

Supporting information

Table S1. Function and operon prediction of HapR regulated genes.

Gene ID	Homolog	Putative function	Operon structure ^a	Regulation by HapR (from this study)	Reference
Motif 1					
VC0166	transcriptional regulator, TetR family	Regulation of multidrug efflux pump	VC0166- VC0164	Repressed	(Bina et al. 2008)
VC0432	malate dehydrogenase	Energy metabolism:TCA cycle	-	Repressed	(Byun et al. 1999)
VC0900	GGDEF family protein	Regulation of biofilm formation	-		(Waters et al. 2008)
VC1000	acetyl-CoA carboxylase	Fatty acid and phospholipid metabolism	-	Repressed	(Liu et al. 2008)
VC1181	ATP-binding protein	cytochrome assembly	VC1181-VC1180	Repressed	(Cruz-Ramos et al. 2004)
VC1222	integration host factor, alpha subunit	Regulation of virulence genes	VC1221-VC1222	Repressed	(Stonehouse et al. 2008)
VC1415	hcp protein	Type VI secreted protein	-	Repressed	(Pukatzki et al. 2006)
VC2370	sensory box/GGDEF family protein	Regulation of biofilm formation	VC2372-VC2369		(Waters et al. 2008)
VC2635	Penicillin-binding protein	peptidoglycan biosynthetic process	-	Repressed	(Sengupta et al. 1990)
VC2762	UDP-N-acetylglucosamine pyrophosphorylase	Cell envelope Biosynthesis	-	Activated	(Mengin-Lecreulx and van Heijenoort 1994)
VCA0017	hcp protein	Type VI secreted protein	-	Repressed	(Pukatzki et al. 2006)
VCA0080	GGDEF family protein	Regulation of biofilm formation	-		(Waters et al. 2008)
VCA0182	sigma-54 dependent transcriptional regulator	Regulation of NO reduction	-	Repressed	(Rodionov et al. 2005)
VCA0183	ferrisiderophore reductase/Flavo-haemoglobin	NO reduction	-	Repressed	(Rodionov et al. 2005)
VCA0246	Protein-N(pi)-phosphohistidine--sugar phosphotransferase	Sugar transport	VCA0246-VCA0244	Repressed	(Beutler et al. 2000)
VCA0247	DeoR family transcriptional regulator	Sugar transport	VCA0247-VCA0248	Repressed	(Beutler et al. 2000)
Motif 2					
VC0089	Cytochrome c peroxidase	Energy metabolism: Electron transport	-	Activated	(Partridge et al. 2007)
VC0583	transcriptional regulator HapR	Quorum sensing master regulator			(Zhu et al. 2002)
VC0934	capsular polysaccharide biosynthesis glycosyltransferase	Biofilm formation	- ^b	Repressed	(Yildiz and Schoolnik 1999)
VC1213	Response regulator VarA	Regulating quorum sensing	VC1213-VC1215	Repressed	(Lenz et al. 2005)
VC2035	conserved hypothetical protein	Multiple antibiotic resistance		Repressed	(Sulavik et al. 1994)
VC2647	Virulence regulator AphA	Virulence activation		Repressed	(Skorupski and Taylor 1999)
VCA0148	TagA-related protein	unknown		Activated	
VCA0684	Sugar phosphate permease	Sugar transport	VCA0682-VCA0684	Activated	(Verhamme et al. 2001)
VCA0865	hemagglutinin/protease HapA	proteolysis		Activated	(Jobling and Holmes 1997)
VCA0880	Hypothetic protein	unknown	VCA0880-VCA0883	Activated	(Zhu and Mekalanos 2003)

a. Operon structure prediction is based on VIMSS operon prediction from <http://www.microbesonline.org>.

b. located between two large vps operons.

Figure Legends

Fig. S1. Alignment highlighting similarity between proposed 16 bp HapR binding sites in *hapR* and *aphA* promoters. Note that the binding sites appear weakly conserved when the direct strands are compared but show strong similarity when the proposed binding site on the direct strand for *aphA* is aligned with the corresponding binding site on the complementary strand upstream of *hapR*.

Fig. S2. Schematic representation of procedure for identification of novel targets of HapR in *V. cholerae*. Note that while experimental validation can be used to iteratively alter the binding motif definitions for both motifs, Motif 1 was not modified based on experimental feedback in the present study since all the predictions were validated.

Fig. S3. Sequence logos corresponding to alignments of 22 bp binding regions for Motif 1 and Motif 2 sites. Since Motif 1 is variable in length, it shows minimal conservation beyond the initial sequence even though almost all the sites have the concluding sequence AATAG. The logo also highlights the lack of conservation for Motif 2 binding sites beyond 16 bp.

