Electronic Supplementary Material (ESI) for Journal of Materials Chemistry B.

Supplemental Figure 1

Experimental: dsDNA was quantified by Quant-iT dsDNA PicoGreen assay (Invitrogen, Grand Island, NY). After incubating C2C12 cells for 3 days at 37°C, cells were lysed with 100 µL lysis buffer and frozen, thawed and sonicated three times to completely dissociate the cells. Cell lysates and standards were incubated in PicoGreen reagent and the fluorescence (ex 485 nm, em 528 nm) was measured (n = 3-5).

Results: After 3 days of incubation, no significant differences in C2C12 dsDNA content were seen between any groups.

Discussion: Treatment with soluble BMP-2, unloaded 10 wt% Hep MPs, or BMP-2 loaded MPs did not significantly affect the total DNA content of C2C12 cells over 3 days.

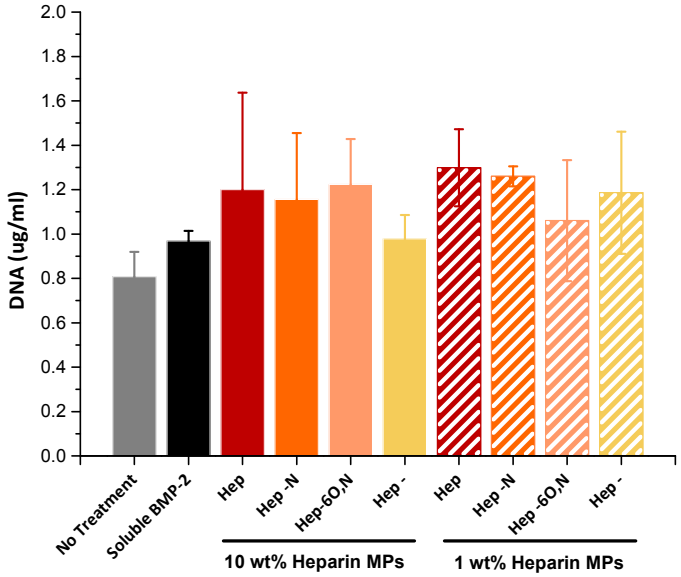

Figure S1. PicoGreen Assay for DNA quantification of C2C12 mouse myoblasts. No significant differences between any group; $p \leq 0.05$; n = 3-5.

Supplemental Figure 2

Experimental: 100 ng BMP-2 was incubated with 0.01 mg soluble Hep, Hep^{-N}, Hep$^{-6O,N}$, Hep-, or PEG-4Ac in 0.5 mL 0.5% v/v BSA PBS solution for 15 minutes or 24 hours at 4°C. BMP-2 was then quantified via ELISA (n = 3-5).

Results: After incubating heparin and BMP-2 for 15 minutes, the ELISA accurately detected approximately 100 ng BMP-2 in all heparin samples (Figure S2A). After incubating for 24 hours at 4°C, the detected levels of BMP-2 remained between 90-100 ng in Hep and Hep^{-N} samples but decreased to between 40-50 ng in Hep$^{-6O,N}$, Hep-, and no heparin samples, significantly lower than Hep and Hep^{-N} (Figure S2B).

Discussion: The ELISA accurately detected approximately 100 ng BMP-2 in samples with each heparin derivative after 15 minutes incubation, indicating that no derivative significantly interfered with the assay. After 24 hours incubation, the detected levels of BMP-2 were maintained in the more sulfated heparin samples (Hep and Hep^{-N}), indicating that these heparin derivatives protected BMP-2 over this timeframe, allowing for accurate detection via ELISA. In contrast, BMP-2 levels were significantly lower in the more desulfated heparin and no heparin samples, indicating that BMP-2 was not well protected by these derivatives.

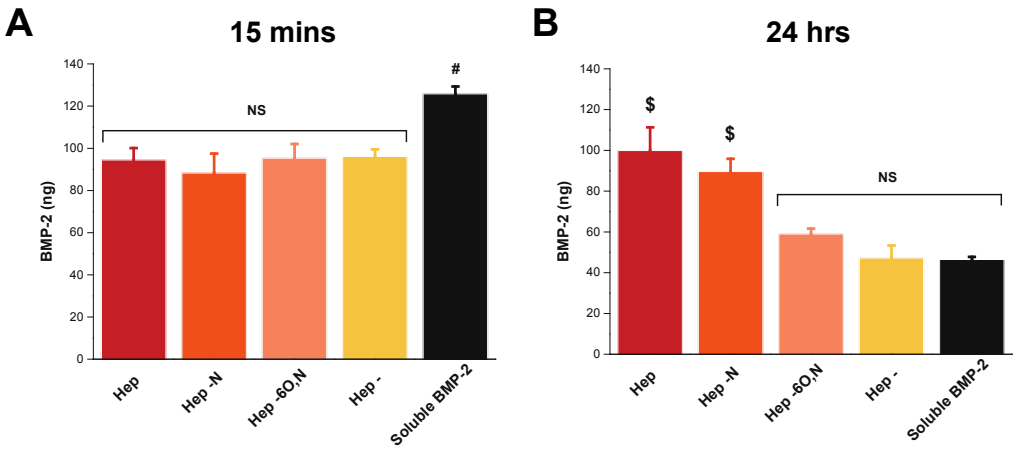

Figure S2. Detected levels of BMP-2 after incubation with soluble heparin derivatives. (A) After 15-min incubation, there was no difference between heparin groups. (B) After 24-hr incubation, there was significantly higher detected levels of BMP-2 in more sulfated heparin groups. [#]Significantly different than all other groups at 15 mins; $p \leq 0.05$. [$]Significantly different than all other groups at 24 hrs; $p \leq 0.05$; $n = 3\text{-}5$.