

Supporting information

Ecotoxicity of isosorbide acrylate and methacrylate monomers and corresponding polymers

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Synthesis of monomers and polymers

Isosorbide 5-methacrylate (**IM**), isosorbide 5-methacrylate-2-acetate (**IMA**) and the corresponding poly(isosorbide 5-methacrylate) (**PIM**) and poly(isosorbide 5-methacrylate-2-acetate) (**PIMA**) were prepared according to the procedure reported by Matt et al.¹ Isosorbide 5-methacrylate propionate (**IMP**) and the corresponding butyrate (**IMB**) were synthesized according to Laanesoo and co-workers.² Isosorbide 5-acrylate (**IA**), isosorbide 5-acrylate-2-acetate (**IAA**) and the corresponding poly(isosorbide 5-acrylate) (**PIA**) and poly(isosorbide 5-acrylate-2-acetate) (**PIAA**) were prepared according to the procedures reported by Nonque et al.³ and Gallagher et al.,⁴ respectively.

The polymers were purified from unreacted monomers and initiator by precipitation. **PIM** and **PIMA** were isolated by precipitation in cold MeOH, while **PIA** was precipitated in a mixture of Et₂O/*i*-PrOH (4:1), and **PIAA** in Et₂O. The solids were filtrated using glass-filters and washed 1-2 times with small volumes of the same solvent to remove traces of the monomer. The polymers in the form of white powders polymers were carefully collected from the glass filters and dried under reduced pressure.

The solubility of the monomers and the polymers was investigated in H₂O. Small samples (about 5 mg) of the compounds were mixed with H₂O (1 mL). The mixture was stirred for ca 1 h at room temperature. If the samples were found to be completely dissolved, they were considered as soluble; if not, they were considered as insoluble.

Monomers and polymers were characterized by NMR (¹H and ¹³C). Based on ¹H NMR, the purity of the monomers was assessed to be $\geq 98\%$. Molecular weight of polymers was determined by SEC.

NMR spectra of monomers and polymers

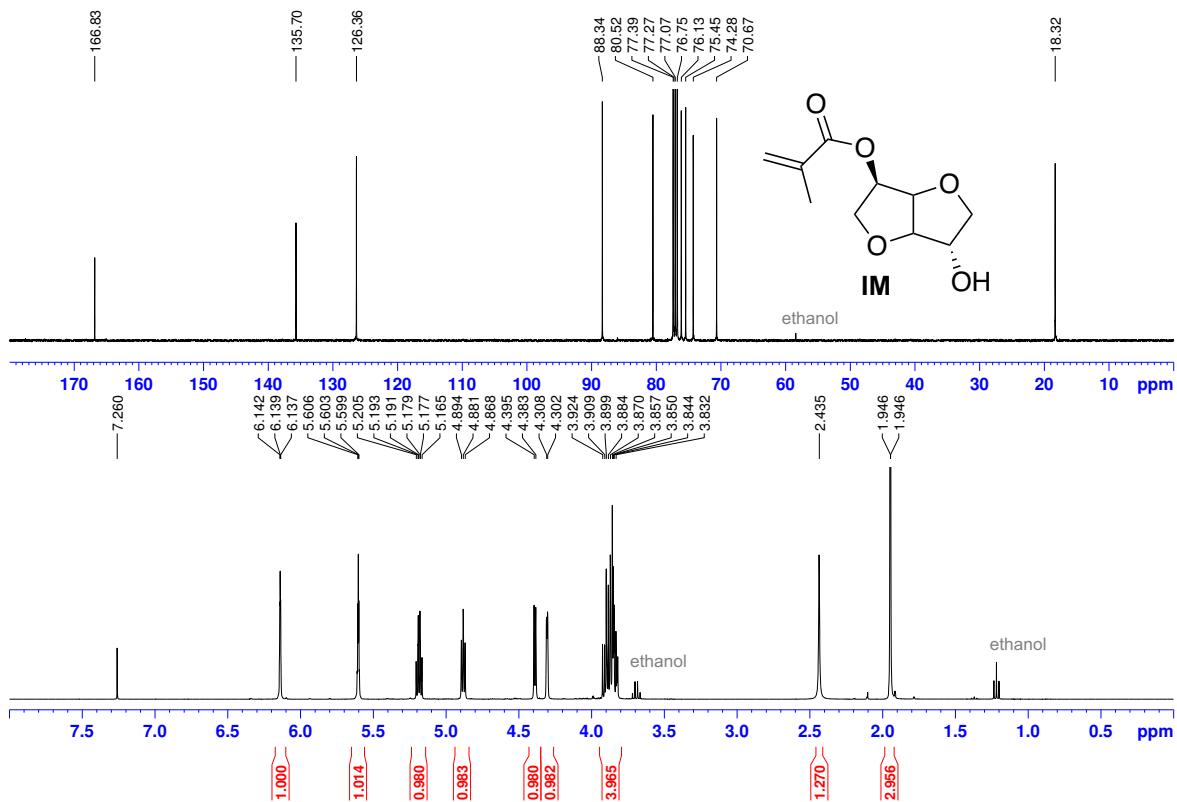


Figure S1. ^{13}C and ^1H NMR spectra of IM in CDCl_3 .

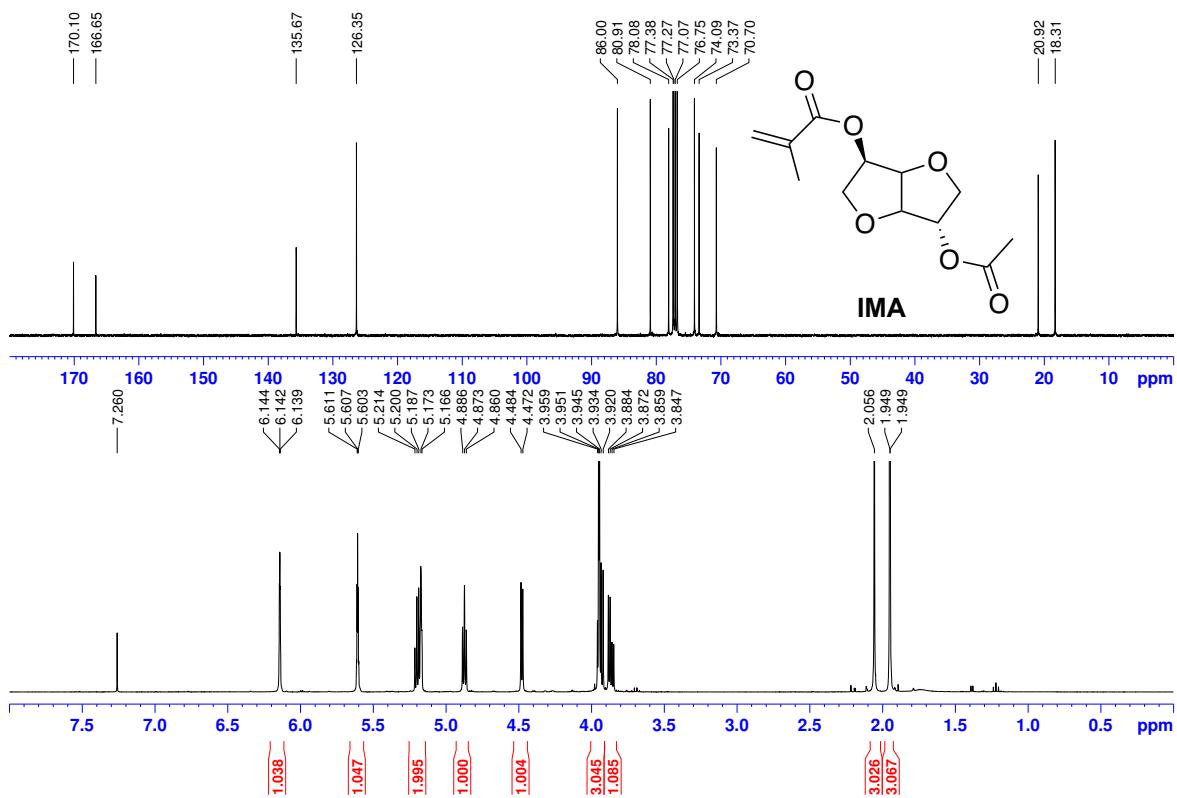


Figure S2. ^{13}C and ^1H NMR spectra of IMA in CDCl_3 .

