

Supporting Information

Upregulation and Identification of Antibiotic Activity of a Marine-Derived *Streptomyces* sp. via Co-Cultures with Challenge Pathogens

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List of Supporting Information:

- Figure S1.** *Styela canopus*, the solitary tunicate collected from mangrove roots in Bastimentos Park, Bocas del Toro, Panama and *Streptomyces* sp. PTY087I2 a bacterium isolated from this tunicate, on YSP+IO agar plate and in YSP+IO liquid media
- Figure S2.** UV chromatogram overlays of monoculture with each co-culture demonstrating upregulation in secondary metabolite production
- Figure S3.** High resolution mass spectral confirmation of granatomycin D, granaticin, and dihydrogranaticin B production by *Streptomyces* sp. PTY087I2
- Table S1.** Granaticin biosynthetic genes from *Streptomyces* sp. PTY087I2 and the % identity to those from *S. violaceoruber* Tu22

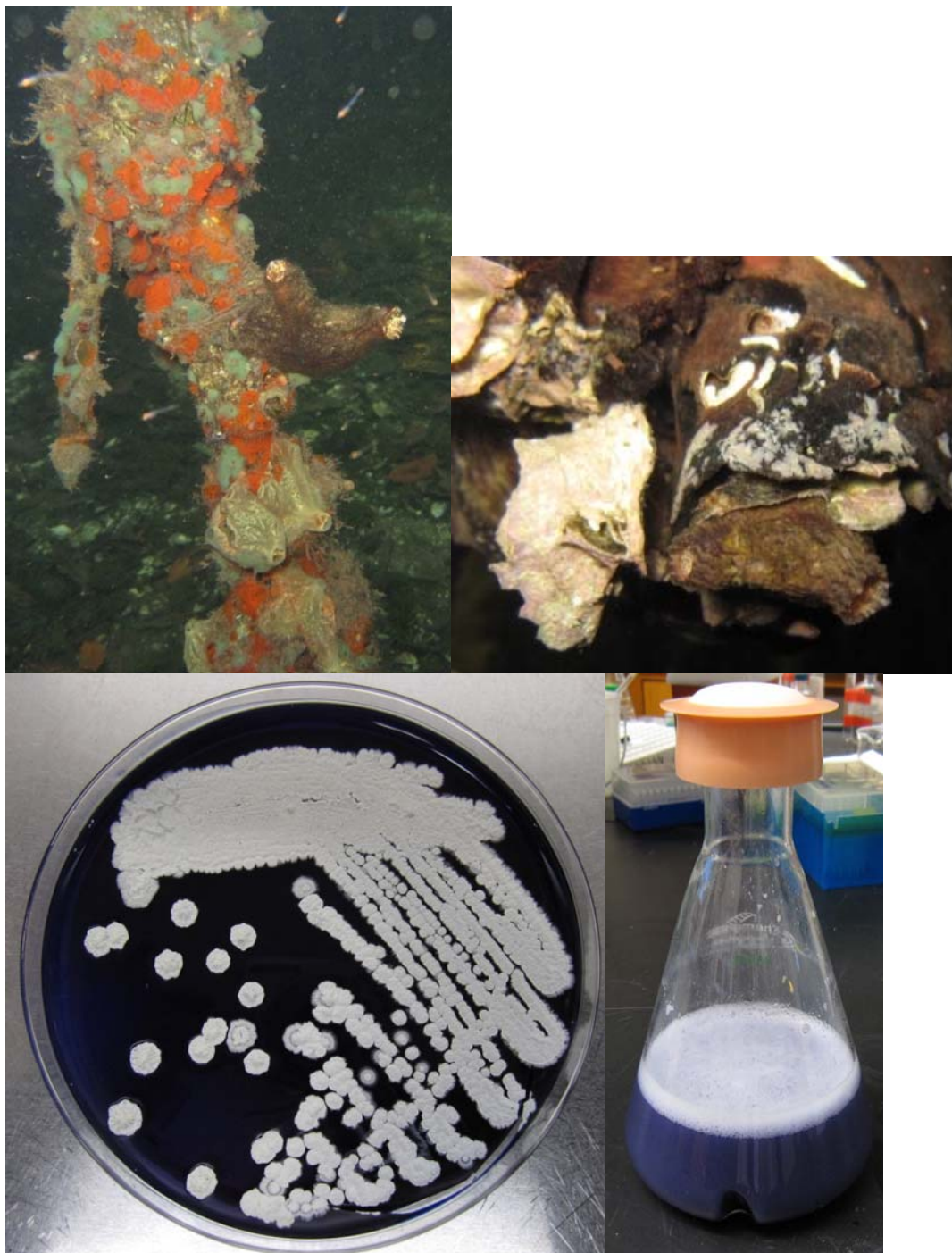


Figure S1. *Styela canopus*, the solitary tunicate collected from mangrove roots in Bastimentos Park, Bocas del Toro, Panama (top) and *Streptomyces* sp. PTY087I2 a bacterium isolated from this tunicate, on YSP+IO agar plate (left bottom) and in YSP+IO liquid media (right bottom).

