A supramolecular pyrenyl glycoside-coated 2D MoS$_2$ composite electrode for selective cell capture†

Mokhtari Wahiba,$^a$ Xue-Qing Feng,$^a$ Yi Zang,$^b$ Tony D. James,$^c$ Jia Li,*$^a$ Guo-Rong Chen$^a$ and Xiao-Peng He*$_a$

Here we demonstrate the simple construction and characterization of a pyrenyl glycoside-coated 2D MoS$_2$ material composite capable of selectively capturing proteins and live cells on an electrode, as determined by differential pulse voltammetry.

The ability to selectively capture a target cell on a solid support is important for the advancement of cell biology and clinical diagnosis. Many bioinspired, well-defined material surfaces are developed, for which bioselectivity relies on unique topological features directed towards specific cell morphologies.$^1$ However, to isolate a cell on the surface, immunosorbent assays that depend on the capture of a cell-surface biomarker by monoclonal antibodies are required. But, the preparation of antibodies is sluggish and costly, and the conventional immunosorbent protocols are accompanied by high technical demand and long detection time. As a result, simpler and more effective methods for selective cell capture are urgently required.

Receptor–ligand interactions are crucial for a number of physiological and pathological events. These interactions are selective and have been shown to be applicable for targeted cell imaging and drug delivery.$^{2-6}$ The coating of ligand arrays onto material surfaces has resulted in effective sensing systems for the selective detection of biomacromolecules and cells/pathogens that express receptors for the attached ligands.$^7,8$ These advanced sensors could be an alternative to traditional immunoassays. Of the many smart sensing systems developed, the construction of 2D graphene composite electro sensors has been of particular interest because of the ease and flexibility in sensor fabrication, high sensitivity and short detection time.$^9-12$ Recently, increasing efforts have been made in the development of 2D graphene analogues (for example 2D transition metal dichalcogenides [TMDs]) as multifunctional materials.$^{13,14}$ These materials have also found application in biosensing and disease theranostics.$^{15-20}$

With continuing interest in the development of functional 2D composite materials,$^{21-26}$ here we illustrate the use of 2D TMD for the simple construction of a composite electrode that selectively captures a target cell over other control cells.

A glycoligand (galactose) that is selectively recognized by a cell-surface galactose receptor (the asialoglycoprotein receptor [ASGPr])$^{27}$ was used to couple with a binder to the 2D material surface. Pyrene was used as the binder for surface attachment due to its planarity.$^6$ Click chemistry$^{28}$ coupling of the glycoligand with a polyethylene glycol (PEG)-grafted pyrene-1-butyric acid produced the glycopyrene ($WXB$) (Fig. 1a and Scheme 1).

The 2D MoS$_2$ sheets were prepared by a liquid exfoliation method.$^{29}$ Subsequently, the components ($WXB$ and 2D MoS$_2$) were mixed in an aqueous solution (Tris-HCl, 0.01 M, pH 7.4) and sonicated for 1 h to facilitate assembly. The formation of the supramolecular $WXB$/2D MoS$_2$ composite is probably driven by the van der Waals interactions between $WXB$ and 2D MoS$_2$.30

Fig. 1 (a) Structure of pyrenyl galactoside ($WXB$) and (b) schematic illustration of the 2D MoS$_2$ composite electrode for selective cell capture.
Fluorescence) was quenched in a concentration-dependent range of 70–400 nm (Fig. 2b). While the composite scattering (DLS) indicated that the particle size of 2D MoS$_2$ was shown to increase with respect to 2D MoS$_2$, the subsequent addition of a galactose-selective lectin (peanut agglutinin [PNA]) further increased the size. This suggests that the fluorescence of the 2D composite could interact with a selective protein receptor to form a larger biomatrix. The fluorescence of WXb (pyrene fluorescence) was quenched in a concentration-dependent manner by 2D MoS$_2$ (Fig. 2c). This is in agreement with the quenching property of the 2D material for closely attached fluorescent species.$^{13-15}$ The quantum yields of WXb in water before and after assembly with 2D MoS$_2$ were determined to be 0.15 and 0.03, respectively.

