Electronic Supplementary Material (ESI) for Polymer Chemistry.

Supporting Information

Functionalized PGMA nanoparticles with aggregation-induced emission characteristics for gene delivery systems

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Fluorescence quantum yields of TPE-PGEA/TPE nanoparticles.

Rhodamine B was used as the standard substance[1] to measure the fluorescence quantum yields of all TPE-PGEA/TPE polymers. The data were summarized in **Table S1**.

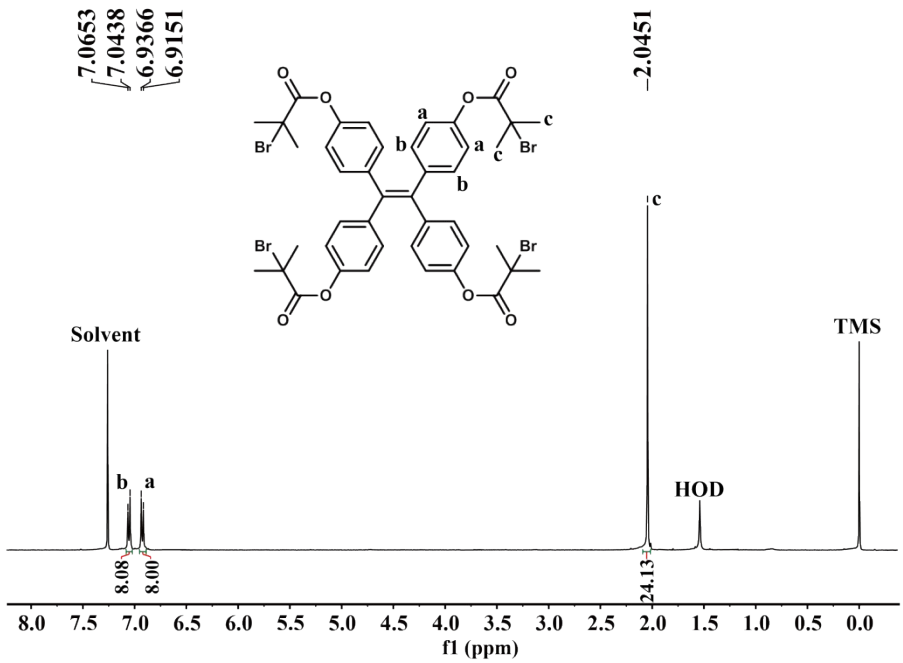
Tab. S1 Fluorescence quantum yields of TPE-PGEA/TPE nanoparticles

Samples	Fluorescence quantum yields
TPE-PGEA/TPE1-1	0.23
TPE-PGEA/TPE1-2	0.22
TPE-PGEA/TPE2-1	0.16
TPE-PGEA/TPE2-2	0.15
TPE-PGEA/TPE3-1	0.19
TPE-PGEA/TPE3-2	0.25

Spectra of ^{1}H NMR

Nuclear magnetic resonance (NMR) was used to confirm the structures of TPE-Br and TPE-$NH_3^+CF_3COO^-$. The ^{1}H NMR spectra were as follows:

(a) TPE-Br



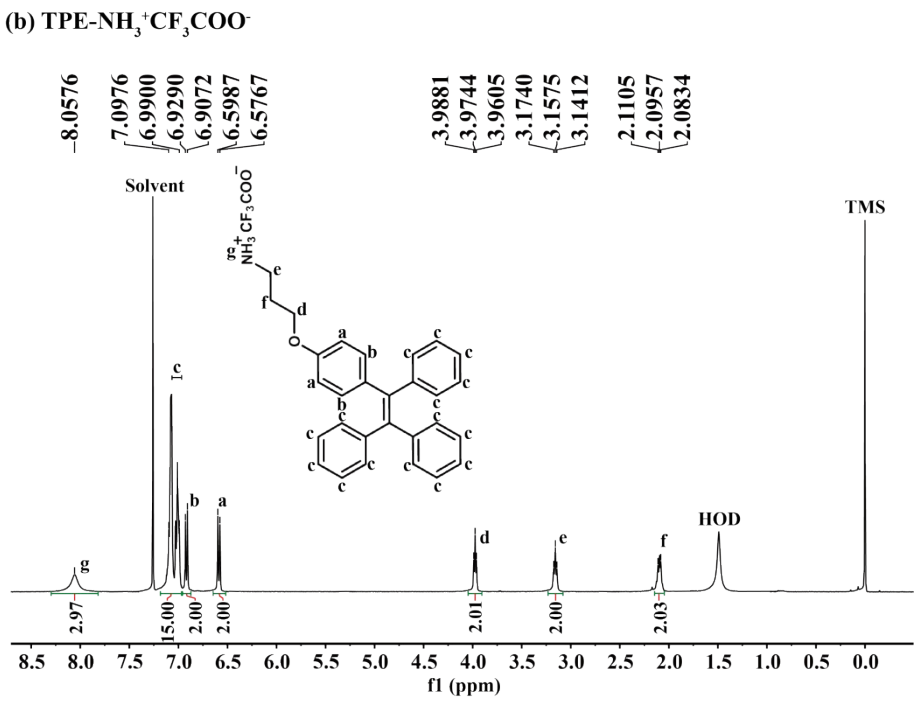
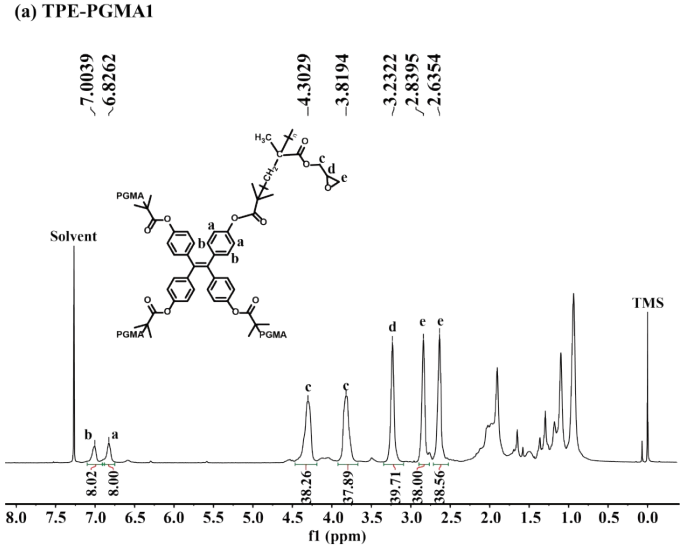


