One Earth, Volume *8*

Supplemental information

Eco-friendly active film and sealant

for underwater drug delivery to diseased corals

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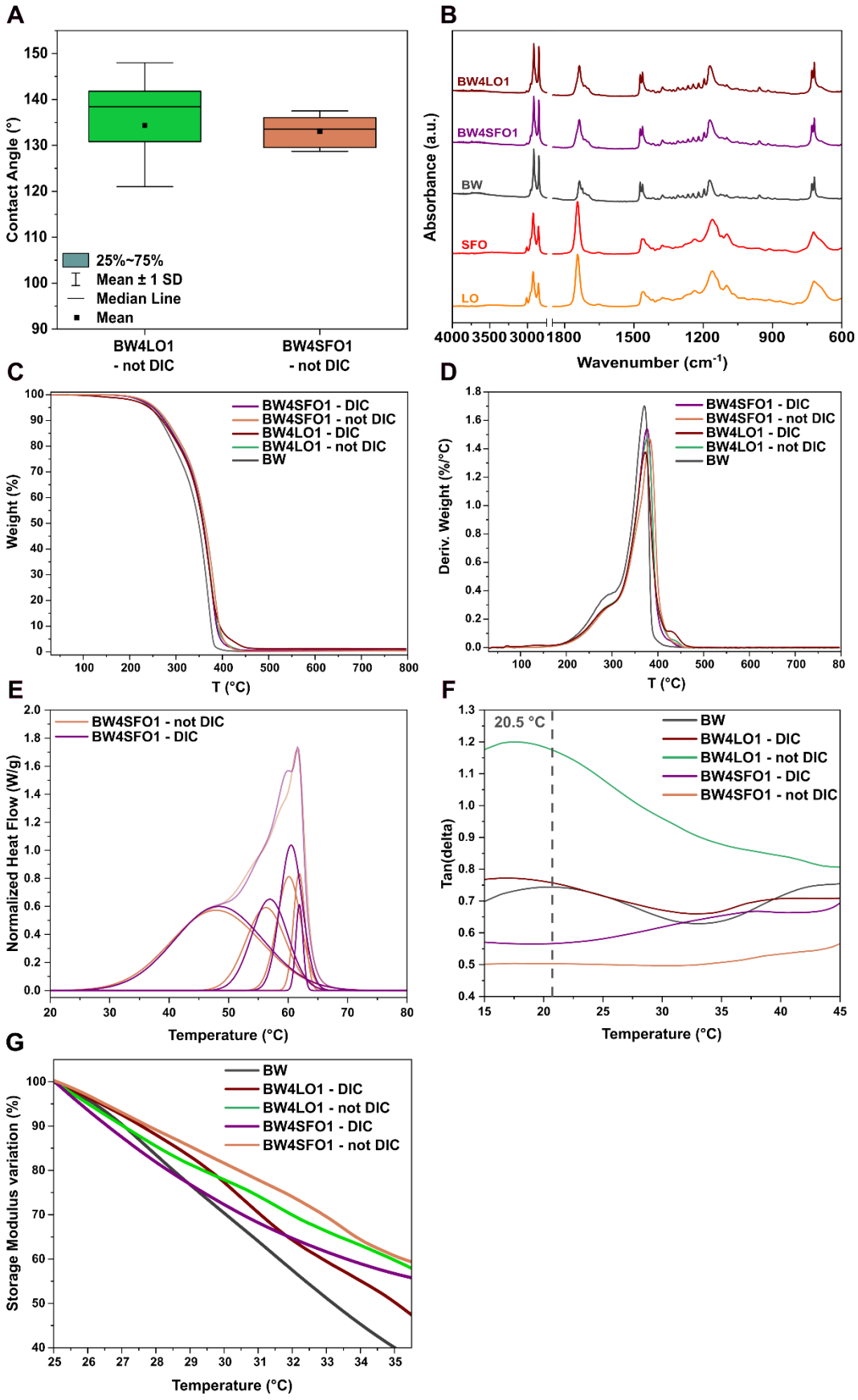
A
Contact Angle (°)
150
140
130
120
110
100
90
25%~75%
Mean ± 1 SD
Median Line
Mean
BW4LO1
- not DIC
BW4SFO1
- not DIC
B
Absorbance (a.u.)
BW4LO1
BW4SFO1
BW
SFO
LO
4000 3500 3000 1800 1500 1200 900 600
Wavenumber (cm-1)
C
Weight (%)
BW4SFO1 - DIC
BW4SFO1 - not DIC
BW4LO1 - DIC
BW4LO1 - not DIC
BW
T (°C)
D
Deriv. Weight (%/°C)
BW4SFO1 - DIC
BW4SFO1 - not DIC
BW4LO1 - DIC
BW4LO1 - not DIC
BW
T (°C)
E
Normalized Heat Flow (W/g)
BW4SFO1 - not DIC
BW4SFO1 - DIC
Temperature (°C)
F
20.5 °C
Tan(delta)
BW
BW4LO1 - DIC
BW4LO1 - not DIC
BW4SFO1 - DIC
BW4SFO1 - not DIC
Temperature (°C)
G
Storage Modulus variation (%)
BW
BW4LO1 - DIC
BW4LO1 - not DIC
BW4SFO1 - DIC
BW4SFO1 - not DIC
Temperature (°C)

