

# Occurrence, Distribution and Ecological Risks of Fluoroquinolone Antibiotics in the Dongjiang River and the Beijiang River, Pearl River Delta, South China

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**Abstract** The occurrence and distribution of five selected fluoroquinolones (FQs) were studied in the Dongjiang River and the Beijiang River, South China. Ciprofloxacin, norfloxacin and enoxacin, used as human and veterinary medicines, were detected with detection frequencies of 75%–100% and average concentrations of 9.5–18.8 ng L<sup>-1</sup> in the two rivers. Meanwhile, enrofloxacin, which is only used as veterinary medicine, was detected at lower levels (2.9–4.0 ng L<sup>-1</sup>) than those of ciprofloxacin, norfloxacin and enoxacin. The spatial distribution of the five FQs exhibited a close relationship with the intensity of local human activity. Certain antibiotics were detected in industrial wastewater and domestic sewage at considerably higher concentrations than those measured in the river water, indicating important sources of antibiotic contamination. Finally, an ecological risk assessment based on the

calculated risk quotient showed that ciprofloxacin could pose high risk to *Microcystis aeruginosa* (*M. aeruginosa*). The two rivers are important sources of drinking water and should arouse the attention of relevant departments. Effective measures must be taken to strengthen the protection of the two rivers.

**Keywords** Antibiotics · Fluoroquinolones · Dongjiang River · Beijiang River · Risk Assessment

Fluoroquinolones (FQs) have been rapidly developed for nearly 30 years as landmark antimicrobial agents. FQs were the most studied and extensively synthetic antimicrobials in recent years (Xu et al. 2006). FQs have a wide antimicrobial spectrum, strong antibacterial properties, simple structures, distinct curative effects, do not have common cross resistances, and come at a low price. Therefore, these agents are widely used in animal and human disease control (Kools et al. 2008). At present, FQs have been developed and put into mass production in China. Ciprofloxacin (CPFX), norfloxacin (NOR) and enoxacin (ENO) are the most consumed antibiotics in China (Wang and Cheng 2009). Many FQs are partially metabolized in the body and are thus excreted along with their metabolites via urine and faeces (Jjemba 2002), then discharged into the environment through various channels, causing both resistance (Hartmann et al. 1998; Wang 2006) and environmental pollution (Kemper 2008; Thiele-Bruhn 2003) problems. In recent years, the residues of FQs in aquatic environments and their potential harm have drawn considerable attention by the authorities and scientific community.

The Pearl River Delta (PRD) located in South China is one of the most urbanized and densely populated regions in China. As previously reported, antibiotics were detected

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in the aquatic environment in the PRD, including in the eight major outlets (Xu et al. 2013), the urban section of the Pearl River at Guangzhou (Peng et al. 2011) and other areas from Heyuan to the Shenzhen River (Chen et al. 2013). However, the level and distribution of FQs in the Beijiang River and Dongjiang River, the two largest tributaries of the Pearl River and two important water sources, have been less studied. The middle and lower reaches of the Dongjiang River are located on the east side of the PRD, which is one of China's most important economic areas and the largest manufacturing production base. In addition, the environment in this area bears enormous pressure because of serious problems related to being highly urbanized, densely populated with a large number of migrant workers and no <1.8 billion tons of treated wastewater draining off into the Dongjiang River annually (Water Resource Department of Guangdong 2009). In particular, the deterioration of the downstream water quality has been listed as a national key river basin water quality protection priority. The Beijiang River is the second largest river of the PRD. Some important cities including Shaoguan, Qingyuan, Zhaoqing, Foshan, and Guangzhou are along the river, and form the northern part of the PRD. In addition to the dense population and a high number of industries, there is also developed livestock industry and aquaculture in the area. Therefore, large amounts of antibiotics are used, and the residues of such antibiotics may reach the nearby aquatic environment. Moreover, the Dongjiang River and the Beijiang River have traditionally played important roles in the water supply of the nearby cities and cities downstream of the PRD. If these rivers were polluted, the populations and production activities in the area would suffer serious harm. Therefore, much attention should be paid to the pollution problems of the Dongjiang River and the Beijiang River, and research on antibiotic pollution is quite necessary.

