ELECTRICAL LABEL-FREE SENSING OF CARDIAC TROPONIN BIOMARKER: FET-BASED INTEGRATION WITH SUBSTRATE-GATE COUPLING

by

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LIST OF ABBREVIATIONS

2D	2-dimension
3D	3-dimension
Ab	Antibody
AFM	Atomic force Microscope
ALD	Atomic layer deposition
ALP	Alkaline phosphatase
AMI	Acute myocardial Infarction
APTES	3-aminopropyltriethoxysilane
ASIC	Application-specific integrated circuit
AuNP	Gold nanoparticles
BNP	B-type natriuretic peptide
BOE	Buffered oxide etch
BOX	Buried oxide
BSA	Bovine serum albumin
CCD	Charge-coupled device
CK-MB	Creatine kinase-MB
CK-MM	Creatine kinase-MM
CL	Chemiluminescence
CLEIA	Chemiluminescence enzyme immunoassay
CLIA	Chemiluminescence immunoassay
CMOS	Complementary metal-oxide semiconductor
CNF	Carbon nanofiber

CNT	Carbon nanotube
CP	Conducting polymer
CRP	C-reactive protein
cTn	Cardiac troponin
cTnC	Cardiac troponin C
cTnI	Cardiac troponin I
cTnT	Cardiac troponin T
CV	Co-efficient of variation
CVD	Chemical vapour deposition
DNA	Deoxyribonucleic acid
ECB	ELISA coating buffer
ECG	Electrocardiography
EDC	1-ethyl-3-(3-dimethylaminopropyl)carbodiimide
EDX	Energy dispersive X-ray
ELISA	Enzyme-linked immunosorbent Assay
EnFET	Enzyme field-effect transistor
EOC	ELISA-on-chip
FESEM	Field-emission scanning electron microscope
FET	Field-effect transistor
FI	Fluorescence immunoassay
FMGC	Fluoro-microbead guiding chip
f-RG	Functionalized reinforcing bar graphene
FTIR	Fourier-transform infrared
GA	Glutaraldehyde

GCE	Glassy carbon electrode
GO	Graphene oxide
HRP	Horseradish peroxidase
IDE	Interdigitated electrode
ISFET	Ion-sensitive field effect transistor
IUPAC	International Union of Pure and Applied Chemistry
LOD	Limit of detection
MAb	Monoclonal antibody
MEA	Monoethanolamine
MHDA	Mercaptohexadecanoic acid
MPA	3-mercaptopropionic acid
MWCNT	Multi-walled carbon nanotube
Myo	Myoglobin
NEA	Nanoelectrode array
NHS	N-hydroxysuccimide
NMO	Nanostructured metal-oxide
NP	Nanoparticle
NSTEMI	Non-ST segment elevation myocardial infarction
OEG	Oligo (ethylene glycol)
PANI	Polyaniline
PBS	Phosphate buffer saline
PDMS	Poly(dimethylsiloxane)
PECVD	Plasma-enhanced chemical vapour deposition
PEDOT	Poly(3,4-ethylenedioxythiophene)

PEI	Polyethyleneimine
POCT	Point-of-care testing
PPY	Polypyrrole
PyBuNHS	1-pyrenebutyric acid N-hydroxysuccinimide ester
PyMe	1-pyrenemethyl
PyMe-NH_2	1-pyrenemethyl hydrochloride
ReBar	Reinforcing bar
RNA	Ribonucleic acid
SAM	Self-assembled monolayer
SDS	Sodium dodecyl sulphate
SEM	Scanning electron microscope
SiNW	Silicon nanowire
SP	Screen-printed
SPE	Screen-printed electrode
SOI	Silicon-on-insulator
SPA	Semiconductor parametric analyzer
SPR	Surface plasmon resonance
SWCNT	Single-walled carbon nanotube
TMAH	Tetramethylammonium hydroxide
Tn	Troponin
UniProtKB	Universal Protein Resources Knowledgebase
XP	X-ray photoelectron
XPS	X-ray photoelectron spectroscopy
XRD	X-ray diffraction

LIST OF SYMBOLS

Al	Aluminium
Al_2O_3	Alumina
Au	Gold
c	Y-intercept
C	Carbon
CH_4	Methane
HCl	Hydrochloric acid
HfO_2	Hafnium (IV) oxide
HNO_3	Nitric acid
I	Current
I_D	Drain current
I_{D0}	Immobilization drain current
IgG	Immunoglobulin G
L	Length
m	Slope
μ	Mean
N	Nitrogen
Ni	Nickel
O	Oxygen
ρ	Resistivity
pI	Isoelectric point
Pt	Platinum

