Supporting information

Design of degradable polyphosphoester networks with tailor-made stiffness and hydrophilicity as scaffolds for tissue engineering

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Synthesis protocols:

Cyclic phosphate monomer synthesis

The commercially available COP was reacted with 1-butanol, 3-buten-1-ol or methanol with formation of the corresponding cyclic phosphates substituted by a n-butyl group (BEP), butenyl group (BenEP) or a methyl group (MEP), respectively. Typically, in a glass flask under N_2 atmosphere, the alcohol (176 mmol) and NEt_3 (25 ml, 176 mmol) were solubilized in 100 mL of anhydrous THF. Then, COP (25 g, 176 mmol) solubilized in 50 mL of anhydrous THF was added drop wise under stirring at 0°C. After complete addition, the resulting mixture was stirred at 0 °C for 6 hrs. Triethylamine hydrochloride salt was then removed by filtration, and the filtrate was concentrated under vacuum before being purified by fractionated distillation under vacuum (10^{-2} Torr) with a final yield of 60% for BEP (T_b= 86°C) and BenEP (T_b= 83°C) monomers and 40% for MEP (T_b= 80°C).

BenEP monomer
^{1}H NMR ($CDCl_3$, 400 MHz): δ= 5.8 ppm (m, 1H, CH_2=C**H**-CH_2), 5.2 ppm (m, 2H, C**H**$_2$=CH-CH_2), 4.4 ppm (m, 4H, O-C**H**$_2$-C**H**$_2$-O), 4.1 ppm (m, 2H, -O-C**H**$_2$-CH_2), 2.4 ppm (m, 2H, CH_2-C**H**$_2$-CH=CH_2).
^{31}P NMR ($CDCl_3$): δ= 17.5 ppm.

BEP monomer
^{1}H NMR ($CDCl_3$, 400 MHz): δ= 4.4 ppm (m, 4H, O-C**H**$_2$-C**H**$_2$-O), 4.15 ppm (m, 2H, -O-C**H**$_2$-CH_2-CH_2), 1.7 ppm (m, 2H, CH_2-C**H**$_2$-CH_2-CH_3), 1.4 ppm (m, 2H, CH_2-CH_2-C**H**$_2$-CH_3), 1.4 ppm (t, 3H, CH_2-CH_2-CH_2-C**H**$_3$).
^{31}P NMR ($CDCl_3$, 400 MHz): δ= 17.6 ppm.

MEP monomer
^{1}H NMR ($CDCl_3$, 400 MHz): δ= 4.4 ppm (m, 4H, O−C**H**$_2$−C**H**$_2$-O), 3.8 ppm (d, 3H, O−C**H**$_3$).
^{31}P NMR ($CDCl_3$, 400 MHz): δ= 18.5 ppm.

Copolymer synthesis

Synthesis of Poly(MEP-*co*-BenEP) PPE1:

TU (15 mg, 0.4 mmol), BenEP (2.8 g, 16 mmol) and MEP (1.2 g, 9 mmol) were transferred in a round bottom flask. The mixture was then dried by three azeotropic distillations with anhydrous toluene. 5 ml of benzylic alcohol stock solution (6.2 mmol of benzylic alcohol in 100 ml of anhydrous CH_2Cl_2) (0.31mmol) was added under a N_2 atmosphere. The mixture was cooled down to 0°C, and DBU (0.12 ml, 0.8 mmol) was finally introduced under a N_2 atmosphere with a syringe equipped with a stainless-steel capillary. The reaction medium was stirred at 0°C for 2 hrs. After concentration of the solution under vacuum, the copolymer was precipitated in cold diethyl ether. After decantation, the recovered copolymer was dissolved in methanol and dialyzed against methanol (MWCO = 3.5 kDa) overnight in order to remove impurities. After evaporation of methanol under vacuum, the copolymer was collected and characterized by NMR and SEC analyses.

^{1}H NMR (CD_2Cl_2) δ= 7.5 (m, 5H, aromatic protons), 5.8 ppm (m, 44 H, –CH_2–C**H**=CH_2) ,5.1 ppm (m, 88 H, C**H₂**=CH-CH_2), 4.25 ppm (m, 264 H, O–C**H₂**–C**H₂**–O and O-C**H₂**-CH_2-CH=CH_2 of BenEP + 108 H, O–C**H₂**–C**H₂**–O of MEP), 4.1 ppm (m, 88 H, CH_2–C**H₂**–CH=CH_2), 3.8 ppm (m, 76 H, O–C**H₃**), 2.41 ppm (m, 88 H, O–CH_2–C**H₂**–CH=CH_2)
^{31}P NMR (CD_2Cl_2) δ= -1.37 ppm.
M_n(^{1}H NMR) = 11400 g/mol, Đ = 1.2 (SEC).

Copolymer	m MEP	m BenEP	n BzOH	TU	DBU
PPE1	1.2 g	2.8 g	0.31 mmol	15 mg	0.12 ml
PPE2	2.0 g	2.0 g	0.31 mmol	15 mg	0.12 ml
PPE3	2.8 g	1.2 g	0.31 mmol	15 mg	0.12 ml

Synthesis of Poly(MEP-*co*-BenEP-*co*-BEP) PPET1:

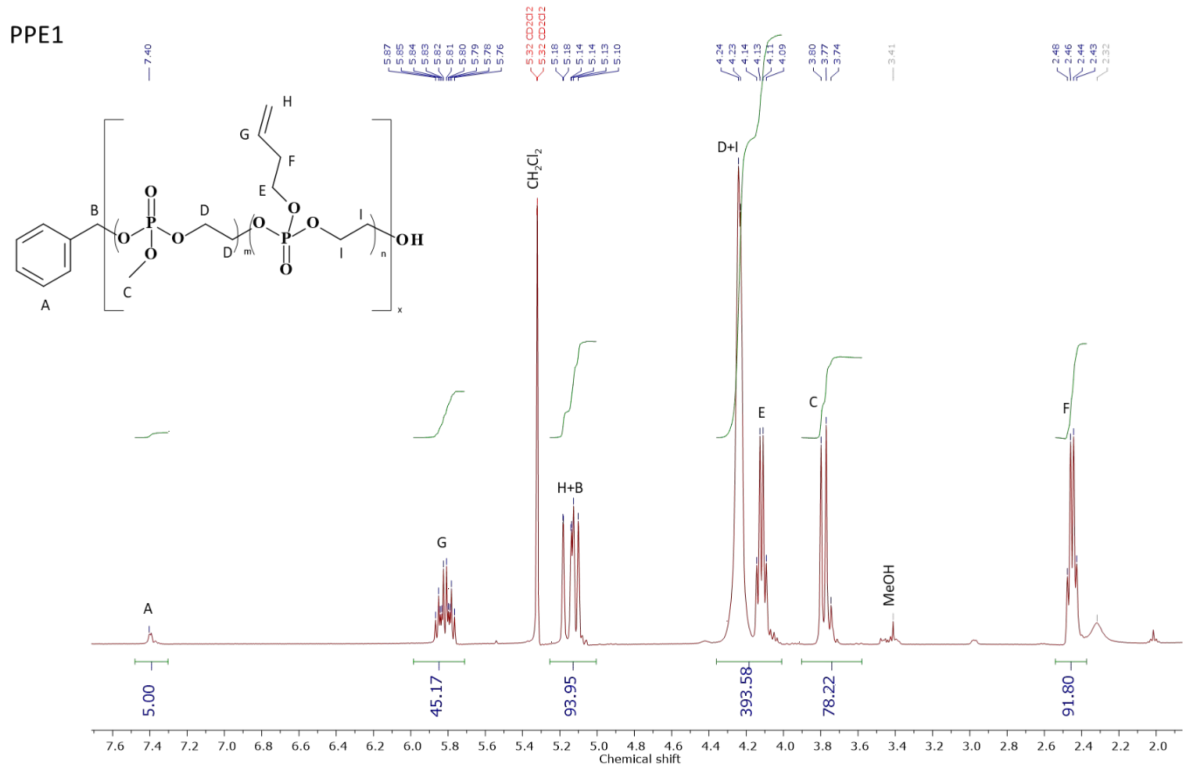
0.15 g (0.4 mmol) of TU, 2 g (14 mmol) of MEP, 1.2 g (6.7 mmol) of BenEP and 0.8 g (4.4 mmol) of BEP were transferred in a round bottom flask. The mixture was then dried by three azeotropic distillations with anhydrous toluene. 5 ml of benzylic alcohol of a stock solution (6.2 mmol BzOH in 100 ml of anhydrous CH_2Cl_2) (0.31mmol) was transferred under a N_2 atmosphere. The mixture was cooled down to 0°C, and DBU (0.12 ml, 0.8 mmol) was finally introduced under a N_2 atmosphere with a syringe equipped with a stainless-steel capillary. The reaction medium was stirred at 0°C for 2 hrs. The terpolymer was purified by precipitation in cold diethyl ether. After decantation, the recovered copolymer was dissolved in methanol and dialyzed against methanol (MWCO = 3.5 kDa) overnight in order to remove impurities. After evaporation of methanol under vacuum, the copolymer was collected and characterized by NMR and SEC analyses.
^{1}H NMR (CD_2Cl_2) δ= 5.8 ppm (m, DP_{BenEP} H, O–CH_2–CH_2-C**H**=CH_2),5.1 ppm (m, 2xDP_{BenEP} H, C**H₂**=CH-CH_2), 4.26 ppm (m, 4xDP_{MEP} H, O–C**H₂**–C**H₂**–O of MEP + 4xDP_{BenEP} H, O–C**H₂**–C**H₂**–O of BenEP + 4xDP_{BEP} H, O–C**H₂**–C**H₂**–O of BEP), 4.1 ppm (m, 2xDP_{BenEP} H, CH_2–C**H₂**–CH=CH_2), 3.8 ppm (m, 3xDP_{MEP} H, O–C**H₃**), 2.46 ppm (m, 2xDP_{BEP} H, O–C**H₂**–CH_2–CH), 1.58 ppm (m,2xDP_{BEP} H, O-CH_2-C**H₂**-CH_2-CH_3), 1.31 ppm (m, 2xDP_{BEP} H CH_3-C**H₂**-CH_2), 0.86 ppm (t, 3xDP_{MEP} H, C**H₃**-CH_2-CH_2).

