Supporting Information

Effect of Variations in Micro-patterns and Surface Modulus on Marine Fouling of Engineering Polymers

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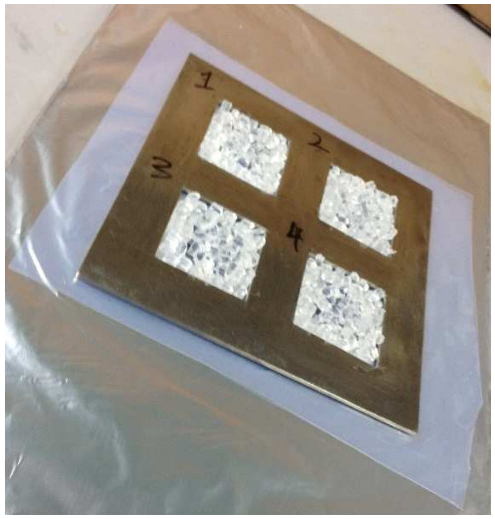

Figure S1. Preparation of polymer and moulds for pattern replication in PU. Image shows metal frame placed on fluorinated sheet (white). Within each opening of the frame (1-4) there is a silicone mould placed (invisible in the image), subsequently covered with PU pellets. The thus prepared frame was subsequently placed between the plates of the hot plate, as described in the manuscript.

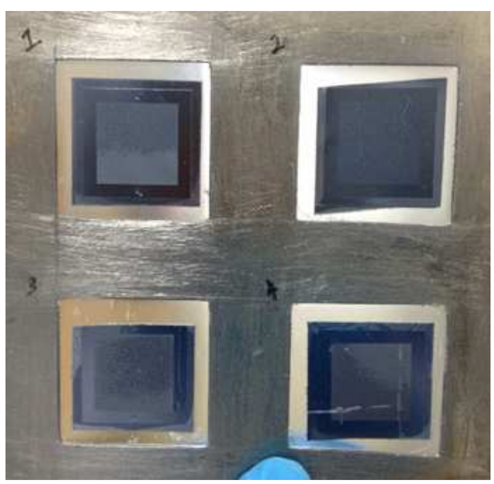

Figure S2. PMMA samples after casting, using hot press, and prior demoulding. In window “4” a mould cracked during the compression moulding is visible.

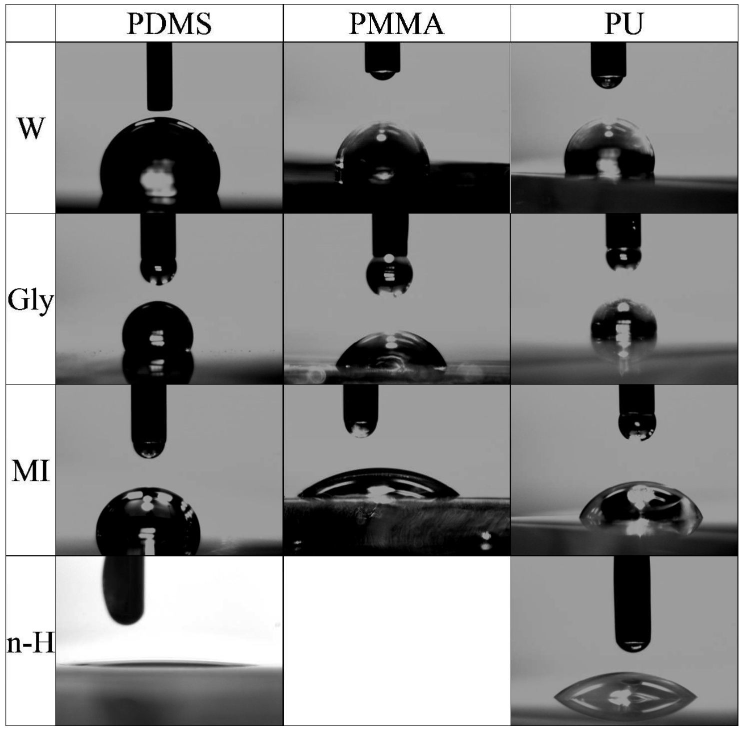

Figure S3. Examples of static contact angle measurements on smooth PDMS, PMMA and PU using water (W), anhydrous glycerol (Gly), methilene iodide (MI) and n-hexadecane (n-H). Static contact angle of n-H on PMMA could not have been determined due to rapid spreading (complete wetting) of the liquid on PMMA surface.

