SUPPLEMENTAL INFORMATION

For

Determination of Three Classes of Antibiotics in a Natural River Basin: Association with Antibiotic-resistant *Escherichia coli*

Qinqin ZHANG[†, ‡], Ai JIA[†], Yi Wan[†], Hong LIU[‡], Kunping WANG[§], Hui Peng[†], Zhaomin Dong[†], and Jianying Hu[†,*]

College of Urban and Environmental Sciences, Peking University, Beijing, 100871 China

[†]Laboratory for Earth Surface Processes, College of Urban and Environmental Sciences, Peking University, Beijing, 100871, China

[‡]Chongqing Institute of Green and Intelligent Technology, Chinese Academy of Sciences Chongqing, 401120, China

[§]School of Urban Construction and Environmental Engineering, Chongqing University, Chongqing, 400030, China

*To whom correspondence may be addressed (hujy@urban.pku.edu.cn).

Text 1-4

Tables 6

Figure 1

Chemicals and materials

SAs and Trimethoprim. Sulfaguanidine (SGD), sulfanilamide (SA), sulfathiazole (STZ), sulfisomidine (SIM), sulfamonomethoxine (SMM), sulfisoxazole (SIA), sulfachloropyridazine (SCP), sulfapyridine (SPD), sulfadiazine (SDZ), sulfamethazine (SMA), sulfamethoxazole (SMX), sulfamerazine (SMR), sulfaquinoxaline (SQX), sulfameter (SME), sulfamethizole (SMT), sulfamoxol (SMO), sulfadimethoxine (SDM), sulfamethoxypyridazine (SMP), sulfanitran (SNT), and trimethoprim (TMP) were all obtained from Sigma-Aldrich (MO, USA). $^{13}C_6$-sulfamethazine ($^{13}C_6$-SMA) was obtained from Cambridge Isotope Laboratories (MA, USA). *N*-acetylsulfapyridine (NAcSPD), *N*-acetylsulfadiazine (NAcSDZ), *N*-acetylsulfamethazine (NAcSMA), *N*-acetylsulfamethoxazole (NAcSMX), *N*-acetylsulfamerazine (NAcSMR), and *N*-acetylsulfamethoxazole-d_5 (NAcSMX-d_5) were purchased from Toronto Research Chemicals (North York, ON, Canada). TMP and SAs are usually used for animal growth promotion agents, together.

Quinolone. Cinoxacin (CINO), lomefloxacin (LOME), pipemidic acid (PIPE), ofloxacin (OFL), danofloxacin (DANO), enrofloxacin (ENRO), ciprofloxacin (CIP), sarafloxacin (SARA), difloxacin (DIF), sparfloxacin (SPAR), moxifloxacin (MOXI), and fleroxacin (FLER) were purchased from Sigma (St. Louis, MO, USA); norfloxacin (NOR), oxolinic acid (OXO), pefloxacin (PEFL), and flumequine (FLUM) were purchased from Dr. Ehrenstorfen (GmbH, Germany); nalidixic acid (NALI) was purchased from Acros Organics (Geel, Belgium); piromidic acid (PIRO) was purchased from Wako (Japan); gatifloxacin (GATI) was purchased from LKT Laboratories Inc. (Minnesota, USA), and norfloxacin-d_5 (NOR-d_5) was purchased from RdH Laborchemikalien GmbH (Germany).

TCs. Six target tetracyclines (purity, %), including tetracycline (TC, 95%), oxytetracycline (OTC, 95%), chlortetracycline (CTC, 80%), doxycycline (DXC, 98%), minocycline (MINO,

93%), and methacycline (MTC), as well as the internal standard demeclocycline (DMC, 98%), and ten products including 4-epitetracycline (ETC,97%), anhydrotetracycline (ATC, 97%), and 4-epianhydrotetracycline (EATC,97%), 4-epioxytetracycline (EOTC, 97%), α-apo-oxytetracycline (α-apo-OTC), β-apo-oxytetracycline (β-apo-OTC), isochlortetracycline (ICTC, 97%), 4-epichlortetracycline (ECTC,97%), anhydrochlortetracycline (ACTC), and 4-epianhydrochlortetracycline（EACTC） were analyzed in this study. These seven chemicals were obtained from Sigma-Aldrich (St. Louis, MO, USA), and the ten degradation products were purchased from Acros Organics (Geel, Belgium).

Isolation Procedure of *E. coli.* Water samples were 10×fold serial diluted and 0.1 mL of each dilution was filtered through nitrocellulose filters (0.45 μm pore-size, 47 mm diameter, 2 Millipore Corporation, America) with the goal of obtaining 30 to 50 colonies per filter. The filters placed onto E. coli chromogenic agar (Chromagar Microbiology, France) and incubated at 44°C for 24 h. After 24 h of incubation, colonies that turned blue on *E. coli* chromogenic agar were chosen and streaked onto LB agar (BD, America), and then incubated at 37°C for 24 6h. Approximately 40 isolates were collected with dilution method using 6-8 disks for each water sample, and their antibiotic susceptibilities were tested. To avoid the clones, all isolates were randomly chosen from independent colonies growing on the disks and the number of 9 selected isolates from each disk was less than 10. The pure cultures were then used to inoculate 1% tryptone water (Oxoid, UK) and EC broth containing 4-methylumbelliferyl-D-glucuronide (Oxoid, UK) and incubated for 24 h at 37 and 44°C, respectively. Isolates that produced indole from tryptophan and that were positive for gas production and fluorescence in EC broth containing 4-methylumbelliferyl-D-glucuronide

were designated as E. coli isolates and used for subsequent studies.

