RESEARCH ARTICLE

The impact and mechanism of quaternary ammonium compounds on the transmission of antibiotic resistance genes

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Abstract

The emergence of antibiotic resistance genes (ARGs) in microbes can be largely attributed to the abuse and misuse of antibiotics and biocides. Quaternary ammonium compounds (QACs) have been used worldwide as common disinfectants and detergents; however, their potential impact on the spread and diffusion of ARGs is still unknown. In this study, we detected the QAC resistance gene ($qacE\Delta1$), the 1 integron gene ($intI1$), and 12 ARGs ($sul1$, $sul2$, cfr, cml, $fexA$, $tetA$, $tetG$, $tetQ$, $tetX$, $ermB$, $bla_{TEM,}$ and $dfrA1$) in 48 water samples from three watersheds by quantitative PCR (qPCR). We investigated the evolution of bacterial antibiotic resistance under QAC and antibiotic environmental pressures by long-term continuous culture. In addition, five QACs were selected to investigate the effect of QAC on the efficiency of conjugation transfer. The changes in bacterial cell membrane and production of reactive oxygen species (ROS) were detected by flow cytometry, revealing the mechanism by which QAC affects the spread of antibiotic resistance. Our results showed that the QAC resistance gene was ubiquitous in watersheds and it had significant correlation with $intI1$ and seven ARGs ($r = 0.999$, $p < 0.01$). QACs could increase the resistance of bacteria to multiple antibiotics. Furthermore, all five QACs promoted the conjugation transfer of the RP4 plasmid; the optimal concentration of QACs was about 10^{-1}–10^{-2} mg/L and their transfer efficiencies were between 1.33×10^{-6} and 8.87×10^{-5}. QACs enhanced membrane permeability of bacterial cells and stimulated bacteria to produce ROS, which potentially promoted the transfer of plasmids between bacteria. In conclusion, this study demonstrated that QACs may facilitate the evolution and gene transfer of antibiotic resistance gene among microbiome.

Keywords Antibiotic resistance gene · Quaternary ammonium compounds · Conjugation transfer

Introduction

Antibiotics are used worldwide to treat infectious diseases (Mcmanus et al. 2002; Singer et al. 2003), but antimicrobial resistance (AMR) can reduce their therapeutic effect and become a critical public health issue. In addition, antibiotic resistance genes (ARGs) carried by antibiotic-resistant bacteria (ARB) can be transferred between bacteria and environments, exacerbating the emergence and spread of AMR globally (Pruden et al. 2006; Zhu et al. 2017). Horizontal gene transfer (HGT) is widely regarded as one of the most important ways by which ARGs spread via mobile genetic elements (MGEs), such as plasmids and integrons (Su et al. 2015; Sentchilo et al. 2013; Gaze et al. 2011). Previous studies reported that antibiotics (Mc Cay et al. 2010), as well as some organics, chlorhexidine, heavy metals, and nutrient elements, can promote the HGT of ARGs (Bakeraustin et al. 2006; Luo et al. 2014; Jutkina et al. 2018; Moriguchi et al. 2016; Qiu et al. 2012). However, quaternary ammonium compounds (QACs) are a common kind of bactericidal chemical, with wide consumer usages and industrial applications (García et al. 1999; Holmberg 2002; Liu et al. 2017; Patrauchan and Oriel 2003), and little is known about the effect of QACs on the spread of AMR.

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QACs are omnipresent pollutants found worldwide in both natural and industrialized settings (Tezel et al. 2006, 2007). Studies have shown that QACs could be widely detected in waste water, surface water, aquatic sediment, and the soil (Liu et al. 2017; Ruan et al. 2014), and that QACs could induce multi-drug resistance in bacteria and gene transfer at sub-inhibitory concentrations (Marathe et al. 2017; Van et al. 2012; Jiao et al. 2017; Pal et al. 2015). The concentration of QACs in urban stormwater, domestic wastewater, treated water, and surface water were 27 mg/L, 0.50 mg/L, 0.050 mg/L, and 0.040 mg/L, respectively (Li and Brownawell 2010; Van et al. 2012). Several identical resistance mechanisms have been reported between QACs and antibiotics. The qac genes encode a large family of bacterial efflux pumps, which can expel many QACs and other compounds from bacterial cells, leading to a decrease in QAC susceptibility (Alekshun and Levy 2007; Jiao et al. 2017). Importantly, qac genes can be horizontally transferred via MGEs to other bacteria and are often transmitted in combination with other ARGs (Yong et al. 2009). Despite these studies, important gaps in our knowledge of the impact of QACs on AMR generation and spread still remain. We hypothesize that (1) QAC exposure may facilitate the evolution of antimicrobial resistance, and (2) the effect of QAC on bacteria may increase the co-occurrence and transfer of QAC resistance gene and ARGs in water environments.