Table S1. Results of static and dynamic contact angle measurements on smooth PDMS, PMMA and PU surfaces using water, glycerol, methyl iodide and n-hexadecane. VCA Optima apparatus, (AST products, Inc.) was used. Each sample was tested in four replicates for each liquid, and each replicate was measured at 4 different areas. We applied the Owens' and Zisman's methods, to evaluate the surface energies and critical surface tensions of casted smooth PDMS, PU and PMMA samples. Numbers in brackets are standard errors. SCA – Static Contact Angle, ACA – Advancing Contact Angle, RCA – Receding Contact Angle. "*" and "**" refer to measurements excluded from calculations of surface energy and critical surface tension due to poor fit, respectively.

	water			glycerol			methyl iodide			n-hexadecane			Surface energy	Critical surface tension
	SCA	ACA	RCA	SCA	ACA	RCA	SCA	ACA	RCA	SCA	ACA	RCA		
PDMS	*114.7 (0.5)	*120.0 (0.7)	*107.6 (1.0)	**116.2 (0.3)	**116.9 (0.4)	**109.2 (0.6)	97.4 (0.4)	101.3 (0.4)	73.1 (0.4)	46.1 (0.2)	40.6 (0.5)	17.6 (0.6)	**19.54 (R^2=0.6310)	*23.36 (R^2=0.9905)
PMMA	96.4 (1.3)	98.4 (1.6)	83.4 (1.7)	77.5 (2.5)	79.4 (2.5)	77.5 (2.0)	*54.3 (2.0)	*61.0 (2.6)	*43.1 (1.8)	32.0 (2.6)	29.4 (2.5)	24.8 (2.6)	*26.07 (R^2=0.9908)	*20.76 (R^2=0.9974)
PU	105.9 (0.2)	114.1 (0.6)	99.4 (0.9)	*99.0 (0.6)	*98.0 (0.3)	*98.0 (1.1)	69.2 (0.5)	79.0 (0.4)	66.1 (0.6)	**37.3 (0.7)	**36.5 (0.8)	**33.2 (0.7)	*22.3 (R^2=0.9753)	**23.9 (R^2=0.9634)

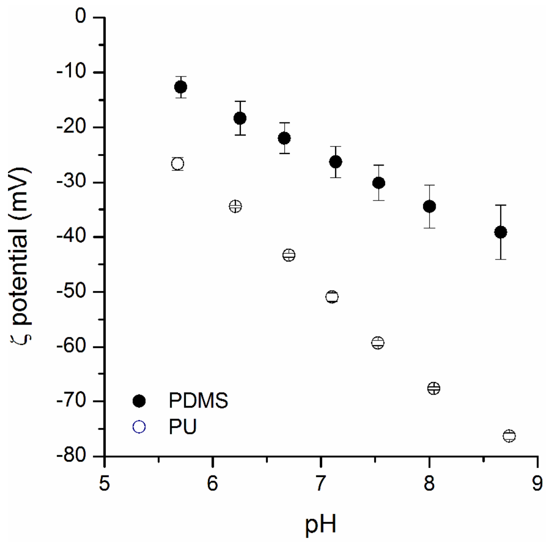

Figure S4. ζ-potentials measured on smooth PDMS and PU substrates.

Laboratory assays

To collect additional information about the fouling performance of the polymers studied we subjected the investigated surfaces to diatom (*Halamphora coffeaeformis*) settlement assays. We compared the settlement on smooth polymer samples with the settlement on smooth microscope cover glass reference samples, and the settlement on smooth and patterned polymer samples. The type of the material used had significant effect on *Halamphora* settlement. Unlike the results of the field test, PMMA was the least prone to the settlement of diatoms under laboratory conditions. The number of diatoms settled on PDMS and PMMA was lower than on the respective glass controls, but the number of organisms settled on PU was higher than on the glass control. Depending on the exact composition, the elastic modulus of glass is at least one order of magnitude larger than that of PMMA (PMMA was the hardest polymer in this study). Thus, considering the mechanical properties only, one could expect that glass would be the most prone to the settlement. In this context, the "unexpectedly" higher settlement on PU indicates the importance, or even the prevailing role, of surface parameters other than modulus in the amphora attachment. To test this hypothesis we assessed *Halamphora* settlement on physically (patterning) modified PDMS and PU surfaces using multi-way ANOVA and post-hoc Tukey's multiple comparisons. We observed no significant difference in number of organisms settled on the tested samples.

Statistical analysis of data obtained during mechanical surface tests (surface hardness and modulus)

Table S2. ANOVA showing the effect of the sample Material (PDMS, PMMA, PU) and of the dimensions of the small features (0 (smooth), 3, or 5 µm) of the surface Pattern on surface hardness.

