Electronic Supplementary Material (ESI) for Soft Matter.
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Control of aggregation temperatures in mixed and blended cytocompatible thermoresponsive block co-polymer nanoparticles

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Supporting Material

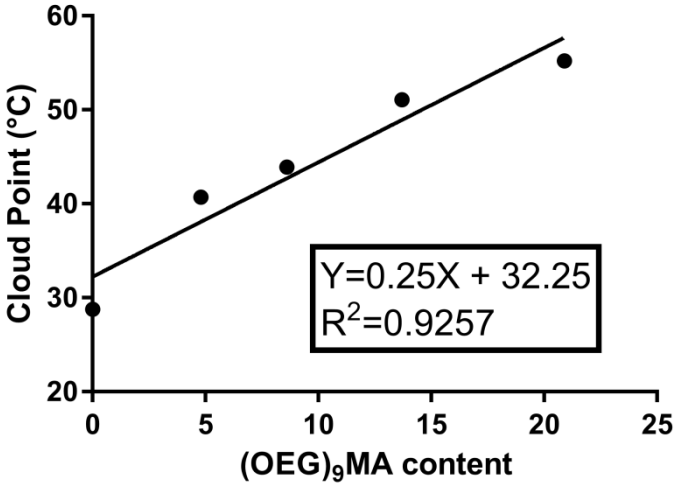

Figure S1: Relationship between the $(OEG)_9MA$ content of each formulation against T_t for single polymer corona nanoparticles.

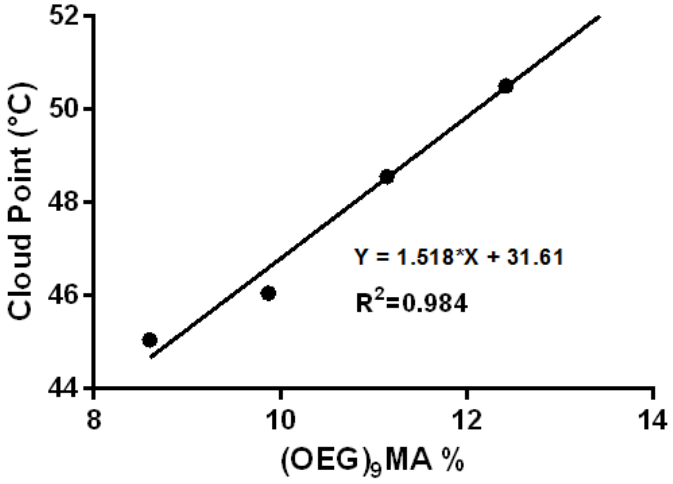

Figure S2: Relationship between the $(OEG)_9MA$ content of each formulation against T_t for nanoparticles blends prepared from PLA-pOEGMA 10 and PLA-pOEGMA 15.

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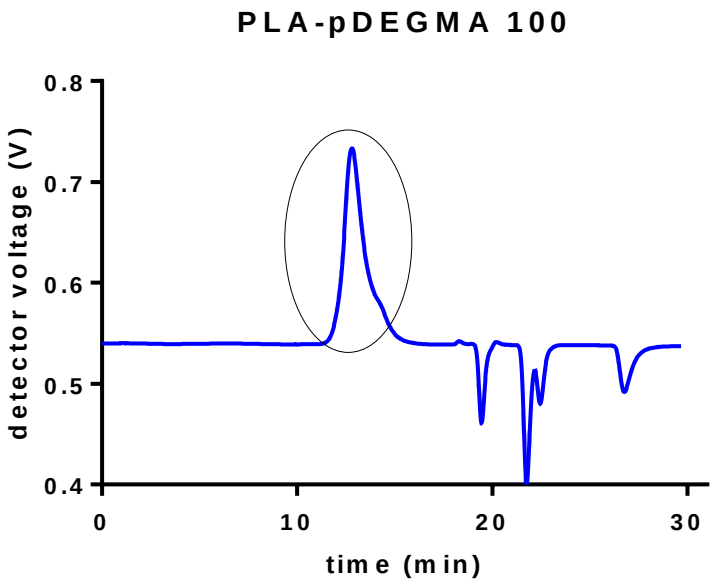
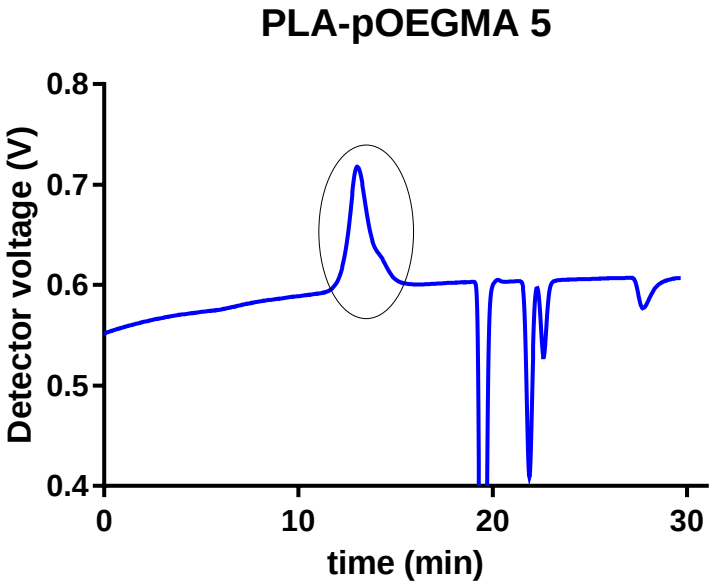
GPC analyses of the polymers



Mn	25090
PDI	1.48

Figure S1: PLA-pDEGMA 100 GPC analysis.



Mn	23000
PDI	1.39

Figure S4: PLA-pOEGMA 5 GPC analysis.

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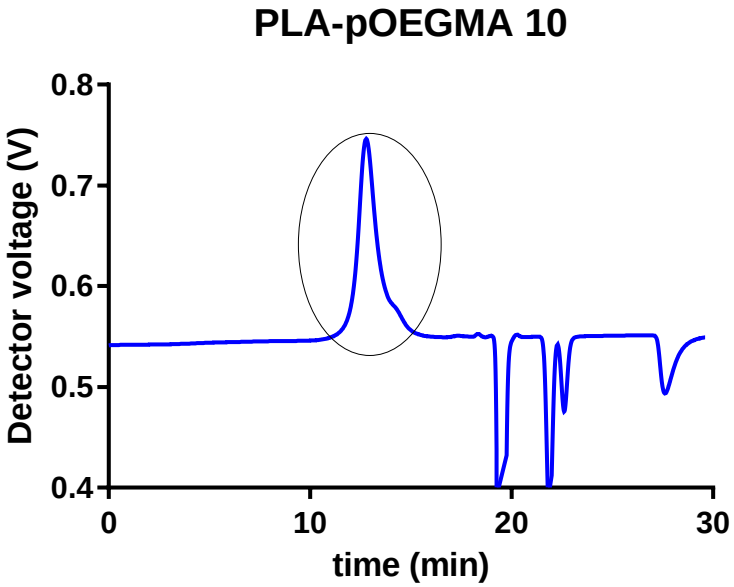
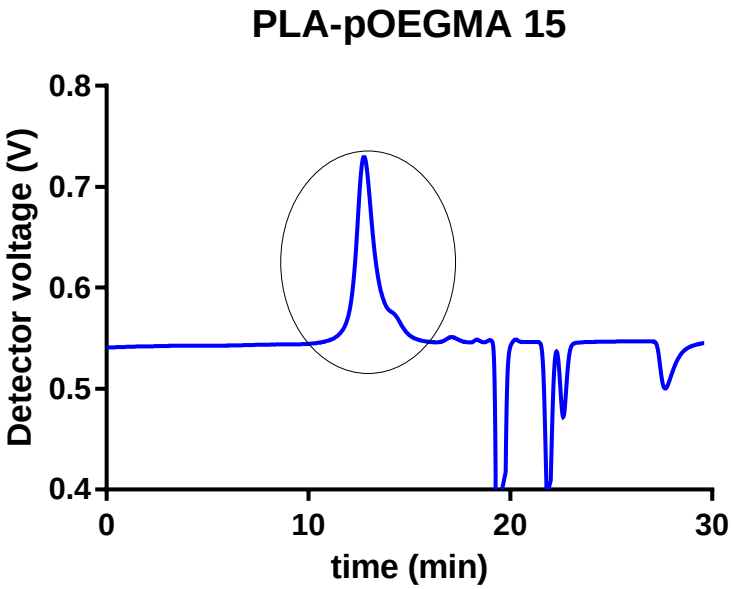


Mn	27710
PDI	1.48

Figure S5: PLA-pOEGMA 10 GPC analysis.



Mn	27390
PDI	1.53

Figure S6: PLA-pOEGMA 15 GPC analysis.

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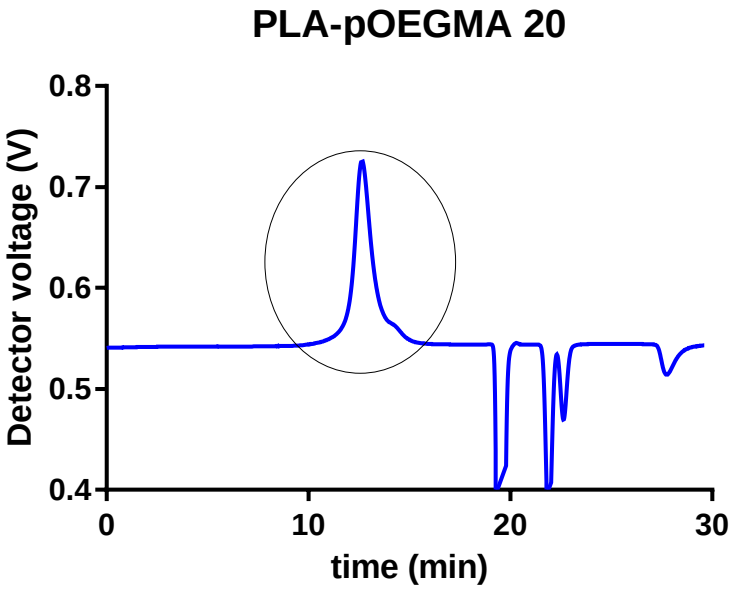

Mn	32840
PDI	1.56

Figure S7: PLA-pOEGMA 20 GPC analysis.

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^{1}H NMR and ^{13}C NMR analyses of the polymers

PLA-pDEGMA 100. ^{1}H NMR: δ 0.9-1.05 (m, DEGMA CH_3), 1.52 (m, lactide CH_3), 1.82-192 (m, DEGMA CH_2C), 3.41 (s, DEGMA CH_3O), 3.56-3.57 (m, DEGMA CH_2O), 4.11 (m, DEGMA $COOCH_2$), 5.17 (m, lactide CH). ^{13}C NMR: δ 17, 45, 59, 64, 69, 70, 72, 77, 169.

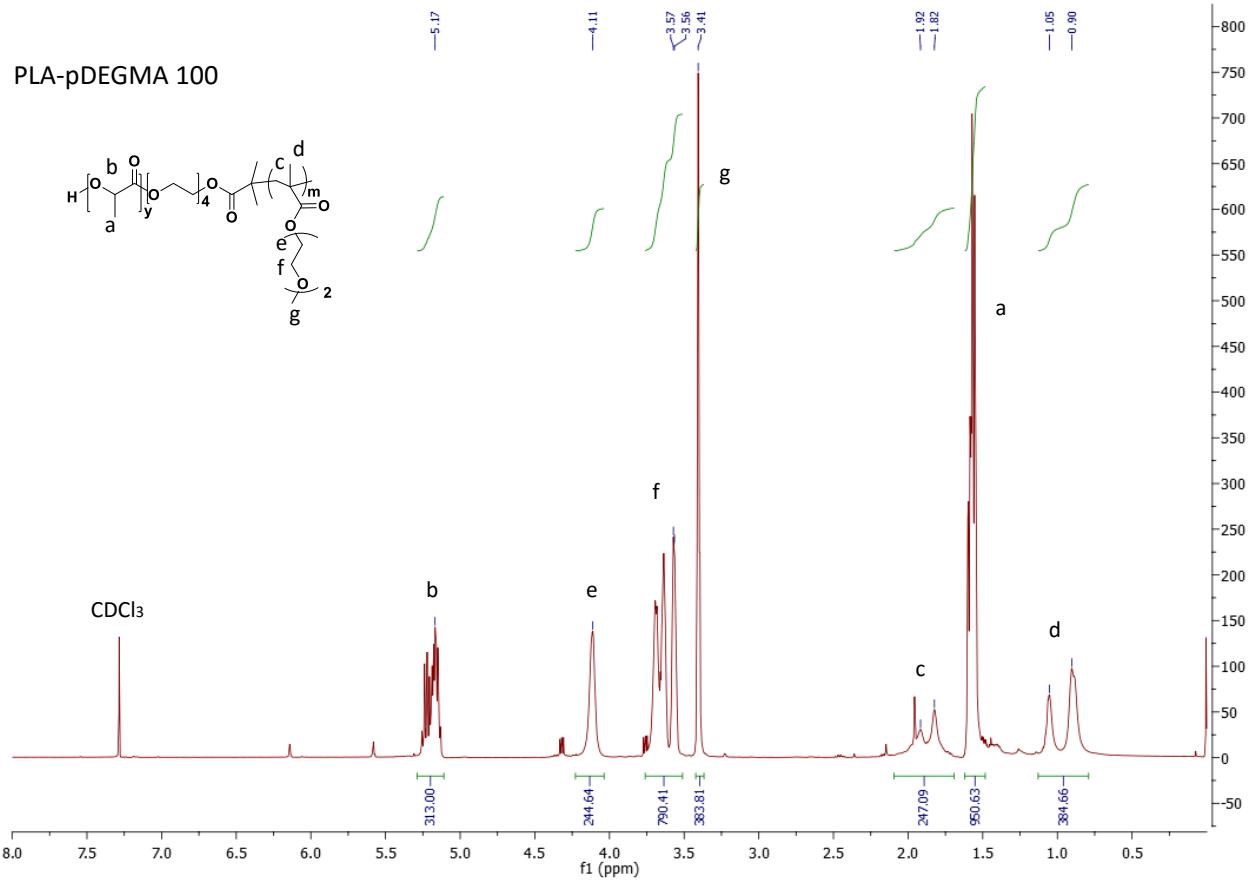

Figure S8: PLA-pDEGMA 100 ^{1}H NMR.

