Supplementary Information

Enhanced Gene Transfection Efficiency of PDMAEMA by Incorporating Hydrophobic Hyperbranched Polymer Cores: Effect of Degree of Branching

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1. Preparation and Characterization of PEHO-g-PDMAEMAs

The synthsis scheme of PEHO-g-PDMAEMAs is shown in Fig. S1. Firstly, PEHOs with different degree of branching (DBs) ranging from 0.48 to 0.07 were synthesized by CROP of EHO in the presence of $BF_3 \cdot Et_2O$ at different reaction temperature, and the higher was the reaction temperature, the higher was the DB.[1] Then, the ATRP macroinitiators of PEHOs-Br were obtained via esterification reaction of PEHOs with 2-bromoisobutyryl bromide.[2] Finally, the PEHOs-Br with different DBs were used as the macroinitiators to initiate the ATRP of DMAEMA monomers to obtain PEHO-g-PDMAEMA copolymers. The detailed characterization data of PEHO cores and PEHO-g-PDMAEMAs are presented in Table S1-2. $D_{m\text{-}n}$ was used as the sample name, where m means the DB of PEHO cores, and n means the PDMAEMA arm length. The detailed syntheses and characterizations PEHOs and PEHO-g-PDMAEMAs should be referred to our previous publications.[1-4] PDMAEMA homopolymer was directly synthesized by atom transfer radical polymerization (ATRP) using ethyl 2-bromoisobutyrate as initiator.[2]

