

PREVALENCE OF ANTIBIOTIC RESISTANT GENES IN THE SAIGON RIVER IMPACTED BY ANTHROPOGENIC ACTIVITIES

Truong Thong^{1,3}, Hoang Thai Loc⁴, Tran Linh Thuoc^{1,3}, Pham Thi Phuong Thuy⁵, Le Thai-
Hoang^{2,3,6*}

¹Faculty of Biology and Biotechnology, University of Science, Ho Chi Minh City, Vietnam

²Department of Environmental Engineering, International University, Ho Chi Minh City,
Vietnam

³Vietnam National University, Ho Chi Minh City, Vietnam

⁴Vietnam Petroleum Institute - Research and Development Centre for Petroleum Safety and
Environment, Vietnam

⁵Faculty of Biotechnology, Ho Chi Minh City University of Food and Industry, Vietnam

⁶Faculty of Environmental and Food Engineering, Nguyen Tat Thanh University, Ho Chi Minh
City, Vietnam

*Corresponding author: Dr. Le Thai-Hoang, ^{2,3}Vietnam National University Ho Chi Minh City,
International University, Department of Environmental Engineering, Ho Chi Minh City,
Vietnam; ⁶Nguyen Tat Thanh University, Faculty of Environmental and Food Engineering, Ho
Chi Minh City, Viet Nam; Email: lthoang.bio@gmail.com, ORCID: 0000-0002-2508-1260

SUPPLEMENTARY MATERIALS

FIGURE S1. MAP OF ALL THE SAMPLING SITES ALONG THE SAIGON RIVER IN HO CHI MINH CITY ..3

TABLE S1. DETAILED INFORMATION OF RT-PCR PRIMERS FOR QUANTIFICATION OF THE ARGS, INTEGRONS, AND 16S RRNA GENE	4
TABLE S2. LIMIT OF DETECTION VALUES FOR ALL TARGET ARGS IN QPCR	5
TABLE S3. THE CONCENTRATION (COPIES/ML) OF THE ARGS, INTEGRONS, AND 16S RRNA GENE IN THE 12 SAMPLING SITES OF THE SAIGON RIVER INFLUENCED BY DIFFERENT ANTHROPOGENIC ACTIVITIES SUCH AS LESS-IMPACTED AREAS (L), AGRICULTURAL (A), INDUSTRIAL (I), AND RESIDENTIAL (R) ACTIVITIES DURING THE WET AND DRY SEASONS. <LOD: LESS THAN THE LIMIT OF DETECTION.	6
TABLE S4. THE RELATIVE ABUNDANCE OF THE ARGS, AND INTEGRONS IN THE 12 SAMPLING SITES OF THE SAIGON RIVER INFLUENCED BY DIFFERENT ANTHROPOGENIC ACTIVITIES SUCH AS LESS-IMPACTED AREAS (L), AGRICULTURAL (A), INDUSTRIAL (I), AND RESIDENTIAL (R) ACTIVITIES DURING THE WET AND DRY SEASONS. NA: NOT AVAILABLE	8
TABLE S5. THE CORRELATION COEFFICIENT VALUES OF PEARSON AND SPEARMAN ANALYSIS BETWEEN WATER QUALITY PARAMETERS AND THE ABSOLUTE ABUNDANCE (COPIES/ML) OF ALL ARGS, INTEGRONS, AND THE 16S RRNA GENE IN THE SURFACE WATER. THE CORRELATION WAS CONSIDERED SIGNIFICANT ONLY IF THE P-VALUE WAS LESS THAN 0.05, AND HIGHLIGHTED WITH BOLD FORMAT.	9
TABLE S6. THE P VALUE OF PEARSON AND SPEARMAN ANALYSIS BETWEEN WATER QUALITY PARAMETERS AND ARGS, INTEGRONS, AND THE 16S RRNA GENE IN THE SURFACE WATER...	10

