Supplementary Information

Light and reduction dual sensitive supramolecular self-assembly gene delivery system based on poly(cyclodextrin) and disulfide-containing azobenzene-terminated branched polycations

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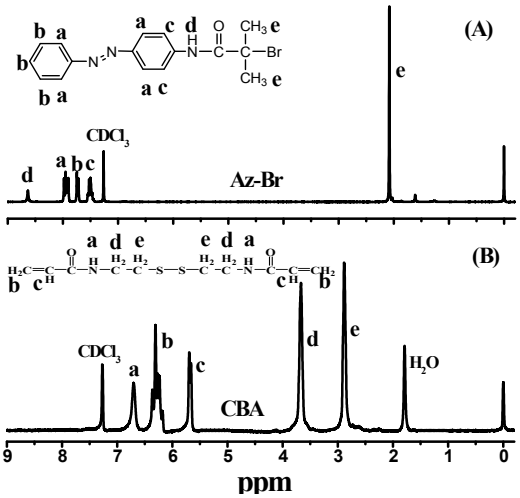


Fig. S1 ^{1}H NMR spectra of (A) tert-butyl-2-bromo-4-phenylazoanilide (Az-Br) and (B) *N,N*-bis(acryloyl)cystamine (CBA) in $CDCl_3$ at 25°C.

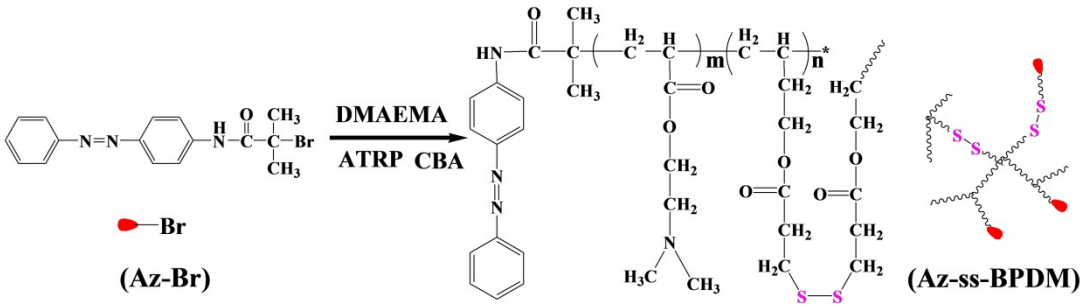


Fig. S2 Schematic preparation process of the Az-ss-BPDMs. Az-Br, *tert*-butyl-2-bromo-4-phenylazoanilide; CBA, *N,N'*-bis(acryloyl)cystamine; DMAEMA, (2-dimethylamino)ethyl methacrylate; Az-ss-BPDM, disulfide-containing azobenzene-terminated branched poly((2-dimethylamino) ethyl methacrylate); ATRP, atom transfer radical polymerization.

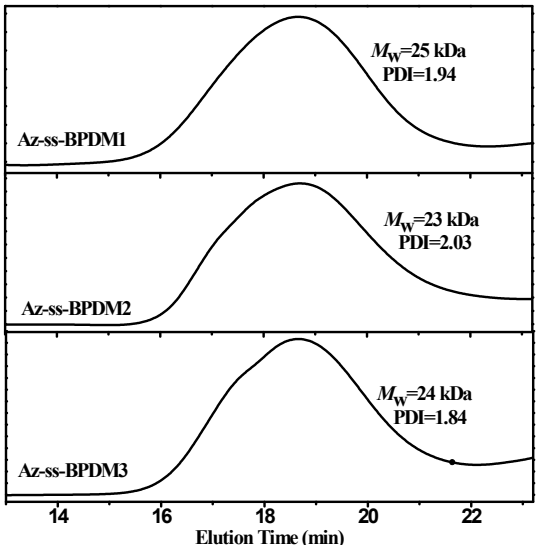


Fig. S3 SEC trace of the three azobenzene-terminated branched polymers in THF with similar M_w.

