

Table SI 1. List of the primers and amplicon sizes in real-time PCR analysis

Gene	Primers name	Primers sequences (5'-3')	Amplicon size (bp)
Metabolic pathway			
<i>eda</i>	edaF	GATGTCGCCCACGCACTG	186
	edaR	CGCCGCAATCGCTTCAT	
<i>edd</i>	eddF	AAAAAGCCGGATTCAAAGTCG	151
	eddR	CAATCCAAGCGCAGCAAATC	
<i>gnd</i>	gndF	TGGCGTGGTGGTTGTATC	168
	gndR	GCCGACTTCCAGCGACTT	
<i>tka</i>	tkaF	CGCCTACGCAAAAACTCA	156
	tkaR	CTGCGCTCGGTGTCGTA	
<i>pfk</i>	pfkF	CACTTGGCGCAGGTTTAG	174
	pfkR	CTTTTCGCGCACTTCAA	
<i>fba</i>	fbaF	TGTGGACGCTGGGATTAT	152
	fbaR	GCGCCGCGTTTATGAC	
<i>aceB</i>	aceBF	CTCGGCATTCCAGACG	180
	aceBR	AATCGCATCACGCAGGTT	
<i>aceA</i>	aceAF	CGTACGCCGCATCAATAA	180
	aceAR	GCGCCCCGTTCAATCA	
Glycogen synthesis			
<i>glgA</i>	glgAF	TGCGCTGCCTGCTTATCG	165
	glgAR	GCCGGGCGGTCAAAAT	
Virulence			
<i>ctxA</i>	ctxAF	TTGTTAGGCACGATGATG	163
	ctxAR	GGACTGTATGCCCTAAT	
<i>hapA</i>	hapAF	GCCAGATTTAACGAAAG	192
	hapAR	CTAGCTTACAATGCAAG	
<i>hlyA</i>	hlyAF	CTGCGCACCTTGATGTTG	171
	hlyAR	GTTGCGCACGGGAGAA	
<i>ompU</i>	ompUF	AACAAACGCTGCCAAGTA	178
	ompUR	GAACAGTCCTGCGAAGTA	
<i>ompT</i>	ompTF	GGAACCGGACAAGGAAT	157
	ompTR	CGTAGCTGACCGTAAAA	
Colonization			
<i>tcpA</i>	tcpAF	TGCAATTATTAACAGC	159
	tcpAR	CAATCGCACGCTGAGC	
<i>mshA</i>	mshAF	GCGTTGCAATCGTCTACA	151
	mshAR	TTCCAGCAACCCCAGTCT	
<i>acf</i>	acfF	AAGCCTGGTATCGGAATC	177
	acfR	ATCAACAAACGCCTTACG	
Pathogenic regulation			
<i>tcpP</i>	tcpPF	GTATGTCCGCGTGATTTA	167
	tcpPR	AGGGGGCAGGATGATA	
<i>tcpH</i>	tcpHF	GGGCGCTACAGGTCTATT	171
	tcpHR	CCTTGTGGATCGGTCTGG	
<i>toxT</i>	toxTF	TGGTGAATTAAGAACGAA	166
	toxTR	GCTAGCAAACCCAGACT	
<i>toxR</i>	toxRF	CTTCCCCTCGCAGTATTA	177
	toxRR	CTTGAGCCCACCAGTATG	
<i>toxS</i>	toxSF	ATCTCCGAATTTGGTGAA	160
	toxSR	CTCGACGACTTTGTTGTG	
<i>aphA</i>	aphAF	AGCGCTTTAGGTGAGTGG	177
	aphAR	AGCGGCTTCGATTTCTG	
<i>aphB</i>	aphBF	TCATCCGCAACCGACTCA	160
	aphBR	TGCGCTGCGAACAACATT	
<i>hns</i>	hnsF	TTGAAGAAGCGTTAGATA	176
	hnsR	TTAGTTTCGCCTGATAG	

