



Design of Cell Surface-Mimicking Materials as Advanced Cell Substrates

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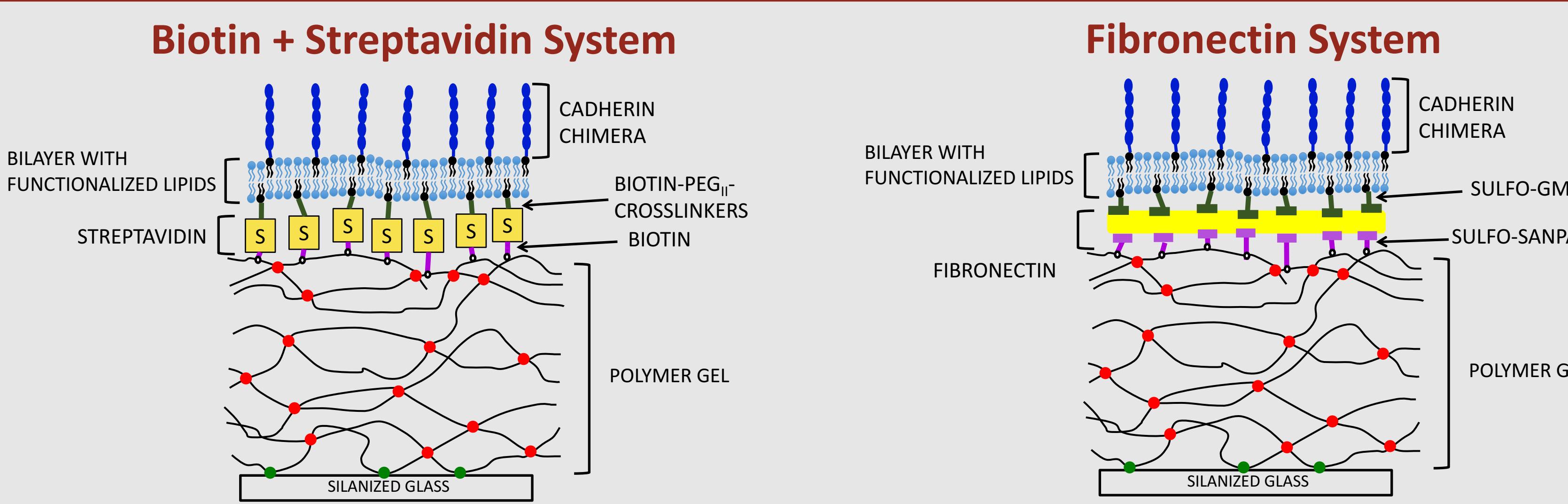
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Results

