

Table S1 Predicted target genes for csRNA1-1

Rank	Locus tag	Gene description (Gene)	Energy	Pvalue
1	SSA_2317	Tfp pilus assembly protein, pilus retraction ATPase PilT	-15.87	0
2	SSA_2345	hypothetical protein	-14.36	0.001
3	SSA_2216	LPS biosynthesis protein (<i>licD1</i>)	-14.01	0.002
4	SSA_1763	molybdenum ABC transporter ATPase	-13.14	0.003
5	SSA_2217	Cps9H	-12.43	0.006
6	SSA_1635	hypothetical protein	-11.89	0.008
7	SSA_0227	collagen-binding surface protein	-11.64	0.009
8	SSA_1820	hypothetical protein	-11.39	0.011
9	SSA_1632	surface protein	-11.36	0.011
10	SSA_0991	deoxyribonuclease	-11.16	0.012
11	SSA_1168	hypothetical protein	-11.06	0.013
12	SSA_1702	hypothetical protein	-10.95	0.014
13	SSA_0926	histone acetyltransferase HPA2-like acetyltransferase	-10.79	0.015
14	SSA_1018	Zinc metalloprotease zmpC	-10.71	0.016
15	SSA_1736	L-cysteine desulfhydrase	-10.67	0.016
16	SSA_0655	cell division protein FtsA (<i>ftsA</i>)	-10.51	0.018
17	SSA_1355	hypothetical protein	-10.45	0.019
18	SSA_0737	arginine deiminase (<i>sagP</i>)	-10.21	0.021
19	SSA_2004	Zinc metalloprotease zmpB (<i>zmpB</i>)	-10.12	0.022
20	SSA_2205	transcription antitermination protein NusG (<i>nusG</i>)	-10.04	0.023
21	SSA_0061	tagatose 1,6-diphosphate aldolase	-9.96	0.024
22	SSA_1544	hypothetical protein	-9.84	0.025
23	SSA_1899	hypothetical protein	-9.84	0.025
24	SSA_0146	DNA repair ATPase	-9.82	0.025
25	SSA_0293	hypothetical protein	-9.7	0.027
26	SSA_1634	Heme utilization/adhesion exoprotein	-9.58	0.029
27	SSA_2121	cell wall surface anchor family protein	-9.55	0.029
28	SSA_1110	hypothetical protein	-9.53	0.029
29	SSA_0770	ribonucleotide-diphosphate reductase subunit alpha (<i>nrdE</i>)	-9.4	0.031
30	SSA_2301	S-layer protein (<i>lytB</i>)	-9.3	0.033
31	SSA_1631	sortase (<i>srtC</i>)	-9.25	0.033
32	SSA_0849	Signal peptidase I (<i>s/p</i>)	-9.1	0.036
33	SSA_2208	preprotein translocase subunit SecE (<i>secE</i>)	-9.08	0.036
34	SSA_1044	homoserine kinase (<i>thrB</i>)	-9.05	0.036
35	SSA_1020	hypothetical protein	-9.01	0.037
36	SSA_2309	fimbrial assembly protein	-8.95	0.038
37	SSA_0718	hypothetical protein	-8.89	0.039
38	SSA_2122	hypothetical protein	-8.85	0.04
39	SSA_1818	very short patch repair endonuclease	-8.79	0.041
40	SSA_0121	hypothetical protein	-8.71	0.042
41	SSA_0758	bifunctional ornithine acetyltransferase/N-acetylglutamate synthase (<i>argJ</i>)	-8.7	0.042
42	SSA_1812	modification methylase	-8.65	0.043
43	SSA_1254	hypothetical protein	-8.64	0.043
44	SSA_2220	galactosyltransferase	-8.59	0.044
45	SSA_1727	branched-chain amino acid ABC transporter permease (<i>livM</i>)	-8.53	0.045
46	SSA_2113	hypothetical protein	-8.52	0.045
47	SSA_2379	signal transduction protein (<i>comD</i>)	-8.5	0.046
48	SSA_0906	CshA-like fibrillar surface protein C (<i>crpC</i>)	-8.5	0.046
49	SSA_0301	hypothetical protein	-8.42	0.048
50	SSA_0905	CshA-like fibrillar surface protein B (<i>crpB</i>)	-8.35	0.049
51	SSA_1849	primosome assembly protein PriA (<i>priA</i>)	-8.33	0.049
52	SSA_1230	hypothetical protein	-8.31	0.05

