

Figure S1: Sampling locations in the watershed used for the study.

Table S1. Sampling Locations for MSAR *Enterococcus* Evaluation Study.

Site #	Zones	No of samples	Site locations	Land use	Geographic positioning system (GPS)
N1	5-Natural sites	12	Ice House Canyon	Open Space	N34°15.057min.;W117° 37.977min;1447 m elevation
N2		12	Cucamonga Creek. @OCWD Ponds	Open Space	San Bernardino County Flood Control District (SBCFCD); 207 m elevation
UR2	1-urban runoff	12	Chino Creek @ Central Ave.	Urban runoff	N33° 58.420min.; W117° 41.302min;174 m elevation
UR3		12	Chino Creek @ Schaefer Ave.	Urban runoff	N34° 00.246min.; W117° 43.628min;207 m elevation
UR4		12	San Antonio Wash @ County Drive	Urban runoff + Commercial wash out	N30° 01.543min.; W117° 43.652min;222 m elevation;
UR5		12	Chino Creek. @ Riverside Drive	Urban runoff	N34° 01.144min.; W117° 44.204min; 207 m elevation;
UR9		12	Big League Dreams storm drain	Urban runoff	N33° 57.364min.; W117° 40.788min;163 m elevation;
UR13		12	Cucamonga Creek @ Regional Water Recycling Plant #1	Urban runoff+ wastewater	N34°;01.853min; W117° 35.946min; Altitude: 246 m
UR12		12	Chino Creek @ Pine Ave.	Urban runoff+ wastewater	N33°56.941min.;W117° 39.986min;155 m elevation;
CAFO6	2-Agri-runoff	12	Cypress Channel @ Schaefer Ave.	Agricultural runoff	N34° 00.262min.; W117° 39.766min, 208 m elevation;

CAFO7	12	Cypress Channel @ Kimball Ave.	Agricultural runoff	N33° 58.113min.; W117° 39.624min, 177m elevation;	
CAFO8	12	Cypress Channel @ Golf Course	Agricultural runoff	N33° 57.057min.; W117° 39.555min;160 m elevation;	
CAFO10	4	Dirt channel on Kimball	Agricultural Runoff	N33°58.109min.;W117° 40.286min 184 m elevation;	
UR13	4-WWTPs	6	Cucamonga Creek @ Regional Water Recycling Plant #1	Effluent from wastewater treatment plant and urban runoff	N34°;01.853min; W117° 35.946min; Altitude:246 m
WW13		6	Inland Empire Utilities Agency (IEUA) Regional Water Recycling Plant #5	Effluent from wastewater treatment plant	N33° 57.840min.; W117° 40.826min;180 m elevation;
WW14		6	IEUA Carbon Canyon Waste Reclamation Facility (CCWRF)	Effluent from wastewater treatment plant	N33° 58.799min.; W117° 41.655min;184 m elevation;
PRADO2	3-Prado Recreational Park	12	Santa Ana River @ Prado Dam	Urban Runoff+Agr	N33°;54.737min; W117° 38.711min Altitude: 141 m.
PRADO33		12	Prado Park outlet	Urban Runoff+ waste water discharge	N33°;56.402min; W117° 38.763min Altitude:166 m
PRADO5		12	Santa Ana River @ River road	Urban Runoff+Agr	N33°;55.405min; W117° 35.894min Altitude:155 m.
PRADO4		12	OCWD (Prado)Wetlands Effluent	Wetland treated (bacteria loaded) Orange County Water District (OCWD	N33°;54.737min; W117° 38.711min Altitude: 141 m