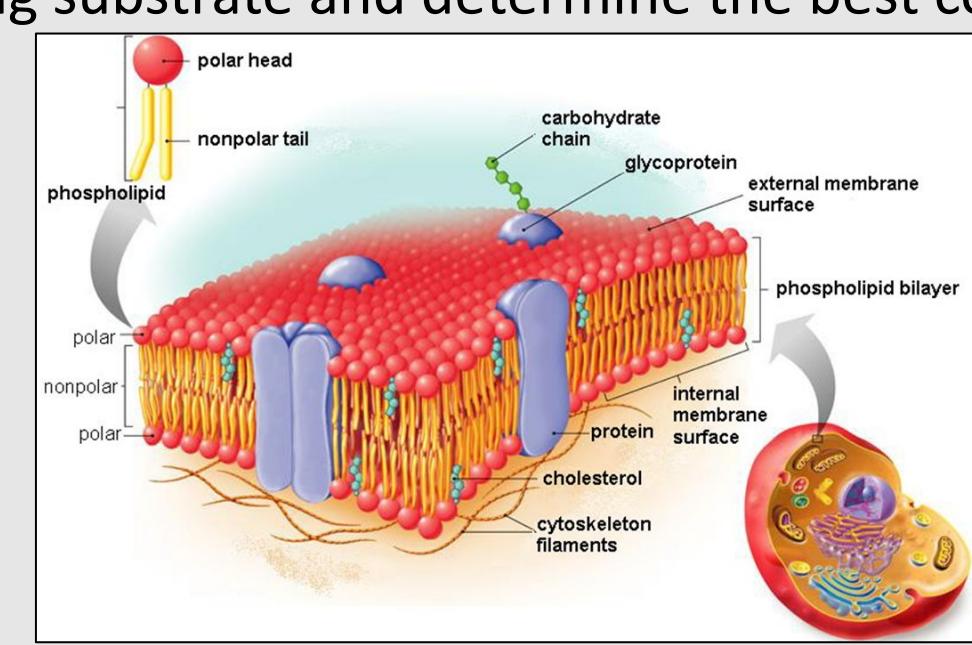
Abstract

In order to analyze major cell processes and gain a better understanding of their mechanics and physiology, cells should be effectively tested in an environment that most resembles the cell surface. The purpose of this design was to establish a cell substrate that parallels the environment in which cells naturally occur. In order to mimic the process of cell adhesion, the following essential constituents were used to construct a substrate: an acrylamide and bis-acrylamide monomer solution for a polyacrylamide (PAA) gel, a lipid bilayer made from a mixture of fluorescent, thiol, and saturated lipids and was connected to the gel through a series of cross linkers and proteins.

Background Information



PURPOSE: Design a cell surface-mimicking substrate and determine the best conditions or method to create polymer gel supported lipid bilayers.



Lipid Bilayer Formation

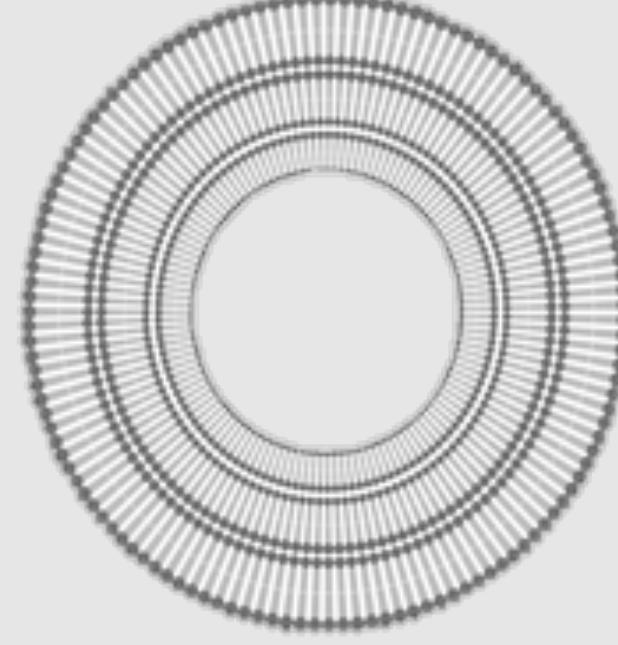


Figure 1 A Multilamellar Vesicle (MLV) is formed by one lipid bilayer making a spherical shape, called the vesicle.