Figure S1. Physical-chemical and thermodynamical characterization of sealants. A, BW4LO1–not DIC, and BW4SFO1–not DIC water contact angle. **B**, FT-IR normalized spectra of the two pastes (BW4LO1-DIC, BW4SFO1–DIC) and the relative single reagents (BW, SFO, LO). **C**, **D**, respectively thermogravimetric curve and the first derivative of the pristine BW and the paste sealants BW4LO1–DIC, BW4SFO1–DIC, BW4LO1–not DIC, BW4SFO–not DIC. **E**, DSC deconvolution of BW4SFO1–DIC and BW4SFO1–not DIC; the semi-transparent curves represent the raw curves, and the more saturated curves are the obtained deconvolved peaks. **F**, tan(delta) of pastes and BW as a function of the temperature. **G**, Stiffness variation of the sealants in the range of temperature between 25 and 35°C. See also Figure 1 and S3.

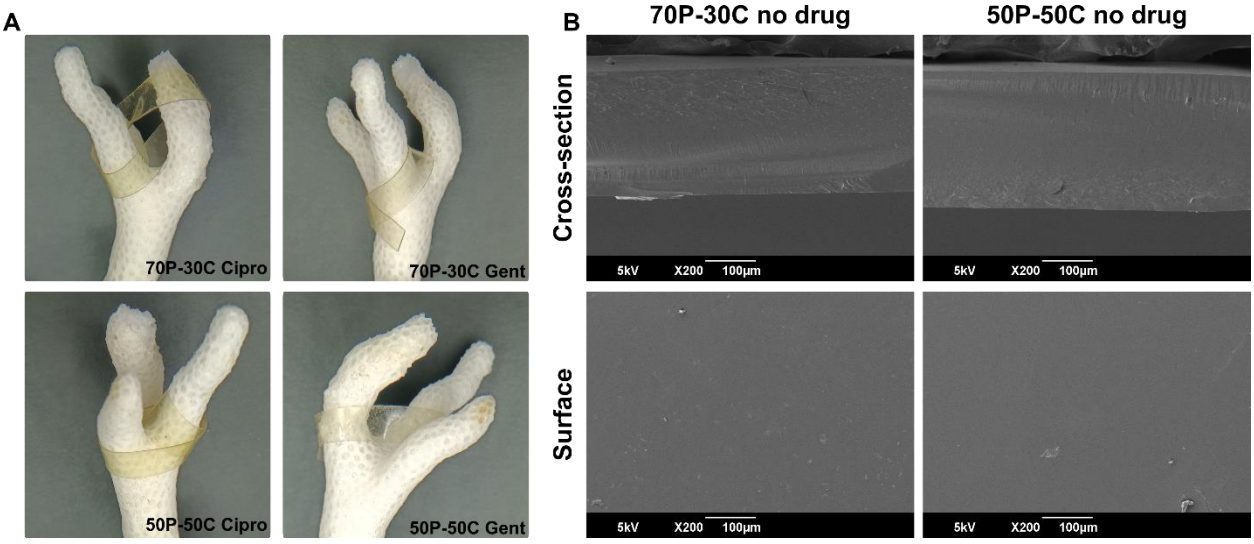

Figure S2. Macroscopical and microscopical film images. **A**, representative photos of the film used; **B**, SEM acquisitions of the 70P-30C and 50P-50C without drugs loaded. See also Figure 2.