Table S2. Primers and PCR conditions used in this study

Primer type	Positive control	Direction	Sequence (5'-3')	Primer conc (pmol)	Annealing temp (°C)	Product size (bp)
Virulence gene (multiplex PCR)						
<i>asa1</i>	<i>E. faecalis</i> MMH 594	F	GCACGCTATTACGAACTATGA	0.1	56	375
		R	TAAGAAAGAACATCACCACGA			
<i>gelE</i>	<i>E. faecalis</i> MMH 594	F	TATGACAATGCTTTTGGGAT	0.1	56	213
		R	AGATGCACCCGAAATAATATA			
<i>cylA</i>	<i>E. faecalis</i> MMH 594	F	ACTCGGGGATTGATAGGC	0.2	56	688
		R	GCTGCTAAAGCTGCGCTT			
<i>esp</i>	<i>E. faecalis</i> MMH 594	F	AGATTTCATCTTTGATTCTTGG	0.2	56	510
		R	AATTGATTCTTTAGCATCTGG			
Antibiotic resistance gene Multiplex PCR						
<i>tet</i> (A) group I	<i>E. coli</i> HB101(RP1)	F	GCTACATCCTGCTTGCCCTTC	1	55	210
		R	CATAGATCGCCGTGAAGAGG			
<i>tet</i> (C) group I	<i>E. coli</i> DO7(pBR322)	F	CTTGAGAGCCTTCAACCCAG	0.25	55	418
		R	ATGGTCGTCATCTACCTGCC			

<i>tet</i> (K) group II	<i>Bacillus subtilis</i> BD99(pT181)	F	TCGATAGGAACAGCAGTA	1.25	55	169
		R	CAGCAGATCCTACTCCTT			
<i>tet</i> (O) group II	Cloned plasmid (O)	F	AACTTAGGCATTCTGGCTCAC	1.25	55	515
	pGEM-tet	R	TCCCACTGTTCCATATCGTCA			
<i>tet</i> (S) group II	Cloned plasmid pAT451	F	CATAGACAAGCCGTTGACC	0.5	55	667
		R	ATGTTTTTGGAAACGCCAGAG			
<i>tet</i> (Q) group I	Cloned plasmid pBT-1	F	TTATACTTCCTCCGGCATCG	1.25	55	904
		R	ATCGGTTTCGAGAATGTCCAC			

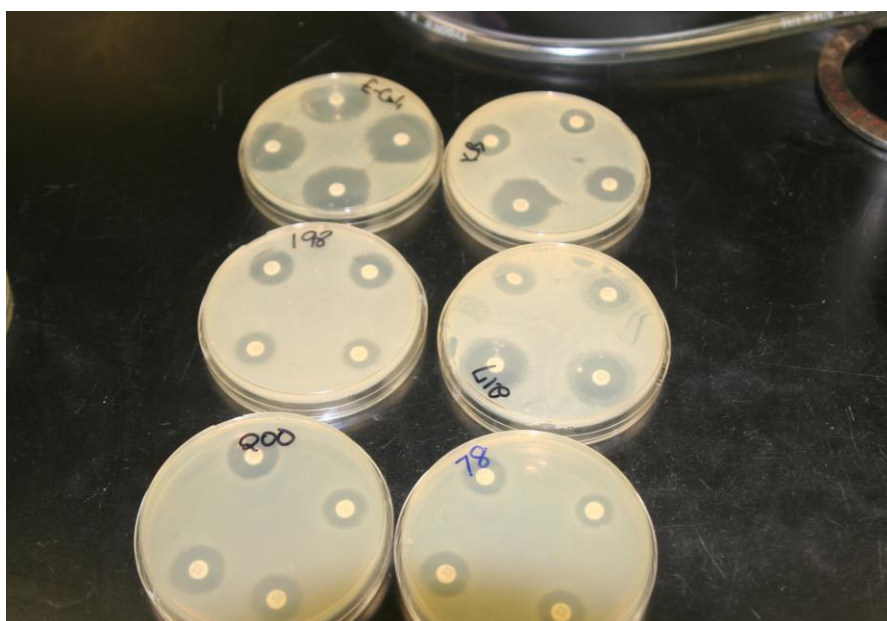


Figure S2. Disk diffusion assay displaying antibiotics resistance