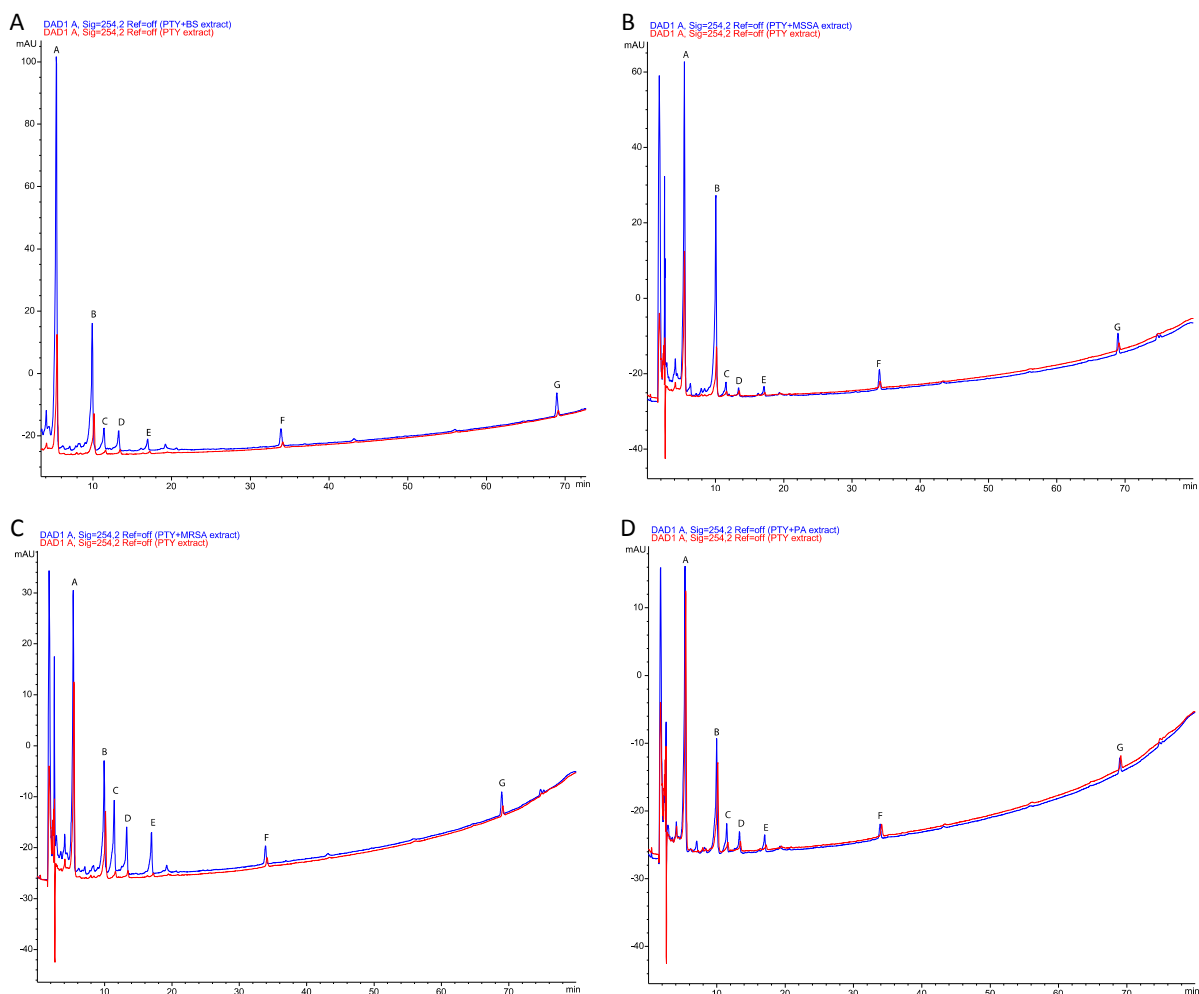


Figure S2. UV chromatogram overlays of monoculture with each co-culture demonstrating upregulation in secondary metabolite production. (A) Monoculture (red) and co-culture with *Bacillus subtilis* (BS, blue); (B) monoculture (red) and co-culture with methicillin sensitive *Staphylococcus aureus* (MSSA, blue); (C) monoculture (red) and co-culture with methicillin resistant *Staphylococcus aureus* (MRSA, blue); and (D) monoculture (red) and co-culture with *Pseudomonas aeruginosa* (PA, blue).

