

Supplementary Information to: “Influence of niche-specific nutrients on secondary metabolism in Vibrionaceae”.

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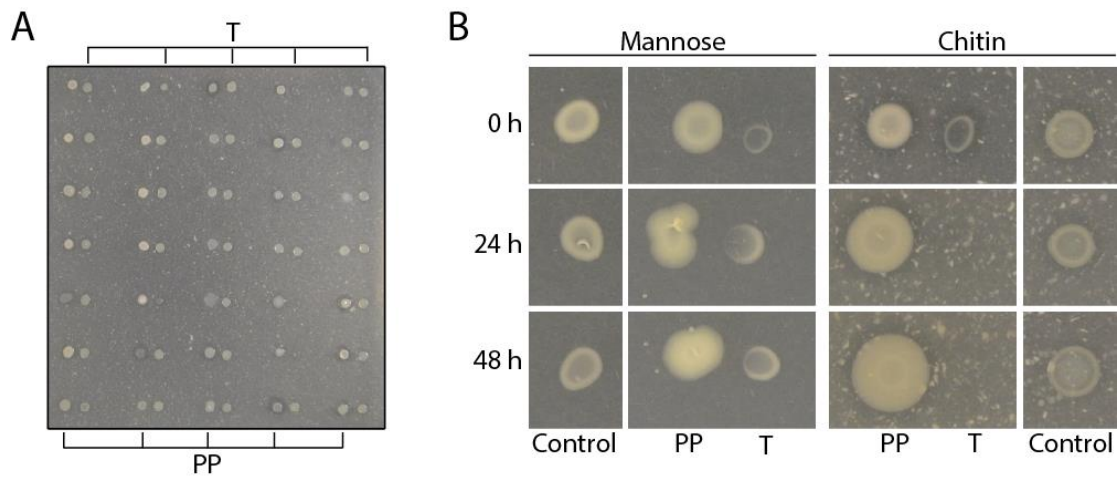


Figure S1. (A) Example of a plate prepared spotting the potential producers (PP) and the target strain *V. anguillarum* (T) on chitin medium. Clear haloes surrounding the colonies indicate chitinolytic activity. (B) Detailed behavior of *V. furnissii* S0821 over time and on the two media. “Control” is the target strain spotted alone on the same two media.

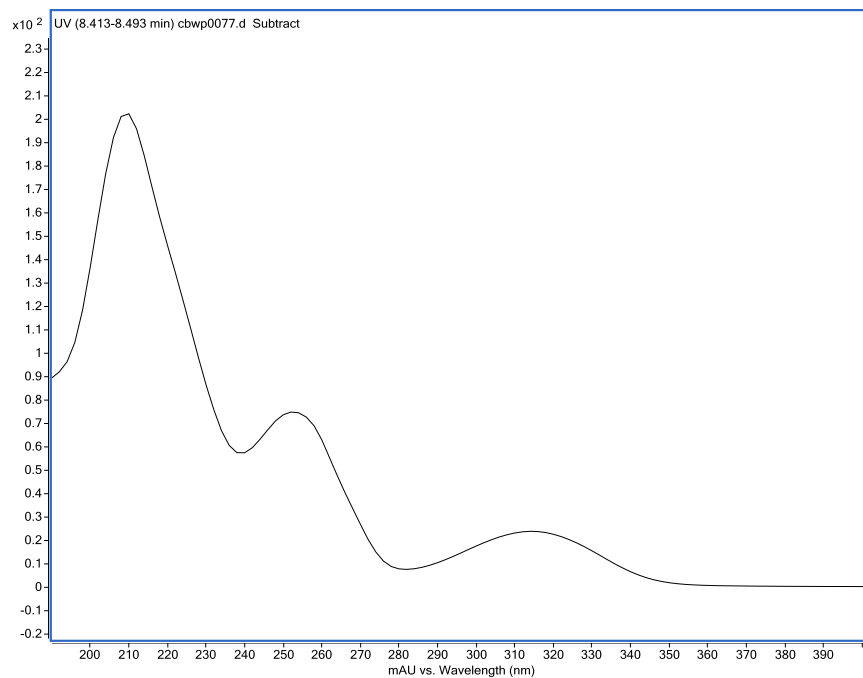


Figure S2 UV/Vis spectra of fluvibactin.