Fig. S1 ^{1}H NMR spectra of (a) TPE-Br and (b) TPE-$NH_3^+CF_3COO^-$



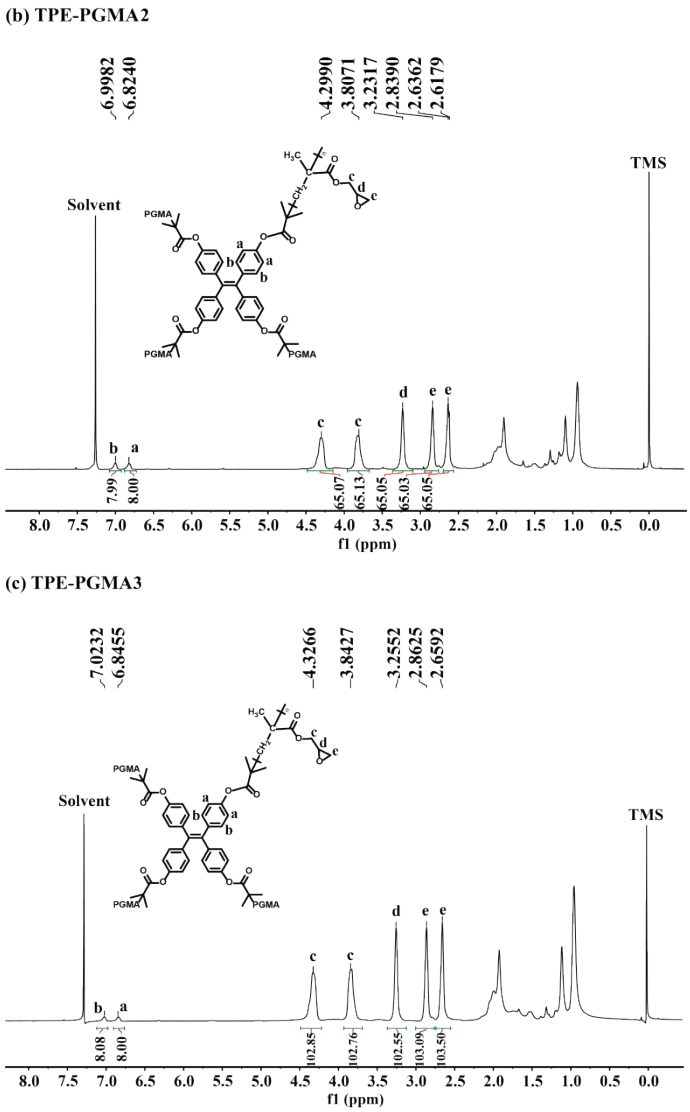


Fig. S2 ^{1}H NMR spectra of (a) TPE-PGMA1, (b) TPE-PGMA2 and (c) TPE-PGMA3.

TPE-PGMA was also analyzed by NMR (Figure S2). The ^{1}H NMR spectra proved the structures of TPE-PGMA. In order to prove the structures of TPE-PGEA and TPE-PGEA/TPE polymers, they were analyzed by NMR (Figures S3-S5). The ^{1}H NMR spectra showed that the TPE-PGEA and TPE-PGEA/TPE polymers were synthesized successfully as expected.

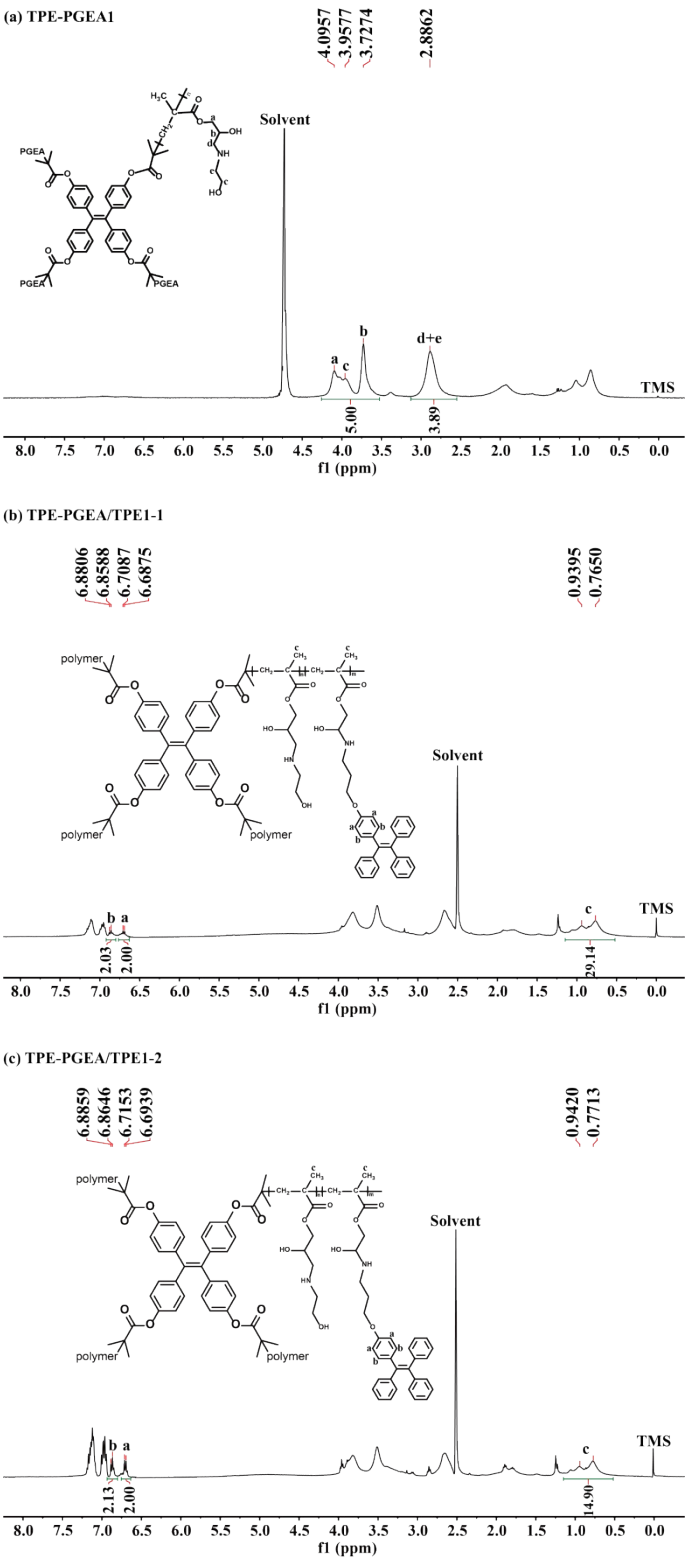


Fig. S3 ^{1}H NMR spectra of (a) TPE-PGEA1, (b) TPE-PGEA/TPE1-1 and (c) TPE-PGEA/TPE1-2.

