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Supplementary material

Co-occurrence of microbial source tracking markers and antibiotic resistance genes in the Chishui River, China

Qianxing Zheng[a, b, 1], Renren Wu[a, 1], Xinnuo Chen[c], Pengxia Liu[d], Kaiming Li[a], Changdong Ke[a], Yongjie Wu[a], Yang Zhang[a*], Shijie Xiao[e], Jianhong Huang[b*]

[a] The Key Laboratory of Water Environment Simulation and Pollution Control, Ministry of Ecology and Environment, South China Institute of Environmental Sciences, Ministry of Ecology and Environment of the People's Republic of China, Guangzhou 510535, China;

[b] Faculty of Environment Science and Engineering, Kunming University of Science and Technology, Kunming 650550, China;

[c] School of Environment, South China Normal University, Guangzhou 510006, China;

[d] Ecological Environment Monitoring and Scientific Research Center, Taihu Basin & East China Sea Ecological Environment Supervision and Administration Bureau, Ministry of Ecology and Environment, Shanghai 200125, China;

[e] Guangdong Basic Research Center of Excellence for Ecological Security and Green Development, Key Laboratory for City Cluster Environmental Safety and Green Development of the Ministry of Education, School of Ecology, Environment and Resources, Guangdong University of Technology, Guangzhou, 510006, China.

Corresponding Author: Yang Zhang; Jianhong Huang

Address: Ruihe Road 18, Huangpu District, Guangzhou 510535, P. R. China; Jing Mingnan Road 727, Chenggong District, Kunming 650550, P. R. China.

Email: zhangyang@scies.org; huangjianhong78@163.com

[1] Qianxing Zheng and Renren Wu are co-first authors.

Table S1 Information on the geographical location of sampling sites

		Number	Sampling sites	Longitude (E)	Latitude (N)
Mainstream	Upstream	M1	Source	104.755395	27.429673
		M2	Banqiao section	104.918048	27.556668
		M3	Luodian Bridge	105.026781	27.681048
		M4	Xiaochi section	105.220302	27.753516
	Midstream	M5	Chahe Ferry	105.305992	27.707859
		M6	Caolian Creek	106.303628	27.806158
		M7	Tianjia Stockade	106.344285	27.942552
		M8	Erlangtan Ferry	106.169343	28.151613
		M9	Kowloon Tung	105.962077	28.344002
	Downstream	M10	Lianghekou section	105.768967	28.483427
		M11	Liyu Creek	105.737255	28.611676
		M12	Terminal point	105.845271	28.798352
Tributary	Upstream	T1	Banqiao section	104.821001	27.505572
		T2	Midstream of Zaxi River	105.09746	27.774846
		T3	Downstream of Zaxi River	105.175226	27.756589
	Midstream	T4	Yanjin River	106.34701	27.816378
		T5	Tongzi River	106.210075	28.136157
		T6	Gulin River	106.035324	28.134109
		T7	Banqiao Bridge tributary	105.963379	28.34387
	Downstream	T8	Datong River	105.683069	28.523964
		T9	Helong Creek	105.716884	28.706052
		T10	Xishui River	105.838414	28.794754

Table S2 Physical and chemical indicators of sampling points during the dryseason