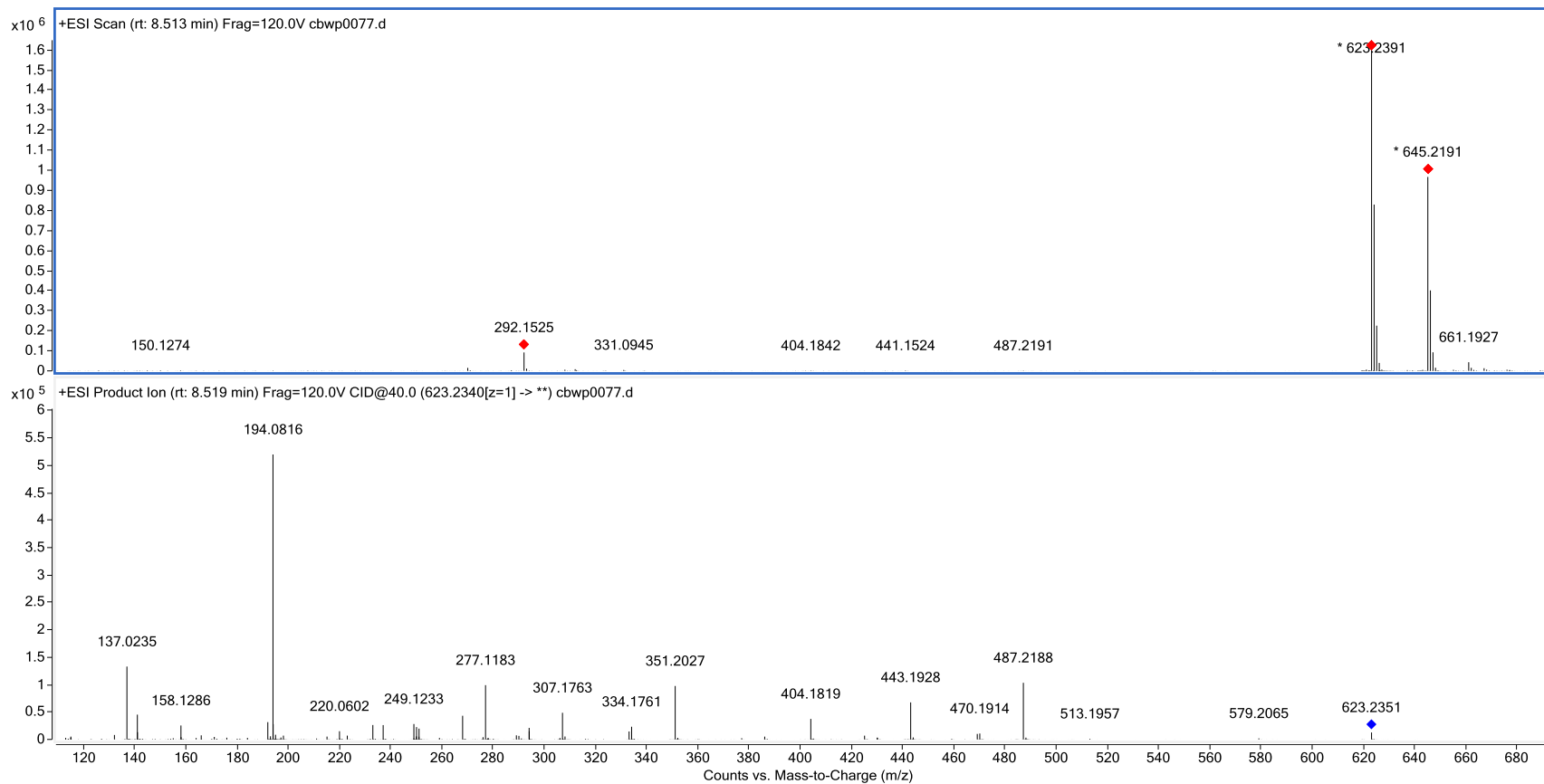


Figure S3 MS and MS/MS spectra of fluvibactin