SUPPLEMENTARY MATERIALS

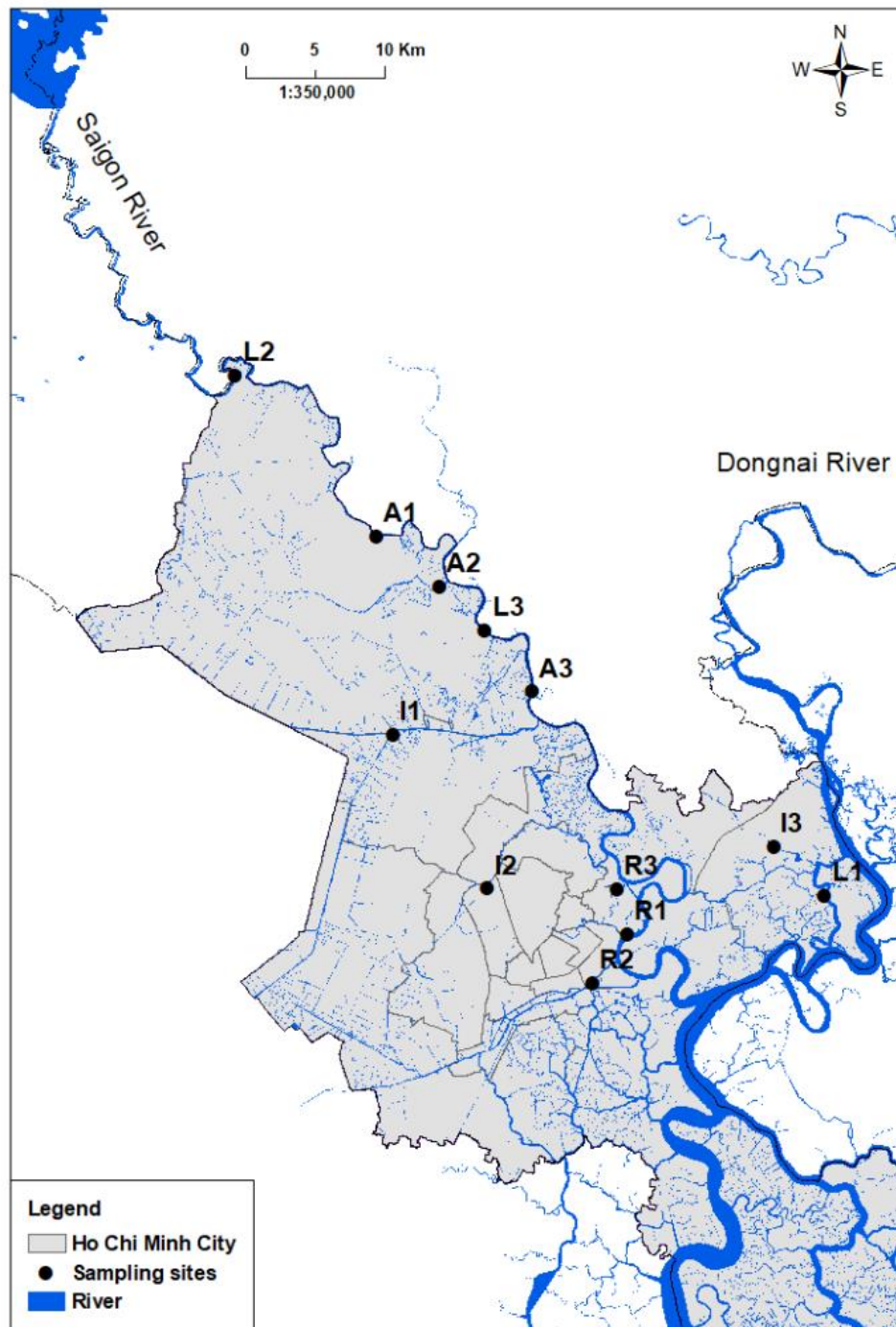


Figure S1. Map of all the sampling sites along the Saigon River in Ho Chi Minh City

Table S1. Detailed information of RT-PCR primers for quantification of the ARGs, integrons, and 16S rRNA gene