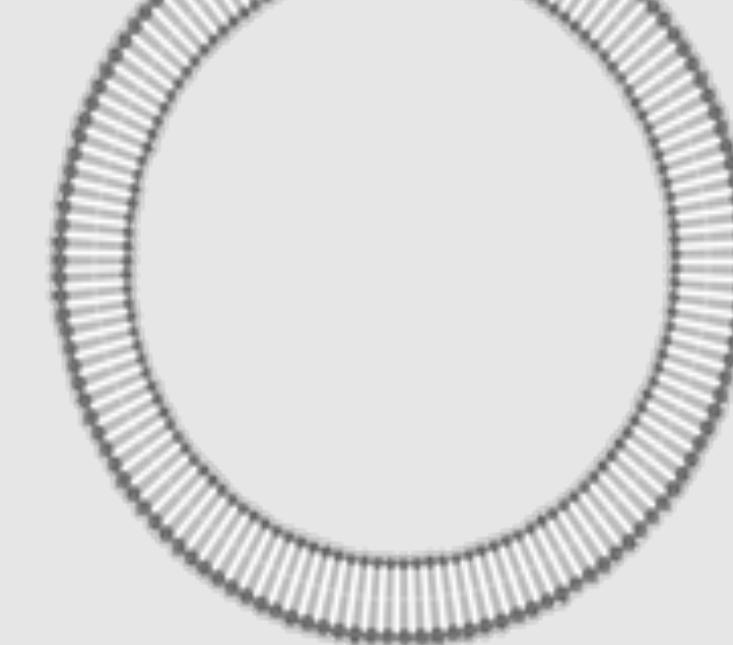


Figure 2 A Giant Unilamellar Vesicle (GUV) is formed by two lipid bilayers making a spherical shape, which is called the vesicle. The GUV is what actually bursts to create a fluid lipid bilayer.