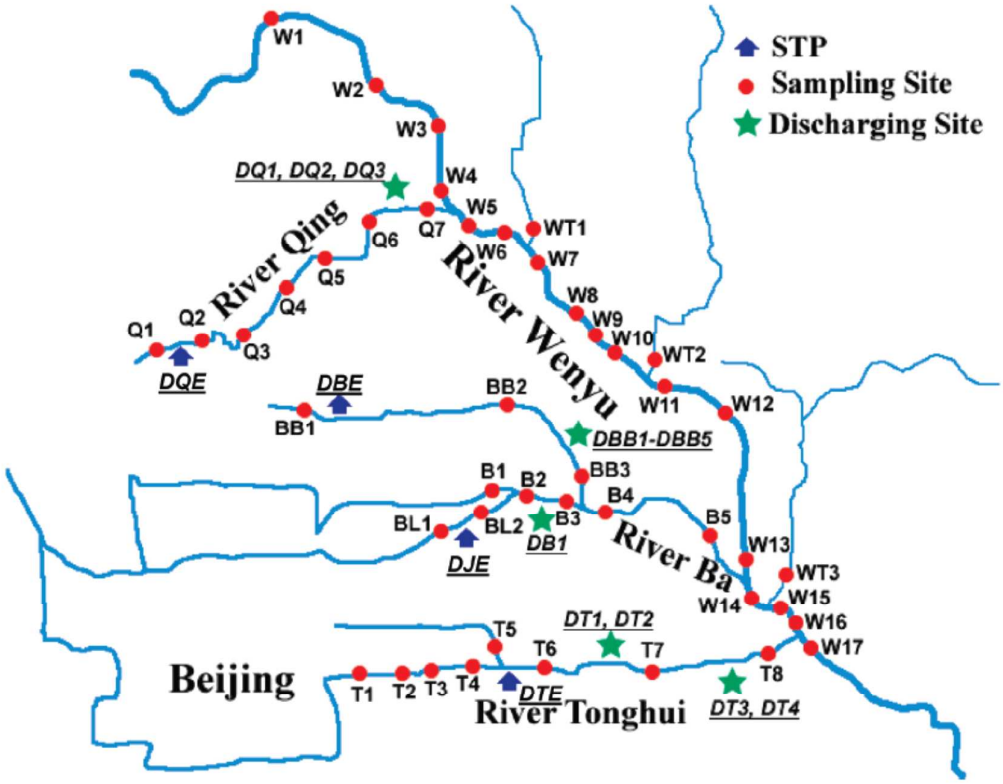

Figure S1. The Sampling Locations in Wenyu Basin of Beijing

Table S1. The Abbreviations of Discharging Sites and STP Effluents.

Sampling site	Abbreviations
STP effluent (Qinghe) in River Qing	DQE
Discharging sites in River Qing	DQ1
	DQ2
	DQ3
STP effluent (Beixiaohe) located River Ba	DBE
STP effluent (Jiuxianqiao) in River Ba	DJE
Discharging sites in River Ba	DB1
	DBB1
	DBB2
	DBB3
	DBB4
	DBB5
STP effluent (Gaobeidian) in River Tonghui	DTE
Discharging sites in River Tonghui	DT1
	DT2
	DT3
	DT4

Table S2. Liquid Chromatography Conditions for Three Classes Antibiotics.

25 sulfonamides and their *N*-acetyl-metabolites			
Time(min）	%A(Methanol）	%B (0.1% formic acid, v/v)	Gradient Curve
0	10	90	1
4.5	48	52	6
5	70	30	6
6	100	0	6
7	100	0	1
10	10	90	1
20 FQs			
0	10	90	1
0.5	20	80	6
4	35	65	6
5	45	55	6
6.5	100	0	6
8	100	0	1
10.5	10	90	1
17 TCs and their degradation products			
0	10	90	1
0.5	20	80	6
3.5	30	70	6
4.5	75	35	6
6	85	15	6
7.5	100	0	1
10	10	90	1

Table S3. Multi-selected Reaction Monitoring (MRM) Conditions of the Target Analytes.

Substance	SRM transition	Cone voltage (V)	Collision energy (eV)	Substance	SRM transition	Cone voltage (V)	Collision energy (eV)
SAs							
SGD	173 > 92	13	16	SMX	254 > 92	31	31
	173 > 156		10		254 > 156		16
SA	215 > 92	25	22	SIA	268 > 92	25	31
	215 > 156		13		268 > 156		16
SPD	250 > 92	31	25	SMP	281 > 92	31	28
	250 > 156		16		281 > 156		16
SDZ	251 > 92	28	25	SMM	281 > 92	31	28
	251 > 156		16		281 > 156		16
STZ	256 > 92	25	25	SCP	285 > 92	25	31
	256 > 156		16		285 > 156		13
SMR	265 > 92	31	28	NAcSMR	307 > 134	36	28
	265 > 110		25		307 > 172		20
SIM	279 > 92	34	31	NAcSMA	321 > 134	40	25
	279 > 124		19		321 > 186		20
SMO	268 > 92	25	28	NAcSMX	296 > 134	35	25
	268 > 156		16		296 > 198		20
SMT	271 > 92	22	28	NAcSDZ	293 > 134	32	25
	271 > 156		13		293 > 198		20
SMA	279 > 92	37	31	SQX	301 > 92	34	28
	279 > 186		16		301 > 156		16
SME	281 > 92	31	28	SDM	311 > 92	31	31
	281 > 156		16		311 > 156		22
$^{13}C_6$-SMA	285 > 98	28	31	SNT	336 > 64	25	43
	285 > 186		16		336 >		13