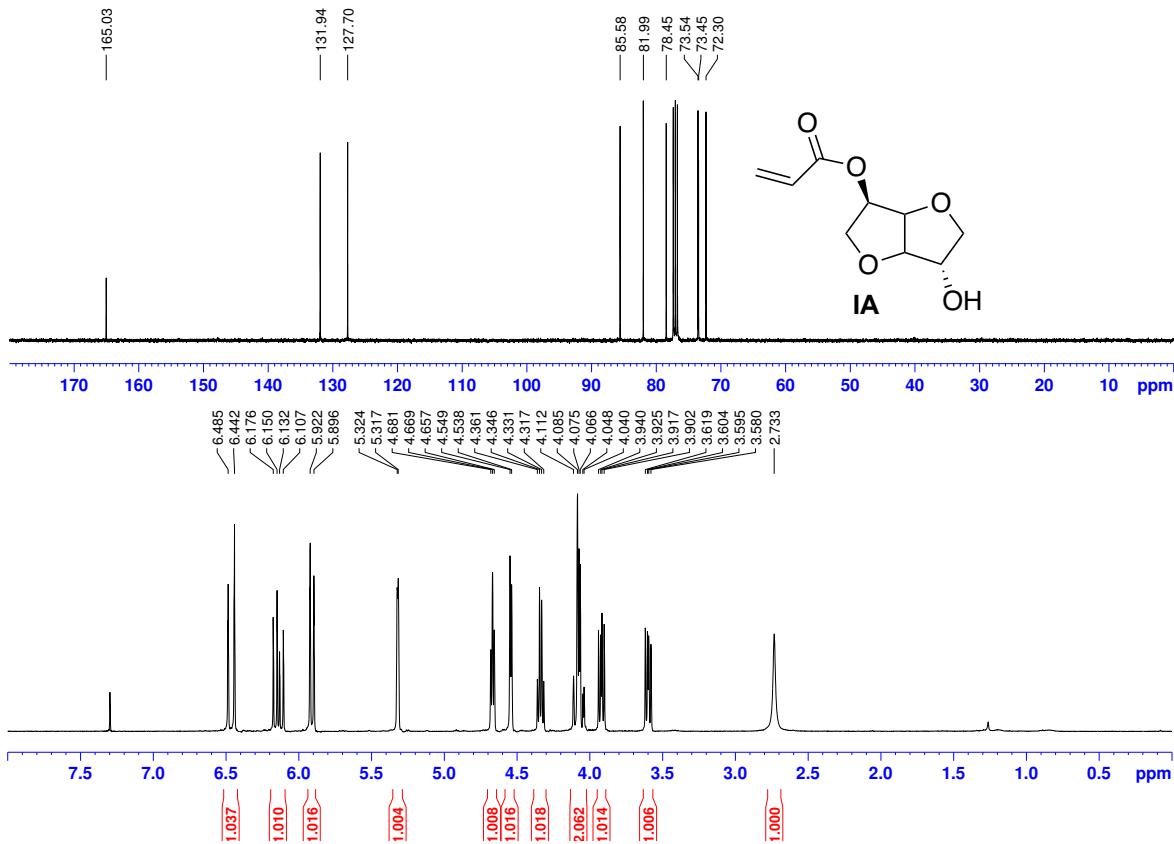


Figure S3. ^{13}C and ^1H NMR spectra of **IA** in CDCl_3 .

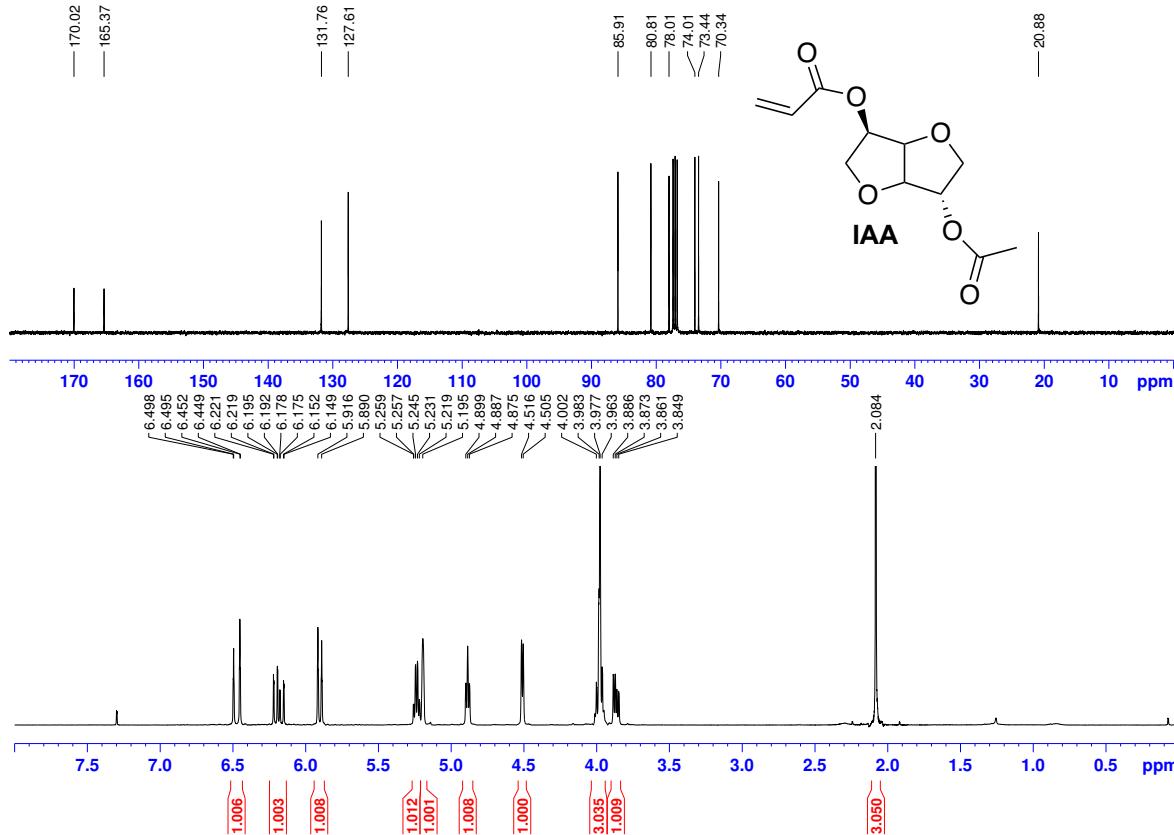


Figure S4. ^{13}C and ^1H NMR spectra of **IAA** in CDCl_3 .