Biofilm regulation

<i>cya</i>	<i>cyaF</i>	AACGCCAGCTGACAATCC	174
	<i>cyaR</i>	TTCGCCCAATCGGTAATG	
<i>crp</i>	<i>crpF</i>	TTGCACGTCAGCCAGATG	180
	<i>crpR</i>	AGCGAGTGCCGTAACCA	
<i>cdg</i>	<i>cdgF</i>	AGGCGCAGGTGGAGTC	170
	<i>cdgR</i>	ACCGGGAGGCAACAAC	
<i>chiA</i>	<i>chiAF</i>	ACGGCGGCTGGAATAACG	163
	<i>chiAR</i>	CCTTGGGCGAGCAGCAGT	
<i>wecB</i>	<i>wecBF</i>	AGCCTGGGCAAACACTCA	178
	<i>wecBR</i>	CTCAAACCCGCTTCAACA	
<i>hapR</i>	<i>hapRF</i>	TGCGAAAGAGGTCATCA	170
	<i>hapRR</i>	CTTGGGCCAGTTTACG	

Flagella

<i>flaC</i>	<i>flaCF</i>	GCGAAAGAAGGGGATGAT	178
	<i>flaCR</i>	GCGCAGCACCGATAACC	
<i>fliI</i>	<i>fliIF</i>	TCTTCCGCCAGTGT	174
	<i>fliIR</i>	ATCGCGGAACCCAGTC	

Iron transport

<i>fbpB</i>	<i>fbpBF</i>	GTGGGGATCCGCTCTGGT	169
	<i>fbpBR</i>	TCATCGCCCGCACAAAAC	

Chemotaxis

<i>cheR</i>	<i>cheRF</i>	CTGCCGATTCCGTGATTA	159
	<i>cheRR</i>	TCTCCGCTGGCTTGAAA	
<i>cheB</i>	<i>cheBF</i>	TCTGCGGTCGATTACAAG	166
	<i>cheBR</i>	ATCGCAAGCTGGTGGTCT	

Bile resistance

<i>bile</i>	<i>bileF</i>	GACGTGAACGGCGACTCT	179
	<i>bileR</i>	TCTGCGGCATTGATTGTG	

Biotin synthesis

<i>bioB</i>	<i>bioBF</i>	GCGGGGCTGGATTATTAC	178
	<i>bioBR</i>	CAGCGCGGTCATTGGT	

In vivo inducible genes

<i>tprG</i>	<i>tprGF</i>	TGCTGCAACGCCTGAAAG	180
	<i>tprGR</i>	CGATCGCCAACGGTGAAG	
<i>acnA</i>	<i>acnAF</i>	GCGGGAGCGGATTATGG	173
	<i>acnAR</i>	AGCGAGTGGCGATTGACG	
<i>gltB1</i>	<i>gltB1F</i>	GTGGCGGTCAGGGTTGTG	173
	<i>gltB1R</i>	TTGGGCGCGTTTATGTTC	
<i>actP</i>	<i>actPF</i>	GCGGGTCTGCTACTGGTT	158
	<i>actPR</i>	CCCGGAGGATTGATGC	
<i>cadA</i>	<i>cadAF</i>	TGGCGCAAGGCATTAC	167
	<i>cadAR</i>	CACGCGACCCACCATCTC	

Real-time PCR internal control

<i>recA</i>	<i>recAF</i>	AAGCAATGCGTAAACTGA	162
	<i>recAR</i>	GGCGAATATCCAAACGAA	

Table SI 2. Selected gene expression profiles of *V. cholerae* strain N16961 grown in M9-Gnt as compared to M9-Glu grown cells and their comparison to microarray analysis of *V. cholerae* genes differentially expressed in rabbit ileal loop as available in GSE24405-07. Real-time PCR based comparative analysis on certain genes have been included.