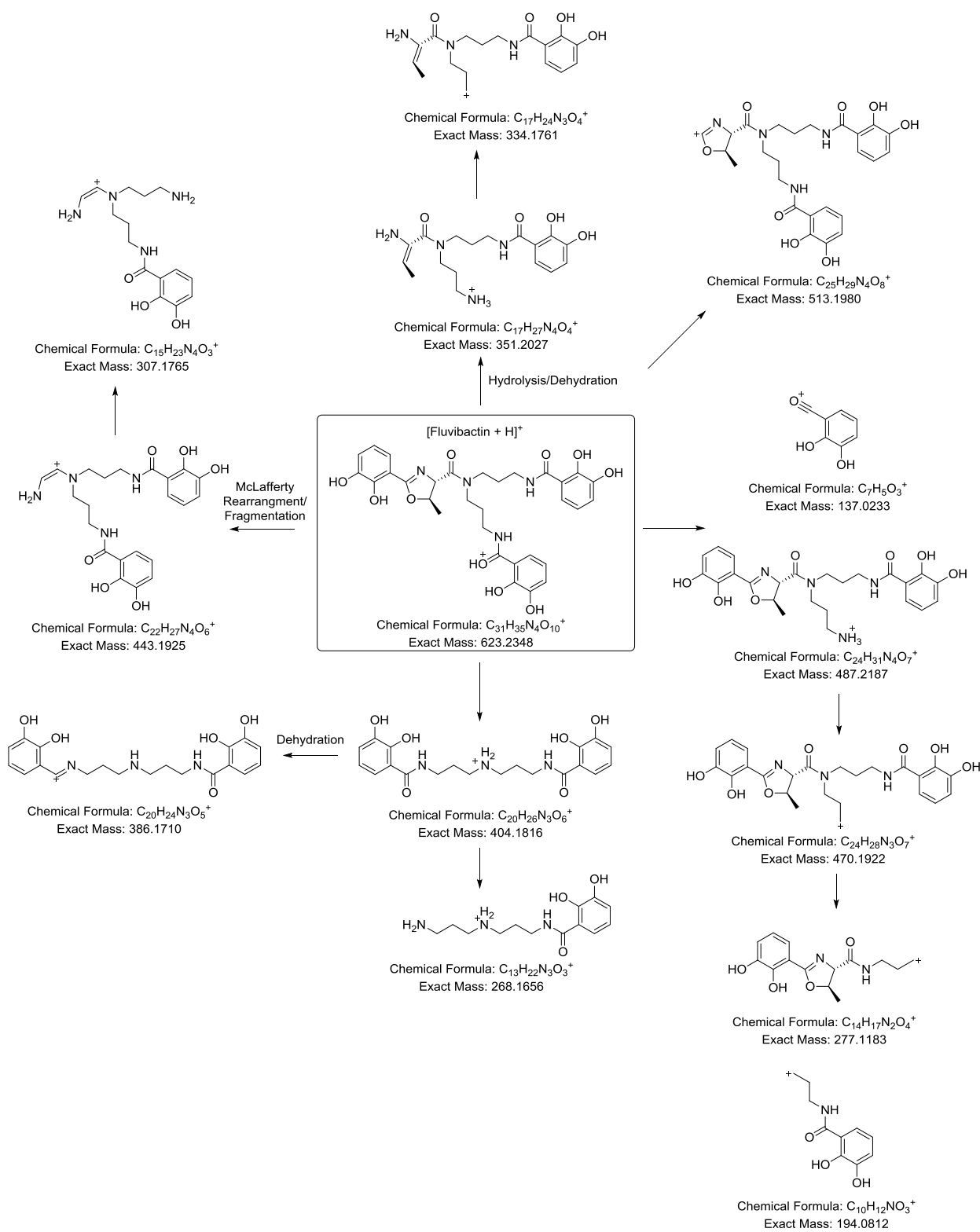


Figure S4 Proposed fragments of fluivibactin