^{31}P NMR (CD_2Cl_2) δ= -1.33 ppm.
M_n NMR= 6750 g/mol, Đ = 1.3 (SEC).

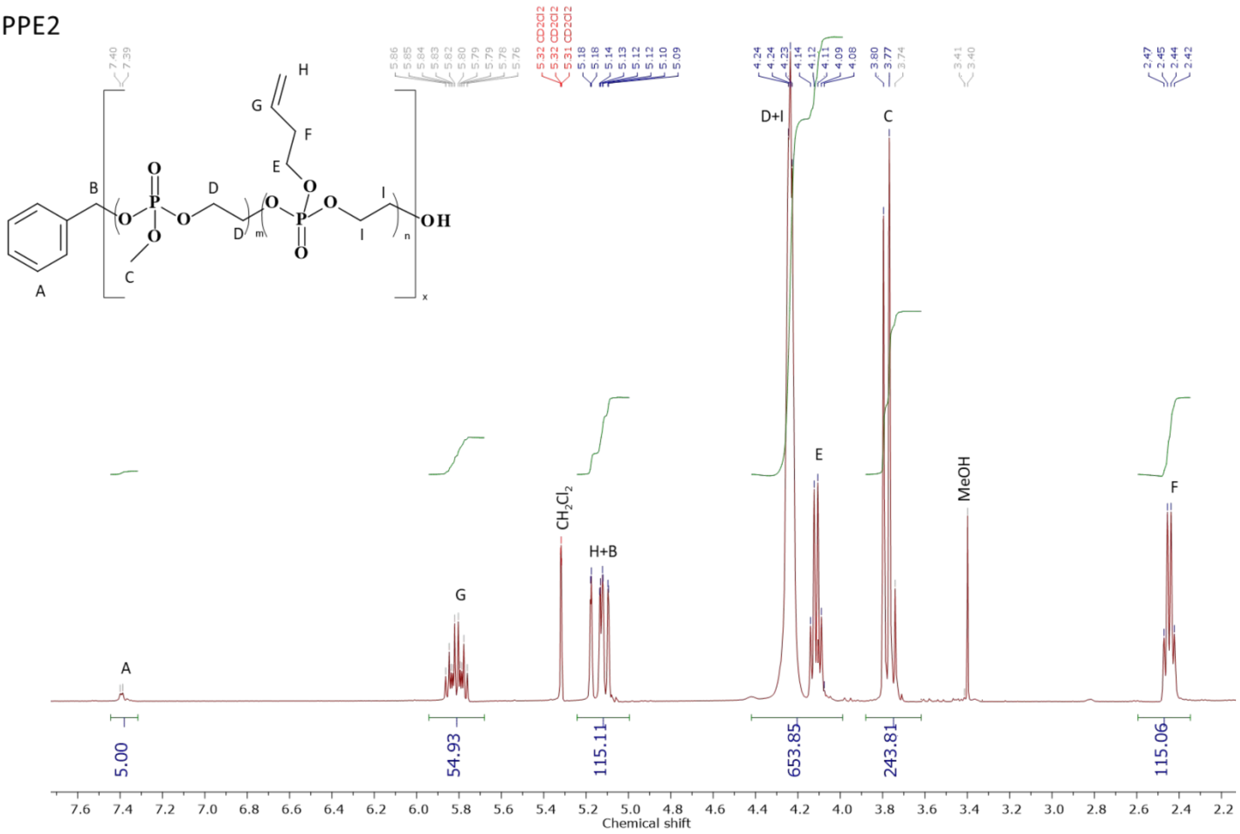
Copolymer	m MEP	m BenEP	m BEP	n BzOH	TU	DBU
PPET1	2.0 g	1.2 g	0.8	0.31 mmol	15 mg	0.12 ml
PPET2	2.0 g	0.8 g	1.2	0.31 mmol	15 mg	0.12 ml

Structural characterization:

^1H and ^{31}P NMR analysis were performed on a Bruker Advance 400 MHz spectrometer in $CDCl_3$ or CD_2Cl_2 using TMS as internal reference at 25 °C in the FT mode. The size exclusion chromatography (SEC) was carried out in DMF (flow rate 1 mL/min) at 40°C using a Water 600 auto sampler liquid chromatograph equipped with a differential refractometer index detector. Waters gel 5 µm (10^5, 10^4, 500, and 100 Å) columns were calibrated with polystyrene standards.