Table S2 Predicted target genes for csRNA1-2

Rank	Locus tag	Gene description (Gene)	Energy	Pvalue
1	SSA_2317	Tfp pilus assembly protein, pilus retraction ATPase PilT	-15.87	0
2	SSA_2345	hypothetical protein	-14.36	0.001
3	SSA_2216	LPS biosynthesis protein (<i>lciD1</i>)	-14.01	0.002
4	SSA_1655	hypothetical protein	-13.19	0.003
5	SSA_2217	Cps9H	-12.43	0.006
6	SSA_1635	hypothetical protein	-11.89	0.008
7	SSA_0227	collagen-binding surface protein	-11.64	0.009
8	SSA_0718	hypothetical protein	-11.59	0.01
9	SSA_1632	surface protein	-11.36	0.011
10	SSA_2177	hypothetical protein	-11.35	0.011
11	SSA_1702	hypothetical protein	-11.2	0.012
12	SSA_0991	deoxyribonuclease	-11.16	0.012
13	SSA_1168	hypothetical protein	-11.06	0.013
14	SSA_0521	ethanolamine utilization protein EutL (<i>eutL</i>)	-10.88	0.015
15	SSA_1018	Zinc metalloprotease zmpC	-10.71	0.016
16	SSA_2131	DNA-binding protein	-10.67	0.016
17	SSA_1736	L-cysteine desulphydrase	-10.67	0.016
18	SSA_1809	PTS system glucose-specific EIIC BA component (<i>ptsG</i>)	-10.64	0.017
19	SSA_0131	30S ribosomal protein S11 (<i>rpsK</i>)	-10.56	0.018
20	SSA_0297	malate dehydrogenase (<i>mleS</i>)	-10.51	0.018
21	SSA_1355	hypothetical protein	-10.45	0.019
22	SSA_0737	arginine deiminase (<i>sagP</i>)	-10.21	0.021
23	SSA_2004	Zinc metalloprotease zmpB (<i>zmpB</i>)	-10.12	0.022
24	SSA_2205	transcription antitermination protein NusG (<i>nusG</i>)	-10.04	0.023
25	SSA_0061	tagatose 1,6-diphosphate aldolase	-9.96	0.024
26	SSA_1544	hypothetical protein	-9.84	0.025
27	SSA_0146	DNA repair ATPase	-9.82	0.025
28	SSA_0535	propanediol utilization: dehydratase, medium subunit	-9.62	0.028
29	SSA_1634	Heme utilization/adhesion exoprotein	-9.58	0.029
30	SSA_1110	hypothetical protein	-9.53	0.029
31	SSA_0133	50S ribosomal protein L17 (<i>rpQ</i>)	-9.45	0.03
32	SSA_1400	pyridoxamine kinase (<i>pdxK</i>)	-9.44	0.031
33	SSA_0448	excinuclease ABC subunit A (<i>uvrA</i>)	-9.31	0.032
34	SSA_2301	S-layer protein (<i>lytB</i>)	-9.3	0.033
35	SSA_0849	Signal peptidase I (<i>sip</i>)	-9.1	0.036
36	SSA_2208	preprotein translocase subunit SecE (<i>secE</i>)	-9.08	0.036
37	SSA_1044	homoserine kinase (<i>thrB</i>)	-9.05	0.036
38	SSA_1020	hypothetical protein	-9.01	0.037
39	SSA_1987	hypothetical protein	-8.98	0.038
40	SSA_2309	fimbrial assembly protein	-8.95	0.038
41	SSA_1261	ribose-5-phosphate isomerase A (<i>rpiA</i>)	-8.84	0.04
42	SSA_1380	hypothetical protein	-8.84	0.04
43	SSA_1818	very short patch repair endonuclease	-8.79	0.041
44	SSA_1699	galactose-6-phosphate isomerase subunit LacA (<i>lacA</i>)	-8.79	0.041
45	SSA_0785	ATP synthase F0F1 subunit delta (<i>uncH</i>)	-8.77	0.041
46	SSA_1563	beta-lactamase superfamily hydrolase (<i>vicX</i>)	-8.75	0.041
47	SSA_0121	hypothetical protein	-8.71	0.042
48	SSA_1758	hypothetical protein	-8.65	0.043
49	SSA_1812	modification methylase	-8.65	0.043
50	SSA_0088	V-type sodium ATPase subunit C (<i>ntpC</i>)	-8.63	0.044
51	SSA_2113	hypothetical protein	-8.52	0.045
52	SSA_0906	CshA-like fibrillar surface protein C (<i>crpC</i>)	-8.5	0.046
53	SSA_0905	CshA-like fibrillar surface protein B (<i>crpB</i>)	-8.35	0.049
54	SSA_0114	50S ribosomal protein L16 (<i>rpP</i>)	-8.34	0.049
55	SSA_1121	cytochrome C-type biogenesis protein	-8.33	0.049
56	SSA_1230	hypothetical protein	-8.31	0.05