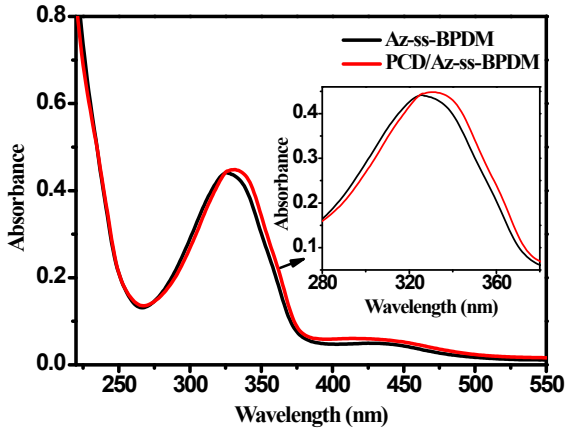


Fig. S4 UV-Vis spectra of (A) Az-ss-BPDM and PCD/Az-ss-BPDM (CD/Az 2:1).

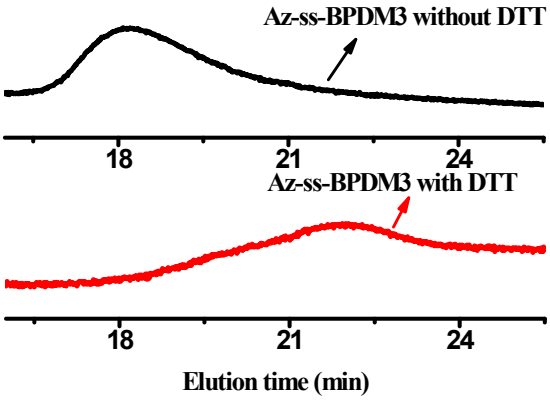


Fig. S5 Aqueous SEC traces (light scattering detection at 90°) of Az-ss-BPDM3 incubated with or without 100 mM DTT.

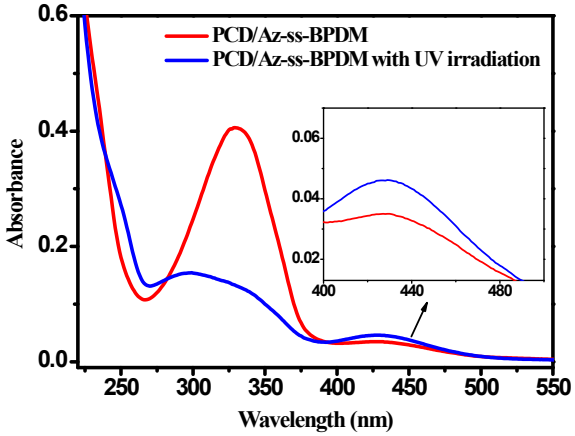
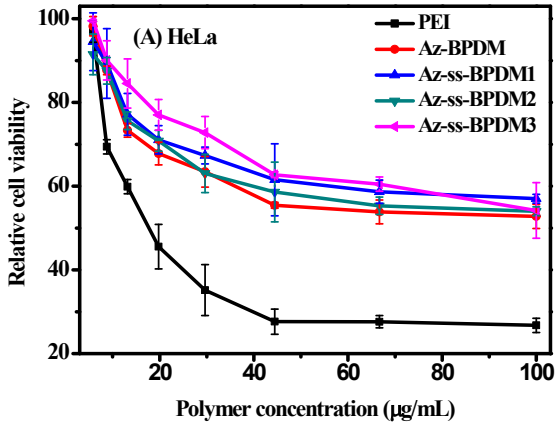
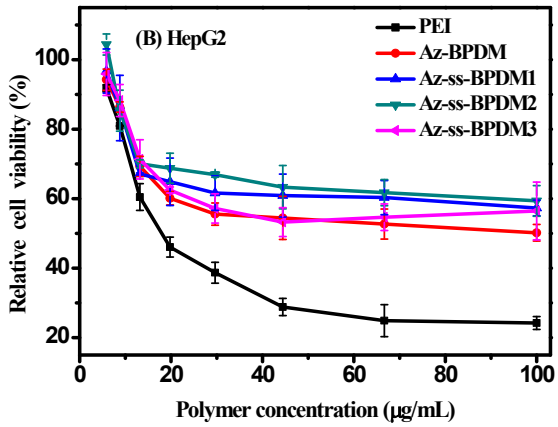


Fig. S6 UV-Vis spectra of PCD/Az-ss-BPDM before and after UV irradiation of 15 min.