Therefore, we characterized the distribution and occurrence of qacEΔ1, intI, and 12 ARGs in three major watersheds in Zhejiang Province, China. The objectives of this research are to (1) investigate the occurrence and distribution of QAC and antibiotic resistance genes in watersheds, (2) evaluate the effects of different QAC compounds on bacterial resistance and the spread of ARGs, and (3) explore the mechanisms of QACs' effect on the conjugational transfer.

Materials and methods

Sample collection and DNA extraction for PCR assays

A total of 48 samples were collected from three watersheds in June 2016 (Fig. S1). There were 20 sample sites in the Zhangxi watershed of Ningbo City recorded as N1–N20, 16 sites in the East Tiaoxi watershed of Huzhou City recorded as H1–H16, and 12 sites along the Wen Ruitang River of Wenzhou City recorded as W1–W12 (Zheng et al. 2017; Zhou et al. 2017). Each water source was sampled in triplicate and stored at 4 °C. Water samples were pretreated within 12 h, water was collected by vacuum filtration of water onto 0.22-μm-pore-size nitrocellulose filters (Millipore, USA) until the filter clogged, and filtered water volume was recorded (Zhou et al. 2018). The DNA from the water samples was extracted using the FastDNA SPIN Kit (MP Bio, USA)

according to the manufacturer's instructions. The concentration of DNA was determined using a NanoDrop spectrophotometer (NanoDrop ND-2000c; Thermo, USA) and stored at − 80 °C until further analysis. In this study, 15 genes were detected by qPCR using a Bio-Rad IQ5 qPCR instrument (Bio-Rad, USA) as described previously (Table S1) (Cui et al. 2016).

Laboratory adaptation of resistance

Briefly, *Pseudomonas aeruginosa* was isolated from river samples and further confirmed bacterial taxon by 16S rRNA gene amplification and sequencing using the universal primers 27F and 1492R (Weisburg et al. 1991) and selected for further research. It was repeated in batch cultures for 30 days under selection pressure of QACs (dodecyl trimethyl ammonium chloride, DTAC; didodecyl dimethyl ammonium chloride, DDDAC; dodecyl dimethyl benzyl ammonium chloride, DDBAC) and antibiotics (tetracycline, Tc; ciprofloxacin, Cip), respectively. The initial concentrations of DTAC, DDDAC, DDBAC, Cip, and Tc were 100 mg/L, 100 mg/L, 100 mg/L, 40 mg/L, and 1 mg/L, respectively. The basic information of QACs is shown in Table S2 (Tezel et al. 2006, 2007). Each generation increased the defined concentration of antimicrobial substances by 20 mg/L, 20 mg/L, 20 mg/L, 1 mg/L, and 20 μg/L, respectively. Controls were continuously cultured for 30 days without selection pressure. Then, the selection pressure was withdrawn, and the bacteria were cultivated for another 30 days. The sensitivity of bacteria to Tc, Cip, and chloramphenicol (Chl) was tested every 5 days using the disk diffusion test. Sensitivity was evaluated based on the diameter of the inhibition zone.

Conjugative transfer system and identification of transconjugants

E. coli HB101 was used as a donor which carrying an RP4 plasmid and resistant to kanamycin (> 50 mg/L), ampicillin (> 60 mg/L), and tetracycline (> 40 mg/L). *E. coli* NK5449 was used as a recipient and resistant to rifampicin (> 160 mg/L) and nalidixic acid (> 50 mg/L). The strains were cultured in liquid LB medium supplemented with corresponding antibiotics. Then, bacteria were washed with PBS buffer and the concentrations of donor and recipient strains were adjusted to OD600 = 0.70 (± 0.02) and mixed 1:1. The conjugation transfer processes were conducted with different concentrations and kinds of QACs (DTAC, DDDAC, DDBAC, dimethyl benzyl tetradecyl ammonium chloride, TDBAC; hexadecyl dimethyl benzyl ammonium chloride, HDBAC). The concentrations of QACs were 10^{-5} mg/L, 10^{-4} mg/L, 10^{-3} mg/L, 10^{-2} mg/L, 10^{-1} mg/L, 1 mg/L, 10 mg/L, and 100 mg/L, respectively. After incubating at 37 °C for 16–20 h, the number of recipients and transconjugants was counted by colony-

forming units (CFU), and the conjugative transfer rate was calculated according to the formula:

$$\text{Conjugative transfer rate} = \text{number of transconjugants (CFU/mL)} / \text{number of recipients (CFU/mL)}$$