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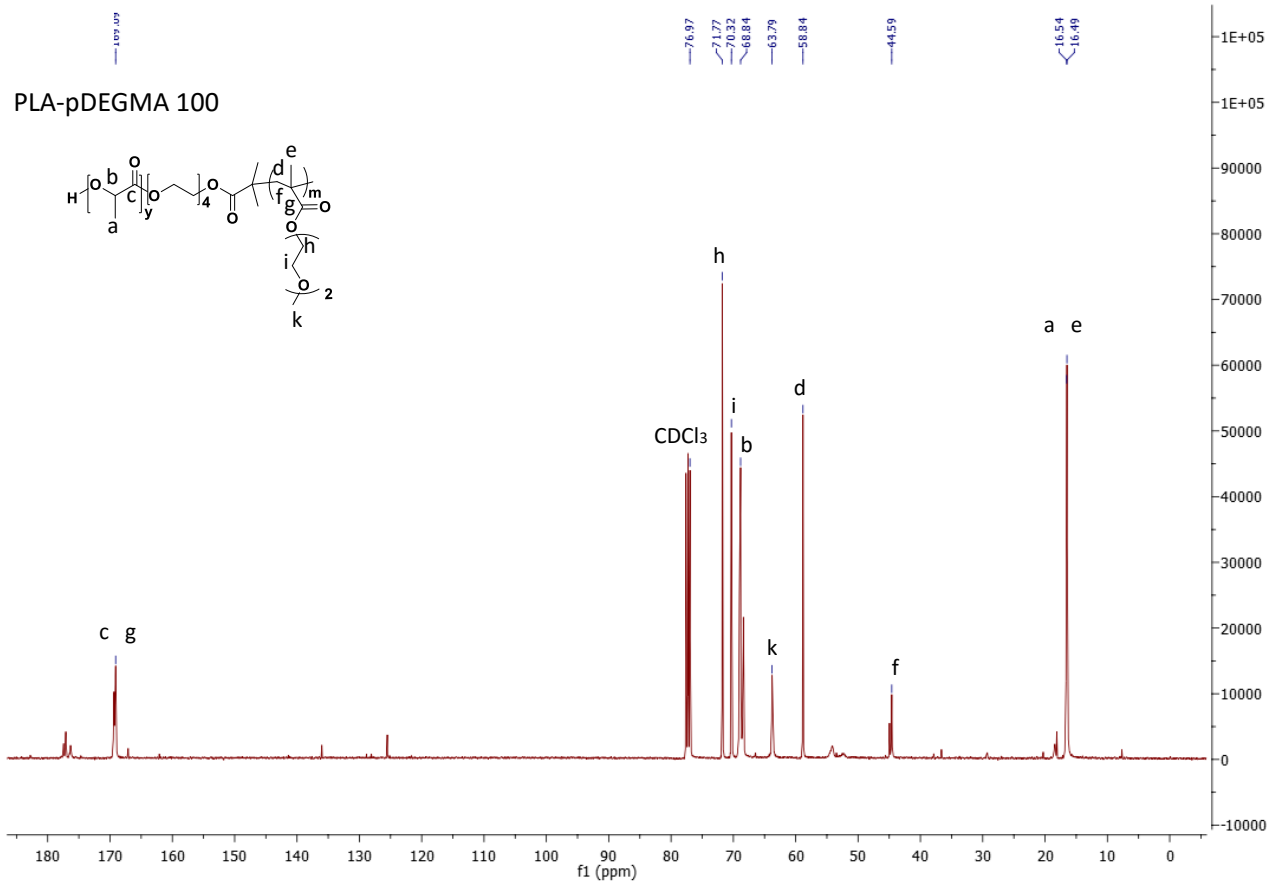

Figure S9: PLA-pDEGMA 100 ^{13}C NMR.

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PLA-pOEGMA 5. ^{1}H NMR: δ 0.89-1.06 (m, DEGMA/OEGMA CH_3), 1.55 (m, lactide CH_3), 1.82-1.92 (m, DEGMA/OEGMA CH_2C), 3.4 (s, DEGMA/OEGMA CH_3O), 3.56-3.69 (m, DEGMA/OEGMA CH_2O), 4.11 (m, DEGMA/OEGMA $COOCH_2$) 5.17 (m, lactide CH). ^{13}C NMR: δ 17, 45, 59, 64, 69, 70, 72, 77, 169.

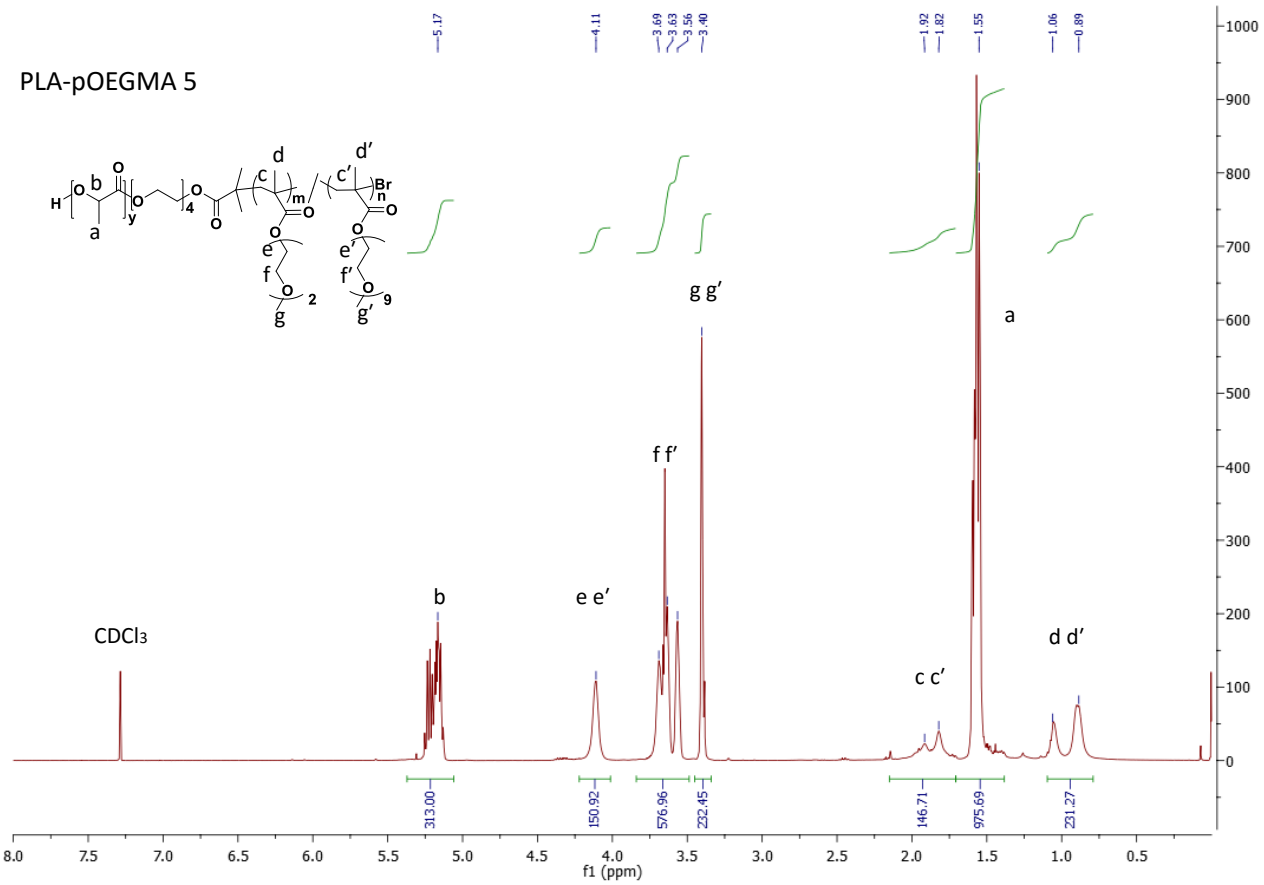

Figure S10: PLA-pOEGMA 5 ^{1}H NMR.

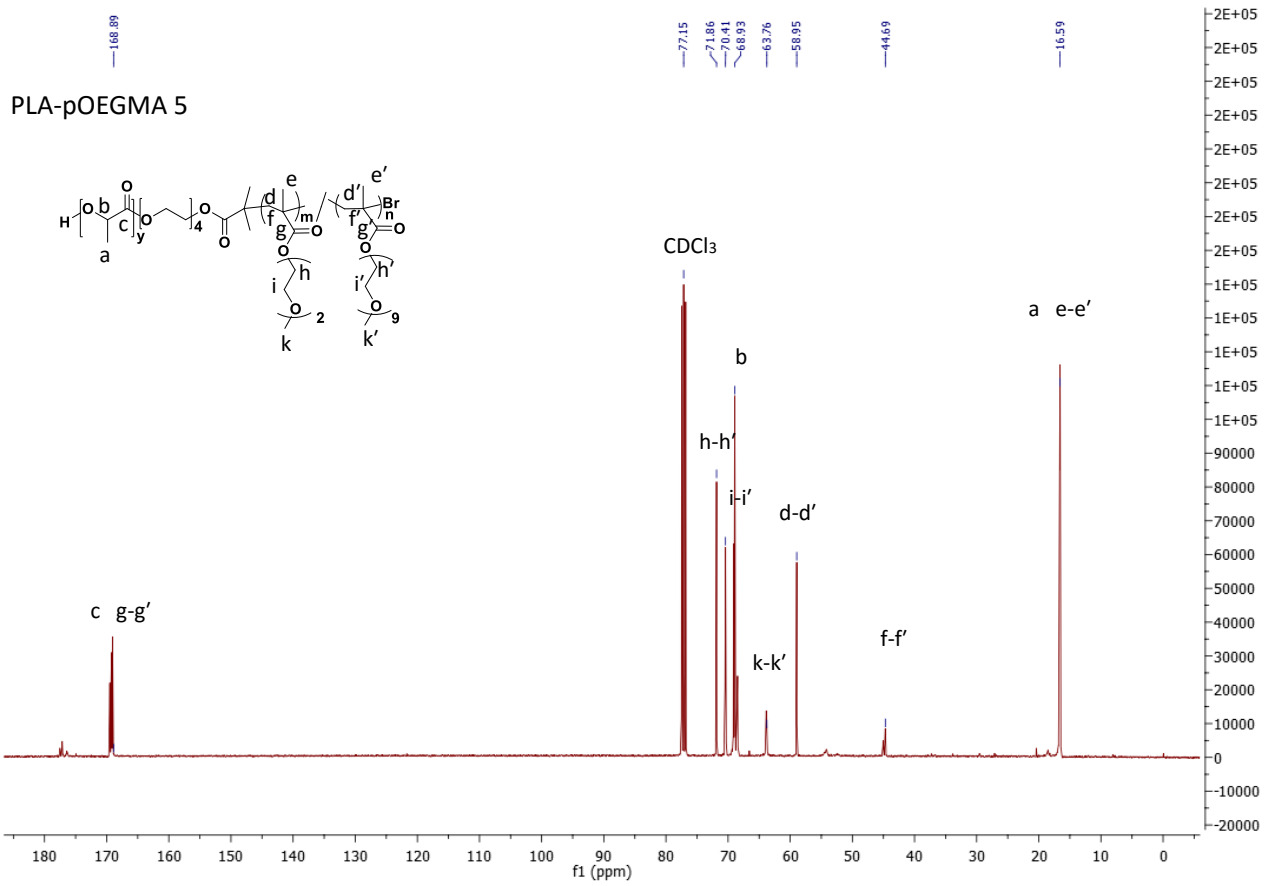

Figure S11: PLA-pOEGMA 5 ^{13}C NMR.

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PLA-pOEGMA 10. ^{1}H NMR: δ 0.88-1.05 (m, DEGMA/OEGMA CH_3), 1.57 (m, lactide CH_3), 1.81-1.91 (m, DEGMA/OEGMA CH_2C), 3.39 (s, DEGMA/OEGMA CH_3O), 3.55-3.68 (m, DEGMA/OEGMA CH_2O), 4.11 (m, DEGMA/OEGMA $COOCH_2$) 5.16 (m, lactide CH). ^{13}C NMR: δ 16, 45, 59, 64, 69, 70, 72, 168.

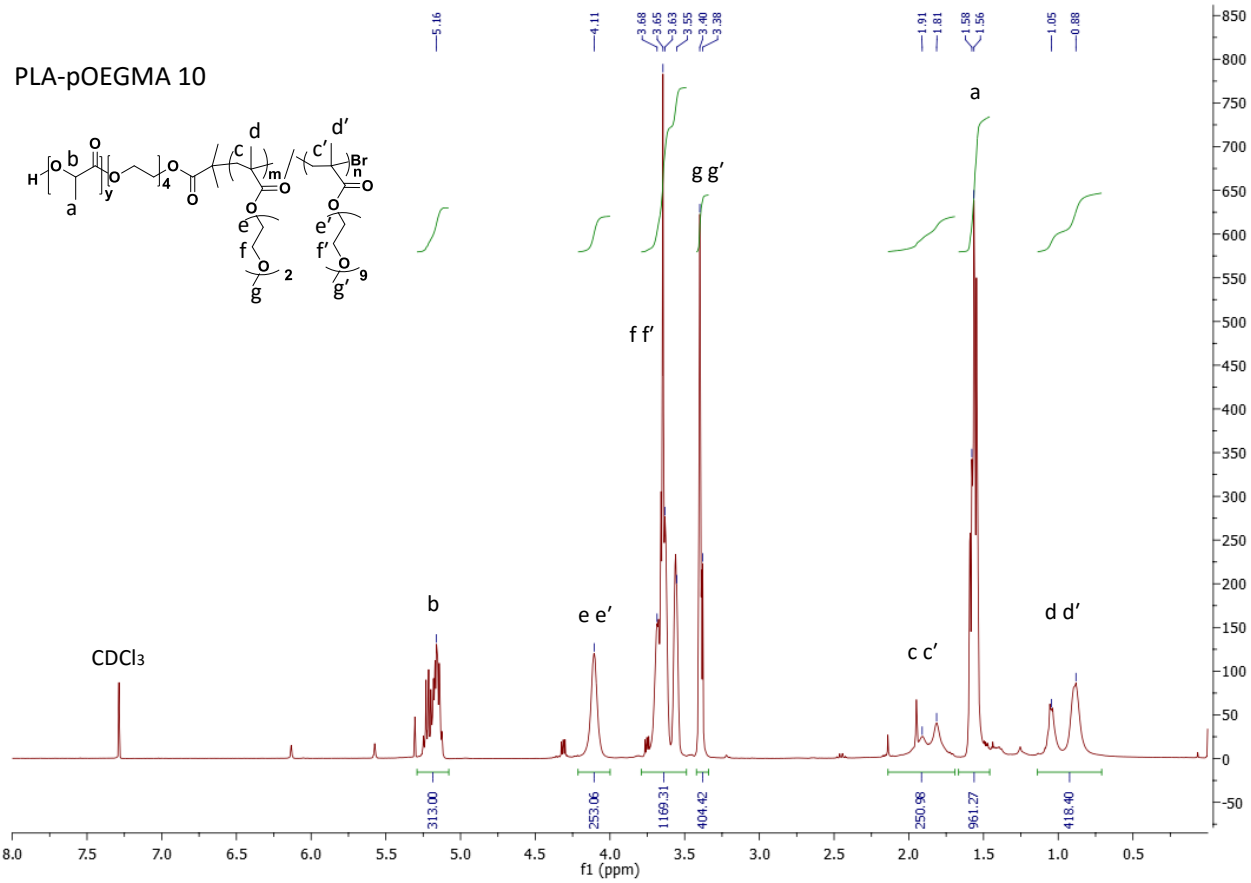

Figure S12: PLA-pOEGMA 10 ^{1}H NMR.