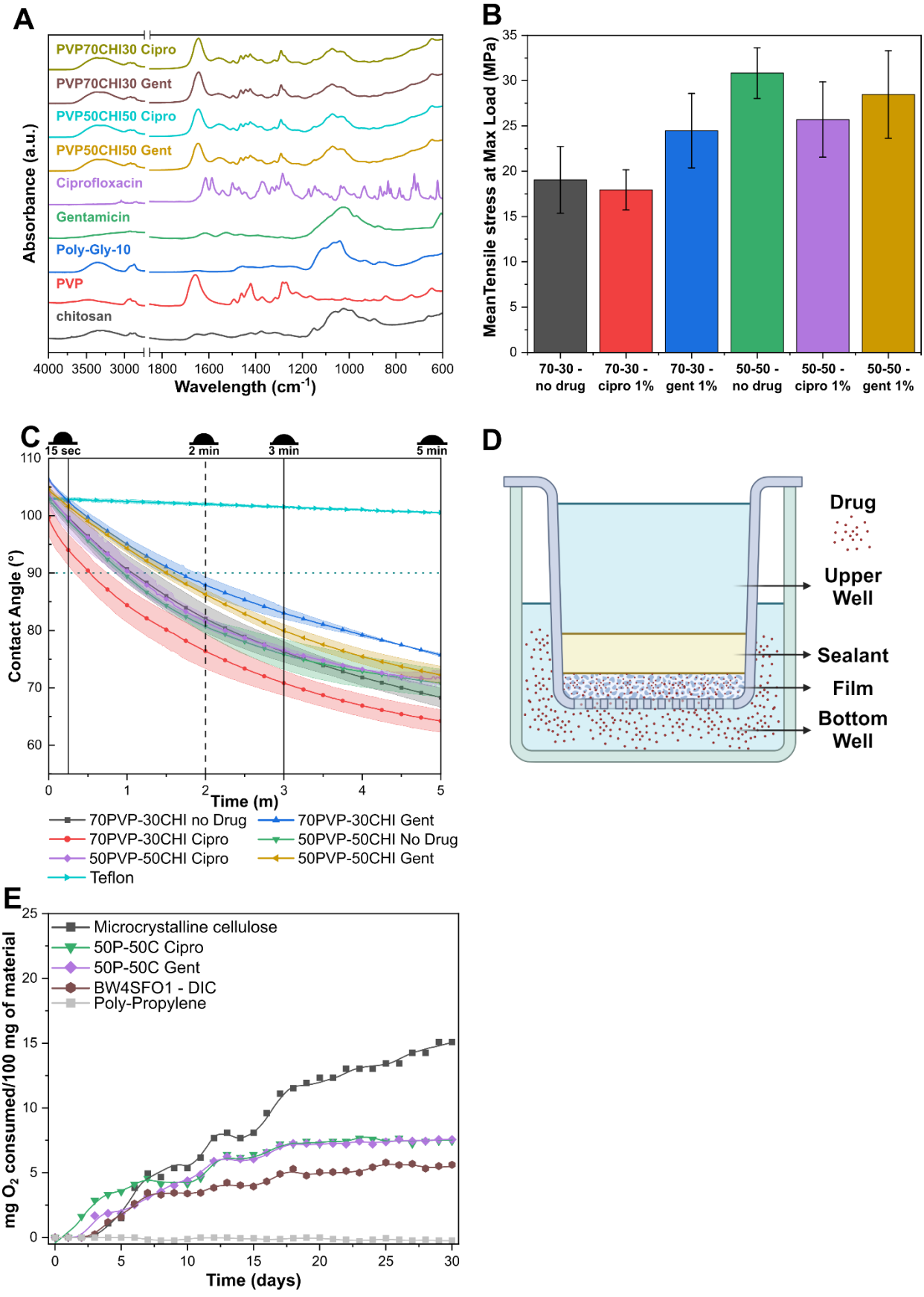
A
PVP70CHI30 Cipro
PVP70CHI30 Gent
PVP50CHI50 Cipro
PVP50CHI50 Gent
Ciprofloxacin
Gentamicin
Poly-Gly-10
PVP
chitosan
Absorbance (a.u.)
4000 3500 3000 1800 1600 1400 1200 1000 800 600
Wavelength (cm-1)
B
MeanTensile stress at Max Load (MPa)
0 5 10 15 20 25 30 35
70-30 - no drug
70-30 - cipro 1%
70-30 - gent 1%
50-50 - no drug
50-50 - cipro 1%
50-50 - gent 1%
C
15 sec
2 min
3 min
5 min
Contact Angle (°)
110 100 90 80 70 60
0 1 2 3 4 5
Time (m)
70PVP-30CHI no Drug
70PVP-30CHI Gent
70PVP-30CHI Cipro
50PVP-50CHI No Drug
50PVP-50CHI Cipro
50PVP-50CHI Gent
Teflon
D
Drug
Upper Well
Sealant
Film
Bottom Well
E
Microcrystalline cellulose
50P-50C Cipro
50P-50C Gent
BW4SFO1 - DIC
Poly-Propylene
mg O2 consumed/100 mg of material
0 5 10 15 20 25
0 5 10 15 20 25 30
Time (days)

