

Supplementary File

Table 1. List of Genes and Primer Sequences used for transcriptome analysis by qRT-PCR.

S. no	Gene name	Gene function	Forward primer sequence	Reverse primer sequence	Reference
1	<i>ftsZ</i>	Cell division protein	CTGAGATGCCTGCTGCTGAA	GATTGCTGTGGCTCAGATG ATG	[1]
2	<i>gyrA</i>	Encodes DNA gyrase subunit A	TTCGTACAAGTGTGCCGA- TATCT	TCTAGGCGCATCACTTT- GACA	[1]
3	<i>gbpB</i>	Glucan binding protein - B	ATGGCGGTTATGGACACGTT	TTTGGCCACCTTGAACACCT	[2]
4	<i>relA</i>	Virulence phenotypes	ACAAAAAGGGTATCGTCCG- TACAT	AATCACGCTTGG- TATTGCTAATTG	[2]
5	<i>gtfC</i>	Extracellular polysaccharide synthesis	GGTTTAACGTCAAAATT- AGCTGTATTAGC	CTCAACCAACCGCCAC- TGTT	[2]
6	<i>atlA</i>	Autolysin like protein	GTTAGTTCTGGTTTTGACCG- CAAT	CCCTCAACAACAACATCAA AGGT	[2]
7	<i>spaP</i>	Adherence to saliva-coated tooth	GACTTTGG- TAATGGTTATGCATCAA	TTTGTATCAGCCG- GATCAAGTG	[2]
8	<i>immB</i>	Bacteriocin immunity proteins	GCTAGAGAGGCAAATGCACA	CAGCAGCAGCTGAGAA- GATG	[3]
10	<i>immA</i>	Bacteriocin immunity proteins	TCTCCCCTGCTTGTTCAGAT	GCTGGCAAATTCGCTTACTT	[3]
11	<i>bsmH</i>	Bacteriocin production	AGACATGTTAGCCGCTGTT- GAAG	AAGCGCCTGTTCCAATCG- TA	[3]
12	<i>bsmI</i>		GAAACAATGGATACAGA- GACG	GGAACAATAAGAGGAT- TTGG	[3]
13	<i>atpD</i>	Essential for acid tolerance	CCAGGCGGTTTCATTCATCTGA C	GGCGGGATTTCGG- TATTTACTG	[4]
14	<i>ldh</i>	Reversible enzyme in tricarboxylic acid cycle	ACTTCACTTGA- TACTGCTCGTT	AACACCAGCTACATTGG- CATGA	[5]
15	<i>dnaK</i>	Stress tolerance protein	GGTACAACAACTCAGCAG- TTGCAGTTCTT	CCCATCTTAGATTT- GATGGAAAGAATTGT	[6]
16	<i>recA</i>	Involved in DNA repair	GGATCCGAGAAAAA- GATTGGCCAAAAGAAT	TAAAGACTCGGGCTTGG- GACCTATTTTTAT	[7]
17	<i>brpA</i>	Cell division, stress tolerance, biofilm formation	GGAGGAGCTGCATCAG- GATTC	AACTCCAGCACATCCAG- CAAG	[2]
18	<i>comDE</i>	Competence histidine kinase and response regulator	ACAATTCCTTGAG- TTCCATCCAAG	TGGTCTGCTGCCTGTTGC	[2]
19	<i>comB</i>	Competence development	CCAGTCCAAACCGTCAGACT	GCTGCTTTCCTTGTCTTTCG	[3]
20	<i>comA</i>		ACGAGCCTAACAAGGGGATT	CCCTGAGGCATTTGTTCAAT	[3]
21	<i>covR</i>	Regulation of EPS synthesizing enzymes	ACACGATTACAGCCTTT- GATGG	CTTCTTAGCCACTTCAA- GACC	[1]
22	<i>vicR</i>	Promotes biofilm architecture	TGACACGATTACAGCCTTT- GATG	CGTCTAGTTCTGG- TAACATTAAGTCCAATA	[2]
23	<i>comX</i>	Competence development	CTGTTTGTCAAGTGGCGGTA	GCATACTTTGCCTTCCCAA	[3]
24	<i>luxS</i>	Autoinducer-2 synthesis	ACTGTTCCCTTTTGGCTGTC	AACTTGCTTT- GATGACTGTGGC	[8]

Table 2. The CI values for individual combinations of Baicalein and Fluoride. For all the tested combinations CI < 1 indicating synergy. (CI=1 indicates addictiveness while CI>1 indicates antagonism).

