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Electrochemical Measurement of Coupled Protein Functionality Across Polymer Membranes

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ABSTRACT

Block copolymer-based membrane technology represents a versatile class of nanoscale materials in which biomolecules, such as membrane proteins, can be reconstituted [1-3]. Among its many advantages over conventional lipid-based membrane systems, block copolymers can mimic natural cell biomembrane environments in a single chain, enabling large-area membrane fabrication using methods like Langmuir-Blodgett deposition [4], or spontaneous protein-functionalized nanovesicle formation [5]. Based on this unique membrane property, a wide variety of membrane proteins possessing unique functionalities including pH/voltage gatable porosity, photon-activated proton pumping, and gradient-dependent production of electricity have been successfully inserted into ABA triblock copolymer systems [6].

Vesicle formation from polymers was confirmed using TEM spectroscopy. Figure 1a shows the formation of vesicles using the 4METH derivative while figure 1b. shows the formation of the 8METH vesicle. Following filtration of the vesicles with 200µm diameter pore sizes, the vesicles were shown to possess similar diameters.

After the protein-functionalized nanovesicles were interfaced with the electrode, current production values increased for both the light and dark-incubated samples as seen in the presence of clear redox peaks (Fig. 2). It is believed that a redox peak was present for the dark-incubated hybrid sample due to the fact that there was possible exposure to some ambient light that could have accounted for the increased current density when expected performance indicates that inactivated BR in the dark should produce no current. However, when comparing light-activated BR/COX vesicles with dark-incubated BR/COX vesicles as well as the initial controls, it could clearly be seen that light-activated samples possessed higher current densities that equated to approximately 12.5µA/cm^2 while controls resided in the pA-nA range.

Amperometric current vs. time plots were also obtained for the samples. Fig. 3 demonstrates the production of current using hybrid functionalized polymeric vesicles measured against the control, which produced no current. Careful attention was paid when setting the scan parameters to set upper and lower boundary limits that were not near the hydrolysis point of water.

Triblock copolymer membrane technology has enabled the formation of stable vesicles that preserve protein functionality. Furthermore, these protein-incorporated vesicles can be fabricated very quickly (~30min.) while conventional lipid vesicle protocol can require as much as a few days to develop protein-functionalized samples. These hybrid bionanodevices represent an exciting push towards the fabrication of fully integrated systems based on biomolecular function.

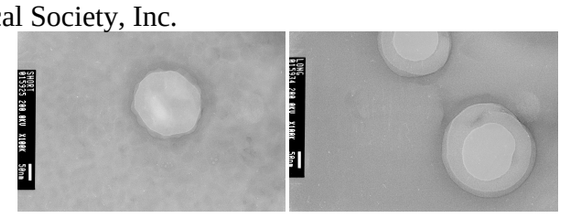

Figure 1. Figure 1a shows the formation of a 4METH nanovesicle while figure 1b shows the formation of 8METH vesicles.

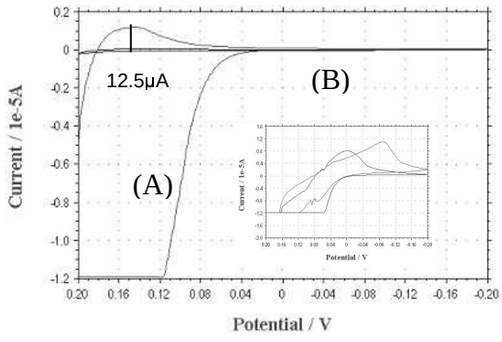

Figure 2. Measurement of hybrid vesicle current (A) against control current (B) reveals microampere-range currents for functionalized vesicles. (Inset: Redox peaks are visible for both light and dark samples, though incubation in light results in increased current).

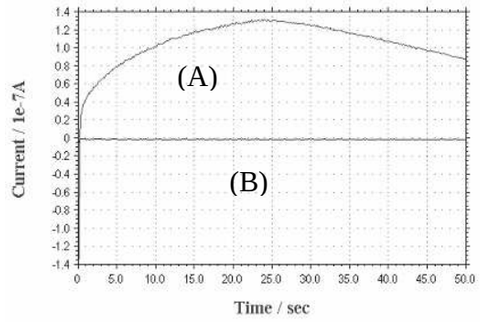

Figure 3. Amperometric detection of current shows an increase in current production (A) over protein-only controls (B).

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