Figure S3. Characterization of the films, transwell model, and BOD degradation. **A**, FT-IR spectra of the films 70P-30C and 50P-50C and their single components. **B**, mean tensile stress at the maximum load of the films; represented as mean ± SEM. **C**, Dynamic Water Contact Angle of the two different film compositions in the presence/absence of the drugs (either Cipro or Gent); transparent bars are the confidence interval within one SD. Teflon is used as a control. **D**, schematic representation of the transwell model. **E**, Biochemical oxygen degradation of the films 50P-50C with Cipro and Gent, and the paste sealant BW4SFO1–DIC; Microcrystalline cellulose and Poly-Propylene are added as positive and negative controls, respectively. See Figure 2 and S2.

BW	Beeswax
LO	Linseed Oil
SFO	Sunflower Oil
DIC	Dry Ice Cooling
not DIC	Not Dry Ice Cooling
P	Polyvinylpyrrolidone
C	Chitosan
Cipro	Ciprofloxacin
Gent	Gentamicin
BW4LO1-DIC	Sealant with Beeswax and Linseed oil prepared with Dry Ice cooling method
BW4LO1-not DIC	Sealant with Beeswax and Linseed oil prepared without Dry Ice cooling method
BW4SFO1-DIC	Sealant with Beeswax and Sunflower oil prepared with Dry Ice cooling method
BW4SFO1-not DIC	Sealant with Beeswax and Sunflower oil prepared without Dry Ice cooling method
50P-50C Cipro	Film with 50% of Polyvinylpyrrolidone, 50% Chitosan and Ciprofloxacin
70P-30C Cipro	Film with 70% of Polyvinylpyrrolidone, 30% Chitosan and Ciprofloxacin
50P-50C Gent	Film with 50% of Polyvinylpyrrolidone, 50% Chitosan and Gentamicin
70P-30C Gent	Film with 70% of Polyvinylpyrrolidone, 30% Chitosan and Gentamicin

Table S1. List of acronyms used in this paper.

Sample Name	**Peak at 48.1 °C**	**Peak at 56.6 °C**	**Peak at 60.3°C**	**Peak at 61.9 °C**
BWSFO1 – DIC	11.15	4.97	4.84	0.92
BW4SFO1 – not DIC	10.93	4.94	4.14	2.02

Table S2. Area of deconvolved peaks for BW4SFO–DIC and BW4SFO1–not DIC. Enthalpy (area under the curve, measured as J/g) of the peaks of the 1st melting round deconvolved curves for BW4SFO1 – DIC and BW4SFO1 – not DIC. See also Figure 1 and Table 1.