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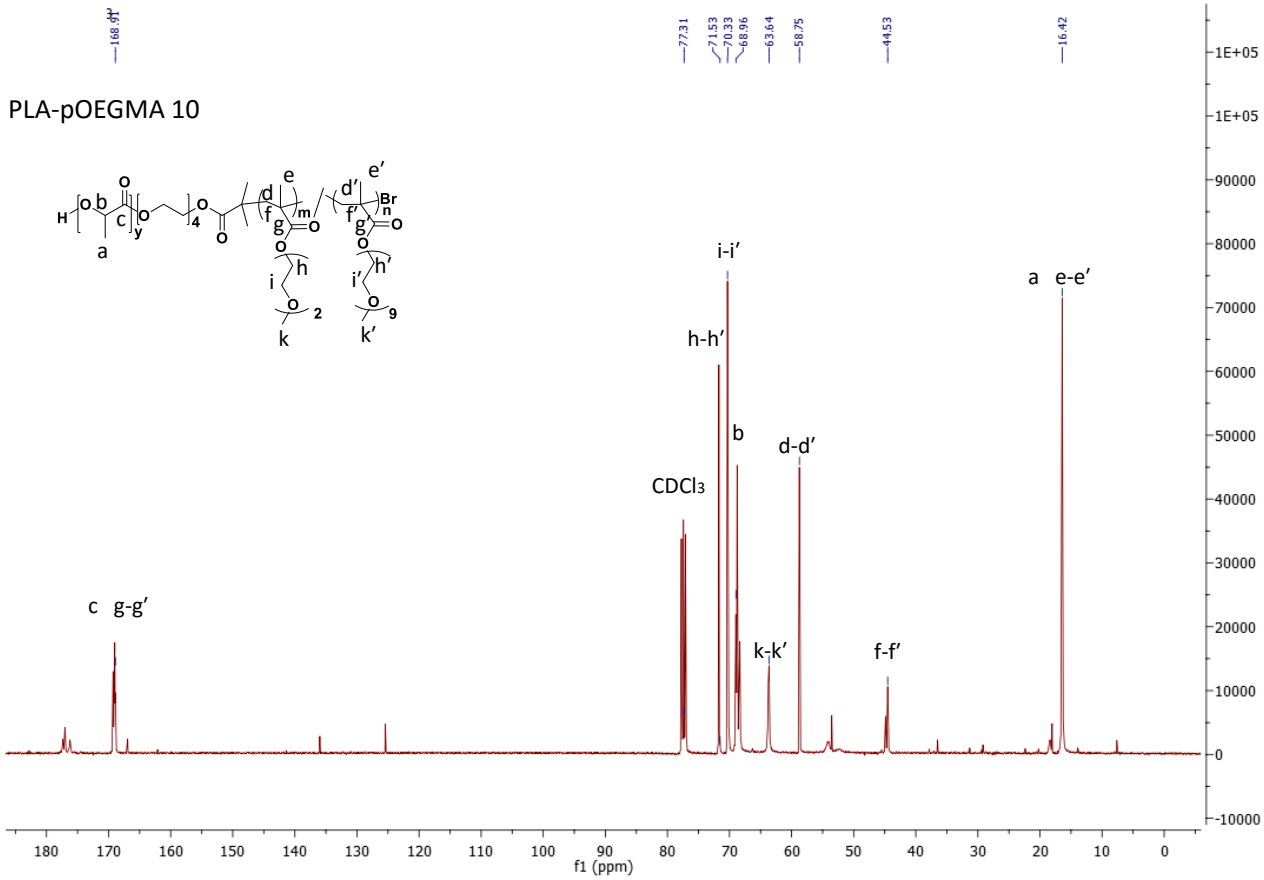

Figure S13: PLA-pOEGMA 10 ^{13}C NMR.

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PLA-pOEGMA 15. ^{1}H NMR: δ 0.91-1.06 (m, DEGMA/OEGMA CH_3), 1.59 (m, lactide CH_3), 1.82-1.92 (m, DEGMA/OEGMA CH_2C), 3.41 (s, DEGMA/OEGMA CH_3O), 3.58-3.69 (m, DEGMA/OEGMA CH_2O), 4.12 (m, DEGMA/OEGMA $COOCH_2$) 5.18 (m, lactide CH). ^{13}C NMR: δ 16, 45, 59, 64, 69, 70, 72, 77, 169.

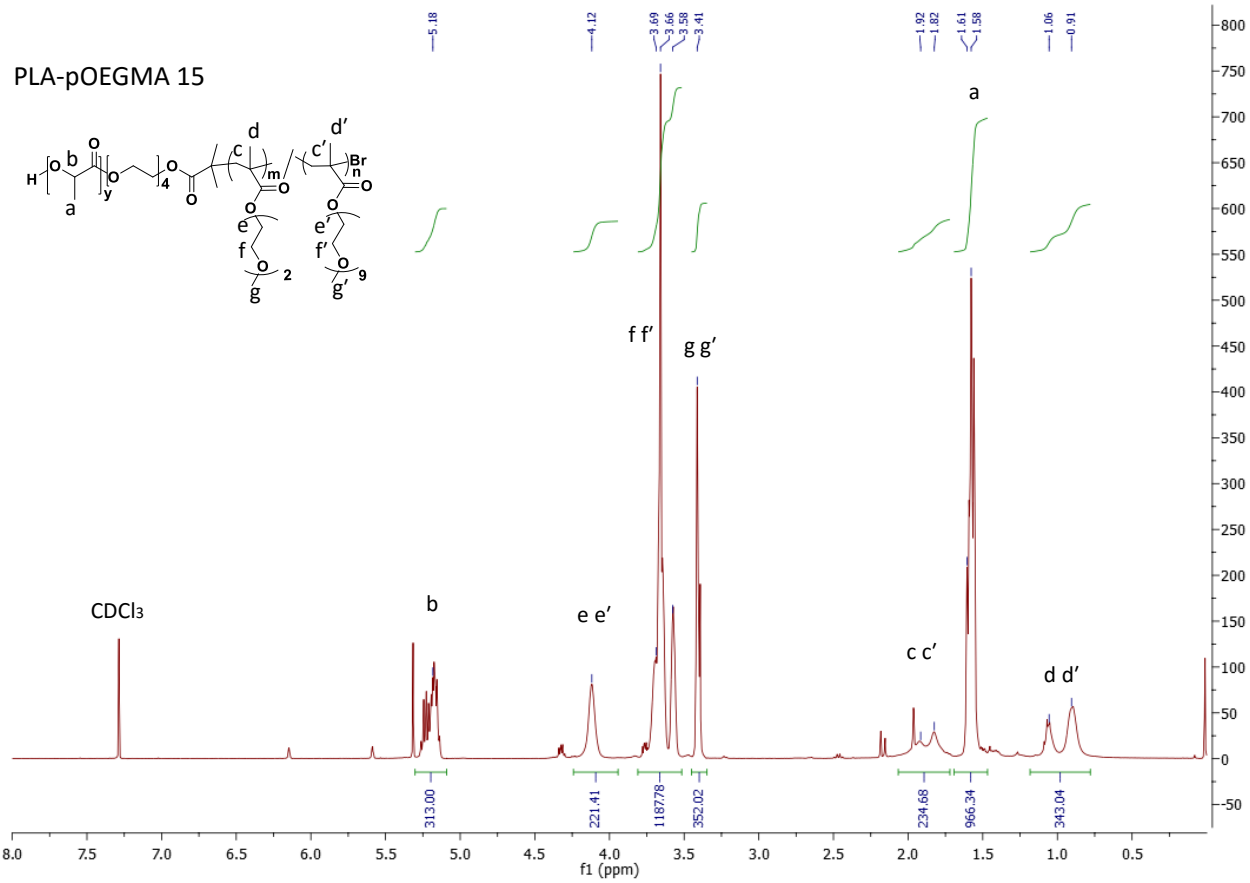

Figure S14: PLA-pOEGMA 15 ^{1}H NMR.

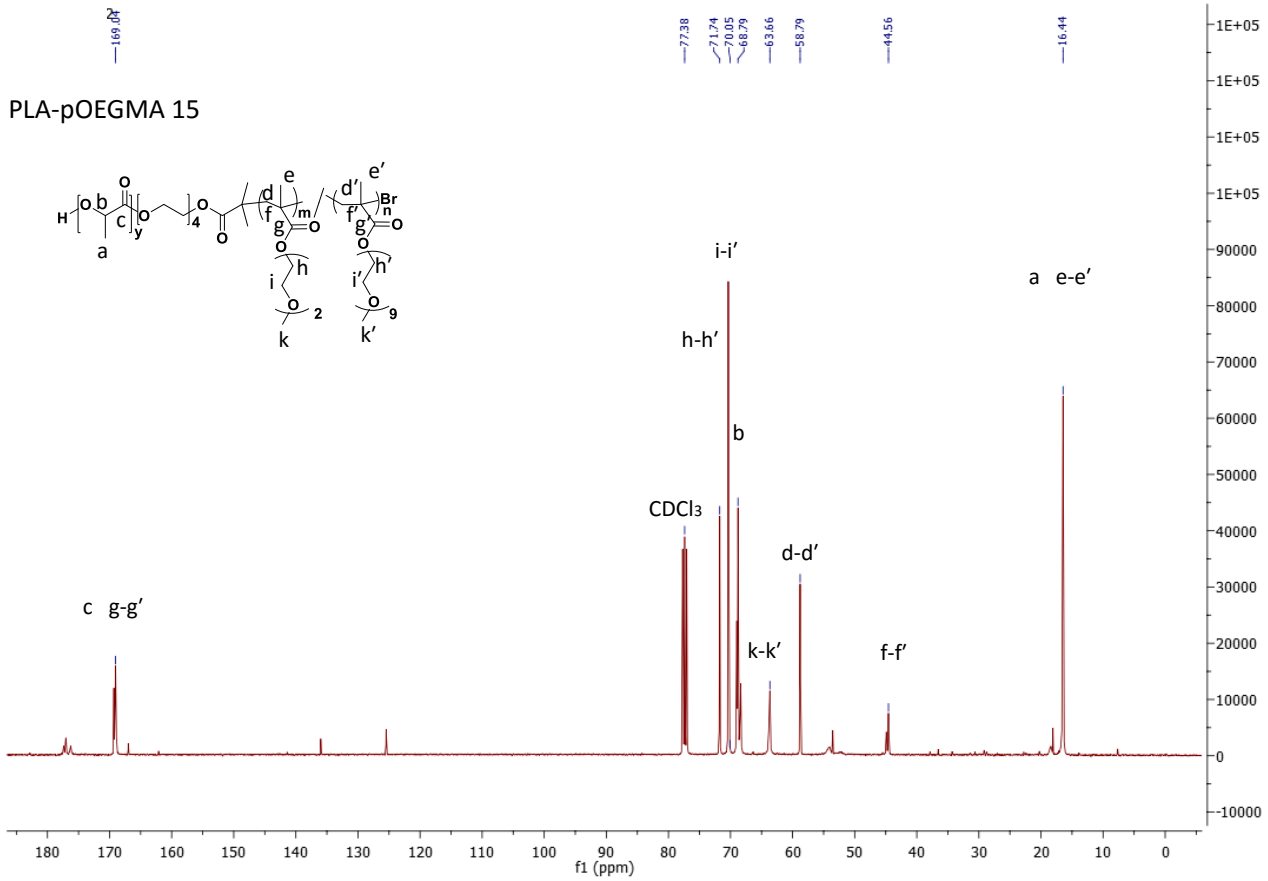

Figure S15: PLA-pOEGMA 15 ^{13}C NMR.

PLA-pOEGMA 20. ^{1}H NMR: δ 0.87-1.03 (m, DEGMA/OEGMA CH_3), 1.56 (m, lactide CH_3), 1.80-1.94 (m, DEGMA/OEGMA CH_2C), 3.39 (s, DEGMA/OEGMA CH_3O), 3.56-3.64 (m, DEGMA/OEGMA CH_2O), 4.1 (m, DEGMA/OEGMA $COOCH_2$) 5.14 (m, lactide CH). ^{13}C NMR: δ 17, 45, 59, 64, 69, 71, 72, 77, 169.

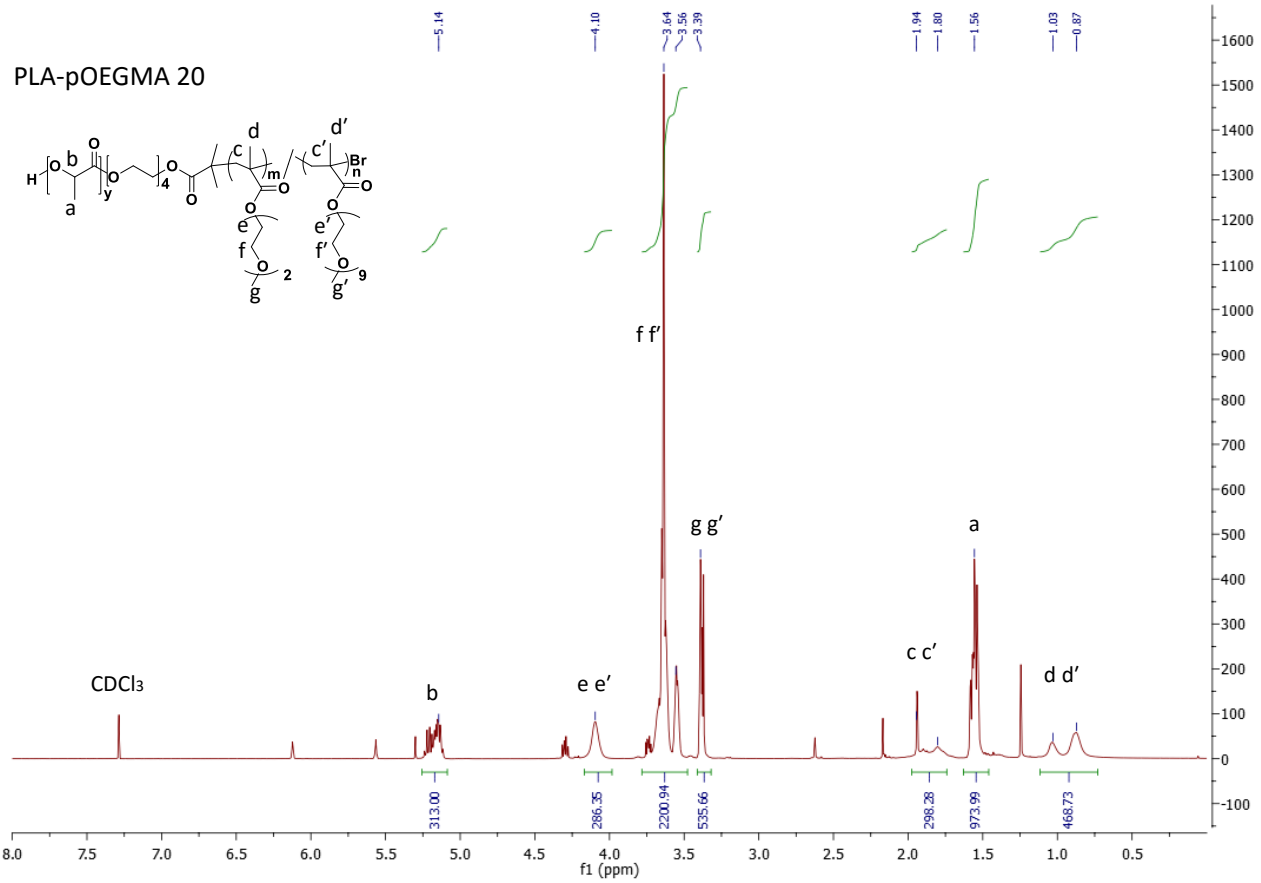

Figure S16: PLA-pOEGMA 20 ^{1}H NMR.

