Supporting Information

Nanoparticle-Mediated Intracellular Delivery Enables Cryopreservation of Human Adipose-Derived Stem Cells Using Trehalose as the Sole Cryoprotectant

*Wei Rao[1,2], Haishui Huang[1,2,3], Hai Wang[1,2,4], Shuting Zhao[1,2], Jenna Dumbleton[1,2], Gang Zhao[5], and Xiaoming He[1,2,4]**

[1]Department of Biomedical Engineering

The Ohio State University, Columbus, OH 43210, US

[2]Davis Heart and Lung Research Institute

The Ohio State University, Columbus, OH 43210, USA

[3]Department of Mechanical Engineering

The Ohio State University, Columbus, OH 43210, US

[4]Comprehensive Cancer Center

The Ohio State University, Columbus, OH 43210, USA

[5]Centre for Biomedical Engineering, Department of Electronic Science and Technology,

University of Science and Technology of China, Hefei, Anhui 230027, China

Table S1. Encapsulation efficiency (EE) and loading content (LC) of propidium iodide (PI) with and without genipin crosslinking using the procedure given in Scheme 1

Feeding ratio PI : NPs	Genipin crosslinking	EE, %	LC, %
1 : 10	No	3.7 ± 0.9	0.4 ± 0.1
1 : 10	Yes	36.4 ± 5.8	4.1 ± 0.2

NPs: Pluronic F127-chitosan nanoparticles

Table S2. Encapsulation efficiency (EE) and loading content (LC) at different feeding ratios of free trehalose (fTre) to Pluronic F127-chitosan NPs in weight and the diameter of the resultant GNCs-encapsulated trehalose (nTre) determined by dynamic light scattering (DLS) at different temperature and pH values. Also shown are the data of size for empty GNPs.

Feeding ratio fTre : NPs	EE, %	LC, %	Diameter of GNPs/nTre, nm			
			pH 5		pH 7	
			22 °C	37 °C	22 °C	37 °C
0 :GNPs	N/A	N/A	82.2 ± 4.6	37.3 ± 2.2	28.0 ± 3.5	24.8 ± 0.4
1 : 50	63.5 ± 4.8	1.4 ± 0.1	38.1 ± 8.1	24.9 ± 2.1	25.8 ± 8.3	19.9 ± 3.3
1 : 20	59.1 ± 3.1	3.5 ± 0.2	57.0 ± 15.0	30.6 ± 3.6	24.0 ± 7.1	22.1 ± 5.6
1 : 10	40.6 ± 5.8	4.6 ± 0.4	91.5 ± 18.2	33.5 ± 1.5	26.8 ± 6.2	20.1 ± 2.4
1 : 1	4.9 ± 0.6	5.6 ± 0.6	93.4 ± 18.3	35.2 ± 3.1	28.5 ± 5.1	21.8 ± 2.9
10 : 1	1.3 ± 0.1	14.4 ± 1.5	105.4 + 14.5	47.0 ± 7.2	36.2 ± 2.6	28.7 ± 4.1
20 : 1	1.2 ± 0.1	26.6 ± 0.8	137.5 ± 16.2	49.5 ± 5.6	42.3 ± 6.1	29.5 ± 6.4

All data are presented as mean ± standard deviation and N/A represents " not applicable".

Table S3. Genes and the corresponding primers used for real time RT-PCR studies

Gene		Primer
Sox2	Forward	5'- CCC ACC TAC AGC ATG TCC TAC TC -3'
	Reverse	5'- TGG AGT GGG AGG AAG AGG TAA C -3'
Klf4	Forward	5'- GAA ATT CGC CCG CTC CGA TGA -3'
	Reverse	5'- CTG TGT GTT TGC GGT AGT GCC -3'
Nanog	Forward	5'- CCT CCT CCA TGG ATC TGC TTA TTC A -3'
	Reverse	5'- CAG GTC TTC ACC TGT TTG TAG CTG AG -3'
Oct4	Forward	5'- GAA ATT CGC CCG CTC CGA TGA -3'
	Reverse	5'- CAT AGT CGC TGC TTG ATC GCT TG -3'
GAPDH	Forward	5'- GAG TCA ACG GAT TTG GTC GTA -3',
	Reverse	5'- ATG GGT GGA ATC ATA TTG GAA -3'