Target genes	Amplicon length (bp)	Description	The sequences of forward and reverse primers	Annealing temp. (°C)	References
<i>aac(6)-Ib-cr</i>	179	Aminoglycoside resistance gene	F: GGAGAGCCGATTGGGTATG R: CATGTACACGGCTGGACCA	60	(Guillard <i>et al.</i> , 2010)
<i>bla_{CTX-M}</i>	232	B-lactam resistance gene	F: GGAATCTGACGCTGGGTAAA R: GGTTGAGGCTGGGTGAAGTA	56	(Ellem, Partridge and Iredell, 2011)
<i>bla_{SHV}</i>	105	B-lactam resistance gene	F: TCCCATGATGAGCACCTTTAAA R: TCCTGCTGGCGATAGTGGAT	55	(Roschanski <i>et al.</i> , 2014)
<i>bla_{TEM}</i>	100	B-lactam resistance gene	F: GCATCTTACGGATGGCATGA R: GTCCTCCGATCGTTGTCAGAA	61	(Roschanski <i>et al.</i> , 2014)
<i>qnrA</i>	159	Quinolone resistance gene	F: ATTTCTCACGCCAGGATTG R: GCAGATCGGCATAGCTGAAG	56	(Marti and Balcázar, 2013)
<i>qnrB</i>	193	Quinolone resistance gene	F: TATGGCTCTGGCACTCGTT R: GCATCTTTCAGCATCGCAC	56	(Hamed <i>et al.</i> , 2018)
<i>oqxB</i>	131	Efflux pump	F: TCCTGATCTCCATTAACGCCCA R: ACCGGAACCCATCTCGATGC	56	(Hong <i>et al.</i> , 2009)
<i>sul1</i>	163	Sulfonamide resistance gene	F: CGCACCAGAAACATCGCTGCAC R: TGAAGTTCGCGCGCAAGGCTCG	60	(Pei <i>et al.</i> , 2006)
<i>sul2</i>	191	Sulfonamide resistance gene	F: TCCGGTGGAGGCCGGTATCTGG R: CGGGAATGCCATCTGCCTTGAG	60	(Pei <i>et al.</i> , 2006)
<i>dfrA</i>	273	Trimethoprim resistance gene	F: ACGGATCCTGGCTGTTGGTTGGACGC R: CGGAATTCACCTTCCGGCTCGATGTC	60	(Lee <i>et al.</i> , 2001)
<i>intI1</i>	196	Integrase class 1	F: GCCTTGATGTTACCCGAGAG R: GATCGGTCTGAATGCGTGT	58	(Barraud <i>et al.</i> , 2010)
<i>intI2</i>	195	Integrase class 2	F: TGCTTTTCCCACCTTACC R: GACGGCTACCCTCTGTTATCTC	58	(Barraud <i>et al.</i> , 2010)
<i>intI3</i> -	138	Integrase class 3	F: GCCACCACTTGTGAGGA R: GGATGTCTGTGCCTGCTTG	58	(Barraud <i>et al.</i> , 2010)
<i>16S rRNA</i>	136	Total bacterial 16S rRNA gene	F: AAACCTCAAAGAATTGACGG R: CTCACRRCACGAGCTGAC	59	(Bacchetti De Gregoris <i>et al.</i> , 2011)

Table S2. Limit of detection values for all target ARGs in qPCR

Antibiotic resistance genes	Limit of detection (gene copies)
<i>aac(6)-Ib-cr</i>	52
<i>blaCTX-M</i>	40
<i>blaTEM</i>	93
<i>blaSHV</i>	88
<i>qnrA</i>	58
<i>qnrB</i>	48
<i>sul1</i>	49
<i>sul2</i>	57
<i>dfrA</i>	39
<i>oqxB</i>	71
<i>intI1</i>	47
<i>intI2</i>	47
<i>intI3</i>	67
<i>16S rRNA</i>	68

Table S3. The concentration (copies/mL) of the ARGs, Integrons, and 16S rRNA gene in the 12 sampling sites of the Saigon River influenced by different anthropogenic activities such as less-impacted areas (L), agricultural (A), industrial (I), and residential (R) activities during the wet and dry seasons. <LOD: Less than the limit of detection.