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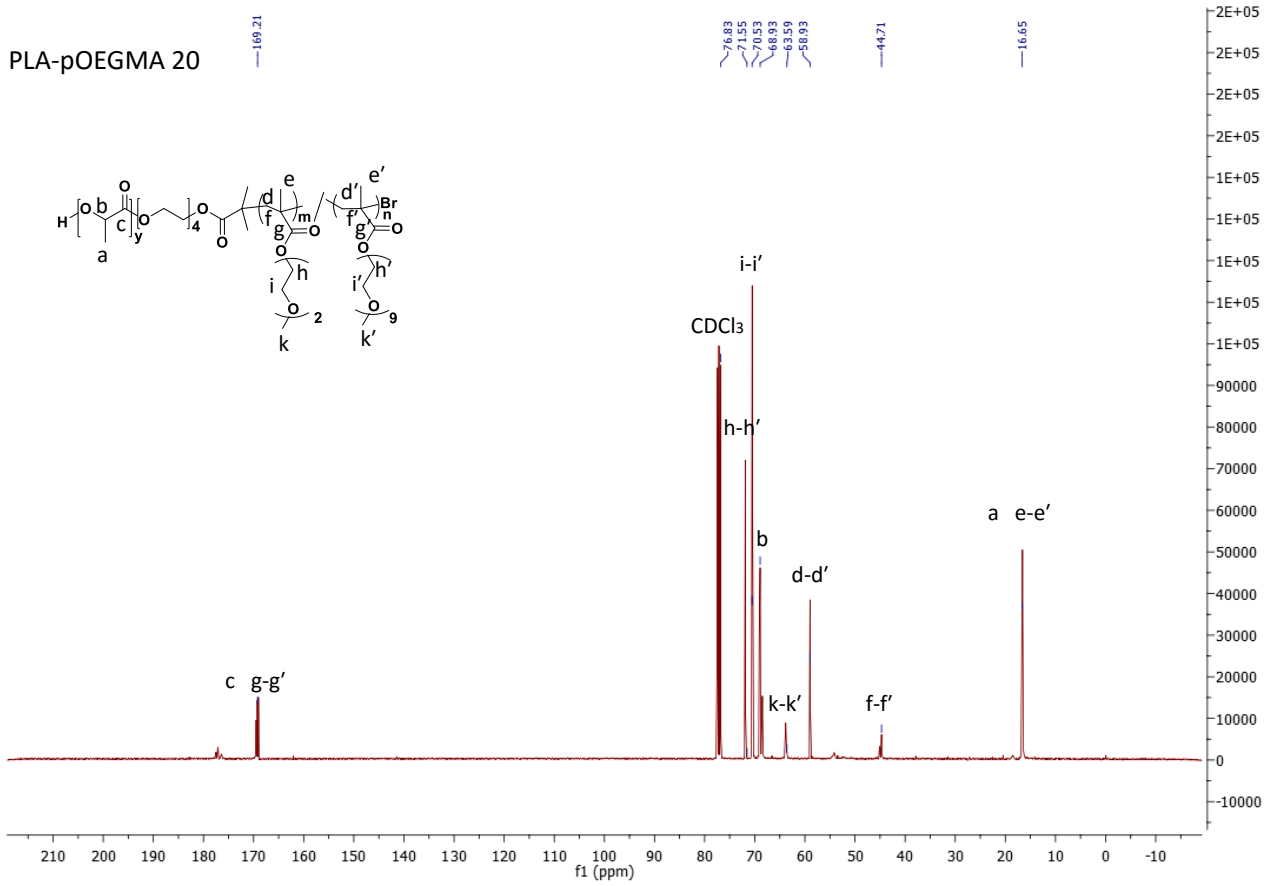

Figure S17: PLA-pOEGMA 20 ^{13}C NMR.

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IR spectra of the polymers

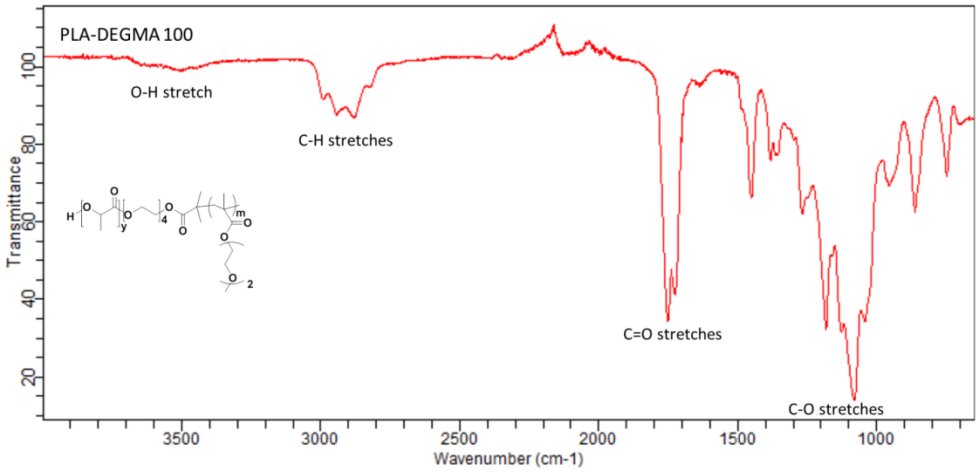

Figure S18: PLA-pDEGMA 100 IR.

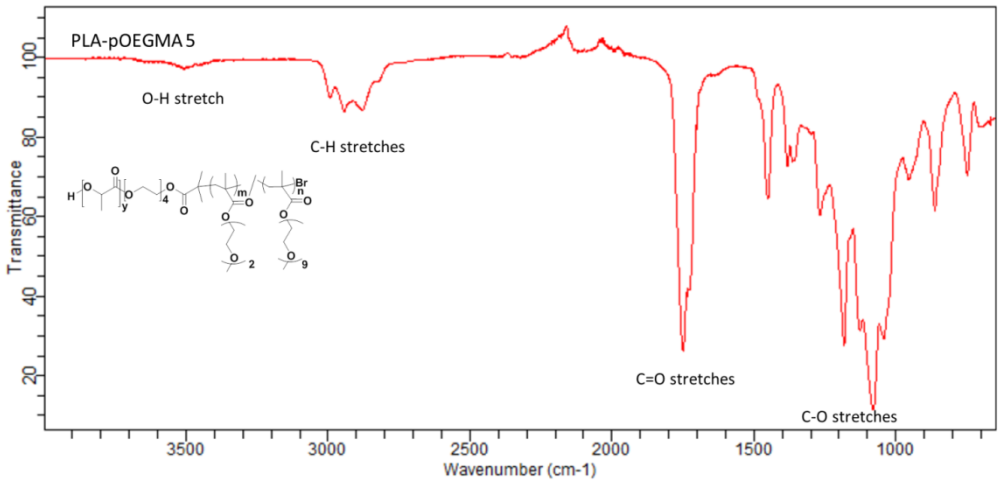

Figure S19: PLA-pOEGMA 5 IR.

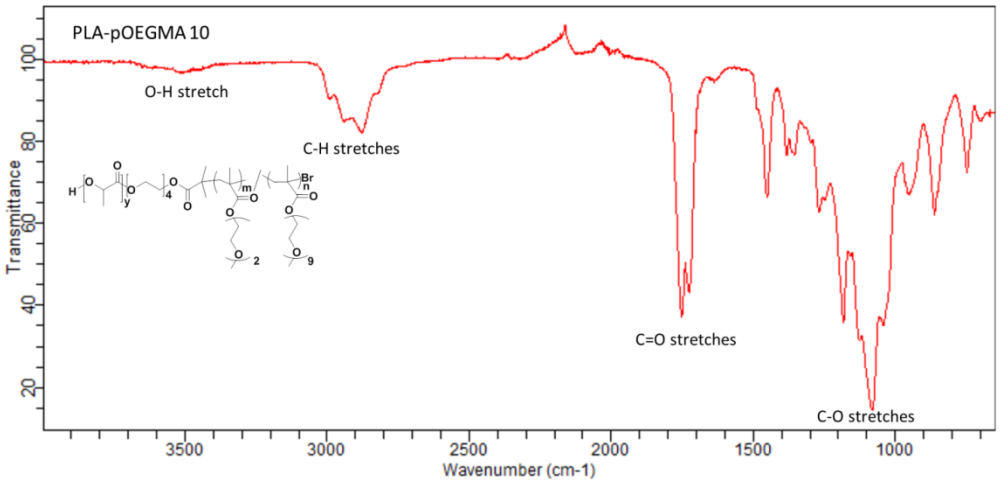

Figure S20: PLA-pOEGMA 10 IR.

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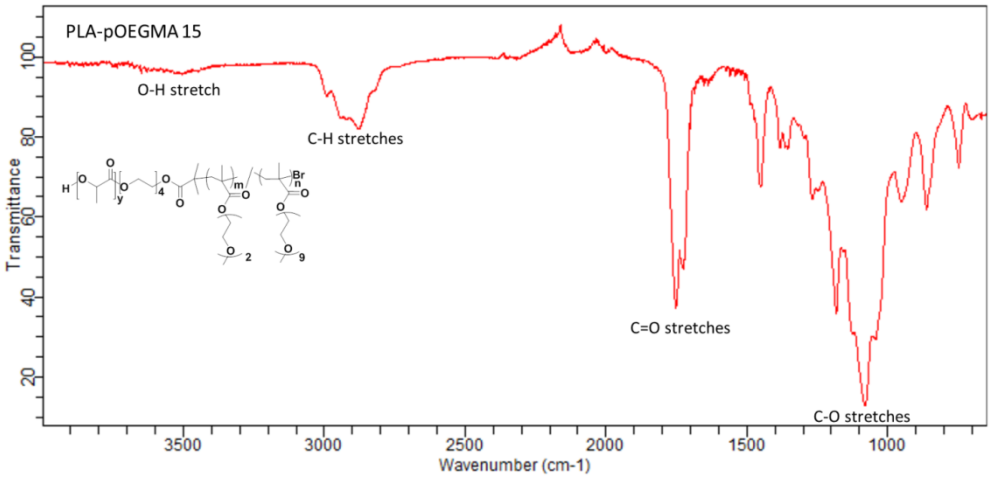

Figure S21: PLA-pOEGMA 15 IR.

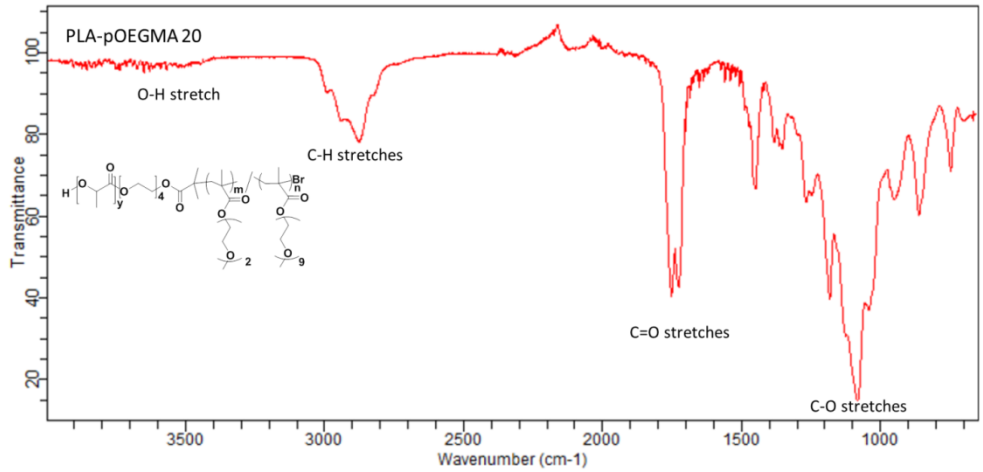

Figure S22: PLA-pOEGMA 20 IR.

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DLS analysis: size and zeta potential of polymer blends

Table 1: DLS and zeta potential of each polymer blend.