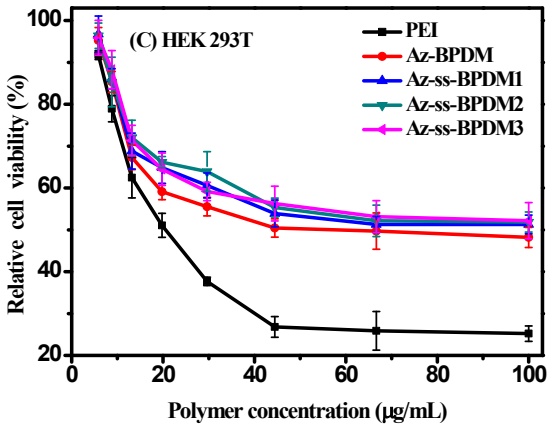


Fig. S7 Cell viability in (A) HeLa, (B) HepG2 and (C) HEK 293T cell lines with different concentrations of azobenzene-terminated polycations in the absence of PCD and PEI (25 kDa). Data was shown as mean ± SD (n=3).

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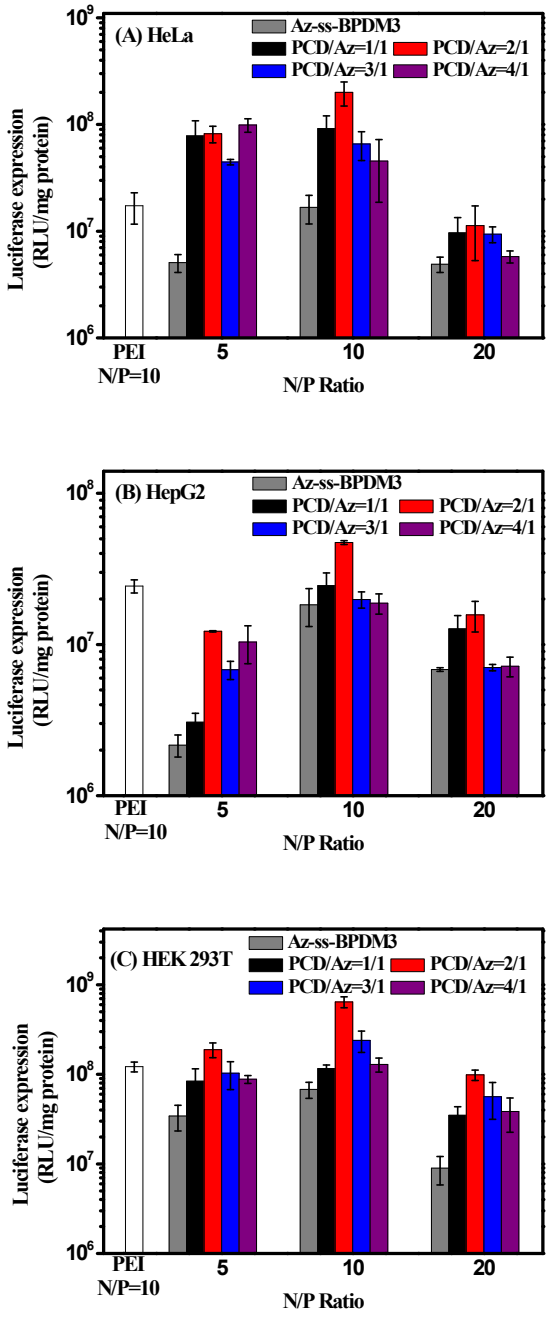

Fig. S8 *In vitro* luciferase protein expression of Az-ss-BPDM3/DNA polyplexes with different PCD content in serum media at various N/P ratios in comparison with that PEI/DNA polyplexes at N/P ratio of 10 in (A) HeLa, (B) HepG2 and (C) HEK 293T cell lines. Data was shown as mean ± SD (n=3).