Detection of bacterial cell membrane permeability and ROS production

Annexin V FITC Apop Dtec Kit I (BD Pharmingen™, USA) was used to detect changes in bacterial cell membrane permeability after addition of QACs. We chose DDBAC as the experimental reagent which had the most obvious promotional effect on horizontal gene transfer. The concentration of bacteria was adjusted to 10^6 CFU/mL with different concentrations of DDBAC and all solutions were incubated at 37 °C for 2 h. Then, 5 μL Annexin-FITC and 5 μL PI, respectively, were added into the bacterial solution and allowed to react in dark for 30 min. The treated bacteria were detected using the BD Biosciences FACSVerse flow cytometer (Becton Dickinson, Franklin Lakes, NJ, USA). A total of 100,000 cells were collected from each sample, and the average fluorescence intensity was obtained using 488 nm excitation wavelength and 525 nm emission wavelength. All assays were performed in triplicate.

The ROS fluorescent dye 2′,7′-dichlorofluorescin (DCFH-DA) (Sigma, USA) was used to detect intracellular ROS production. The DCFH-DA probe was diluted with ultrapure water and added to the suspension of donor and recipient bacteria, respectively, to make a final concentration of 10 μM. After 30 min of dark reaction at 37 °C, the solution was washed three times with PBS and then DDBAC was added. After 15 min of dark reaction, BD flow cytometry was used to detect the organism at 488 nm excitation wavelength and 525 nm emission wavelength.

Statistical analysis

The Pearson correlations between the abundance of ARGs and class 1 integron-integrase gene (intI1) were analyzed using SPSS v.22.0 (IBM, USA) at $p < 0.05$ level of significance. Linear regression of ARG abundance with intI1 was conducted and visualized using R3.4.2 (R Foundation for Statistical Computing) with the vegan and ggplot2 packages. Histograms and line graphs were generated by Origin 9.0 (OriginLab Corporation, Northampton, MA). A heatmap of log-transformed ARG abundances was constructed in R3.4.2 with the pheatmap package, and ARG abundances were normalized to the corresponding absolute abundance of 16S rRNA genes. The results of flow cytometry were analyzed using FlowJo software (version 10.0; Tree Star, Ashland, OR).

Results and discussion

Occurrence and distribution of resistance genes

The similar distribution of ARGs' absolute abundance at three watersheds is shown in a heatmap (Fig. 1). ARG profiles were divided into two distinct parts according to the results of cluster analysis. The qacEΔ1 was detected in all samples, ranging 10^5–10^{11} copies/L. In most samples from the three watersheds, abundances of cfr, blaTEM, dfrA1, and fexA were lower, and abundances of ermB, intI1, sul1, sul2, tetA, tetG, tetX, cml, and qacEΔ1 were higher. According to the resistance mechanisms of ARGs, all of cfr, blaTEM, and dfrA1 encoded
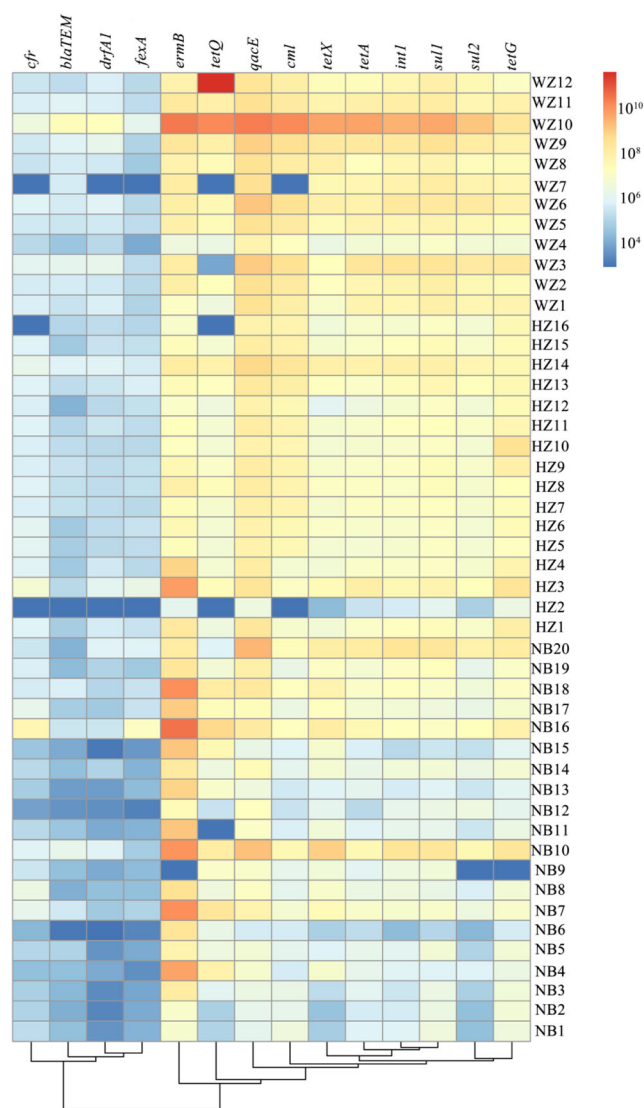