Table S3 Predicted target genes for csRNA1–3

Rank	Locus tag	Gene description (Gene)	Energy	P value
1	SSA_2317	Tfp pilus assembly protein, pilus retraction ATPase PilT	-15.9	0
2	SSA_2216	LPS biosynthesis protein (<i>licD1</i>)	-14	0.002
3	SSA_1655	hypothetical protein	-13.2	0.003
4	SSA_1044	homoserine kinase (<i>thrB</i>)	-12.7	0.005
5	SSA_1230	hypothetical protein	-12.6	0.005
6	SSA_0297	malate dehydrogenase (<i>mleS</i>)	-12.5	0.005
7	SSA_1634	Heme utilization/adhesion exoprotein	-12.5	0.005
8	SSA_2217	Cps9H	-12.4	0.006
9	SSA_1635	hypothetical protein	-11.9	0.008
10	SSA_2345	hypothetical protein	-11.8	0.009
11	SSA_0718	hypothetical protein	-11.6	0.01
12	SSA_1359	arginine/histidine ABC transporter permease	-11.5	0.01
13	SSA_1168	hypothetical protein	-11.5	0.01
14	SSA_0227	collagen-binding surface protein	-11.4	0.011
15	SSA_0961	ketopantoate reductase PanE/ApbA	-11.4	0.011
16	SSA_0521	ethanolamine utilization protein EutL (<i>eutL</i>)	-11.2	0.012
17	SSA_1702	hypothetical protein	-11.2	0.012
18	SSA_0991	deoxyribonuclease	-11.2	0.012
19	SSA_1023	von Willebrand factor A	-10.9	0.014
20	SSA_1102	hypothetical protein	-10.7	0.016
21	SSA_1758	hypothetical protein	-10.7	0.016
22	SSA_0131	30S ribosomal protein S11 (<i>rpsK</i>)	-10.6	0.018
23	SSA_1961	amino acid ABC transporter substrate-binding protein	-10.4	0.019
24	SSA_0077	oxidoreductase	-10.4	0.02
25	SSA_1809	PTS system glucose-specific EIIC BA component (<i>ptsG</i>)	-10.3	0.02
26	SSA_1987	hypothetical protein	-10.3	0.02
27	SSA_0737	arginine deiminase (<i>sagP</i>)	-10.2	0.021
28	SSA_0905	CshA-like fibrillar surface protein B (<i>crpB</i>)	-10.2	0.021
29	SSA_2245	recombinase A (<i>recA</i>)	-10.2	0.021
30	SSA_2208	preprotein translocase subunit SecE (<i>secE</i>)	-10.2	0.022
31	SSA_1340	Zn/Mn ABC transporter	-10.1	0.022
32	SSA_0061	tagatose 1,6-diphosphate aldolase	-9.96	0.024
33	SSA_1261	ribose-5-phosphate isomerase A (<i>rpiA</i>)	-9.89	0.025
34	SSA_0066	hypothetical protein	-9.72	0.027
35	SSA_1018	Zinc metalloprotease zmpC	-9.7	0.027
36	SSA_0535	propanediol utilization: dehydratase, medium subunit	-9.62	0.028
37	SSA_2121	cell wall surface anchor family protein	-9.55	0.029
38	SSA_1110	hypothetical protein	-9.53	0.029
39	SSA_2318	PilB-like pili biogenesis ATPase	-9.46	0.03
40	SSA_0906	CshA-like fibrillar surface protein C (<i>crpC</i>)	-9.34	0.032
41	SSA_0343	DNA polymerase IV (<i>dinP</i>)	-9.33	0.032
42	SSA_1148	Beta-glucosides PTS, EIIC	-9.33	0.032
43	SSA_0448	excinuclease ABC subunit A (<i>uvrA</i>)	-9.31	0.032
44	SSA_2301	S-layer protein (<i>lytB</i>)	-9.3	0.033
45	SSA_0541	acetate kinase	-9.13	0.035
46	SSA_1248	hypothetical protein	-8.99	0.037
47	SSA_0067	acyltransferase	-8.97	0.038
48	SSA_2113	hypothetical protein	-8.96	0.038
49	SSA_1380	hypothetical protein	-8.84	0.04
50	SSA_1317	hypothetical protein	-8.75	0.041
51	SSA_0121	hypothetical protein	-8.71	0.042
52	SSA_0617	membrane protease subunit	-8.65	0.043
53	SSA_0064	hypothetical protein	-8.61	0.044
54	SSA_0425	glycosyltransferase	-8.41	0.048
55	SSA_0114	50S ribosomal protein L16 (<i>rplP</i>)	-8.34	0.049
56	SSA_1121	cytochrome C-type biogenesis protein	-8.33	0.049
57	SSA_1633	FimA fimbrial subunit-like protein	-8.31	0.05

Table S4 Predicted target genes for csRNA2

Rank	Locus tag	Gene description (Gene)	Energy	Pvalue
1	SSA_1229	hypothetical protein	-12.65	0.005
2	SSA_1475	hypothetical protein	-12.43	0.006
3	SSA_1184	DNA topoisomerase I (<i>topA</i>)	-11.54	0.01
4	SSA_1367	hypothetical protein	-11.29	0.012
5	SSA_1164	hypothetical protein	-10.87	0.015
6	SSA_0230	hypothetical protein	-10.82	0.015
7	SSA_1878	hypothetical protein	-9.18	0.034
8	SSA_1422	hypothetical protein	-9.06	0.036
9	SSA_1859	Holliday junction-specific endonuclease (<i>recU</i>)	-8.6	0.044
10	SSA_0666	hypothetical protein	-8.31	0.05