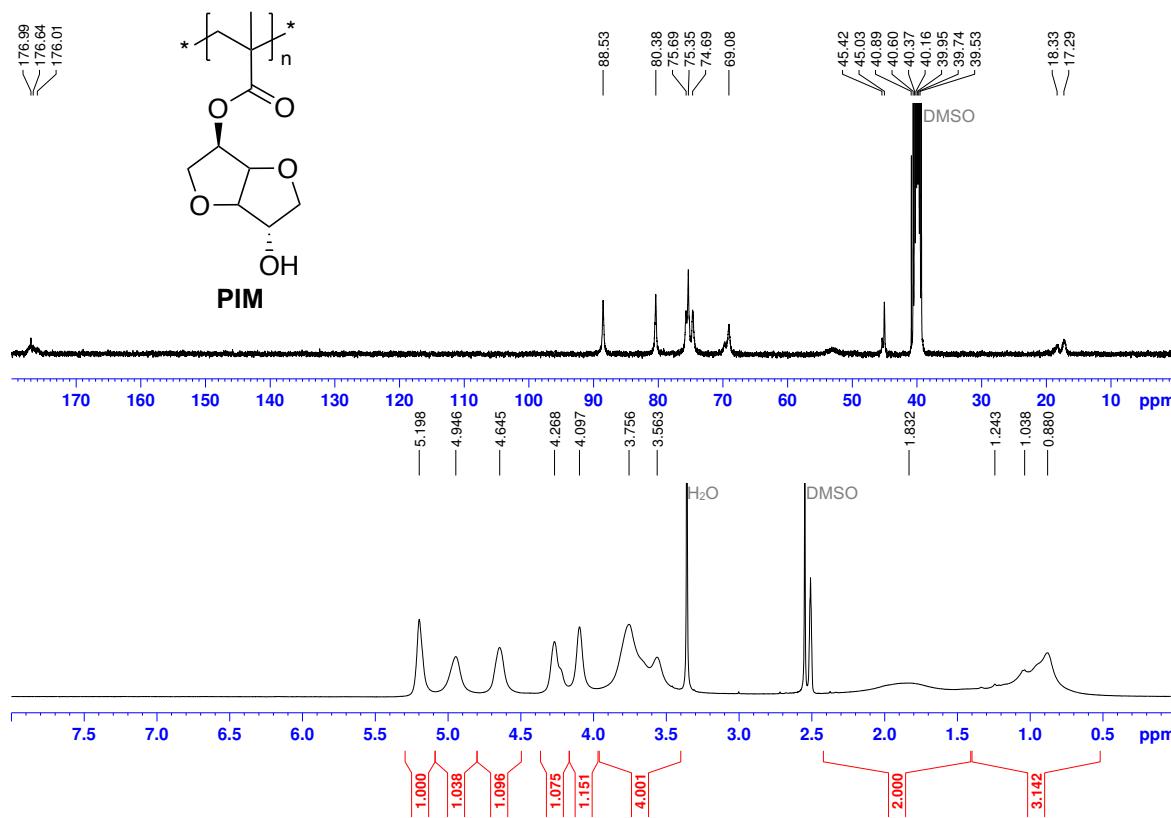


Figure S5. ^{13}C and ^1H NMR spectra of PIM in $\text{DMSO}-d_6$.

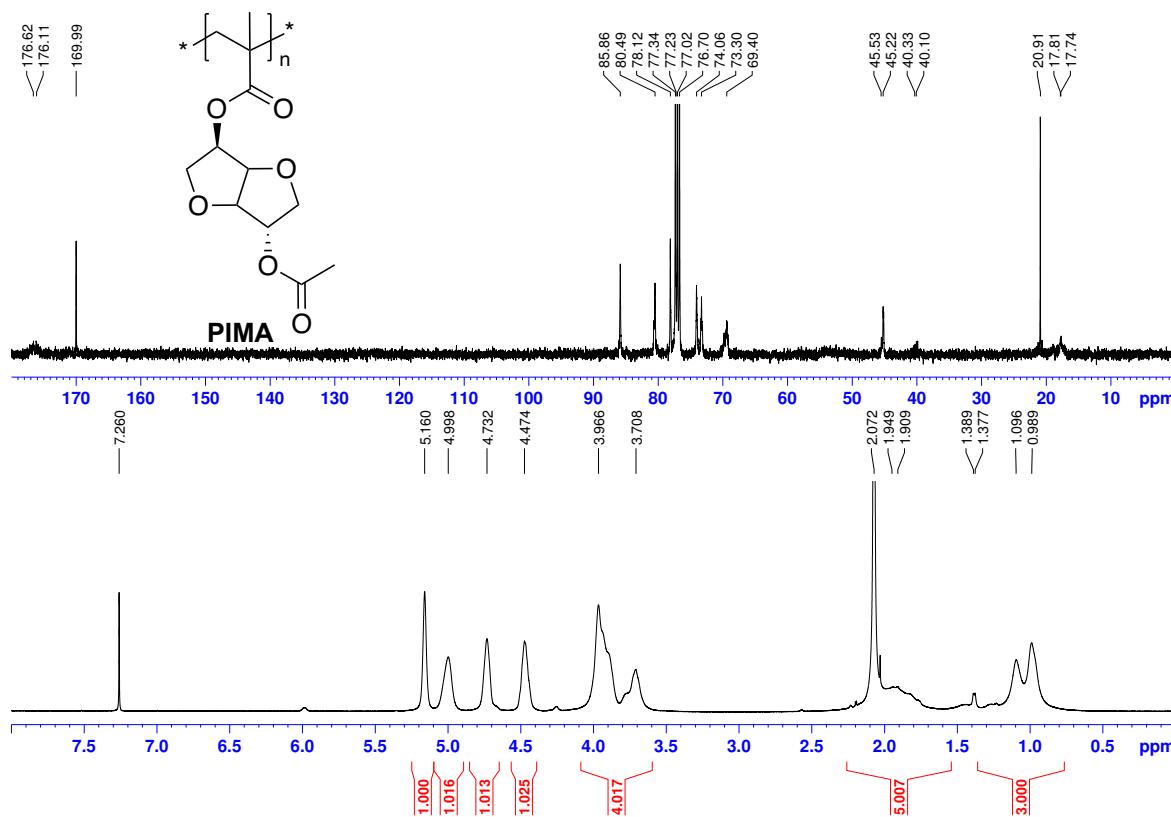


Figure S6. ^{13}C and ^1H NMR spectra of PIMA in CDCl_3 .

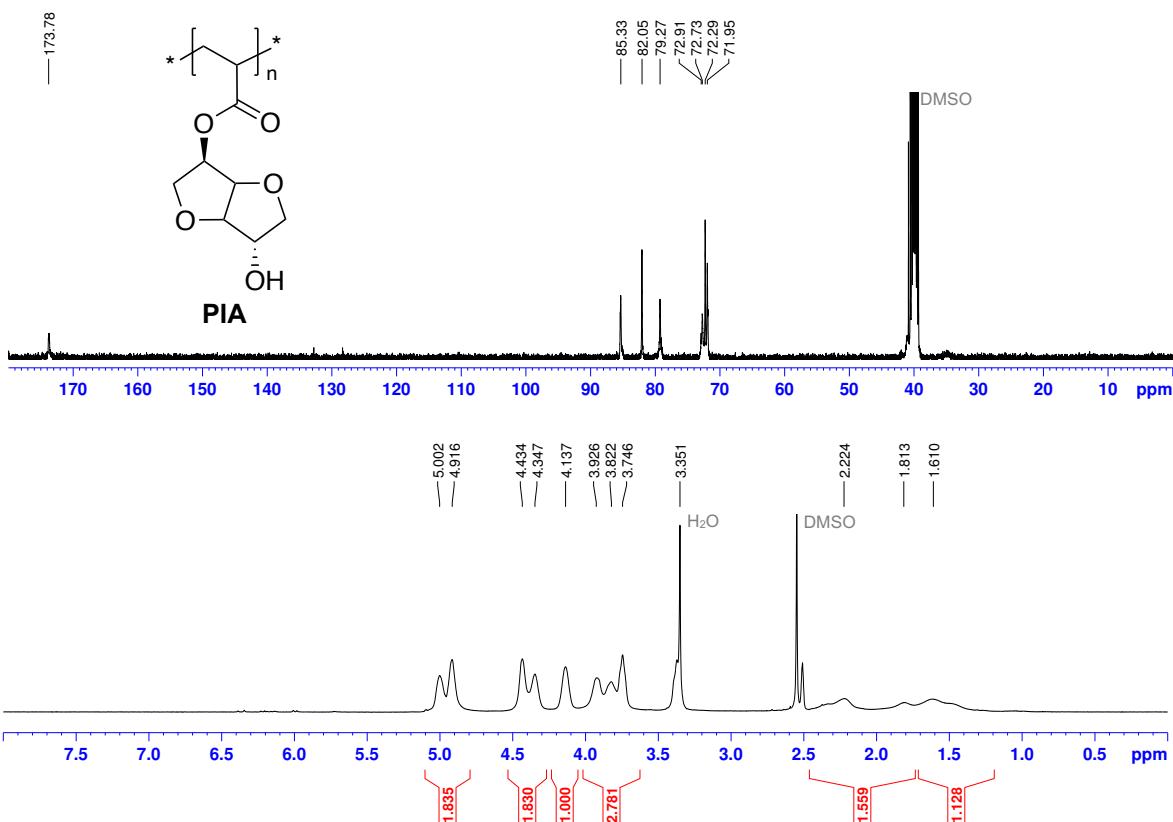


Figure S7. ¹³C and ¹H NMR spectra of PIA in DMSO-*d*₆.