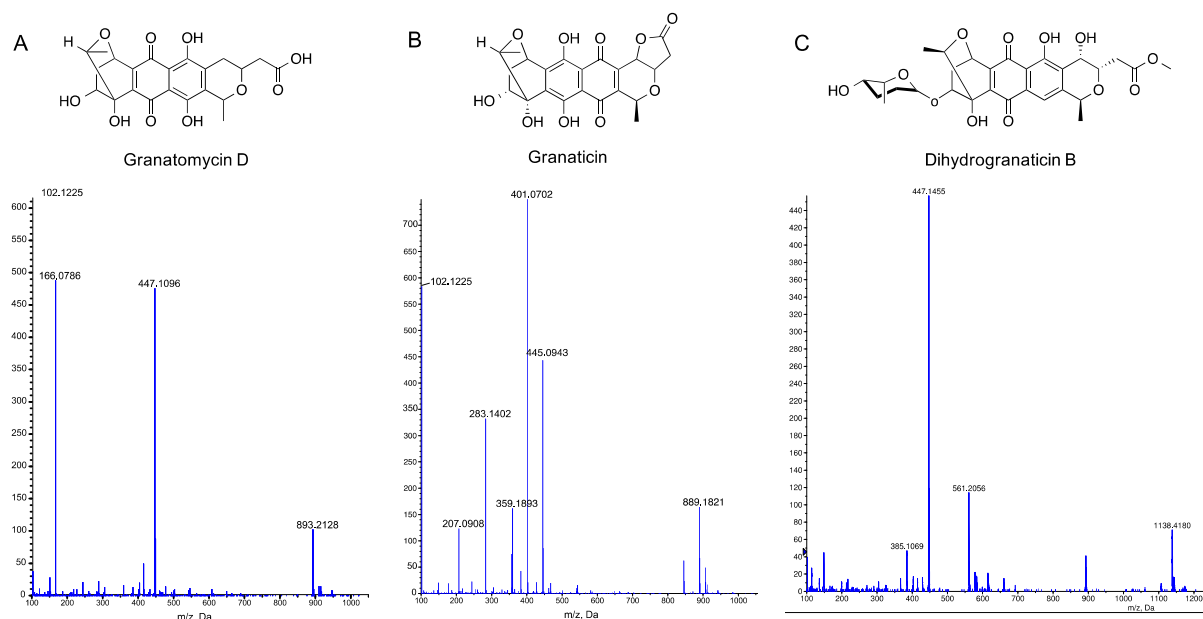


Figure S3. High resolution mass spectral confirmation of granatomecin D, granaticin, and dihydrogranaticin B production by *Streptomyces* sp. PTY08712. High resolution mass spectral data was collected on Qstar Elite system as described in the Materials and Methods. (A) Granatomecin D (stereochemistry not defined in literature) eluted at retention time (t_R) 11.4 min. The m/z of $[M+H]^+$ 447.1096 is consistent with a molecular formula of $C_{22}H_{22}O_{10}$, confirming presence of granatomecin D. (B) Granaticin eluted at t_R 13.2 min. The m/z of $[M+H]^+$ 445.0943 is consistent with a molecular formula of $C_{22}H_{20}O_{10}$, confirming presence of granaticin. (C) Dihydrogranaticin B eluted at retention time (t_R) 17.0 min. The m/z of $[M+H]^+$ 561.2056 is consistent with a molecular formula of $C_{28}H_{32}O_{12}$, confirming presence of dihydrogranaticin B.

Table S1. Granaticin biosynthetic genes from *Streptomyces* sp. PTY087I2 and their % identity to those from *S. violaceoruber* TŮ22 (ORF, gene name, and deduced role from [1]).

ORF	Gene	Deduced role	% identity
7	<i>pkaA</i>	Serine threonine protein kinase	72
8	-	-	n.p.
9	<i>actII-4</i>	Pathway-specific transcriptional activator	n.p.
10	<i>degU</i>	Response regulator of two-component system	n.p.
11	<i>degS</i>	Sensor kinase of two-component system	n.p.
12	-	-	n.p.
13	-	-	n.p.
14	<i>dnrS</i>	Glycosyl transferase	93
15	<i>actII-2</i>	Transmembrane protein	84
16	<i>strD</i>	dTDP-1-glucose synthase	84