PPE2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5.32 CD2Cl2
5.31 CD2Cl2
5.18
5.18
5.14
5.13
5.12
5.12
5.10
5.09
4.24
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F
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D
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I
C
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O
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OH
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n
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MeOH
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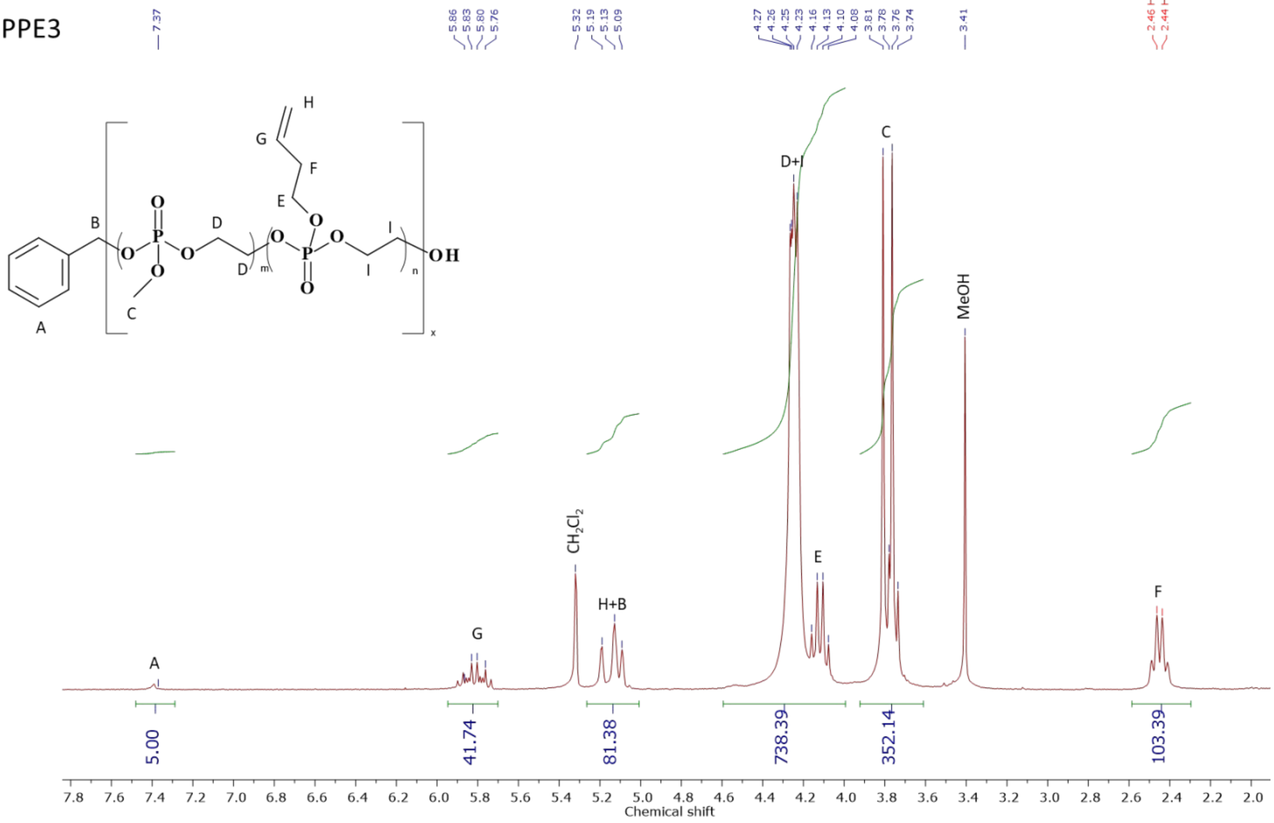
2.2
Chemical shift

PPE3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3.78
3.76
3.74
3.41
2.46 HDO
2.44 HDO
H
G
F
E
B
D
D
I
I
C
A
O
P
OH
m
n
x
D+I
C
MeOH
E
F
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H+B
G
A
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738.39
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7.8
7.6
7.4
7.2
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4.2
4.0
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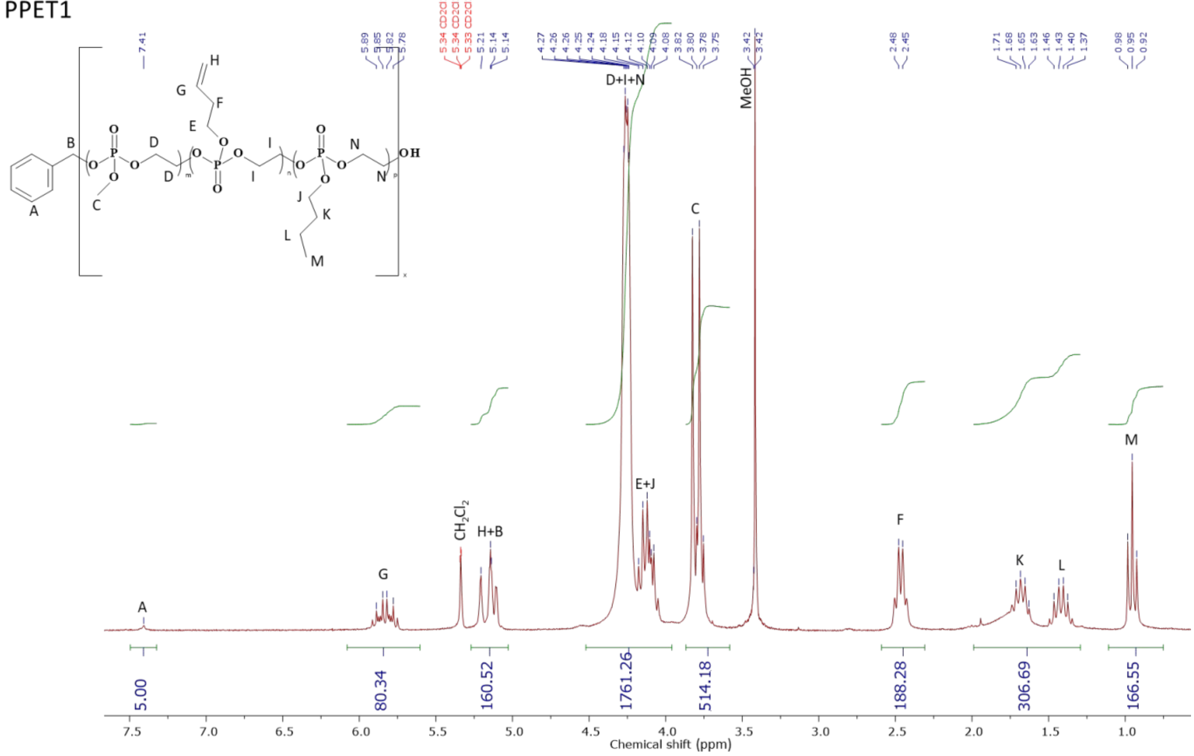
2.2
2.0
Chemical shift

