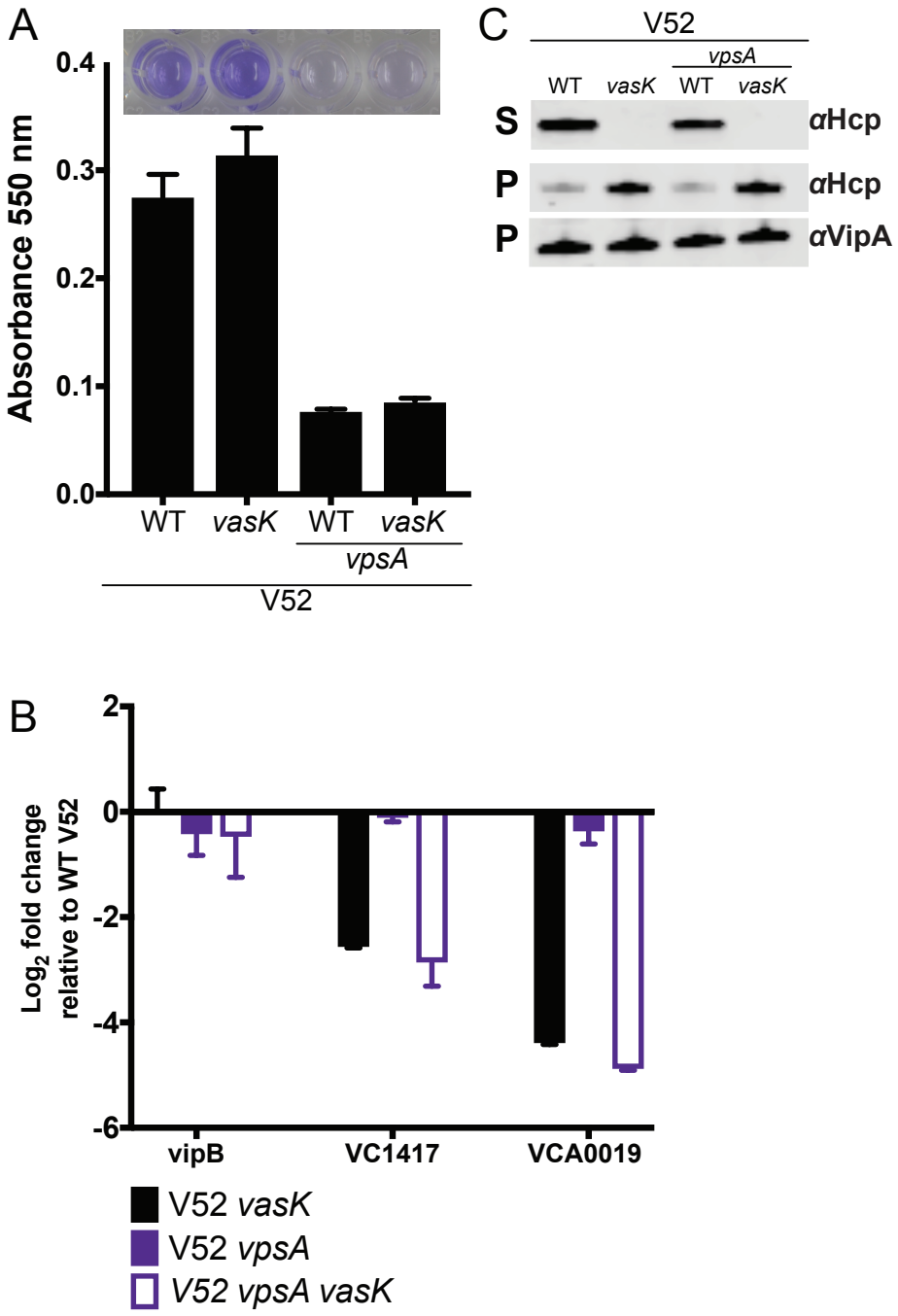

Figure S1. Deletion of vpsA from V. cholerae impairs biofilm formation independent of T6SS functionality. Data A-B represent the mean ± standard deviation of two biological replicates. (A) Crystal violet staining of vpsA mutants in wild type V52 and V52 vasK (T6SS–). (B) Expression of indicated genes in vpsA mutant backgrounds were measured by qRT-PCR with respect to wild type V52. (C) Supernatant (S) and cell pellets (P) were collected for wild type and vpsA mutants in the presence and absence of a functional T6SS and analyzed by western blot using anti-Hcp antibody. Blot with anti-vipA is a loading control.