Sites	DO	TN	TP	NH_3-N	COD	Turbidity (NTU)	pH	Conductivity (mS/cm)	T (℃)
	(mg/L)								
M1	11.87±0.61	2.55±0.86	0.29±0.08	0.06±0.01	4.33±0.58	4.93±0.38	7,41±0.05	0.30±0.01	12.27±0.12
M2	12.83±0.45	0.86±0.27	0.13±0.02	0.05±0.01	16.33±4.16	5.48±0.34	8.35±0.03	0.18±0.01	18.17±0.06
M3	11.47±0.85	1.33±0.23	0.14±0.04	0.03±0.00	6.33±1.53	13.40±1.14	8.58±0.10	0.32±0.00	16.80±0.00
M4	12.43±0.67	3.45±0.47	0.12±0.02	0.04±0.02	9.50±0.71	61.03±0.71	8.24±0.10	0.33±0.00	16.80±0.00
M5	13.17±0.06	1.69±0.71	0.12±0.03	0.03±0.00	5.67±0.58	24.10±2.95	8.41±0.01	0.33±0.00	17.50±0.00
M6	15.07±1.24	0.37±0.13	0.04±0.02	0.03±0.00	21.67±10.07	6.47±1.55	8.56±0.05	0.39±0.00	18.43±0.06
M7	10.43±0.25	2.02±0.09	0.06±0.04	0.05±0.01	17.67±3.51	2.80±0.53	8.14±0.01	0.41±0.00	18.20±0.00
M8	9.77±1.72	1.66±0.19	0.05±0.01	0.08±0.00	11.67±0.58	1.63±1.07	7.90±0.12	0.44±0.00	17.93±0.15
M9	12.03±1.01	2.44±0.28	0.06±0.02	0.03±0.00	12.33±3.51	9.80±0.33	8.22±0.06	0.43±0.00	18.53±0.23
M10	1.73±0.50	1.46±0.30	0.19±0.10	0.05±0.01	9.00±0.00	12.03±1.51	8.08±0.11	0.39±0.00	18.03±0.06
M11	10.87±0.32	1.59±0.58	0.14±0.01	0.05±0.02	8.33±3.06	7.60±0.85	7.89±0.12	0.35±0.00	18.40±0.10
M12	11.13±0.38	1.29±0.22	0.12±0.01	0.05±0.00	6.00±1.73	14.07±1.55	7.85±0.09	0.30±0.00	18.00±0.00
T1	10.60±0.00	4.20±1.40	0.14±0.07	0.05±0.02	14.67±2.52	34.60±0.00	8.01±0.00	0.35±0.00	16.30±0.00
T2	11.43±0.40	6.85±0.63	0.29±0.02	1.34±0.10	18.67±2.52	9.65±4.82	8.32±0.01	0.39±0.00	17.40±0.00

Continued Table S2 Physical and chemical indicators of sampling points during the dryseason

Sites	DO	TN	TP	NH_3-N	COD	Turbidity	pH	Conductivity	T (℃)
	(mg/L)					(NTU)		(mS/cm)	
T3	12.77±0.29	3.16±0.34	0.27±0.02	1.07±0.02	12.00±3.00	36.80±4.59	8.34±0.02	0.36±0.00	16.77±0.06
T4	11.07±0.29	8.78±0.65	0.29±0.02	1.58±0.08	22.33±0.58	7.53±1.27	8.12±0.04	0.59±0.00	16.60±0.00
T5	13.37±0.55	2.37±0.55	0.02±0.01	0.02±0.02	35.00±36.43	14.87±1.68	8.23±0.17	0.40±0.00	16.40±0.00
T6	12.30±0.87	3.07±1.53	0.06±0.03	0.06±0.02	18.33±0.58	64.63±7.52	8.15±0.03	0.44±0.01	18.30±0.00
T7	12.23±0.42	4.39±1.20	0.14±0.01	0.03±0.00	6.00±0.00	15.90±0.98	8.23±0.01	0.37±0.01	14.40±0.00
T8	12.77±0.97	3.21±0.97	0.09±0.02	0.07±0.00	4.00±0.00	14.73±3.19	7.69±0.12	0.11±0.00	16.70±0.10
T9	9.80±0.53	1.45±0.46	0.10±0.03	0.07±0.02	12.00±5.29	29.33±7.10	7.84±0.19	0.32±0.00	18.50±0.10
T10	10.20±0.40	2.02±0.33	0.11±0.01	0.05±0.01	8.50±0.71	17.03±1.14	7.16±0.05	0.23±0.01	18.13±0.06

Table S3 Physical and chemical indicators of sampling points during the wetseason