PPET1



PPET2

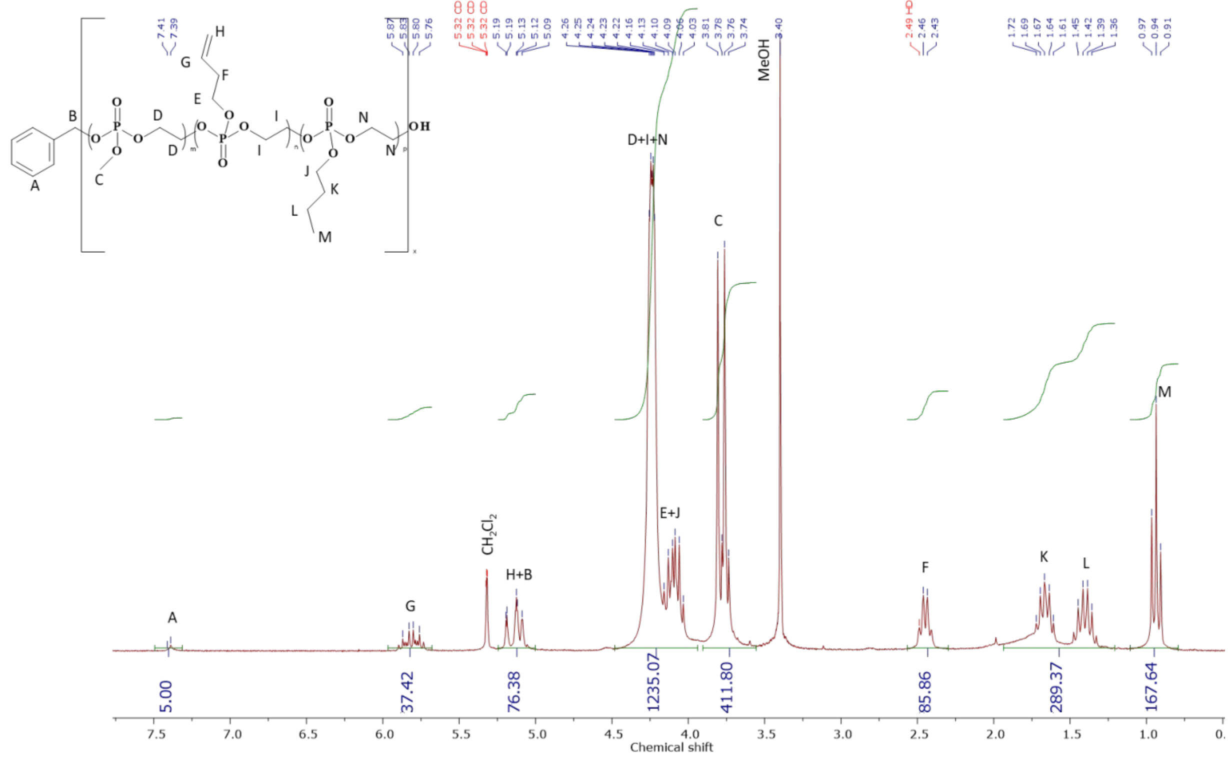
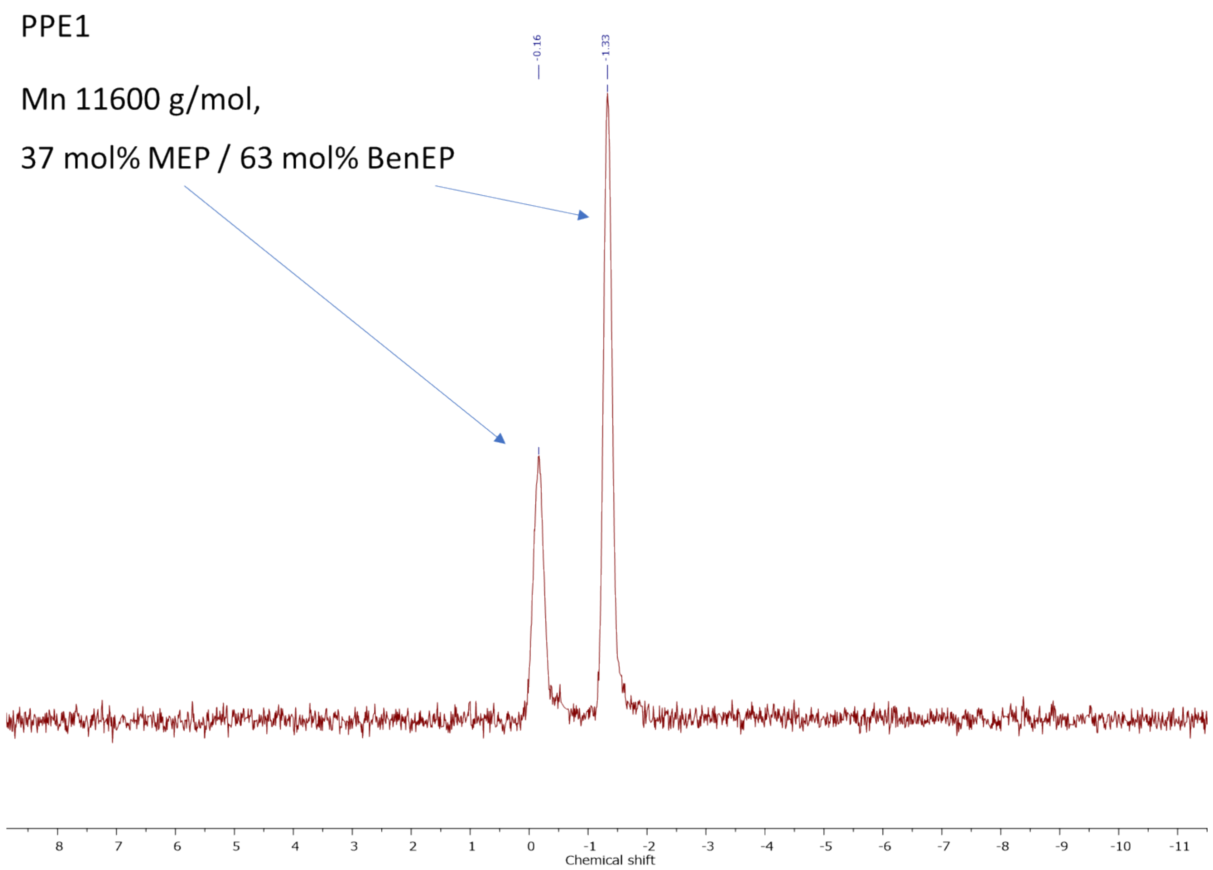
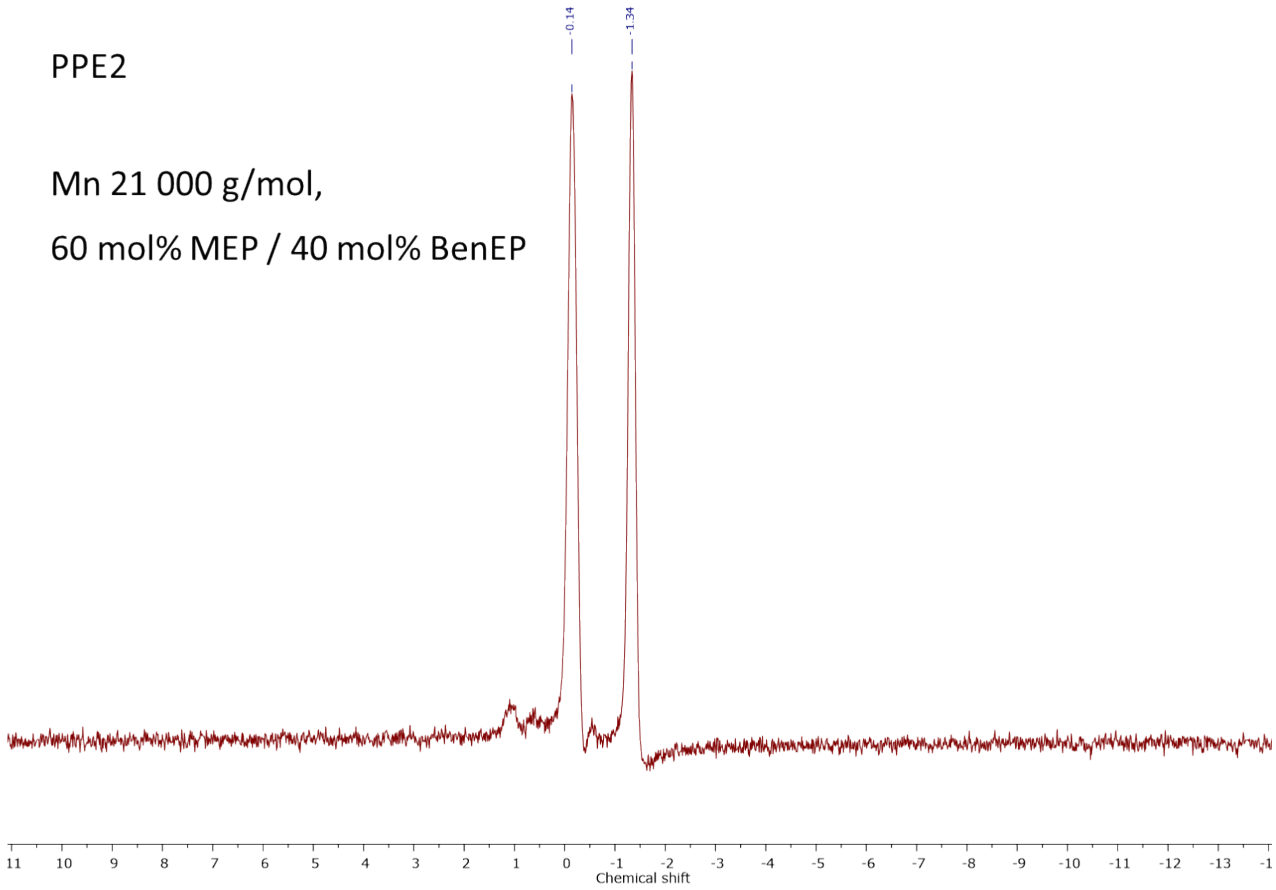