Seasons	Sites	<i>aac(6)-Ib-cr</i>	<i>blaCTX</i>	<i>blaTEM</i>	<i>blaSHV</i>	<i>qnrA</i>	<i>qnrB</i>	<i>sulI</i>	<i>sul2</i>	<i>dfrA</i>	<i>intI1</i>	<i>intI2</i>	<i>intI3</i>	<i>oqxB</i>	<i>16S rRNA</i>
Wet season	L1	1.9E+02	3.7E+00	1.0E+03	9.1E+02	8.8E+01	9.5E+01	2.5E+04	3.5E+03	8.8E+02	2.5E+04	2.7E+01	6.5E+02	5.0E+02	5.1E+05
	L2	<LOD	6.3E+00	3.3E+02	4.0E+02	7.4E+01	6.3E+00	1.5E+03	8.5E+01	2.7E+02	1.6E+03	2.9E+00	3.4E+00	5.3E+01	7.3E+04
	L3	1.9E+02	2.5E+00	5.3E+02	1.4E+01	7.9E+01	8.2E+01	1.5E+04	1.8E+03	5.7E+02	2.0E+04	2.7E+01	3.4E+02	2.5E+02	6.2E+05
	A1	2.4E+02	7.2E-03	2.4E+02	1.7E+02	1.2E+02	6.7E+01	1.2E+04	1.4E+03	1.2E+03	1.2E+04	1.6E+01	5.2E+02	1.8E+02	7.0E+05
	A2	4.6E+02	6.3E-01	4.6E+02	1.8E+04	2.7E+02	1.2E+02	3.7E+04	7.3E+03	2.1E+03	3.6E+04	7.8E+01	2.5E+03	2.2E+02	2.2E+06
	A3	1.5E+02	2.9E-02	2.2E+02	8.0E+02	1.2E+02	2.7E+02	1.9E+04	2.4E+03	4.0E+02	1.8E+04	3.5E+01	3.1E+02	2.3E+02	9.4E+05
	I1	2.4E+02	4.3E+00	6.9E+02	2.2E+02	2.8E+02	1.3E+02	2.0E+04	1.9E+03	3.7E+02	2.2E+04	4.2E+01	4.2E+02	3.3E+02	8.1E+05
	I2	5.8E+05	1.3E+03	6.8E+05	9.6E+03	7.9E+04	3.4E+03	6.4E+06	4.6E+05	2.9E+05	1.1E+07	1.0E+04	6.5E+04	2.2E+05	1.1E+08
	I3	1.6E+06	9.5E+02	6.7E+05	1.5E+04	1.6E+05	2.2E+03	8.9E+06	8.7E+05	4.9E+05	1.9E+07	3.5E+04	1.9E+05	2.9E+05	1.6E+08
	R1	6.0E+03	5.4E+01	5.0E+03	6.3E+01	5.7E+02	2.0E+03	1.3E+05	2.1E+04	3.8E+03	1.8E+05	1.0E+02	1.6E+03	3.0E+03	1.2E+07
	R2	1.2E+05	3.6E+02	1.5E+05	6.0E+02	1.2E+04	2.5E+03	1.7E+06	4.3E+05	5.2E+04	3.1E+06	2.7E+02	3.8E+04	5.8E+04	7.8E+07
	R3	5.5E+05	8.9E+02	3.3E+05	2.0E+02	3.3E+04	2.2E+04	4.8E+06	3.6E+05	1.9E+05	7.3E+06	5.5E+03	6.7E+04	1.1E+05	9.4E+07
Dry season	L1	5.7E+02	4.3E+01	8.8E+02	6.7E+03	3.9E+02	3.7E+01	3.4E+02	5.7E+03	1.3E+03	1.6E+03	2.3E+02	4.4E+02	8.0E+02	3.3E+04
	L2	8.1E+01	8.0E+00	8.3E+02	1.8E+03	6.2E+02	1.4E+02	1.1E+03	4.4E+03	2.7E+03	1.5E+03	2.4E+02	2.2E+02	9.8E+02	2.6E+06
	L3	2.8E+03	4.7E+00	1.6E+03	4.9E+02	1.7E+02	1.0E+01	2.7E+04	3.1E+05	2.0E+04	3.2E+04	2.2E+01	3.8E+02	1.7E+03	1.6E+04
	A1	1.5E+03	5.8E+01	5.1E+04	6.3E+03	9.9E+02	1.3E+02	1.8E+04	6.8E+04	1.3E+04	1.5E+04	2.6E+02	3.4E+02	7.5E+02	3.7E+05
	A2	1.4E+04	2.6E+02	1.6E+03	1.7E+04	2.5E+03	1.1E+02	1.0E+05	3.7E+05	9.1E+04	7.3E+04	2.3E+02	1.5E+03	1.6E+03	1.3E+06
	A3	1.1E+04	6.4E+02	9.6E+03	6.2E+03	2.4E+03	3.5E+03	1.8E+05	7.0E+05	6.2E+04	2.2E+05	2.7E+02	4.8E+03	4.5E+03	2.4E+06
	I1	2.1E+04	7.1E+02	2.7E+03	9.1E+03	8.8E+03	8.6E+02	4.1E+05	1.3E+06	1.1E+05	2.6E+05	3.0E+02	2.0E+03	5.5E+03	5.3E+06
	I2	1.5E+07	2.0E+04	1.2E+07	1.4E+05	2.6E+05	5.3E+03	4.4E+07	3.8E+07	5.6E+06	2.7E+07	1.8E+04	1.6E+05	4.6E+05	1.6E+08
	I3	5.7E+06	8.7E+03	2.9E+06	8.1E+04	1.2E+05	2.7E+03	1.5E+07	1.0E+07	2.1E+06	1.3E+07	7.9E+03	5.1E+04	2.1E+05	6.2E+07
	R1	7.7E+03	1.4E+02	8.2E+03	1.5E+03	1.9E+02	7.1E+03	4.6E+04	2.2E+05	7.9E+03	6.1E+04	5.1E+01	9.8E+02	1.6E+03	1.1E+06
	R2	9.5E+05	2.9E+03	1.0E+06	6.3E+04	1.2E+04	4.2E+03	3.9E+06	9.7E+06	5.5E+05	3.3E+06	2.0E+03	4.7E+04	8.0E+04	1.8E+07
	R3	2.7E+06	7.8E+03	1.9E+06	2.9E+05	4.4E+04	2.8E+04	2.1E+07	2.2E+07	1.1E+06	1.1E+07	6.5E+03	1.3E+05	2.3E+05	5.8E+07
Average	L	6.2E+02	1.1E+01	8.6E+02	1.7E+03	2.4E+02	6.2E+01	1.2E+04	5.5E+04	4.4E+03	1.4E+04	9.2E+01	3.4E+02	7.1E+02	6.3E+05
	A	4.6E+03	1.6E+02	1.1E+04	8.0E+03	1.1E+03	7.0E+02	6.2E+04	1.9E+05	2.8E+04	6.1E+04	1.5E+02	1.7E+03	1.3E+03	1.3E+06