Polymer formulation	Diameter (nm)	ζ (mV)
PLA-pDEGMA 100	118.00 ± 7.37	-18.1
PLA-pDEGMA 100 (75% w/w) - PLA-pOEGMA 5 (25% w/w)	113.27 ± 2.97	-14
PLA-pDEGMA 100 (50% w/w) - PLA-pOEGMA 5 (50% w/w)	105.77 ± 1.37	-12.6
PLA-pDEGMA 100 (25% w/w) - PLA-pOEGMA 5 (75% w/w)	109.67 ± 1.31	-13.3
PLA-pOEGMA 5	120.93 ± 1.38	-24.5
PLA-pDEGMA 100 (75% w/w) - PLA-pOEGMA 10 (25% w/w)	106.47 ± 0.83	-11.5
PLA-pDEGMA 100 (50% w/w) - PLA-pOEGMA 10 (50% w/w)	114.27± 2.4	-7.35
PLA-pDEGMA 100 (25% w/w) - PLA-pOEGMA 10 (75% w/w)	109.53 ± 2.05	-6.98
PLA-pOEGMA 10	111.40 ± 5.11	-12
PLA-pDEGMA 100 (75% w/w) - PLA-pOEGMA 15 (25% w/w)	106.30 ± 1.55	-13.6
PLA-pDEGMA 100 (50% w/w) - PLA-pOEGMA 15 (50% w/w)	114.03 ± 1.4	-12.1
PLA-pDEGMA 100 (25% w/w) - PLA-pOEGMA 15 (75% w/w)	118.13 ± 3.94	-5.32
PLA-pOEGMA 15	110.87 ± 3.7	-5.51
PLA-pDEGMA 100 (75% w/w) - PLA-pOEGMA 20 (25% w/w)	109.73 ± 2.75	-6.67
PLA-pDEGMA 100 (50% w/w) - PLA-pOEGMA 20 (50% w/w)	127.57 ± 8.8	-5.14
PLA-pDEGMA 100 (25% w/w) - PLA-pOEGMA 20 (75% w/w)	137.23 ± 1.72	-4.3
PLA-pOEGMA 20	138.53 ± 9.28	-4.04
PLA-pOEGMA 10 (75% w/w) - PLA-pOEGMA 15 (25% w/w)	123.00 ± 6.26	-6.29
PLA-pOEGMA 10 (50% w/w) - PLA-pOEGMA 15 (50% w/w)	100.20 ± 0.92	-5.24
PLA-pOEGMA 10 (25% w/w) - PLA-pOEGMA 15 (75% w/w)	99.78 ± 0.89	-4.66
Mixtures of pre-formed nanoparticles		
PLA-pDEGMA 100 (75% w/w) - PLA-pOEGMA 5 (25% w/w) NP	108.93 ± 1.4	-16.2
PLA-pDEGMA 100 (50% w/w) - PLA-pOEGMA 5 (50% w/w) NP	109.17 ± 1.6	-15.5
PLA-pDEGMA 100 (25% w/w) - PLA-pOEGMA 5 (75% w/w) NP	110.33 ± 1.95	16.5
PLA-pDEGMA 100 (75% w/w) - PLA-pOEGMA 10 (25% w/w) NP	83.51 ± 1.53	-10.1
PLA-pDEGMA 100 (50% w/w) - PLA-pOEGMA 10 (50% w/w) NP	87.49 ± 1.54	-12.5
PLA-pDEGMA 100 (25% w/w) - PLA-pOEGMA 10 (75% w/w) NP	90.43 ± 0.8	-9.01
PLA-pDEGMA 100 (75% w/w) - PLA-pOEGMA 15 (25% w/w) NP	98.09 ± 1.13	-12.7
PLA-pDEGMA 100 (50% w/w) - PLA-pOEGMA 15 (50% w/w) NP	103.23 ± 1.67	-10.4
PLA-pDEGMA 100 (25% w/w) - PLA-pOEGMA 15 (75% w/w) NP	108.23 ± 2.39	-7.06
PLA-pDEGMA 100 (75% w/w) - PLA-pOEGMA 20 (25% w/w) NP	118.03 ± 1.52	-15.3
PLA-pDEGMA 100 (50% w/w) - PLA-pOEGMA 20 (50% w/w) NP	121.10 ± 2.5	-8.74
PLA-pDEGMA 100 (25% w/w) - PLA-pOEGMA 20 (75% w/w) NP	124.67 ± 1.47	-5.56
PLA-pOEGMA 10 (75% w/w) - PLA-pOEGMA 15 (25% w/w) NP	90.57 ± 1.36	-5.11
PLA-pOEGMA 10 (50% w/w) - PLA-pOEGMA 15 (50% w/w) NP	88.23 ± 0.95	-5.24
PLA-pOEGMA 10 (25% w/w) - PLA-pOEGMA 15 (75% w/w) NP	87.32 ± 1.39	-4.6

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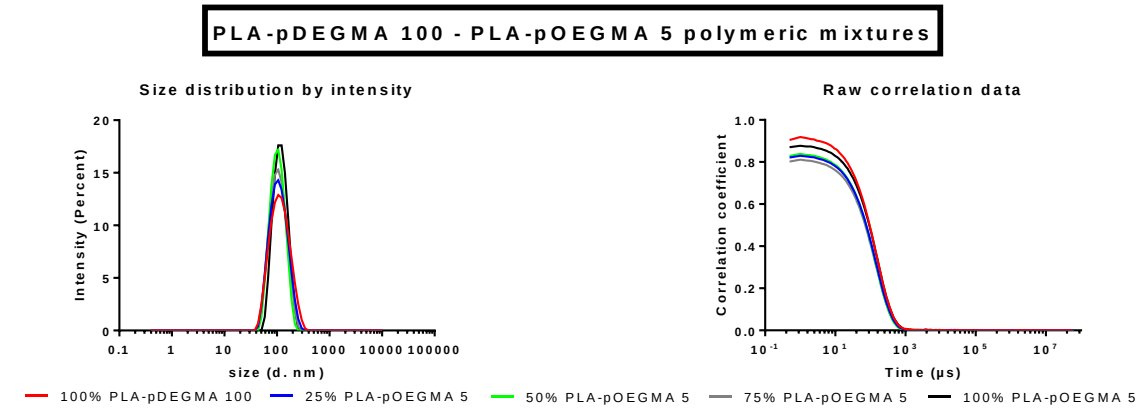

Figure S23: particle size of PLA-DEGMA 100 – PLA-pOEGMA 5 polymeric blends.

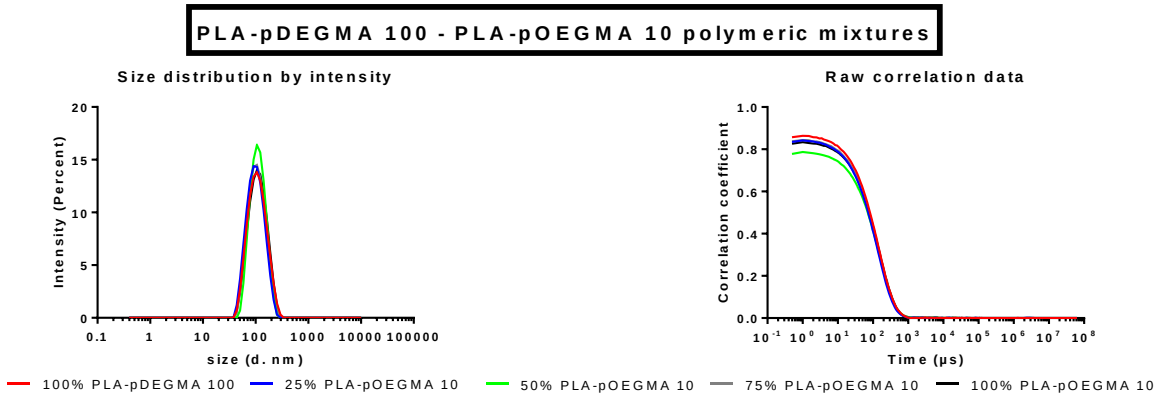

Figure S24: particle size of PLA-DEGMA 100 – PLA-pOEGMA 10 polymeric blends.

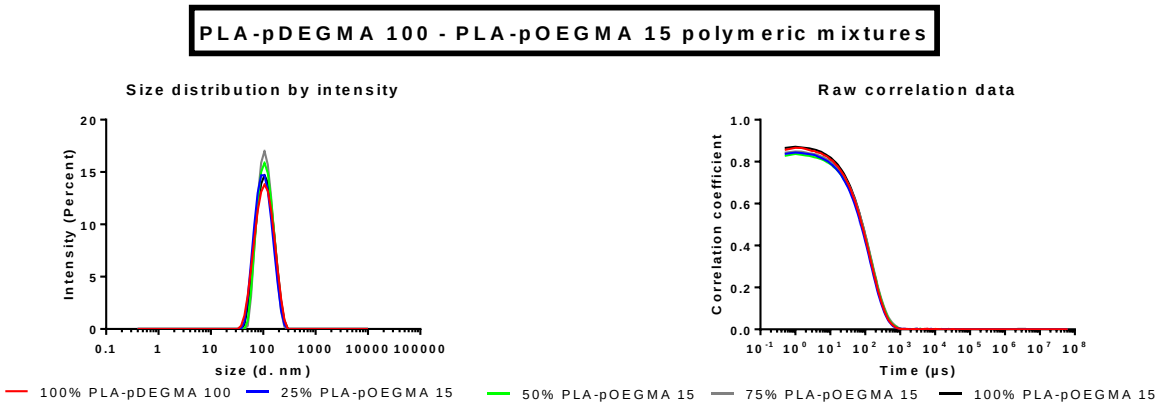

Figure S25: particle size of PLA-DEGMA 100 – PLA-OEGMA 15 polymeric blends.

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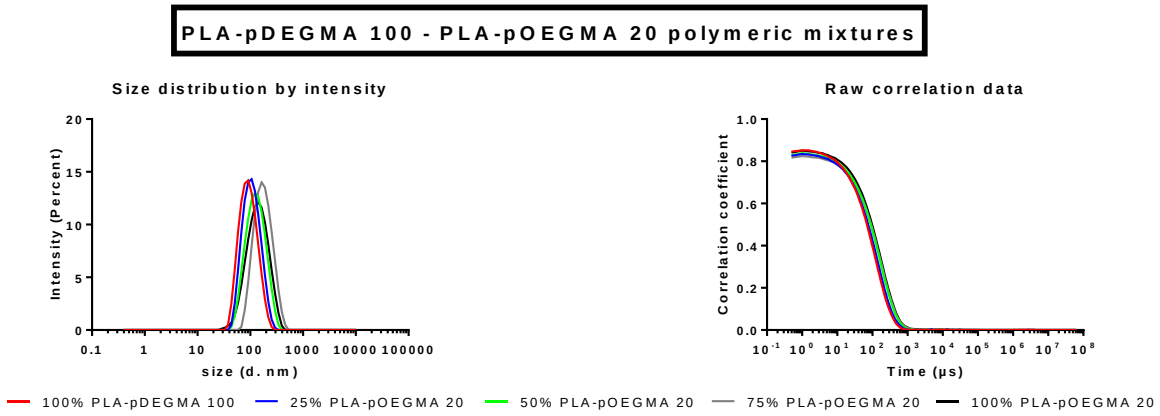

Figure S26: particle size of PLA-DEGMA 100 – PLA-OEGMA 20 polymeric blends.

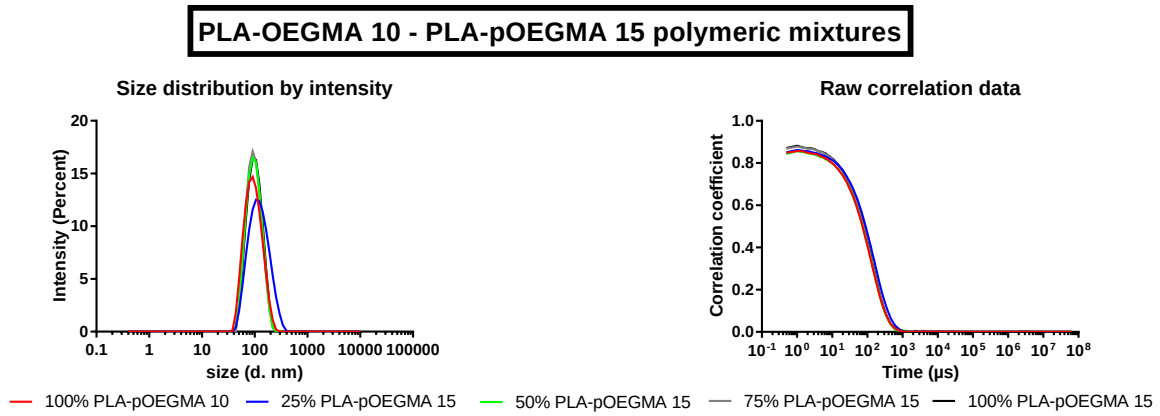

Figure S27: particle size of PLA-OEGMA 10 – PLA-OEGMA 15 polymeric blends.

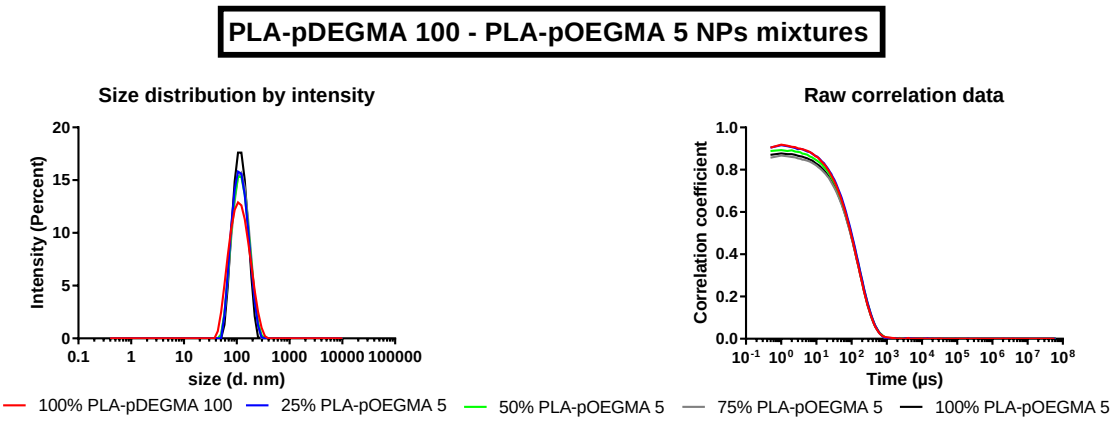

Figure S28: particle size of PLA-pDEGMA 100 – PLA-pOEGMA 5 nanoparticles blends.

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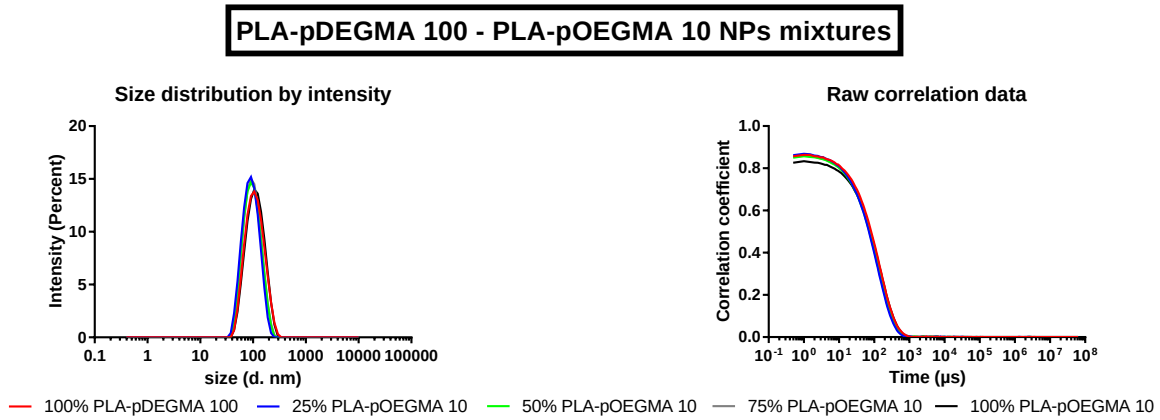

Figure S29: particle size of PLA-pDEGMA 100 – PLA-pOEGMA 10 nanoparticles blends.