	Df	Sum Sq	Mean Sq	F value	Pr(>F)	Sig.codes
Material	2	1.3732	0.6866	529.3832	< 0.0001	***

Pattern	2	0.0183	0.0092	7.0532	0.0015	**
Material : Pattern	4	0.0489	0.0122	9.4290	< 0.0001	***
Residuals	81	0.1051	0.0013			

Tukey's Tests (see S4):
Pattern: PDMS: 0 µm = 3 µm = 5 µm; PU: 0 µm = 3 µm = 5 µm; PMMA: 3 µm >***0 µm = 5 µm
Material: 0 µm, 3 µm, and 5 µm: PMMA > PU = PDMS

Significance codes: '***' < 0.001; '**' < 0.01; '*' < 0.05; '.' < 0.1; ' ' < 1
Barlett's test: p = 0.0348, Shapiro – Wilk test: p < 0.0001

Table S3. ANOVA showing the effect of the sample Material (PDMS, PMMA, PU) and of the dimensions of the small features (0 (smooth), 3, or 5 µm) of the surface Pattern on surface modulus.

	Df	Sum Sq	Mean Sq	F value	Pr(>F)	Sig.codes
Material	2	412.85	206.426	1236.7032	< 0.0001	***
Pattern	2	2.14	1.069	6.4068	0.0026	**
Material : Pattern	4	4.00	0.999	5.9851	0.0003	***
Residuals	81	13.52	0.167			

Tukey's tests (see S5):
Pattern: PDMS: 0 µm = 3 µm = 5 µm; PU: 0 µm = 3 µm = 5 µm; PMMA: 0 µm = 3 µm > 5 µm
Materials: PMMA > PU = PDMS

Significance codes: '***' < 0.001; '**' < 0.01; '*' < 0.05; '.' < 0.1; ' ' < 1
Bartlett's test: p = 0.4418, Shapiro – Wilk test: p < 0.0001

Table S4. The results of post-hoc Tukey's multiple comparison test of data collected during mechanical test, comparing the effect of the sample Material (PDMS, PMMA, PU) and of the dimensions of small features (0 (smooth), 3, or 5 µm) of the surface Pattern on surface hardness. Significance levels are marked with colors: green (p < 0.001), orange (0.001 < p < 0.01), and yellow (0.01 < p < 0.05).

	PDMS : 0	*PDMS : 3*	*PDMS : 5*	*PU : 0*	*PU : 3*	*PU : 5*	*PMMA : 0*	*PMMA : 3*	*PMMA : 5*
PDMS : 0		1.0000	1.0000	1.0000	0.9993	1.0000	< 0.0001	< 0.0001	< 0.0001
PDMS : 3			1.0000	1.0000	0.9997	1.0000	< 0.0001	< 0.0001	< 0.0001
PDMS : 5				1.0000	0.9943	0.9997	< 0.0001	< 0.0001	< 0.0001
PU : 0					0.9981	1.0000	< 0.0001	< 0.0001	< 0.0001
PU : 3						1.0000	< 0.0001	< 0.0001	< 0.0001
PU : 5							< 0.0001	< 0.0001	< 0.0001
PMMA : 0								0.0008	0.2091
PMMA : 3									< 0.0001
PMMA : 5									

***	p < 0.001
**	0.001 < p < 0.01
*	0.01 < p < 0.05

Table S5. The results of post-hoc Tukey's multiple comparison test of data collected during mechanical test, comparing the effect of the sample Material (PDMS, PMMA, PU) and of the dimensions of small features (0 (smooth), 3, or 5 µm) of the surface Pattern on surface modulus. Significance levels are marked with colors: green (p < 0.001), orange (0.001 < p < 0.01), and yellow (0.01<p<0.05).

	PDMS : 0	PDMS : 3	PDMS : 5	PU : 0	PU : 3	PU : 5	PMMA : 0	PMMA : 3	PMMA : 5
PDMS : 0		1.0000	1.0000	1.0000	0.9999	1.0000	> 0.0001	> 0.0001	> 0.0001
PDMS : 3			1.0000	1.0000	1.0000	1.0000	> 0.0001	> 0.0001	> 0.0001
PDMS : 5				1.0000	1.0000	1.0000	> 0.0001	> 0.0001	> 0.0001
PU : 0					1.0000	1.0000	> 0.0001	> 0.0001	> 0.0001
PU : 3						1.0000	> 0.0001	> 0.0001	> 0.0001
PU : 5							> 0.0001	> 0.0001	> 0.0001
PMMA : 0	***	p < 0.001						0.9987	0.0002
PMMA : 3	**	0.001 < p < 0.01							> 0.0001
PMMA : 5	*	0.01 < p < 0.05							

Statistical analysis of data collected during static field immersion tests

Table S6. ANOVA showing the effect of the sample material (PDMS, PMMA, PU), of the dimensions of small features (0 (smooth), 3, or 5 µm) of the surface Pattern, and of the Pressure (0, 50, and 100 Psi) of water jet applied for surface cleaning on surface fouling during the first immersion test.