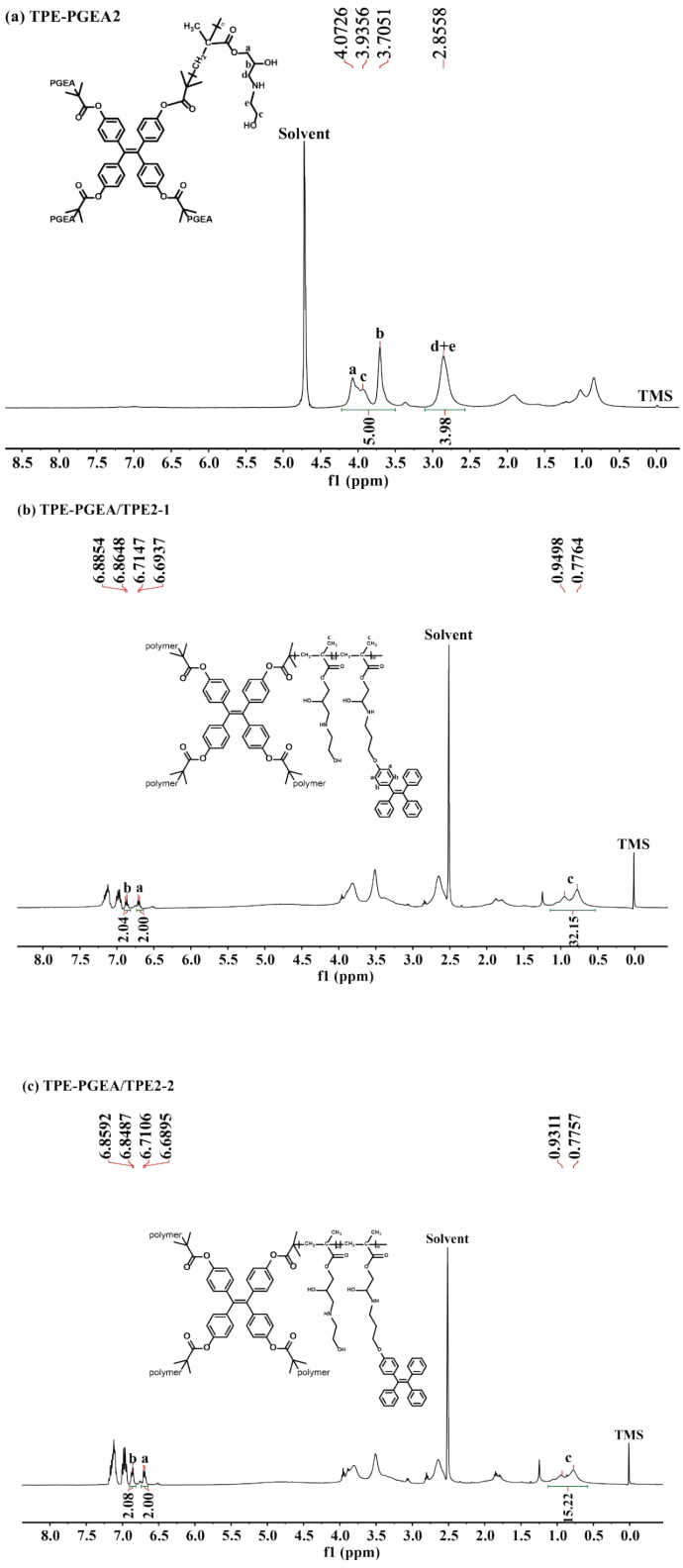


Fig. S4 ^{1}H NMR spectra of (a) TPE-PGEA2, (b) TPE-PGEA/TPE2-1 and (c) TPE-PGEA/TPE2-2.