Fig. 1 Heatmap of ARG profiles showing distinct pattern between ARGs in three watersheds. The clustered ARGs are divided into two distinct clusters: cluster I, the ARGs are negatively detected in three watersheds; cluster II, the ARGs are positively detected in three watersheds

enzyme inactivation, while *sul*1, *sul*2, and *erm*B all encoded ribosome protective protein.

The Pearson correlations between genes calculated by SPSS are shown in Table 1. The correlation between *qacE*Δ1 and *intI*1 was the strongest ($r = 0.999$, $p < 0.01$); one reason is possibly due to that *qacE*Δ1 existing in integron gene cassette and co-transferred with *intI*1 (Gillings et al. 2009a, b, 2015; Kholodii et al. 1995; Pal et al. 2015). Further, both *qacE*Δ1 and *intI*1 had significant correlations with *sul*1, *su*l2, *bla*$_{TEM}$, *tet*A, *tet*X, *cml*, and *dfr*A1 ($r = 0.997$, 0.993, 0.996, 0.992, 0.988, 0.990, and 0.995, respectively, $p < 0.01$), indicating the concurrence of those environmental resistance genes in integron gene cassette and the co-selection of residual bactericides such as QAC agents in environments.

Based on the resistance mechanisms, we divided those 12 ARGs into three classes; there were efflux pump resistance-mechanism genes (*tet*A, *tet*G, *cml*), enzyme activity inhibition resistance-mechanism genes (*tet*X, *dfr*A1, *bla*$_{TEM,}$ *cfr*), and ribosomal protective protein resistance-mechanism genes (*sul*1, *sul*2, *tet*Q, *erm*B). The network of correlations between all these genes is shown in Fig. S2a, where the thickness of the line indicates the degree of correlation between each pair of genes. QAC resistance gene had the strongest correlation with some ARGs which employed the resistance mechanism of inhibiting enzyme activity, followed by ARGs with the ribosomal protection protein resistance mechanism, while the correlation was found relatively weak with the efflux pump resistance-mechanism class of genes. Furthermore, 7 of the 12 resistance genes had a strong correlation with *qacE*Δ1 and *intI*1. The other five ARGs all had no significant correlation with *qacE*Δ1 and *intI*1 ($p > 0.05$), but the result further demonstrated the strong correlation between QAC resistance gene and *intI*1.

The linear regression graphs of *qacE*Δ1 and *intI*1 are shown in Fig. 2a; the linear fitting curve formula was $y = 6.13x + 2.10 \times 10^7$, $r = 0.999$. The degree of fitting was high, consistent with the results of Pearson correlation analysis. Studies had shown that *qacE*Δ1 was located in the conserved region of the type I integron, which is a mobile recombination element capable of acquiring and expressing the ARGs (Gaze et al. 2011; Gillings et al. 2009a, b). The integron-captured antibiotic-resistant gene was located in the gene cassette, which contained genes *qacE*Δ1 and *sul*1 (Gillings et al. 2009b; Hegstad et al. 2010; Recchia and Hall 1995). The presence of QACs in an environment may cause some stress to the bacteria in that environment, which could increase the abundance of bacteria carrying *qacE*Δ1 and type I integrons, resulting in the capture and co-transfer of additional ARGs. The linear regression graphs of *qacE*Δ1 and other ARGs (*sul*1, *sul*2, *bla*$_{TEM}$, *tet*A, *tet*X, *cml*, *dfr*A1) are shown in Fig. 2b–d, respectively. The high degree of model fit confirmed the results of Pearson correlation analyses and potentially suggested the environmental co-selection of *qacE*Δ1 and ARGs in watersheds.