Figure S1: ^{1}H-NMR spectra of the copolymers


PPE1

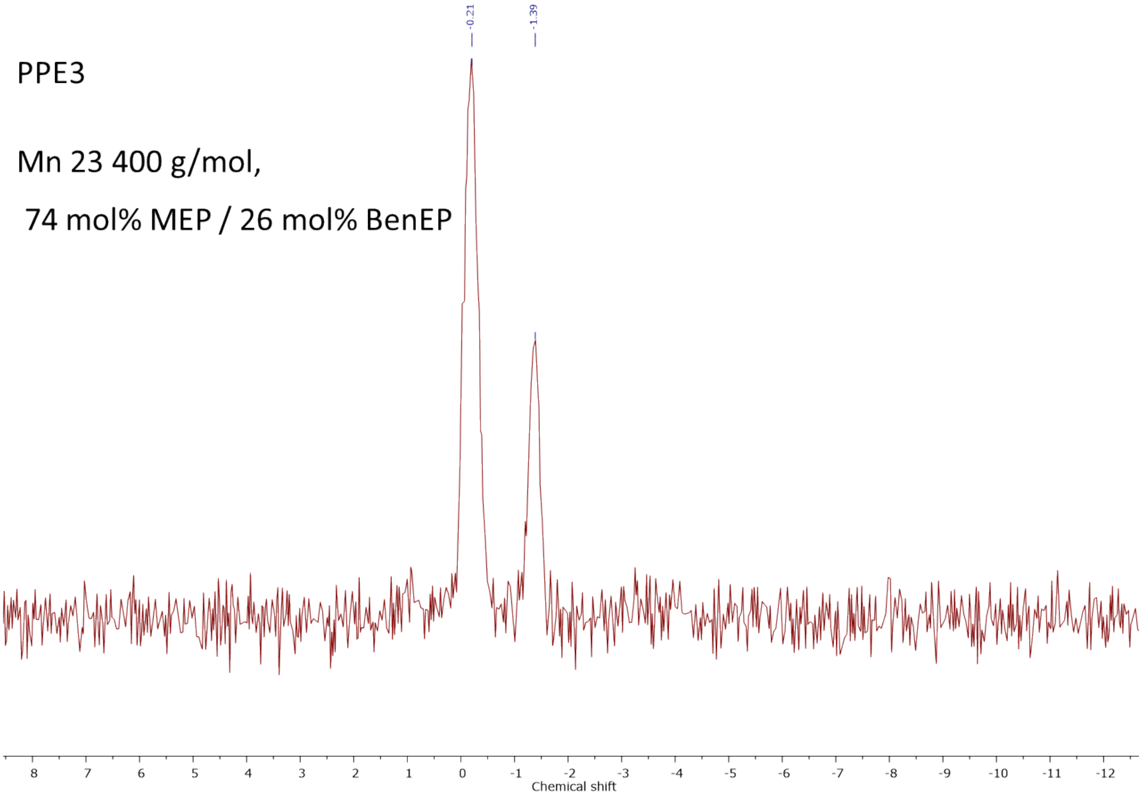
Mn 11600 g/mol,

37 mol% MEP / 63 mol% BenEP


PPE2

Mn 21 000 g/mol,

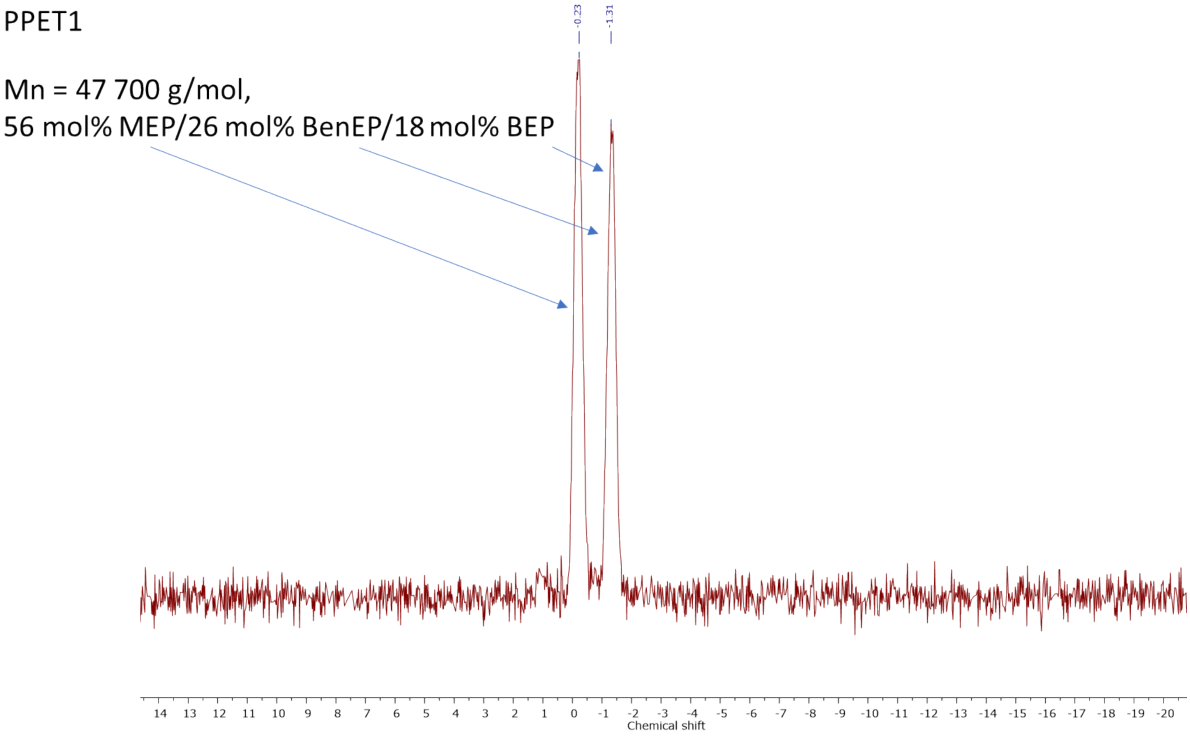
60 mol% MEP / 40 mol% BenEP