Table S5 Predicted target genes for csRNA7

Rank	Locus tag	Gene description (Gene)	Energy	P value
1	SSA_0847	6-phosphofructokinase (<i>pfk</i>)	-12.69	0.005
2	SSA_1380	hypothetical protein	-12.48	0.005
3	SSA_1601	SsrA-binding protein (<i>smpB</i>)	-12.15	0.007
4	SSA_1390	hypothetical protein	-12.15	0.007
5	SSA_2345	hypothetical protein	-12.09	0.007
6	SSA_2352	nitrate/sulfonate/bicarbonate ABC transporter periplasmic protein	-12.09	0.007
7	SSA_1327	hypothetical protein	-11.88	0.008
8	SSA_0061	tagatose 1,6-diphosphate aldolase	-11.47	0.01
9	SSA_0684	fibril-like structure subunit FibA	-11.21	0.012
10	SSA_2309	fimbrial assembly protein	-10.86	0.015
11	SSA_1474	lipoprotein	-10.55	0.018
12	SSA_1984	cell surface SD repeat-containing protein	-10.48	0.018
13	SSA_2301	S-layer protein (<i>lytB</i>)	-10.18	0.021
14	SSA_0303	surface protein C (<i>sspC</i>)	-10.03	0.023
15	SSA_1022	group 1 glycosyl transferase	-9.91	0.024
16	SSA_0258	hypothetical protein	-9.78	0.026
17	SSA_2299	hypothetical protein	-9.78	0.026
18	SSA_1657	hypothetical protein	-9.77	0.026
19	SSA_2313	hypothetical protein	-9.74	0.027
20	SSA_0784	ATP synthase FOF1 subunit B (<i>uncF</i>)	-9.59	0.029
21	SSA_2279	hypothetical protein	-9.56	0.029
22	SSA_2254	Holliday junction DNA helicase RuvA (<i>rvuA</i>)	-9.5	0.03
23	SSA_1632	surface protein	-9.39	0.031
24	SSA_0967	hypothetical protein	-9.35	0.032
25	SSA_2217	Cps9H	-9.34	0.032
26	SSA_0035	bifunctional phosphoribosylaminoimidazolecarboxamide formyltransferase (<i>purH</i>)	-9.29	0.033
27	SSA_2222	tyrosine-protein kinase Wze	-9.23	0.034
28	SSA_1019	collagen-binding surface protein	-9.16	0.035
29	SSA_1225	branched-chain amino acid aminotransferase (<i>ilvE</i>)	-9.15	0.035
30	SSA_2216	LPS biosynthesis protein (<i>licD1</i>)	-9.12	0.035
31	SSA_2017	hypothetical protein	-9.12	0.035
32	SSA_1466	prephenate dehydrogenase (<i>tyrA</i>)	-9.11	0.035
33	SSA_1985	hypothetical protein	-9.02	0.037
34	SSA_1231	hypothetical protein	-9.02	0.037
35	SSA_0737	arginine deiminase (<i>sagP</i>)	-8.8	0.041
36	SSA_0565	hypothetical protein	-8.73	0.042
37	SSA_0227	collagen-binding surface protein	-8.71	0.042
38	SSA_1113	two-component response transcriptional regulator	-8.65	0.043
39	SSA_1989	ABC transporter ATPase	-8.63	0.044
40	SSA_2312	hypothetical protein	-8.55	0.045
41	SSA_0761	XRE family transcriptional regulator	-8.54	0.045
42	SSA_1492	hypothetical protein	-8.47	0.047
43	SSA_2282	Phage infection protein	-8.33	0.049
44	SSA_1230	hypothetical protein	-8.31	0.05
45	SSA_1547	HPr kinase/phosphorylase (<i>ptsK</i>)	-8.29	0.05

Table S6 Predicted target genes for csRNA8

Rank	Locus tag	Gene description (Gene)	Energy	Pvalue
1	SSA_1655	hypothetical protein	-14.6	0.001
2	SSA_0158	hypothetical protein	-12.57	0.005
3	SSA_2230	anaerobic ribonucleoside triphosphate reductase (<i>nrdD</i>)	-12.51	0.005
4	SSA_1842	two-component response transcriptional regulator	-11.11	0.013
5	SSA_0678	ArgR family transcriptional regulator (<i>ahrC</i>)	-10.81	0.015
6	SSA_0515	propanediol utilization protein PduU	-10.35	0.02
7	SSA_1612	diacylglycerol kinase (<i>dgk</i>)	-10.14	0.022
8	SSA_0830	glycosyltransferase	-10.09	0.022
9	SSA_1533	glutathione reductase (<i>gor</i>)	-9.96	0.024
10	SSA_0718	hypothetical protein	-9.89	0.025
11	SSA_0162	hypothetical protein	-9.78	0.026
12	SSA_1044	homoserine kinase (<i>thrB</i>)	-9.53	0.029
13	SSA_0785	ATP synthase F0F1 subunit delta (<i>uncH</i>)	-9.49	0.03
14	SSA_1173	lipoate protein ligase A (<i>lpA</i>)	-9.21	0.034
15	SSA_0139	copper transport operon or penicillinase transcriptional repressor	-9.2	0.034
16	SSA_1230	hypothetical protein	-9	0.037
17	SSA_1634	Heme utilization/adhesion exoprotein	-8.97	0.038
18	SSA_1221	L-lactate dehydrogenase (<i>ldh</i>)	-8.95	0.038
19	SSA_1929	macrophage infectivity potentiator protein (<i>mip</i>)	-8.87	0.039
20	SSA_2312	hypothetical protein	-8.62	0.044
21	SSA_1762	permease	-8.6	0.044
22	SSA_1995	hypothetical protein	-8.48	0.046
23	SSA_1835	late competence protein (<i>comFC</i>)	-8.46	0.047
24	SSA_0507	hypothetical protein	-8.43	0.047
25	SSA_2119	pentose-5-phosphate-3-epimerase (<i>rpe</i>)	-8.42	0.047
26	SSA_0121	hypothetical protein	-8.33	0.049