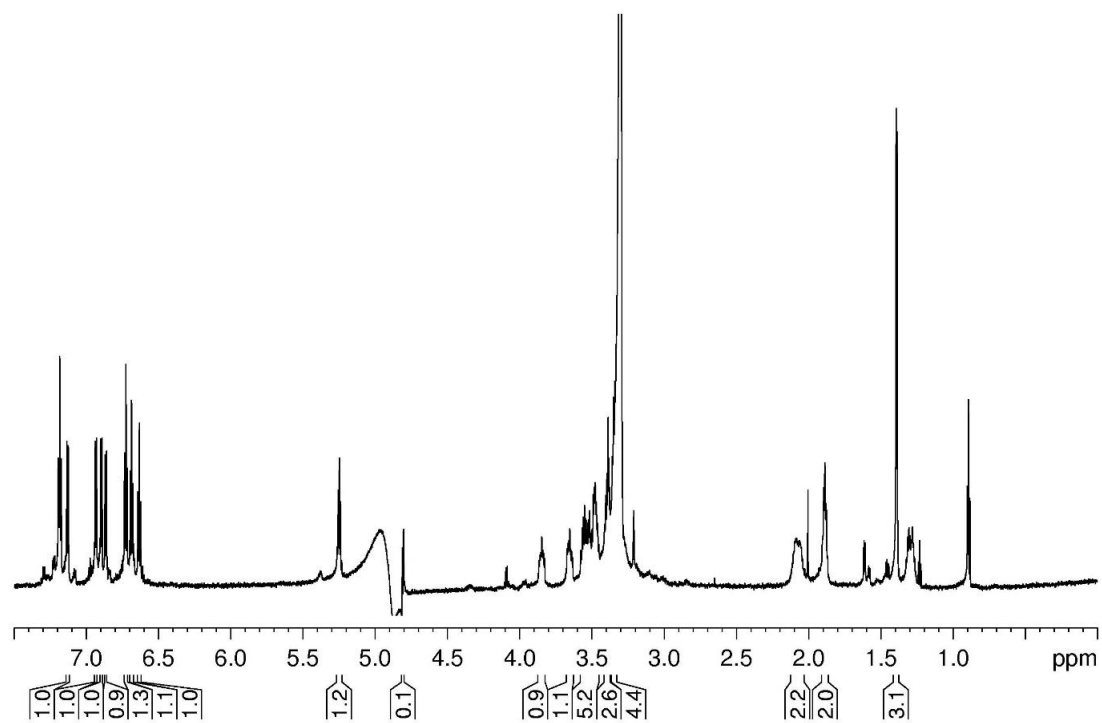


Figure S5 1D ¹H NMR spectrum of fluvibactin at 800 MHz.