	I	3.8E+06	5.2E+03	2.7E+06	4.2E+04	1.1E+05	2.4E+03	1.2E+07	8.5E+06	1.4E+06	1.2E+07	1.2E+04	7.9E+04	2.0E+05	8.4E+07
	R	7.3E+05	2.0E+03	5.7E+05	6.0E+04	1.7E+04	1.1E+04	5.2E+06	5.5E+06	3.1E+05	4.2E+06	2.4E+03	4.7E+04	8.1E+04	4.3E+07

Table S4. The relative abundance of the ARGs, and Integrons in the 12 sampling sites of the Saigon River influenced by different anthropogenic activities such as less-impacted areas (L), agricultural (A), industrial (I), and residential (R) activities during the wet and dry seasons. NA: not available

Seasons	Sites	<i>aac(6)-Ib-cr</i>	<i>blaCTX</i>	<i>blaTEM</i>	<i>blaSHV</i>	<i>qnrA</i>	<i>qnrB</i>	<i>sul1</i>	<i>sul2</i>	<i>dfrA</i>	<i>intI1</i>	<i>intI2</i>	<i>intI3</i>	<i>oqxB</i>
Wet season	L1	3.6E-04	7.2E-06	2.0E-03	1.8E-03	1.7E-04	1.8E-04	4.9E-02	6.8E-03	1.7E-03	4.9E-02	5.2E-05	1.3E-03	9.7E-04
	L2	NA	8.6E-05	4.6E-03	5.4E-03	1.0E-03	8.6E-05	2.0E-02	1.2E-03	3.7E-03	2.2E-02	3.9E-05	4.7E-05	7.3E-04
	L3	3.1E-04	4.1E-06	8.5E-04	2.3E-07	1.3E-04	1.3E-04	2.5E-02	2.9E-03	9.2E-04	3.2E-02	4.3E-05	5.5E-04	4.0E-04
	I1	3.0E-04	5.3E-06	8.5E-04	2.8E-04	3.4E-04	1.6E-04	2.5E-02	2.3E-03	4.6E-04	2.8E-02	5.2E-05	5.3E-04	4.1E-04
	I2	5.2E-03	1.2E-05	6.1E-03	8.6E-05	7.1E-04	3.0E-05	5.8E-02	4.2E-03	2.6E-03	9.7E-02	9.4E-05	5.8E-04	2.0E-03
	I3	9.6E-03	5.8E-06	4.1E-03	9.1E-05	9.9E-04	1.3E-05	5.4E-02	5.3E-03	3.0E-03	1.2E-01	2.1E-04	1.2E-03	1.8E-03
	A1	3.4E-04	1.0E-08	3.5E-04	2.5E-04	1.7E-04	9.7E-05	1.7E-02	2.0E-03	1.8E-03	1.7E-02	2.3E-05	7.5E-04	2.6E-04
	A2	2.1E-04	2.9E-07	2.1E-04	8.2E-03	1.3E-04	5.8E-05	1.7E-02	3.4E-03	9.8E-04	1.7E-02	3.6E-05	1.2E-03	1.0E-04
	A3	1.6E-04	3.0E-08	2.3E-04	8.5E-04	1.3E-04	2.9E-04	2.0E-02	2.6E-03	4.2E-04	1.9E-02	3.7E-05	3.3E-04	2.5E-04
	R1	4.9E-04	4.4E-06	4.1E-04	5.2E-06	4.7E-05	1.7E-04	1.1E-02	1.7E-03	3.1E-04	1.5E-02	8.6E-06	1.3E-04	2.4E-04