TMP	291 > 110 291 > 123	40	34 22	NAcSMX-d_5	156 301 > 139 301 > 203	35	25 18
NAcSPD	292 > 134 292 > 198	40	25 20				
TCs							
MINO	458>352	31	30	ETC	445>410	28	19
	458>441		19		445>427		15
EOTC	461>426	22	19	OTC	461>426	22	19
	461>444		16		461>444		16
α-apo-OTC	443>408	31	25	TC	445>154	28	26
	443>426		16		445>410		20
DMC	465>430	34	25	ICTC	479>462	34	15
	465>448		19		479>197		40
ECTC	479>444	34	22	EATC	427>154	31	34
	479>462		15		427>410		16
ATC	427>154	31	34	MTC	443>201	28	31
	427>410		16		443>426		16
β-apo-OTC	443>408	31	25	DXC	445>154	28	34
	443>426		16		445>428		16
EACTC	461>154	28	28	ACTC	461>154	28	28
	461>444		16		461>444		16
CTC	479>444	34	22				
	479>462		15				
FQs							
PIPE	304>217	31	22	LOME	352>265	34	22
	304>286		16		352>308		16
FLER	370>269	37	25	DIF	400>299	45	28
	370>326		19		400>356		20
OFL	362>261	34	25	SARA	386>299	45	28
	362>318		19		386>342		18
PEFL	334>290	37	19	GATI	376>289	37	28
	334>316		19		376>332		19
NOR	320>276	31	16	SPAR	393>292	40	24
	320>302		19		393>349		20
NOR-d_5	325>281	31	19	MOXI	402>358	34	19
	325>307		22		402>384		22
CIP	332>231	37	36	CINO	263>189	30	28
	332>288		18		263>245		15
DANO	358>283	40	25	OXO	262>216	32	25
	358>340		25		262>244		18

ENRO	360>316	42	20	NALI	233>187	30	25
	360>342		22		233>215		14
ENO	321>232	35	30	FLUM	262>202	28	34
	321>303		35		262>244		22
				PIRO	289>243	32	30
					289>271		18

1

Table S4. Recoveries (%), Instrument Detection Limits (IDLs, μg/L), Limits of Quantification (LOQs, ng/L) and Matrix Effects for Target Antibiotics in Various Water Matrices.

compounds	IDLs (μg/L)	Recovery ± RSD (%)			LOQs (ng/L)			Matrix effects （%）
		discharging sites	STP effluents	river water	discharging sites	STP effluents	river	
TCs								
MINO	0.5	95±15.7	81±5.2	91±3.9	19.1	30.6	32.6	32.7
ETC	0.03	101±19.6	98±6.7	87±3.4	4.7	1.6	1.4	8
EOTC	0.05	94±6.4	103±7.7	90±5.3	6.8	6.5	6.0	11.7
OTC	0.05	90±8.1	93±6.7	88±2.7	3.0	3.3	2.3	5.3
α-apo-OTC	0.1	120±10.1	73±10.1	68±9.4	11.7	4.4	6.3	20.3
TC	0.03	86±4.5	97±10.2	89±1.1	4.9	1.4	1.4	8.3
DMC	0.1	85±9.7	82±8.5	95±2.9				-
ICTC	0.05	108±2.4	95±7.8	113±1.3	11.7	8.8	2.5	20.1
ECTC	0.1	80±2.0	87±3.9	85±7.5	16.3	7.5	6.5	27.9
EATC	0.05	83±9.7	61±2.4	64±11.5	7.5	3.7	4.2	12.8
ATC	0.05	75±14.2	76±6.8	71±12.2	7.9	4.4	2.8	13.6
MTC	0.05	79±8.4	84±5.0	67±6.1	7.5	4.0	4.4	12.9
β-apo-OTC	0.1	78±1.0	73±12.7	68±1.2	14.0	12.8	5.1	24.1
DXC	0.1	113±5.6	80±1.7	91±7.1	9.5	7.2	4.0	16.4
EACTC	0.1	57±6.9	54±6.2	54±2.5	16.6	8.6	8.9	28.5
ACTC	0.05	50±16.4	55±8.3	48±6.0	15.1	9.1	8.2	25.9
CTC	0.1	82±10.7	77±5.2	86±1.5	11.7	5.8	4.6	20.2
SAs								
SGD	1	76±14.4	86±10.4	93±9.1	2	1.75	1.45	9
SA	0.5	79±18.5	87±20.0	95±20.0	1.75	1.25	0.6	(+)27.4
SPD	0.08	84±8.8	91±2.9	102±11.9	1	0.75	0.5	5
SDZ	0.2	78±10.1	89±7.6	93±10.0	1.5	1	1	12.2
STZ	0.15	82±10.6	92±5.2	89±16.4	2.5	1.3	1.3	15.2
SMR	0.07	80±9.0	86±1.7	105±10.9	1.5	1.3	1.1	6.5
SIM	0.06	81±10.8	88±3.7	102±12.3	0.8	0.5	0.5	13.3

