

Supporting Information

Substantial differentiation of human neural stem cells into motor neurons on biomimetic polyurea

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+ These authors contributed equally to this work

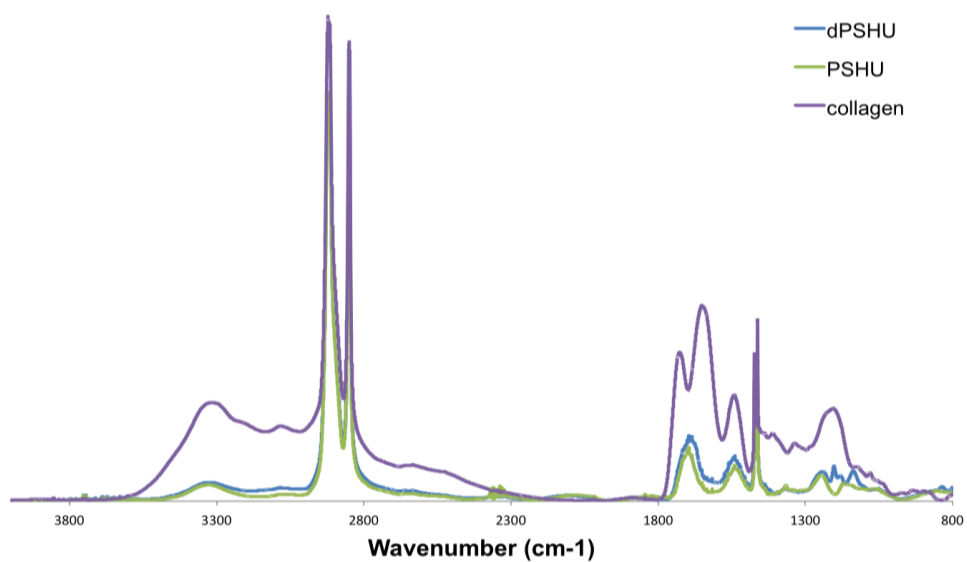


Figure S1. FT-IR of dPSHU (blue), PSHU (green), and collagen (purple).

Characterizations: We began by confirming and characterizing the synthesis process of Poly (serinol hexamethylene urea). NMR and FT-IR were both used to confirm the overall polymer structure and to detect any remaining BOC-protecting groups. NMR analysis was able to show complete deprotection of the BOC-protecting groups from the PSHU backbone as well as the expected polymer structure. The NMR analysis of the polymer structure can be seen in **Figure S2**. The NMR spectrum of PSHU confirmed the expected copolymer structure, with peaks at 1.3 (–CH₂–), 1.5 (–NH–CH₂–CH₂–), and 3.2 (–NH–CH₂–) associated with HDI, at 1.4 (–C–(CH₃)₃), and 4.1 (–CH–NH–) associated with N-BOC-serinol. You can see in the NMR spectrum that the deprotected PSHU does not show a peak at 4.1 confirming the removal of the N-BOC-serinol groups (**Figure S3**).