Generation of antibiotic resistance under QAC exposure

In order to further verify whether the presence of QACs imposed stress on bacteria and improved the resistance of bacteria to antibiotics, we continuously cultured bacteria in media with QACs and antibiotics, respectively. The results are shown in Fig. 3a–c. Among the three selected QACs (DTAC, DDBAC, DDDAC), the bacteria cultured in DTAC generated the highest resistance to the three antibiotics, including Cip (Fig. 3a), Chl (Fig. 3b), and Tc (Fig. 3c), reducing the bacteriostatic circle diameter by an average of 23.6%. The more bacteriostatic circle diameter reduced, the more antibiotic resistance they had. The bacteriostatic circle diameter reduction rate of bacteria cultured in DDDAC was quite close to that of DTAC, with the average reduction of 23.5%, while the reduction rate of bacteria cultured in DDBAC was 20.3%. With the increased concentration of QACs, bacteria generated robust resistance to QACs and antibiotics. When QAC environmental pressures were withdrawn, over the subsequent 30 days, the bacteria's antibiotic resistance generally decreased and gradually returned to its initial level, but some of them showed stronger resistance than before (Fig. 3). These results further demonstrated that QACs can increase the resistance of bacteria to antibiotics and lead to multi-drug resistance. Under tetracycline exposure (Fig. 3b), the decrease in resistance to chloramphenicol may be related to the metabolism compensatory of certain resistance genes or drug resistance mechanisms. Sometimes the expression of the resistance gene may produce a certain toxic effect or energy consumption to the bacteria, so the bacteria under the environmental pressure may prefer to express the specific resistance gene to relevant environmental stress and would weaken or discard other unrelated resistance genes, in which case the resistance of bacteria to certain antibiotics

Table 1 Person correlation between qacE△1, intI1, and ARGs

	qacEΔ1	IntI1	sul1	sul2	tetX	tetA	cml	drfA1	blaTEM	ermB	tetQ	tetG	cfr	fexA
qacEΔ1	1	0.999**	0.997**	0.993**	0.988**	0.992**	0.990**	0.995**	0.996**	0.218	0.016	0.281	0.019	0.048
IntI1	0.999**	1	0.997**	0.995**	0.986**	0.991**	0.988**	0.995**	0.997**	0.312	0.017	0.115	0.016	0.045