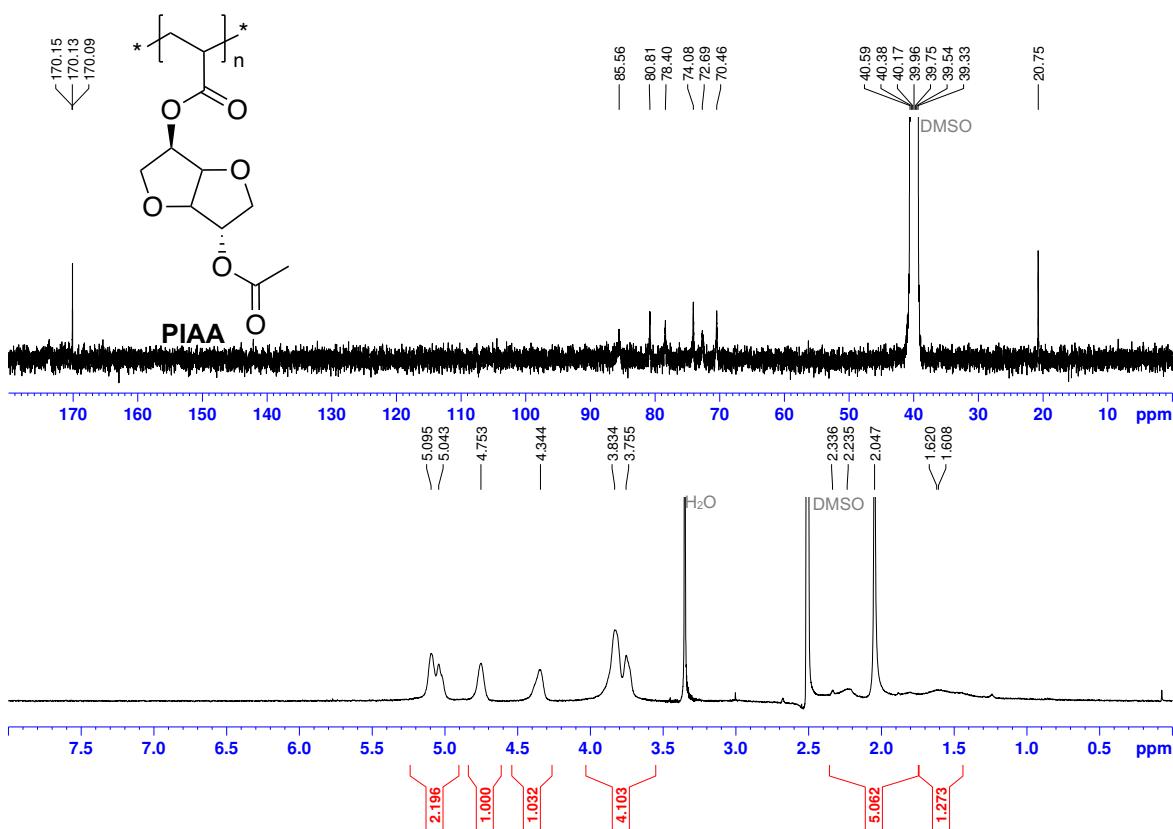


Figure S8. ¹³C and ¹H NMR spectra of PIAA in DMSO-*d*₆.

SEC curves of polymers

The molecular weights of the polymethacrylates and polyacrylates were determined by size-exclusion chromatography (SEC) in THF or in DMF. The SEC setup included three Shodex columns coupled in series (KF-805, -804, and -802.5) for THF or two Shodex columns coupled in series (KD-804, -802.5) for DMF situated in a Shimadzu CTO-20A prominence column oven, a Shimadzu RID-20A refractive index detector, with Shimadzu LabSolution software. Samples were run at 40 °C in THF and at an elution rate of 1 mL min⁻¹, and at 50 °C in DMF and at an elution rate of 0.5 mL min⁻¹. Calibration was done by using poly(ethylene oxide) standards ($M_n = 3\ 860, 21\ 160, 49\ 390$, and $96\ 100\ g\ mol^{-1}$) for both solvents.

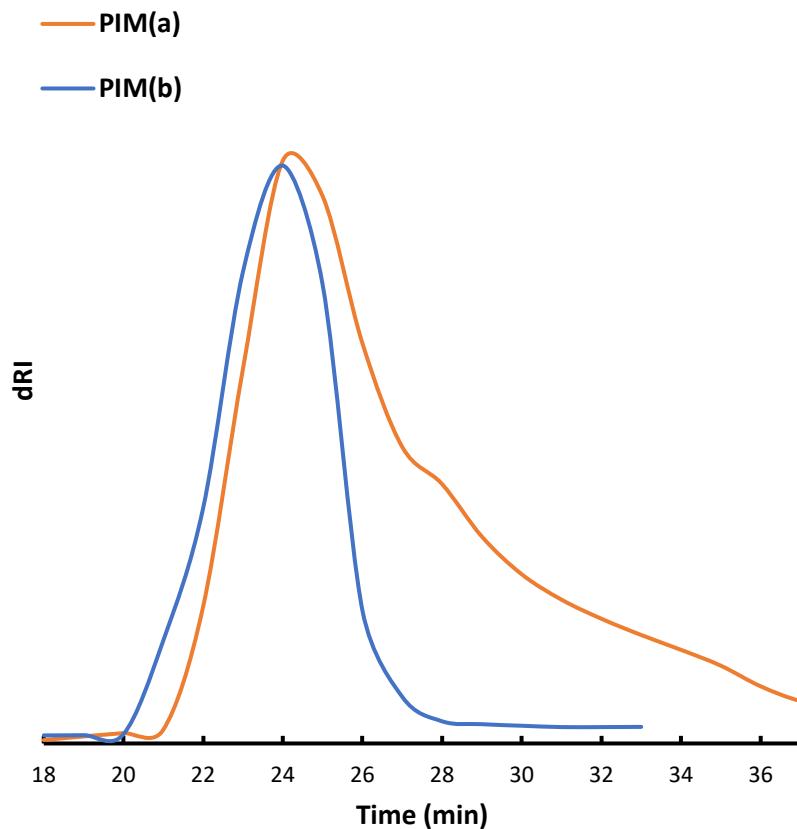


Figure S9. SEC curves in DMF with differential refractive index (dRI) detector of polymers **PIM(a,b)**.