In the past, the occurrence and distribution of macrolide and sulphonamide antibiotics in the Dongjiang River were reported in an international journal (Zhang et al. 2012a) and a Chinese monograph (Ying et al. 2012). Results for FQs were only reported in a Chinese book (Ying et al. 2012) for the Dongjiang River and no report about FQs in the Beijiang River are available. Therefore, we believe the study about FQs in the Dongjiang River and Beijiang River will contribute to a more comprehensive understanding of the concentration of antibiotics on the surface of the PRD and do much contribution to the antibiotics database of the global rivers. Therefore, five most frequently prescribed FQ antibiotics for human and veterinary medicine in China were studied in the aquatic environment of the Dongjiang River and the Beijiang River in this study. The objectives of the present study were not only to survey the occurrence and distribution of the five antibiotics in the aquatic environment but also to identify the potential source of

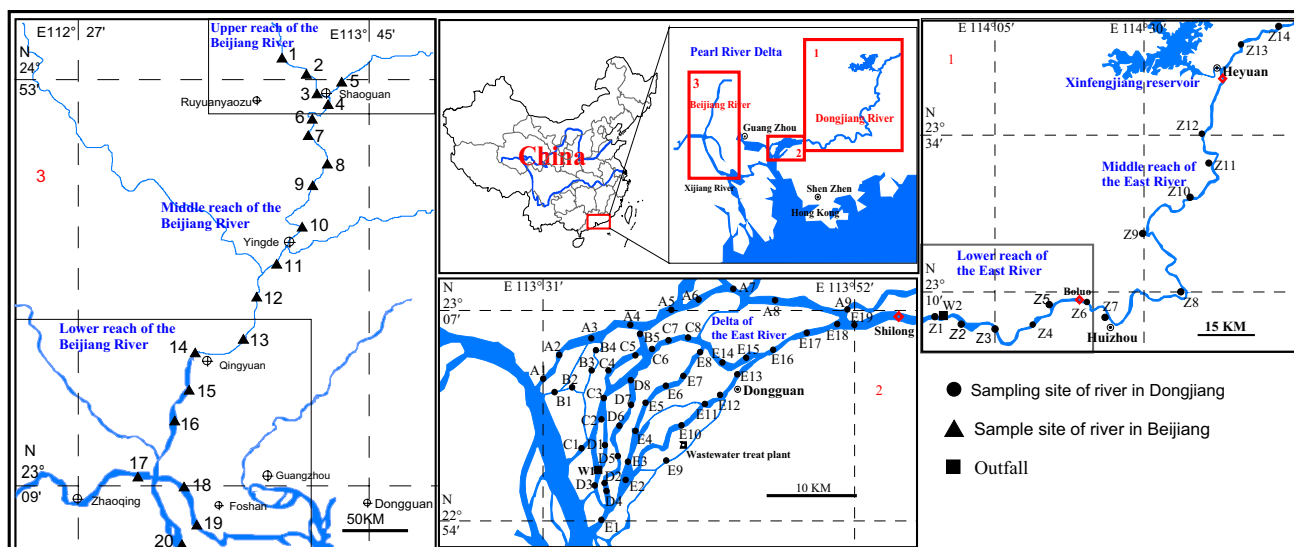
antibiotics. Meanwhile, the ecological risks of FQs to the organisms in the aquatic environment were also discussed.

## Materials and Methods

Five FQs standards were purchased from Sigma-Aldrich Co. (St. Louis, Mo, USA). They were CPF, NOR, ENO, enrofloxacin (ENRO) and ofloxacin (OFL). The physico-chemical properties and primary usage of the target compounds were shown in Supplementary Table S1 in our previous paper (Zhang et al. 2012b). The  $^{13}\text{C}_3$ -caffeine solution was purchased from Cambridge Isotope Labs (1 mg mL<sup>-1</sup> in methanol, USA) and used as a surrogate standard. All antibiotic compounds were dissolved in methanol and stored in a freezer. Methanol (HPLC grade) was obtained from Merck (Darmstadt, Germany). Formic acid and ammonium acetate were all of HPLC grade and purchased from CNW (Germany). Ethylenediamine tetraacetic acid disodium (Na<sub>2</sub>EDTA) was analytical grade and purchased from Tianjin Chemical (Tianjin, China). High purity water was prepared using a Milli-Q ultra-pure water apparatus, (Millipore, Bedford, MA, USA). Unless otherwise instructed, the chemicals used in the analysis were analytical grade or above.