SMO	0.07	75±11.8	85±6.6	96±12.1	0.8	0.5	0.5	17
SMT	0.03	75±10.0	84±9.0	75±19.2	2.5	1.5	1.5	1.7
SMA	0.08	82±9.8	90±4.6	113±12.0	1.3	0.8	0.8	8.9
SME	0.01	82±8.2	88±2.8	97±11.4	1.5	0.8	1.3	6.8
TMP	0.05	84±9.0	92±11.0	110±7.3	2.3	1.5	1.3	21
SMX	0.15	82±9.1	89±4.2	86±9.4	2.5	1.5	1.3	3.8
SIA	0.12	67±18.6	90±9.7	71±10.6	2.8	1.0	1.5	24.6
SMP	0.2	83±7.1	88±10.0	120±15.4	2.0	1.5	1.5	11
SMM	0.2	84±9.4	83±12.3	103±11.8	2.3	1.3	1.5	5.5
SCP	0.35	80±8.4	88±7.9	81±16.4	3.8	2.3	2.0	9.8
SQX	0.04	80±10.0	87±11.2	102±6.0	3.3	2.3	1.5	8.6
SDM	0.11	83±7.7	89±10.5	107±3.9	3.3	3.0	1.3	11.6
SNT	0.19	82±10.7	73±5.8	62±0.3	17.5	6.8	7.5	2.8
NAcSPD	0.11	81±4.2	116±7.2	110±3.2	5.0	2.5	2.5	5.7
NAcSDZ	0.12	75±1.2	91±12.4	92±9.8	6.3	4.0	2.5	(+)9.6
NAcSMA	0.06	85±2.9	117±8.6	115±2.8	2.5	1.8	1.3	14.1
NAcSMX	0.5	91±5.0	98±7.9	120±8.9	5.0	4.3	3.3	12.3
NAcSMR	0.5	77±3.3	91±6.0	110±0.1	7.5	7.5	3.5	11.7
$^{13}C_6$-SMA	0.05	82±4.9	89±0.4	110±10.8	-	-	-	3.6
NAcSMX-d_5	0.5	89±11.1	92±6.9	109±8.4	-	-	-	15.8
FQs								
PIPE	0.15	57±2	53±11	63±8	4.4	11.0	2.3	(+)4.0
FLER	0.06	80±7	81±3	94±5	0.6	1.5	0.5	4.3
OFL	0.1	61±21	61±6	71±7	6.0	15.0	7.5	(+)15.7
PEFL	0.1	94±4	80±11	112±6	3.5	8.8	2.3	(+)19.6
ENO	0.2	58±2	59±9	61±17	1.2	3.0	1.5	(+)17.0
NOR	0.15	76±2	64±13	88±9	10.4	26.0	6.5	(+)1.4
NOR-d_5	1.2	72±1	91±13	86±10	-			(+)14.4
CIPRO	0.25	59±3	75±18	63±9	1.3	3.3	1.0	(+)18.4
DANO	0.1	84±4	107±10	98±11	1.7	4.3	1.5	(+)26.3

ENRO	0.05	92±2	83±6	111±4	1.1	2.8	1.0	(+)23.9
LOME	0.3	63±5	62±11	72±14	0.4	1.0	0.5	15.5
DIF	0.15	68±3	81±5	80±9	2.7	6.8	3.3	3.1
SARA	0.1	58±3	66±17	62±17	9.0	22.5	5.5	13.4
GATI	0.15	56±4	46±2	59±20	1.5	3.8	0.8	5.2
SPAR	0.03	67±2	60±18	77±18	2.6	6.5	1.5	25.1
MOXI	0.1	91±4	99±14	106±3	9.6	24.0	4.8	(+)19.5
CINO	0.15	62±5	67±19	66±11	3.8	9.5	3.3	(+)1.3
OXO	0.55	61±3	54±13	64±16	1.8	4.5	1.3	22.9
NALI	0.05	66±3	71±18	68±2	3.3	8.3	2.0	21.8
FLUM	0.05	66±4	68±12	78±4	1.5	3.8	2.0	6.1
PIRO	0.05	57±3	55±14	58±14	3.9	9.8	3.3	20.8

Table S5. Number of *E. coli* Resistant to FQs, TCs, and SAs in Sampling Sites, and the Level of All Target Chemicals.

	Sampling sites	Q1	DQE	Q2	Q3	Q4	Q5	Q6	Q7	DQ2	DQ3	W1	W2	W3	W4	W5
FQs	FLER		9.5													
	OFL	71.9	854.0	54.9	116.7	81.3	39.5	30.0	251.3	18.5	2.1	73.4	39.7	25.1	42.4	208.2
	DANO															
	PEFL		5.9													
	CIP		15.0	12.7			6.7	4.5								
	NOR	28.8	140.3	160.0	199.4	81.8	95.9	41.5	33.2		11.1					60.7
	PIPE	15.3	10.2	1.3	19.5	12.7	9.1	4.9	10.2							4.3
	DIF															
	LOME	2.4	27.2					1.2	1.8						1.5	
	GATI	8.8	56.5	8.8	14.7	11.3	8.1	5.2	27.5	2.6		3.1	3.2	1.6	4.6	16.5
	MOXI															
	OXO	9.4	4.5	7.3	12.0	11.9	11.1	4.9	6.9	3.7		5.3				7.3
	PIRO	129.1		11.0	16.4		12.3								12.0	8.9
	FLUM	48.4	131.8	74.3	104.4	96.3	54.6	41.4	109.6	52.9		54.6	25.4	29.8	45.1	88.1
	NALI	78.1	5.2	6.1												
TCs	MINO	0.0	0.0	1.4	0.0	1.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	ETC	4.8	7.7	4.8	3.7	4.5	3.0	0.0	0.0	0.0	0.0	1.1	0.0	0.0	0.0	0.0
	OTC	30.9	41.1	37.2	34.9	30.2	32.1	31.4	11.5	9.8	0.0	39.1	0.0	0.0	0.0	8.5
	TC	7.1	12.6	7.9	5.9	7.2	3.4	5.2	3.0	2.2	0.0	2.1	0.0	0.0	1.4	1.4
	ICTC	29.1	3.7	6.2	6.1	4.4	13.0	12.3	5.5	14.1	0.0	3.4	1.9	0.0	0.0	2.3
	DXC															
	ATC															
	EACTC	0.0										0.0	0.0	0.0	0.0	0.0
SAs	SGD			0.7					0.9	1.6		1.2	0.9	1.0	1.0	1.3
	SA								0.5	1.3		0.9	1.6	0.7	1.1	1.0