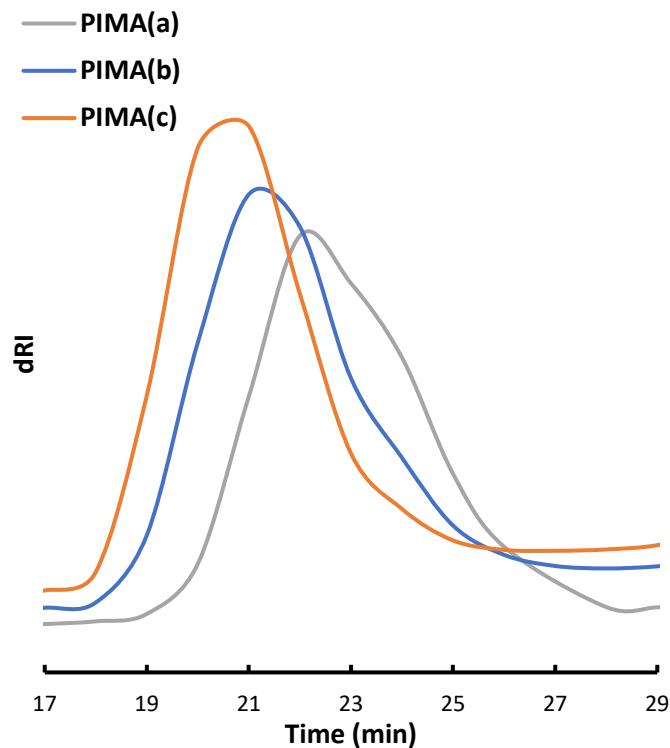


Figure S10. SEC curves in THF with differential refractive index (dRI) detector of polymers PIMA(a-c).

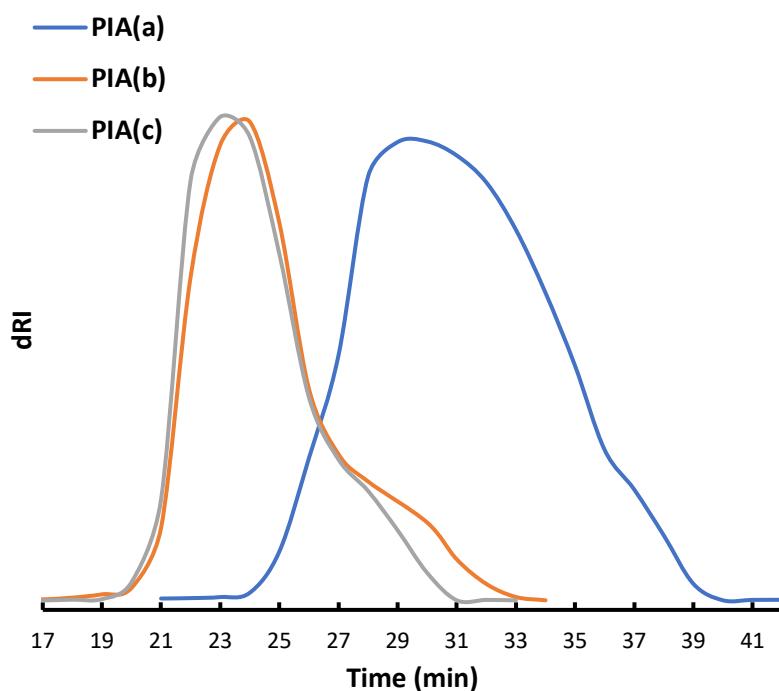


Figure S11. SEC curves in DMF with differential refractive index (dRI) detector of polymers PIA(a-c).

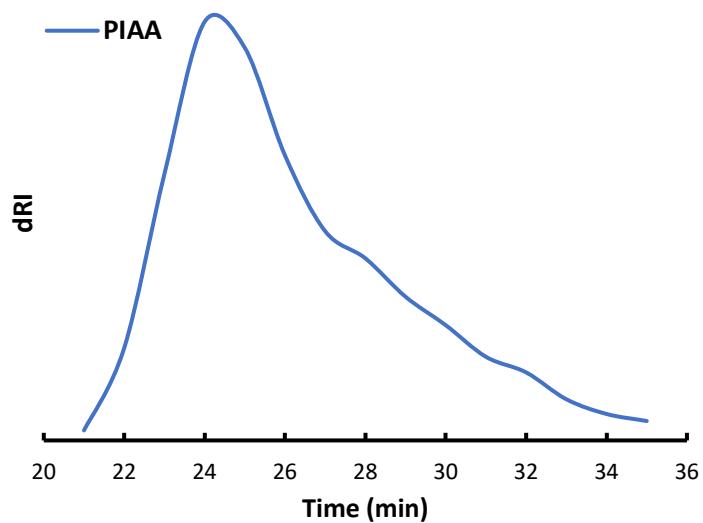


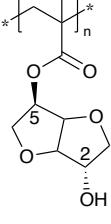
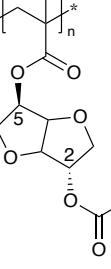
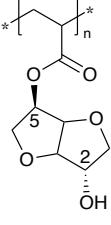
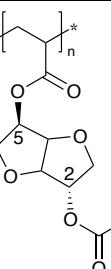
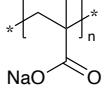
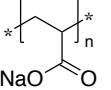
Figure S12. SEC curve in DMF with differential refractive index (dRI) detector of polymer **PIAA**.

Solubility of the test compounds in the mixture of water and DMSO

First, we attempted to dissolve all compounds in water without the addition of DMSO. Monomers **IM** and **IA** with hydroxyl groups in the position 2 in isosorbide structure easily dissolved in water, as well as polymer **PIA** obtained from monomer **IA**. For all other compounds, the mixture of DMSO/water was necessary for full solubility, and we used the lowest possible DMSO content. The solubility of the monomers and the polymers was investigated in H₂O. Small samples (about 5 mg) of the compounds were mixed with H₂O (1 mL). The mixture was stirred for ca 1 h at room temperature. If the samples were found to be completely dissolved, they were considered as soluble; if not, they were considered as insoluble.

Table S1. Name, chemical structure, abbreviation, and solubility in water/DMSO mixture of the tested compounds.

Name	Structure	Abbreviation	DMSO, %	Water, %	Highest tested concentration, mg mL ⁻¹
Isosorbide 5-methacrylate		IM	-	100	25
Isosorbide 5-methacrylate-2-acetate		IMA	15	85	40
Isosorbide 5-acrylate		IA	-	100	10
Isosorbide 5-acrylate-2-acetate		IAA	15	85	4

Poly(isosorbide 5-methacrylate)		PIM	30	70	2.5
Poly(isosorbide 5-methacrylate-2-acetate)		PIMA	15	85	5
Poly(isosorbide 5-acrylate)		PIA	-	100	20
Poly(isosorbide 5-acrylate-2-acetate)		PIAA	15	85	2
Poly(methacrylic acid, sodium salt) solution		PMMA	-	100	132.5
Poly(acrylic acid, sodium salt) solution		PAA	-	100	195

DMSO ecotoxicity tests

The potential effect of DMSO on tested organisms was estimated.

Table S2. Ecotoxicology results of DMSO on bacterial tests with *E. coli* and *A. fischeri*.

Test	Dose, mg L ⁻¹	Response, %	Mean response, %	Standard deviation
<i>E. coli</i>	$3.3 \cdot 10^5$	58.95	56.2	2.39
	$3.3 \cdot 10^5$	54.62		
	$3.3 \cdot 10^5$	55.04		
	$1.65 \cdot 10^5$	49.61	46.47	3.72
	$1.65 \cdot 10^5$	47.45		
	$1.65 \cdot 10^5$	42.35		
	$8.25 \cdot 10^4$	20.91	23.6	3.36
	$8.25 \cdot 10^4$	27.36		
	$8.25 \cdot 10^4$	22.52		
<i>A. fischeri</i>	$3.3 \cdot 10^5$	63.5	61.27	2.01
	$3.3 \cdot 10^5$	59.6		
	$3.3 \cdot 10^5$	60.7		
	$2.2 \cdot 10^5$	45.5	45.2	2.4
	$2.2 \cdot 10^5$	42.7		
	$2.2 \cdot 10^5$	47.4		
	$1.65 \cdot 10^5$	32.8	34.9	2.6
	$1.65 \cdot 10^5$	34.2		
	$1.65 \cdot 10^5$	37.8		
	$1.1 \cdot 10^5$	12.9	12.93	3.3
	$1.1 \cdot 10^5$	16.2		
	$1.1 \cdot 10^5$	9.69		

Prediction of EC₅₀ for monomers.