The location of the study area and sampling sites are shown in Fig. 1. During the wet season, a total of 63 river water samples were collected from 12 waterways (49 samples) of the Dongjiang River delta, as well as from the middle and lower reaches (14 samples) in July 2009. Two additional wastewater samples were collected from outfalls to the river for comparison purposes. One wastewater sample (W2) was discharged from domestic sewage outfall and the other (W1) from an outfall near an industrial area including producers of antibiotics and other industries. In addition, 20 river water samples from the upper stream, via Lishi, Shaoguan, Wushi and Qingyuan of the Beijiang River were collected in December 2009. All samples were surface samples (within 50 cm under surface) collected using a stainless steel bucket in a fishing vessel at the centre of flow. Wastewater samples were obtained from the outfalls at the river bank. Samples were immediately transferred to a 5-L pre-cleaned amber glass bottle, and each bottle was rinsed with sample water three times prior to collection. The samples were kept at 4°C in a refrigerator and processed within 24 h.

The antibiotics in the water samples were concentrated by pre-conditioned solid-phase extraction (SPE) using the Oasis HLB cartridge (500 mg, 6 mL, Waters Corporation). Before the extraction, a specific volume of a water sample (2 L river water or 0.5 L sewage water) was filtered through 0.7 µm glass-fibre filters (GF/F, Whatman) and then acidified to pH=3.0 with 3.0 mol L<sup>-1</sup> H<sub>2</sub>SO<sub>4</sub>, followed by the



**Fig. 1** Sampling locations of water samples from the Dongjiang River and the Beijiang River

addition of 0.2 g  $\text{Na}_2\text{EDTA}$  as the chelating agent and 100 ng of  $^{13}\text{C}_3$ -caffeine as the surrogate to monitor the possible losses during the analytical procedure. Each water sample was passed through the SPE cartridge. The analytes were eluted with 2 mL of methanol three times. The volume of each of the analytes was reduced to approximately 20  $\mu\text{L}$ , and then, the analyte was dissolved in 40% aqueous methanol to the final volume of 1.0 mL.

For the recovery experiments, the five target compounds were determined for river water using the standards addition method (Zhang et al. 2012b), i.e., 2 L of filtered river water fortified with 100 ng of target analytes was processed in the same procedure as the field samples.

The extracted antibiotics were analysed using high-performance liquid chromatography–electrospray ionization tandem mass spectrometry (HPLC-ESI-MS-MS) with multiple reaction monitoring (MRM). The instrumental analysis method was published in Supplementary Table S4 of a previous paper (Zhang et al. 2012b). The separation of the FQ compounds was performed with Agilent 1200 series on an ODS-P (Dikma, USA) (4.6  $\times$  250 mm, 3.5  $\mu\text{m}$ ) chromatograph column. For mass spectrometric analysis, a Sciex API 3200 mass spectrometer (Applied Biosystems, Foster City, USA) equipped with an electrospray ionization source in the positive mode (ESI<sup>+</sup>) was used to analyse the FQs.

A quantitative analysis of each compound was performed using HPLC-ESI-MS-MS with MRM mode using one or two of the highest characteristic precursor ion/product ion transitions. Together with the retention times, the characteristic ions were used to ensure correct peak assignment and peak purity. The limits of quantification (LOQ) for each compound in the river water were defined as a signal-to-noise (S/N) ratios of ten (Xu et al. 2007). The LOQs

for these antibiotics selected in this study were from 1.3 to 5.0 ng  $\text{L}^{-1}$  in the river water (Table 1). Recovery for the surrogate was  $80\% \pm 9\%$ . The recovery rates of these spiked antibiotics ranged from 70% to 76%.

In this study, the antibiotic content in the sample data was processed by the software (MassHunter) of the Agilent HPLC-MS/MS. Distribution histogram and correlated analysis were performed using Statistical Package for Social Science (SPSS version 17.0).

## Results and Discussion

The concentrations of antibiotics in the surface water from the Dongjiang River and the Beijiang River are summarized in Table 1. The concentrations of antibiotics detected in the Dongjiang River water ranged from nd (not detected) to 53.6 ng  $\text{L}^{-1}$  with high detection frequencies of at least 87% for all compounds except for OFL. NOR and ENO were detected in all samples with average concentrations of 18.8 and 17.7 ng  $\text{L}^{-1}$ , respectively. CPFX also exhibited high detection frequencies of 91% with an average concentration of 10.6 ng  $\text{L}^{-1}$ , followed by ENRO with a detection rate of 87% and an average concentration of 2.9 ng  $\text{L}^{-1}$ . OFL was below the detection limits in the Dongjiang River water.