Fig. 2 The correlations between
$qacE\Delta1$ and $int1I$ (**a**), $sul1$ (**b**),
$sul2$ (**c**), cml (**c**), $tetA$ (**c**), bla_{TEM}
(**d**), $drfA1$ (**d**), and $tetX$ (**d**). The
shadow indicates the 95%
confidence intervals
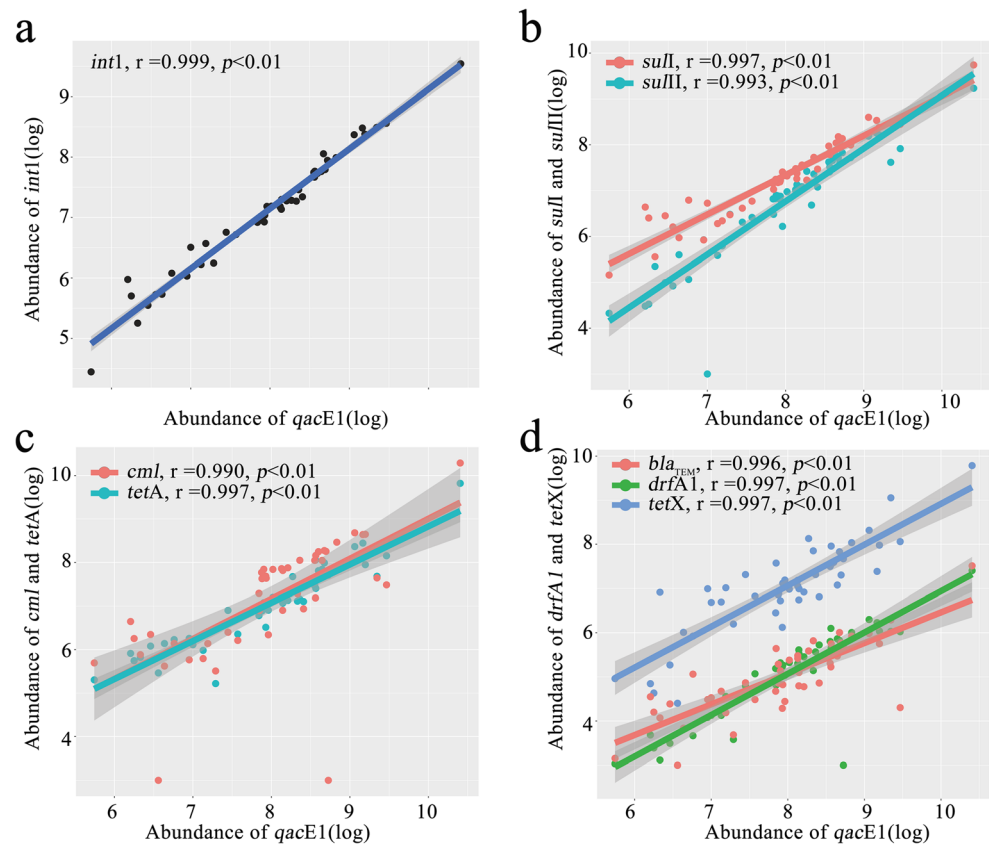
would be reduced. QACs and antibiotics are widely present in environments, and antibiotic concentrations were found lower than those of QACs in water environments (Li and Brownawell 2010). Previous studies have demonstrated the development of high-level resistance during antibiotic exposure (Wistrand-Yuen et al. 2018; Levin-Reisman et al. 2017). Our results showed that the presence of QACs can lead to co-resistance to QACs and antibiotics, indicating that QACs in the environment may increase the resistance of bacteria to antibiotics even in the absence of antibiotics. QACs can also persist in environments (Li and Brownawell 2010), which may greatly promote the circulation and emergence of ARGs in the environment.

Effects of QAC on conjugational transfer

Five kinds of QACs promoted the transconjugative efficiency of the bacteria (Fig. 3d); the transconjugative efficiencies were between 1.33×10^{-6} and 8.87×10^{-5}, and the highest transconjugative efficiency was 15 times higher than that of the blank control group. With the increase of QAC concentration, the transconjugative efficiency increased initially and then decreased, and DDBAC, TDBAC, and HDBAC reached the highest

transconjugative efficiency at 10^{-1} mg/L, while DTAC and DDDAC peaked at 10^{-2} mg/L. When the concentration of QAC was 10 mg/L, the number of recipient bacteria significantly decreased and there were few transconjugant bacteria. Continually adding to 100 mg/L, few bacteria remained, indicating that concentrations of QAC higher than 100 mg/L had a lethal effect on experimental bacteria. The effect of DDBAC on the transconjugative efficiency was the most obvious among the five QACs, with the complete ranking being DDBAC > TDBAC > DTAC > DDDAC > HDBAC. The results showed that all five QAC substances could promote the transfer of RP4-resistant plasmids in *E. coli*, but their promoting efficiencies were various, which may be related to the structure and toxicity of organic matter. The concentration of QACs which optimally promoted transconjugative efficiency was about 10^{-1}–10^{-2} mg/L, while the average concentrations of QACs in domestic wastewater, treated water, and surface water were 0.5 mg/L, 0.05 mg/L, and 0.04 mg/L, respectively (Li and Brownawell 2010), suggesting environmental residual QACs could promote antibiotic resistance gene transfer by plasmids and exacerbate the problem of environmental AMR, which should attach more attention. Our results provide empirical genetic evidences that