Modelling was done using Ecological Structure Activity Relationships (ECOSAR) predictive model (Tracy Wright, U.S. EPA Existing Chemicals Risk Assessment Division); a software for estimating a chemical's acute (short-term) toxicity and chronic (long-term or delayed) toxicity to aquatic organisms.

Table S3. ECOSAR toxicity prediction for monomers.

Name	Organism	Duration	End Point	Concentration, mg L ⁻¹
IM	Fish	96 h	LC ₅₀	1960
	Daphnid	48 h	LC ₅₀	2200
	Green Algae	96 h	EC ₅₀	1920
IMA	Fish	96 h	LC ₅₀	575
	Daphnid	48 h	LC ₅₀	1570
	Green Algae	96 h	EC ₅₀	1160
IA	Fish	96 h	LC ₅₀	13.5
	Daphnid	48 h	LC ₅₀	25.8
	Green Algae	96 h	EC ₅₀	24.2
IAA	Fish	96 h	LC ₅₀	15.0
	Daphnid	48 h	LC ₅₀	28.7
	Green Algae	96 h	EC ₅₀	25.5

Values of the mean effective concentration (EC₅₀, mg L⁻¹) of the tested monomers and polymers with 95% CI values

Confidence interval (CI) is the mean of the estimate plus and minus the variation of the estimate. 95% CI gives 95% certainty that the expected values of the experiment will be within this interval. The 95% CI is calculated by a standard formula:

$$\bar{x} = \pm 1.96 \frac{s}{\sqrt{n}}$$

Where: \bar{x} – mean value

s – standard deviation

n – sample size

The EC₅₀ values were determined at 50% response level on the X-axis of the dose-response curves.

Table S4. EC₅₀ values of the tested monomers and polymers with 95% CI values.

Code name	Toxi-Chromo Test TM <i>E. coli</i>	WaterTOX TM <i>A. fischeri</i>	DuckWeed Toxkit <i>S. polystachya</i>	Thamnotoxkit <i>T. platyurus</i>
	EC ₅₀ , mg L ⁻¹ (95% CI)			
IM	2850 (1093; 4606)	1938 (1799; 2075)	>1000	>1000
IMA	6100 (4431; 7768)	2721 (2072; 3371)	139.3 (115.8; 162.9)	>1000
IA	16 (8.8; 24)	456 (418; 511)	9 (5.8; 12.2)	8.7 (6.5; 11.1)
IAA	125 (114; 158)	585 (420; 749)	9.1 (6.8; 11.3)	15.6 (8.46; 22.88)
PIM	374 (280; 468)	>2000	>1000	>1000
PIMA	1081 (800; 1364)	14533 (12540; 16525)	>1000	>1000
PIA	12100 (5757; 18443)	12533 (8357; 16709)	>1000	>1000
PIAA	514 (433; 595)	>4000	>1000	>1000
PMMA	35650 (18611; 52688)	33667 (20919; 46414)	-	-
PAA	18675 (7872; 29477)	45633 (40462; 50804)	-	-

Statistical analysis by one-way ANOVA

Table S5. One-way ANOVA and post hoc pairwise comparison (Tukey test) between EC₅₀ values of studied monomers and polymers. *p* values are adjusted for multiple testing.

	<i>p</i> values										
	IA	IAA	IM	IMA	PAA	PIA	PIAA	PIM	PIMA	PMAA	
IA	NA	NA									
IAA	<i>p</i> > 0.9999	NA	NA								
IM	<i>p</i> = 0.1732	<i>p</i> = 0.2145	NA	NA							
IMA	<i>p</i> = 0.0035	<i>p</i> = 0.0049	<i>p</i> = 0.9267	NA	NA	NA	NA	NA	NA	NA	
PAA	<i>p</i> < 0.0001	<i>p</i> < 0.0001	<i>p</i> < 0.0001	<i>p</i> < 0.0001	NA	NA	NA	NA	NA	NA	
PIA	<i>p</i> < 0.0001	NA	NA	NA	NA	NA					
PIAA	<i>p</i> = 0.2293	<i>p</i> = 0.2792	<i>p</i> > 0.9999	<i>p</i> = 0.8775	<i>p</i> < 0.0001	<i>p</i> < 0.0001	NA	NA	NA	NA	
PIM	<i>p</i> = 0.7941	<i>p</i> = 0.8471	<i>p</i> = 0.9873	<i>p</i> = 0.3156	<i>p</i> < 0.0001	<i>p</i> < 0.0001	<i>p</i> = 0.9953	NA	NA	NA	
PIMA	<i>p</i> < 0.0001	<i>p</i> < 0.0001	<i>p</i> < 0.0001	<i>p</i> = 0.0396	<i>p</i> < 0.0001	<i>p</i> < 0.0001	<i>p</i> < 0.0001	<i>p</i> < 0.0001	NA	NA	
PMAA	<i>p</i> < 0.0001	<i>p</i> < 0.0001	<i>p</i> < 0.0001	<i>p</i> < 0.0001	<i>p</i> > 0.9999	<i>p</i> < 0.0001	<i>p</i> < 0.0001	<i>p</i> < 0.0001	<i>p</i> < 0.0001	NA	

Toxi-Chromo Test™: chromo inhibition test with bacteria *Escherichia coli*

For the instrumental analysis of the results, the absorption at 600 nm was measured on BioTek Synergy™ Mx monochromator-based multi-mode microplate reader.

The results were evaluated according to the standard protocol:

$$TF = 100 - 100[(A_{600S} - A_{600BL_{avg}})/(A_{600NC_{avg}} - A_{600BL_{avg}})]$$

Where: TF – toxicity factor of tested sample

A_{600S} – the value of absorption at 600 nm of test sample

$A_{600BL_{avg}}$ – the average value of absorption at 600 nm for the blank samples

$A_{600NC_{avg}}$ – the average value of absorption at 600 nm for the negative control samples

Table S6. Ecotoxicology results of the test compounds on bacteria *E. coli*.

Code name	Dose, mg L ⁻¹	Response, %	Mean response, %	Standard deviation
IM	12500	71.38	77.27	4.69
	12500	68.01		
	12500	77.27		
	6250	67.34	65.01	2.18
	6250	64.65		
	6250	63.03		
	3125	49.16	48.16	7.26
	3125	47.16		
	3125	60.61		
	1563	37.61	41.31	5.21
	1563	39.06		
	1563	47.27		
IMA	781	18.18	21.57	3.33
	781	22.90		
	781	23.64		
	40000	88.77	84.21	2.28
	40000	86.19		
	40000	84.21		
	20000	76.38	78.19	4.78
	20000	69.16		
	20000	78.19		
	10000	60.38	63.68	2.06
	10000	59.87		
	10000	63.68		
	5000	49.55	45.64	3.13
	5000	43.35		
	5000	45.64		
	2500	41.81	39.85	2.27
	2500	44.39		
	2500	39.85		

