



**Fig. S01.** *S. Typhimurium* biofilm formation in human bile (HB). Biofilm formation in M9 media supplemented with different amounts of human bile or glucose (10 mM) measured by crystal violet.

**Supplementary Table 3.** Differentially regulated operons in biofilms grown in *in vitro* human GS conditions by RNASeq

Operon function	Gene name	Locus number	Fold Change	P value	Function
SPI6	STM14_0320	STM14_0320	57.86	0	putative cytoplasmic protein
	STM14_0321	STM14_0321	20.61	0	putative cytoplasmic protein
Fimbriae	stbB	STM14_0395	-10.93	0.00010127	Putative fimbriae chaparone;stbB;ortholog
	stbA	STM14_0396	-4.8	0.0502858	Putative fimbriae major subunit;stbA;ortholog
Purine metabolism	purA	STM14_5248	2.09	2.723E-07	Adenylosuccinate synthetase;purA;ortholog
	purB	STM14_1410	2.73	0.00166837	Adenylosuccinate lyase;purB;ortholog
	purC	STM14_3050	8.23	0	Phosphoribosylaminoimidazole-succinocarboxamide synthase;purC;ortholog
	purD	STM14_5016	7.78	0	Phosphoribosylamine--glycine ligase;purD;ortholog
	purE	STM14_0624	16.47	0	N5-carboxyaminoimidazole ribonucleotide mutase;purE;ortholog
	purF	STM14_2909	14.3	0	Amidophosphoribosyltransferase;purF;ortholog
	purG	STM14_3145	16.15	0	#N/A
	purH	STM14_5017	3.65	0	Bifunctional purine biosynthesis protein PurH;purH;ortholog
	purK	STM14_0623	25.24	0	N5-carboxyaminoimidazole ribonucleotide synthase;purK;ortholog
	purM	STM14_3064	13.05	0	Phosphoribosylformylglycinamide cyclo-ligase;purM;ortholog
	purN	STM14_3065	7.87	0	Phosphoribosylglycinamide formyltransferase;purN;ortholog
	purR	STM14_1727	1.84	0.02341246	HTH-type transcriptional repressor PurR;purR;ortholog
	purT	STM14_2288	22.79	0	Phosphoribosylglycinamide formyltransferase 2;purT;ortholog
	purU	STM14_2122	1.81	3.6244E-05	Formyltetrahydrofolate deformylase;purU;ortholog
	yicE	STM14_4513	14.08	0	Putative NCS2 family purine/xanthine transport protein;yicE;ortholog
Fimbriae	fimA	STM14_0635	-10.15	4E-10	Type-1 fimbrial protein, A chain;fimA;ortholog
	fimI	STM14_0636	-9.22	1.667E-07	Fimbrin-like protein FimI;fimI;ortholog
	fimC	STM14_0637	-14.46	9.89E-08	Chaperone protein FimC;fimC;ortholog
	fimD	STM14_0638	-3.03	0.00151554	Outer membrane usher protein FimD;fimD;ortholog
	fimZ	STM14_0641	-15.8	0	Fimbriae Z protein;fimZ;ortholog
	fimY	STM14_0642	-4.03	7.2768E-05	Fimbriae Y protein;fimY;ortholog
	STM14_0643	STM14_0643	-4.86	0.00043957	#N/A
	fimW	STM14_0644	-1.8	0.04777227	Fimbriae W protein;fimW;ortholog
SPI2 effector	ssel	STM14_1193	-7.32	0	Secreted effector protein Ssel;ssel;ortholog
	ssel	STM14_1193	-15.02	0	Secreted effector protein Ssel;ssel;ortholog
SPI2 effector	pipA	STM14_1232	-2.44	0.00193477	Pathogenicity island encoded protein: SPI5;pipA;ortholog
	pipB	STM14_1233	-37.41	0	Secreted effector protein PipB;pipB;ortholog
	STM14_1235	STM14_1235	-9.76	0.00231626	#N/A
	serX	STM14_1297	-10.39	0.00070678	tRNA-Ser
Curli fimbriae	csgG	STM14_1303	8.24	0	Curli production assembly/transport component CsgG;csgG;ortholog
	csgF	STM14_1304	20.13	0	Curli production assembly/transport component CsgF;csgF;ortholog
	csgE	STM14_1305	26.57	0	Curli production assembly/transport component CsgE;csgE;ortholog