Table S7 *Streptococcus sanguinis* strains used in this study

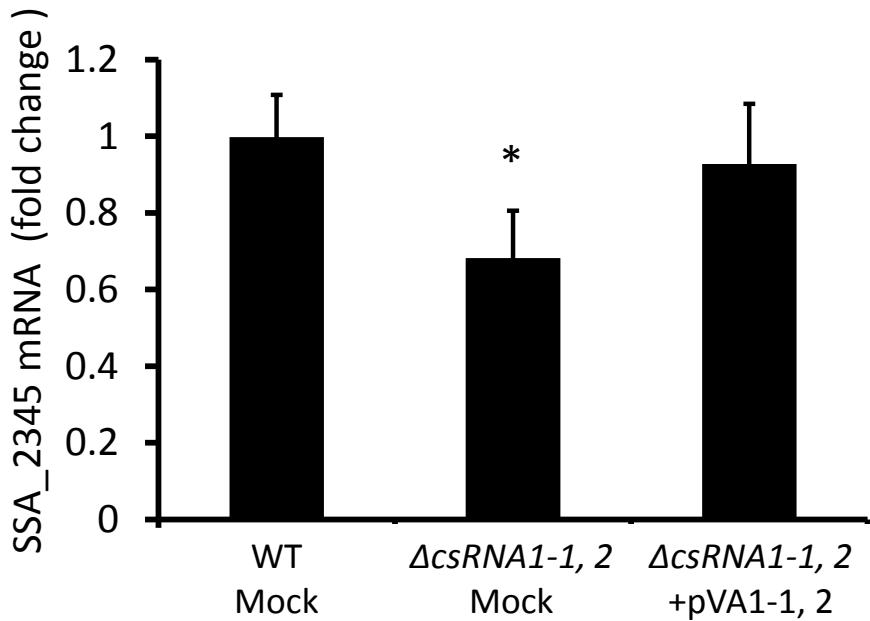
Strain Name	Information	Reference or source
10556	<i>Streptococcus sanguinis</i> type strain	ATCC
WT Mock	10556 containing empty shuttle vector pVA838	This study
$\Delta ciaRH$ Mock	10556 derivative in which the <i>ciaRH</i> gene has been replaced with a spectinomycin resistance cassette; also contains empty shuttle vector pVA838	This study
$\Delta ciaRH$ +pVAciaRH	$\Delta ciaRH$ containing the pVA838 derivative pVAciaRH, which contains a wild-type <i>ciaRH</i> allele downstream of the natural promoter	This study
$\Delta csRNA1\text{-}1, 2$ Mock	10556 derivative in which the <i>csRNA1\text{-}1</i> and <i>csRNA1\text{-}2</i> genes have been replaced with a spectinomycin resistance cassette; also contains empty shuttle vector pVA838	This study
$\Delta csRNA1\text{-}1, 2$ +pVA1\text{-}1, 2	$\Delta csRNA1\text{-}1, 2$ containing the pVA838 derivative pVA1\text{-}1, 2, which contains wild-type <i>csRNA1\text{-}1</i> and <i>csRNA1\text{-}2</i> alleles downstream of their natural promoters	This study
WT-luc Mock	10556 derivative in which promoter-less luciferase (<i>NanoLuc</i>) gene and a tetracycline resistance cassette have been substituted with <i>pilT</i> gene; also contains empty shuttle vector pVA838	This study
$\Delta csRNA1\text{-}1, 2-luc Mock$	WT-luc derivative in which the <i>csRNA1\text{-}1</i> and <i>csRNA1\text{-}2</i> genes have been replaced with a spectinomycin resistance cassette; also contains empty shuttle vector pVA838	This study
$\Delta csRNA1\text{-}1, 2-luc+pVA1\text{-}1, 2$	$\Delta csRNA1\text{-}1, 2$ -luc containing the pVA838 derivative pVA1\text{-}1, 2	This study
$\Delta csRNA1\text{-}1, 2-luc+pVA1\text{-}1$	$\Delta csRNA1\text{-}1, 2$ -luc containing the pVA838 derivative pVA1\text{-}1, which contains wild-type <i>csRNA1\text{-}1</i> allele downstream of the natural promoter	This study
$\Delta csRNA1\text{-}1, 2$ -luc+pVA1\text{-}2	$\Delta csRNA1\text{-}1, 2$ -luc containing the pVA838 derivative pVA1\text{-}2, which contains wild-type <i>csRNA1\text{-}2</i> allele downstream of the natural promoter	This study
$\Delta csRNA1\text{-}1, 2-luc+pVA1\text{-}1 mut\text{-}1$	$\Delta csRNA1\text{-}1, 2$ -luc containing the pVA1\text{-}1 derivative pVA1\text{-}1 mut\text{-}1, which contains seven nucleotide substitutions within the putative region for base-pairing with <i>pilT</i> mRNA	This study
$\Delta csRNA1\text{-}1, 2$ -luc+pVA1\text{-}1 mut\text{-}2	$\Delta csRNA1\text{-}1, 2$ -luc containing the pVA1\text{-}1 derivative pVA1\text{-}1 mut\text{-}2, which contains four nucleotide substitutions within the putative region for base-pairing with <i>pilT</i> mRNA	This study
$\Delta csRNA1\text{-}1, 2$ -luc+pVA1\text{-}1 mut\text{-}3	$\Delta csRNA1\text{-}1, 2$ -luc containing the pVA1\text{-}1 derivative pVA1\text{-}1 mut\text{-}3, which contains three nucleotide substitutions within the putative region for base-pairing with <i>pilT</i> mRNA	This study