IAA	1000	99.18	98.37	0.82
	1000	97.55		
	1000	98.37		
	500	95.91	94.69	1.22
	500	94.69		
	500	93.47		
	250	80.82	77.28	4.19
	250	72.65		
	250	78.37		
	125	56.32	50.88	4.73
	125	47.75		
	125	48.57		
	62.5	33.06	31.29	4.93
	62.5	35.10		
	62.5	25.71		
IA	78	95.68	94.56	2.22
	78	92		
	78	96		
	39	77.07	73.28	3.87
	39	69.33		
	39	73.33		
	19.5	60.47	58.38	7.55
	19.5	50		
	19.5	64.67		
	9.75	27.91	28	6.05
	9.75	23.33		
	9.75	35.33		
	4.8	21.93	21.53	3.35
	4.8	18		
	4.8	24.67		
PIM(a)	1000	82.65	84.85	3.11
	1000	87.05		
	500	56.77	57.04	0.38
	500	57.31		
	250	49.33	46.44	4.09
	250	43.54		
	125	18.76	17.8	1.36
	125	16.83		
	62.5	0.86	1.41	0.78
	62.5	1.97		
PIM(b)	2500	122.01	124.9	4.97
	2500	122.03		
	2500	130.62		
	1250	74.64	77	2.19
	1250	78.94		
	1250	77.51		
	625	70.33	65.6	7.08
	625	57.41		
	625	68.89		
	312.5	48.80	46.4	2.98
PIMA(a)	312.5	47.36		
	312.5	43.06		
	3125	98.08	103.33	4.55
	3125	105.74		
	3125	106.18		

PIMA(b)	1563	65.9	68.47	7.21
	1563	76.63		
	1563	62.89		
	781	30.65	37.87	7.83
	781	36.78		
	781	46.02		
	391	24.52	20.42	3.86
	391	19.92		
	391	16.83		
PIMA(c)	5000	80.35	83.68	4.71
	5000	87.01		
	2500	58.75	57.91	1.19
	2500	57.06		
	1250	59.65	51.83	1.66
	1250	53.01		
	625	22.71	21.76	1.34
	625	20.81		
	312.5	18.73	17.71	1.46
	312.5	16.67		
PIAA	5000	87.02	86.34	0.96
	5000	85.66		
	2500	59.65	63.16	4.96
	2500	66.66		
	1250	29.19	34.03	6.94
	1250	38.94		
	625	28.42	24.73	5.21
	625	21.05		
	312.5	11.22	15.08	5.45
	312.5	18.94		
	1000	64	71.89	14.21
	1000	63.38		
	1000	88.31		
PIA(a)	500	48.61	52.1	4.54
	500	50.46		
	500	57.23		
	250	33.84	34.67	6.81
	250	28.31		
	250	41.85		
	125	25.84	26.05	3.39
	125	22.77		
	125	29.54		
	20000	57.71	61.86	3.99
PIA(b)	20000	65.67		
	20000	62.19		
	10000	41.79	46.27	4.25
	10000	50.25		
	10000	46.76		
	5000	27.36	29.02	4.69
	5000	34.33		
	5000	25.38		
	2500	25.87	27.19	4.62
	2500	32.34		
	2500	23.58		
PIA(b)	20000	62.02	64.93	4.11
	20000	67.83		

	10000	24.98	23.76	1.72
	10000	22.54		
PIA(c)	5000	17.11	16.33	1.08
	5000	15.57		
PMAA	2500	10.96	11.07	0.15
	2500	11.18		
PMAA	20000	55.14	53.88	1.78
	20000	52.62		
	10000	15.53	16.68	1.63
	10000	17.83		
	5000	4.88	4.55	0.46
	5000	4.22		
	2500	1.59	2.11	0.72
	2500	2.61		
	195000	99.51	98.43	0.99
	195000	97.56		
PAA	195000	98.21		
	97500	90.73	85.85	6.9
	97500	80.98		
	97500	89.33		
	48750	75.12	66.34	12.42
	48750	57.56		
	48750	68.18		
	24375	40.98	38.05	4.14
	24375	35.12		
	24375	40.23		
PAA	12187	24.39	21.51	2.84
	12187	18.71		
	12187	21.42		
PAA	132500	90.77	92.04	2.9
	132500	90.52		
	132500	89.74		
	66250	78.48	81.14	4.3
	66250	77.69		
	66250	87.18		
	33125	62	58.84	3.86
	33125	62.26		
	33125	56.41		
PAA	16562	41.33	43.75	2.87
	16562	43.43		
	16562	42.39		
	8281	19.08	22.56	4.44
	8281	29.06		
	8281	20.73		

Water-TOXTM STD: luminescence inhibition test with bacteria *Aliivibrio fischeri*

The luminescence intensity was measured in GloMax® 20/20 Luminometer.

The results were evaluated according to ISO 11348-3 as follows:

- 1) $KF = IC_t/IC_0$
- 2) $INH\% = 100 - 100[IT_t/(KF \times IT_0)]$

Where: KF – Correction factor

IC_t – Luminescence intensity of negative control after contact time

IC_0 – Initial luminescence intensity of control sample

$INH\%$ – Inhibition percentage of luminescence

IT_t – Luminescence intensity of test sample after contact time

IT_0 – Initial luminescence intensity of test sample

Table S7. Ecotoxicology results of the test compounds on bacteria *A. fischeri*.

Code name	Dose, mg L ⁻¹	Response, %	Mean response, %	Standard deviation
IM	5200	78.13	81.21	2.69
	5200	82.35		
	5200	83.15		
	3500	70.93	74.46	4.75
	3500	79.86		
	3500	72.59		
	2100	54.78	53.56	1.19
	2100	53.51		
	2100	52.39		
	1750	45.09	45.52	2.37
	1750	48.08		
	1750	43.41		
IMA	875	35.88	35.71	2.33
	875	37.93		
	875	33.28		
	7500	91.2	89.82	1.24
	7500	89.49		
IMA	7500	88.77		
	3750	59.68	65.37	6.35
	3750	64.21		
	3750	72.24		
	1875	38.01	32.89	8.56
	1875	23.02		
	1875	37.67		

	937.5	17.66	19.14	4.46
	937.5	15.62		
	937.5	24.15		
IA	468.7	8.25	7.01	1.08
	468.7	6.55		
	468.7	6.24		
IAA	1000	95.38	92.77	2.31
	1000	91.96		
	1000	90.97		
	500	78.52	76.29	4.11
	500	71.55		
	500	78.81		
	250	26.21	26.51	3.58
	250	30.22		
	250	23.07		
	125	17.01	14.81	2.08
	125	12.88		
	125	14.52		
PIMA	1000	89.62	92.01	2.87
	1000	95.19		
	1000	91.22		
	600	50.36	46.19	5.21
	600	47.88		
	600	40.34		
	500	41.65	35.5	7.6
	500	37.85		
	500	26.99		
	250	9.93	7.89	1.76
	250	7.51		
	250	6.49		
PIA	20000	91.4	89.3	2.9
	20000	87.3		
	16000	53.6	56.1	3.5
	16000	58.6		
	13000	41.6	45.5	5.5
	13000	49.4		
	8000	9.5	10.3	1.2
	8000	11.2		
	3000	1.5	1.9	0.6
	3000	2.3		

	5000	22.9		
PMAA	2500	14.6	11.1	4.9
	2500	5.5		
	2500	13.1		
	1250	9.2	6.8	2.6
PAA	1250	6.7		
	1250	4.5		
	39000	58.06	53.46	4.1
	39000	50.33		
	39000	51.99		
	19500	45.77	43.74	2.1
	19500	41.64		
	19500	43.81		
	9750	34.36	36.04	1.5
	9750	37.35		
	9750	36.4		
	4875	31.03	30.35	1.8
	4875	31.73		
	4875	28.31		
	2437	26.1	26.57	0.8
	2437	26.19		
	2437	27.44		
IMA-latex	53000	59.6	60.3	1.1
	53000	59.7		
	53000	61.1		
	26500	23.3	25.1	6.1
	26500	20.03		
	26500	31.8		
	13250	18.4	18.3	0.14
	13250	18.1		
	13250	18.3		
	6625	9.3	9.9	1.2
	6625	11.3		
	6625	9.2		
	3312	18.6	18.7	2.9
	3312	15.9		
	3312	21.7		

	62500	23.5		
IMP-latex	31000	17.7	21.9	2.3
	31000	21.6		
	31000	21.9		
	500000	92.6		
IMB-latex	500000	88.4	91.5	2.7
	500000	93.6		
	250000	48.03		
	250000	47.3	50.5	4.8
	250000	56.1		
	125000	43.9		
	125000	34.5	39.7	4.7
	125000	40.7		
	62500	36.5	32.6	3.5
	62500	30.8		
	62500	30.3		
	31000	30.2	30.3	1.6
	31000	31.9		
	31000	28.6		
IMB-latex	500000	49.7	55.9	6.3
	500000	62.3		
	500000	55.6		
	250000	32.5	31.1	1.9
	250000	31.8		
	250000	28.9		
	125000	20.5	22.9	2.5
	125000	25.4		
	125000	23.3		
	62500	15.5	16.6	1.6
	62500	18.4		
	62500	16.1		
	31000	15.5	17.4	1.7
	31000	18.8		
	31000	17.9		

DuckWeed Toxkit F: growth inhibition test with vascular plants *Spirodela polyrhiza*

The 72 h EC₅₀ is the concentration of tested substances that will affect 50% of the vegetative buds in the test population after 3 days of exposure to samples.

