Supporting Information

Experimental:

Cytotoxicity via quantification of DNA content

Cells were seeded at 8,000 cells/cm^2 and allowed to adhere overnight. The next day, NP-siRNA treatments were prepared using non-targeting *Silencer* Negative Control siRNA 1 (Ambion) and cells were washed 2x with 1x DPBS. NP-siRNA treatments were added directly to wells containing fresh Opti-Mem media and incubated for 24 hours at 37 °C and 5% CO_2. Treated cells were washed 2x using 1x DPBS and lysed using 5x Luciferase Cell Culture Lysis reagent (Promega) diluted to 1x using dH_2O. Lysates were sonicated for 10 s using a tip sonicator to homogenize samples. 10 µL of cell lysate was diluted in 90 µL of 1x Tris-EDTA (TE) buffer and DNA content was quantified using Quant-iT PicoGreen dsDNA Assay Kit (Invitrogen) according to the manufacturer's protocol.

Determination of critical micelle concentration (CMC) by PRODAN assay

6-propionyl-2-(dimethylamino)naphthalene (PRODAN) was used to measure CMC as previously described, as the spectral emission of PRODAN displays a significant shift when partitioned into hydrophilic versus hydrophobic regions of NPs.[1, 2] Briefly, PRODAN was dissolved in methanol at 24 µM. 10 µL of PRODAN solution was loaded into each well of a black 96 well plate, and plates were incubated overnight in a chemical fume hood to allow the methanol to evaporate. The next day, NP were diluted using 1 x PBS at concentrations ranging from 0.1 ng/mL to 0.5 mg/mL, and 100 µL of each sample was loaded into the 96 well plates containing PRODAN. All samples were loaded in triplicate. Plates containing NPs and PRODAN were sealed and incubated at 4 °C overnight. PRODAN fluorescence was analyzed using a plate reader using 360 nM excitation and 435 nm emission for the hydrophobic and 520 nm emission for hydrophilic detection. The ratio of hydrophilic/hydrophobic emission was plotted against the $\log_{10}$ polymer concentration. The first data point above 20% of the average baseline was used to designate the first point of the hydrophobic regime, and the baseline was used as the hydrophilic regime. Each were fit using a linear regression, and the intersection of the two lines was defined as the CMC.

Results:

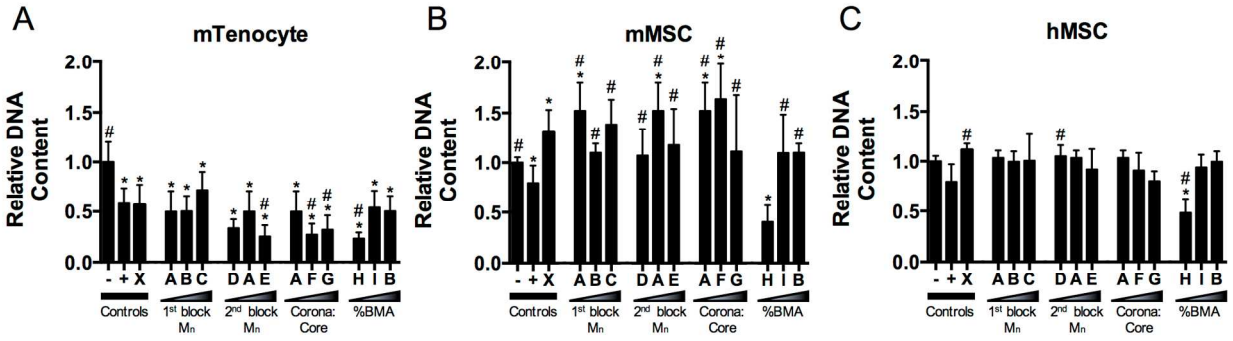

Figure S1: NP-siRNA treatments result in varying degrees of cytocompatibility as measured via relative quantification of DNA content. Murine tenocytes (mTenocyte) (A), murine MSCs (mMSC) (B), and human MSCs (hMSC) (C) were treated with 30 nM non-targeting negative control siRNA complexed to NP at a charge ratio of 4:1. DNA content was quantified as a measure of cytotoxicity and normalized to negative controls. *$p < 0.05$ compared to negative (-) control, $^{\#}p < 0.05$ compared to positive (+) control. Negative controls (-) are comprised of untreated cells and positive controls (+) are cells treated with 30 nM non-targeting negative control siRNA using Lipofectamine2000.