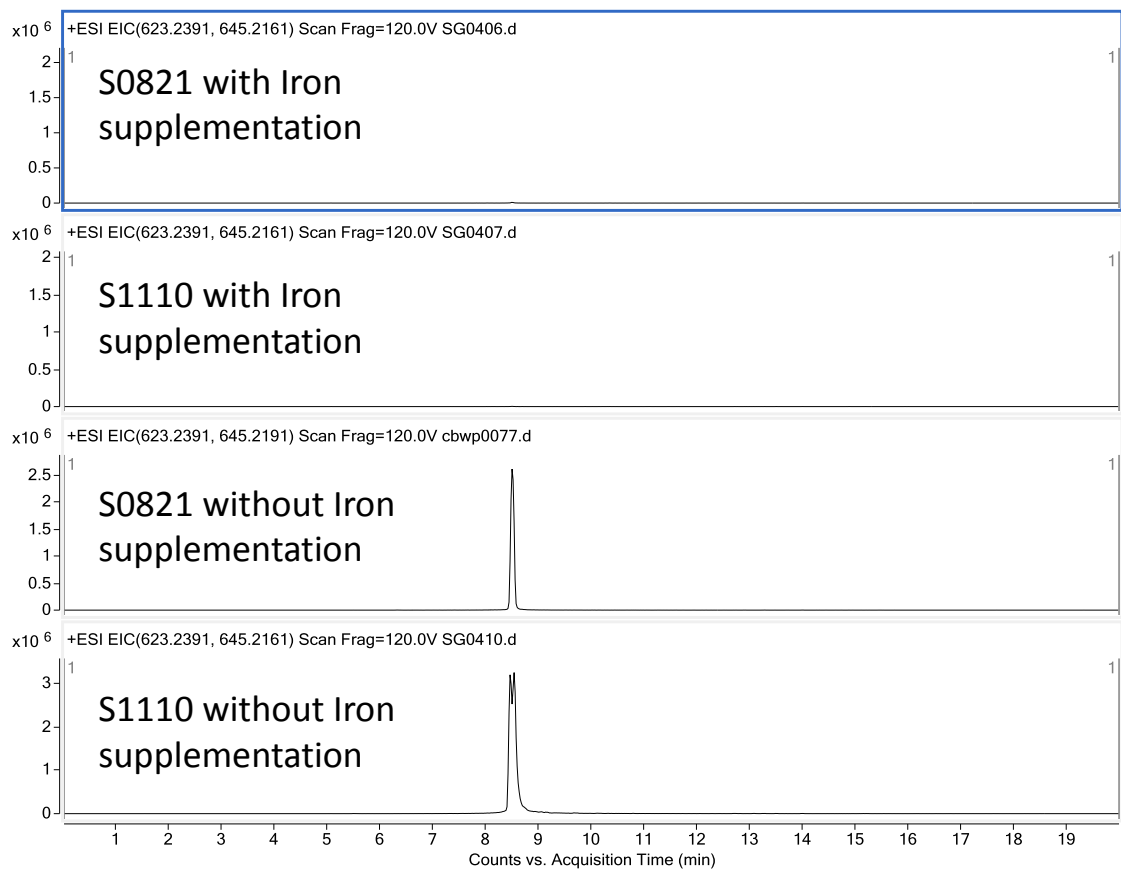


Figure S6 EIC of fluvibactin for S0821 and S1110 grown with and without iron supplementation

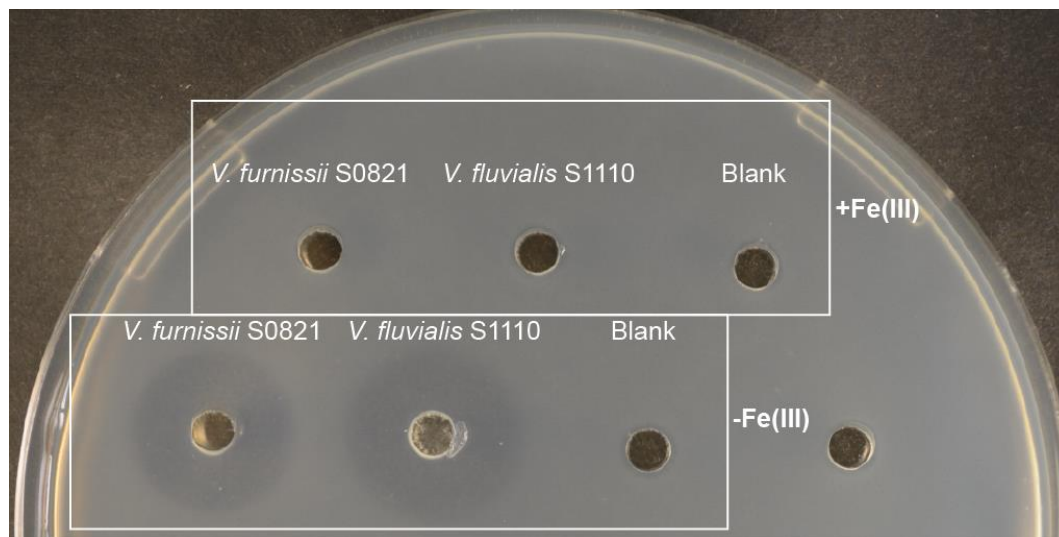


Figure S7 Test of the antibacterial activity of ethyl acetate extracts obtained from cultures of *V. furnissii* S0821 and *V. fluvialis* S1162 grown in presence (top panel) and in absence (lower panel) of Fe(III).