PPE3

Mn 23 400 g/mol,

74 mol% MEP / 26 mol% BenEP



PPET1

Mn = 47 700 g/mol,
56 mol% MEP/26 mol% BenEP/18 mol% BEP

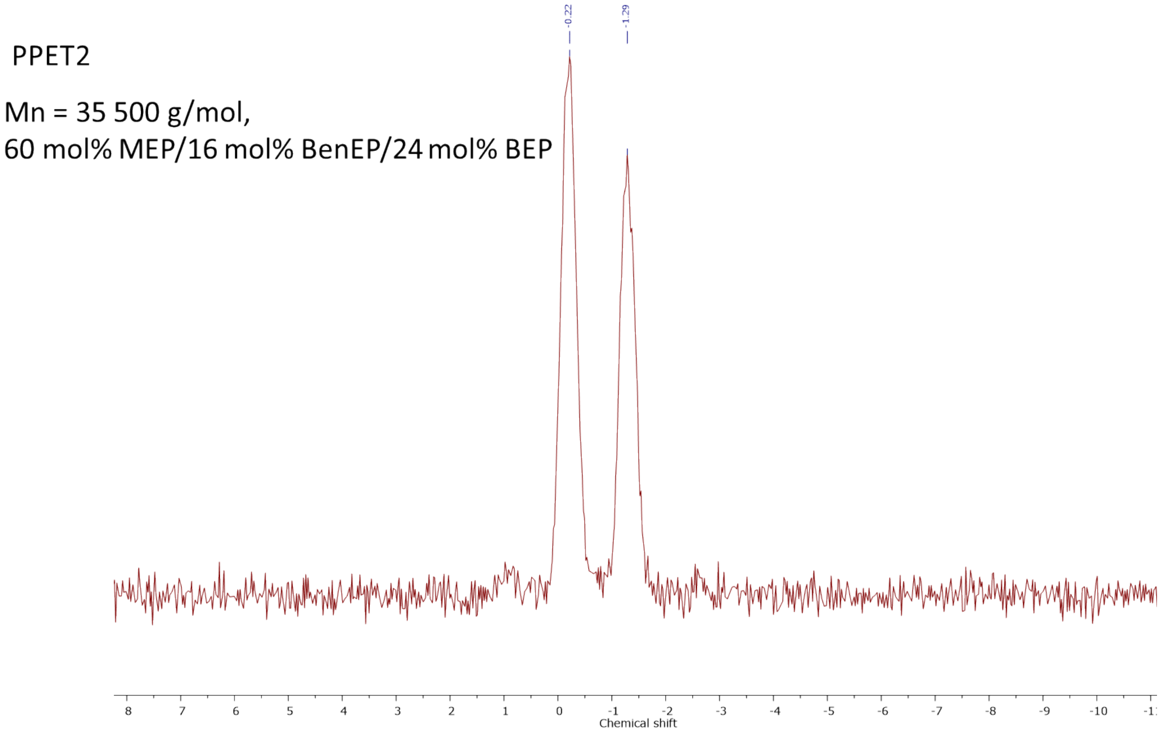
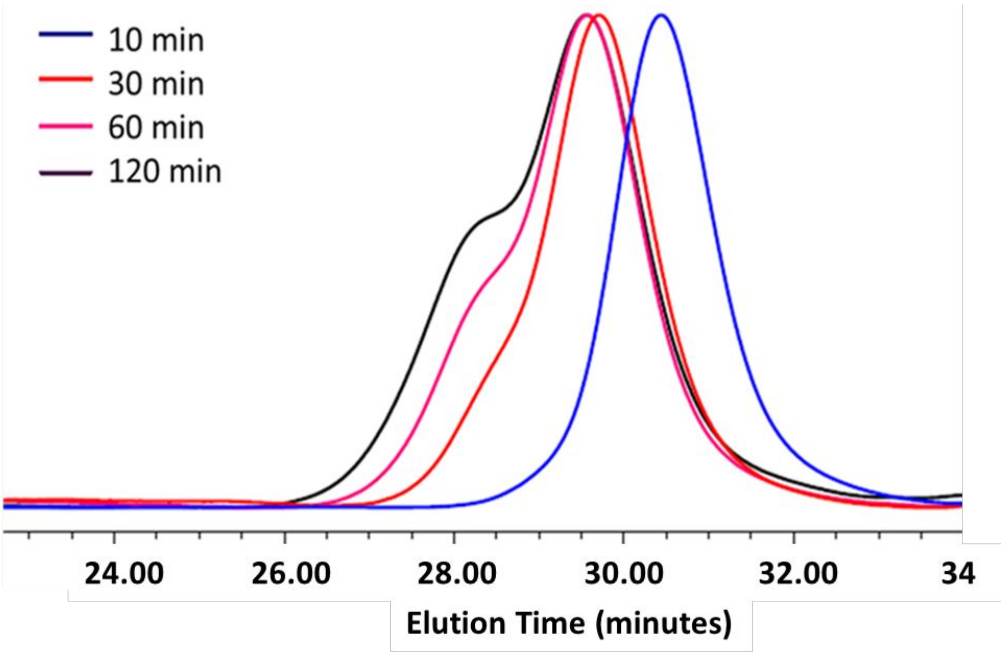
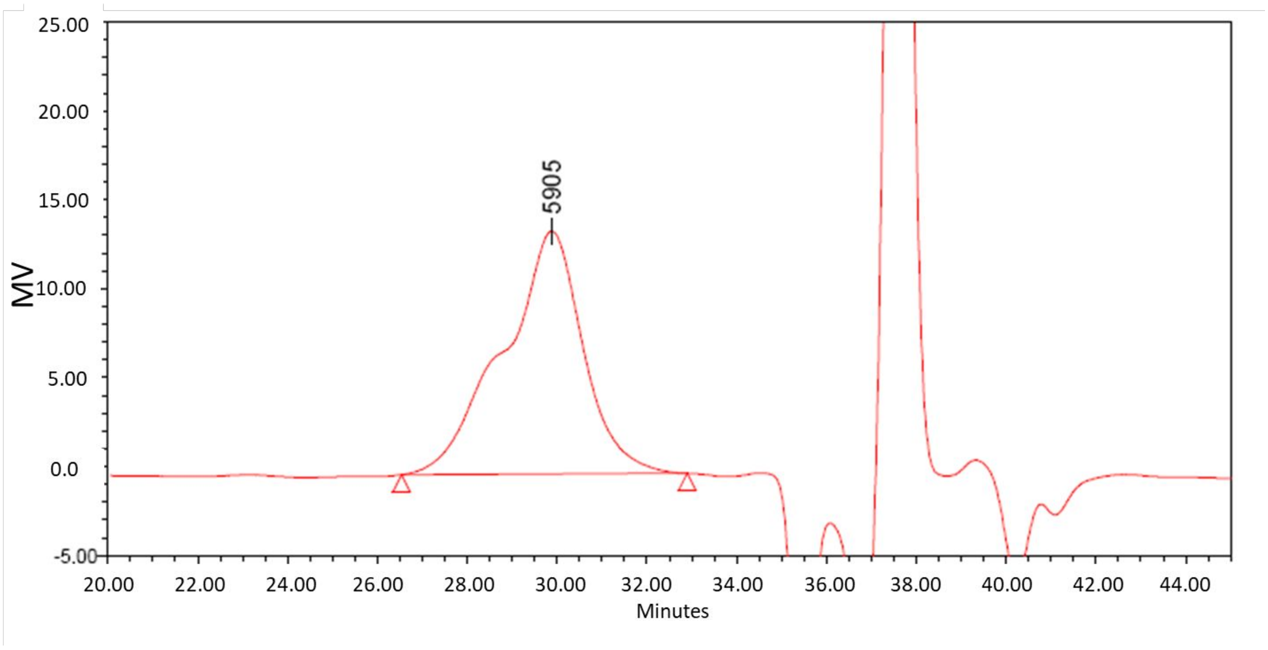