A

Multi-Parameter Regression Model of Relative DNA Content

	mTenocyte	mMSC	hMSC
p	0.10	0.05	**< 0.0001**
R^2	0.14	0.17	0.39

B

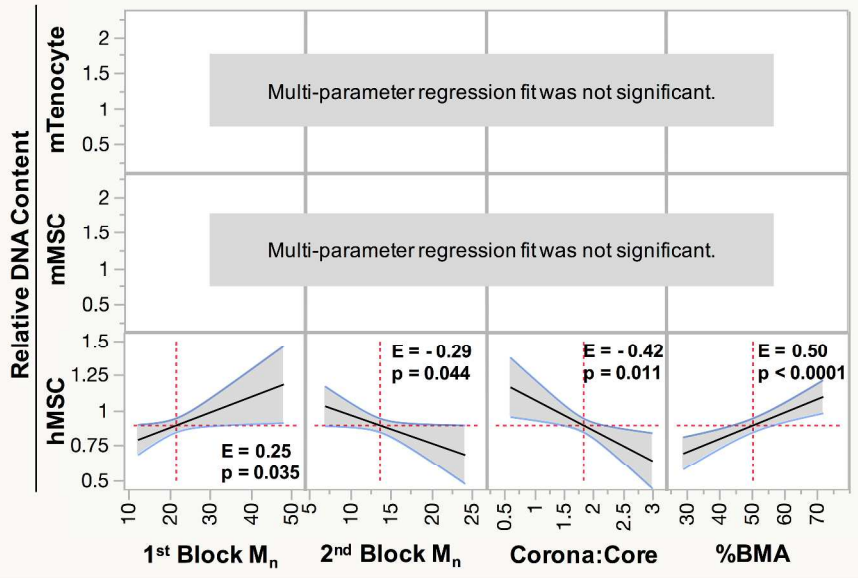

Figure S2: MPR analysis of relative DNA content. Relative DNA content in treated cells was fit using a multi-parameter linear regression to determine if cell viability is dependent on NP properties. A) p-values less than 0.05 (bold) indicates that cell viability is dependent on NP parameters, and the R^2 value describes what percentage of the behavior can be adequately described by the predictive model. B) Effect sizes (E) from significant regression models show the extent that a single property correlates with cell viability. $p < 0.05$ indicates the polymer property significantly influences cell viability. Significant effects are bolded.

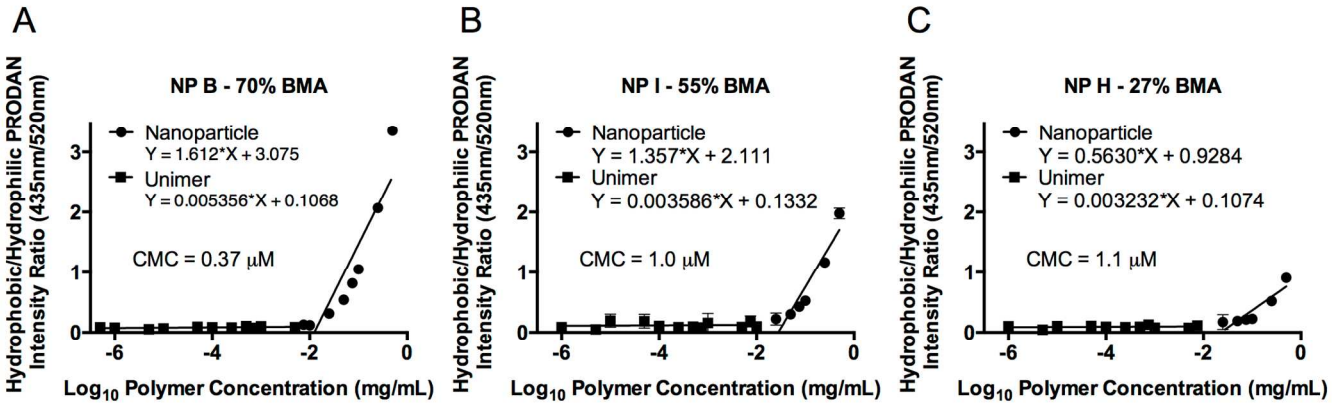

Figure S3: Determination of critical micelle concentration (CMC) via PRODAN assay of NP B (A), NP I (B), and NP H (C) containing varying amounts of BMA. PRODAN partitions into the hydrophobic micelle cores versus aqueous solution of the unimers. Therefore, CMC is calculated by finding the intersection

of the nanoparticle and unimer regimes via linear regression. n=3 from a single experiment run in triplicate. Error bars represent the standard deviation.