	SPD	28.6	91.1	51.9	35.1	29.3	31.1	40.4	49.4	13.2	0.2	6.8	3.7	1.3	4.8	21.5
	SDZ	176.5	245.4	216.9	106.3	121.6	192.6	221.1	216.3	142.9	0.8	46.9	136.4	107.7	131.5	151.5
	STZ				0.8		0.3		0.5	2.0		0.2	0.7		0.5	0.5
	SMA	0.9	2.0	1.4	4.6	1.4	1.4	2.2	4.2	8.8	0.9	37.2	37.0	29.6	39.4	19.1
	SME		0.0		1.2	0.5	0.6	1.0	0.5	1.7		4.4	9.5	5.4	6.0	2.9
	TMP	77.1	138.7	67.3	81.3	77.0	50.7	71.5	58.6	44.2	27.7	16.1	34.1	22.8	23.8	54.8
	SMX	138.2	443.5	272.0	221.0	246.9	234.1	293.0	284.2	136.0		71.3	97.5	83.6	95.3	164.6
	SMM	1.4	0.7	1.1	1.4	0.8	1.2	0.8	1.3	3.7		5.5	11.6	9.3	10.8	6.4
	SCP		0.0		0.5	1.5	1.1	1.8	3.9	1.9		1.4	3.1	2.8	2.3	7.4
	SQX		1.1		0.7	1.4	0.6					0.8	1.0	2.1	1.0	1.0
	NAcSPD	37.9	119.0	107.3	70.4	76.8	65.2	116.6	87.9	13.2		8.7	7.8	4.5	7.5	48.9
	NAcSDZ	175.1	218.2	213.6	106.4	152.2	220.9	317.7	207.9	77.3	0.8	33.2	61.5	51.1	63.5	146.8
	NAcSMA	3.4	4.1	3.8	4.0	2.9	3.8	8.0	3.7	2.7		21.7	35.6	25.1	31.3	20.3
	NAcSMX	434.5	313.5	459.3	352.4	461.4	595.0	753.2	520.7	241.3	1.1	218.6	229.5	205.8	201.6	319.3
	Numbers of *E. coli* resistant to three antibiotics (E.coli/ml)	57000	1700	41000	15000	13000	17000	2000	1000	300	6	2800	17	10	4	13
	Numbers of *E. coli* resistant to SXT(SAs, E. coli/ml)	13100	290	6400	4500	1700	2800	300	180	170	1	860	4	1	2	2
	Numbers of *E. coli* resistant to TC(TCs, E.coli/ml)	6400	440	3600	600	600	200	400	130	200	2	290	1	0	0	3
	Numbers of *E. coli* resistant to LEV(FQs, E.coli/ml)	4100	100	2000	1600	800	1200	100	60	50	1	130	2	0	0	5
	sampling sites	**W6**	**WT1**	**W7**	**W8**	**W10**	**WT2**	**W11**	**W12**	**W13**	**B5**	**W14**	**WT3**	**W16**	**T8**	**W17**
FQs	FLER							1.1				0.6		1.0		3.3
	OFL	225.9	27.0	28.7	62.2	45.2	110.0	53.5	116.7	116.3	345.6	296.7	77.6	185.7	202.4	1092.4

	DANO														3.4	
	PEFL															6.4
	CIP			6.9				5.5							18.1	7.0
	NOR			41.0	34.5			51.4			79.7	46.6		23.4	144.9	1.8
	PIPE		4.0				10.0							3.1	19.4	7.0
	DIF														2.6	
	LOME				2.9		4.5	6.9	3.6					2.9	4.9	7.0
	GATI	19.7	2.0	4.8	9.1	5.1	3.6	6.4	19.5	19.7	66.4	31.5	13.1	16.8	34.6	90.5
	MOXI															7.4
	OXO	8.5	12.2	5.9	8.6	3.7		1.7	5.6	4.0	20.8	2.1		3.7	14.0	3.1
	PIRO					4.1		4.1					5.7	2.1	72.7	
	FLUM	113.9	28.4	95.2	67.1	40.5	50.3	51.4	127.9	106.0	113.7	87.3	41.9	48.7	114.1	56.0
	NALI														69.7	
	MINO															2.0
	ETC	1.4		1.7					0.8	0.0	2.2			1.3	26.3	7.6
	OTC	11.9	20.1	12.7	13.3	7.4		4.9	5.6	9.0	12.3	8.6	20.2	8.8	110.2	46.8
TCs	TC	2.4	1.9	2.4	1.7	1.6		2.7	1.7	1.7	3.6	2.6	1.3	2.9	44.6	11.4
	ICTC	3.8	3.8	3.5	4.9	2.7		3.5	2.7	3.7	5.4	1.8	1.8	1.8	8.0	1.6
	DXC														13.4	
	ATC														0.0	
	EACTC														2.9	
	SGD	1.1	0.7	1.2	1.1	0.9	1.3	1.4	2.8	1.3	1.4	1.2	2.0	2.2	1.4	4.4
	SA	0.5	0.7	0.8	0.8		1.7	0.7	1.7	1.1	0.8	0.5	2.5	2.0	1.0	4.4
	SPD	25.4	18.9	23.6	14.1	18.1	2.6	22.3	51.7	20.8	17.3	19.7	4.9	32.0	129.3	48.1
	SDZ	174.5	177.9	146.1	121.4	141.8	54.4	175.9	342.0	188.0	123.1	97.5	215.6	185.1	739.2	460.1
SAs	STZ	0.4		0.4	2.6			0.4	0.9	0.4	0.3	0.3		0.4	0.8	0.5
	SMA	18.6	3.2	12.1	25.6	13.4	43.5	14.9	18.4	10.5	2.4	9.8	16.2	9.7	17.4	22.4
	SME	3.0	1.9	1.7	2.0	2.2		2.9	2.9	1.8	0.4	1.4	2.8	1.7		0.7
	TMP	63.9	46.7	47.2	39.0	49.8	12.8	39.4	119.7	61.4	30.4	48.3	14.5	48.2	79.2	82.6
	SMX	220.0	130.1	166.1	113.4	153.8	33.2	183.1	359.8	201.1	106.3	200.6	144.8	188.0	528.1	370.5