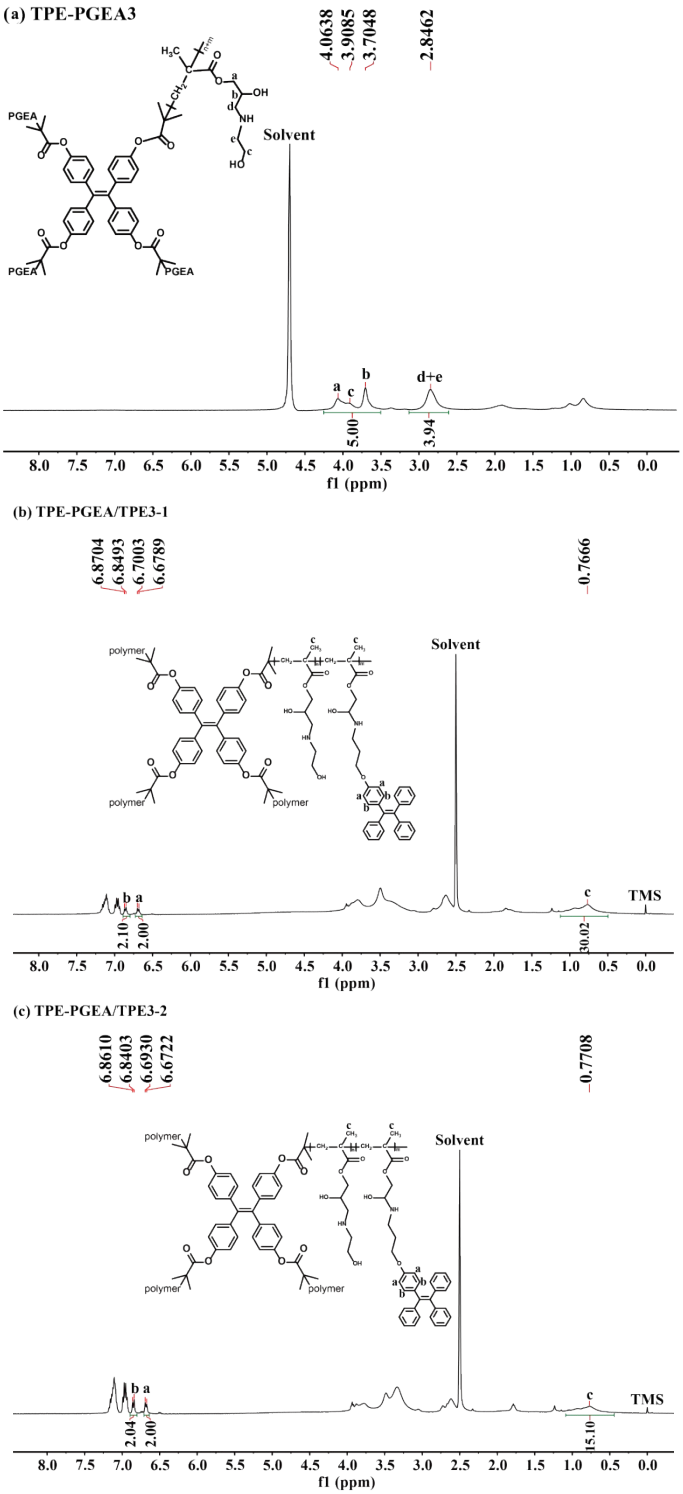


Fig. S5 ^{1}H NMR spectra of (a) TPE-PGEA3, (b) TPE-PGEA/TPE3-1 and (c) TPE-PGEA/TPE3-2.

Cell viability assay

HeLa and HepG2 cell lines from two common cancers were selected to evaluate the performance of gene carriers. Both of cell lines were cultured in Dulbecco's modified eagle medium (DMEM), which was supplemented with 10% fetal bovine serum (FBS), 100 IU/mL of penicillin and 100 mg/mL of streptomycin. The growing environmental requires a 5% CO_2 atmosphere with 95% relative humidity at 37^{o}C. All cell culture mentioned in this article was taken care at the same condition. The cytotoxicity of TPE-PGEA/pDNA and TPE-PGEA/TPE/pDNA complexes at series of N/P ratios was evaluated using a Bio-Rad model 680 microplate reader at a wavelength of 570 nm through the MTT assay method outlined in our previous paper.[2] For each sample, the final absorbance was the average of those measured from six wells in parallel.