In the Beijiang River, all five FQs were detected in most of the samples and the concentrations ranged from nd to 43 ng  $\text{L}^{-1}$  (Table 1). It should be noted that NOR and ENO were also the most frequently detected compounds, with high detection frequencies of 95% and high average concentrations of 17.3 and 17.5 ng  $\text{L}^{-1}$ , respectively. CPFX, which was next highest compared to NOR and ENO, was

**Table 1** Summary results for the selected FQs in the Dongjiang River and the Beijiing River

	CPFEX	ENRO	NOR	ENO	OFL
Dongjiang River					
River water					
DF (%)	91	87	100	100	0
Range (ng L <sup>-1</sup> )	nd to 23.1	nd to 6.1	<LOQ to 41.3	<LOQ to 53.6	nd
Mean <sup>a</sup> ± STD (ng L <sup>-1</sup> )	10.6 ± 6.1	2.9 ± 2.2	18.8 ± 8.2	17.7 ± 14.0	nd
Median (ng L <sup>-1</sup> )	11.3	<LOQ	18.3	12.0	nd
Wastewater					
W1 (ng L <sup>-1</sup> )	49.3	9.8	39.7	247.4	20.1
W2 (ng L <sup>-1</sup> )	42.1	<LOQ	74.4	13.7	42.3
Dilution ratios = W2/ River water <sup>b</sup>	4.0	0.86	4.0	0.8	–
Beijiing River					
DF (%)	75	35	95	95	5
Range (ng L <sup>-1</sup> )	nd to 40.4	nd to 43.0	nd to 35.3	nd to 38.9	nd to 36.7
Mean ± STD (ng L <sup>-1</sup> )	9.5 ± 8.7	4.0 ± 9.6	17.3 ± 7.6	17.5 ± 8.7	1.8 ± 8.2
Median (ng L <sup>-1</sup> )	10.1	nd	15.8	17.6	nd

DF detection frequencies, nd no detected, LOQ limits of quantification

<sup>a</sup>Mean values were calculated using the measured values if it was above the LOQ, the 1/2 LOQ if it <LOQ or 0 if not detected

<sup>b</sup>Ratios of selected antibiotics concentrations in W2 to those in the river water

detected with detection frequencies of 75% and mean concentration of 9.5 ng L<sup>-1</sup>. The detection frequency of ENRO was only 35%, with its concentration ranged from nd to 43 ng L<sup>-1</sup> (mean 4.0 ng L<sup>-1</sup>). Otherwise, OFL was only detected in the sample R1, which was located in Wujiang, with high concentrations of 36.7 ng L<sup>-1</sup>. Concentrations of the other four kinds of FQs were also higher than the other sites. They were CPFEX, ENRO, NOR and ENO with concentration of 40.4, 43.0, 35.3 and 38.9 ng L<sup>-1</sup>, respectively. This may be caused by some point pollution, such as hospital wastewater, livestock industrial wastewater or certain factory wastewater. These results indicate that the Beijiing River may be polluted by the discharge of some sewage or wastewater.

The detection frequencies and mean concentrations of NOR and ENO were both the highest in the two rivers, followed by CPFEX and ENRO, while OFL was scarcely detected. The differences of their concentrations may be related to their degrees of usage. CPFEX, NOR and ENO were the predominant antibiotics, which presented higher medical consumption than other FQ antibiotics in China (Wang and Cheng 2009; Zhang and Liu 2010). CPFEX and NOR are also used extensively in animal husbandry and aquaculture. In particular, NOR is widely utilized in the livestock industry and aquaculture due to the low cost. Therefore, the detection rates of the three antibiotics are higher and may be widely present in the environment. As ENRO is only used as a veterinary drug and OFL is mainly used as human medicine, the consumption of these

antibiotics is limited. ENRO can be eliminated through hydration, adsorption, sedimentation, biodegradation and bioaccumulation after it reaches the water (Wu et al. 2006b). The degradation of ENRO is rapid, with the ability to fall below 50% of the original concentration within 5 h (Wu et al. 2006b). In addition, ENRO can be partly metabolized to CPFEX in the body of animals and can be easily photodegraded in the natural environment (Wu et al. 2006a). As a result, ENRO was detected at low frequency and low concentrations. Moreover, OFL possesses a higher curative effect, shorter course of treatment and better tolerance than NOR, but its usage is limited because of its relatively high price.