Q_F	Interface charge density
R	Electrical resistance
R_a	Average roughness
R_{et}	Electron transfer resistance
ΔR_{et}	Difference in electron transfer resistance
RMS	Root mean square
RSD	Relative standard deviation
Si	Silicon
SiO_2	Silicon dioxide
σ	Standard deviation
SnO_2	Tin oxide
t	Thickness
V	Voltage
V_D	Drain voltage
V_{SG}	Substrate-gate voltage
V_T	Threshold voltage
W	Width
ZnO	Zinc oxide

Pengesanan Elektrik tanpa Label untuk Penanda Biologi Troponin Jantung: Berasaskan Transistor Kesan Medan Disepadukan Gandingan Get-substratum

ABSTRAK

Infaksi myokardium akut (AMI) merupakan punca utama kematian di seluruh dunia walaupun dengan adanya kemajuan terapi. Oleh itu, kaedah diagnosis awal menggunakan biopenanda-biopenanda jantung adalah diperlukan supaya tindakan yang tepat dapat dilaksanakan. Troponin jantung I (cTnI) merupakan salah satu biopenanda jantung untuk diagnosis awal AMI dan dianggap sebagai "piawaian emas" untuk menentukan kecederaan otot jantung. Pengesanan cTnI melalui biopenderia berasaskan elektrikal membolehkan pengesanan tanpa label dengan menukarkan pengikatan biomolekul kepada isyarat elektrikal yang ketara melalui sebuah pemindaharuh semikonduktor. Biopenderia ini memanfaatkan keberaliran untuk menentukan kewujudan biomolekul. Salah sebuah biopenderia berasaskan elektrikal ini yang dikenali sebagai biopenderia berasaskan transistor kesan medan (FET) telah menarik banyak perhatian kerana memiliki konsep pemindaharuhan cas; di mana ia membolehkan diagnosis segera biopenanda jantung dengan kadar sensitiviti yang tinggi secara khusus pada kepekatan rendah di peringkat awal. Dalam kajian ini, biopenderia berasaskan FET-zink oksida (ZnO) digandingkan dengan get-substratum telah direka bentuk dan difabrikasi untuk pengesanan cTnI. Saput nipis ZnO sebagai bahan separa-pengalir jenis-n dan juga merupakan pemindaharuh serasi dengan biologi telah diendapkan menggunakan teknik-teknik gel-sol dan penyalutan putar di antara terminal punca dan salir jenis-p, yang terletak di atas substratum silikon-atas-penebat (SOI) untuk menghasilkan simpang *p-n-*p, sebuah peranti yang berupaya untuk aplikasi pengesanan biologi. Morfologi permukaan salut nipis ini telah dicirikan melalui mikroskop daya atom (AFM) dan mikroskop elektron imbasan pancaran medan (FESEM). Saput nipis ini memperlihatkan fasa wurzit heksagon seperti yang telah dipaparkan oleh analisa belauan sinar-X (XRD) adalah bersesuaian dengan interaksi biomolekul. Permukaan saput nipis ZnO ini telah ditetapkan dengan antibodi monoklonal cTnI (MAb-cTnI) melalui kaedah pengikatan kovalen untuk mengesan biopenanda cTnI. Proses ini telah dibuktikan melalui infra-merah jelmaan fourier (FTIR) dan spektroskopi fotoelektron sinar-X (XPS). Struktur peranti ini telah diselakukan di dalam perisian penyelaku 2-dimensi Silvaco ATLAS bertujuan untuk menghuraikan ciri elektrikal peranti tersebut, secara khususnya kepekatan elektron di dalam terusan dan permukaan oksida tertanam/substratum. Peranti ini mempamerkan strategi baru melalui pencirian elektrikal, apabila digandingkan dengan get-substratum yang mempertingkatkan pembentukan lapisan pengaliran lubang pada saluran yang terletak di antara kawasan saliran dan punca. Akhirnya, biopenderia ini menunjukkan peningkatan pada perubahan nisbi aras arus saliran yang ketara dalam julat lelurus daripada 6.2 ke 16.5% dengan peningkatan kepekatan biopenanda cTnI yang bercas positif daripada 1 ng/ml ke 10 µg/ml. Sensitiviti pengesanan peranti ini adalah pada 2.51 %·$(g/ml)^{-1}$ dengan had pengesanan (LOD) serendah 3.24 pg/ml.