Film name	Conc. mg/mL	Film name	Conc. mg/mL	Alpha	Significance	Prob	p-value
50P/50C Cipro	1	TEFLON	1	0.05	1	8.92E-28	0.001
50P/50C Cipro	2.5	TEFLON	2.5	0.05	1	5.81E-27	0.001
50P/50C Cipro	5	TEFLON	5	0.05	1	8.92E-28	0.001
50P/50C Gent	1	TEFLON	1	0.05	1	1.76E-07	0.001
50P/50C Gent	1	50P/50C Cipro	1	0.05	1	3.05E-25	0.001
50P/50C Gent	2.5	TEFLON	2.5	0.05	1	2.58E-25	0.001
50P/50C Gent	2.5	50P/50C Cipro	2.5	0.05	1	8.40E-04	0.001
50P/50C Gent	5	TEFLON	5	0.05	1	9.03E-28	0.001
50P/50C Gent	5	50P/50C Cipro	5	0.05	0	1	1
70P/30C Cipro	1	TEFLON	1	0.05	1	8.92E-28	0.001
70P/30C Cipro	1	50P/50C Cipro	1	0.05	0	1	1
70P/30C Cipro	1	50P/50C Gent	1	0.05	1	3.05E-25	0.001
70P/30C Cipro	2.5	TEFLON	2.5	0.05	1	5.81E-27	0.001
70P/30C Cipro	2.5	50P/50C Cipro	2.5	0.05	0	1	1
70P/30C Cipro	2.5	50P/50C Gent	2.5	0.05	1	8.40E-04	0.001
70P/30C Cipro	5	TEFLON	5	0.05	1	8.92E-28	0.001
70P/30C Cipro	5	50P/50C Cipro	5	0.05	0	1	1
70P/30C Cipro	5	50P/50C Gent	5	0.05	0	1	1
70P/30C Gent	1	TEFLON	1	0.05	1	2.11E-12	0.001
70P/30C Gent	1	50P/50C Cipro	1	0.05	1	4.88E-31	0.001
70P/30C Gent	1	50P/50C Gent	1	0.05	1	3.82E-18	0.001
70P/30C Gent	1	70P/30C Cipro	1	0.05	1	4.88E-31	0.001
70P/30C Gent	2.5	TEFLON	2.5	0.05	1	1.22E-24	0.001
70P/30C Gent	2.5	50P/50C Cipro	2.5	0.05	1	3.22E-06	0.001
70P/30C Gent	2.5	50P/50C Gent	2.5	0.05	0	1	1
70P/30C Gent	2.5	70P/30C Cipro	2.5	0.05	1	3.22E-06	0.001
70P/30C Gent	5	TEFLON	5	0.05	1	9.49E-28	0.001
70P/30C Gent	5	50P/50C Cipro	5	0.05	0	1	1
70P/30C Gent	5	50P/50C Gent	5	0.05	0	1	1
70P/30C Gent	5	70P/30C Cipro	5	0.05	0	1	1

Table S3. Results of the Two-way ANOVA test followed by the Bonferroni post-hoc for all the comparisons among the materials tested in the antibacterial liquid test. See also Figure 3.

ASV	STYLO-I1	STYLO-I2	STYLO-I3	STYLO-D1	STYLO-D2	STYLO-D3	STYLO-H1	STYLO-H2	STYLO-H3	Best hit
ASV_30	45.19	49.30	0.26	19.33	15.95	21.51	19.35	19.51	12.82	*Vibrio harveyi/owensii/parahaemolyticus/alginolyticus*
ASV_580	3.59	3.26	0.02	2.17	2.28	1.30	1.01	1.42	2.76	*Vibrio alginolyticus/fluvialis/maritimus/furnissii*
ASV_1353	1.20	1.54	0.00	0.00	1.08	0.58	0.00	0.00	0.00	*Vibrio natriegens/rotiferianus/astriarenae/ishigakensis*
ASV_1994	1.26	0.87	0.00	0.72	0.42	0.26	0.87	0.00	0.00	*Vibrio gallaecicus/mediterranei/splendidus/echinoideorum*
ASV_4320	1.07	0.29	0.00	0.08	0.13	0.09	0.03	0.33	0.00	*Vibrio gazogenes/alfacsensis/salilacus*
ASV_15901	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.88	0.00	*Vibrio europaeus/alfacsensis/marinisediminis/gangliei*

Table S4 – Best hit species for ASVs classified as belonging to genus *Vibrio*; relative abundance (%) of each ASV in each sample. See also Table 3, 4 and Figure 4.