The detection rate and the content of FQs had similar spatial variation trends along the river flow direction in the Dongjiang River and the Beijiing River, and the concentration ranges of the same FQs changed slightly. However, the samples in the Dongjiang River were sampled during the rainy season while the samples in the Beijing River were sampled during the dry season. It has been found that antibiotics are usually present a relatively lower level during the rainy season than during the dry season (Luo et al. 2011; Xu et al. 2007). Therefore, higher concentration can be expected in the Dongjiang River than that of the Beijiing River in the same season.

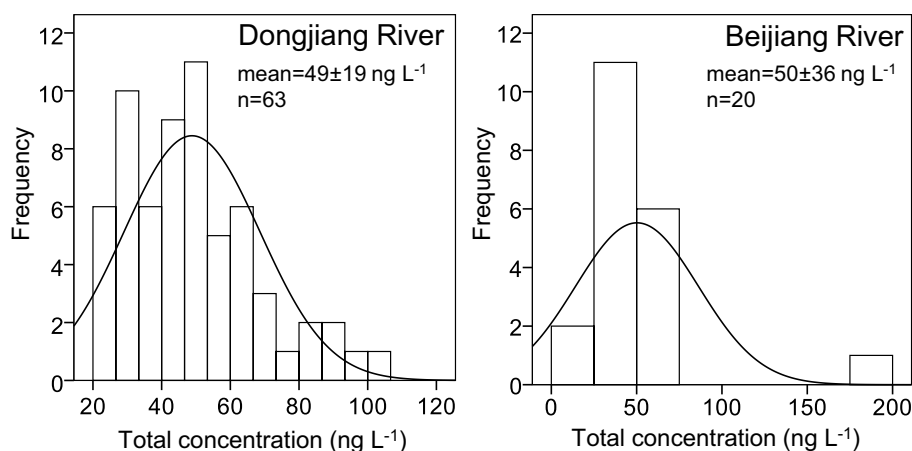
Compared to other rivers or segments of the Pearl River in the PRD, the average concentration of NOR was lower than that in the Shenzhen Reservoir (38.5 ng L<sup>-1</sup>), while higher than that in Heyuan, Dongguan and

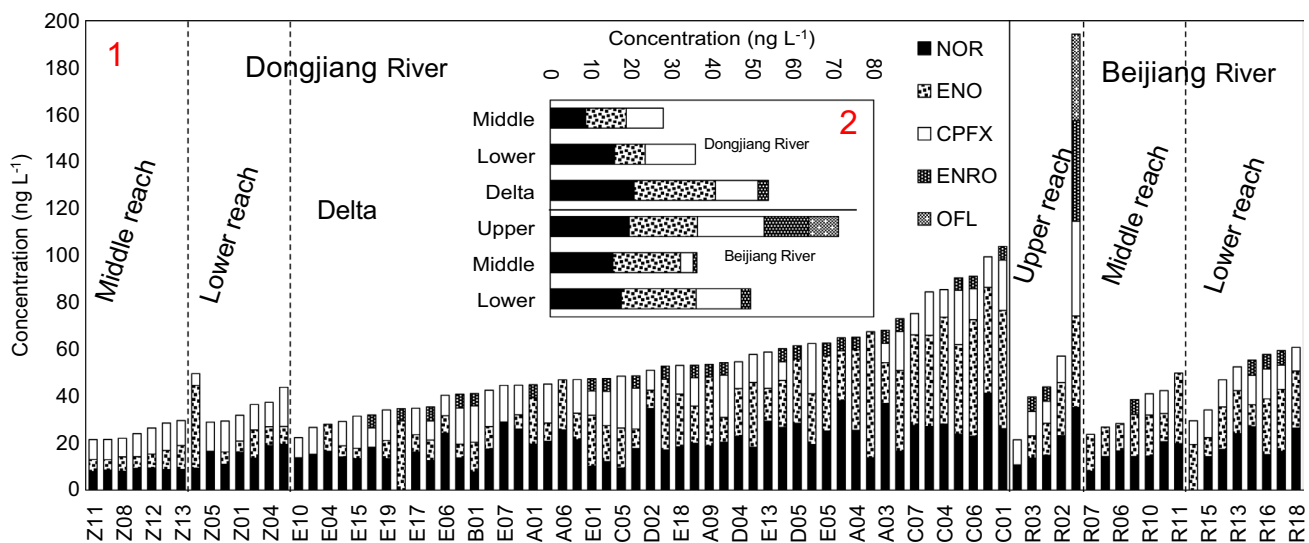
Huizhou (not detected) (Chen et al. 2013), and the Yangtze Estuary (nd to  $14.2 \text{ ng L}^{-1}$ ) (Yan et al. 2013) and Lake Taihu (not detected) (Zhou et al. 2016). It was higher than those measured in the Guangzhou segment of the Pearl River during the high water season (nd to  $13 \text{ ng L}^{-1}$ ) but was lower than that during the low water season ( $117\text{--}251 \text{ ng L}^{-1}$ ) (Xu et al. 2007). The concentration of ENRO in this study was comparable to the levels in the Yangtze Estuary (nd to  $4.77 \text{ ng L}^{-1}$ ) (Yan et al. 2013) and Lake Taihu (nd to  $7.57 \text{ ng L}^{-1}$ ) (Zhou et al. 2016). The concentration of CPFX in these two rivers was higher than those measured in the Yangtze Estuary (nd to  $2.27 \text{ ng L}^{-1}$ ) (Yan et al. 2013) and Lake Taihu (not detected) (Zhou et al. 2016). OFL was barely detected in the two rivers, which was comparable to Heyuan, Dongguan and Huizhou (not detected), but it was lower than that in Shenzhen Reservoir ( $52.4 \text{ ng L}^{-1}$ ) (Chen et al. 2013), the Yangtze Estuary (nd to  $12.4 \text{ ng L}^{-1}$ ) (Yan et al. 2013) and Lake Taihu (nd to  $26.3 \text{ ng L}^{-1}$ ) (Zhou et al. 2016). Otherwise, it was detected with a concentration ranging from lower LOQ to  $16 \text{ ng L}^{-1}$  (rainy season) and  $53\text{--}108 \text{ ng L}^{-1}$  (dry season) in the Guangzhou segment of the Pearl River (Xu et al. 2007). Compared to another result which was detected in the Dongjiang River at the similar time, the concentrations of CPFX and ENRO were at the same order of magnitude (Ying et al. 2012). While NOR presented higher concentration in five sampling sites ( $100\text{--}289 \text{ ng L}^{-1}$ ) in the study reported by Ying et al. (2012). As a whole, the antibiotic contamination in the two rivers was significantly lower than that in Guangzhou segment of the Pearl River. That may be caused by the differences in human activity intensity. In addition, the antibiotic contamination in surface water of the two rivers was higher than that detected in the Lake Taihu and Yangtze River in the Yangtze River Delta. It indicates that the two rivers in the study area were seriously affected by industrial and human contamination.