ORF	Gene	Gene ID#	Gnt microarray†	Expression significance in Gnt grown cells‡	Expression significance in 4h/ 12h in mucus	Real Time PCR±
Metabolic pathway						
2-keto-3-deoxy-6-phosphogluconate aldolase (EC 4.1.2.14)	<i>eda</i>	VC0285	2.96	+	0/0	16.87
Gluconate permease	<i>gntP</i>	VC0286	3.85	+	+*/0	ND
Gluconate kinase (EC 2.7.1.12)	<i>gntK</i>	VC0287	2.81	+	0/0	ND
6-phosphogluconate dehydratase (EC 4.2.1.12)	<i>edd</i>	VC0288	3.27	+	+*/0	31.76
Gnt-I regulator	<i>gntR</i>	VC0289	1.34	+	0/0	ND
Glucose-6-phosphate dehydrogenase (EC 1.1.1.49)	<i>zwf</i>	VCA0896	-4.49	-	0/0	ND
6-phosphogluconolactonase (EC 3.1.1.31)	<i>pgls</i>	VCA0897	-5.09	-	0/-*	ND
6-phosphogluconate dehydrogenase (EC 1.1.1.44)	<i>gnd</i>	VCA0898	-3.55	-	0/-*	0.25
Ribulose-5-phosphate epimerase (EC 5.1.3.1)	<i>rpe</i>	VC2625	-0.42	0	-*/0	ND
Ribose-5-phosphate isomerase (EC 5.3.1.6)	<i>rpiA</i>	VC2480	-1.11	-	-*/0	ND
Transaldolase (EC 2.2.1.2)	<i>tka</i>	VCA0623	-1.81	-	-*/0	0.60
Transketolase (EC 2.2.1.1)	<i>tkt</i>	VCA0624	-0.72	-	0/-*	ND

Glucose-6-phosphate isomerase (EC 5.3.1.9)	<i>pgi</i>	VC0374	-0.58	0	0/-*	ND
6-phosphofructokinase (EC 2.7.1.11)	<i>pfk</i>	VC2689	-0.48	0	-*/0	0.05
Fructose-1-6-bisphosphate aldolase (EC 4.1.2.13)	<i>fba</i>	VC0478	-1.75	-*	0/0	0.57
Malate synthase (EC 2.3.3.9)	<i>aceB</i>	VC0734	4.50	+	0/0	3.55
Isocitrate lyase (EC 4.1.3.1)	<i>aceA</i>	VC0736	4.45	+	0/+*	2.80
Glycogen synthesis						
Glycogen synthase (EC 2.4.1.21)	<i>glgA</i>	VC1726	2.32	+	0/+*	9.45
Glycogen operon protein	<i>glgX</i>	VCA1029	1.17	+	0/0	ND
Glycogen branching enzyme (EC 2.4.1.18)	<i>glgB</i>	VCA0016	0.40	0	0/+*	ND
Virulence						
Cholera toxin subunit B	<i>ctxB</i>	VC1456	1.73	+	+*/0	ND
Cholera toxin subunit A (EC 2.4.2.36)	<i>ctxA</i>	VC1457	0.23	0	+*/0	58.03
Zonula occludens toxin	<i>zot</i>	VC1458	0.34	0	0/0	ND
Accessory cholera enterotoxin	<i>ace</i>	VC1459	0.74	+	0/0	ND
Repetitive toxin transporter	<i>rtxA</i>	VC1448	1.15	+	0/0	ND
Haemagglutinin protease	<i>hapA</i>	VCA0446	-5.98	-*	0/0	0.49
Haemolysin	<i>hlyA</i>	VC0489	0.06	0	0/0	0.14
Outer membrane protein U	<i>ompU</i>	VC0633	-3.91	-*	0/0	0.86
Outer membrane protein T	<i>ompT</i>	VC1854	3.21	+	0/+*	10.52
Colonization						
Toxin coregulated pilus	<i>tcpA</i>	VC0828	1.47	+	+*/0	472.06