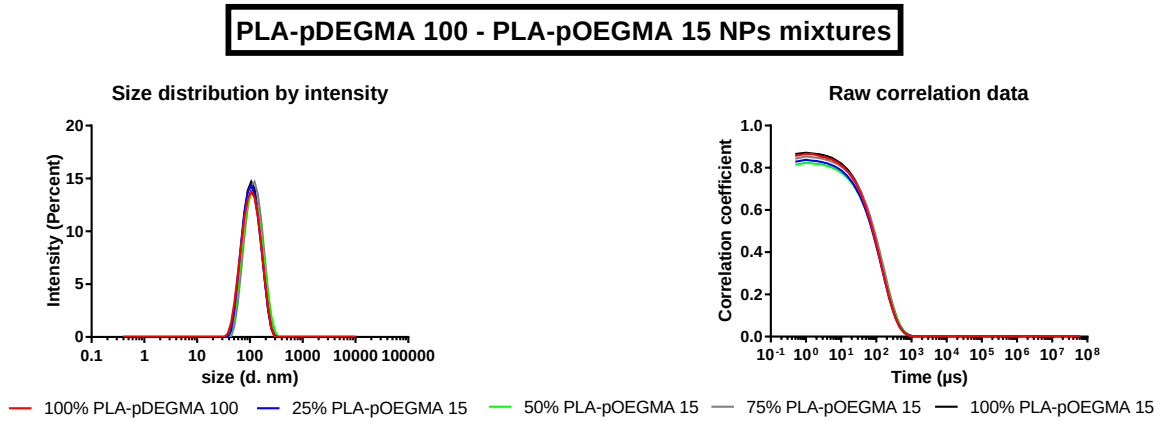

Figure S30: particle size of PLA-pDEGMA 100 – PLA-pOEGMA 15 nanoparticles blends.

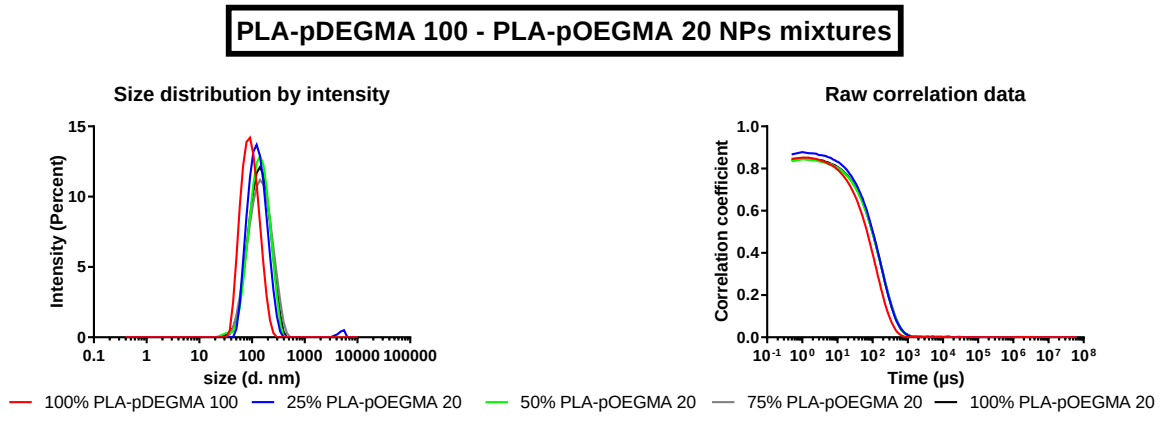

Figure S31: particle size of PLA-pDEGMA 100 – PLA-pOEGMA 20 nanoparticles blends.

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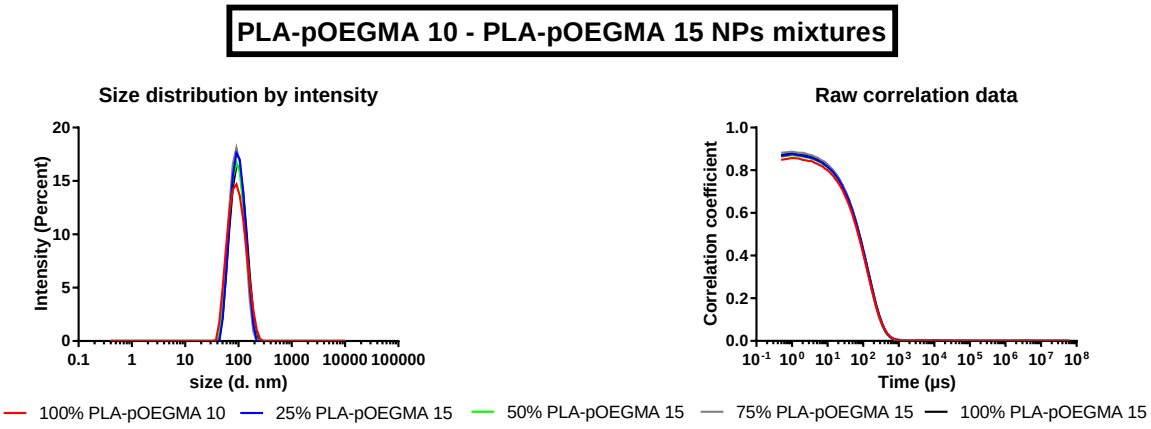
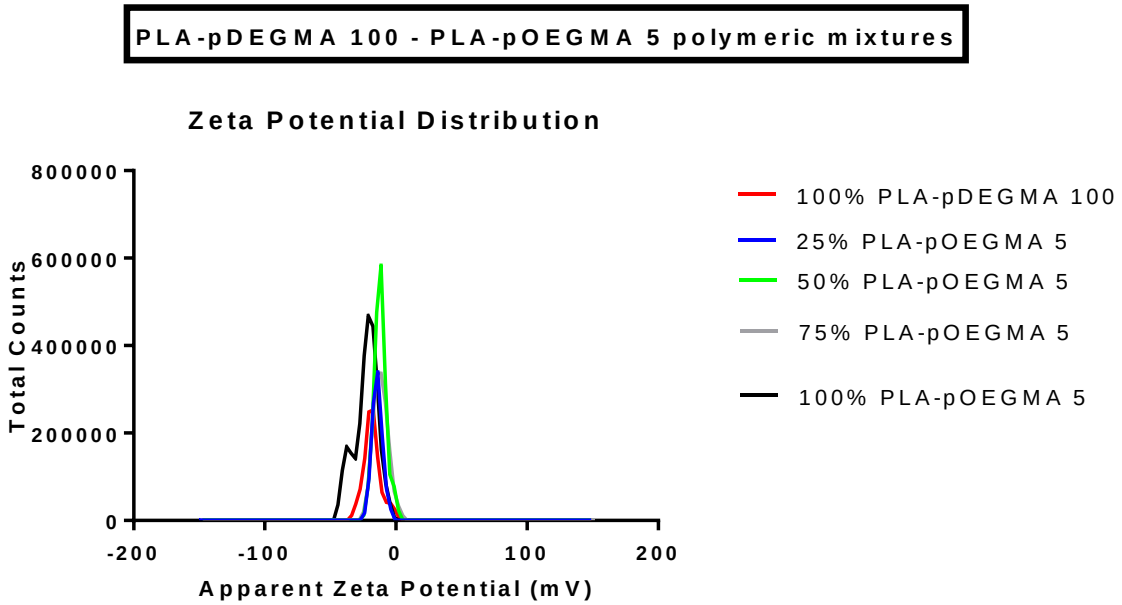


Figure S32: particle size of PLA-pOEGMA 10 -PLA-pOEGMA 15 nanoparticles blends.



FigureS33: zeta potential of PLA-pDEGMA 100 - PLA-pOEGMA 5 polymeric blends.

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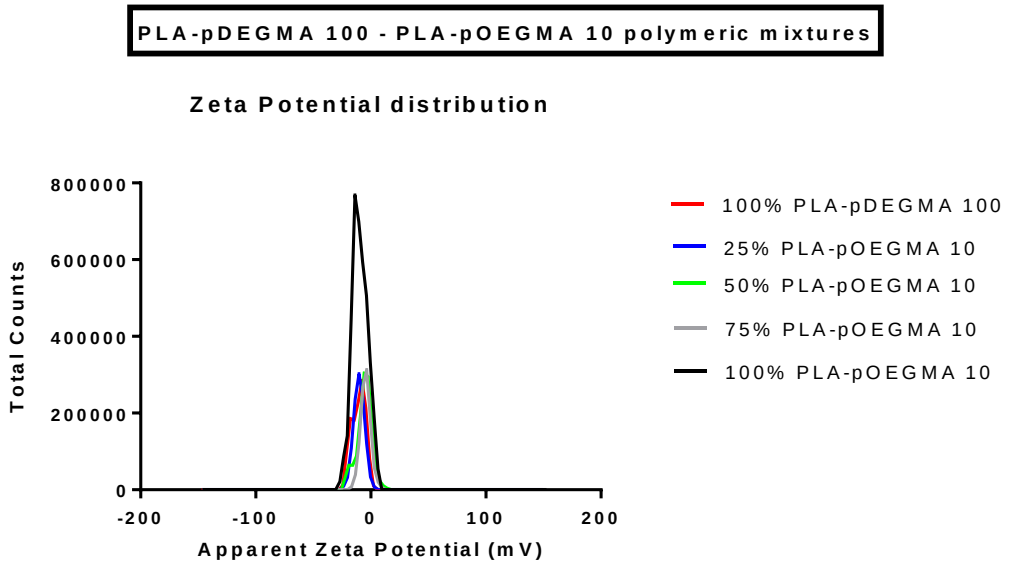

Figure S34: zeta potential of PLA-pDEGMA 100 - PLA-pOEGMA 10 polymeric blends.

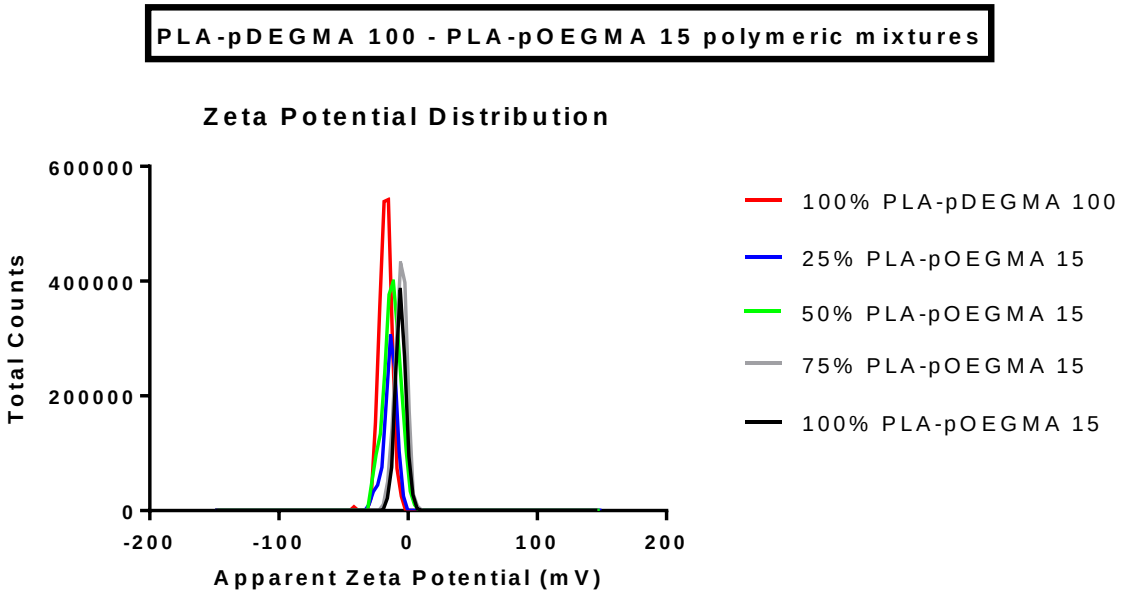

Figure S35: zeta potential of PLA-pDEGMA 100 - PLA-pOEGMA 15 polymeric blends.

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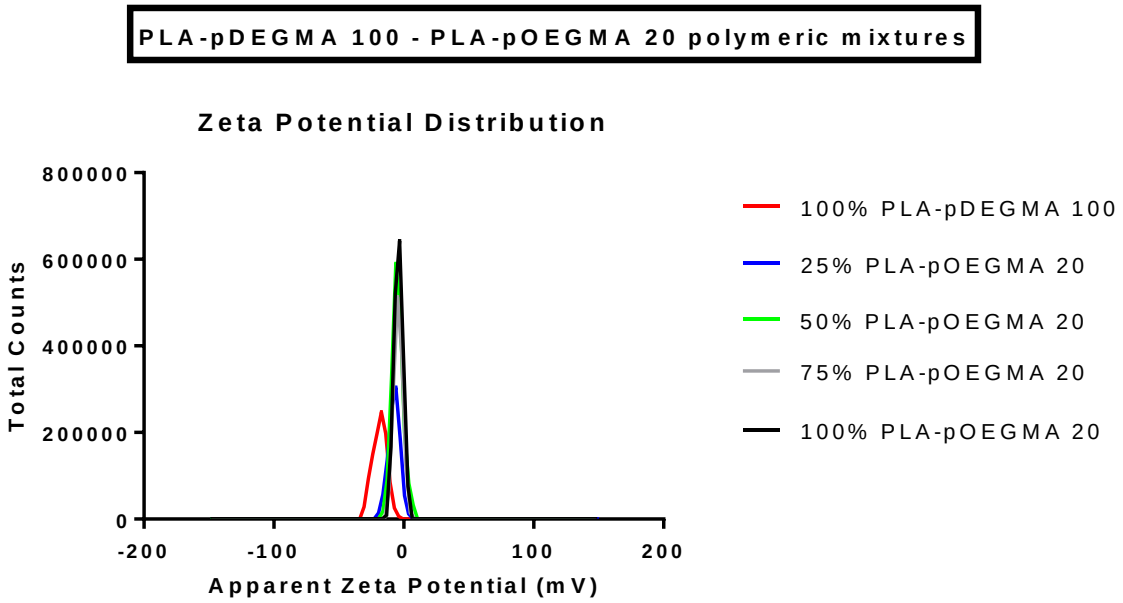

Figure S36: zeta potential of PLA-pDEGMA 100 - PLA-pOEGMA 20 polymeric blends.