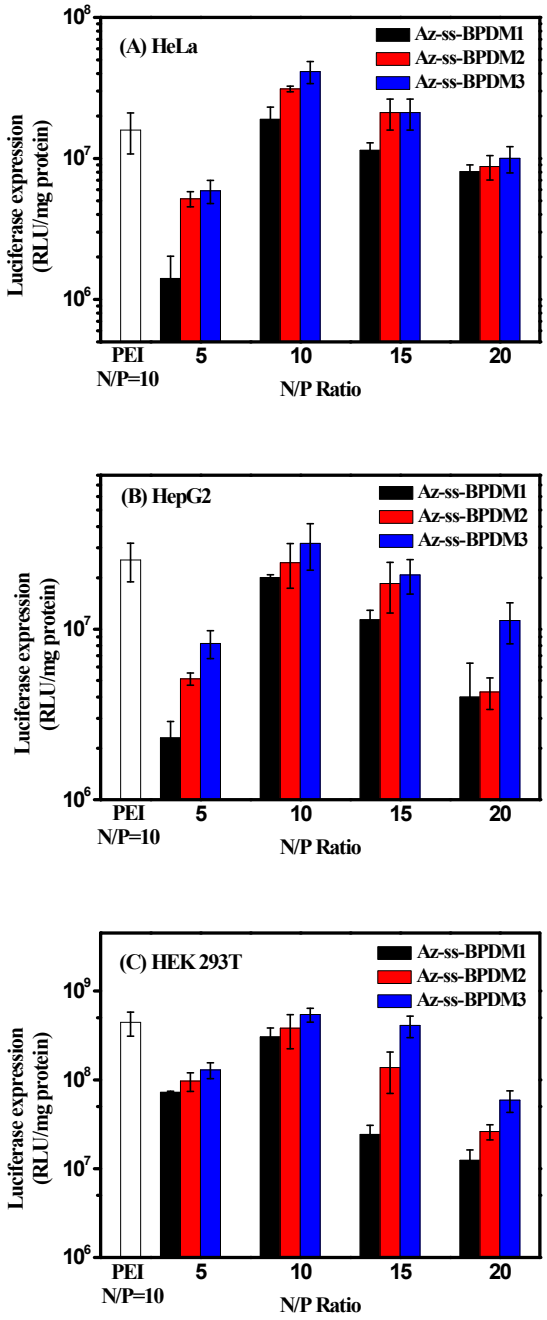

Fig. S9 Effect of branching degree of disulfide-containing azobenzene-terminated polycations without PCD on luciferase protein expression in serum media at various N/P ratios in comparison with that PEI/DNA polyplexes at N/P ratio of 10 in (A) HeLa, (B) HepG2 and (C) HEK 293T cell lines. Data was shown as mean ± SD (n=3).