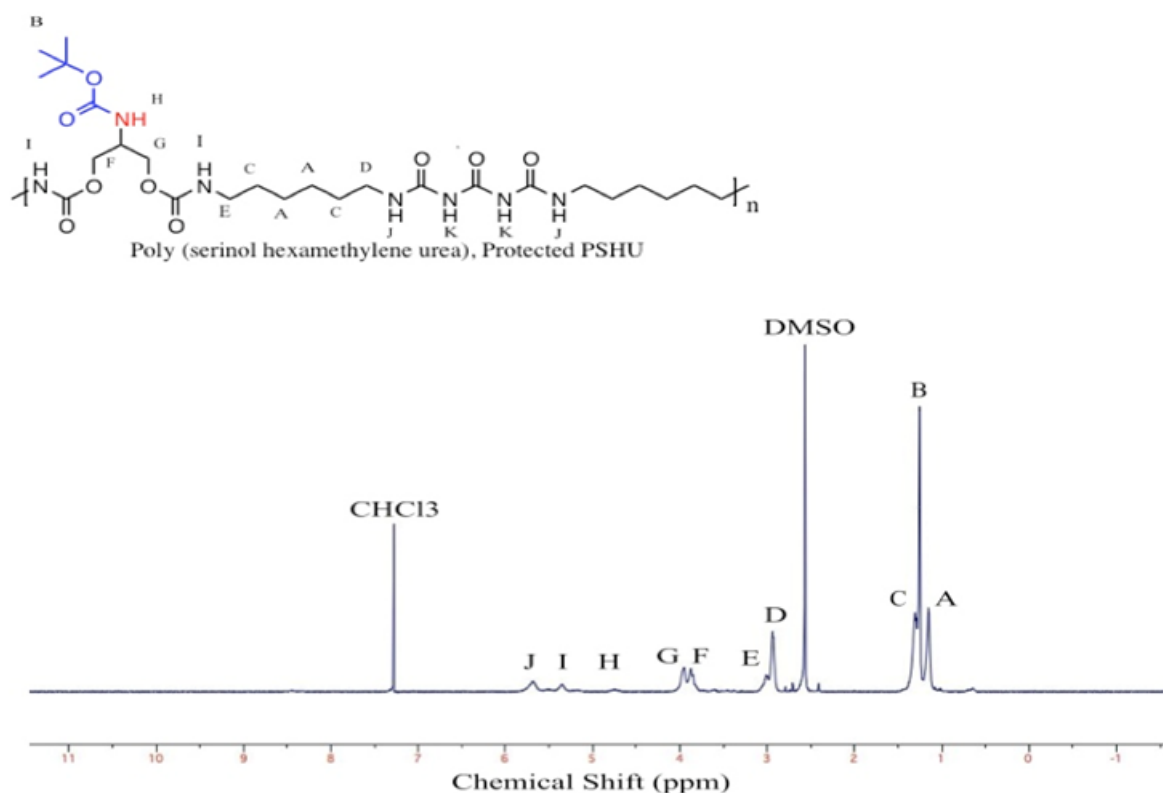
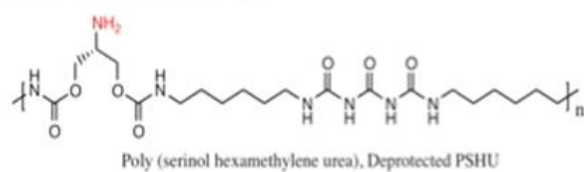


Figure S2. ¹H NMR (500 MHz, CDCl₃) spectrum of PSHU to confirm overall structure of polymer chain.

Removal of BOC Groups



Disappearance of peak at 1.4

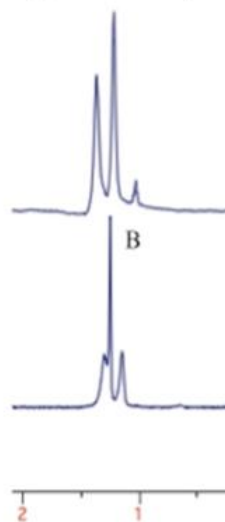
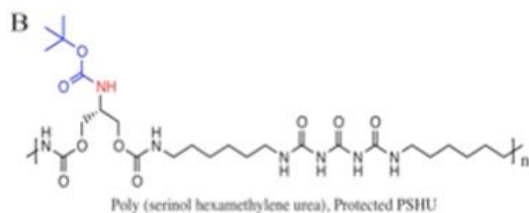


Figure S3. ¹H NMR (500 MHz, CDCl₃) spectrum of PSHU and deprotected PSHU to confirm removal of the BOC protecting groups during deprotection process.