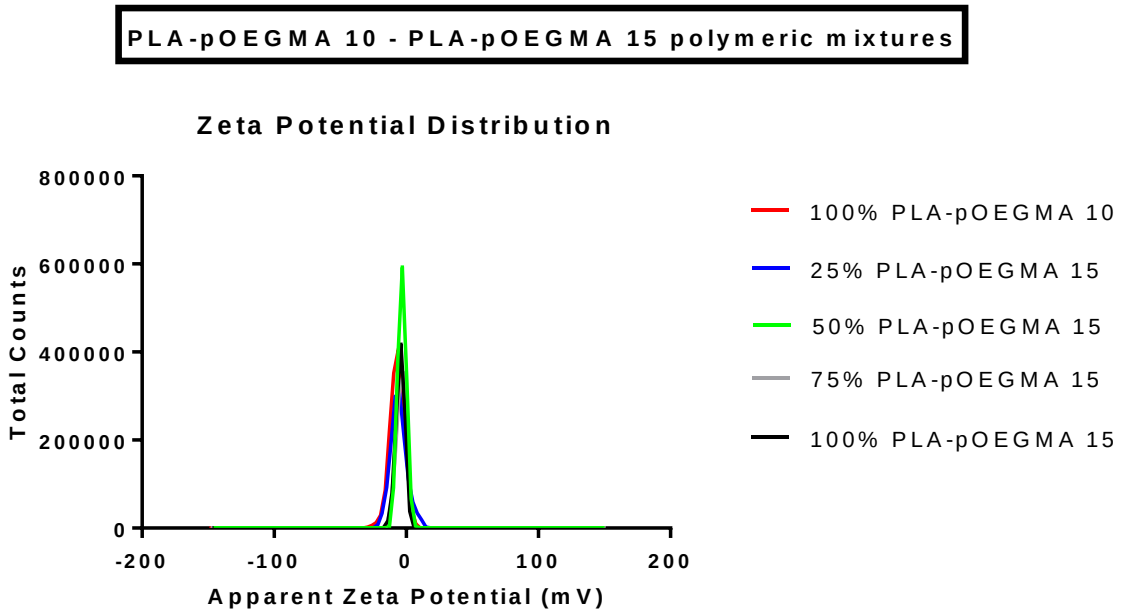

Figure S37: zeta potential of PLA-pOEGMA 10 - PLA-pOEGMA 15 polymeric blends.

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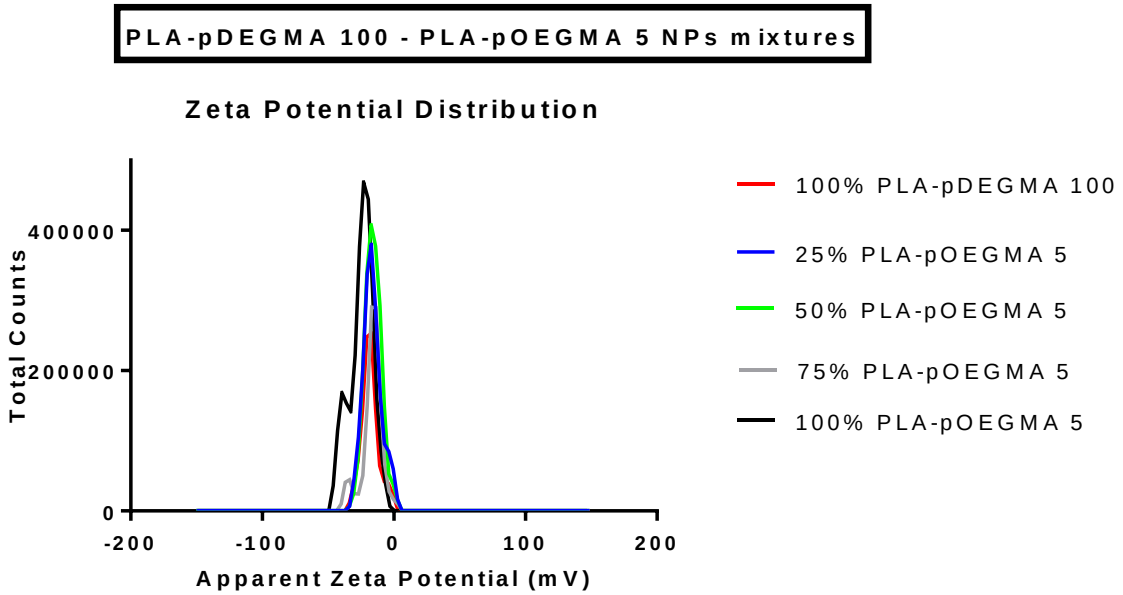

FigureS38: zeta potential of PLA-pDEGMA 100 - PLA-pOEGMA 5 nanoparticles blends.

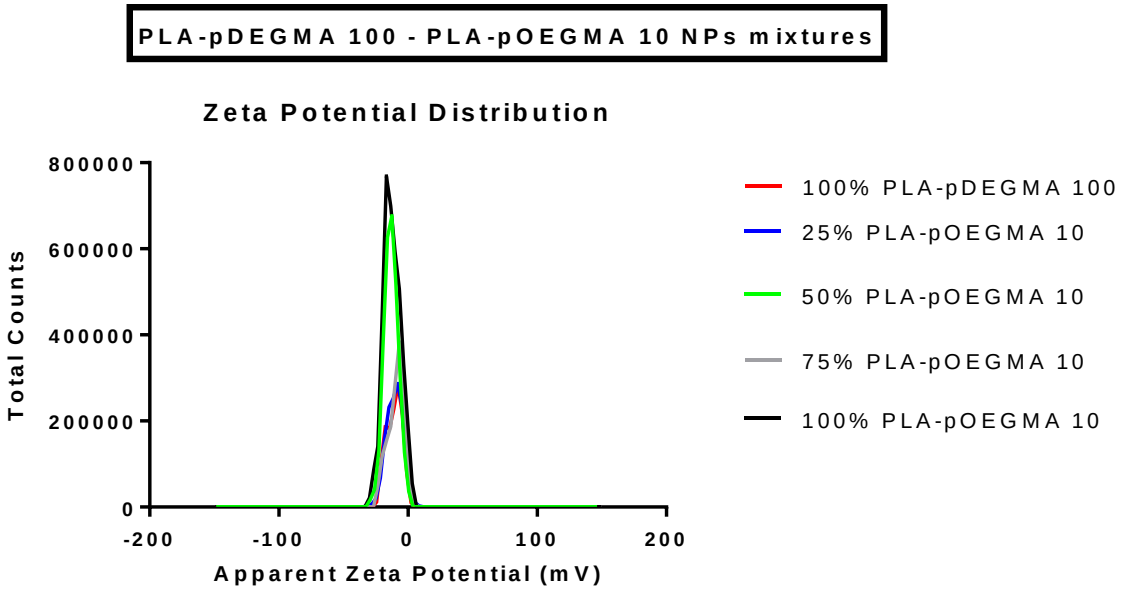

Figure S39: zeta potential of PLA-pDEGMA 100 - PLA-pOEGMA 10 nanoparticles blends.

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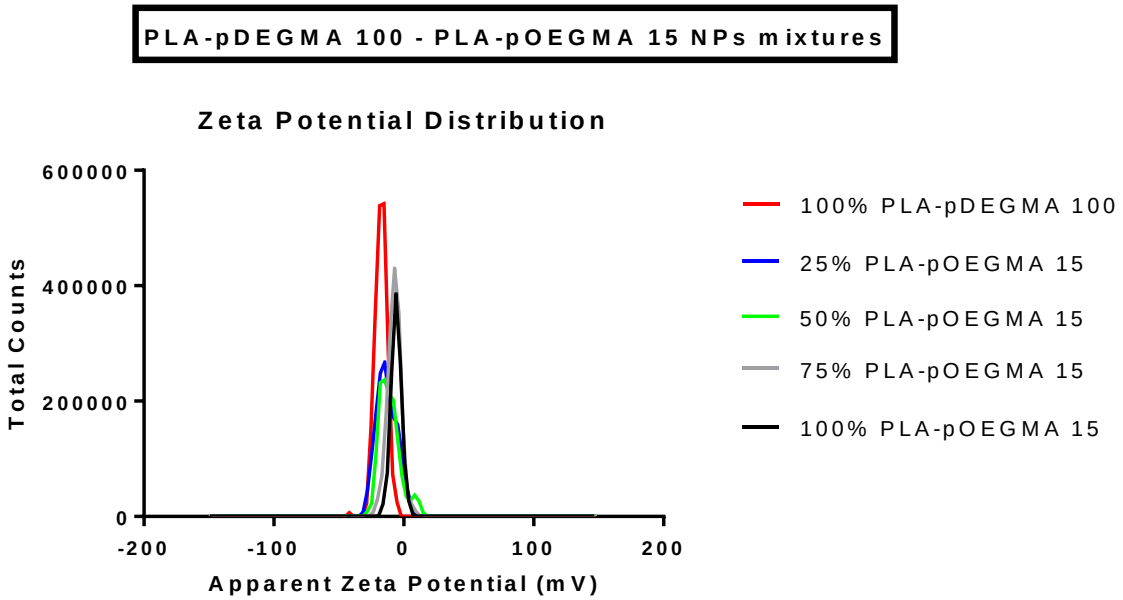

Figure S40: zeta potential of PLA-pDEGMA 100 - PLA-pOEGMA 15 nanoparticles blends.

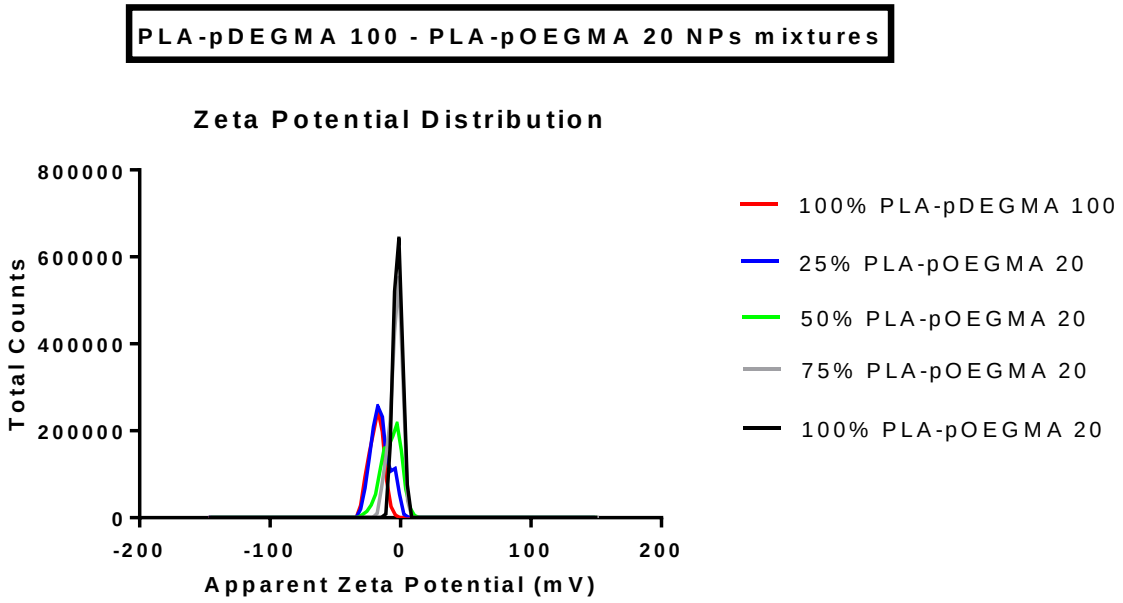

Figure S41: zeta potential of PLA-pDEGMA 100 - PLA-pOEGMA 20 nanoparticles blends.

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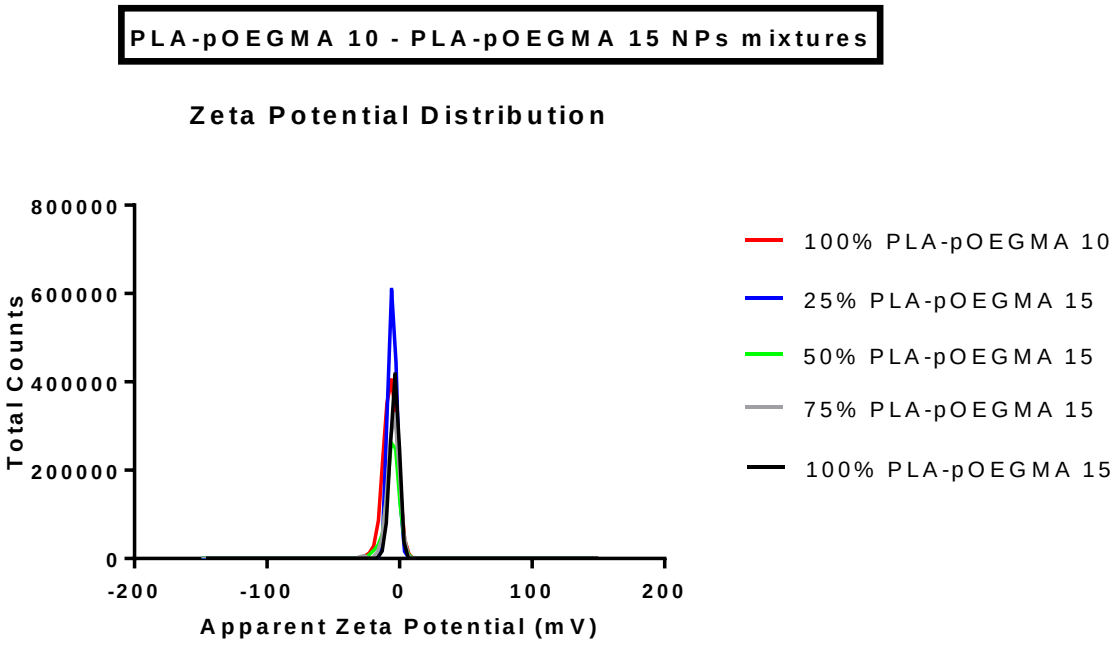

Figure S42: zeta potential of PLA-pOEGMA 10 - PLA-pOEGMA 15 nanoparticles blends.