	SMM	5.4	3.3	0.3	4.9	4.1	0.9	5.3	5.1	3.8	1.3	2.7	17.8	4.0	1.6	1.4
	SCP	10.4	0.3	3.7	2.4	2.0	0.9	4.7	9.5	5.9		4.8	0.3	3.5	1.8	1.3
	SQX	1.6	0.7	0.6	1.1	0.7		1.0	1.1				0.5	0.7	1.0	1.0
	NAcSPD	48.7	22.5	66.7	21.7	44.6	6.0	38.8	54.0	39.1	37.1	45.9	6.9	29.3	192.6	89.9
	NAcSDZ	131.5	114.0	191.4	64.4	153.9	83.5	110.0	180.1	162.1	101.3	154.2	135.6	130.0	638.9	407.0
	NAcSMA	12.4	1.3	13.9	7.3	16.9	2.7	12.4	8.7	5.6	1.3	8.3	6.5	4.6	5.9	5.5
	NAcSMX	322.5	285.3	446.7	165.6	391.6	239.4	245.7	358.0	340.5	207.5	438.9	282.2	262.3	826.0	670.0
	Numbers of *E. coli* resistant to three antibiotics（E.coli/ml）	2100	20000	1300	8	50	31	68	22	8	1050	40	11	315	55000	17000
	Numbers of *E. coli* resistant to SXT（SAs, E.coli/ml）	340	2600	430	1	9	3	16	1	0	340	6	1	83	11400	4000
	Numbers of *E. coli* resistant to TC（TCs, E.coli/ml）	110	700	150	0	7	5	11	0	2	90	5	2	37	3800	650
	Numbers of *E. coli* resistant to LEV（FQs, E.coli/ml）	50	600	110	0	3	0	6	1	0	44	3	0	22	5950	900
	sampling sites	**T1**	**T3**	**T5**	**T4**	**DTE**	**T6**	**T7**	**BB1**	**DBE**	**BB2**	**BB3**	**DBB4**	**BL1**	**DJE**	
FQs	FLER		2.2			5.0		1.3		52.2					2.4	
	OFL	375.9	543.9	598.3	436.3	1308.6	1213.6	163.3	38.8	1110.1	33.5	88.1	11.3	34.1	159.3	
	DANO										1.0					
	PEFL					9.1				21.7						
	CIP					14.2	3.7	10.0		13.1	23.4	6.0		9.9	66.3	
	NOR		22.2	30.8		102.2	32.2	132.0	141.4	49.3	146.4	23.6		115.2	512.5	

	PIPE		4.9			8.9	1.8	18.4	15.8			2.9		13.6	19.5
	DIF										2.7				
	LOME		4.6			7.3	6.2	12.8	3.4	37.7	0.9	2.5		3.9	15.2
	GATI	44.2	46.9	38.6	39.8	72.5	91.9	26.3	6.9	28.2	14.3	19.5		5.2	28.4
	MOXI						10.6								
	OXO	5.3		1.5			1.6	7.1	8.7	2.6	12.7	5.0		7.1	6.6
	PIRO	16.8					4.8	22.7		7.1	12.6			17.1	57.4
	FLUM	24.2	56.2	69.8	79.8	93.8	64.0	90.4	39.4	139.2	50.0	30.5	28.0	33.2	154.6
	NALI														112.7
	MINO									2.7					5.8
	ETC	3.2	3.1	4.0	3.6	10.5	5.5	5.9	46.5	16.1	5.6	4.1	3.5	4.5	21.0
	OTC	14.3	5.1	7.9	7.8	74.0	37.7	53.5	101.6	92.3	52.2	27.6	4.3	36.8	214.3
TCs	TC	4.2	5.8	5.8	4.9	15.8	9.8	9.7	90.7	31.0	12.3	5.6	6.7	5.9	32.5
	ICTC	3.0	2.1	1.3	1.7	2.6	3.5	3.4	6.4	4.0	7.3	4.9	35.7	5.0	22.0
	DXC			8.4						8.1					1.5
	ATC														
	EACTC												50.1		
	SGD		1.6	1.5	0.9	2.0	2.5	2.9	0.7		1.2	0.7	1.1	0.6	2.6
	SA		0.6	0.7		0.6	0.8	2.7					1.7	0.5	1.0
	SPD	5.9	31.2	25.8	24.1	52.7	51.2	95.1	8.9	46.6	54.7	15.9		2.0	60.9
	SDZ	67.9	201.1	194.6	197.3	304.5	292.7	180.7	115.0	187.7	238.6	108.2	6.3	76.6	321.0
	STZ		0.5	0.5	0.5	0.3	0.3	1.6						0.2	
	SMA		4.0	3.7	3.7	9.1	9.8	267.3	0.8	0.8	1.1	1.7		1.6	1.5
SAs	SME		0.7				0.6			0.0	1.6	1.1		0.7	0.3
	TMP	38.1	23.8	28.3	17.5	6.1	19.8	57.1	67.6	165.5	59.7	25.4	1.3	37.1	39.1
	SMX	58.4	228.1	214.6	180.6	355.0	341.4	395.4	115.5	282.5	164.6	74.5	5.0	78.1	202.4
	SMM		0.9	1.1	0.9	1.1	1.1	1.1	0.5	0.0	0.8	0.8			1.2
	SCP		0.9	0.7	0.7	1.1	1.3	2.3		0.5					
	SQX						0.6	0.8		0.0					
	NAcSPD	11.7	47.0	37.7	41.1	107.2	84.1	107.5	24.5	93.7	139.5	40.2		5.9	87.0