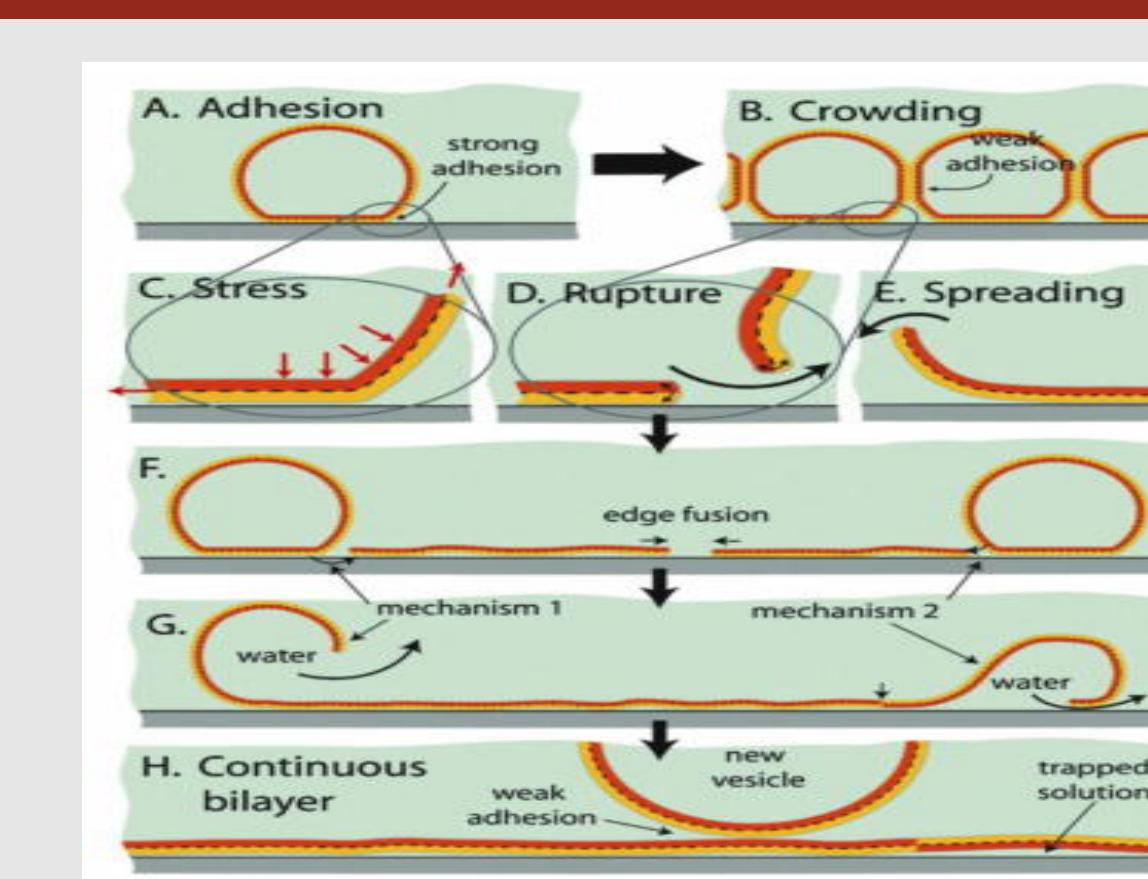


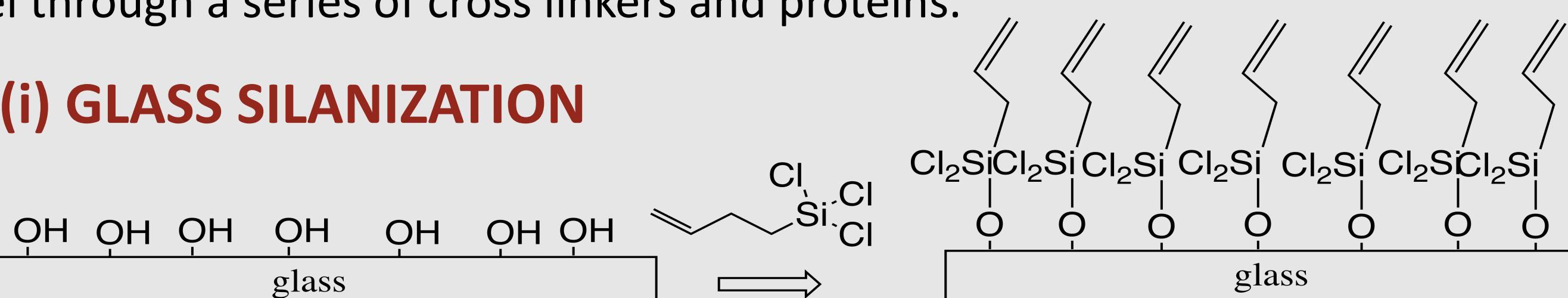
Figure 3 This diagram above demonstrates the steps of vesicle rupture leading to vesicle fusion resulting in a continuous bilayer.

When the lipid solution is first made and dried, the lipids that remain after drying are in the MLV form. After they are heated in a water bath, large chunks of lipid are visible, called the GUVs. These GUVs were used to create the lipid bilayers on polymer gels.