Figure 2 shows the results of the frequency histogram of the total concentration of antibiotics in the Dongjiang River and the Beijiang River. The total concentration of FQs at each sampling site in this study is mainly less than  $60 \text{ ng L}^{-1}$ , accounting for 84% of the samples in the Dongjiang River. Moreover, in the Beijiang River, the total concentration mainly focused on between 40 and  $60 \text{ ng L}^{-1}$ , accounting for 55% of the samples. FQs did not accord with the normal distribution in the Dongjiang River and the Beijiang River based on the “K-S single-sample nonparametric test.” It suggested that their concentrations were influenced by point sources.

Figure 3 shows the spatial distribution of antibiotics in the two rivers. The total antibiotic concentrations in the three parts of the Dongjiang River presented in the following order:  $C_{\text{Delta}} > C_{\text{Lower}} > C_{\text{Middle}}$ . In particular, the concentrations of NOR and ENO, the two most common antibiotics, were significantly higher in the river delta than in the middle or lower reaches ( $p < 0.5$ ). As shown in Fig. 1, the river delta is mainly located in Dongguan City. It was estimated that there were more than 7 million residents and the population density of this city reached nearly 3000 people per square kilometre by the end of 2009. The wastewater discharged into the river from Dongguan was 930 million tons in 2008 (Water Resource Department of Guangdong 2009). Therefore, the NOR and ENO in the delta may mainly come from domestic sewage. In the main agricultural area through the mountains and the east shelter-forest region, these antibiotics may mainly come from livestock breeding or aquaculture wastewater. CPFX was used extensively in human medicine, livestock breeding, and aquaculture, so the content of CPFX was concentrated near the emissions of domestic sewage, livestock breeding, and aquaculture wastewater, and the levels in the three parts were not significantly different ( $p > 0.3$ ). In Dongguan, 707 million  $\text{m}^3$  of water was for domestic use, 1003 million  $\text{m}^3$  for industry and 141 million  $\text{m}^3$  for agriculture in

**Fig. 2** The FQs distribution histogram of the Dongjiang River and the Beijiang River





**Fig. 3** Distribution of fluoroquinolones in the Dongjiang River and the Beijiang River (1 the concentration of antibiotics in each sampling site in the two rivers; 2 the average concentration distribution of antibiotics in different regions of the two rivers)

