GRAPHENE BIOSENSOR FOR DETECTION OF hCG BIOMARKER

Sofia Teixeira[1], A. Castaing[1], G. Burwell[1], R.S. Conlan[2], O.J. Guy[1]

[1]College of Engineering, Swansea University, SA2 8PP Swansea, UK

[2] College of medicine, Swansea University, SA2 8PP Swansea, UK

r.s.r.teixeira.607010@swansea.ac.uk

Abstract

Human chorionic gonadotropin (hCG), a 37 kDa glycoprotein hormone [1], is an important biomarker for pregnancy and one of the most important tumor markers for several cancers [2]. Most modern assay systems for hCG are optical based ELISAs. Though advances have been made in miniaturization of sensor diagnostics, in point of care detection, and even automated/continuous monitoring systems, the desired level of sensitivity for early detection and monitoring for cancer has not yet been achieved.

A novel chemically-modified graphene diagnostic sensor has been developed for ultrasensitive detection of a hCG (human chorionic gonadotropin) biomarker. Multi-layer epitaxial graphene (MEG), grown on silicon carbide substrates, has been patterned using electron beam lithography to produce channel based devices. Silicon Carbide (SiC) is a suitable substrate for graphene growth [3-7]. Graphene is grown by sublimating the silicon from the SiC substrate. Graphene, essentially a single atomic layer of graphite, has some exceptional electronic properties such as carrier mobilities of more than 15000 $cm^2 V^{-1}s^{-1}$ at room temperature and ballistic transport of carriers [4]. Modifying the growth conditions yields variations in the graphene layer thickness and consequently its electronic properties.

Diagnostics based on bio-functionalised semiconductor devices are an important development in ultrasensitive sensors for early detection and monitoring of disease biomarkers. The chemical sensor uses chemically-modified graphene channels, functionalized with a “bio-receptor” antibody to detect the presence of the disease target biomarker, anti-hCG. It is then able to interact with the surface-bound bioreceptor and produce a change in the charge properties of the biosensor device. This surface charge modification is measured by amperometry on our graphene-based channel devices.

The attachment of the anti-hCG onto the graphene surface requires two parallel processes: the surface modification and the antibody modification.

The surface modification is performed by firstly terminating the graphene with -OH groups using the Fenton reaction [8], then reacted with 3-Aminopropyl-triethoxysilane (APTES) [9] in order to obtain an amine-terminated surface. This is illustrated in Figure 1 (a to d) where the presence of the silane and the carbon chain from the Aptes molecule are clearly detected using XPS. Amine groups, along with oxygen in the C-O-Si configuration were also detected (Table 1).

In order to be able to react with the surface amine groups, the carboxylic acids on the antibody are activated. However, in order to prevent the antibody from cross-linking, the majority of amine groups on the antibody are blocked using Di-*tert*-butyl dicarbonate. The antibody can now be reacted with the amine on the surface. The groups blocking the amines on the antibody are subsequently removed by mild acidic treatment.

The sensitivity of the obtained device was then tested against varying concentration of hCG by measuring the resistance across the modified graphene channel (Figure 2). The obtained result showed a sensitivity of the sensor of 142 Ω/(ng/ml), and a detection limit around 1ng/ml.

References

[1] J. Chen, J.H. Tang, *et al*. Biomaterials 27 (2006) 2313–2321.
[2] M. Aizawa, A. Morioka, *et al*. Anal. Biochem. 94 (1979) 22–28.
[3] O.J. Guy, M. Lodzinski, *et al.* Appl. Surf. Sci., Vol. 254, pp.8098-8105, 2008.

[4] C. Berger, Z. Song, *et al.* Science, Vol. 312, pp.1191-1196, 2006.
[5] A. Charrier, A. Coati, *et al.* J. Appl. Phys., Vol. 92,pp.2479-2484, 2002.
[6] C. Berger, Z. Song, *et al.* J. Phys. Chem. B, Vol. 108, pp.19912-19916,2004.
[7] A. Castaing, O.J. Guy, *et al.* Mater. Sci. Forum, Vols. 615-617, pp.223-226, 2009.
[8] R.H. Bradley, K. Cassity, *et al*. Applied Surface Science 258 (2012) 4835–4843.
[9] J. Kathi, K. Y. Rhee. J Mater Sci (2008) 43:33–37.