Sites	DO	TN	TP	NH_3-N	COD	Turbidity (NTU)	pH	Conductivity (mS/cm)	T (℃)
	(mg/L)								
M1	10.06±0.10	2.55±0.91	0.22±0.09	0.05±0.01	5.67±0.33	6.56±1.34	7.90±0.05	0.33±0.00	17.47±0.06
M2	10.6±0.30	4.20±0.02	0.14±0.08	0.05±0.03	14.67±1.33	121.67±2.08	8.31±0.08	0.31±0.00	19.43±0.06
M3	10.87±0.83	1.33±0.11	0.14±0.01	0.03±0.00	6.33±0.33	18.57±0.38	8.55±0.01	0.34±0.00	23.10±0.00
M4	13.10±0.96	3.45±0.36	0.12±0.07	0.04±0.00	9.50±2.75	690.00±42.72	8.49±0.02	0.34±0.00	19.10±0.00
M5	12.90±0.36	0.37±0.08	0.04±0.01	0.03±0.00	21.67±7.83	45.37±2.61	8.67±0.15	0.36±0.00	18.00±0.00
M6	12.00±0.56	1.69±0.36	0.12±0.01	0.04±0.00	5.67±0.33	15.63±1.21	8.53±0.01	0.41±0.00	22.83±0.23
M7	10.00±0.36	2.02±0.53	0.06±0.01	0.05±0.01	17.67±3.83	79.23±3.19	8.38±0.10	0.39±0.00	21.63±0.06
M8	10.50±0.00	1.66±0.85	0.05±0.02	0.08±0.02	11.67±2.83	43.47±2.72	8.33±0.01	0.40±0.00	22.10±0.17
M9	11.40±0.36	2.44±0.20	0.06±0.02	0.03±0.00	12.33±3.16	59.13±2.55	8.41±0.03	0.41±0.00	23.40±0.00
M10	9.87±0.15	1.46±0.31	0.19±0.09	0.05±0.02	9.00±2.00	16.90±0.17	8.36±0.05	0.42±0.00	24.07±0.21
M11	10.30±0.00	1.59±0.36	0.14±0.00	0.05±0.01	8.33±1.66	41.30±2.62	8.38±0.02	0.39±0.00	24.50±0.00
M12	10.00±1.21	1.29±0.15	0.12±0.02	0.05±0.03	6.00±0.00	18.63±1.86	8.32±0.06	0.39±0.01	24.43±0.15
T1	10.06±0.30	4.39±0.11	0.14±0.10	0.03±0.00	6.00±3.00	121.67±2.08	8.31±0.08	0.31±0.00	19.43±0.06
T2	11.37±0.47	6.85±1.89	0.29±0.09	1.34±0.65	18.67±5.83	9.65±0.99	8.64±0.10	0.44±0.00	13.40±0.35

Continued Table S3 Physical and chemical indicators of sampling points during the wetseason

Sites	DO	TN	TP	NH_3-N	COD	Turbidity	pH	Conductivity	T (℃)
	(mg/L)					(NTU)		(mS/cm)	
T3	11.67±0.72	3.76±0.59	0.21±0.06	0.76±0.3	10.5±1.5	12.85±0.92	8.52±0.01	0.40±0.00	19.80±0.00
T4	10.13±0.06	5.38±3.39	0.26±0.03	0.82±0.75	14.16±8.16	8.05±0.90	8.49±0.07	0.49±0.00	22.93±0.06
T5	10.60±0.72	2.81±0.44	0.02±0.00	0.02±0.02	20.00±15.00	5.67±1.01	8.32±0.07	0.37±0.00	21.60±0.00
T6	9.43±0.25	3.27±0.21	0.04±0.01	0.06±0.01	12.16±6.16	27.00±2.54	8.40±0.01	0.55±0.00	27.00±0.00
T7	8.93±0.55	1.71±0.85	0.24±0.11	0.15±0.11	13.66±2.66	62.97±0.15	8.52±0.04	0.25±0.02	28.70±0.00
T8	10.97±0.21	2.33±0.87	0.09±0.00	0.12±0.05	8.00±4.00	34.33±2.30	8.27±0.09	0.12±0.00	24.20±0.00
T9	8.87±1.78	1.67±0.23	0.09±0.00	0.14±0.07	12.00±0.00	16.97±0.58	8.18±0.06	0.32±0.00	26.90±0.00
T10	9.43±1.03	2.09±0.07	0.12±0.01	0.11±0.06	7.25±1.25	59.83±1.27	8.32±0.01	0.35±0.01	25.70±0.17