Fig. 3 The evolution of bacteria resistant to antibiotics (CIP (**a**), CHL (**b**), and Tc (**c**)) under DTAC, DDDAC, DDBAC, CIP, and Tc selection pressures and the changes of transconjugative efficiency in bacteria under five QACs selection pressures (**d**) (dodecyl trimethyl ammonium chloride, DTAC; didodecyl dimethyl ammonium chloride, DDDAC; dodecyl dimethyl benzyl ammonium chloride, DDBAC; dimethyl benzyl tetradecyl ammonium chloride, TDBAC; hexadecyl dimethyl benzyl ammonium chloride, HDBAC)
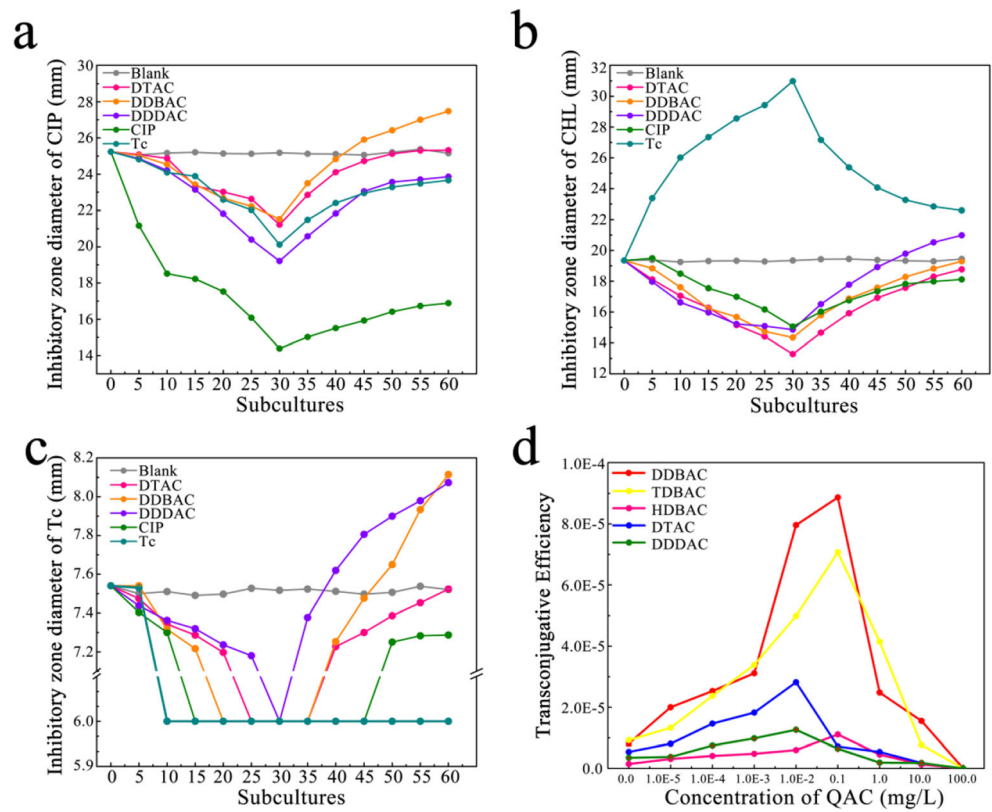
Mechanisms of QAC promote the conjugational transfer

The cell membranes of the donor (Fig. 4a–c) and the recipient bacteria (Fig. 4d–f) both showed obvious changes upon addition of QAC. Compared with the blank control group (Fig. 4a, d), the bacterial cell membrane permeability increased upon addition of 10^{-1} mg/L of QAC. QAC exposure damaged donor (Fig. 4b) and recipient cell membranes (Fig. 4e) to some extent and resulted in bacterial apoptosis. With additional QAC, the phenomenon of bacterial apoptosis became more obvious. The proportion of bacteria (Fig. 4c) with cell membrane damage increased from 2.49% to 95% in case of donor bacteria and from 3.81% to 97.52% of recipient bacteria. QAC can stimulate bacteria to produce ROS and change the bacterial cell membrane permeability. ROS could, therefore, induce the horizontal transfer of transmissible plasmids and integrons that contain both disinfectant and antibiotic resistance genes (Bouzada et al. 2010; Chapman 2003; Li et al. 2016). Compared with the blank control group (Fig. 5a, d), QAC stimulated bacteria to produce a large amount of ROS; the amount of ROS produced by donor bacteria (Fig. 5b, c) increased from 0.31% to 16.72%, and the amount of ROS produced by recipient bacteria (Fig. 5e, f)

increased from 0.01% to 6.23%. The variation of ROS levels in bacteria may lead the potential change of conjugational transfer. ROS could also promote DNA damage, activate error-prone reactive oxygen free radicals to inhibit the SOS response, or cause mutations which might affect multiple drug efflux pump genes, resulting in bacteria with co-resistance to QACs and antibiotics (Brazas and Hancock 2005).