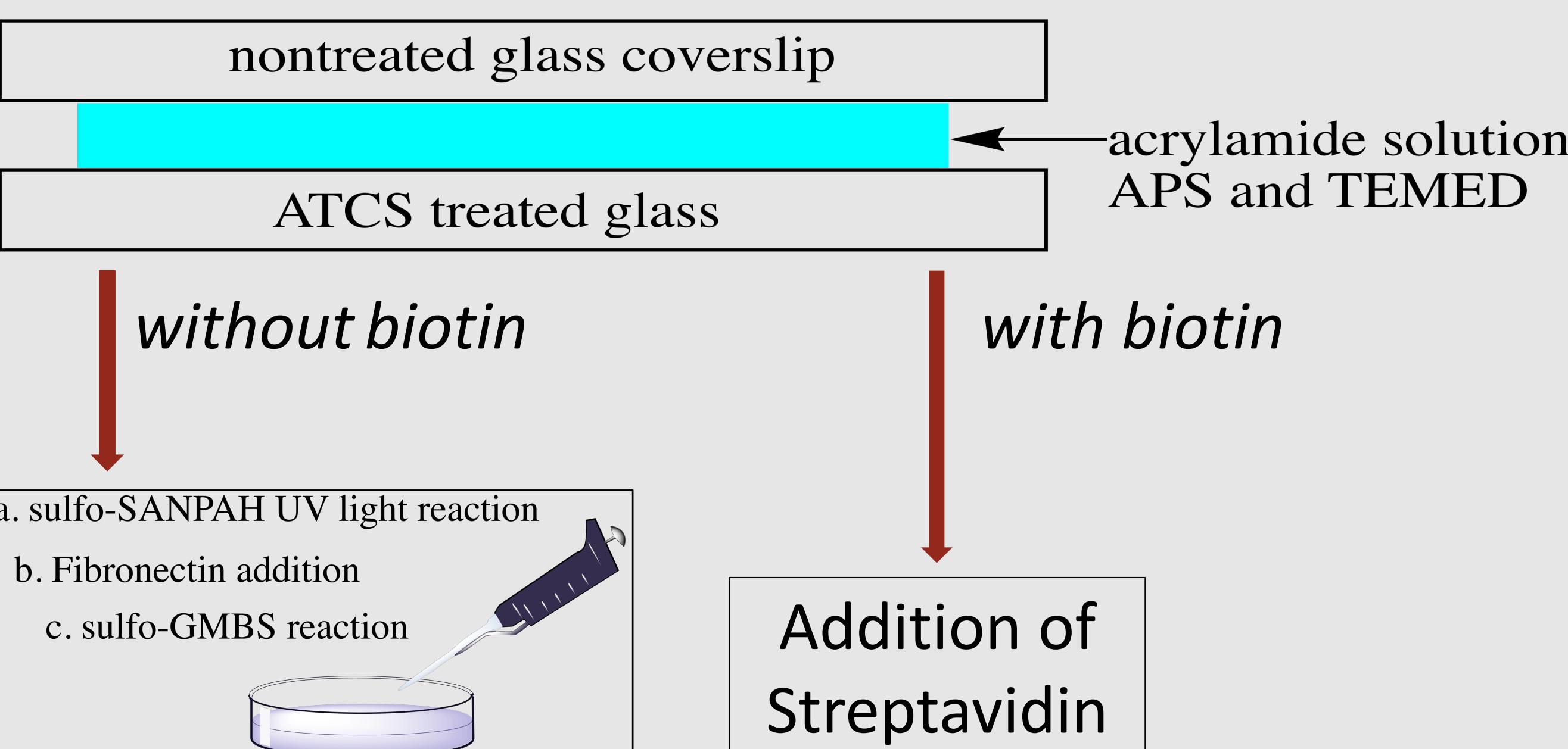
Method

Polyacrylamide gels were cast on silanized glass slides. The lipid bilayer consisted of fluorescent, thiol, and saturated lipids and was connected to the gel through a series of cross linkers and proteins.