Mannose sensitive haemagglutinin pilin subunit B	<i>mshB</i>	VC0408	2.44	+*	0/+*	ND
Mannose sensitive haemagglutinin pilin subunit A	<i>mshA</i>	VC0409	0.65	+*	0/0	1.80
Mannose sensitive haemagglutinin pilin subunit C	<i>mshC</i>	VC0410	2.63	+*	0/0	ND
Mannose sensitive haemagglutinin pilin subunit D	<i>mshD</i>	VC0411	3.15	+*	0/+*	ND
Accessory colonizing factor	<i>acf</i>	VC0841	1.20	+*	+*/0	4.93
Core encoded pilus	<i>cep</i>	VC1461	2.23	+*	0/0	ND
Pathogenic regulation						
<i>toxR</i> activated gene A lipoprotein	<i>tagA</i>	VC0820	0.35	0	+*/0	ND
Toxin coregulated pilus biosynthesis protein P	<i>tcpP</i>	VC0826	1.43	+*	0/0	1.98
Toxin coregulated pilus biosynthesis protein H	<i>tcpH</i>	VC0827	1.90	+*	0/0	2.76
Toxin coregulated pilus biosynthesis protein S	<i>tcpS</i>	VC0834	1.78	+*	0/0	ND
Toxin coregulated pilus biosynthesis protein T	<i>tcpT</i>	VC0835	2.79	+*	+*/0	ND
Toxin coregulated pilus virulence regulatory protein	<i>toxT</i>	VC0838	2.24	+*	+*/0	22.47
Cholera toxin transcriptional activator	<i>toxR</i>	VC0984	2.72	+*	+*/0	5.45
Transmembrane regulatory protein	<i>toxS</i>	VC0983	1.69	+*	0/0	3.87
PadR family transcriptional regulator	<i>aphA</i>	VC2647	1.24	+*	0/+*	1.46
LysR family transcriptional regulator	<i>aphB</i>	VC1049	1.03	+*	0/0	1.60
DNA binding H-NS protein	<i>hns</i>	VC1130	-0.80	-*	0/0	0.32
Biofilm regulation						
Adenylate cyclase (EC 4.6.1.1)	<i>cya</i>	VC0122	1.53	+*	0/0	3.06
Catabolic repressor protein	<i>crp</i>	VC2614	1.87	+*	0/0	2.31

C-di-GMP phosphodiesterase		VC0658	2.87	+*	0/0	1.91
Di-guanylate cyclase		VCA0956	-1.19	-*	0/0	ND
GGDEF		VCA0939	-1.78	-*	-*/0	ND
Sigma E factor regulatory protein	<i>rseC</i>	VC2464	-1.62	-*	0/0	ND
Sigma 54 dependent transcriptional regulator		VC1817	1.75	+*	0/0	ND
Chitinase (EC 3.2.1.14)	<i>chiA</i>	VCA0027	-0.47	0	-*/0	0.18
Chitinase (EC 3.2.1.14)	<i>chiA-I</i>	VC1073	-1.51	-*	0/0	ND
Transcriptional regulator	<i>vpsT</i>	VCA0952	-0.78	-*	0/0	ND
Polysaccharide biosynthesis glycosyltransferase	<i>vpsL</i>	VC0934	-0.94	-*	-*/0	ND
UDP-N-acetylglucosamine 2-epimerase (EC 5.1.3.14)	<i>wecB</i>	VC0917	-1.00	-*	0/0	0.42
UDP-N-acetylmannosamine dehydrogenase	<i>WecC</i>	VC0918	-0.20	0	0/0	ND
Serine acetyltransferase related protein (EC 2.3.1.30)	<i>cysE</i>	VC0923	-0.24	0	0/0	ND

Flagella

Flagellin C	<i>flaC</i>	VC2187	1.02	+*	+*/0	1.50
Flagellar basal body P-ring biosynthesis protein	<i>flgA</i>	VC2203	0.67	+*	0/0	ND
Negative regulator of flagellin biosynthesis	<i>flgM</i>	VC2204	-0.72	-*	0/0	ND
Flagellum specific ATP synthase (EC:3.6.3.14)	<i>fliI</i>	VC2130	1.38	+*	0/0	1.46
Flagellar biosynthesis sigma factor	<i>fliA</i>	VC2066	0.46	0	0/0	ND

Iron transport

Ferrous iron transport protein B	<i>feoB</i>	VC2077	1.02	+*	0/0	ND
Ferrous iron transport protein A	<i>feoA</i>	VC2078	0.60	0	0/+*	ND

Hemin ABC transporter	<i>hutB</i>	VCA0913	1.07	+*	0/0	ND
Hemin ABC transporter	<i>hutC</i>	VCA0914	1.43	+*	0/0	ND
Ferric vibriobactin transporter	<i>viuP</i>	VC0776	3.15	+*	+*/0	ND
Ferric vibriobactin transporter	<i>viuD</i>	VC0777	0.76	+*	+*/0	ND
Ferric vibriobactin transporter	<i>viuG</i>	VC0778	1.53	+*	+*/0	ND
Ferric vibriobactin transporter	<i>viuH</i>	VC0779	1.35	+*	+*/0	ND
Iron (III) ABC transporter	<i>fbpA</i>	VCA0685	5.41	+*	0/+*	ND
Iron (III) ABC transporter	<i>fbpB</i>	VCA0686	6.92	+*	+*/0	4.40
Iron (III) ABC transporter	<i>fbpC</i>	VCA0687	2.27	+*	0/0	ND

