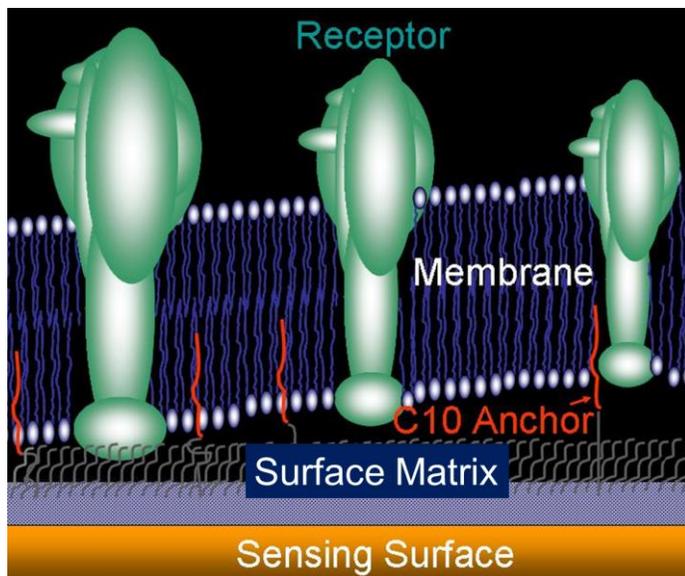


VESICLE CAPTURE FOR MEMBRANE-BOUND RECEPTOR INTERACTIONS (VesCap BIOSENSOR)

Label-free binding experiments interactions with cell membranes, native membrane-bound receptors or liposomes are now possible using the SensiQ Technologies Vesicle Capture (VesCap) sensor.



The VesCap sensor uses a hydrophobic anchor to stably adsorb membranes for binding studies.

On the VesCap chip, the lipid bilayer is maintained just as in the natural cellular environment, and membrane components in their native conformation can diffuse freely in this *ex vivo* model of the cell membrane.

We have not confirmed that the vesicles fuse into a single membrane bilayer but this is a likely case.

- VesCap is an excellent model for binding experiments with drugs, toxins or peripheral membrane-associated proteins involved in cell signaling^{1,2,3}.
- Membrane proteins and their partners interact just as in real cell membranes.⁴
- Receptor/ligand embedded in liposomes may be immobilized.
- VesCap chemistry is especially suitable for cell lines over-expressing a surface receptor.

VesCap Sensor Properties

- The lipid bilayer and the native structure of membrane proteins are maintained. Capture of vesicles onto the sensing surface is non-covalent, allowing free diffusion of membrane components in all directions.
- Preparation of vesicles, or liposomes, for attachment to this surface is straightforward.
 - Purification steps can often be eliminated as samples can be relatively impure.
- Regeneration of the VesCap surface is simple and complete allowing reuse of the biosensor

VesCap Mechanism

The VesCap chemistry uses a covalently linked lipophilic hydrocarbon chain in a hydrogel matrix to insert into the phospholipid bilayer of vesicles. Membrane fragments, liposomes, micelles are thereby captured.

¹Kim et al. Pharm Res **21** (2004) p. 1233-1238

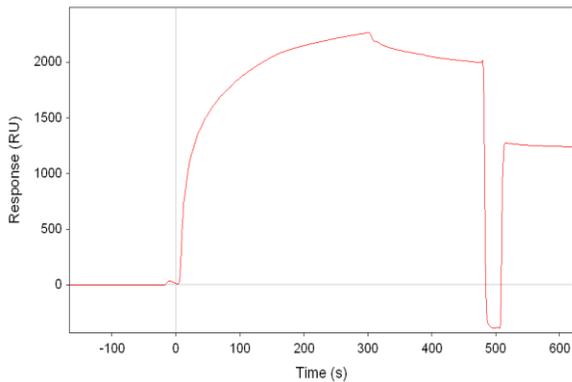
²Anderluh et al. Anal. Biochem. **344** (2005) p. 43-52

³Gopinath et al. Biochem J **204** (2007) p. 351-357

⁴Williams and Addona. Trends Biotechnol. **18** (2000) p. 45-48

Attachment Of Membrane To VesCap Surface

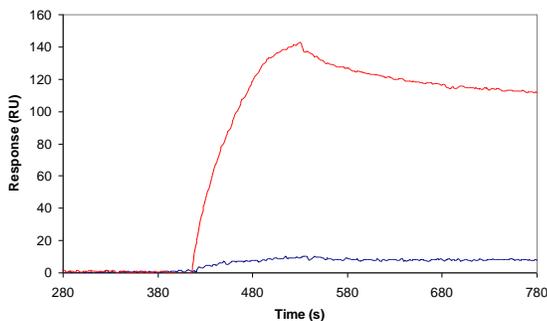
In the following example cell membrane fragments are immobilized. In brief, cells are lysed in hypotonic solution and the membrane recovered by centrifugation. The membrane pellet is washed to remove soluble protein and is then diluted in buffer. The membrane will spontaneously degrade producing small vesicles that have similar composition to the original parent cells. This preparation is sufficient for surface coating.



- Receptor-positive membrane solution was coated onto this surface yielding >2,000RU bound membrane vesicles as shown above.
- The surface was then exposed to HCL to stabilize the baseline by removing weakly attached material.
- Alternatively the HCl wash may be avoided if the surface is allowed to stabilize for 30 minutes prior to use.

In this example, a mass equivalent to 1000 RU of receptor membrane was immobilized. We recommend that a receptor negative membrane should be coated onto the reference sensing surface.

Demonstration of VesCap Chip Binding of Antibody to Membrane Associated Receptor



- As a control non-specific antibody (blue curve) yielded a negligible binding response of 8 RU.
- Anti-receptor antibody yielded a binding response of 136 RU.

The data set shown above is the product of reference curve subtraction where the sample response curve from the reference surface (coated with receptor negative membrane) was subtracted from the sample response curve over the receptor positive surface. Note that this procedure eliminates baseline drift yielding flat baselines that are necessary for accurate data analysis.

Sensiq Technologies, Inc.
800 Research Pkwy
Suite 100
Oklahoma City, OK 73104

T+ 1.405.239.8600
F+ 1.405.235.8608

www.sensiqtech.com