References

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		-82		-67		Similarity
5'→3'	aphA		TTATTGAGAATAATGT			
5'→3'	hapR		ACAAAATAATCATTAG			4/16
		+18		+33		
5'→3'	aphA	-82	TTATTGAGAATAATGT	-67		
3'→5'	hapRc	+33	CTAATGATTATTTTGT	+18		10/16

Fig. S1. Tsou, et al.

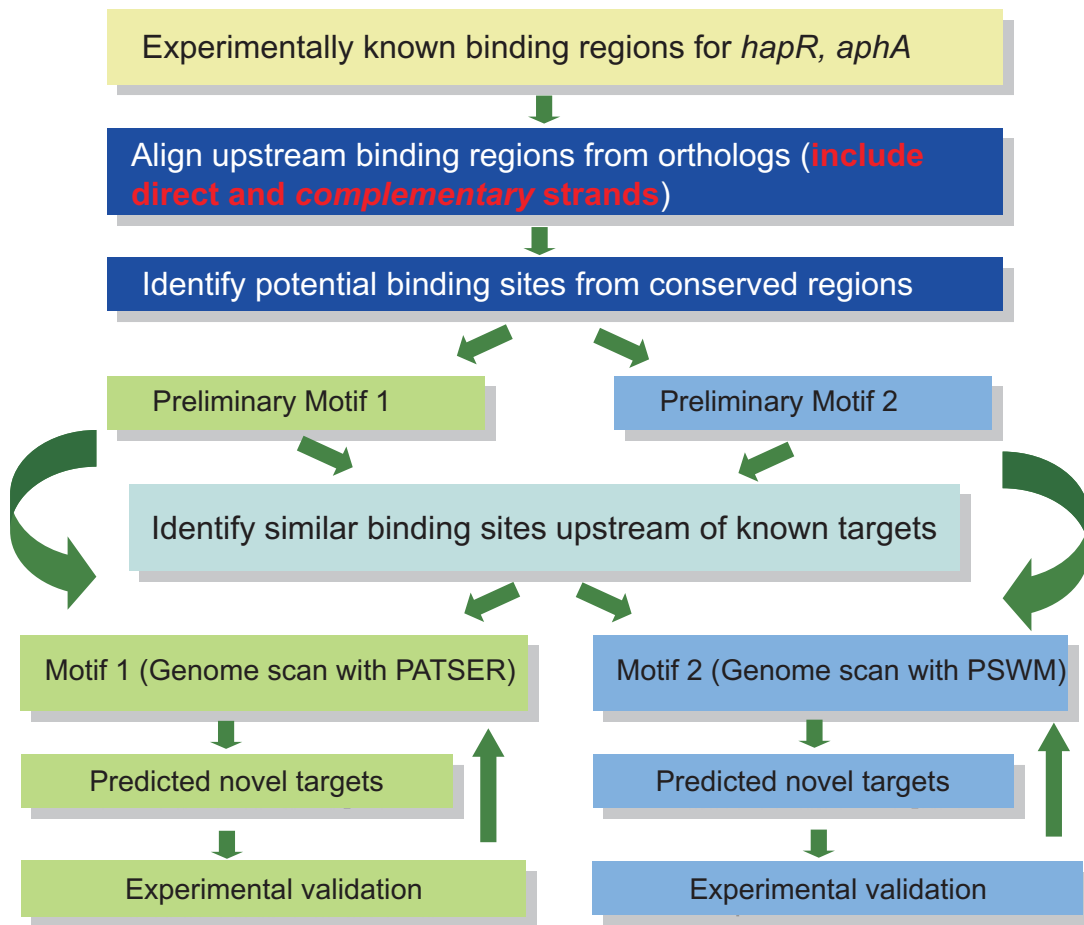


Fig. S2. Tsou, et. al.

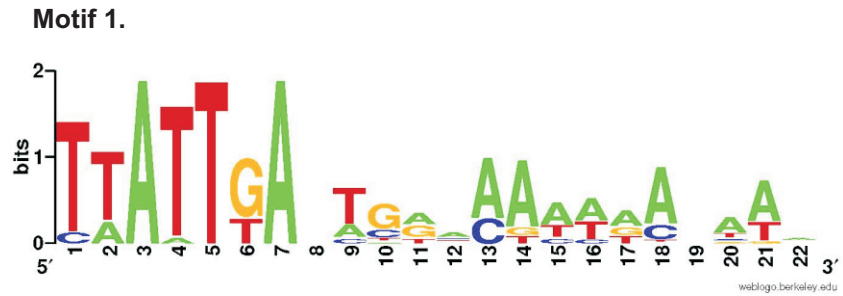


Fig. S3. Tsou, et al.