	csgD	STM14_1306	27.14	0	Probable csgAB operon transcriptional regulatory protein;csgD;ortholog
	csgB	STM14_1309	76.33	0	Minor curlin subunit;csgB;ortholog
	csgA	STM14_1310	82.93	0	Major curlin subunit;csgA;ortholog
	csgC	STM14_1311	31.74	0	Curli assembly protein CsgC;csgC;ortholog
	ymdA	STM14_1312	11.43	0	Uncharacterized protein YmdA;ymdA;ortholog
	rpmF	STM14_1364	56.75	0	50S ribosomal protein L32;rpmF;ortholog
SPI2 effector	sifA	STM14_1400	-23.66	0	Secreted effector protein SifA;sifA;ortholog
Arginine utilization in Nitrogen limited env	argD_1	STM14_1582	-9.76	1.2E-08	#N/A
	astA	STM14_1583	-9.89	2.5E-09	Arginine N-succinyltransferase;astA;ortholog
	astD	STM14_1584	-5.59	6.532E-07	N-succinylglutamate 5-semialdehyde dehydrogenase;astD;ortholog
	astB	STM14_1585	-3.24	7.1393E-05	N-succinylarginine dihydrolase;astB;ortholog
	spy	STM14_1588	-2.28	3.58E-08	Periplasmic protein related to spheroblast formation;spy;ortholog
	osmE	STM14_1592	-12.77	0	Transcriptional activator of ntrL protein;osmE;ortholog
SPI2	ssrB	STM14_1686	-15.15	0	Secretion system regulator: transcriptonal activator;ssrB;ortholog
	ssrA	STM14_1687	-21.9	0	Histidine kinase;ssrA;ortholog
	ssaC	STM14_1689	-35.23	0	Secretion system apparatus protein;ssaC;ortholog
	ssaD	STM14_1690	-26.6	0	Secretion system apparatus protein;ssaD;ortholog
	ssaE	STM14_1691	-19.92	0	Secretion system effector;ssaE;ortholog
	STM14_1693	STM14_1693	-32.8	0	#N/A
	STM14_1694	STM14_1694	-28.11	0	#N/A
	sscA	STM14_1695	-21.96	0	Secretion system chaparone;sscA;ortholog
	sseC	STM14_1696	-24.49	0	Secreted effector protein SseC;sseC;ortholog
	sseD	STM14_1697	-22.19	0	Secreted effector protein SseD;sseD;ortholog
	sseE	STM14_1698	-26.24	0	Secretion system effector;sseE;ortholog
	sscB	STM14_1699	-21.2	0	Secretion system chaparone;sscB;ortholog
	sseF	STM14_1700	-9.63	0	Secretion system effector;sseF;ortholog
	sseG	STM14_1701	-3.41	3.07E-08	Secretion system effector;sseG;ortholog
	ssaG	STM14_1702	-22.16	0	Secretion system apparatus;ssaG;ortholog
	ssaG	STM14_1702	-25.24	0	Secretion system apparatus;ssaG;ortholog
	ssaI	STM14_1704	-27.82	0	Secretion system apparatus;ssaI;ortholog
	ssaJ	STM14_1705	-29.28	0	Secretion system apparatus lipoprotein SsaJ;ssaJ;ortholog
	STM14_1706	STM14_1706	-23.52	0	#N/A
	ssaK	STM14_1707	-28.58	0	Secretion system apparatus protein SsaK;ssaK;ortholog
	ssaL	STM14_1708	-20.35	0	Secretion system apparatus protein SsaL;ssaL;ortholog
	ssaM	STM14_1709	-17.63	0	Secretion system apparatus protein SsaM;ssaM;ortholog
	ssaV	STM14_1710	-20.53	0	Secretion system apparatus protein SsaV;ssaV;ortholog
	ssaN	STM14_1711	-11.75	0	Probable secretion system apparatus ATP synthase SsaN;ssaN;ortholog
ssaO	STM14_1712	-6.87	1.282E-07	Secretion system apparatus protein SsaO;ssaO;ortholog	
ssaP	STM14_1713	-6.69	0	Secretion system apparatus protein SsaP;ssaP;ortholog	