17	<i>strE</i>	dTDP-glucose-4,6-dehydratase	84
18	<i>actVI-3</i>	Cyclase dehydratase	77
19	-	Disulphide bond forming protein	76
20	<i>soxR</i>	Transcript activator in redox control	n.p.
21	<i>actVA-5</i>	Hydroxylase	79
22	<i>dnmV</i>	dTDP-4-keto-6-deoxyhexose-reductase	80
23	<i>rfbH (ascC)</i>	CDP-4-keto-6-deoxyglucose-3-dehydratase E1	94
24	-	-	n.p.
25	<i>strM</i>	dTDP-4-keto-6-deoxyglucose-3,5-epimerase in streptomycin biosynthesis	87
26	<i>rdmF</i>	Rhodomyacin biosynthesis	n.p.
27	<i>dnmT</i>	dTDP-4-keto-6-deoxyglucose-2,3-dehydratase	91
28	<i>actVA-3</i>	unknown	n.p.
29	<i>lmbY</i>	FMN dependent monooxygenase in lincomycin biosynthesis	91
6	<i>actIII</i>	Keto reductase	94
5	<i>actIII</i>	Keto reductase for C-9	
1	<i>actI-1</i>	Keto acyl synthase	95
2	<i>actI-2</i>	Chain Length Factor	80
3	<i>actI-3</i>	Acyl carrier protein	80
4	<i>actVII</i>	First ring aromatisation (aromatase)	85
30	<i>actVA-3</i>	Unknown	n.p.
31	<i>actVI-A</i>	Unknown	n.p.
32	-	-	n.p.
33	<i>actIV</i>	Second ring cyclisation (cyclase)	83
34	<i>actVB</i>	FMN:NADH oxidoreductase	82
35	Unknown	-	n.p.
36	Unknown	-	n.p.
37	<i>nshA</i>	Transcriptional activator	n.p.
	-	tRNA-ala	92
	-	YcaO-like family protein	77

n.p. = not present in *Streptomyces* sp. PTY087I2; 1. Ichinose, K.; Bedford, D.J.; Tornus, D.; Bechthold, A.; Bibb, M.J.; Revill, W.P.; Floss, H.G.; Hopwood, D.A. The granaticin biosynthetic gene cluster of *streptomyces violaceoruber* tu22: Sequence analysis and expression in a heterologous host. *Chem. Biol.* **1998**, *5*, 647-659.