	R2	1.6E-03	4.6E-06	1.9E-03	7.7E-06	1.6E-04	3.2E-05	2.1E-02	5.5E-03	6.6E-04	3.9E-02	3.4E-06	4.9E-04	7.3E-04
	R3	5.9E-03	9.5E-06	3.5E-03	2.2E-06	3.5E-04	2.3E-04	5.1E-02	3.8E-03	2.1E-03	7.8E-02	5.9E-05	7.2E-04	1.2E-03
Dry season	L1	1.7E-02	1.3E-03	2.7E-02	2.0E-01	1.2E-02	1.1E-03	1.0E-02	1.7E-01	4.0E-02	4.8E-02	7.1E-03	1.3E-02	2.4E-02
	L2	3.2E-05	3.2E-06	3.2E-04	6.9E-04	2.4E-04	5.6E-05	4.4E-04	1.7E-03	1.1E-03	5.7E-04	9.5E-05	8.7E-05	3.8E-04
	L3	1.7E-01	2.9E-04	9.7E-02	3.0E-02	1.0E-02	6.1E-04	1.7E+00	1.9E+01	1.2E+00	1.9E+00	1.4E-03	2.3E-02	1.0E-01
	I1	4.0E-03	1.3E-04	5.0E-04	1.7E-03	1.7E-03	1.6E-04	7.7E-02	2.4E-01	2.1E-02	4.9E-02	5.6E-05	3.8E-04	1.0E-03
	I2	9.4E-02	1.2E-04	7.3E-02	8.7E-04	1.6E-03	3.3E-05	2.7E-01	2.4E-01	3.5E-02	1.7E-01	1.1E-04	1.0E-03	2.8E-03
	I3	9.1E-02	1.4E-04	4.7E-02	1.3E-03	2.0E-03	4.4E-05	2.4E-01	1.7E-01	3.3E-02	2.1E-01	1.3E-04	8.2E-04	3.4E-03
	A1	4.1E-03	1.6E-04	1.4E-01	1.7E-02	2.7E-03	3.7E-04	5.0E-02	1.9E-01	3.5E-02	4.1E-02	7.1E-04	9.3E-04	2.1E-03
	A2	1.0E-02	1.9E-04	1.2E-03	1.2E-02	1.8E-03	7.9E-05	7.8E-02	2.8E-01	6.8E-02	5.5E-02	1.7E-04	1.1E-03	1.2E-03
	A3	4.7E-03	2.6E-04	3.9E-03	2.6E-03	9.7E-04	1.4E-03	7.5E-02	2.9E-01	2.5E-02	8.9E-02	1.1E-04	2.0E-03	1.8E-03
	R1	6.7E-03	1.2E-04	7.1E-03	1.3E-03	1.6E-04	6.2E-03	4.0E-02	1.9E-01	6.9E-03	5.3E-02	4.5E-05	8.5E-04	1.4E-03
	R2	5.2E-02	1.6E-04	5.7E-02	3.5E-03	6.9E-04	2.3E-04	2.2E-01	5.3E-01	3.0E-02	1.8E-01	1.1E-04	2.6E-03	4.4E-03
	R3	4.7E-02	1.4E-04	3.3E-02	5.1E-03	7.7E-04	4.8E-04	3.6E-01	3.8E-01	1.8E-02	1.9E-01	1.1E-04	2.2E-03	4.1E-03

Table S5. The correlation coefficient values of Pearson and Spearman analysis between water quality parameters and the absolute abundance (copies/ml) of all ARGs, integrons, and the 16S rRNA gene in the surface water. The correlation was considered significant only if the p-value was less than 0.05, and highlighted with bold format.

