



FIG S1. Bacterial killing requires contact. Chloramphenicol resistant *E. coli* prey were incubated with *V. cholerae* predator strains separated by a 0.22- μ m filter to measure contact dependence of killing. Prey alone (“None”), isogenic *E. coli* control, and negative (“C6706 T6SS”) and positive (“C6706 maximum killer”) controls are represented by white bars. Shown are average prey survival values \pm standard deviation after triplicate encounters of *E. coli* prey with *V. cholerae* clinical isolates (black bars) and environmental isolates (grey bars) for all isolates that demonstrated constitutive killing previously. One representative experiment is shown of three performed.

TABLE S1: List of strains used in this study and the lab from which they were acquired

Strains used in this study	Source
C6706	Lab stocks
C6706 TfoX*	
C6706 Δ <i>cytR</i>	
C6706 TfoX* HapR* QstR* CytR ⁺ (C6706 maximum killer)	
C6706 TfoX* HapR* QstR* CytR ⁺ Δ <i>vasK</i> (C6706 T6SS ⁻)	
*denotes constitutive expression of corresponding gene	
2010EL-1749, 2012V-1001, 3582-05, 3566-08, 3541-04, 3558-04, 2011V-1043, 3500-05, 3546-06, 2011EL-1141, 3554-08, 2009V-1085, 2009V-1096, 2011EL-1137, 2009V-1046, 2011EL-1938, Nepal 25, 2010EL-1786, Nepal 14, 2010EL-1941, 2010V-1014, 2011EL-1939, 2633-78, 2631-78, E8498, 2479-86, 3223-74, 3272-78, 1074-78, 2559-78, 2512-86, 3225-74, 692-79, 1496-86, 2497-86, 2523-87	C. Tarr and C. Bopp, Centers for Disease Control and Prevention
NCTC8457, MZO-2, MAK757, CA401, O395, MO10, 857, 2740-80	J. Zhu, University of Pennsylvania
VC56, VC22, VC53, VC48	A. DePaulo, Food and Drug Administration
SIO, TP	D. Bartlett, Scripps Institution of Oceanography
HE46	R. Colwell, University of Maryland

TABLE S2. Sequences of primers used in cloning experiments during this study.

Primer	Sequence, 5'→3'
pQT_1	AGTTGTCAGCAGCGTTAAGTTAAAGGATCCAGGAGCTAAGGAAGCTAAAATGCA ACGAGCCAACCTATGC
pQT_2	ATAAAGCTTGCTCAATCAATCACCTTACAGTAAATTTTGATTAACCCAAGC
pQT_3	TTGACAATTAATCATCGGCTCG
pQT_4	TTTTTTCACAAAACGGTTTACAAGC
vasK_1	AATTCCCGGGAGAGCTCGATATCGGGATGAGTTACACCATTGCC
vasK_2	AAAACCTCGCTCTGGCAACCTAGAATTGTGTCCTTGTTTACTCTG
vasK_3	CAGAGTAAACAAGGACACAATTCTAGGTTGCCAGAGCGAGTTTT
vasK_4	CAGGTCGACGGATCCCAAGCCAGCTGATGTAACCGCTCG
vasK_5	CCGAGTTTAGTGGAACAAATTC
vasK_6	CAACTTCGTCCTAAAAAGCCAG

TABLE S3. Pearson correlation matrix generated from principal component analysis (PCA) on clinical and environmental data, clinical data only, or environmental data only. Each variable was assessed for its correlation with all other variables. Variables include clinical (C) or environmental (E), year of isolation, serogroup, CTX status, chitinase activity (Chi), natural transformation (TF), and fold reduction in prey survival due to contact-dependent constitutive bacterial killing (Killing). Values in bold represent a correlation between variables that are different from 0 with a significance level $\alpha \leq 0.05$.

	Variables	C/E	Year	Serogroup	CTX	Chi	TF	Killing
Clinical and Environmental	C/E	-----	-0.252	0.351	-0.778	-0.105	0.088	0.212
	Year	-0.252	-----	0.230	0.170	0.206	-0.106	-0.161
	Serogroup	0.351	0.230	-----	-0.309	-0.168	-0.002	-0.051
	CTX	-0.778	0.170	-0.309	-----	0.146	-0.157	-0.238
	Chi	-0.105	0.206	-0.168	0.146	-----	0.069	0.047
	TF	0.088	-0.106	-0.002	-0.157	0.069	-----	0.072
	Killing	0.212	-0.161	-0.051	-0.238	0.047	0.072	-----
Clinical only	Year	N/A	-----	0.261	-0.025	0.373	-0.030	0.008
	Serogroup	N/A	0.261	-----	-0.212	0.205	0.042	0.205
	CTX	N/A	-0.025	-0.212	-----	-0.036	-0.366	-1.000
	Chi	N/A	0.373	0.205	-0.036	-----	0.013	0.015
	TF	N/A	-0.030	0.042	-0.366	0.013	-----	0.681
	Killing	N/A	0.008	0.205	-1.000	0.015	0.681	-----
Environmental only	Year	N/A	-----	0.641	-0.089	-0.547	-0.231	-0.280
	Serogroup	N/A	0.641	-----	-0.005	-0.334	-0.090	-0.174
	CTX	N/A	-0.089	-0.005	-----	0.155	-0.048	-0.128
	Chi	N/A	-0.547	-0.334	0.155	-----	0.123	0.088
	TF	N/A	-0.231	-0.090	-0.048	0.123	-----	0.071
	Killing	N/A	-0.280	-0.174	-0.128	0.088	0.071	-----