	ssaQ	STM14_1714	-2.81	0.00013326	Secretion system apparatus protein SsaQ;ssaQ;ortholog
	ssaR	STM14_1715	-32.78	0	#N/A
	ssaS	STM14_1716	-31.12	0	Secretion system apparatus protein SsaS;ssaS;ortholog
	ssaT	STM14_1717	-29.77	0	Secretion system apparatus protein SsaT;ssaT;ortholog
	ssaU	STM14_1718	-11.57	0	Secretion system apparatus protein SsaU;ssaU;ortholog
	valV	STM14_1721	10.54	6.1801E-06	#N/A
	STM14_1804	STM14_1804	-10.89	0	#N/A
	STM14_1805	STM14_1805	-12.07	0	#N/A
	STM14_1937	STM14_1937	-12.34	0	#N/A
SPI2 effector	sifB	STM14_1940	-20.99	0	Secreted effector protein SifB;sifB;ortholog
	yncJ	STM14_1941	-10.69	0	Putative periplasmic protein;yncJ;ortholog
	STM14_1971	STM14_1971	-25.25	0	#N/A
SPI2 effector	sseJ	STM14_1974	-27.72	0	Secreted effector protein SseJ;sseJ;ortholog
propanediol utilization (pdu) operon	pduM	STM14_2537	51.58	0	Propanediol utilization protein;pduM;ortholog
	pduP	STM14_2540	8.14	0	Propanediol utilization CoA-dependent propionaldehyde dehydrogenase;pduP;ortholog
	pduS	STM14_2542	7.65	2E-10	Propanediol utilization protein;pduS;ortholog
	STM14_2637	STM14_2637	-20.79	0	putative ABC transporter periplasmic component
	STM14_2638	STM14_2638	-16.49	0	putative inner membrane protein
	STM14_2756	STM14_2756	-5.63	1.094E-05	#N/A
	STM14_2759	STM14_2759	-32.27	0	#N/A
	STM14_2760	STM14_2760	-29.99	0	#N/A
	STM14_2761	STM14_2761	-32.74	0	#N/A
	STM14_2762	STM14_2762	-39.8	0	#N/A
	STM14_2763	STM14_2763	-30.43	0	#N/A
SPI2 effector	sseL	STM14_2824	-33.69	0	Deubiquitinase SseL;sseL;ortholog
Thiamine synthesis	purF	STM14_2909	14.3	0	Amidophosphoribosyltransferase;purF;ortholog
Colicin V production	cvpA	STM14_2910	12.97	0	Membrane protein required for colicin V production;cvpA;ortholog
Phage	STM14_3166	STM14_3166	-10.74	3.6E-09	DNA polymerase V subunit
	STM14_3168	STM14_3168	-6.02	0.00079487	phage tail assembly-like protein
	STM14_3169	STM14_3169	-2.77	0.05396389	phage tail assembly-like protein
	STM14_3170	STM14_3170	-1.71	0.0323214	tail fiber-like protein
	STM14_3171	STM14_3171	-10.78	0.02949509	hypothetical protein
	STM14_3172	STM14_3172	-3.8	0.00217499	host specificity protein-J-like protein
	STM14_3192	STM14_3192	-31.83	0.00113684	DNA packaging-like protein
	STM14_3331	STM14_3331	-8.9	0.00817208	#N/A
	STM14_3336	STM14_3336	1.63	0.03114065	#N/A
	iroN	STM14_3348	-19.64	5.1E-09	TonB-dependent siderophore receptor protein;iroN;ortholog
SPI2 effector	pipB2	STM14_3350	-35.71	0	Secreted effector protein pipB2;pipB2;ortholog
Virulence protein	virK	STM14_3352	-4.74	0	VirK-like protein;virK;ortholog
tricarboxylic transport	STM14_3361	STM14_3361	13.82	7.88E-08	tricarboxylic transport
	STM14_3362	STM14_3362	8.72	0.00019185	tricarboxylic transport