Percent inhibition of growth rate (I_r) were calculated for each test concentration according to the following formula:

$$I_r = \frac{\mu C - \mu T}{\mu C} \times 100\%$$

Where: I_r – percent inhibition in average specific growth rate

m – the size of the vegetative buds measured by Image Analysis software

μC – mean value for μ in the control

μT – mean value for μ in the treatment group

Table S8. Ecotoxicology results of the test compounds on vascular plants *S. polyrhiza*.

Code name	Dose, mg L ⁻¹	Response, %	Mean response, %	Standard deviation
IM	1	98.4	101.4	11.8
	1	114.5		
	1	91.4		
	10	102.1		
	10	89.8	87.6	15.7
	10	70.9		
	25	86.8		
	25	74.2		
	50	83.3	93	13.7
	50	102.7		
	100	104.5	90.9	111.8
	100	82.7		
	100	85.8		
IMA	10	82.2	93.7	10
	10	99.3		
	10	99.7		
	50	73.8	72.7	1.5
	50	71.6		
	100	62.2	54.6	6.7
	100	51.8		
	100	49.7		
	500	10.5		
	500	11.6	11	0.8

	1000	7.7	7.2	0.7
	1000	6.7		
IA	1	103.3	102.9	4.1
	1	106.8		
	1	98.5		
	10	56.3		9.5
	10	45.3		
	10	37.3		2.3
	25	21.1		
	25	17.7		1.4
	50	1.8		
	50	4.8		
	50	3.2		
IAA	1	100.7	100.4	6.6
	1	93.7		
	1	106.9		
	10	68.4	52.9	13.4
	10	44.9		
	10	45.3		
	25	22.5	21.8	1.03
	25	21.1		
	50	11.3	11.7	0.7
	50	12.2		
	100	8.2	5.9	2.1
	100	5.4		
	100	4.0		
PIM	1	102.7	102.9	2.1
	1	105.3		
	1	100.9		
	10	83.5	91.9	8.3
	10	100.2		
	10	92.2		
	25	99.5	101.3	12.3
	25	95.7		
	25	108.7		
	100	96.1	113.9	15.7
	100	120		
	100	125.7		
PIMA	1	89.2	92.6	3.4
	1	96.1		
	1	92.7		
	10	86.8	88.7	2.7
	10	90.7		
	25	122.5	120.5	2.8
	25	118.5		
	100	77.6		
	100	108	107	32.2
	100	142.3		

	1000	73.5	73.8	0.4
	1000	74.1		
PIA	0.1	105.9	105.9	0.06
	0.1	106.1		
	1	138.8	133.9	6.9
	1	129.1		
	10	128.3	130.3	2.8
	10	132.3		
	100	125.2	123.7	2.7
	100	121.3		
	1000	126.6	126.6	2.01
	1000	129.5		
PIAA	0.1	96.1	92.1	5.6
	0.1	88.1		
	1	102.2	105.4	3.5
	1	104.4		
	1	109.1		
	10	109.1	106.9	5.9
	10	100.2		
	10	111.3		
	100	89.1	90.1	2.9
	100	87.9		
	100	93.4		
	1000	74.1	73.6	0.6
	1000	73.2		

**Thamnotoxkit F: crustacean toxicity screening test with invertebrates
*Thamnocephalus platyurus***

To estimate the amount of alive and dead crustaceans dissection microscope was used.

Percent mortalities were calculated for each test concentration according to the following formula:

$$\%mortality = \frac{Nd}{NT} \times 100\%$$

Where: Nd – total number of dead crustaceans

NT – total number of tested crustaceans

Table S9. Ecotoxicology results of the test compounds on invertebrates *T. platyurus*.

Code name	Dose, mg L ⁻¹	Response, %	Mean response, %	Standard deviation
IA	5	8	10.6	6.4
	5	6		
	5	18.75		
	10	63.6	58.8	6.5
	10	51.3		
	10	61.5		
	25	100	-	-
	25	100		
	50	100		
	50	100	14.6	3.6
IAA	10	18.8		
	10	12.9		
	10	12.1		
	12	29.4	30.7	1.9
	12	32.1		
	15	56.2		
	15	63.4	61.5	4.8
	15	65		
	25	100		
	25	100	-	-
	25	100		

Technical Data Sheet of CHP BAR 1400



Technical Data Sheet and
Product Specification

CHP BAR 1400

Description	Anionic aqueous dispersion of styrene acrylate copolymer for barrier coating of paper and board.	
Application	CHP BAR 1400 is a barrier binder for general purposes. It gives good balance of properties regarding water, water vapour and grease resistance. The binder is stable against yellowing induced by heat and UV-light. The product complies with FDA (§ 176.170 and 176.180) and BfR XIV and XXXVI requirements.	
Specification	Solids content, %	50 ± 1
	pH	7± 1
	Brookfield viscosity, mPas (LVTDV – II, 60 rpm, spindle 2)	< 500
Characteristics of the dispersion	Appearance	milky white
	Stabilization	anionic
	Density (g/cm3)	1,0
Packaging, storage & safety	CHP BAR 1400 is supplied in bulk by road tanker. Consult us about delivery in containers or drums. The dispersion should be kept in the original containers or in stainless steel, aluminium or plastic tanks. Ordinary steel tanks with a corrosion -proof lining can also be used. The containers should be kept closed to prevent evaporation of the water and the formation of a skin on the surface. The product should not be exposed to frost or to temperatures exceeding 30°C. Under normal conditions, the product can be stored for six months with no significant loss of its properties, but it cannot be guaranteed for a longer time. For safety issues, please refer to the material safety data sheet.	
Technical Service	Trained and experienced field sales and technical service representatives of CH-Polymers are ready to provide advice and assistance with laboratory tests and plant trials, to determine the best application conditions.	
Contact	CH-Polymers Oy Tel. +358 9 5024 4150 Info@ch-polymers.com www.ch-polymers.com	

This information is based on our laboratory tests, experience and best knowledge for the moment. We recommend that the prospective user determine the usage of our raw materials and recommendations before adopting them on a commercial scale.

References

1. L. Matt, J. Parve, O. Parve, T. Pehk, T. H. Pham, I. Liblikas, L. Vares and P. Jannasch, *ACS Sustainable Chem. Eng.*, 2018, **6**, 17382-17390.
2. S. Laanesoo, O. Bonjour, J. Parve, O. Parve, L. Matt, L. Vares and P. Jannasch, *Biomacromolecules*, 2021, **22**, 640-648.
3. F. Nonque, A. Sahut, N. Jacquel, R. Saint-Loup, P. Woisel and J. Potier, *Polym. Chem.*, 2020, **11**, 6903-6909.
4. J. J. Gallagher, M. A. Hillmyer and T. M. Reineke, *ACS Sustainable Chem. Eng.*, 2016, **4**, 3379-3387.