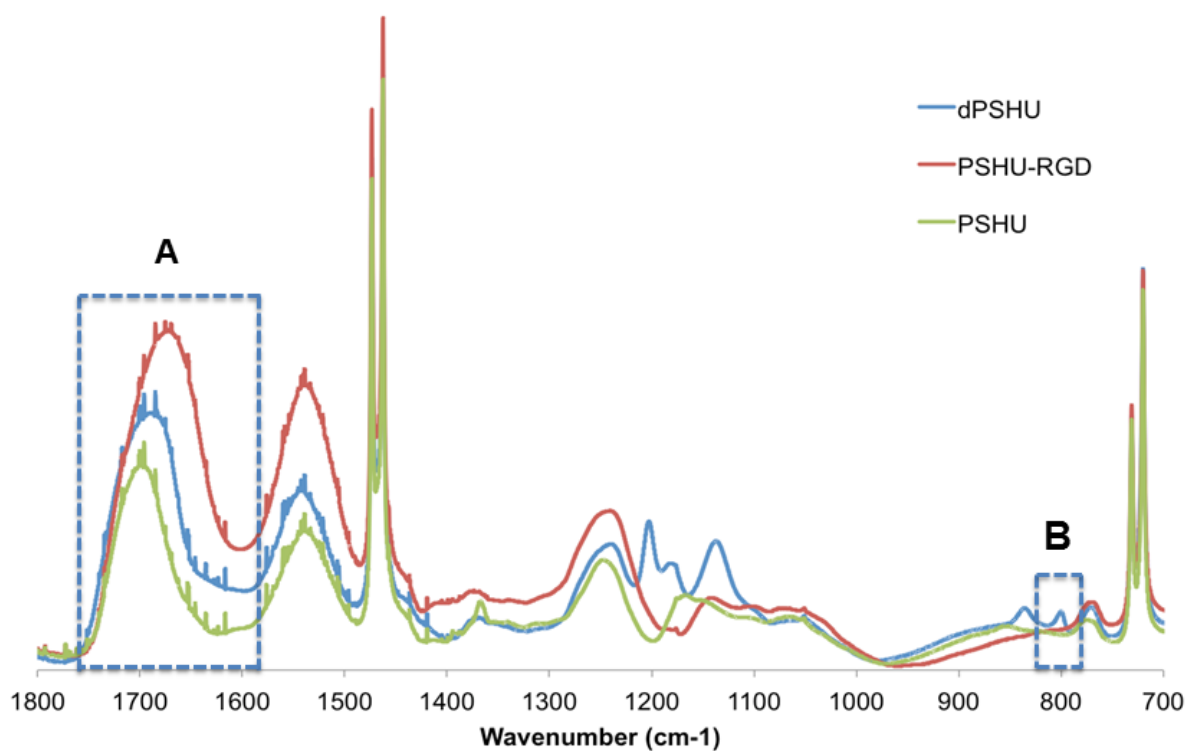


Figure S4. FT-IR of dPSHU, PSHU, and PSHU-RGD. Confirmation of free amine groups on dPSHU after deprotection (B). Shift in carbonyl absorbance to confirm attachment of RGD to the polymer backbone (A).