Figure S2: ^{31}P-NMR spectra of the copolymers

a) PPET1 polymerization kinetics



b) PPE1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-5.00
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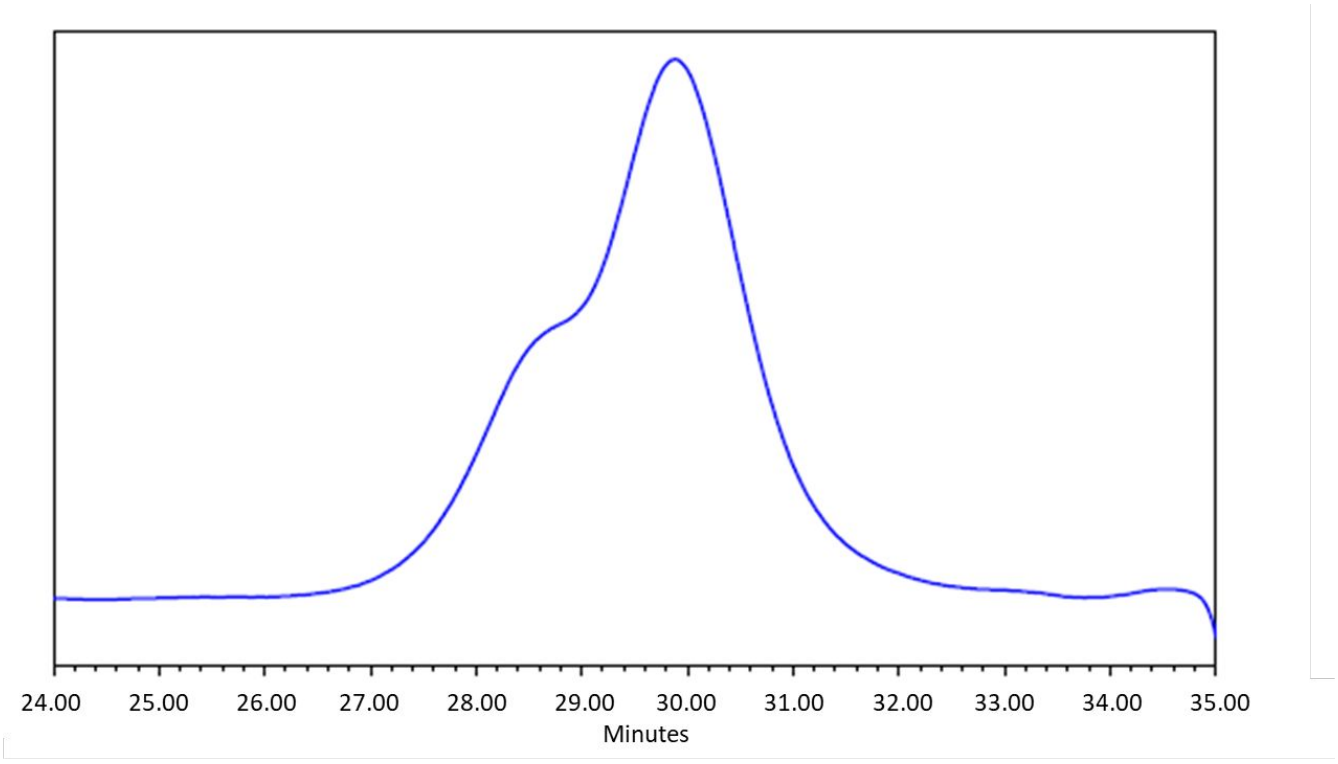
22.00
24.00
26.00
28.00
30.00
32.00
34.00
36.00
38.00
40.00
42.00
44.00
Minutes

c) PPE2


24.00
25.00
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29.00
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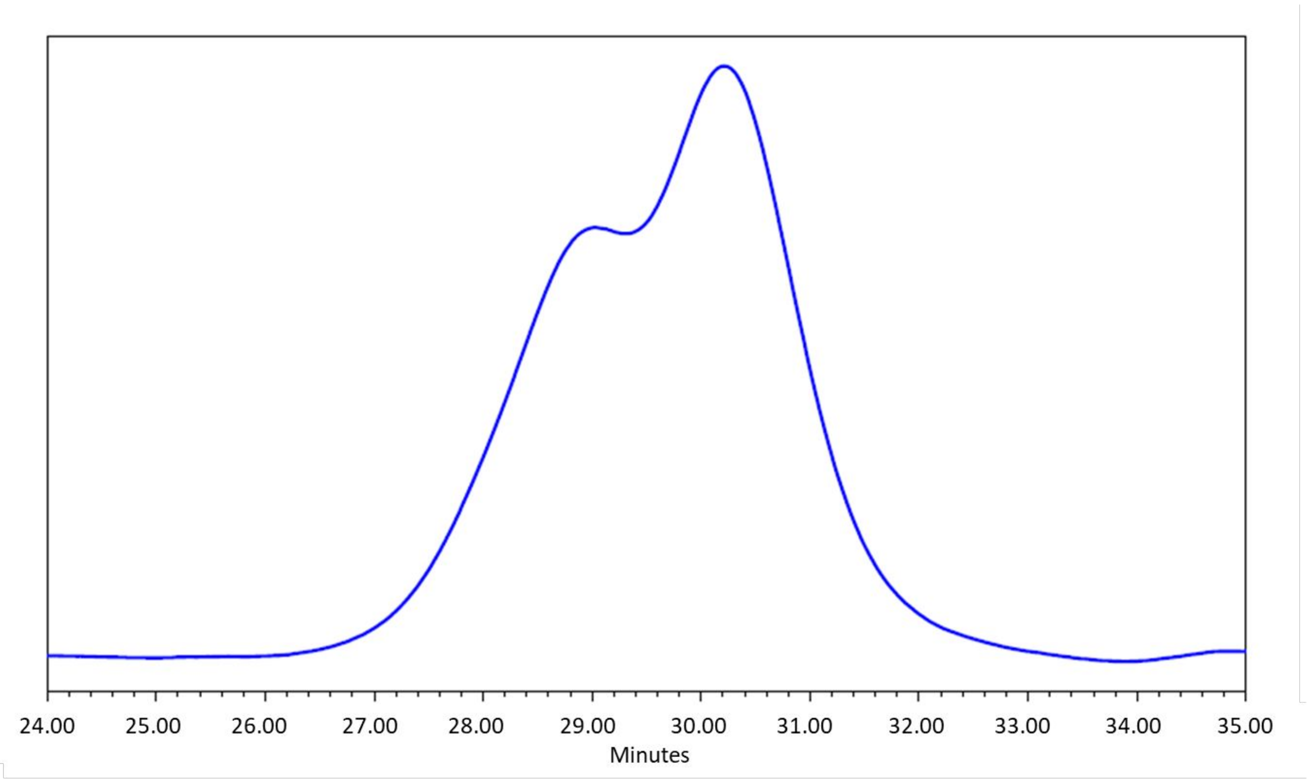
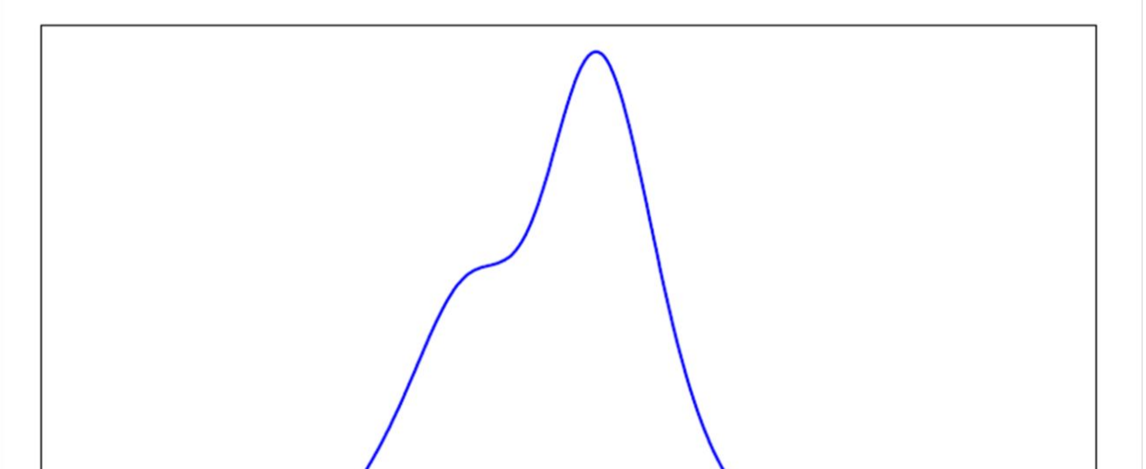
33.00
34.00
35.00
Minutes

