Acylation of electrospun chitosan membranes with medium chain fatty acids

Landon R. Choi[1], Carlos M. Wells[1], Zoe L. Harrison[1], Joel D. Bumgardner, PhD[1], Tomoko Fujiwara, PhD[2], J. Amber Jennings, PhD[1]

[1]University of Memphis Department of Biomedical Engineering, Memphis, TN

[2]University of Memphis Department of Chemistry, Memphis, TN

Statement of Purpose: Wounds resulting from musculoskeletal trauma can introduce infection and significant pain. Wound dressings can be loaded with therapeutics, such as local anesthetics or antimicrobials, and be used as wraps over soft tissue wounds to alleviate pain and prevent bacterial contamination. Chitosan nanofibrous membranes are well suited for tissue healing and drug delivery applications due to their increased surface area, high degree of biocompatibility, and ability to mimic the extracellular matrix. Previous work has demonstrated acylation of short chain fatty acids to protect nanofibers from swelling (1), with the finding that increased hydrophobic characteristics extended drug release of hydrophobic molecules. In this study, we investigated the characteristics of chitosan membranes modified through acylation reactions with hexanoic, octanoic, and decanoic acids through scanning electron microscopy and Fourier transform infrared (FTIR).

Methods: Membranes were electrospun using a 311.5 kDa chitosan (Primex) with a 71%-degree deacetylation. Chitosan was dissolved overnight at 5.5 (w/v) %, of 70% (v/v) trifluoroacetic acid & 30% (v/v) dichloromethane. Solution was centrifuged to remove particulates and transferred to syringe to be electrospun at a rate of 0.015 ml/min and a range of 14-26 kV with constant monitoring of the Taylor Cone to ensure good quality membranes. Membranes were spun to 15 cm diameters and ~ 0.7 mm (30 ml spinning solution) thickness. After membranes were fabricated, punch-outs of 1 cm diameter discs were made to be treated with pyridine and different anhydride solutions (Hexanoic-HA, Octanoic-OA, & Decanoic-DA). Membrane disks were weighed and divided by 5 mg/ml to calculate the total volume used for treatment. The ratio of solution, used for treatment, consisted of 1:1(pyridine:desired anhydride). Disks were washed 3 times in copious amounts of DI water, with a 10-minute rinse in 70% absolute ethanol after the first wash step. Treated membranes were observed via a PerkinElmer Frontier FT-IR spectrometer in Attenuated Total Reflectance (ATR) mode and imaged with Scanning electron microscopy (Nova NanoSEM650, FEI) to characterize fibers compared with untreated membranes.

Results: SEM images indicate that successful modification occurred, as minimal to no fiber swelling was observed (Figure 1). FTIR analysis also confirms the presence of ester bonding at peaks 1740 cm^{-1} and removal of TFA salts at peaks <1000 cm^{-1}, indicating successful acylation reaction for all three acyl lengths (Figure 2). Peaks at 2900 cm^{-1} also confirm the presence of acyl carbon chains at the surface of the membrane.

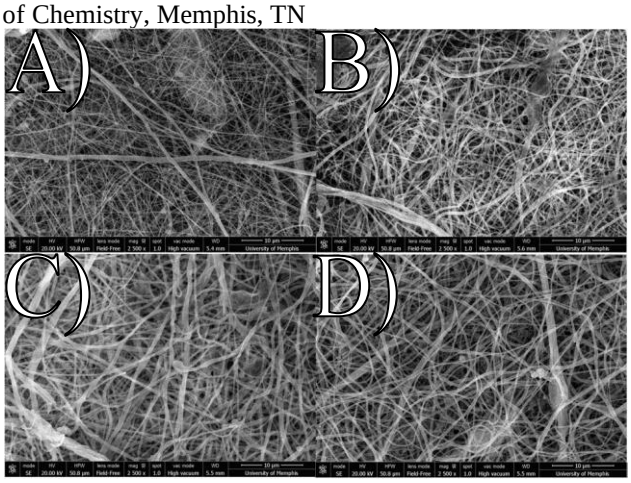

Figure 1.) SEM of chitosan (A) Untreated membranes, (B) HA treated membranes, (C) DA treated membranes, (D) OA treated membranes at 2500x.

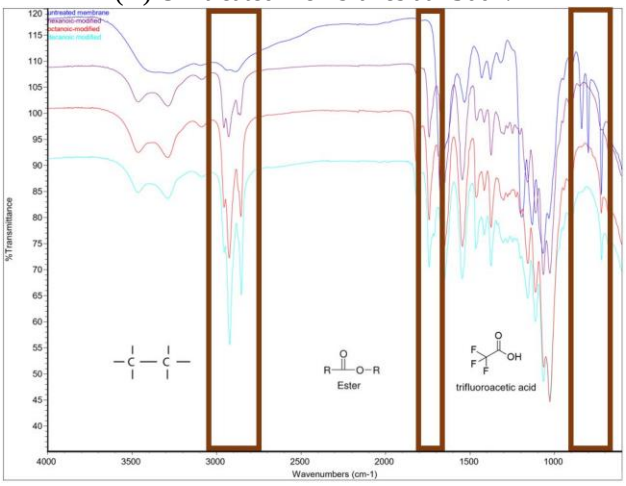

Figure 2.) FTIR of Untreated, HA, OA, DA chitosan membranes.

Conclusions: From observing the SEM images, untreated and treated membranes have similar nanofiber structures with minimal to no swelling. This preliminary study demonstrated that pyridine can be used to catalyze acylation of up to 10 carbon fatty acids on chitosan membranes. It further indicates that the fibers are modified by formation of ester bonds with -OH functional groups on chitosan. Future work will measure release of hydrophobic molecules such as bupivacaine and cis-2-decenoic acid from all 3 acyl length groups. Further, evaluations of cell compatibility and efficacy as wound dressing materials are planned.

Acknowledgements: Funding was provided by the Military Burn Research Program under award number: W81XWH-19-MBRP-IDA

References:

1. Murali V. Priya, et al *Inter J of Pharm.* (2020): 119438.