The cytotoxicity of complexes condensed the pRL-CMV as a reporter gene was evaluated by MTT assay in HeLa and HepG2 cell lines. In general, the results exhibited a dose-dependent tendency of the cell viability which decreased with increasing N/P ratios. At the same N/P ratio, the cell viability of TPE-PGEA/TPE polymers was lower than TPE-PGEA polymers, probably because the cytotoxocity was caused by the TPE units. With the increased molecular weight and ratio of ring-opening of TPE-NH_2 moieties relative to epoxy groups, the cationic charge density and lipophilicity of polymers increased, which was able to induce damage to cell membranes and thus increase cytotoxicity.[3] As N/P ratio increased, the cell viability decreased gradually due to the increasing content of free cationic polymers. However, TPE-PGEA and TPE-PGEA/TPE polymers had much lower cytotoxicity than branched polyethylenimine (PEI, Mw = 25 KDa), the gold standard of nonviral gene

vectors.

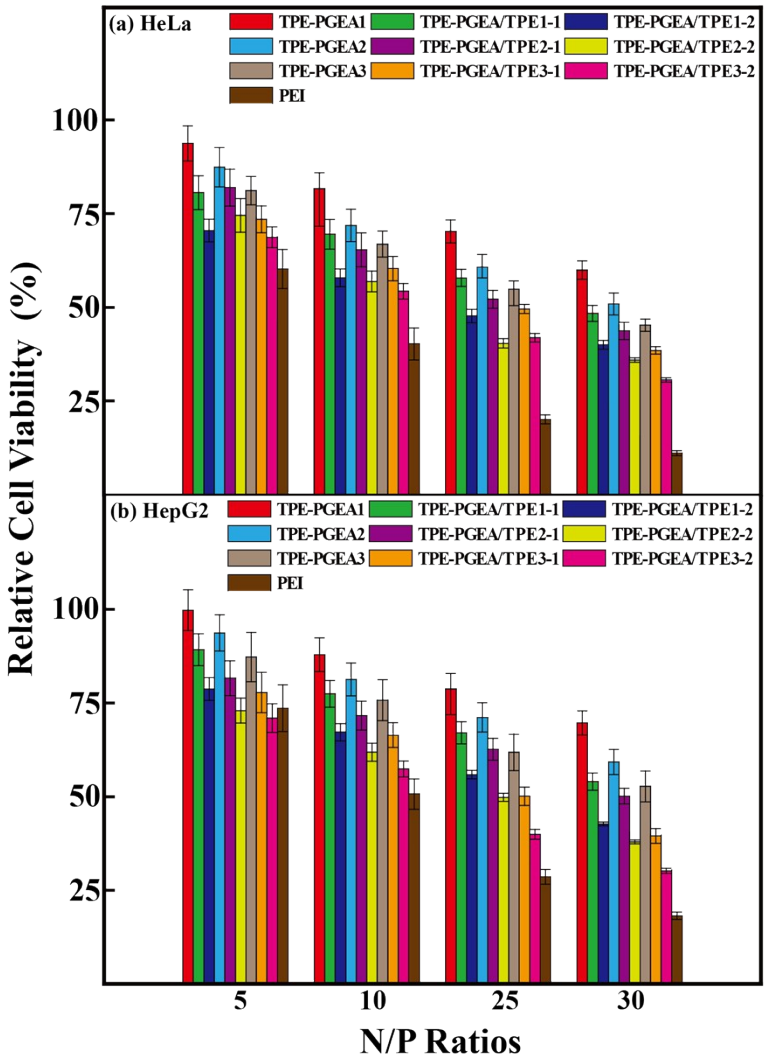


Fig. S6 Cytotoxicity of TPE-PGEA/pDNA, TPE-PGEA/TPE/pDNA and PEI/pDNA complexes at different N/P ratios in (a) HeLa and (b) HepG2 cell lines.

1 T. Karstens and K. Kobs, *J. Phys. Chem.*, 1980, **84**, 1871-1872.

2 H. M Yuan, C. Xu, Y. Zhao, B. Yu, G. Cheng and F. J. Xu, *Adv. Funct. Mater*, 2016. DOI: 10.1002/adfm.201504980.

3 R. Elisabetta, F. Paolo, L. Ettore, M. Amedea, R. Manuela, P. R. Mussini, C. Federica and B. Cristina, *J. Polym. Sci. Polym. Chem.*, 2009, **47**, 6977-6991.