d) PPE3



e) PPET1



f) PPET2

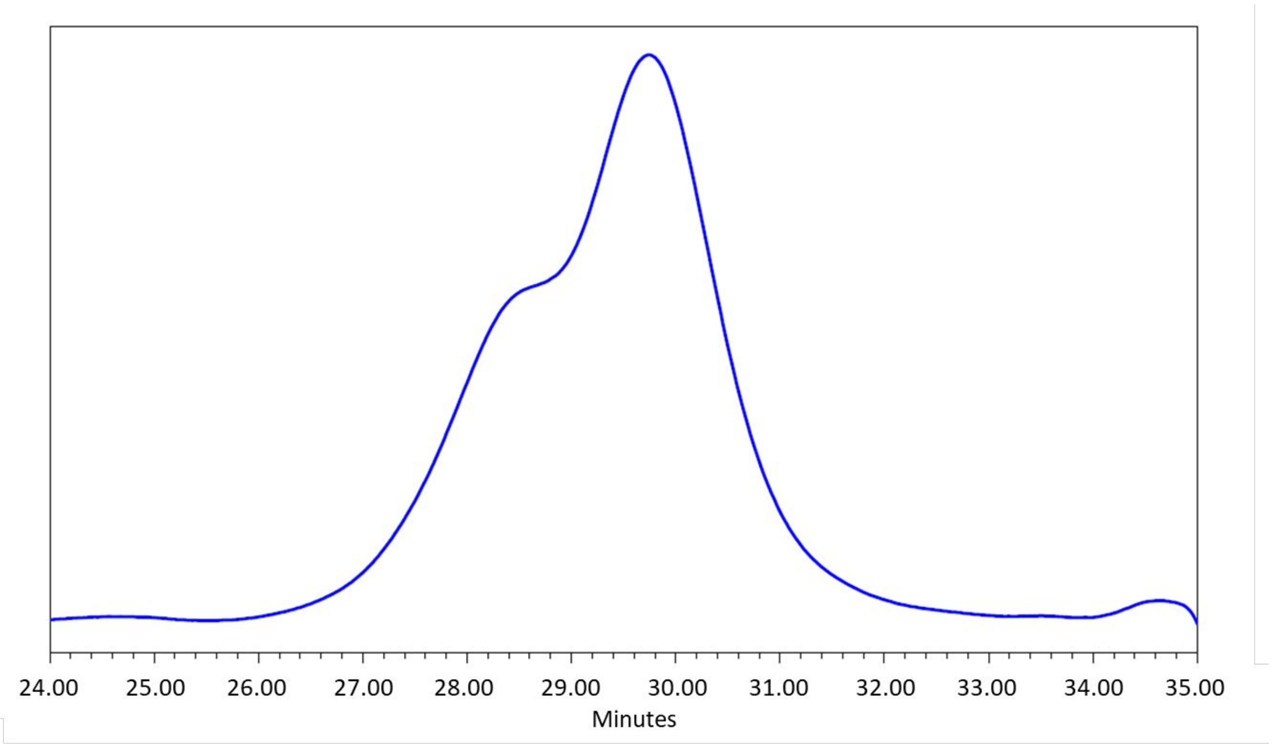


Figure S3: SEC traces a) following the polymerization kinetics for the PPET1 terpolymer synthesis and b-f) of the copolymers after 2h of polymerization

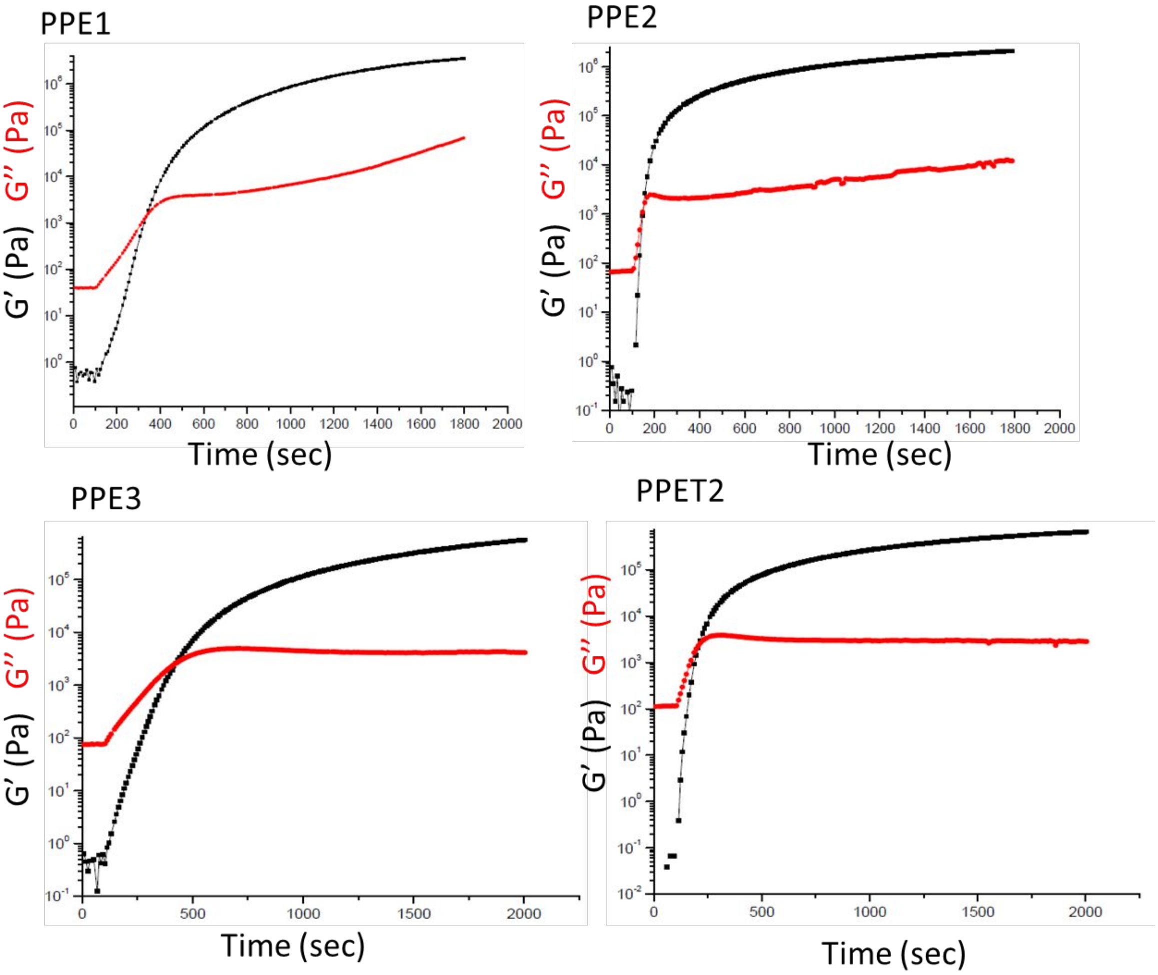


Figure S4: evolution of loss and elastic moduli vs. UV irradiation time as measured in the rheometer

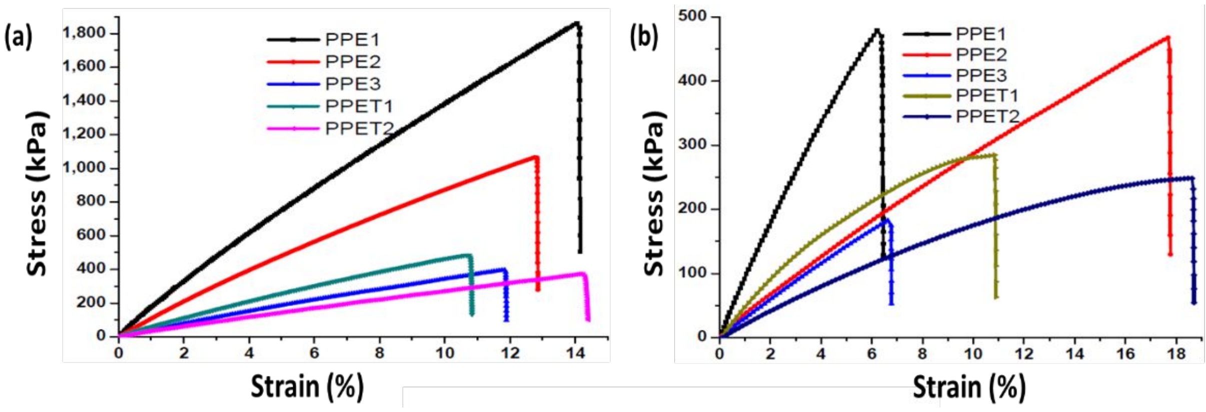


Figure S5: Stress vs strain curves of the networks in a) dry state and b) swollen state