	NAcSDZ	70.3	93.6	92.2	113.3	110.8	198.5	205.4	96.1	109.4	268.5	105.1	1.7	78.7	225.4
	NAcSMA	0.9	1.4	1.4	1.4	2.3	2.9	4.2	0.6	1.8	1.5	1.5		3.4	1.8
	NAcSMX	214.6	285.7	256.9	286.9	213.9	87.4	623.4	640.6	162.7	571.2	239.2	7.7	562.0	329.2
	Numbers of *E. coli* resistant to three antibiotics（E.coli/ml）	1650	560	700	560	410	750	27000	1300	300	900	540	28	180	360000
	Numbers of *E. coli* resistant to SXT（SAs, E.coli/ml）	435	150	205	86	101	235	5150	1040	108	340	143		52	34000
	Numbers of *E. coli* resistant to TC（TCs, E.coli/ml）	65	130	45	76	102	35	1000	260	39	300	54	17	10	11000
	Numbers of *E. coli* resistant to LEV（FQs, E.coli/ml）	165	54	60	25	31	70	1500	170	19	80	33	1	8	12300
	sampling sites	**BL2**	**B1**	**B2**	**B3**	**B4**	**DQ1**	**DBB1**	**DBB2**	**DBB3**	**DBB5**	**DT1**	**DT2**	**DT3**	**DT4**
FQs	FLER	7.3											16.05	60.01	11.08
	OFL	685.9	195.5	972.4	89.5	105.0	61.17	368.09	0.78	35.30		966.34	758.90	1717.34	176.23
	DANO														
	PEFL											7.08			
	CIP	24.1		9.6	14.3			24.98		13.29		6.35	404.72	304.55	46.52
	NOR	142.3		74.2	41.4	19.5	68.02	1182.42	4.73	386.39		58.86	1710.70	1773.23	412.48
	PIPE	15.3	5.7			7.3	6.18			3.37		9.50		16.42	24.15
	DIF	2.1													
	LOME	15.7		8.7		1.6	1.65			2.13		1.79	60.51	1735.39	195.19
	GATI	76.9	36.6	116.4	12.1	21.7	8.53			2.42		101.03	24.84	259.35	17.25

	MOXI	14.9										8.18			
	OXO	3.3	7.7	6.7	6.9	9.1	8.65	5.58		1.69		0.36	9.86	36.04	27.62
	PIRO	20.2		6.4	13.6	8.1		79.72		11.34		8.08	127.85	395.58	203.89
	FLUM	65.4	27.1	62.1	109.7	93.5	88.72	114.52	2.98	39.25	11.51	62.64	106.34	66.69	78.11
	NALI	37.5	7.3					499.50		36.39			153.45	224.83	206.59
TCs	MINO	3.4									0.86				
	ETC	8.7	0.0	5.2	3.0	2.6	3.38	9.41	11.00	1.01	0.79	4.52	3.71	53.94	44.39
	OTC	60.8	13.6	41.7	30.0	19.7	26.15	704.93	47.19	26.06	9.38	30.67	33.28	153.47	42.49
	TC	13.7	1.8	8.0	5.0	5.6	5.31	15.76	16.06	1.60	0.95	7.55	6.11	89.36	67.24
	ICTC	7.2	5.7	5.7	6.0	4.6	2.90	10.51	0.92	3.58	1.49	3.06	43.21	70.74	9.41
	DXC							4.02				6.88			5.37
	ATC		0.4									4.13	5.35		
	EACTC														
SAs	SGD	2.8	1.0	1.5	0.8	1.2	1.1	0.9		1.5		3.4	3.0	2.6	9.6
	SA	2.0	0.8	1.1	0.6	0.6		0.5		1.0		1.6	3.6	0.6	2.1
	SPD	47.8	6.2	23.0	19.9	18.1	32.7		1.5	3.6	1.1	46.7	4.5	1970.7	47.6
	SDZ	260.8	129.1	193.4	189.3	163.9	173.6	311.6	40.7	1500.2	41.4	330.0	965.9	2544.3	2931.2
	STZ			0.3	0.3					1.3		1.3	0.6		2.3
	SMA	1.4	1.5	2.3	1.6	2.2	2.4	0.6		1.4	1.0	143.3	213.7	2.0	484.1
	SME		0.3	0.4	0.7	0.7	0.5							0.7	
	TMP	61.6	58.3	40.0	37.8	27.7	54.4	49.1	22.5	74.7	9.4	20.9	48.3	53.5	461.4
	SMX	162.9	90.4	118.9	229.1	128.6	208.0	183.4	27.2	174.1	25.8	346.1	827.8	486.5	1294.3
	SMM	1.1	1.9	2.6	1.5	0.9	1.0	0.4		0.3		1.0	0.6	0.9	
	SCP						9.8					1.7	1.6		
	SQX				0.6							0.8			
	NAcSPD	62.9	7.5	39.4	18.6	32.8	50.5	3.7	1.4	0.8	2.2	73.5	6.5	1011.7	92.6
	NAcSDZ	124.2	80.2	134.8	76.0	103.5	171.8	398.5	36.2	165.5	13.5	178.3	2442.3	1228.5	378.8
	NAcSMA	0.8	0.6	1.2	0.9	4.6	2.7	0.6		2.6		3.0	8.3	3.5	2.0
	NAcSMX	289.3	310.0	345.8	231.5	198.2	384.6	824.2	67.5	373.6	69.6	335.2	7800.1	2053.1	606.7
	Numbers of *E.*	298000	4800	300000	7100	1400									