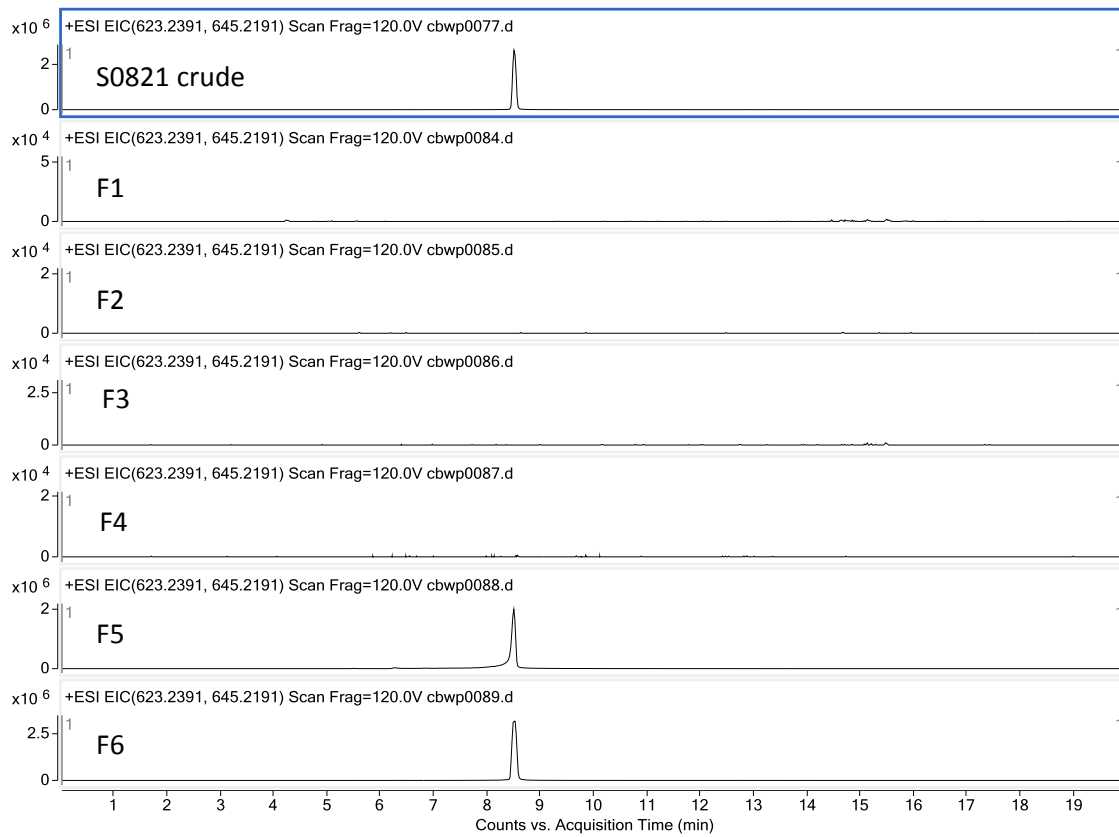


Figure S8 EIC of fluvibactin in SPE fractions of S0821 culture

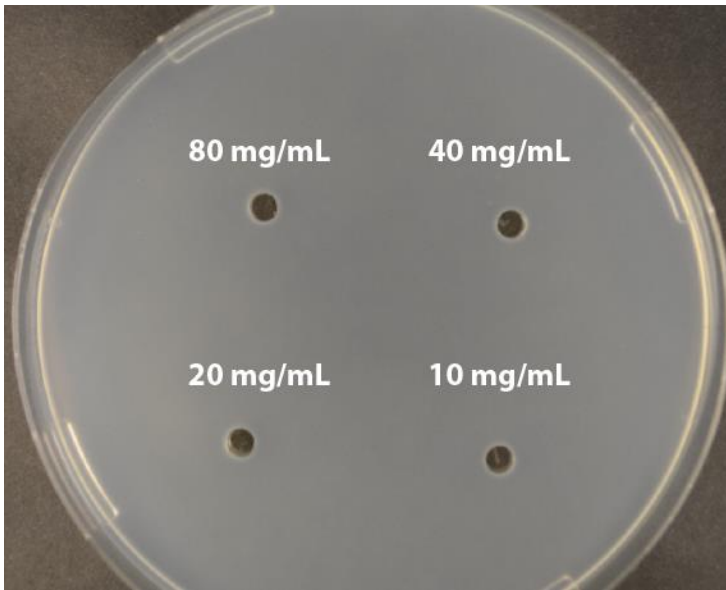


Figure SI 9 Assessment of antibacterial activity of ectoine (Fluka 81619, dissolved in sterile milliQ water). 50 μ L of 10, 20, 40, 80 mg/mL solutions were transferred to wells punched in solid medium seeded with *V. anguillarum* 90-11-287. No growth inhibition of the pathogen was observed after 48 hours of incubation of the plate at 25 °C.

Table S1 Amino acid sequences of the putative bacteriocins predicted by Bagel3 based on the analysis of the genomic sequences of *V. furnissii* S0821 and *V. fluvialis* S1110.

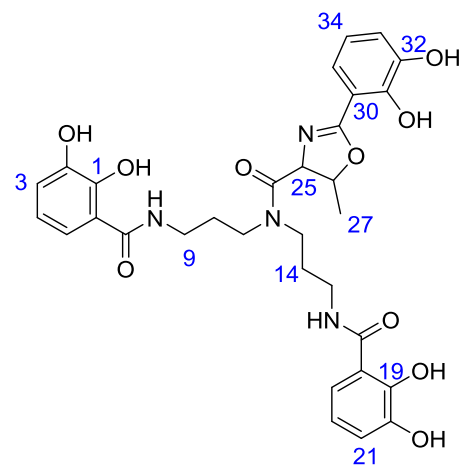
Species	Strain	Pfam	Predicted amino acid sequence