	Df	Sum Sq	Mean Sq	F value	Pr(>F)	Sig.codes
Material	2	1798.68	899.3	60.715	< 0.0001	***
Pressure	2	136.82	68.4	4.619	0.013	*
Pattern	2	1443.90	722.0	48.739	< 0.0001	***
Material : Pressure	4	615.67	153.9	10.391	< 0.0001	***
Material : Pattern	4	573.95	143.5	9.687	< 0.0001	***
Pressure : Pattern	4	13.65	3.4	0.230	0.920	
Material : Pressure : Pattern	8	43.33	5.4	0.366	0.935	
Residuals	72	1066.50	14.8			

Tukey’s tests (see S7 & S8):
Material x Pressure - Material: 0 Psi: PDMS = PMMA = PU; 50 Psi & 100 Psi: PDMS < PU < PMMA
Material x Pressure - Pressure: PDMS: 0 Psi > 50 Psi = 100 Psi; PU: 0 Psi = 50 Psi, 50 Psi = 100 Psi, 0 Psi > 100 Psi; PMMA: 0 Psi = 50 Psi = 100 Psi
Material x Pattern - Material: 0 µm: PDMS = PMMA < PU; 3 µm: PDMS = PU > PMMA; 5 µm: PDMS < PMMA = PU
Material x Pattern – Pattern: PDMS: 0 µm = 3 µm = 5v; PMMA: 0 µm < 3 µm, 0 µm = 5 µm, 3 µm = 5 µm; PU: 0 µm = 3 µm, 0 µm = 5 µm, 3 µm < 5 µm

Significance codes: '***' <0.001; '**' <0.01; '*' <0.05; '.' <0.1; ' ' <1
Bartlett’s test: p = 0.0258; Shapiro – Wilk test: p > 0.05.

Table S7. The results of post-hoc Tukey's multiple comparison test of data collected during the first immersion test, comparing effect of the sample Material (PDMS, PMMA, PU) and of the Pressure (0, 50, and 100 Psi) used during water jet cleaning on surface fouling. Significance levels are marked with colors: green ($p < 0.001$), orange ($0.001 < p < 0.01$), and yellow ($0.01 < p < 0.05$).

	PDMS : 0	PDMS : 50	PDMS : 100	PMMA : 0	PMMA : 50	PMMA : 100	PU : 0	PU : 50	PU : 100
PDMS : 0		> 0.0001	> 0.0001	0.9864	1.0000	0.9996	0.8816	0.9359	0.0453
PDMS : 50			0.7137	> 0.0001	> 0.0001	> 0.0001	> 0.0001	> 0.0001	0.0002
PDMS : 100				> 0.0001	> 0.0001	> 0.0001	> 0.0001	> 0.0001	> 0.0001
PMMA : 0					0.9990	0.8656	1.0000	0.4420	0.0049
PMMA : 50						0.9967	0.9775	0.8781	0.0427
PMMA : 100							0.6059	0.9996	0.2967
PU : 0	***	p < 0.001						0.1646	0.0004
PU : 50	**	0.001 < p < 0.01							0.5739
PU : 100	*	0.01 < p < 0.05							

Table S8. The results of post-hoc Tukey's multiple comparison test of data collected during the first immersion test, comparing effect of the sample Material (PDMS, PMMA, PU) and of the dimensions of small features (0 (smooth), 3, or 5 μm) of the surface Pattern on surface fouling. Significance levels are marked with colors: green ($p < 0.001$), orange ($0.001 < p < 0.01$), and yellow ($0.01 < p < 0.05$).

	PDMS : 0	PDMS : 3	PDMS : 5	PMMA : 0	PMMA : 3	PMMA : 5	PU : 0	PU : 3	PU : 5
PDMS : 0		1.0000	0.9703	0.1466	> 0.0001	0.0003	0.0012	0.1302	> 0.0001
PDMS : 3			0.9840	0.1152	> 0.0001	0.0002	0.0008	0.1017	> 0.0001
PDMS : 5				0.0069	> 0.0001	> 0.0001	> 0.0001	0.0058	> 0.0001
PMMA : 0					> 0.0001	0.0793	0.7772	1.0000	0.0081
PMMA : 3						0.9993	0.0029	> 0.0001	0.5739
PMMA : 5							0.5626	0.0862	0.9995
PU : 0	***	p < 0.001						0.8064	0.4330
PU : 3	**	0.001 < p < 0.01							0.0096
PU : 5	*	0.01 < p < 0.05							

Table S9. ANOVA showing the effect of the coating Material (PDMS, PMMA, PU), of the dimensions of small features (0 (smooth), 3, or 5 μm) of the surface Pattern, and of the Pressure (0, 50, and 100 Psi) of water jet applied for surface cleaning on surface fouling during the second immersion test.