2008, while the corresponding values were 251 million, 537 million and 1274 million in Huizhou, respectively (Water Resource Department of Guangdong 2009). Therefore, domestic sewage may contribute the major part of CPFX residues in the delta and parts of the lower reach in Dongguan, while agricultural wastewater (including aquaculture wastewater) may be the major contamination source in the Huizhou segment of the river. The average concentration of ENRO was  $2.9 \text{ ng L}^{-1}$ , obviously lower than that of NOR, CPFX, and ENO. ENRO is only used as veterinary drug, and it is widely used in livestock industry as an antimicrobial agent. Moreover, ENRO was discharged into the sewers with livestock and poultry excreta, so the discharge of wastewater was the main source of ENRO. As a whole, the concentration of ENRO in the delta was significantly higher than in the middle and lower reaches ( $p < 0.05$ ). Guangzhou and Dongguan were important aquaculture bases (Gao et al. 2010), thus the concentration of ENRO was affected by thriving aquaculture in the delta.

The average concentrations of total FQs in different segments of the Beijiang River were as follows:  $C_{\text{upper}} (71.3 \pm 62.5 \text{ ng L}^{-1}) > C_{\text{lower}} (49.6 \pm 11.1 \text{ ng L}^{-1}) > C_{\text{middle}} (35.8 \pm 8.9 \text{ ng L}^{-1})$  (Fig. 3), but their differences were significant only between that of upper reach and middle reach ( $p < 0.1$ ). Among the five FQs, NOR and ENO presented relatively higher concentrations than the other three antibiotics. Furthermore, the concentrations of NOR and ENO were evenly distributed throughout all sampling sites. CPFX, used as a human medicine and veterinary drug, had higher ( $p < 0.05$ ) concentrations in the upper ( $16.5 \text{ ng L}^{-1}$ ) and lower reaches ( $11.2 \text{ ng L}^{-1}$ ) compared to the middle section of the river ( $3.1 \text{ ng L}^{-1}$ ). This difference may be

because the upper and lower reaches are near Shaoguan and Foshan, where human activity is relatively concentrated, and the river is vulnerable to industrial wastewater, municipal sewage, the livestock industry and aquaculture wastewater. The terrain of the middle section was mainly covered hills and less affected by human activities. ENRO was only detected in a few samples of R1, R3, R5, R9, R14, R16 and R19. These samples were mainly collected from the upper and lower reaches and may be polluted by some point sources of contamination. For example, R19 was adjacent to pig farms. This result showed that the point source contamination could directly affect the surrounding rivers. As a whole, the antibiotic contamination sources in the upper and lower reaches were more severe than in the middle section of the Beijiang River.

W1 and W2 were industrial wastewater and domestic wastewater samples, respectively. As shown in Table 1, the five FQs were detected in W1 and W2 with concentrations ranging from  $9.8$  to  $247.4 \text{ ng L}^{-1}$  and from  $< \text{LOQ}$  to  $74.4 \text{ ng L}^{-1}$ . The concentrations were higher than the corresponding average concentrations in the Dongjiang River, except for ENRO ( $< \text{LOQ}$ ) and ENO in W2.

The correlated relationship of the concentrations of some antibiotics in the river water and sewage may be used to estimate the sources. The ratios of some antibiotic concentrations in the possible source of the sewage and river water may serve as an index to estimate the source of the antibiotics to the river water (Zhang et al. 2012a). The ratios of selected FQs in the domestic sewage and river are shown in Table 1. The dilution ratios of CPFX and NOR in the domestic sewage and the Dongjiang River are similar in both environments. Therefore, CPFX and NOR may come from the same

source of domestic sewage. Other compounds, such as ENRO and ENO, present lower dilution ratios, indicating that these compounds might come from contamination sources other than domestic sewage. Of course, the discussion above was based on the hypothesis that these FQs are subject to similar environmental fates in river water and sewage. Therefore, a detailed study should be conducted for a better understanding of the sources and fates of antibiotics in the two rivers.