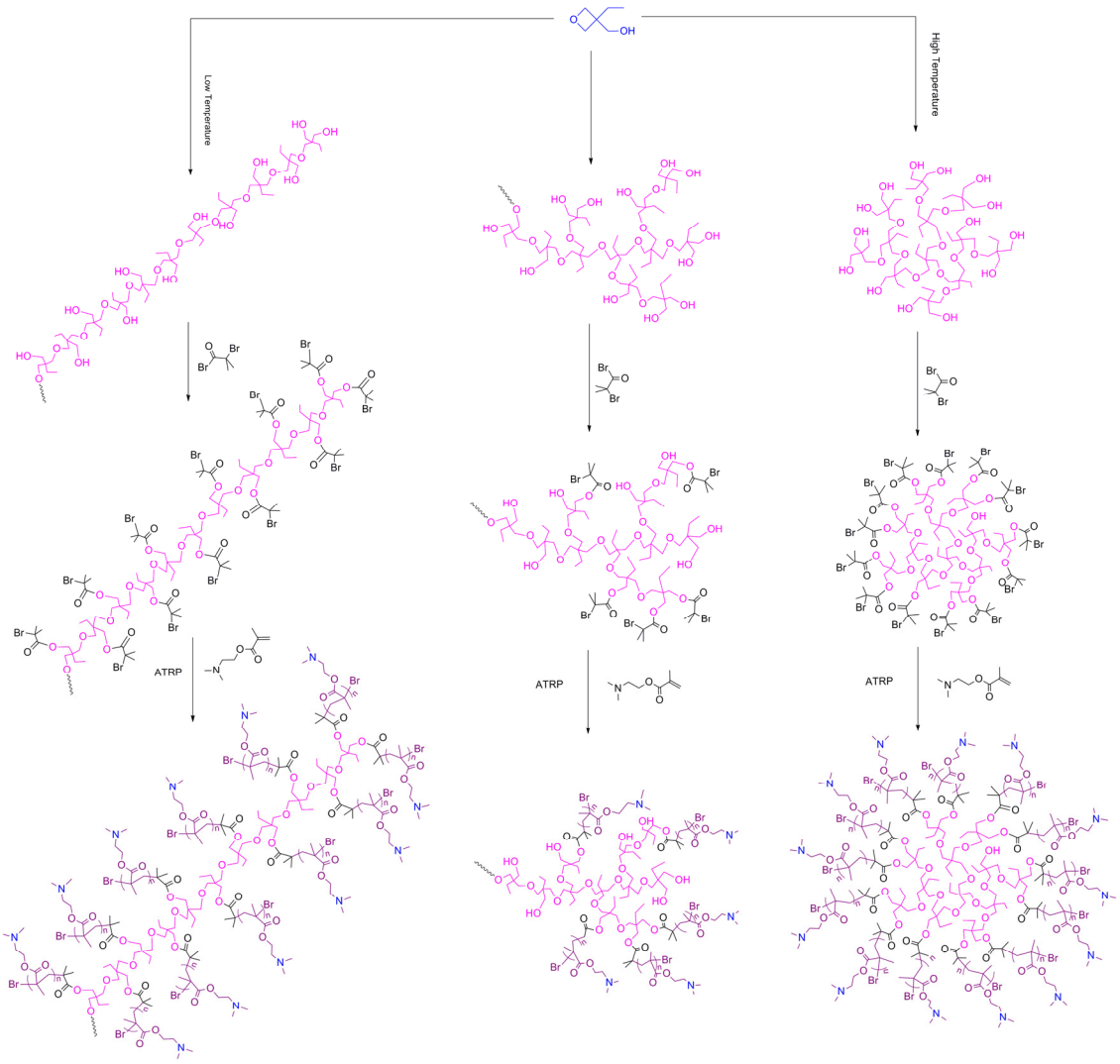

Fig. S1 Preparation of PEHO-g-PDMAEMAs with DB-variable PEHO cores.

Table S1. Characterizations of PEHOs[4]

Sample	Reaction temp (°C)	DB (%)[a]	M_n[b]	Polydispersity[b]
1	30	48	5,400	1.81
2	0	35	5,600	1.82
3	-25	23	6,500	1.90
4	-50	7	6,800	1.87

[a] DB (%) is determined by ^{1}H NMR.

[b] Determined by SEC.

Table S2. Characterizations of PEHO-g-PDMAEMAs[4]

Sample ($D_{m\text{-}n}$)	DB_{core} (%)	Arm length[a]	M_n[b]	Polydispersity[b]
$D_{0.48\text{-}4}$	48	4	10,800	1.73
$D_{0.35\text{-}4}$	35	4	16,700	1.78
$D_{0.07\text{-}4}$	7	4	21,600	1.83
$D_{0.48\text{-}7}$	48	7	19,500	1.75
$D_{0.35\text{-}7}$	35	7	22,700	1.79
$D_{0.07\text{-}7}$	7	7	30,800	1.77
$D_{0.48\text{-}12}$	48	12	40,300	1.68
$D_{0.35\text{-}12}$	35	12	40,700	1.78
$D_{0.23\text{-}12}$	23	12	43,800	1.85
$D_{0.07\text{-}12}$	7	12	47,800	1.76

[a] Arm length is determined by ^{1}H NMR.

[b] Determined by SEC.

2. Zeta potential Measurements of PEHO-g-PDMAEMAs in PBS buffer

The ζ-potentials of PEHO-g-PDMAEMAs in PBS buffers were measured at 25℃ on a Malvern Zetasizer NanoS equipped with a DTS5001 cell. The polymer solutions were directly filled into the cuvette before the measurements, and the measurements were performed in the ζ-model for a minimum of 10 cycles and a maximum of 100 cycles.

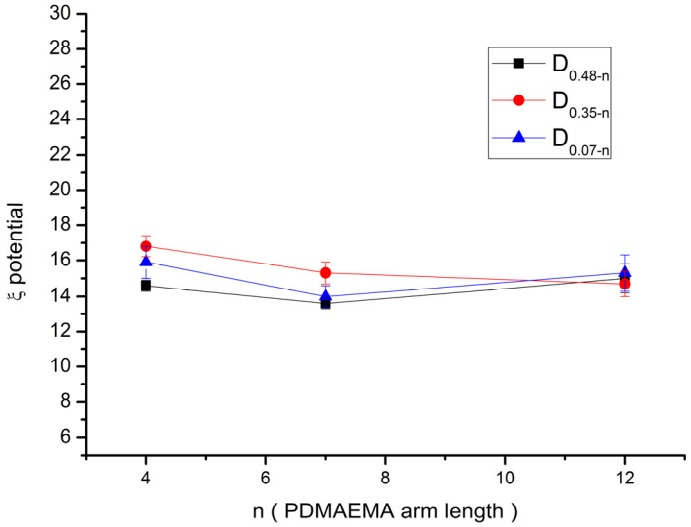

Fig. S2 ζ-Potential of PEHO-g-PDMAEMAs solutions.