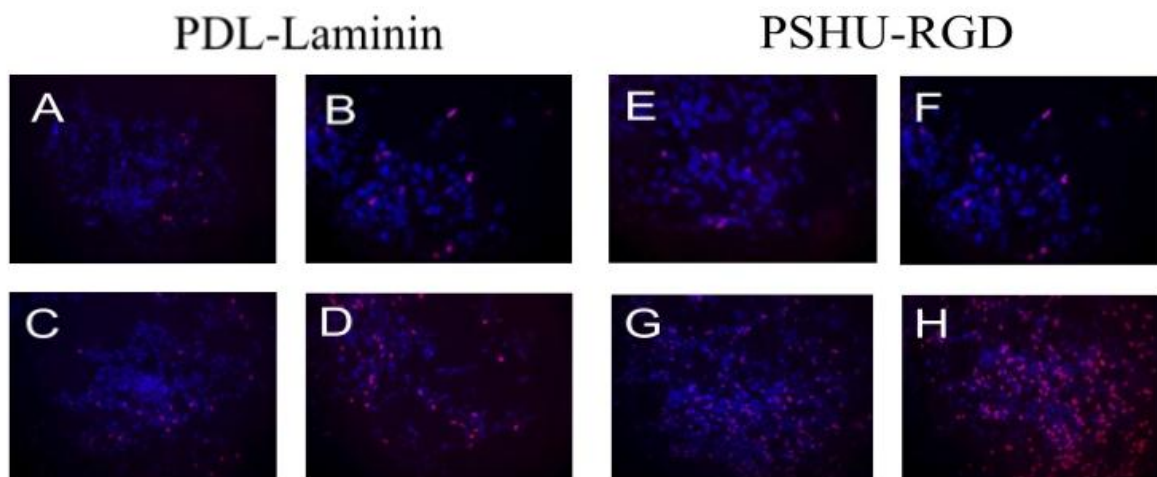


Figure S5. Fluorescent microscopy images of hNSCs cultured on PDL-Laminin(**A-D**) and PSHU-RGD (**E-H**). After culture for 14 days (**A,B,E,F**) and 21 days (**C,D,G,H**), cells were stained with Hoechst 3342 and HB9+ (**A,C,E,G**), and Hoechst 3342 and Isl1+ (**B,D,F,H**). Red dots indicate HB9+ and Isl1+ cells while blue dots indicate Hoechst 3342. These images were used to perform quantitative analysis comparing the ratio of cells expressing Isl1 and HB9 (Figure 4).

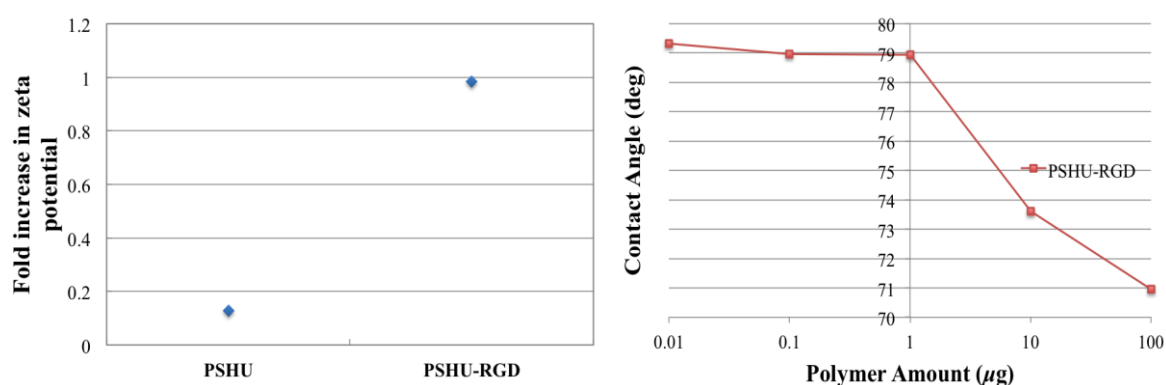


Figure S6. Left: Zeta potential measurements normalized to PSHU show an increase in charge with PSHU-RGD when compared to PSHU. Right: Contact angle shows an increase in hydrophilicity as the amount of PSHU-RGD is increase.

$$(1 \mu\text{g PSHU} - \text{RGD}) * \left(\frac{1 \text{ mole}}{9600 \text{ g}}\right) * \left(\frac{18 \text{ moles NH}_2}{1 \text{ mole PSHU} - \text{RGD}}\right) * \left(\frac{6.022e + 23 \text{ molecules}}{1 \text{ mole}}\right)$$

$$=1.12931e+15 \text{ RGD Biomolecules}$$

$$(4.5 \mu\text{g Laminin}) * \left(\frac{1 \text{ mole}}{400000 \text{ g}}\right) * \left(\frac{6.022e + 23 \text{ molecules}}{1 \text{ mole}}\right)$$

$$=6.77621e+12 \text{ RGD Biomolecules}$$

Equation S1. Calculations to determine theoretical density of integrin binding RGD biomolecules. 1 μg of PSHU-RGD coating possesses $\sim 1e+15$ RGD biomolecules whereas 4.5 μg of PDL-Laminin coating possesses $\sim 6e+12$ RGD biomolecules.