		pH	Temperature	Conductivity	Salinity	Turbidity	DO	P-PO43-	N-NH4+	N-NO2-	N-NO3-	BOD5
<i>aac(6)-Ib-cr</i>	Pearson	0.448	0.449	0.025	0.061	0.205	-0.433	0.153	0.013	-0.157	-0.074	0.582
	Spearman	0.645	0.524	0.777	0.787	-0.071	-0.409	0.526	0.461	-0.429	-0.328	0.783
<i>blaCTX</i>	Pearson	0.491	0.489	0.113	0.159	0.210	-0.441	0.208	0.057	-0.168	-0.092	0.581
	Spearman	0.648	0.502	0.754	0.766	-0.161	-0.295	0.456	0.451	-0.512	-0.473	0.752
<i>blaTEM</i>	Pearson	0.438	0.406	0.049	0.084	0.114	-0.394	0.241	0.000	-0.144	-0.071	0.499
	Spearman	0.640	0.447	0.732	0.745	-0.186	-0.339	0.512	0.434	-0.456	-0.353	0.754
<i>blaSHV</i>	Pearson	0.410	0.357	0.273	0.343	0.038	-0.401	0.235	0.104	-0.147	-0.099	0.295
	Spearman	0.465	0.330	0.457	0.479	-0.100	-0.291	0.389	0.235	-0.310	-0.363	0.301
<i>qnrA</i>	Pearson	0.476	0.428	-0.050	-0.024	0.206	-0.569	0.304	0.287	-0.218	-0.106	0.752
	Spearman	0.556	0.405	0.640	0.646	-0.099	-0.496	0.578	0.466	-0.527	-0.502	0.833
<i>qnrB</i>	Pearson	0.301	0.237	0.294	0.338	-0.204	-0.330	0.358	0.427	-0.122	-0.118	0.144
	Spearman	0.618	0.444	0.709	0.704	-0.254	-0.355	0.526	0.477	-0.220	-0.212	0.666
<i>sul1</i>	Pearson	0.497	0.443	0.072	0.120	0.117	-0.525	0.366	0.157	-0.194	-0.099	0.583
	Spearman	0.615	0.445	0.625	0.633	0.098	-0.520	0.567	0.505	-0.401	-0.168	0.774
<i>sul2</i>	Pearson	0.483	0.455	0.217	0.271	0.065	-0.382	0.218	0.009	-0.149	-0.089	0.414
	Spearman	0.572	0.502	0.736	0.750	-0.095	-0.279	0.397	0.365	-0.356	-0.399	0.663
<i>dfrA</i>	Pearson	0.456	0.456	0.049	0.085	0.201	-0.419	0.160	0.017	-0.158	-0.080	0.577
	Spearman	0.531	0.444	0.697	0.709	-0.164	-0.367	0.415	0.354	-0.382	-0.472	0.723
<i>intI1</i>	Pearson	0.517	0.442	0.014	0.048	0.136	-0.630	0.419	0.416	-0.240	-0.125	0.747
	Spearman	0.625	0.456	0.624	0.634	0.129	-0.511	0.543	0.527	-0.407	-0.133	0.786
<i>intI2</i>	Pearson	0.354	0.266	-0.051	-0.036	0.041	-0.534	0.280	0.407	-0.204	-0.101	0.604
	Spearman	0.568	0.384	0.648	0.654	-0.123	-0.473	0.528	0.505	-0.482	-0.501	0.797
<i>intI3</i>	Pearson	0.487	0.388	0.094	0.131	-0.026	-0.595	0.402	0.460	-0.233	-0.130	0.582
	Spearman	0.619	0.369	0.633	0.639	0.061	-0.512	0.587	0.521	-0.396	-0.196	0.721
<i>oqxB</i>	Pearson	0.541	0.440	0.057	0.095	0.104	-0.632	0.476	0.457	-0.243	-0.132	0.736
	Spearman	0.626	0.478	0.730	0.740	-0.155	-0.361	0.492	0.468	-0.513	-0.398	0.805
<i>16s rRNA</i>	Pearson	0.488	0.359	-0.054	-0.035	0.031	-0.625	0.471	0.627	-0.219	-0.141	0.699
	Spearman	0.598	0.368	0.485	0.486	0.029	-0.580	0.522	0.534	-0.331	-0.237	0.848

Table S6. The p value of Pearson and Spearman analysis between water quality parameters and ARGs, integrons, and the 16S rRNA gene in the surface water.