<i>V. furnissii</i>	S0821	Peptidase_M23	FNQLGFSYQELMKIMETDLNYLALDTLKPGNVLRFWRSQDGRSLAKMELK FSLVERAVYVRTDDGSFEFKDVKIPGTWKEYPLIGEIQGSFSQSANQLGLGS SDIDQIVTLKDKINFVRDVRAGDRFEVLSRQFVGDQLTGNSEIQAIKIFSR SNDVTAYLYKDGQYYDKNGESLQRAFQRYPTTGKWRLSSGFDPNRRHPVT GRIAPHNGTDFAAPGTGPVVSTGDGVVVMTRNHPYAGNYVVIQHGSTYM TRYLHLSKILVSKGQKVSQRIGLSGATGRVTGPHIHYELIVRGRPVDAMK ANIPMANSVPPKDMANFTARRNELDRMLAHQEGLLASTNSQATPES
<i>V. fluvialis</i>	S1110	Peptidase_M23	TDLNYLALDTLKPGNILRFWRGQDGHSLAKMELEFSLVERAVYARTDDGSFE FKDVKIPGKWKEYPLIGEIQGSFSQSANQLGLGSSDIDQIVSLKDKINFVR DIRAGDRFEVLSRQFVGEKMTGNSEIQAIKIFSRNEVTAYLYKDGQYYDK NGESLQRAFQRYPTTQKWRMSSGFDPNRHHPVTGRIAPHNGTDFAAPIG TPVVSTGDGVVVMTRNHPYAGNYVVIQHGSTYMTRYLHLSKILVRKGQKV SRGQRIGLSGATGRVTGPHIHYELIVRGRPVDAMKANIPMANSVPPKEMA SFVSRRNELDKMLAHQESLLASNSSPDNPES