Table S8 Oligonucleotide primers used in the study

Primer name	Sequence (5'-3')	Description
CiaR-up-Hind-F	TATAAGCTTCCTTGCACAGCTATTACGG	Primer to amplify the upstream fragment of <i>ciaRH</i> in the creation of <i>ciaRH</i> deletion mutant and to amplify <i>ciaRH</i> containing fragment in the creation of plasmid pVAciaRH
CiaR-up-Pst-R	TATCTGCAGTAAGTTTTCTACGACTGCGAC	Primer to amplify the upstream fragment of <i>ciaRH</i> in the creation of <i>ciaRH</i> deletion mutant
CiaR-down-Sma-F	TATCCCGGGCTCTCGTAGTGTGGCTAC	Primer to amplify the downstream fragment of <i>ciaRH</i> in the creation of <i>ciaRH</i> deletion mutant
CiaR-down-Eco-R	TATGAATTCTGCTGACCAAAGCTGTTGCC	Primer to amplify the downstream fragment of <i>ciaRH</i> in the creation of <i>ciaRH</i> deletion mutant
CiaH-down-Sal-R	CCTTGCTCTAAAGTCGACAGC	Primer to amplify <i>ciaRH</i> containing fragment in the creation of plasmid pVAciaRH
csRNA1-1_real-F	TGAAGCCCCCTAACCTTGTTC	RT-PCR primer for csRNA1-1
csRNA1-1_real-R	CCAGATTGGGTGGCCTTAT	RT-PCR primer for csRNA1-1
csRNA1-2-real-F	GAAGACCTCTAACCTTATTTG	RT-PCR primer for csRNA1-2
csRNA1-2-real-R	CCGGATTGGGTGGCCTTAT	RT-PCR primer for csRNA1-2
csRNA1-3-real-F	CACTGGGTGACTTCAGGA	RT-PCR primer for csRNA1-3
csRNA1-3-real-R	CAAAGACCTCTAACCTTGT	RT-PCR primer for csRNA1-3
csRNA2-real-F	TTGGGCGGTTCTTATAGGGA	RT-PCR primer for csRNA2
csRNA2-real-R	GATCTCCTAACCTTGTTC	RT-PCR primer for csRNA2
csRNA7-real-F	TTAACTTAACCTCTAAAACCTTTTC	RT-PCR primer for csRNA7
csRNA7-real-R	AAACCACCGTTTGGTGGCT	RT-PCR primer for csRNA7
csRNA8-real-F	AGCTTGAAGACTAGAAGAC	RT-PCR primer for csRNA8
csRNA8-real-R	GCTTGTGATTGTTAGGAG	RT-PCR primer for csRNA8
SSA_2345-real-F	AAACGCCGAAAAACCAAGGC	RT-PCR primer for SSA_2345
SSA_2346-real-R	GGCTGATACCAGCAGCTTGA	RT-PCR primer for SSA_2345
licD1-real-F	GTTCTTGCACATGGAGGC	RT-PCR primer for <i>licD1</i>
licD2-real-R	CGCATCCGCAGGAGAGATTA	RT-PCR primer for <i>licD1</i>
Sang16S-F	CGTAAACGATGAGTGCTAGGTG	RT-PCR primer for 16S ribosomal RNA
Sang16S-R	TAGAGCGGTAGAGGGATGT	RT-PCR primer for 16S ribosomal RNA
csRNA1-up-Sph-F	TATGCATCGGCTGATGCAGAGCGTGAC	Primer to amplify the upstream fragment of <i>csRNA1-1</i> in the creation of <i>csRNA1-1</i> , <i>csRNA1-2</i> double deletion mutant
csRNA1-up-Sal-R	TATGTCGACGGAGGCTTCCCTGCTC	Primer to amplify the upstream fragment of <i>csRNA1-1</i> in the creation of <i>csRNA1-1</i> , <i>csRNA1-2</i> double deletion mutant
csRNA1-down-Sma-F	TATCCCGGGTTCAGACGTGGATTAG	Primer to amplify the downstream fragment of <i>csRNA1-2</i> in the creation of <i>csRNA1-1</i> , <i>csRNA1-2</i> double deletion mutant
csRNA1-down-Eco-R	TATGAATTCGCGACTGGCTCACCTGAG	Primer to amplify the downstream fragment of <i>csRNA1-2</i> in the creation of <i>csRNA1-1</i> , <i>csRNA1-2</i> double deletion mutant
csRNA-Sph-F	TATGCATCGAAACAATCGCGAAAGAGC	Used in the creation of <i>csRNA1-1</i> and/or <i>csRNA1-2</i> complementation plasmid pVA1-1, 2 and pVA1-1
csRNA-Sal-R	TATGTCGACTAACCGACGTCTGAAC	Used in the creation of <i>csRNA1-1</i> and/or <i>csRNA1-2</i> complementation plasmid pVA1-1, 2 and pVA1-2
csRNA1-1-Sal-R	TATGTCGACAATCCGATAAACCTCCCGTC	Used in the creation of <i>csRNA1-1</i> complementation plasmid pVA1-1