Chemotaxis

Chemotaxis protein methyltransferase	<i>cheR</i>	VC1399	4.38	+*	0/0	9.77
Chemotaxis protein	<i>cheA</i>	VC1400	1.73	+*	0/0	ND
Chemotaxis protein methylesterase	<i>cheB</i>	VC1401	2.51	+*	0/0	2.48
Purine binding chemotaxis protein	<i>cheW</i>	VC1402	0.16	0	0/0	ND
Methyl accepting chemotaxis protein		VC1535	0.96	+*	0/0	ND
Methyl accepting chemotaxis protein		VC1248	1.43	+*	+*/0	ND
Methyl accepting chemotaxis protein		VC1298	2.11	+*	0/0	ND
Methyl accepting chemotaxis protein		VC1394	3.70	+*	0/+*	ND
Methyl accepting chemotaxis protein		VC1403	0.55	0	0/+*	ND
Methyl accepting chemotaxis protein		VC1643	3.17	+*	+*/0	ND

Bile resistance

Bile resistance protein		VCA1008	0.91	+*	0/0	ND
Terrulite resistance protein		VC0813	1.66	+*	0/+*	ND
Bile resistance protein		VCA0877	2.15	+*	0/0	2.75
Biotin synthesis						
Biotin biosynthesis protein B (EC 2.8.1.6)	<i>bioB</i>	VC1112	1.80	+*	0/0	2.19
Biotin biosynthesis protein C (EC 2.1.1.197)	<i>bioC</i>	VC1114	2.44	+*	+*/0	ND
Biotin biosynthesis protein D (EC 6.3.3.3)	<i>bioD</i>	VC1115	1.25	+*	+*/0	ND
<i>In vivo</i> inducible genes						
Anthranilate synthase (EC 4.1.3.27)	<i>tprG</i>	VC1173	4.07	+*	0/0	6.72
Aconitate hydratase (EC 4.2.1.3)	<i>acnA</i>	VC1338	3.43	+*	0/0	3.84
Glutamate synthase (EC 1.4.1.13)	<i>gltB1</i>	VC2373	2.85	+*	0/0	4.13
Phosphoenolpyruvate carboxylase (EC 4.1.1.31)	<i>ppc</i>	VC2646	0.51	0	0/0	ND
Adenylyltransferase (EC 2.7.7.73)	<i>thiF</i>	VC0063	1.61	+*	0/0	ND
Serine transporter	<i>sdaC2</i>	VC1658	0.93	+*	0/0	ND
Cation/acetate symporter	<i>actP</i>	VC2705	7.48	+*	+*/0	5.90
Histidine sensor kinase		VC0622	2.24	+*	0/0	ND
Extra-cellular nuclease related protein		VC2621	1.24	+*	+*/0	ND
RNase BN	<i>rbn</i>	VC2742	0.34	0	0/0	ND
Recombinase protein	<i>xerD</i>	VC2419	0.33	0	0/0	ND
Paraquat-inducible protein A	<i>pqiA</i>	VC1500	0.79	+*	0/0	ND
Lysine decarboxylase (EC 4.1.1.18)	<i>cadA</i>	VC0281	1.39	+*	+*/0	2.91

#, Gene ID is based on annotation of N16961 whole genome sequence; †, Fold change in microarray analysis (Gnt grown cells as compared to Glu grown cells) where - indicates decrease of transcript levels as compared to Glu grown cells; ‡, Fold change (increase or decrease) by 0.6 or more (\log_2 expression ratio) have been considered for expression significance and +* denotes increase significantly while -* denotes decrease significantly; † Microarray based relative expression adopted from GSE24405-07 for a comparison; ‡Relative fold in transcript level of Gnt grown cells as compared to Glu grown cells and that was determined by real-time PCR; ND, not done