	STM14_3363	STM14_3363	12.05	4.799E-07	tricarboxylic transport
	STM14_3365	STM14_3365	-30.32	0	hypothetical protein
	ygaF	STM14_3366	-24.91	0	L-2-hydroxyglutarate oxidase LhgO;ygaF;ortholog
	gabD	STM14_3367	-15.76	0	NADP-dependent succinate-semialdehyde dehydrogenase I;gabD;ortholog
	gabT	STM14_3368	-12.21	3.3E-09	4-aminobutyrate aminotransferase;gabT;ortholog
Hydrogenase	hypF	STM14_3434	-2.04	0.0049248	Hydrogenase maturation protein;hypF;ortholog
	hydN	STM14_3436	-28.56	0	Electron transport protein (FeS center) from formate to hydrogen;hydN;ortholog
	STM14_3438	STM14_3438	-28.12	0	#N/A
	hycl	STM14_3439	-31.77	0	Protease involved in processing C-terminal end of HycE;hycl;ortholog
	hycH	STM14_3440	-27.25	0	Protein for processing of HycE;hycH;ortholog
	hycG	STM14_3441	-33.57	0	Hydrogenase activity;hycG;ortholog
	hycF	STM14_3442	-2.33	0.005573	Hydrogenase 3;hycF;ortholog
	hycE	STM14_3443	-36.23	0	Hydrogenase 3, large subunit;hycE;ortholog
	hycD	STM14_3444	-30.15	0	Hydrogenase 3, membrane subunit;hycD;ortholog
	hycC	STM14_3445	-22.18	0	Hydrogenase 3, membrane subunit;hycC;ortholog
	hycB	STM14_3446	-36.07	0	Hydrogenase-3, iron-sulfur subunit;hycB;ortholog
	hycA	STM14_3448	-24.88	0	Transcriptional repressor of hyc and hyp operons;hycA;ortholog
	hypB	STM14_3451	-6.57	0	Hydrogenase-3 accessory protein;hypB;ortholog
	hypC	STM14_3452	-4.22	2.334E-06	Putative hydrogenase expression/formation protein;hypC;ortholog
	hypD	STM14_3453	-4.52	1.41E-08	Putative hydrogenase expression/formation protein;hypD;ortholog
	hypE	STM14_3454	-6.16	9E-10	Putative hydrogenase expression/formation protein;hypE;ortholog
	fhlA	STM14_3455	-3.12	1.6341E-05	Formate hydrogenlyase transcriptional activator;fhlA;ortholog
	ygbA	STM14_3456	-4.19	2.458E-07	Putative cytoplasmic protein;ygbA;ortholog
Iron transport	sitA	STM14_3458	-7.2	0	Fur regulated Salmonella iron transporter;sitA;ortholog
	sitB	STM14_3459	-5.1	0	Fur regulated Salmonella iron transporter;sitB;ortholog
	sitC	STM14_3460	-5.99	0	Fur regulated Salmonella iron transporter;sitC;ortholog
	sitD	STM14_3461	-9.92	0	Fur regulated Salmonella iron transporter;sitD;ortholog
	STM14_3787	STM14_3787	-7.68	0	#N/A
	STM14_3787	STM14_3787	-14.47	0	#N/A
	STM14_3787	STM14_3787	-17.11	0	#N/A
putative cation/proton antiporter	yjcO	STM14_5153	-10.25	0	Putative TPR repeat protein;yjcO;ortholog
	nrdG	STM14_5342	14.72	0	Anaerobic ribonucleoside-triphosphate reductase-activating protein;nrdG;ortholog
	treC	STM14_5345	-8.44	1.3E-09	Trehalose-6-P hydrolase;treC;ortholog
	treB	STM14_5346	-12.47	0	#N/A