csRNA1-2-Sph-F	TATGCATGCGAATAAGACGGGAAGTTTATCGG	Used in the creation of <i>csRNA1-1</i> complementation plasmid pVA1-2
csRNA1-1-mut-1-F	CCCCTAACTTTGTTGATAATT CGGGTTTTCTATAATAATCTCCCC	Used in the creation of <i>csRNA1-1</i> mutant expression plasmid pVA1-1 mut-1
csRNA1-1-mut-1-R	GGGAGATTATTATGAAAAAACCGAAATTATCAAACAAAGTTAGGGG	Used in the creation of <i>csRNA1-1</i> mutant expression plasmid pVA1-1 mut-1
csRNA1-1-mut-2-F	CTAACTTTGTTGATAATT CGGGTTTTCTATAATAATCT GGG	Used in the creation of <i>csRNA1-1</i> mutant expression plasmid pVA1-1 mut-2
csRNA1-1-mut-2-R	GGGAGATTATTATGAAAAAACCCAGGATTATCAAACAAAGTTAGG	Used in the creation of <i>csRNA1-1</i> mutant expression plasmid pVA1-1 mut-2
csRNA1-1-mut-3-F	CTAACTTTGTTGATAATT CGGGTTTTCTATAATAATCT GGGAGATTATTATGAAAAAGTTGAAATTATCAAACAAAGTTAGG	Used in the creation of <i>csRNA1-1</i> mutant expression plasmid pVA1-1 mut-3
csRNA1-1-mut-3-R	GGGAGATTATTATGAAAAAGTTGAAATTATCAAACAAAGTTAGG	Used in the creation of <i>csRNA1-1</i> mutant expression plasmid pVA1-1 mut-3
pilT-real-F	AAACAAC TGCGGGATTGCC	RT-PCR primer for <i>pilT</i>
pilT-real-R	GCTTCCTGTTGGCCCCGGTAA	RT-PCR primer for <i>pilT</i>
pilT-up-Sph-F	TATGCATGCTGGACTGACCTCGCGAGTG	Primer to amplify the upstream fragment of <i>pilT</i> fused to <i>NanoLuc</i> in the creation of PilT reporter strain
pilT-up-luc-R	CAACGAAATCTCGAGTGTGAAGACCATCCTAAACTTCCCCTTCTAGACTAAAGGC	Primer to amplify the upstream fragment of <i>pilT</i> fused to <i>NanoLuc</i> in the creation of PilT reporter strain
pilT-up-luc-F	GCCTTAGTCTAGAAGGGGAAGTTAGGATGGTCTTCACACTCGAAGATTTCGTTG	Primer to amplify <i>NanoLuc</i> fused to the upstream fragment of pilT in the creation of PilT reporter strain
luc-Sal-R	TATGTCGACTTACGCCAGAATCGGTTCGCACAGC	Primer to amplify <i>NanoLuc</i> fused to the upstream fragment of <i>pilT</i> in the creation of PilT reporter strain
pilT-down-Sma-F	TATCCCGGCCTACTCAGTGGATCGCGCTCAAGTTG	Primer to amplify the downstream fragment of <i>pilT</i> in the creation of PilT reporter strain
pilT-down-Eco-R	TATGAATTCCCTTCCAGCACCTCGTCC	Primer to amplify the downstream fragment of <i>pilT</i> in the creation of PilT reporter strain
T7-pilT-F	CTAATACGACTCACTATAGGGAGAGCCTTACTAGTCTAGAAGGGAAAG	Primer to amplify <i>pilT</i> (WT) and place downstream of T7 promoter for use as template in the <i>in vitro</i> transcription
T7-pilT-mut-F	CTAATACGACTCACTATAGGGAGAGCCTTACTAGTCTAGAAGG GGAAACCCAGGATGAATTGGATGAGTTAAGCAGGC	Primer to amplify <i>pilT</i> (mut) and place downstream of T7 promoter for use as template in the <i>in vitro</i> transcription
pilT-R	ATATACTGTCATGCTCTAGTACC	Primer to amplify <i>pilT</i> (WT) or (mut) and place downstream of T7 promoter for use as template in the <i>in vitro</i> transcription
T7-csRNA1-1-R	AAAAAAAGACCACCAAGATTGGGTGGCCTTATGGGAGATTA TTATGAAAAAGTTAGGATTATCAAACAAAGTTAGGGGGCT TCATTTGATTCTCCCTATAGTGAGTCGTATTAG	Used with T7-F as template for <i>in vitro</i> transcription of csRNA1-1
T7-F	CTAATACGACTCACTATAGGGAGA	Used with T7-csRNA1-1-R or with T7-csRNA1-1-mut-2-R as template for <i>in vitro</i> transcription of csRNA1-1 or csRNA1-1 mut-2 respectively
T7-csRNA1-1-mut2-R	AAAAAAAGACCACCAAGATTGGGTGGCCTTATGGGAGATTA TTATGAAAAAACCCAGGATTATCAAACAAAGTTAGGGGGCT TCATTTGATTCTCCCTATAGTGAGTCGTATTAG	Used with T7-F as template for <i>in vitro</i> transcription of csRNA1-1 mut-2