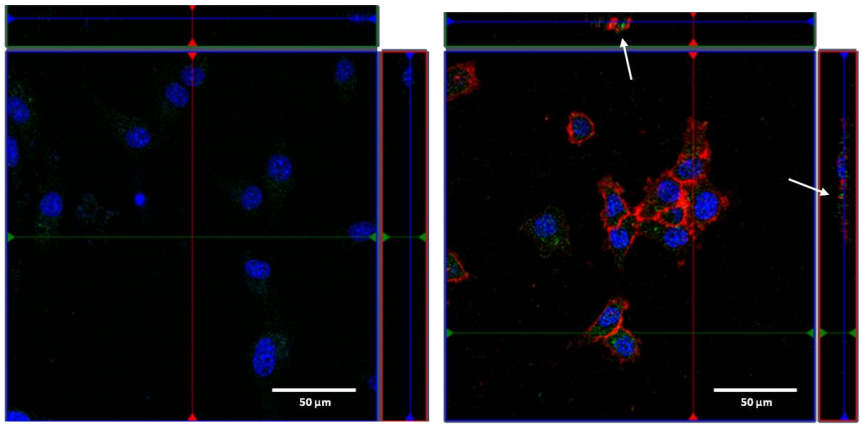

Figure S43: orthogonal projection of a 3D image acquired on 3T3 (left) HCT116 cells (right) treated with 250 µg/ml of PLA-pOEGMA 5 overnight at 37°C. The arrows point at the internalization of the nanoparticles (Green) where its fluorescence can be localised within the boundaries of the red membrane. Nuclei: Blue. Scale bars represent 50 µm.

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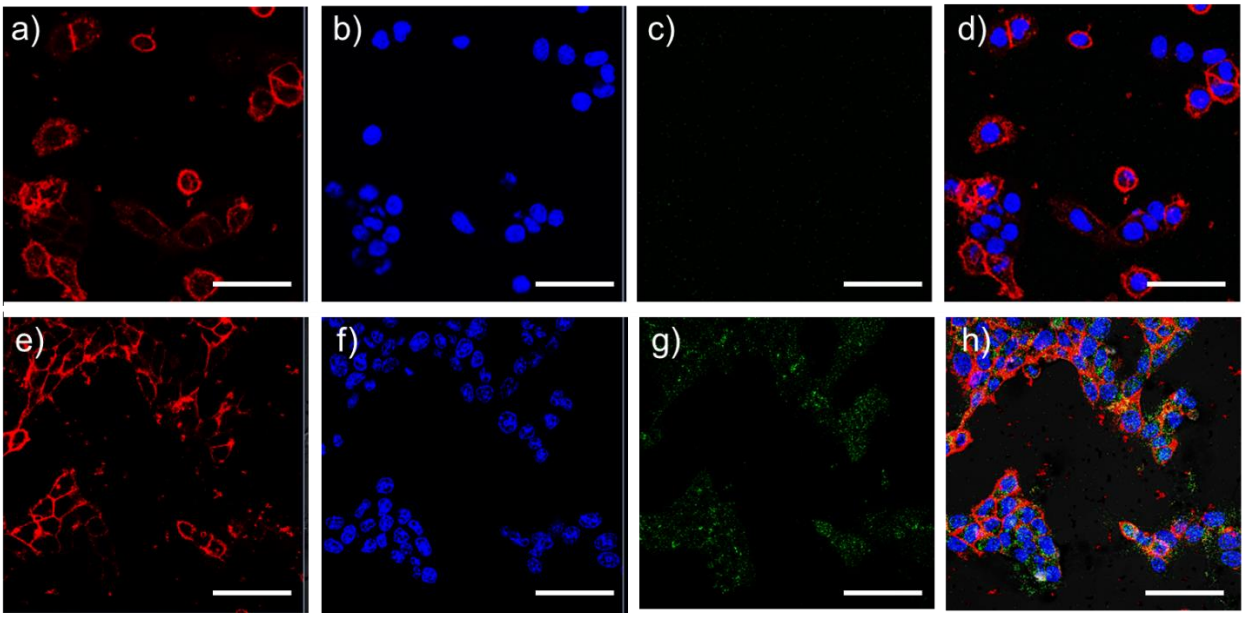

Figure S44: confocal microscopy images of HCT116 cells incubated without, and with PLA-b-P((DEGMA)-stat-(OEGMA)) polymer (PLA-pOEGMA 5 nanoparticles) at a concentration of 250 µg/ml. Plates a-d show HCT116 cells labelled with CellMask deep red membrane stain (red), Hoechst nuclear stain (blue). Plates e-h show HCT116 cells incubated with polymers for 1 h at 25 °C then overnight at 37 °C prior to imaging. Polymers were labelled with Rhodamine B which has been re-coloured in green for easy distinction of the micelles from the Deep Red CellMask dye. Scale bars represent 50 µm.

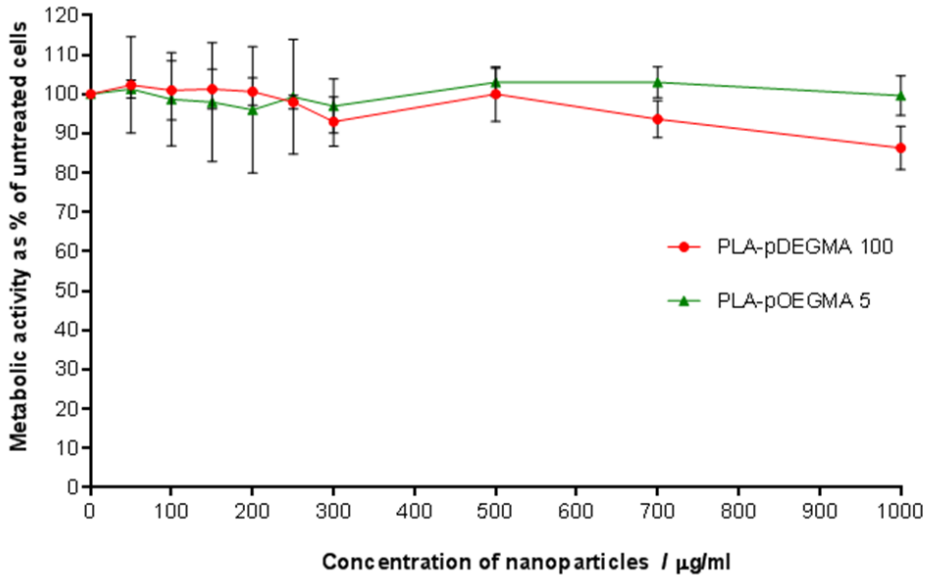

Figure S45: Metabolic activities of HCT116 cells incubated with PLA-b-pDEGMA or PLA-b-P((DEGMA)-stat-(OEGMA)) polymer (PLA-pDEGMA 100 and PLA-pOEGMA 5 nanoparticles).

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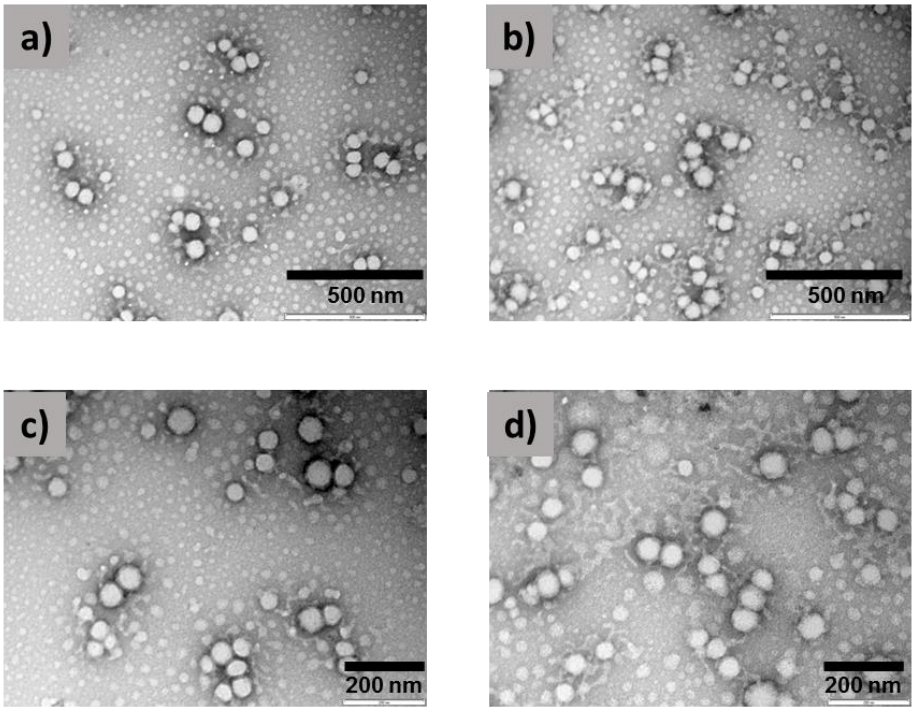

Figure S46: Transmission Electron Microscopy of representative PLA-pOEGMA 10 (75% w/w) - PLA-pOEGMA 15 (25% w/w) nanoparticles). Samples were imaged before (a, c), or after (b, d) heating above their thermal transition temperature. Sample magnification – x 60,000 (a, b) or x 87,000 (c, d).

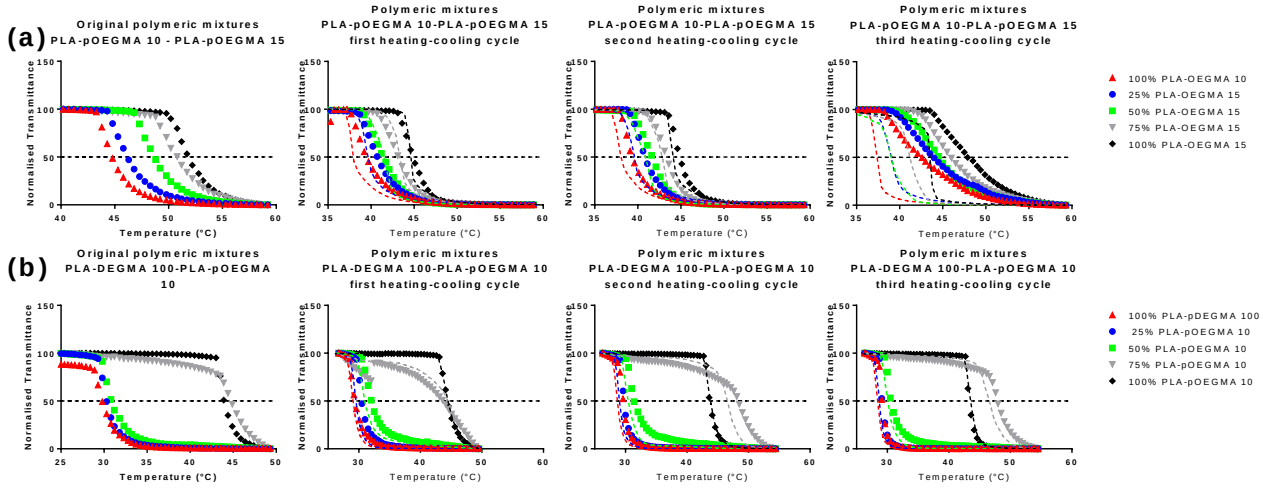

Figure S47: turbidimetric heating-cooling cycles, normalised transmittance. (a) PLA-pOEGMA 10 – PLA-pOEGMA 15 blends. (b) PLA-pOEGMA 0 – PLA-pOEGMA 10 blends.

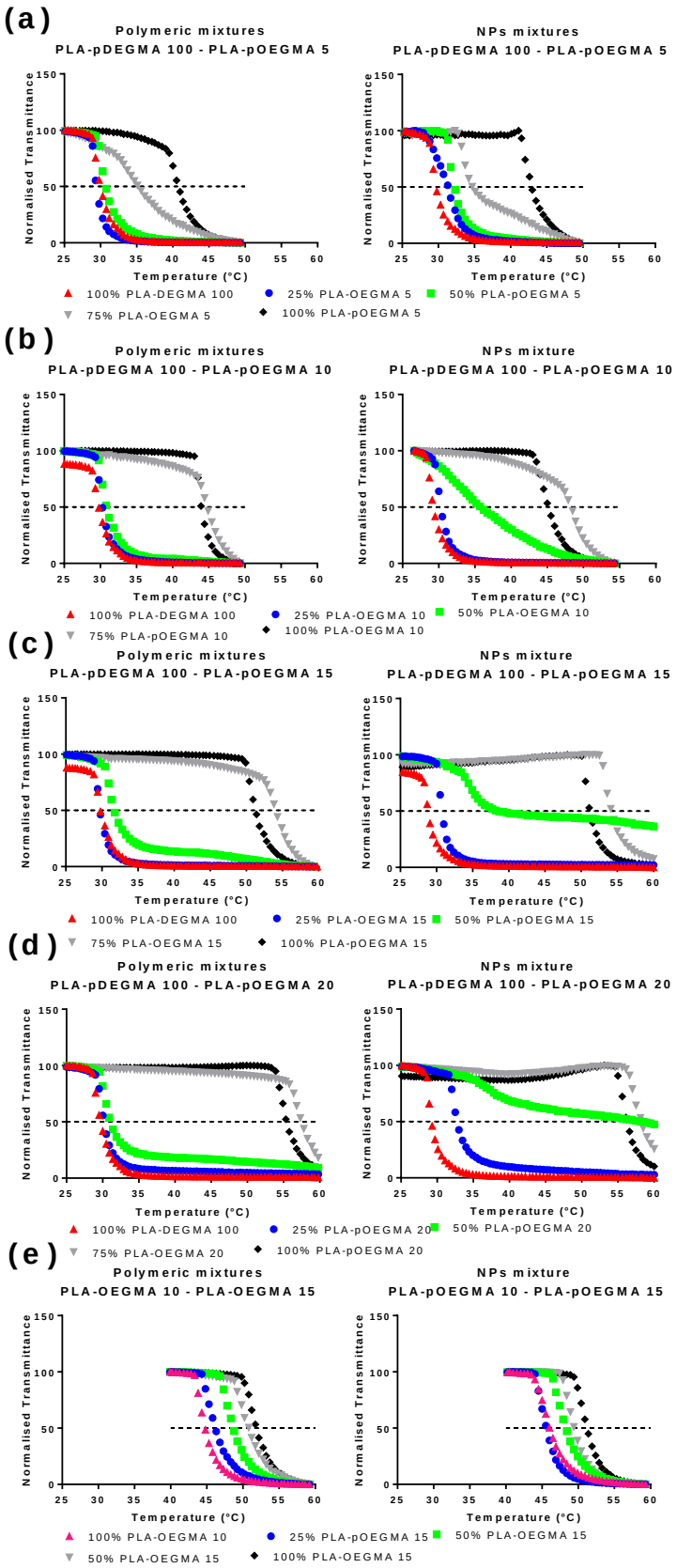

Figure S48: Turbidimetric analysis, normalised transmittance, of: (a) PLA-pDEGMA 100 – PLA-pOEGMA 5, (b) PLA-pDEGMA 100 – PLA-pOEGMA 10, (c) PLA-pDEGMA 100 – PLA-pOEGMA 15, (d) PLA-pDEGMA 100 – PLA-pOEGMA 20 and (e) PLA-pOEGMA 10 – PLA-pOEGMA 15 blends. On the left are temperature-turbidity curves for the nanoparticles prepared from binary polymer blends, on the right are the corresponding curves for mixtures of pre-formed single polymer corona nanoparticles.