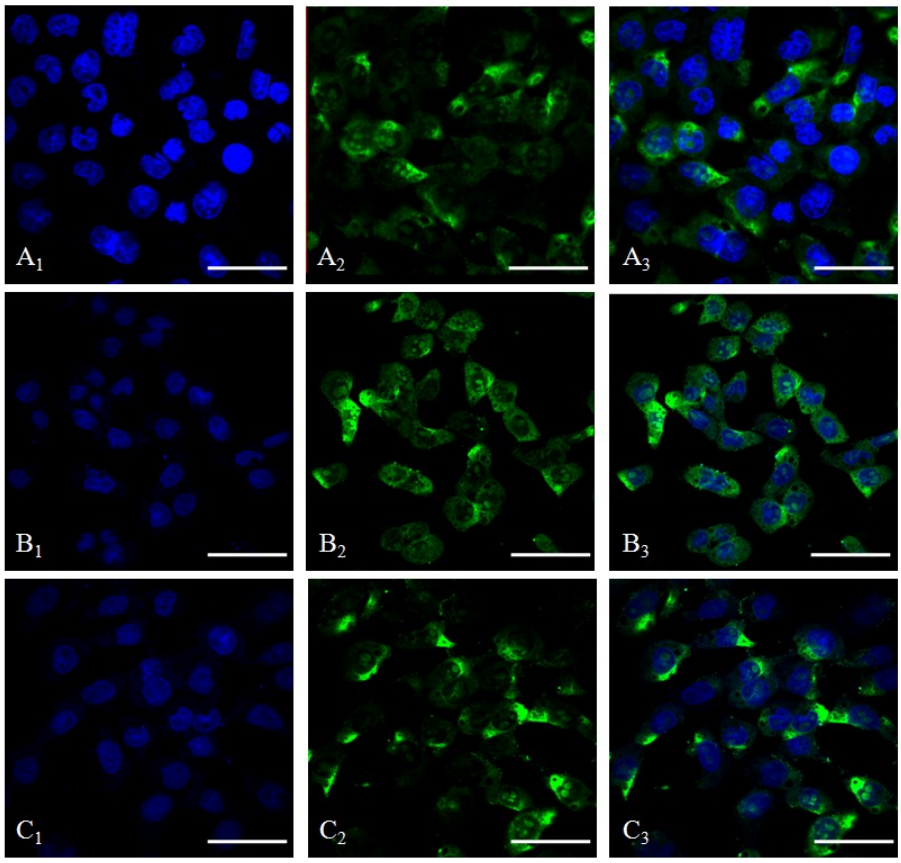

Fig. S10 Confocal laser scanning images of HepG2 cells after 4 h incubation with polyplexes, (A) Az-ss-BPDM3, (B) PCD/Az-ss-BPDM3=2/1, (C) PCD/Az-ss-BPDM3=4/1 at N/P ratio of 10. (A_1−C_1) Blue fluorescence of nuclei stained with Hoechst. (A_2−C_2) Green fluorescently stained plasmid pcDNA3-Luc with YOYO-1 (10^{-5} M). (A_3−C_3) Merged images of green and blue fluorescence fields. The scale bar is 40 μm.