Typical Raman shifts of 2D MoS$_2$ were observed at ca. 405 and 383 cm$^{-1}$, which are assigned to the out-of-plane vibration of S (A$_{1g}$) and in-plane relative motion between S and Mo (E$_{1g}$) modes of the MoS$_2$ crystal (Fig. 2d).$^{31}$ We observed that the E$_{1g}$/A$_{1g}$ ratio of the composite increased with respect to that of 2D MoS$_2$ alone, suggesting a perturbation towards the in-plane relative motion between S and Mo by the molecular coating.$^{32}$ In addition, typical UV shifts (621 and 682 nm, which are ascribed to the A1 and B1 direct exciton transitions of 2D MoS$_2$, respectively) were observed for both 2D MoS$_2$ and the composite (Fig. 2e).$^{31}$ These data suggest the successful formation of the pyrenyl glycoside-coated 2D MoS$_2$ composite.

With the composite in hand, we then tested its ability to capture cells on an electrode surface (Fig. 1b). Our previously developed screen-printed electrode (SPE) was used.$^{6,33}$ To the working electrode area, 2D MoS$_2$ and pyrenyl glycoside were dripped sequentially, forming a supramolecular composite on the surface. On the basis of the DLS result that the composite might interact selectively with specific lectins, we used differential pulse voltammetry (DPV) to measure the recognition using [Fe(CN)$_6$]$^{3-}/^{4-}$ as a redox probe.$^{34}$ We observed the typical DPV signal of the redox probe, which was gradually decreased with increasing PNA, a galactose-selective lectin (Fig. 3a). The quenched signal could be reasoned by the adhesion of the protein onto the glycoside layer of the composite electrode, thereby compromising electron transfer (Fig. 1b).$^{33-35}$ A good linearity was observed over a wide PNA concentration range (Fig. 3b), and the limit of detection (LOD) for the electrode...
towards PNA was determined to be 373 nM. A selectivity test showed that the current decrease of the redox probe was specific for the selective lectin (PNA), over other non-selective proteins including the mannoseselective concanavalin A, the $N$-acetyl glucosamine-selective wheat germ agglutinin, bovine serum albumin and pepsin (Fig. 3c and Fig. S1, ESI†). With these promising outcomes in hand we set out to evaluate cell capture using the composite electrode.

A hepatoma cell line that highly expresses ASGPr, which is selective for galactose, was used. An established sh-ASGPr cell line was used.36-39 With added analytes suggesting a gradual increase in electron-transfer resistance of the composite electrode. Clearly, we determined that preincubation and pepsin (Fig. 3c and Fig. S1, ESI†) did not alter the sensitivity of the electrode to Hep-G2 cells (Fig. S3, ESI†). These pieces of evidence suggest the good biospecificity of our 2D composite system for cell capture in a receptor-targeting manner.

In order to test the reversibility of the composite, a useful attribute for the isolation of captured cells, we carried out competition assays. Thus, we determined that preincubation with increasing concentrations of free $\alpha$-galactose and WXB with Hep-G2 caused a gradual current increase of the electrode (Fig. S4, ESI†), implying that the receptor-mediated capture of cells is reversible. We also used electrochemical impedance spectroscopy to investigate both protein and cell capture. Nyquist plots of the 2D composite electrode in the presence of increasing PNA and Hep-G2 cells (Fig. S5, ESI†) show increasing capacitive loops with added analytes suggesting a gradual increase in electron-transfer resistance of the composite electrode. Clearly indicating a coating of proteins/cells on the electrode surface as a result of ligand–receptor recognition.40

In summary, we have demonstrated that a simple 2D MoS$_2$ based pyrenyl$^{41,42}$ glycomosaic composite material can be used for the selective capture of cells on an electrode surface. This research may help the development of 2D-material composite based sensors for solid-phase analysis of cells and disease diagnostics.43-50

This research was supported by the 973 project (2013CB733700), the Science and Technology Commission of Shanghai Municipality (15540723800), the National Natural Science Foundation of China (21572058, 21576088 and 81302820) and the Shanghai Rising-Star Program (16QA1401400) (X.-P. H.). The Catalysis and Sensing for Environment (CASE) network is thanked for research exchange opportunities. T. D. J. thanks ECUST for a guest professorship.

Notes and references