Table S4 Primers sequence used in this study

Gene	Primer	Sequence(5'→3')	Reference
BacHum	BacHum-160f	TGAGTTCACATGTCCGCATGA	(Kapoor et al., 2015)
	BacHum-241r	CGTTACCCCGCCTACTATCTAATG	
CPQ_064	064F1	TGTATAGATGCTGCTGCAACTGTACTC	(Stachler et al., 2018)
	064R1	CGTTGTTTTCATCTTTATCTTGTCCAT	
Pig-1-Bac	Bac32-f	AACGCTAGCTACAGGCTTAAC	(Mieszkin et al., 2009)
	Bac108r	CGGGCTATTCCTGACTATGGG	
P.ND5	P.ND5-f	ACAGCTGCACTACAAGCAATGC	(He et al., 2016)
	P.ND5-r	GGATGTAGTCCGAATTGAGCTGATTAT	
Rum-2-Bac	BacB2-590f	ACAGCCCGCGATTGATACTGGTAA	(Mieszkin et al., 2010)
	Bac708Rm	CAATCGGAGTTCTTCGTGAT	
BacCow	CF128-f	CCAACYTTCCCGWTACTC	(Kildare et al., 2007)
	305r	GGACCGTGTCTCAGTTCCAGTG	
GFD	GFD-f	TCGGCTGAGCACTCTAGGG	(Green et al., 2012)
	GFD-r	GCGTCTCTTTGTACATCCCA	
AllBac	AllBac296f	GAGAGGAAGGTCCCCCAC	(Alice et al., 2006)
	AllBac412r	CGCTACTTGGCTGGTTCAG	
E. coli	EC23S857F	GGTAGAGCACTGTTTTGGCA	(Chern et al. 2011)
	EC23S857R	TGTCTCCCGTGATAACTTTCTC	
intI1	*intI1*-F	CCTCCCGCACGATGATC	(Zeng et al. 2019)
	intI1-R	TCCACGCATCGTCAGGC	
sul1	*sul1*-F	CACCGGAAACATCGCTGCA	(Luo et al. 2010)
	sul1-R	AAGTTCCGCCGCAAGGCT	
sul2	*sul2*-F	CTCCGATGGAGGCCGGTAT	(Luo et al. 2010)
	sul2-R	GGGAATGCCATCTGCCTTGA	
tetW	*tetW*-F	GAGAGCCTGCTATATGCCAGC	(Aminov et al. 2001)
	tetW-R	GGGCGTATCCACAATGTTAAC	
tetO	*tetO*-F	TACGGARAGTTTATTGTATACC	(Aminov et al. 2001)
	tetO-R	TGGCGTATCTATAATGTTGAC	
ermF	*ermF*-F	TCGTTTTACGGGTCAGCACTT	(Knapp et al., 2010)
	ermF-R	CAACCAAAGCTGTGTCGTTT	

Table S5. Comparison between log-transformed gene concentrations in water and sediment using paired *t*-test. For comparison between water and sediment, Log_{10}GC/g sediment was converted to $\log_{10}$ GC/100g.Null hypothesis (H_O) is $\mu_{sediment} = \mu_{water}$ and alternative hypothesis (H_A) is $\mu_{sediment} > \mu_{water.}$

Gene	μ(sediment-water)
BacHum	**0.93 ($p < 0.001$)**
CPQ-064	**1.66 ($p < 0.001$)**
Pig-1-Bac	**1.85 ($p < 0.001$)**
P.ND5	**4.24 ($p < 0.001$)**
Rum-2-Bac	**2.38 ($p < 0.001$)**
BacCow	**2.40 ($p < 0.001$)**
GFD	**2.54 ($p < 0.001$)**
E. coli	**1.64 ($p < 0.001$)**
*sul*1	**2.77 ($p < 0.001$)**
sul2	**3.08 ($p < 0.001$)**
tetO	**2.01 ($p < 0.001$)**
tetW	**2.16 ($p < 0.001$)**
ermF	**2.28 ($p < 0.001$)**
intI1	**2.93 ($p< 0.001$)**