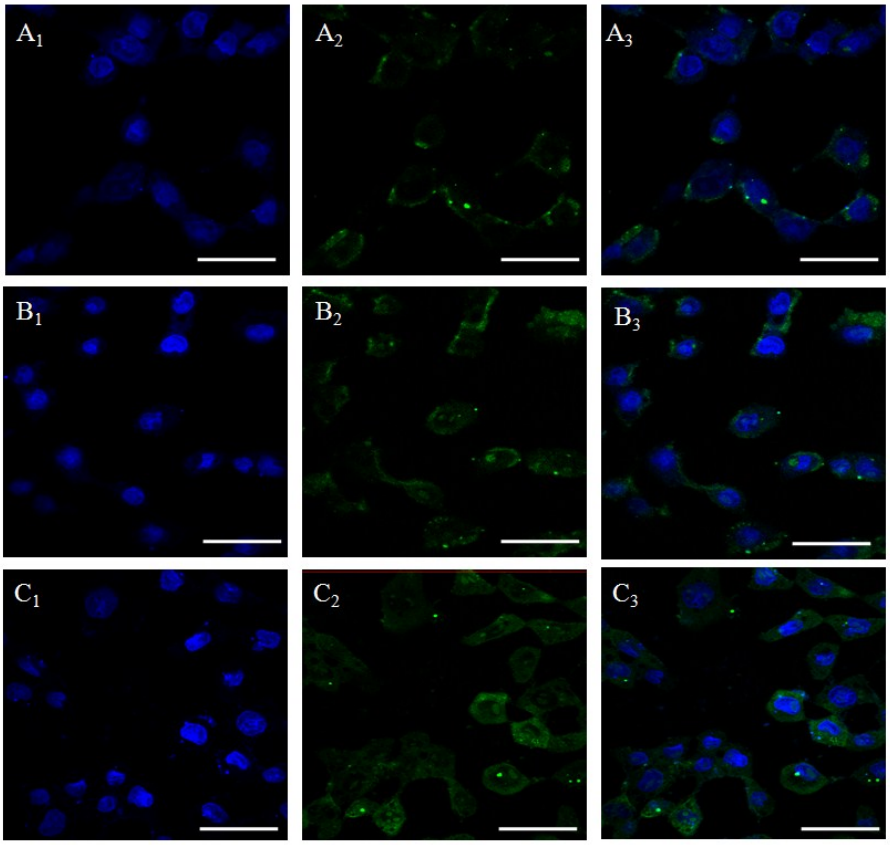

Fig. S11 Influence of branching degree of disulfide-containing azobenzene-terminated polycations without PCD on cellular internalization in HepG2 cells after 4 h incubation with polyplexes by confocal laser scanning images: (A) Az–ss-BPDM1, (B)Az–ss–BPDM2 and (C) Az–ss–BPDM3 at N/P ratio of 10 in HepG2 cells. (A_1−C_1) Blue fluorescence of nuclei stained with Hoechst. (A_2−C_2) Green fluorescently stained plasmid pcDNA3-Luc with YOYO-1(10^{-6} M). (A_3−C_3) Merged images of green and blue fluorescence fields. The scale bar is 40 μm.

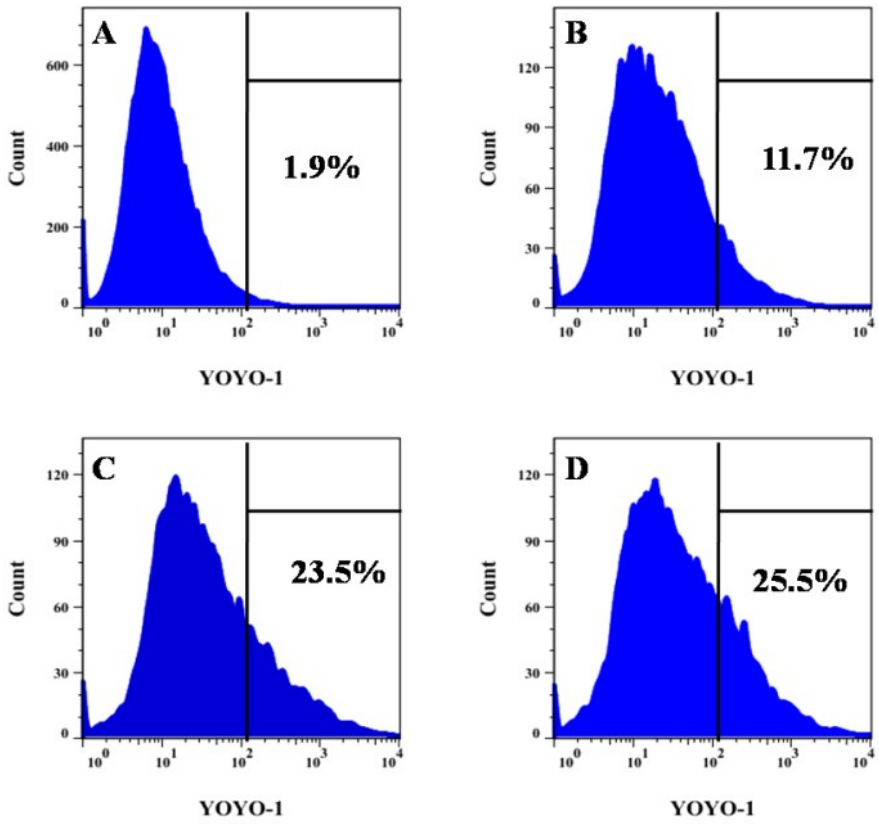

Fig. S12 Flow cytometry images of (A) untreated cells (control), (B) Az-ss-BPDM1/DNA, (C) Az-ss-BPDM2/DNA and (D) Az-ss-BPDM3/DNA polyplexes at N/P ratio of 10 in HepG2 cells.

Eq. S1

The NMR spectrum was used to determine the composition of ratio within the polymers structure *via* the following equations:

DMAEMA component (k) = $S_i/2$

CBA component (m+n) = $(S_h)/2$

CBA with free vinyl (m) = $(S_d + S_e + S_f)/3$

Branching CBA (n) =$[(S_h)/2]-[(S_d + S_e +S_f)/3]$

Initiator component (I) =$(S_a+S_b+S_c)/9$

Total content = k +n + m + I

S_a, S_b, S_c, S_d, S_e, S_f, S_h and S_i represent the peak areas of the related signals (Fig. 1). Therefore the percentage of each component within the polymer structure is a percentage of the total content, for instance:

Degree of branching (DB) = n/ (k +n + m + I)

Pendent vinyl group residue (R_p,%) = m/(m+n)