Risk assessments in terms of risk quotients (RQs) were calculated for *Microcystis aeruginosa* (*M. aeruginosa*) and *Pseudokirchneriella subcapitata* (*P. subcapitata*), all of which can be found in the river water. According to the European Technical Guidance Document on Risk Assessment (TGD), the effective concentration 50% ( $EC_{50}$ ), the lethal (European commission 2003) concentration 50% ( $LC_{50}$ ), the no observed effect concentration (NOEC), and the lowest observed effect concentration (LOEC) were used to calculate the predicted no-effect concentrations (PNEC). The assessment factor (AF) for the short-term/acute PNEC calculation was equal to 1,000 for each of the three trophic levels (algae, daphnia and fish). For the long-term/chronic PNEC calculation, an AF of 100, 50, or 10 is used according to two or three trophic levels (EC 2003; Saling et al. 2005).

$$\text{Acute PNEC calculation: PNEC} = \frac{EC_{50} \text{ or } LC_{50}}{1000}$$

$$\text{Chronic PNEC calculation: PNEC} = \frac{NOEC \text{ or } LOEC}{AF}$$

After calculating PNEC values, RQs were estimated for each antibiotic using the measured environmental concentrations (MEC) and PNEC values:

$$RQ = \frac{MEC}{PNEC}$$

In this study, PNEC values calculated for acute or chronic toxicity data of the target antibiotics were collected from literature and are shown in Table 2. The results of the RQs for each target antibiotic, which were calculated using the MEC in the surface water of the studied area, are listed in Table 3.

To characterize the risk levels, the ratios were classified into three risk levels: low risk ( $RQ < 0.1$ ), medium risk ( $0.1 \leq RQ < 1$ ) and high risk ( $RQ \geq 1$ ) (Hernando et al. 2006). As is shown in Table 3, the RQs for ENRO, NOR and OFL were all  $< 0.1$  and posed a low risk in this study. While CPFEX showed high ecological risk to the relevant sensitive aquatic organisms (*M. aeruginosa*) both in the Dongjiang River and the Beijiang River, and similar result was reported in the rivers adjacent the Laizhou Bay, North China (Zhang et al. 2012b). Thus, it is worthwhile to focus the attention of the relevant departments to the detection of antibiotics in the surface waters and control the abuse of these compounds because of their potential ecological risk or health risk.

This study investigated the occurrence, spatial distribution and sources of FQs in the Dongjiang River and the Beijiang River in South China. Five FQ antibiotics were measured in water samples from the different segments of the two rivers. The results showed that four selected FQs widely occurred and the total concentrations were mainly  $< 60 \text{ ng L}^{-1}$ . Two wastewater samples were also analysed for comparison in the river water, and the five FQs were all detected in higher concentrations compared to the river

**Table 2** Aquatic toxicity data of five antibiotics to the most sensitive aquatic species

Compound	Non-target organism	Toxicity data ( $\text{mg L}^{-1}$ )	Toxicity	AF	References
CPFEX	<i>M. aeruginosa</i>	$EC_{50} = 0.005$	Acute	1000	Halling-Sorensen et al. (2000)
ENRO	<i>P. subcapitata</i>	$EC_{50} = 3.1$	–	1000	Robinson et al. (2005)
NOR	<i>P. subcapitata</i>	$EC_{50} = 16.6$	Acute	1000	Eguchi et al. (2004)
OFL	<i>P. subcapitata</i>	$EC_{50} = 1.44$	Chronic	1000	Isidori et al. (2005)

AF assessment factor,  $EC_{50}$  half maximal effective concentration

**Table 3** The risk quotients of fluoroquinolones in the Dongjiang River and the Beijiang River

Compound	PNEC ( $\text{ng L}^{-1}$ )	Risk quotients					
		The Dongjiang River			The Beijiang River		
		Middle	Lower	Delta	Upper	Middle	Lower
CPFEX	5	2.32	3.34	4.61	8.08	1.96	2.56
ENRO	3100	<b>0.00</b>	<b>0.00</b>	<b>0.00</b>	<b>0.01</b>	<b>0.00</b>	<b>0.00</b>
NOR	16,600	<b>0.09</b>	<b>0.00</b>	<b>0.00</b>	<b>0.00</b>	<b>0.00</b>	<b>0.00</b>
OFL	1440	<b>0.00</b>	<b>0.00</b>	<b>0.00</b>	<b>0.03</b>	<b>0.00</b>	<b>0.00</b>

Italics—high risk, bold—low risk

water samples. In addition, a risk assessment based on the calculated RQ showed that CPFX posed high risks to the relevant sensitive aquatic organisms in the Dongjiang River and the Beijiing River. In conclusion, as important sources of drinking water, the two rivers should arouse the attention of relevant departments. Effective measures must be taken to strengthen the protection of the two rivers.

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