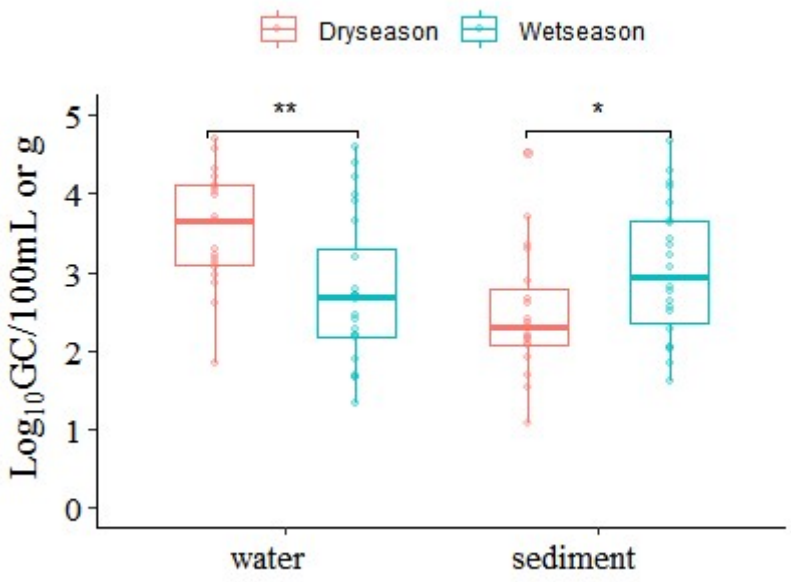

Fig. S1 The concentrations of *E. coli* in water and sediment. * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$.

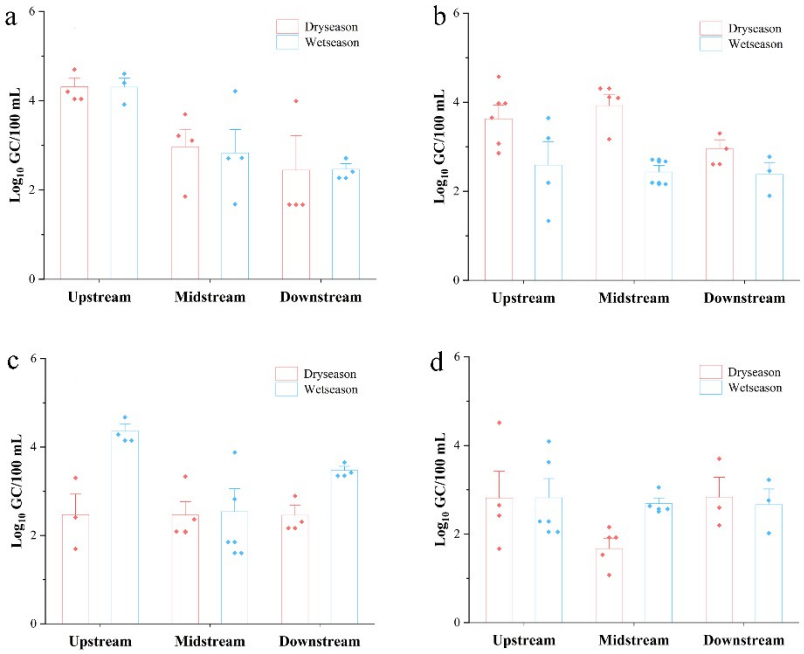

Fig. S2 The concentration of *E. coli* in the water and sediment of the up, middle, and downstream of the mainstream and tributaries.