[a] Data represent mean standard deviation (n = 3)

3. DLS measurements of PEHO-g-PDMAEMAs/pDNA complexes

Preparation of copolymer/pDNA complexes was based on the above procedures. The complex solution at N/P ratio of 10 was diluted with PBS (pH=7.4) to a final sample concentration of about 0.2 mg/mL. The particle size was measured at 25 ^{o}C by using a Malvern Zetasizer NanoS. The results (Fig. S3) indicate the size of the complexes decreases from 257 nm to 95.3 nm with the increase of DB from 0.07 to 0.48, which agree well with the statistical size distribution from the AFM image (Figure 4 in main text). It should be noted that the size of the complexes measured from AFM images is smaller than that measured from DLS due to the collapse of the particles in the dried state for the AFM measurements.

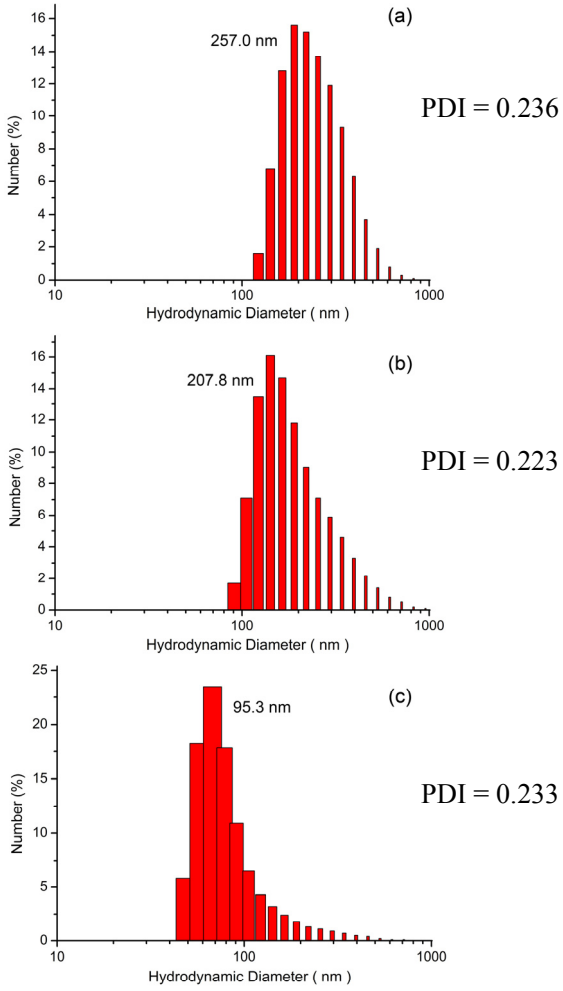

Fig. S3 Particle size distributions of copolymer/pDNA complexes with different DBs in PEHO-g-PDMAEMA (DB=0.07(a), 0.35(b), and 0.48(c) respectively) at N/P =10.

4. Cytotoxicity evaluation

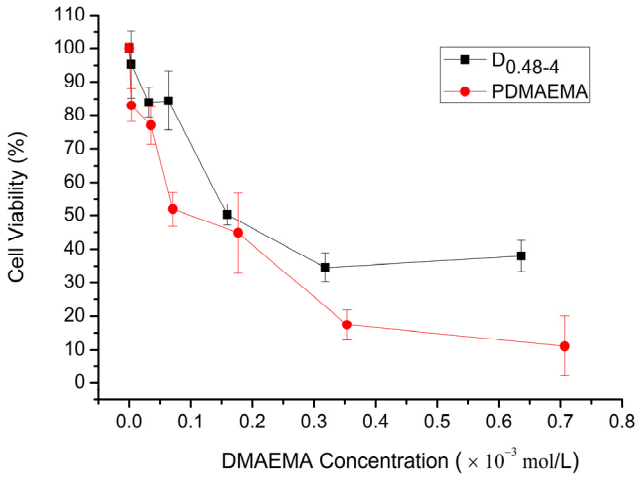

Fig. S4 Comparison the cytotoxicity of $D_{0.48\text{-}4}$ and PDMAEMA(6000Da) homopolymer at different DMAEMA concentrations in polymers in vitro.

3. References

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(4) H. Cheng, S. Xie, Y. Zhou, W. Huang, D. Yan, J. Yang, and B. Ji, *J. Phys. Chem. B.*, 2010, **114**, 6291.