

**Table S1. Related to Figure 2. Bacteria tested in swarming bioassay.**

Species	Strain	Genotype	Live bacteria	Conditioned media	Concentrated conditioned media	Accession information
<i>Vibrio fischeri</i>	ES114	WT	+	+	+	ATCC 700601
		ΔLuxO	+	+	+	Lupp et al. 2003
		ΔAinS	+	+	+	Lupp et al. 2003
		ΔLuxI	+	+	+	Lupp et al. 2003
		ΔAinSΔLuxS	+	+	+	Lupp and Ruby 2005
		ΔLuxR	+	+	+	Lupp and Ruby 2005
		ΔSypC	+	+	+	Shibata et al. 2012
		ΔSypH	+	+	+	Shibata et al. 2012
		ΔSypI	+	+	+	Shibata et al. 2012
		ΔSypK	+	+	+	Shibata et al. 2012
		ΔSypM	+	+	+	Shibata et al. 2012
		ΔSypO	+	+	+	Shibata et al. 2012
		ΔSypQ	+	+	+	Shibata et al. 2012
<i>Vibrio fischeri</i>	MJ11		+	+	+	BAA-1741
<i>Vibrio tubiashii</i>			+	+	+	ATCC 19105
<i>Vibrio orientalis</i>			+	+	+	ATCC 33934
<i>Vibrio harveyi</i>			-	-	-	ATCC 14126
<i>Vibrio natriegens</i>			-	-	-	ATCC 8110
<i>Vibrio parahaemolyticus</i>		Ψ	Ψ	Ψ	Ψ	ATCC 17802
<i>Vibrio alginolyticus</i>		Ψ	Ψ	Ψ	Ψ	PRJNA13571
<i>Vibrio anguillarum</i>			-	-	-	ATCC 19181
<i>Vibrio ordalii</i>			-	-	-	ATCC 33509
<i>Vibrio cholerae</i>	YB1A01		n.t.	-	-	PRJNA281423
<i>Vibrio metoecus</i>	YB4D01		-	-	-	PRJNA281423
<i>Vibrio mimicus</i>			-	-	-	ATCC 33653
<i>Echinicola pacifica</i>			-	-	-	DSM 19836
<i>Echinicola pacifica</i>	From starved <i>S. rosetta</i> co-culture		n.t.	-	-	ATCC PRA-390

+: swarming observed; -: no swarming observed; Ψ: settling observed; n.t.: not tested

**Table S2. Related to Figure 2. Purified molecules tested in swarming bioassay.**

Molecule	Induces Swarming?	Source
3-O-C <sub>6</sub> -(L)-HSL	–	Cayman Chemical (10011207)
C8-HSL	–	Cayman Chemical (10011199)
Cyclic di-GMP	–	Sigma SML1228
Chondroitinase ABC	+	Proteus vulgaris (Sigma C3667)
Chondroitinase AC	+	Flavobacterium heparinum (Sigma C2780)
Chondroitinase B	–	Flavobacterium heparinum (Sigma C8058 )
Heparinase I and III	–	Flavobacterium heparinum (Sigma H3917 )
Hyaluronate lyase	–	Staphylococcus hyalurolyticus (Sigma H113)
Chitinase	–	Streptomyces griseus (Sigma C9830)
O-Glycosidase	–	Streptococcus pneumonia (Sigma G1163)
Lysozyme	–	Sigma L67876
Collagenase	–	Clostridium histolyticum (Sigma C0130)
Unsulfated chondroitin	–	Sigma C3920
Chondroitin-6-sulfate	–	Sigma C4170
Chondroitin sulfate (shark cartilage)	–	Sigma C4384

**Table S3. Related to Figure 3. Chondroitinase-induced mating in *S. rosetta***

Inducing factor	# clonal isolates	% thecate isolates (presumably diploid)	% outcrossed diploid isolates
<i>E. pacifica</i> conditioned media (5% vol/vol)	88	0	0
EroS ( <i>V. fischeri</i> , 50pM)	53	17%	15%
Chondroitinase ABC ( <i>P. vulgaris</i> , 0.0035 units)	62	21%	17%
Chondroitinase AC ( <i>F. heparinum</i> , 0.0035 units)	52	13%	11%

**Table S4. Related to Figure S4. Quantification of EroS secreted by *V. fischeri*.**

	Purified EroS	Low nutrient media	High nutrient media
MS2 Peak intensity (sequence TQITDDTYQNFFDK)	1.43E+05	9.08E+02	2.77E+03
MS2 Peak intensity (1 pMol standard)	1.07E+04	2.24E+04	1.86E+04
pMol EroS in excised gel band	13.4	0.0405	0.149
Sample volume in gel band (μL)	2.0	18.0	6.0
[EroS] loaded on gel	6.7 μM	2.3 nM	25 nM
[EroS] in culture supernatant		6.8 pM	75 pM

**Table S5. Related to Figure S4. Vibrio-induced swarming is not observed in other tested choanoflagellate species**

	<i>V. fischeri</i>	<i>V. tubiashii</i>	<i>V. orientalis</i>	<i>P. vulgaris</i>
Choanoflagellate species	Live bacteria Conditioned media EroS	Live bacteria Conditioned media	Live bacteria Conditioned media	ABC chondroitinase
<i>S. rosetta</i>	+++	++	++	+
<i>S. roanoaka</i>	---	--	--	-
<i>S. infusionum</i>	---	--	--	-
<i>S. minuta</i>	---	--	--	-
<i>S. pyxidium</i>	---	--	--	-
<i>M. brevicollis</i>	---	--	--	-

+: swarming observed; -: no swarming observed