Table S2 MS/MS fragments of Fluvibactin

Observed Mass	Predicted Formula	Assignment	Predicted Mass	Error (ppm)
645.2158	C ₃₁ H ₃₄ N ₄ O ₁₀ Na	[M+Na] ⁺	645.2167	-1.394880201
623.2342	C ₃₁ H ₃₅ N ₄ O ₁₀	[M+H] ⁺	623.2348	-0.962719027
513.1963	C ₂₅ H ₂₉ N ₄ O ₈	See figure S4	513.198	-3.312561623
487.2186	C ₂₄ H ₃₁ N ₄ O ₇	See figure S4	487.2187	-0.205246638
470.1918	C ₂₄ H ₂₈ N ₃ O ₇	See figure S4	470.1922	-0.850715941
443.192	C ₂₂ H ₂₇ N ₄ O ₆	See figure S4	443.1925	-1.128177936
404.1813	C ₂₀ H ₂₆ N ₃ O ₆	See figure S4	404.1816	-0.742240617
386.1708	C ₂₀ H ₂₄ N ₃ O ₅	See figure S4	386.171	-0.51790528
351.2023	C ₁₇ H ₂₇ N ₄ O ₄	See figure S4	351.2027	-1.138943408
334.1758	C ₁₇ H ₂₄ N ₃ O ₄	See figure S4	334.1761	-0.897730269
307.176	C ₁₅ H ₂₃ N ₄ O ₃	See figure S4	307.1765	-1.627728684
277.1182	C ₁₄ H ₁₇ N ₂ O ₄	See figure S4	277.1183	-0.360856717
268.1656	C ₁₃ H ₂₂ N ₃ O ₃	See figure S4	268.1656	0
194.0812	C ₁₀ H ₁₂ N ₃ O	See figure S4	194.0812	0
137.0234	C ₇ H ₅ O ₃	See figure S4	137.0233	0.729802888

Table SI3 NMR assignment for Fluvibactin in CD₃OD

Atom assignment	¹³ C chemical shift [ppm]	¹ H chemical shift [ppm], Integral,multiplicity, <i>J</i> [Hz]
1	150.2	-
2	147.3	-
3	119.7*	6.90, 1H, dd, 7.8, 1
4	119.6*	6.69, 1H, t, 8
5	118.6#	7.18, 1H, br. d, 8
6	116.7	-
7	171.5	-
9a	37.8 ^α	3.39, 1H, m
9b	37.8 ^α	3.35, 1H, m
10	28.4	1.89, 2H, p, 7
11a	45	3.56, 1H, m
11b	45	3.49, 1H, m
13a	46.7	3.84, 1H, m
13b	46.7	3.65, 1H, m
14a	30.3	2.09, 1H, m
14b	30.3	2.05, 1H, m
15a	37.9 ^α	3.52, 1H, m
15b	37.9 ^α	3.47, 1H, m
17	171.8	-
18	116.7	-
19	150.2	-
20	147.3	-
21	119.6*	6.86, 1H, dd, 7.8,1
22	119.6*	6.63, 1H, t, 8
23	118.6#	7.19, 1H, br. d, 8
24	171.4	-
25	73	4.81, 1H, d, 6.4
26	79.8	5.25, 1H, p, 6.4
27	20.2	1.39, 3H, d, 6.4
28	167.8	-
30	111.8	-
31	149.4	-
32	146.7	-
33	120.2	6.93, 1H, dd, 8,1.4
34	119.9 [^]	6.72, 1H, t, 8
35	119.9 [^]	7.13, 1H, dd, 8,1.4



[^],*,^α,# : indicates overlap and thereby specific assignment impossible.

All spectra were acquired on a Bruker Advance 800 MHz NMR spectrometer using standard pulse sequenced. Chemical shifts are reported in ppm relative to deuterated solvent peaks as internal standards (δH , CD_3OD 3.30 ppm; δC , CD_3OD 49 ppm). Coupling constants (J) are given in hertz (Hz). Multiplicities of ^1H NMR signals are reported as follows: d, doublet; br.d, broad doublet; t, triplet; p, pentet; m, multiplet.