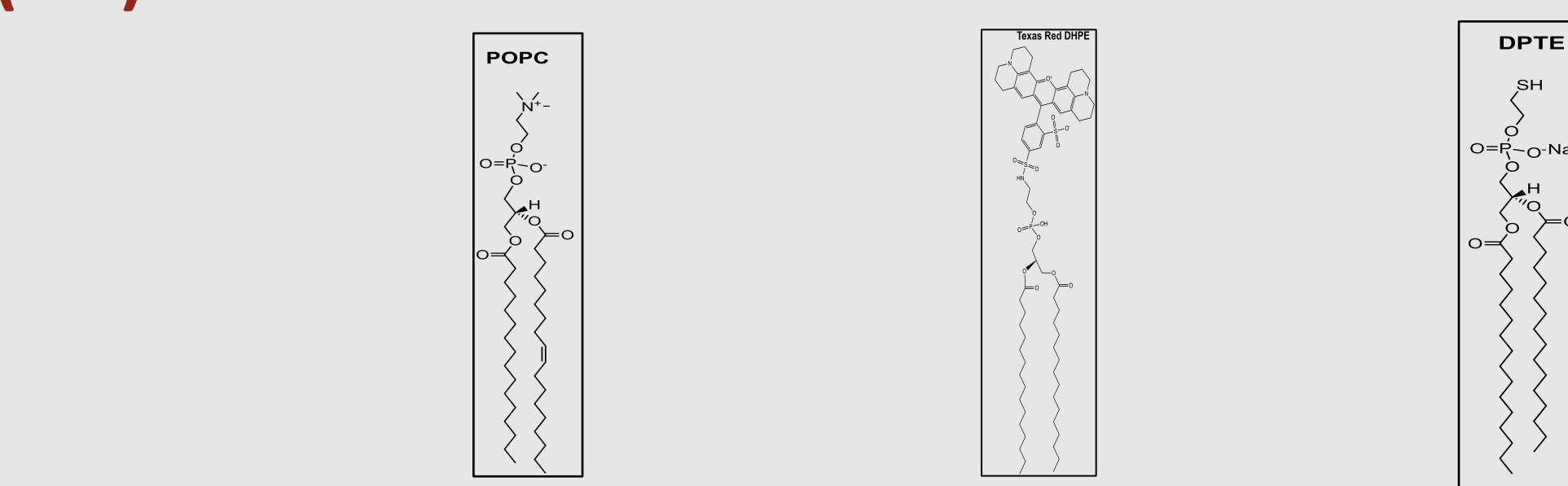
(i) GLASS SILANIZATION



(ii) POLYMERIZATION OF HYDROGEL



(iiia) BILAYER ADDITION- GUV VESICLES



(iib) α -HELICAL PEPTIDE SOLUTION ADDITION

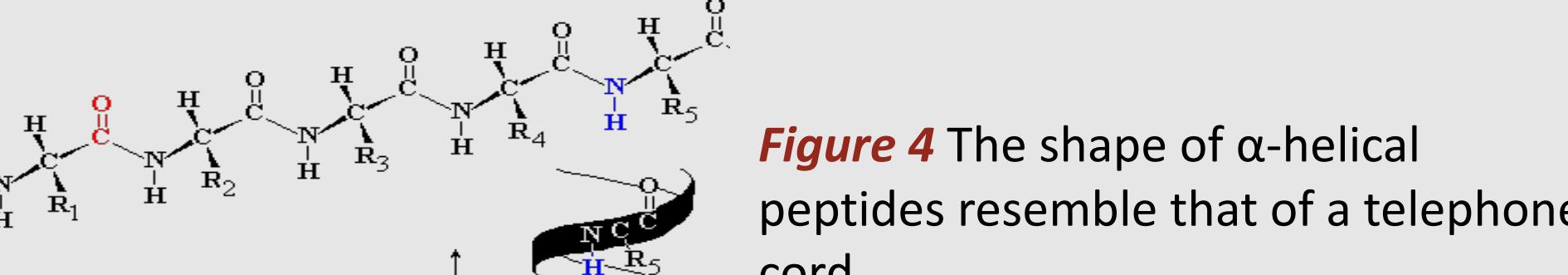


Figure 4 The shape of α -helical peptides resemble that of a telephone cord.

(iv) IMAGING



Figure 5 The Epifluorescence microscope was used to image the bilayers.