Table S1. Strains used in this study

Organism	Genotype	Plasmid	Relevant Characteristics	Reference
V. cholerae				
2740-80	ClpV-mCherry2		Parental strain, lacZ$^-$, Str^r	(1)
	lacZ$^-$, Str^r, ΔvipA ClpV-mCherry2		*vipA*(VCA0107) in frame deletion, lacZ$^-$, Str^r	(1)
	lacZ$^-$, Str^r, ΔvpsA ClpV-mCherry2		*vpsA* (VC1917) in frame deletion, lacZ$^-$, Str^r	this study
	lacZ$^-$, Str^r, ΔvpsA ΔvipA ClpV-mCherry2		*vpsA* (VC1917) in frame deletion in *vipA* background, lacZ$^-$, Str^r	this study
	lacZ$^-$, Str^r, ΔflaA ClpV-mCherry2		*flaA* (VC2188) in frame deletion, lacZ$^-$, Str^r	this study
	lacZ$^-$, Str^r, ΔmotX ClpV-mCherry2		*motX* (VC2601) in frame deletion, lacZ$^-$, Str^r	this study
	lacZ$^-$, Str^r, ΔflaA ΔvpsA ClpV-mCherry2		*vpsA* (VC1917) in frame deletion in *flaA* background, lacZ$^-$, Str^r	this study
V52	Δrhh		*rtxA hlyA hapA* parental strain, Str^r	(2)
	Δrhh ΔvasK		*vasK* (VCA0120) in frame deletion, Str^r	(2)
	Δrhh ΔvpsA		*vpsA* (VC1917) in frame deletion, Str^r	this study
	Δrhh ΔvpsA ΔvasK		*vpsA* in frame deletion in *vasK* background, Str^r	this study
	Δrhh ΔflaA		*flaA* (VC2188) in frame deletion, Str^r	this study
	Δrhh ΔmotX		*motX* (VC2601) in frame deletion, Str^r	this study
	Δrhh	pBAD33	parental strain with empty pBAD33 plasmid, Str^r, Cm^r	this study
	Δrhh ΔvpsA	pBAD33	*vpsA* deletion in parental strain with empty pBAD33 plasmid, Str^r, Cm^r	this study
	Δrhh ΔVgrG-3ΔVCA0124		*VgrG3-VCA0124* in frame deletion, Str^r	(2)
	Δrhh ΔVC1417-VC1421		*VC1417-VC1421* in frame deletion, Str^r	(2)
	Δrhh ΔVCA0019-VCA0021		*VCA0019-VC0021* in frame deletion, Str^r	(2)
	Δrhh ΔVgrG-3ΔVCA0124 ΔvpsA		*vpsA* in frame deletion in *VgrG3-VCA0124* background, Str^r	this study
	Δrhh ΔVC1417-VC1421 ΔvpsA		*vpsA* in frame deletion in *VC1417-VC1421* background, Str^r	this study
	Δrhh ΔVCA0019-VCA0021 ΔvpsA		*vpsA* in frame deletion in *VCA0019-VC0021* background, Str^r	this study
	Δrhh ΔVgrG-3ΔVCA0124	pCR-XL-TOPO	*VgrG3-VCA0124* in frame deletion with empty pCR-XL-TOPO plasmid, Str^r, Kan^r	this study
	Δrhh ΔVC1417-VC1421	pCR-XL-TOPO	*VC1417-VC1421* in frame deletion with empty pCR-XL-TOPO plasmid, Str^r, Kan^r	this study
	Δrhh ΔVCA0019-VCA0021	pCR-XL-TOPO	*VCA0019-VCA0021* in frame deletion with empty pCR-XL-TOPO plasmid, Str^r, Kan^r	this study

	Δrhh ΔVgrG-3ΔVCA0124 ΔvpsA	pCR-XL-TOPO	*vpsA* deletion in *VgrG3-VCA0124* background with empty pCR-XL-TOPO plasmid, Str^r, Kan^r	this study
	Δrhh ΔVC1417-VC1421 ΔvpsA	pCR-XL-TOPO	*vpsA* deletion in *VC1417-VC1421* background with empty pCR-XL-TOPO plasmid, Str^r, Kan^r	this study
	Δrhh ΔVCA0019-VCA0021 ΔvpsA	pCR-XL-TOPO	*vpsA* deletion in *VCA0019-VCA0021* background with empty pCR-XL-TOPO plasmid, Str^r, Kan^r	this study
	Δrhh ΔrbmA		*rbmA* (VC0928) in frame deletion, Str^r	this study
	Δrhh ΔrbmC		*rbmC* (VC0930) in frame deletion, Str^r	this study
	Δrhh Δbap1		*rbmA* (VC1888) in frame deletion, Str^r	this study
	ΔvgrG3	pNucSe1	*vgrG3* in frame deletion with NucSe1 expressed on pBAD33	(3)
	ΔvgrG3 ΔvipA	pNucSe1	*vgrG3 vipA* in frame deletion with NucSe1 expressed on pBAD33	(3)
E. coli				
	MG1655		Gm^r	(1)
A. baylyi				
	ADP1		ATCC 33305 parental strain	(4)
	ADP1 ΔT6SS		T6SS deletion, Kan^r	(4)
P. aeruginosa				
	PA01 ΔretS ClpV-GFP		parental strain, Irg^r	(1)

Table S2. Plasmids used in this study

Plasmid	Relevant Characteristics	Source
pDS132	suicide vector used for allelic exchange, sacB, Cm^r	(5)
pWM91	suicide vector used for allelic exchange, sacB, $Carb^r$	(6)
pCR-XL-TOPO	TOPO cloning vector, used to make strains kanamycin resistant, Kan^r	Invitrogen
pBAD33	expression vector, Cm^r	(1)
pWM91-ΔvpsA	*vpsA* suicide vector, $Carb^r$	this study
pDS132-ΔflaA	*flaA* suicide vector, Cm^r	this study
pDS132-ΔmotX	*motX* suicide vector, Cm^r	this study

References

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