	coli resistant to three antibiotics（E.coli/ml）					
	Numbers of *E. coli* resistant to SXT（SAs, E.coli/ml）	11500	980	8600	2200	470
	Numbers of *E. coli* resistant to TC（TCs, E.coli/ml）	3100	260	2000	1000	170
	Numbers of *E. coli* resistant to LEV（FQs, E.coli/ml）	3800	220	1800	320	50
	sampling sites	**T2**	**W9**	**W15**	**DB1**	
FQs	FLER	0.8				
	OFL	421.5	100.9	226.4	61.14	
	DANO					
	PEFL					
	CIP	10.2		2.7	11.14	
	NOR	24.3			182.91	
	PIPE	3.5		2.1	25.20	
	DIF					
	LOME	3.4			1.29	
	GATI	39.3	14.5	16.7	3.33	
	MOXI					
	OXO	1.2	12.3	2.9	3.26	
	PIRO			3.7	30.41	
	FLUM	42.3	137.0	54.4	11.75	

	NALI	11.3			22.99
TCs	MINO	1.2	0.0	0.0	
	ETC	4.0	0.0	1.9	
	OTC	14.5	12.2	12.5	
	TC	5.6	2.7	2.2	
	ICTC		3.3	1.1	
	DXC				
	ATC				
	EACTC				
SAs	SGD	0.8	1.2	1.3	1.2
	SA	0.4	1.3	0.8	0.6
	SPD	21.2	17.6	19.5	37.3
	SDZ	159.4	148.1	239.5	172.7
	STZ		0.2		0.6
	SMA	3.7	14.3	12.2	2.1
	SME		1.8	2.5	3.0
	TMP	17.1	44.5	56.7	78.0
	SMX	168.6	105.9	233.0	239.8
	SMM	0.7	3.9	9.0	1.7
	SCP	0.7	2.4	2.7	
	SQX		0.6		
	NAcSPD	43.7	80.9	20.6	64.1
	NAcSDZ	127.8	227.1	100.0	84.6
	NAcSMA	1.8	23.3	4.8	3.5
	NAcSMX	345.8	447.1	296.6	682.0
	Numbers of *E. coli* resistant to three antibiotics （E.coli/ml）				
	Numbers of *E.*				

	coli resistant to SA（SAs, E.coli/ml）	
	Numbers of *E. coli* resistant to TC（TCs, E.coli/ml）	
	Numbers of *E. coli* resistant to levofloxacin（FQs, E.coli/ml）	

Sampling sites in red: No data on *E. coli* Isolation, including 3 river samples (T2, W9, W15), 10 discharging sites samples (DQ1, DB1, DBB1, DBB2, DBB3, DBB5, DT1, DT2, DT3, DT4). Sampling sites in blue: data reported in our previous papers.[1, 2] Sampling sites in black: data were collected in the present study.

Table S6. Concentrations and Detection Frequencies of Target Antibiotics in Wenyu Rivers.

Compound[a]	n[b]	Frequency (%)	Median (ng/L)	Range (ng/L)
FQs				
OFL	45	100	110	25.1-1213.6
GATI	45	100	16.5	1.6-116.4
FLUM	45	100	56.2	24.2-137.0
OXO	38	84.4	6.9	ND-20.8
NOR	29	64.4	46.6	ND-199.4
PIPE	24	53.3	7.2	ND -19.5
LOME	22	48.9	3.5	ND -15.7
PIRO	21	46.7	12	ND-129.1
CIP	17	37.8	9.6	ND-24.1
FLER	8	17.8	1.2	ND-7.3
DIF	3	6.7	2.6	ND-2.7
MOXI	3	6.7	10.6	ND-14.9
DANO	2	4.4	2.2	ND-3.4
PEFL	1	2.2	6.4	ND-6.4
NALI	6	13.3	24.4	ND-78.1
Total	45	100	287.5	56.5-1430.3
TCs				
TC	42	93	3.6	ND-90.7
OTC	41	91	14.3	ND-110.2
ICTC	41	91	3.5	ND-29.1
ETC	29	64	2.2	ND-46.5
DXC	2	7		ND-13.4
MINO	5	11		ND-3.4
EACTC	1	2		ND-2.9
ATC	1	2		ND-0.4
Total	45	100	23.6	ND-296.6
SAs				
SPD	45	100	21.5	1.3-129.3
SDZ	45	100	175.9	46.9-739.2
TMP	45	100	47.2	12.8-119.7
SMX	45	100	166.1	33.2-528.1
NAcSPD	45	100	40.2	4.5-192.6
NAcSDZ	45	100	127.8	33.2-638.9

NAcSMA	45	100	4.0	0.6-35.6
NAcSMX	45	100	319.3	87.4-826.0
SMA	44	98	7.2	ND-267.3
SMM	43	96	1.4	ND-17.8
SGD	39	87	1.2	ND-4.4
SME	34	76	1.6	ND-9.5
SA	33	73	0.8	ND-4.4
SCP	32	71	2.1	ND-10.4
STZ	27	60	0.4	ND-2.6
SQX	23	51	0.8	ND-2.1
Total	45	100	967	468-3164

[a]Full names and structures of chemicals are listed in Table 1. [b]Number detected.
ND=Not detected.

References

(1) Hu, J.; Shi, J.; Chang, H.; Li, D.; Yang, M.; Kamagata, Y. Phenotyping and Genotyping of Antibiotic-Resistant Escherichia coli Isolated from a Natural River Basin. *Environ. Sci. Technol.* **2008**, *42*, 3415-3420.

(2) Shi, J.C.; Hu, J.Y.; Chang, H.; Wan, Y.; Zhang, Z.B.; Xiang, Y. Investigation on the antibiotic-resistance E.coli in Wenyu river in Beijing. *China Environmental Science*, **2008**, 28, 39~42