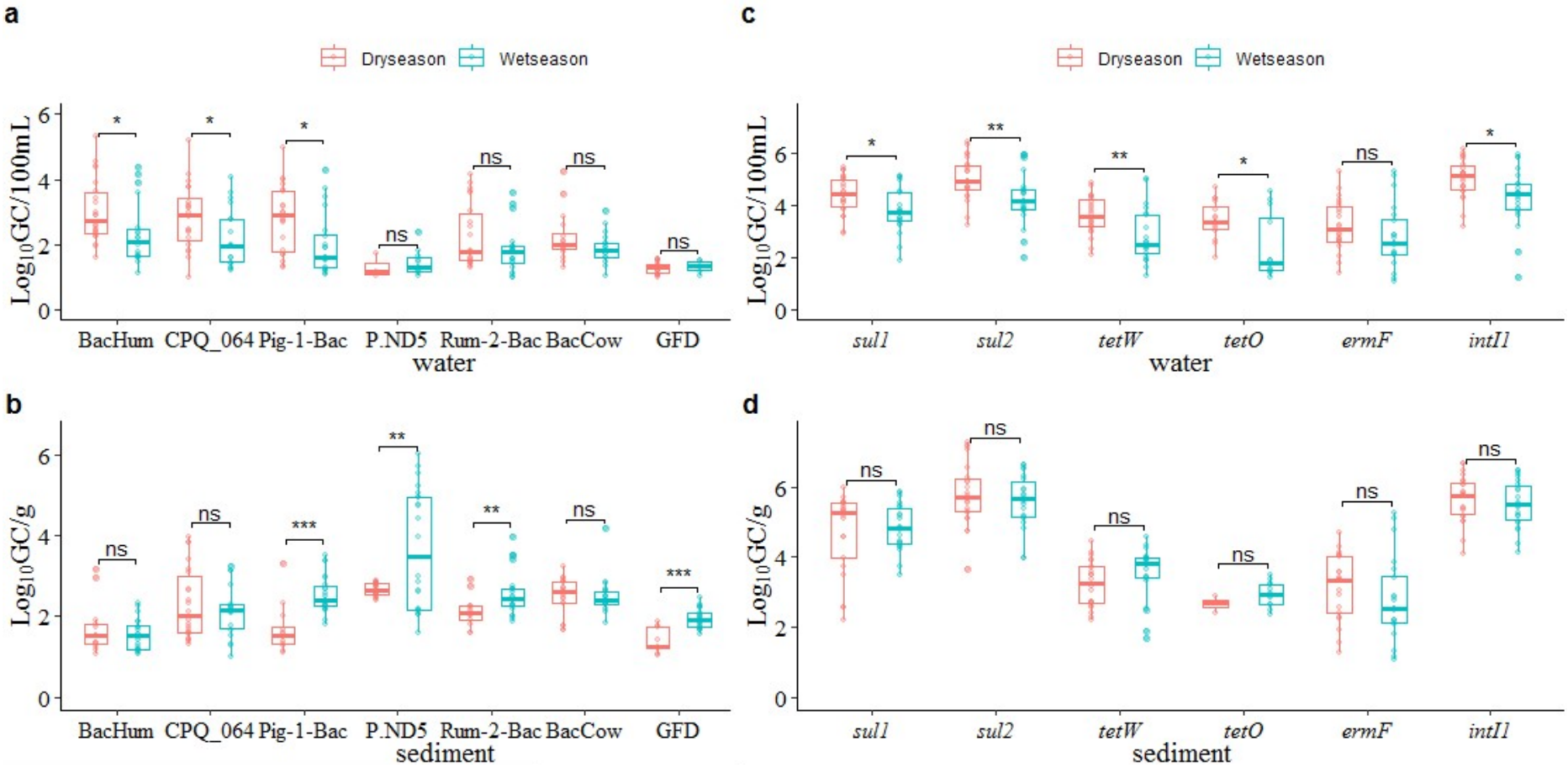

Fig. S3 The concentrations of MST markers, ARGs and MGE during the dryseason and wetseason. (a) MST markers in water samples; (b) MST markers in sediment samples; (c) ARGs in water samples; (d) ARGs in sediment samples. * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$.

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