Electrical Label-Free Sensing of Cardiac Troponin Biomarker: FET-based Integration with Substrate-gate Coupling

ABSTRACT

Acute myocardial infarction (AMI) is a leading cause of death worldwide despite the existence of therapy's advances. Therefore, an early diagnosis method by using cardiac biomarkers is essential to enable correct countermeasures. Cardiac Troponin I (cTnI) is one of the cardiac biomarkers for early diagnosis of AMI and considered as 'gold standard' for cardiac muscle injury determination. The detection of cTnI through an electrical-based biosensor allows label-free detection by converting biomolecular binding event into a significant electrical signal via a semiconductor transducer. It utilizes conductivity to specify the existence of biomolecules. One of the electrical-based biosensors known as field-effect transistor (FET)-based biosensor has drawn much attention for owning the concept of charge transduction; thus, allows early, high sensitivity, high selectivity, and rapid diagnosis of the specific cardiac biomarker at low concentrations. In this work, the zinc oxide (ZnO)-FET biosensor coupled with substrate-gate has been designed and fabricated for the detection of cTnI biomarker. ZnO thin film, as n-type biocompatible semiconductor material, and also as transducer was deposited via sol-gel and spin coating techniques between p-type source and drain terminal on SOI substrate, forming a *p*-*n*-p junction, a device capable of bio-sensing application. The surface morphology of the thin film was characterized by using atomic force microscopy (AFM) and field emission scanning electron microscopy (FESEM). The thin film, which demonstrated hexagonal wurtzite phase as shown by X-ray diffraction (XRD) analysis appropriate for biomolecules interaction. The surface of the ZnO thin film was immobilized with cTnI monoclonal antibody (MAb-cTnI) as biological receptor via covalent binding technique for capturing cTnI biomarker. The process was validated by Fourier transform-infrared (FTIR) and X-ray photoelectron spectroscopy (XPS). The device structure was simulated in Silvaco Atlas 2D-simulator, to elucidate its electrical characteristic, by means of hole and electron concentration in the channel and buried oxide/substrate interface, respectively. The device demonstrated a new strategy via electrical characterization with the introduction of substrate-gate coupling that enhanced the formation of hole conduction layer at the channel between drain and source region. Finally, the biosensor shown significant increment in relative changes of drain current level in a linear range of 6.2 to 16.5 % with the increase of positively charge cTnI biomarker concentrations from 1 ng/ml to 10 µg/ml. The device sensitivity of the detection is at 2.51 %·$(g/ml)^{-1}$ with limit of detection (LOD) down to 3.24 pg/ml.

CHAPTER 1

INTRODUCTION

1.1 Background

Biosensors are frequently defined as integrated diagnostic devices comprising of three elements, which are bio-receptor, transducer, and a signal processing unit (Conroy, Hearty, Leonard, & O'Kennedy, 2009). Generally, a suitable transducer surface of biosensor is immobilized with a biological receptor material (i.e. antibody (Ab), deoxyribonucleic acid (DNA), or ribonucleic acid (RNA)). It produces a measureable signal upon bio-receptor interaction with the specific biomolecules (Goode, Rushworth, & Millner, 2015; Qureshi, Gurbuz, & Niazi, 2012). The generated signals is in the mode of either electrochemical (Gomes-Filho, Dias, Silva, Silva, & Dutra, 2013; Horak, Dincer, Qelibari, Bakirci, & Urban, 2015), optical (H.-Z. He et al., 2013; C.-H. Leung et al., 2013; K.-H. Leung et al., 2015; Lu et al., 2014), mass change (piezoelectric/acoustic wave) (Joonhyung Lee et al., 2013), or magnetic (J. Liu, Zhang, Wang, Zheng, & Sun, 2014). The development of biosensors for diversity of biomolecules detection has cover many field, including medicine (J. Wang, 2006), food testing (Huet et al., 2010), environmental (Weller, Schuetz, Winklmair, & Niessner, 1999), and process control monitoring (Venugopal, 2002). The developed biosensors that come with several advantages (i.e. portable, inexpensive tools for the rapid detection of pathogens, proteins and other biomolecules) are intended to provide as an alternative method to the conventional bioanalytical approaches (Fathil et al., 2016). Commonly, conventional bioanalytical