**Supplementary Table 4.** Strains and plasmids used in this study.

Strains	Genotype or relevant phenotype	Reference or source
JSG210	<i>Salmonella enterica</i> sv. Typhimurium ATCC 14028	ATCC
JSG3736	<i>S. Typhimurium</i> $\Delta$ <i>csgA</i>	(1)
JSG1149	<i>S. Typhimurium</i> pFPV25.1	This study
JSG4136	<i>S. Typhimurium</i> $\Delta$ <i>csgA</i> pFPV25.1	This study
JSG4237	<i>S. Typhimurium</i> WT (JSG210) + pCS26	This study
JSG4238	<i>S. Typhimurium</i> WT (JSG210) + pBA	This study
JSG4239	<i>S. Typhimurium</i> WT (JSG210) + pDEFG	This study
JSG4277	<i>S. Typhimurium</i> $\Delta$ <i>csgD</i>	This study
JSG4292	<i>S. Typhimurium</i> $\Delta$ <i>csgD</i> (JSG4277) + pCS26	This study
JSG4293	<i>S. Typhimurium</i> $\Delta$ <i>csgD</i> (JSG4277) + pBA	This study
JSG4294	<i>S. Typhimurium</i> $\Delta$ <i>csgD</i> (JSG4277) + pDEFG	This study
JSG1811	<i>S. Typhimurium</i> $\Delta$ <i>rpos</i>	(2)
JSG4272	<i>S. Typhimurium</i> $\Delta$ <i>rpos</i> (JSG1811) + pCS26	This study
JSG4273	<i>S. Typhimurium</i> $\Delta$ <i>rpos</i> (JSG1811) + pBA	This study
JSG4274	<i>S. Typhimurium</i> $\Delta$ <i>rpos</i> (JSG1811) + pDEFG	This study
JSG4302	<i>S. Typhimurium</i> <i>ompR</i> ::Cam	(3)
JSG4303	<i>S. Typhimurium</i> <i>ompR</i> ::Cam (JSG4302) + pCS26	This study
JSG4304	<i>S. Typhimurium</i> <i>ompR</i> ::Cam (JSG4302) + pBA	This study
JSG4305	<i>S. Typhimurium</i> <i>ompR</i> ::Cam (JSG4302) + pDEFG	This study
JSG4355	<i>S. Typhimurium</i> <i>cpxR</i> ::Cam	(6)
JSG4356	<i>S. Typhimurium</i> <i>hns</i> ::Cam	(2)
JSG4360	<i>S. Typhimurium</i> <i>cpxR</i> ::Cam mutant (JSG4355) + pCS26	This study
JSG4361	<i>S. Typhimurium</i> <i>cpxR</i> ::Cam mutant (JSG4355) + pBA	This study
JSG4362	<i>S. Typhimurium</i> <i>cpxR</i> ::Cam mutant (JSG4355) + pDEFG	This study
JSG4363	<i>S. Typhimurium</i> <i>hns</i> ::Cam mutant (JSG4356) + pCS26	This study
JSG4364	<i>S. Typhimurium</i> <i>hns</i> ::Cam mutant (JSG4356) + pBA	This study
JSG4365	<i>S. Typhimurium</i> <i>hns</i> ::Cam mutant (JSG4356) + pDEFG	This study
Plasmid	Characteristics	Source
pFPV25.1	GFP tagging; pfpv25 p <sub>rpsM</sub> -gfpmut3	(4)
pCS26	Empty pCS26 plasmid with <i>luxCDABE</i> and Km resistance cassette	(5)
pBA	pCS26 with curli <i>csgBA</i> operon promoter	(5)
pDEFG	pCS26 with Curli <i>csgDEFG</i> operon promoter	(5)

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**Supplementary table 5.** Oligonucleotide primers used in this study

<b>Primer</b>	<b>Sequence</b>	<b>Purpose</b>
JG2674	5'-CCAATATTACCCAGTACGGTACGCAGAAAACAGC-3'	Forward <i>csgA</i>
JG2675	5'-GGTTCGTTTAATGTGACCTGAGGGATCACCG-3'	Reverse <i>csgA</i>
JG2676	5'-GCTTGTTTTTCGCCAGCATGAATTACTCTGC-3'	Forward <i>wcaM</i>
JG2677	5'-CGTGGTATCTACCGTGCATAGCGGTATTCC-3'	Reverse <i>wcaM</i>
JG2682	5'-GGCAGCATTACGCCCCGTCGTCAATACGGG-3'	Forward <i>bscE</i>
JG2683	5'-CGGCGCAGAGTTCCGGCTGGTTTTACATAACG-3'	Reverse <i>bscE</i>
JG3010	5'-TGTGTCACGTCTTGTGCGTA-3'	Forward pBA
JG3011	5'-TCGGGAAAGATTTCAACCTG-3'	Reverse pBA
JG3012	5'-CAGCTGTCAGATGTGCGATT-3'	Forward pDEFG
JG3013	5'-TCATCCATGATATGGCGAGA-3'	Reverse pDEFG
JG2993	5'-ATGGGCTATTTCAACCCACA-3'	Forward Kn
JG2994	5'-GTCGCTGAATGCACGATAGA-3'	Reverse Kn
JG2658	5'-TCGACATTGCAGGCAGTAAG-3'	Forward RpoS
JG2659	5'-ACTTCGCCGGTAAAAGGAGT-3'	Reverse Rpos
JG2991	5'-gtgcgattaaaaaagtggagttcatcATGTTTAATGAAGTGTAGGCTGGAGCTGCTTC-3'	Forward <i>csgD</i>
JG2992	5'-aatccaggtcagatagcgtttcatggccTTACCGCCTGAGCATATGAATATCCTCCTTAG-3'	Reverse <i>csgD</i>