

## Hydrophobic Elastomeric Fiber Mats for Soft Tissue Engineering and Wound Care

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**Statement of Purpose:** Hydrophobic surfaces play an important role in promoting the adsorption of proteins and nucleic acids, and thereby encourage the formation of the lipid bilayer of a cell membrane to bind cells<sup>1</sup>. For medical purposes, hydrophobic surfaces can be applied as implanted wound patches to promote healing, and as tissue scaffolds to grow cells for tissue transplant and reconstruction. Electrospinning is a unique nano-processing technology<sup>2</sup> to manipulate surface properties, like wettability, by producing polymeric fibers with diameters in the range of tens of nanometers to microns. Dendritic poly(isobutylene-*b*-styrene) (D\_IBS) block copolymers are a promising class of biocompatible thermoplastic elastomers with a favorable combination of properties suitable for soft tissue implant devices<sup>3</sup>. This paper describes the creation of hydrophobic surfaces using electrospun D\_IBS fibers and the evaluation of fiber morphology, water contact angle and protein adsorption capability.

**Materials and Methods:** D\_IBS used in this study had a molecular weight ( $M_n$ ) of 220,300 g/mol, a polydispersity index of 1.87 and a polystyrene content of 29.4 wt%. The polymer was separately compression molded into 100- $\mu$ m thick film, and dissolved in a solvent mixture of tetrahydrofuran and toluene (95:5 w/w) at a polymer content of 10 wt% for electrospinning. In the electrospinning experiment, a positive voltage of 20 kV was applied via copper wired connections at a distance of 20 cm between the spinning glass pipette tip and a grounded metal collector. Samples were prepared for scanning electron microscopy (SEM) and water contact angle (WCA) evaluation.

For the protein adsorption study, polymer samples were incubated in buffers containing different proteins (insulin - 10  $\mu$ g/ $\mu$ l; ubiquitin - 10  $\mu$ g/ $\mu$ l; lysozyme 10  $\mu$ g/ $\mu$ l) separately for 24 hr at 25 °C in an incubator shaker. The pH of buffers was controlled at 4.4, 6.9 and 8.7. The isoelectric points (pI) of insulin, ubiquitin and lysozyme are at pH = 5.3, 5.2 and 11.0, respectively. After incubation, the samples were washed thrice with the buffer. Bound proteins were removed from the sample by adding 200  $\mu$ l of urea (6 M) and heating the mixture at 55 °C for 10 min. The protein solution was desalted by ZipTips C<sub>18</sub> before being analyzed with MALDI TOF/TOF mass spectrometry. Calibration curves were prepared to quantify the protein adsorption of the samples.

**Results:** The electrospun D\_IBS fibers assumed a circular cross section with an average diameter of 1.61  $\mu$ m (Fig. 1). The hierarchical fiber morphology in D\_IBS resulted in a high WCA of 122 ° (avg.), compared to 93 ° (avg.) for bulk film. Table 1 shows the adsorption of insulin on D\_IBS bulk polymer film and fiber mats. In a consistent agreement for all three proteins and across the pH range

of 4.4 to 8.7, the data show that electrospun fiber mats adsorb more protein than a polymer film due to a larger surface area per unit mass. The surface areas per unit mass of fiber mat and polymer film for a plane surface are estimated to be 360 and 220 cm<sup>2</sup>/g, respectively. Puskas et al. reported that lower adsorption occurs at the pH below and above the pI of the protein due to the electrostatic repulsion between the protein and surface<sup>4</sup>. In agreement with this finding, one can observe that adsorption on both the D\_IBS polymer film and fiber mat fell with pH for insulin and ubiquitin, but increased with pH for lysozyme.

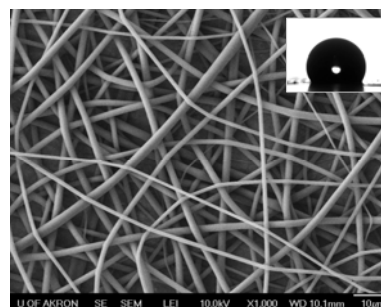


Figure 1. SEM and WCA images of D\_IBS fibers.

Table 1. Protein adsorption on D\_IBS materials.

Material	pH	Adsorption per unit area (pmol/cm <sup>2</sup> )		
		Insulin (pI = 5.3)	Ubiquitin (pI = 5.2)	Lysozyme (pI = 11.0)
D_IBS bulk polymer film	4.4	146.5	10.4	0.1
	6.9	102.7	8.5	0.7
	8.7	49.0	5.1	0.9
D_IBS fiber mat	4.4	355.9	40.1	1.1
	6.9	224.2	13.5	1.7
	8.7	79.0	6.2	3.3

**Conclusions:** Our study demonstrates the usefulness of electrospinning to increase the surface hydrophobicity of a biopolymer such as D\_IBS<sup>3</sup>. The large surface area per unit mass of the fiber mat allows an increased protein adsorption. Further effort is currently planned to study the cell and tissue interactions of hydrophobic D\_IBS fiber mats via *in vitro* and *in vivo* biological testing and explore relevant biomedical applications.

**Acknowledgements:** This work is supported by NSF under grants DMR #0804878 and #0821313.

### References:

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