Abstract

Hydrophobically modified carbohydrates were incorporated into CTAT/SDBS vesicles and the carbohydrate presence was quantified via sulfuric acid assay. Two different glucose conjugates were used to test the functionalization of vesicles. The first one, n-octylβ-D-glucoside did not incorporate into the vesicles. The second conjugate n-dodecyl-β-D-glucopyranoside had a 41.3% incorporation into the vesicles with a concentration of 28.1 µM. Hydrophobically modified glucose reacted with acrylonitile chloride in order to produce an acrylic ester. A TLC of this reaction mixture showed two major spots. TLC mass spec analysis shows peaks at 369.9 m/z and 424.0 m/z representative of the product. A TLC mass spec analysis of the cysteine conjugation showed a peak at 470.1 m/z, which is only 0.2% off from the expected peak for the monocysteine conjugate. The monocysteine conjugate was incorporated into bare vesicles with a concentration of 73.4 µM.

Background

Vesicles are formed spontaneously when two surfactants having oppositely charged polar heads are mixed in the correct ratio. These vesicles are soft and spherical. In this experiment vesicles were prepared with CTAT and SDBS in PBS. These particular vesicles form a lipid bilayer and have an interior lumen. They will be either cationic or anionic depending on which surfactant is used in excess. The anionic vesicles are about 150 nm in diameter and cationic vesicles are roughly 250 nm in diameter.

Vesicles are easy to functionalize by attaching molecules with a polar head and long hydrophobic tail, by anchoring the tail into the lipid bilayer. By doing this carbohydrates can be attached and built upon to attack larger proteins and peptides. Hydrophobically modified carbohydrates can be incorporated into the lipid bilayer of the vesicles. Their presence can be determined via lectin binding experiments (park et al.). These vesicle systems have potential for being vaccine platforms and drug delivery devices. This research mostly targets the use for vaccine platforms.

Other uses for these vesicles include the separation of charged organic solutes, vaccine and drug delivery, colloid and interface science.

Method

Mono and diester conjugates were made in a two step synthesis. In the first step octyl glucose and DMAP were dissolved in DCM and reacted with triethylamine and acrolyl chloride. The mixture produced in this step was then reacted with L-cysteine HCL and TCEP HCL, after being dissolved in PBS. NaOH was added to increase the pH to 7. The mixture from step one and step two were analyzed with TLC MS to verify the identity of the reactants.

Vesicles were formed by mixing SDBS and CTAT and the reaction mixture with the monoester in PBS and stirring overnight until vesicles were formed. Vesicles were purified using size exclusion column in 1.0 mg increments on 1.0 column.

Viability

Vesicles were formed by mixing SDBS and CTAT and the reaction mixture with the monoester in PBS and stirring overnight until vesicles were formed. Vesicles were purified using size exclusion column in 1.0 mg increments on 1.0 column.

Concentration

Vesicles were formed by mixing SDBS and CTAT and the reaction mixture with the monoester in PBS and stirring overnight until vesicles were formed. Vesicles were purified using size exclusion column in 1.0 mg increments on 1.0 column.

References

Kaler et al. Science 1989, 245, 1371-1374
Oppolzer et al. Helvetica Chimica Acta 1981, 64, 2802-2807

Conclusion

The dodecyl glucose conjugate was successfully incorporated into the SDBS/CTAT vesicles. This along with the desired monocysteine conjugate show promising results. The next step is to incorporate the monocysteine conjugate with the vesicles. This will then allow for further functionalization and may help with creating vaccine platforms that mimic the body’s cells with targeting agents that only attack specific targets. The monocysteine conjugation showed good incorporation into glucose vesicles with a concentration of 73.4 µM.