	Df	Sum Sq	Mean Sq	F value	Pr(>F)	Sig.codes
Material	2	15548.1	7774.1	51.078	< 0.0001	***
Pressure [psi]	2	4965.6	2482.8	16.313	< 0.0001	***
Pattern [um]	2	3440.1	1720	11.301	< 0.0001	***
Material : Pressure	4	1674	418.5	2.750	0.0345	*
Material : Pattern	4	5807.7	1451.9	9.540	< 0.0001	***
Pressure : Pattern	4	814.6	203.6	1.338	0.2642	
Material : Pressure : Pattern	8	524.2	65.5	0.431	0.8990	
Residuals	72	10958.5	152.2			

Tukey's tests (see S10 & S11):
Material x Pressure - Material: 0 Psi: PDMS = PMMA = PU; 50 Psi & 100 Psi: PDMS < PU = PMMA
Material x Pressure - Pressure: PDMS: 0 Psi = 50 Psi, 50 Psi = 100 Psi, 0 Psi > 100 Psi; PU & PMMA: 0 Psi = 50 Psi = 100 Psi
Material x Pattern - Material: 0 μm : PDMS = PMMA > PU; 3 μm & 5 μm: PDMS < PU = PMMA
Material x Pattern – Pattern: PDMS & PU: 0 μm = 3 μm = 5 μm; PMMA: 0 μm <

3 µm = 5 µm
Significance codes: '***' <0.001; '**' <0.01; '*' <0.05; '.' <0.1.
Bartlett & Shapiro – Wilk tests: $p > 0.05$

Table S10. The results of post-hoc Tukey's multiple comparison test of data collected during the second immersion test, comparing effect of the sample Material (PDMS, PMMA, PU) and of the Pressure (0, 50, and 100 Psi) used during water jet cleaning on surface fouling. Significance levels are marked with colors: green ($p < 0.001$), orange ($0.001 < p < 0.01$), and yellow ($0.01 < p < 0.05$).

	PDMS : 0	PDMS : 50	PDMS : 100	PMMA : 0	PMMA : 50	PMMA : 100	PU : 0	PU : 50	PU : 100
PDMS : 0		0.0337	< 0.0001	0.2813	0.2510	0.9797	0.0842	0.0776	0.9999
PDMS : 50			0.3276	< 0.0001	< 0.0001	0.0028	< 0.0001	< 0.0001	0.0081
PDMS : 100				< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001
PMMA : 0					1.0000	0.9284	1.0000	1.0000	0.5543
PMMA : 50						0.9100	1.0000	1.0000	0.5130
PMMA : 100							0.7288	0.7097	0.9995
PU : 0	***	p < 0.001						1.0000	0.2424
PU : 50	**	0.001 < p < 0.01							0.2273
PU : 100	*	0.01 < p < 0.05							

Table S11. The results of post-hoc Tukey's multiple comparison test of data collected during the second immersion test, comparing effect of the sample Material (PDMS, PMMA, PU) and of the dimensions of small features (0 (smooth), 3, or 5 µm) of the surface Pattern on surface fouling. Significance levels are marked with colors: green ($p < 0.001$), orange ($0.001 < p < 0.01$), and yellow ($0.01 < p < 0.05$).

	PDMS : 0	PDMS : 3	PDMS : 5	PMMA : 0	PMMA : 3	PMMA : 5	PU : 0	PU : 3	PU : 5
PDMS : 0		0.9997	0.9998	0.8643	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001
PDMS : 3			1.0000	0.9917	< 0.0001	< 0.0001	0.0003	0.0002	< 0.0001
PDMS : 5				0.9895	< 0.0001	< 0.0001	0.0002	0.0002	< 0.0001
PMMA : 0					< 0.0001	< 0.0001	0.0065	0.0050	0.0014
PMMA : 3						0.9889	0.0629	0.0776	0.1859
PMMA : 5							0.1011	0.1144	0.1941
PU : 0	***	p < 0.001						1.0000	0.9999
PU : 3	**	0.001 < p < 0.01							1.0000
PU : 5	*	0.01 < p < 0.05							