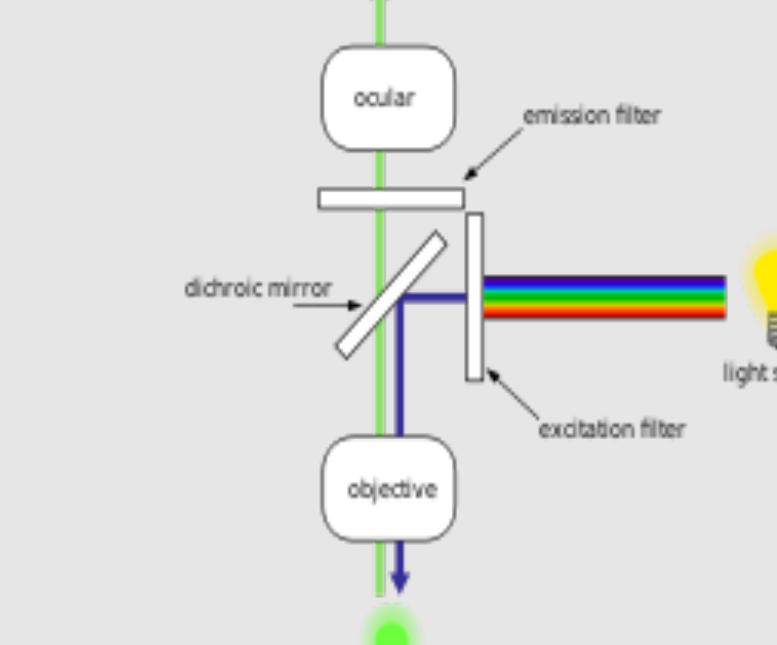
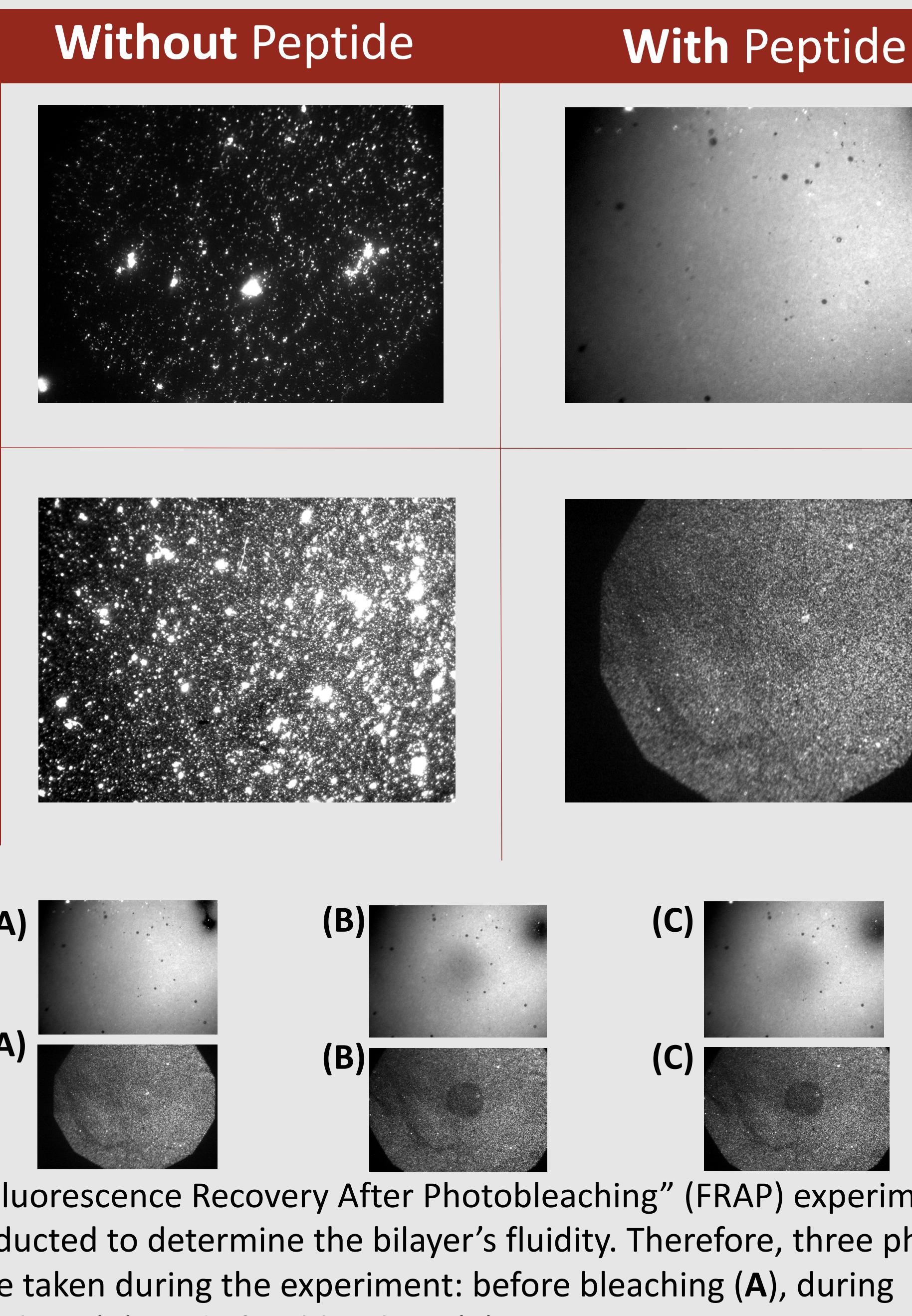
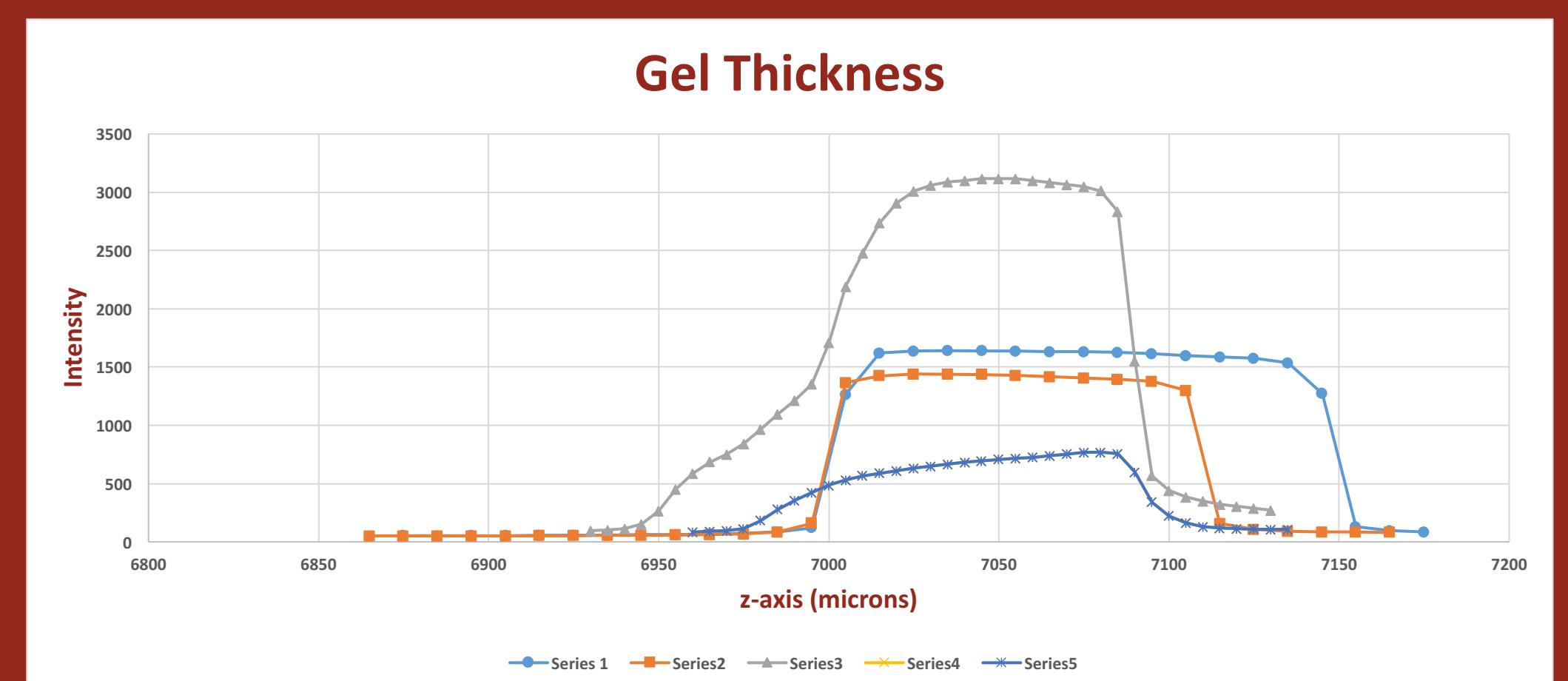


Figure 6 Above is a schematic of the microscope. Green fluorescent light was chosen to view the lipids.

References

- [1] G.J. Hardy et al. / Current Opinion in Colloid & Interface Science 18 (2013) 448-458



Conclusion

*Fibronectin Method

Because the microscope showed us that there was recovery through vesicle diffusion after photo bleaching, we could assume that our bilayer was fluid.

*Biotin + Streptavidin Method

It was observed that there was no recovery during FRAP, therefore it cannot be proven that the created lipid bilayer was fluid.

*Peptide Solution Addition

The addition of 30 μM peptide solution resulted in a more homogeneous lipid bilayer by ensuring more vesicle rupture. Therefore it can be concluded that the addition of a peptide solution is an important and helpful step to ensure a fluid and planar bilayer.

*Gel Thickness

Average: $106.4 \pm 25.8 \mu\text{m}$

Acknowledgements

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