A



B

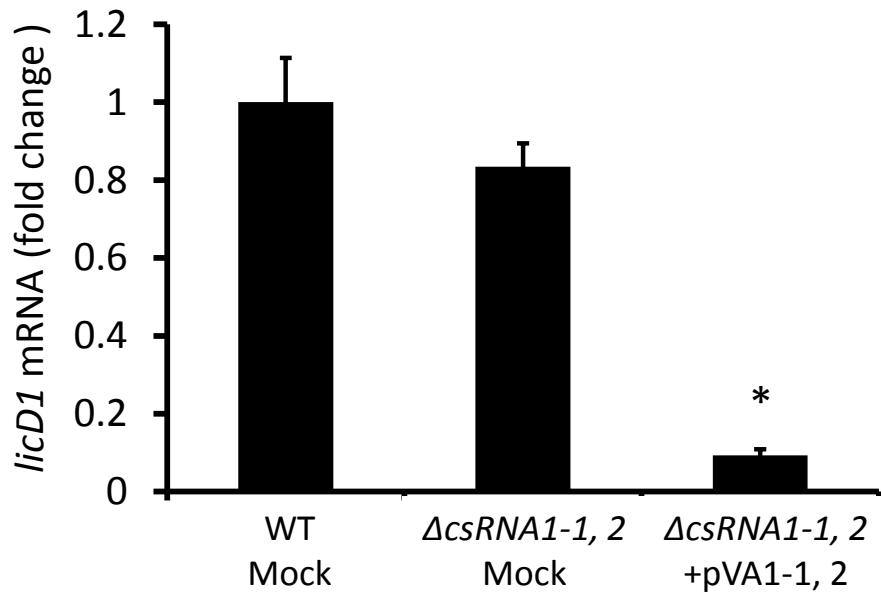


FIG S1 Quantitative RT-PCR analysis of *SSA_2345* (A) and *licD1* (B) expressions in *S. sanguinis*. $\Delta csRNA1-1, 2$ strain was transformed with either complementation plasmid pVA1-1,2 or empty vector pVA838 (Mock). Strains were grown in BHI broth to the early exponential phase, and RNA was isolated and used in quantitative RT-PCR analysis. Data are expressed as mean \pm SD from triplicate experiments. *, P<0.01.

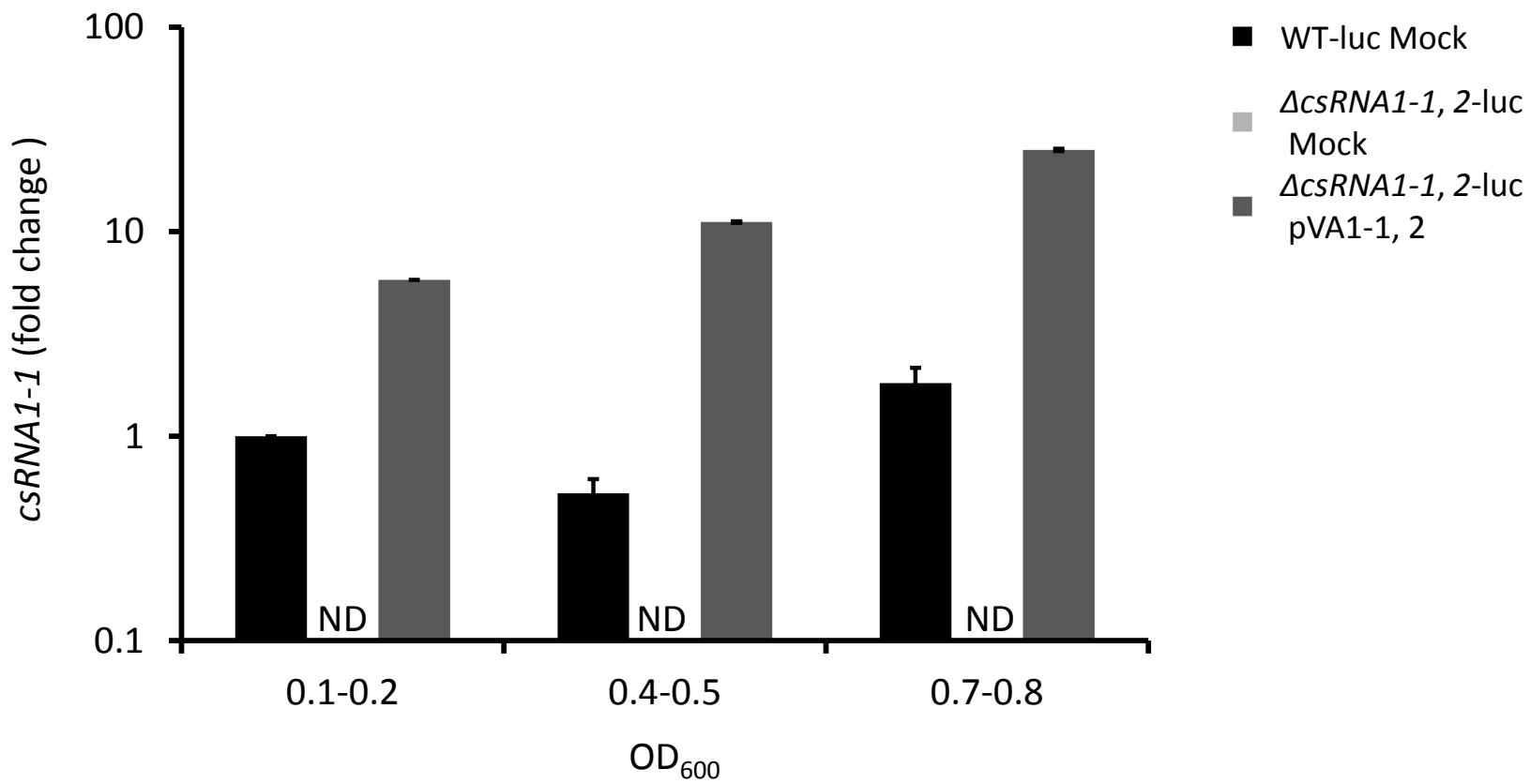


FIG S2 Growth phase-dependent expression of *csRNA1-1*. PilT-luciferase reporter strains were grown over a 6-h period, and samples were collected for RNA preparation at OD₆₀₀ of 0.1–0.2 (early-log phase), 0.4–0.5 (mid-log phase) and 0.7–0.8 (late-log phase). The extracted RNA was used in quantitative RT-PCR analysis of *csRNA1-1* expression. Data are shown as fold change relative to WT-luc Mock strain at OD₆₀₀=0.1–0.2, and are expressed as mean ± SD from triplicate experiments. The vertical axis is shown in logarithmic scale. ND, not detected.