Baicalein Conc. (μM)	Fluoride Conc. (ppm)	Effect	CI	Interpretation
200.0	31.25	0.85	0.21	Synergistic
200.0	15.63	0.99	5.12E-6	Synergistic
200.0	7.81	0.93	0.09	Synergistic
200.0	3.9	0.89	0.16	Synergistic
200.0	1.95	0.96	0.05	Synergistic
100.0	31.25	0.63	0.57	Synergistic
100.0	15.63	0.81	0.16	Synergistic
100.0	7.81	0.87	0.095	Synergistic
100.0	3.9	0.69	0.32	Synergistic
100.0	1.95	0.77	0.20	Synergistic
50.0	31.25	0.62	0.40	Synergistic
50.0	15.63	0.8	0.08	Synergistic
50.0	7.81	0.79	0.09	Synergistic
50.0	3.9	0.62	0.24	Synergistic
50.0	1.95	0.47	0.72	Synergistic
25.0	31.25	0.63	0.25	Synergistic
25.0	15.63	0.69	0.1	Synergistic
25.0	7.81	0.69	0.09	Synergistic
25.0	3.9	0.53	0.32	Synergistic
25.0	1.95	0.49	0.38	Synergistic
12.5	15.63	0.6	0.20	Synergistic
12.5	7.81	0.68	0.05	Synergistic
12.5	3.9	0.53	0.24	Synergistic

References:

- [1] S. Hasan, K. Singh, M. Danisuddin, P.K. Verma, A.U. Khan, Inhibition of Major Virulence Pathways of *Streptococcus mutans* by Quercitrin and Deoxynojirimycin: A Synergistic Approach of Infection Control, *PLoS One*. 9 (2014) e91736. <https://doi.org/10.1371/journal.pone.0091736>.
- [2] M. Shemesh, A. Tam, D. Steinberg, Expression of biofilm-associated genes of *Streptococcus mutans* in response to glucose and sucrose, *J. Med. Microbiol.* 56 (2007) 1528–1535. doi:<https://doi.org/10.1099/jmm.0.47146-0>.
- [3] G. Kaur, P. Balamurugan, S.A. Princy, Inhibition of the Quorum Sensing System (ComDE Pathway) by Aromatic 1,3-di-m-tolylurea (DMTU): Cariostatic Effect with Fluoride in Wistar Rats, *Front. Cell. Infect. Microbiol.* . 7 (2017) 313. <https://www.frontiersin.org/article/10.3389/fcimb.2017.00313>.
- [4] V. Gabe, T. Kacergius, S. Abu-Lafi, P. Kalesinskas, M. Masalha, M. Falah, B. Abu-Farich, A. Melnikaitis, M. Zeidan, A. Rayan, Inhibitory Effects of Ethyl Gallate on *Streptococcus mutans* Biofilm Formation by Optical Profilometry and Gene Expression Analysis, *Molecules*. 24 (2019) 529. doi:10.3390/molecules24030529.
- [5] Z. He, Z. Huang, W. Jiang, W. Zhou, Antimicrobial Activity of Cinnamaldehyde on *Streptococcus mutans* Biofilms, *Front. Microbiol.* . 10 (2019) 2241. <https://www.frontiersin.org/article/10.3389/fmicb.2019.02241>.
- [6] Y. Matsumi, K. Fujita, Y. Takashima, K. Yanagida, Y. Morikawa, M. Matsumoto-Nakano, Contribution of glucan-binding protein A to firm and stable biofilm formation by *Streptococcus mutans*, *Mol. Oral Microbiol.* 30 (2015) 217–226. doi:10.1111/omi.12085.
- [7] S. Inagaki, M. Matsumoto-Nakano, K. Fujita, K. Nagayama, J. Funao, T. Ooshima, Effects of recom-

binase A deficiency on biofilm formation by *Streptococcus mutans*, *Oral Microbiol. Immunol.* 24 (2009) 104–108. doi:10.1111/j.1399-302X.2008.00480.x.

- [8] Z.T. Wen, D. Yates, S.-J. Ahn, R.A. Burne, Biofilm formation and virulence expression by *Streptococcus mutans* are altered when grown in dual-species model, *BMC Microbiol.* 10 (2010) 111. doi:10.1186/1471-2180-10-111.