Table S1: Primer sequences used in this study.

Species	Gene	Forward Primer (5' → 3')	Reverse Primer (5' → 3')
Human (homo sapiens)	*PPIB*	GTCCGTCTTCTTCCTGCTG	CATCTTCATCTCCAATTCGTAGG
	GAPDH	GCAAGAGCACAAGAGGAAGAG	AAGGGGTCTACATGGCAACT
Mouse (mus musculus)	*β-Actin*	CCCCACTGAAGCCTACAAAA	GGGAGGCTCCTCCATTC
	Gapdh	AGGTCGGTGTGAACGGATTTG	TGTAGACCATGTAGTTGAGGT

Table S2: Pearson's correlation of MFI vs GAPDH Expression and DNA Content. Significant correlations are bolded ($p < 0.05$).

NP-siRNA Uptake (MFI) Pearson Correlations			
		GAPDH Expression	**DNA Content**
mTenocyte	r	-0.28	**0.89**
	R^2	0.08	**0.80**
	p	0.47	**0.001**
mMSC	r	-0.56	-0.19
	R^2	0.31	0.04
	p	0.12	0.10
hMSC	r	-0.23	0.55
	R^2	0.05	0.30
	p	0.55	0.59

Table S3: Pearson's correlation of polymer mass used in NP treatments vs DNA content and metabolic activity. Significant correlations are bolded ($p < 0.05$).

Polymer Mass Peasron Correlations			
		DNA Content	**Metabolic Activity**
mTenocyte	r	-0.35	-0.35
	R^2	0.12	0.12
	p	0.36	0.35
mMSC	r	0.08	-0.06
	R^2	0.01	0.004
	p	0.84	0.87
hMSC	r	0.02	0.10
	R^2	0.0004	0.01
	p	0.96	0.81

Table S4: Pearson's correlation of zeta-potential vs MFI, GAPDH expression, and DNA content. Significant correlations are bolded ($p < 0.05$).

NP Zeta-Potetial Pearson Correlations				
		MFI	**GAPDH Expression**	**DNA Content**
mTenocyte	r	0.08	0.37	-0.15
	R^2	0.01	0.14	0.02
	p	0.83	0.32	0.70
mMSC	r	0.53	0.12	-0.54
	R^2	0.28	0.01	0.29
	p	0.14	0.76	0.13
hMSC	r	0.13	0.46	-0.27
	R^2	0.02	0.21	0.07
	p	0.74	0.22	0.48

Table S5: Pearson's correlation of NP diameter vs MFI, GAPDH expression, and DNA content. Significant correlations are bolded ($p < 0.05$).

NP Diameter Pearson Correlations				
		MFI	**GAPDH Expression**	**DNA Content**
mTenocyte	r	0.57	-0.64	**0.82**
	R^2	0.32	0.41	**0.67**
	p	0.11	0.06	**0.01**
mMSC	r	-0.19	-0.40	0.48
	R^2	0.04	0.16	0.23
	p	0.62	0.28	0.19
hMSC	r	0.52	**-0.67**	**0.70**
	R^2	0.27	**0.44**	**0.49**
	p	0.15	**0.05**	**0.04**

References:

1. Wong, J. E.; Duchscherer, T. M.; Pietraru, G.; Cramb, D. T., Novel fluorescence spectral deconvolution method for determination of critical micelle concentrations using the fluorescence probe PRODAN. *Langmuir* **1999,** 15, (19), 6181-6186.
2. Baranello, M. P.; Bauer, L.; Benoit, D. S., Poly(styrene-alt-maleic anhydride)-based diblock copolymer micelles exhibit versatile hydrophobic drug loading, drug-dependent release, and internalization by multidrug resistant ovarian cancer cells. *Biomacromolecules* **2014,** 15, (7), 2629-41.