Figures

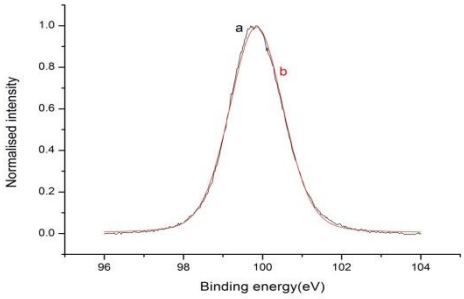

Figure 1(a). XPS core level spectrum of the Si2p peak of an epitaxial graphene sample (grown on SiC) before functionalisation (a) measured data, (b) fitted peak attributed to SiC.

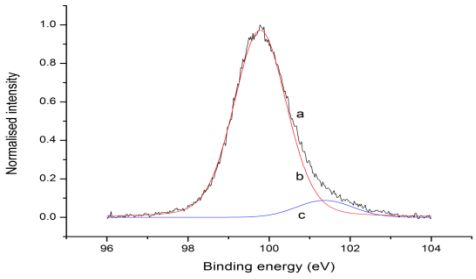

Figure 1(b). XPS core level spectrum of the Si2p peak of an epitaxial graphene sample after functionalisation (a) measured data, (b) fitted peak attributed to SiC and (c) fitted peak attributed to the silicon atom of the APTES molecule.

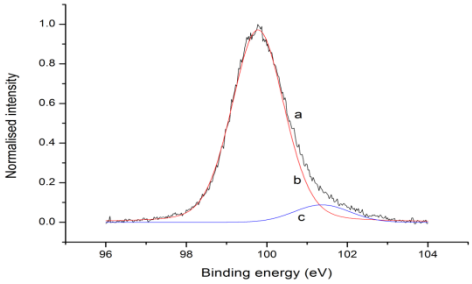

Figure 1(c). XPS core level spectrum of the C1s peak of a graphene sample before functionalisation (a) measured data, (b) fitted SiC peak and (c) fitted epitaxial graphene peak.

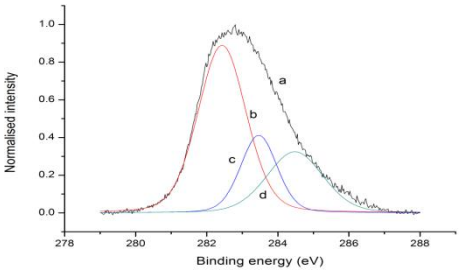

Figure 1(d). XPS core level spectrum of the C1s peak of a graphene sample after functionalisation (a) measured data, (b) fitted graphene peak(c) fitted SiC peak, (d), fitted APTES peak.

		Graphene		APTES	
peak	sensitivity factor	measured area	actual contribution	measured area	actual contribution
C1s SiC	0.25	3915	39.5	1858	35.9
C1s graphene	0.25	2146	21.6	657	12.7
C1s Aptes	0.25	0		790	15.3
Si SiC	0.27	4172	38.9	1725	30.9
Si Aptes	0.27	0		156	2.8
N Aptes	0.43	0		212	2.4

Table 1. Description of bonds and atomic abundances calculated from the fitted components of the C, Si and N core peaks from XPS measurement before and after functionalisation with APTES.

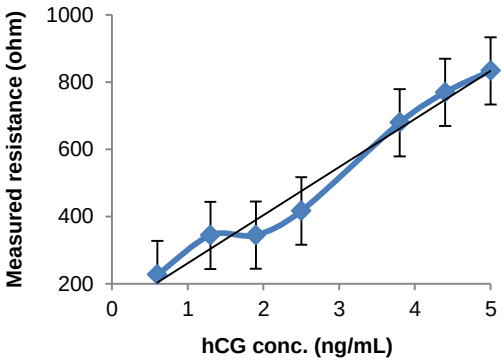

Figure 2: hCG concentration plotted as a function of channel resistance across a 100 µmx 4mm graphene channel.