Anal. Chim. Acta 431, 219–225.

Wang, J., Ibanez, A., Chatrathi, M., Escarpa, A., 2001a. Electrochemical immunoassays on microchip platforms. Anal. Chem. 73, 5323–5328.

Wang, J., Xu, D., Kawde, A., Polsky, R., 2001b. Metal nanoparticles based electrochemical stripping potentiometric detection of DNA hybridization. Anal. Chem. 73, 5576–5582.

Wang, J., Polsky, R., Merkoci, A., Turner, K., 2003a. Electroactive beads for ultrasensitive DNA detection. Langmuir 19, 989–992.

Wang, J., Liu, G., Merkoçi, A., 2003b. Electrochemical coding technology for simultaneous detection of multiple DNA targets. J. Am. Chem. Soc. 125, 3214–3215.

Wang, J., 2005. Nanomaterial-based electrochemical biosensors. Analyst 130, 421–426.

Weisner, A., 2004. Detection of tumor markers with protein chip technology. Curr. Pharm. Biotechnol. 5, 45–67.

Wilson, M.S., 2005. Electrochemical immunosensors for the simultaneous detection of two tumor markers. Anal. Chem. 77, 1496–1502.

Zheng, G., Patolsky, F., Cui, Y., Wang, W.U., Lieber, C.M., 2005. Multiplexed electrical detection of cancer markers with nanowire sensor arrays. Nat. Biotechnol. 23, 1294.