		pH	Temperature	Conductivity	Salinity	Turbidity	DO	P-PO43-	N-NH4+	N-NO2-	N-NO3-	BOD5
<i>aac(6)-Ib-cr</i>	Pearson	0.0282	0.0279	0.9081	0.7773	0.3371	0.0347	0.4959	0.9527	0.4844	0.7300	0.0028
	Spearman	0.0007	0.0086	0.0000	0.0000	0.7406	0.0472	0.0120	0.0267	0.0466	0.1179	0.0000
<i>blaCTX</i>	Pearson	0.0148	0.0152	0.6007	0.4583	0.3246	0.0309	0.3541	0.7969	0.4538	0.6676	0.0029
	Spearman	0.0006	0.0124	0.0000	0.0000	0.4527	0.1616	0.0331	0.0310	0.0148	0.0196	0.0000
<i>blaTEM</i>	Pearson	0.0324	0.0491	0.8193	0.6959	0.5956	0.0568	0.2800	0.9999	0.5214	0.7415	0.0131
	Spearman	0.0008	0.0285	0.0000	0.0000	0.3840	0.1056	0.0148	0.0386	0.0331	0.0906	0.0000
<i>blaSHV</i>	Pearson	0.0468	0.0871	0.1972	0.1012	0.8599	0.0520	0.2927	0.6366	0.5139	0.6468	0.1616
	Spearman	0.0220	0.1150	0.0249	0.0179	0.6420	0.1682	0.0735	0.2801	0.1603	0.0816	0.1533
<i>qnrA</i>	Pearson	0.0186	0.0370	0.8151	0.9118	0.3347	0.0037	0.1694	0.1847	0.3302	0.6219	0.0000
	Spearman	0.0048	0.0495	0.0008	0.0007	0.6449	0.0137	0.0049	0.0249	0.0118	0.0125	0.0000
<i>qnrB</i>	Pearson	0.1530	0.2645	0.1628	0.1061	0.3394	0.1154	0.1019	0.0423	0.5874	0.5829	0.5016
	Spearman	0.0013	0.0299	0.0001	0.0001	0.2312	0.0886	0.0120	0.0213	0.3260	0.3196	0.0004
<i>sulI</i>	Pearson	0.0135	0.0302	0.7396	0.5765	0.5868	0.0085	0.0936	0.4733	0.3876	0.6452	0.0028
	Spearman	0.0014	0.0292	0.0011	0.0009	0.6478	0.0091	0.0059	0.0140	0.0640	0.4331	0.0000
<i>sul2</i>	Pearson	0.0169	0.0254	0.3084	0.2003	0.7621	0.0654	0.3295	0.9664	0.5092	0.6802	0.0441
	Spearman	0.0035	0.0125	0.0000	0.0000	0.6595	0.1876	0.0674	0.0872	0.1036	0.0533	0.0004
<i>dfrA</i>	Pearson	0.0250	0.0253	0.8210	0.6927	0.3458	0.0415	0.4760	0.9388	0.4821	0.7118	0.0032
	Spearman	0.0075	0.0296	0.0002	0.0001	0.4429	0.0775	0.0548	0.0977	0.0791	0.0198	0.0001
<i>intI1</i>	Pearson	0.0097	0.0306	0.9485	0.8236	0.5249	0.0010	0.0522	0.0481	0.2822	0.5603	0.0000
	Spearman	0.0011	0.0252	0.0011	0.0009	0.5490	0.0107	0.0091	0.0098	0.0600	0.5354	0.0000
<i>intI2</i>	Pearson	0.0897	0.2086	0.8115	0.8666	0.8482	0.0072	0.2062	0.0538	0.3625	0.6393	0.0018
	Spearman	0.0038	0.0640	0.0006	0.0005	0.5654	0.0195	0.0116	0.0140	0.0232	0.0127	0.0000
<i>intI3</i>	Pearson	0.0158	0.0611	0.6627	0.5433	0.9042	0.0022	0.0634	0.0272	0.2957	0.5435	0.0029
	Spearman	0.0013	0.0763	0.0009	0.0008	0.7775	0.0106	0.0041	0.0108	0.0682	0.3596	0.0001
<i>oqxB</i>	Pearson	0.0064	0.0316	0.7911	0.6580	0.6288	0.0009	0.0250	0.0282	0.2752	0.5377	0.0000
	Spearman	0.0011	0.0180	0.0001	0.0000	0.4702	0.0829	0.0201	0.0242	0.0146	0.0539	0.0000
<i>16s rRNA</i>	Pearson	0.0156	0.0851	0.8028	0.8715	0.8860	0.0011	0.0271	0.0014	0.3275	0.5101	0.0001
	Spearman	0.0021	0.0771	0.0162	0.0160	0.8941	0.0030	0.0127	0.0087	0.1318	0.2640	0.0000