The main mechanisms of QAC on bacterial cells included interfering with the lipid bilayer of bacterial cytoplasmic membrane and the outer membrane of gram-negative bacteria, leading to loss of cytoplasmic components and cell lysis (Gilbert and Moore 2005; Heerklotz 2008). Although the primary disinfection mechanism of QAC was lysing bacterial cell, it only played a role when the concentration was equal to or higher than critical micelle concentration (CMC) (Gilbert and Moore 2005; Maillard 2002; Mcdonnell and Russell 1999). When the bacteria were exposed to the sub-inhibitory concentration of QAC, they would reduce the cell protein number of surface, and change the structure of the pore protein and the cell surface composition and polarity, causing the bacterial cell membrane permeability and mobility to be changed accordingly (Loughlin et al. 2002; Nagai et al. 2003; Tezel et al. 2007; Jansen et al. 2013). The conjugation transfer was the process which the donor bacteria were ligated with the recipient bacteria to form a channel through which the DNA of the donor was transferred into the acceptor bacteria (Chen et al. 2005; Waters 2001). The permeability of the
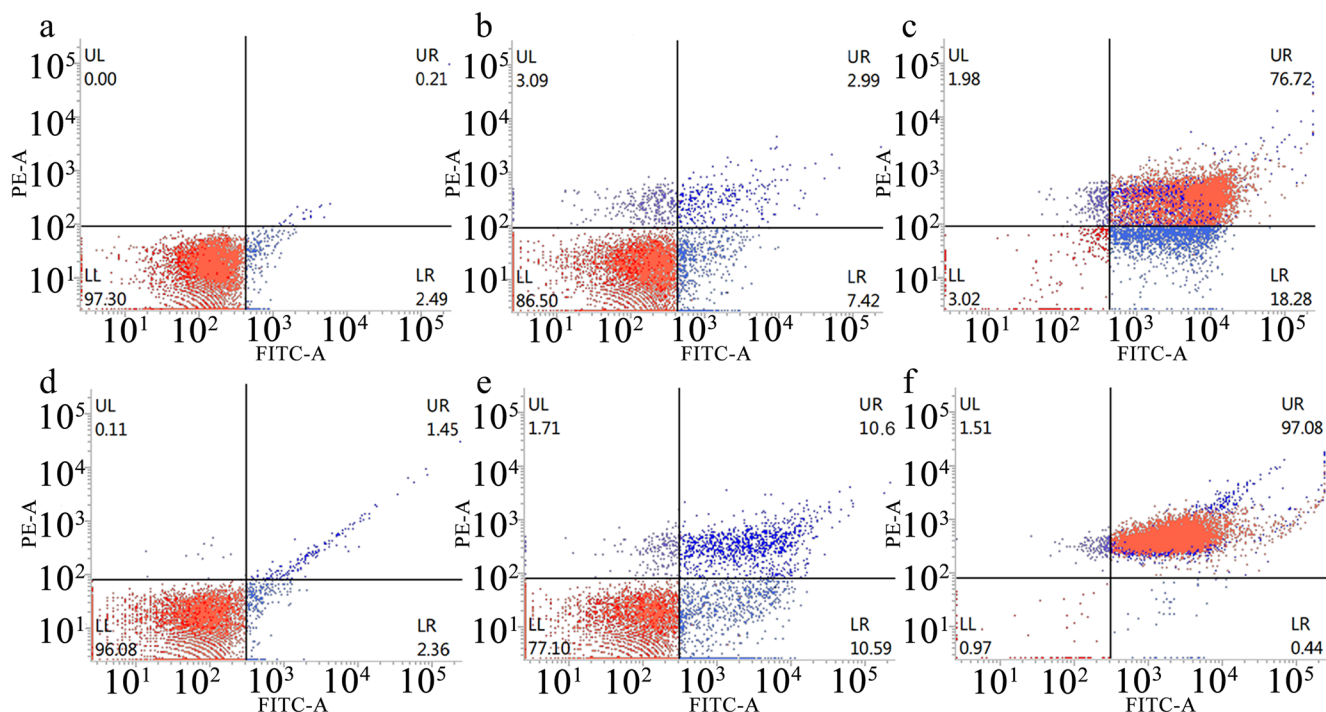
Fig. 4 The changes of donor (**a**, **b**, **c**) and recipient (**d**, **e**, **f**) cell membrane permeability under the influence of QAC. **a**, **d**, Blank control group. **b**, **e**, The addition of QAC concentration of 10^{-1} mg/L. **c**, **f**, The addition of QAC concentration of 100 mg/L. Quadrant LL: negative signal (normal cells). Quadrant LR: PI-positive cells (with increased cell membrane permeability). Quadrant UR: dead cells
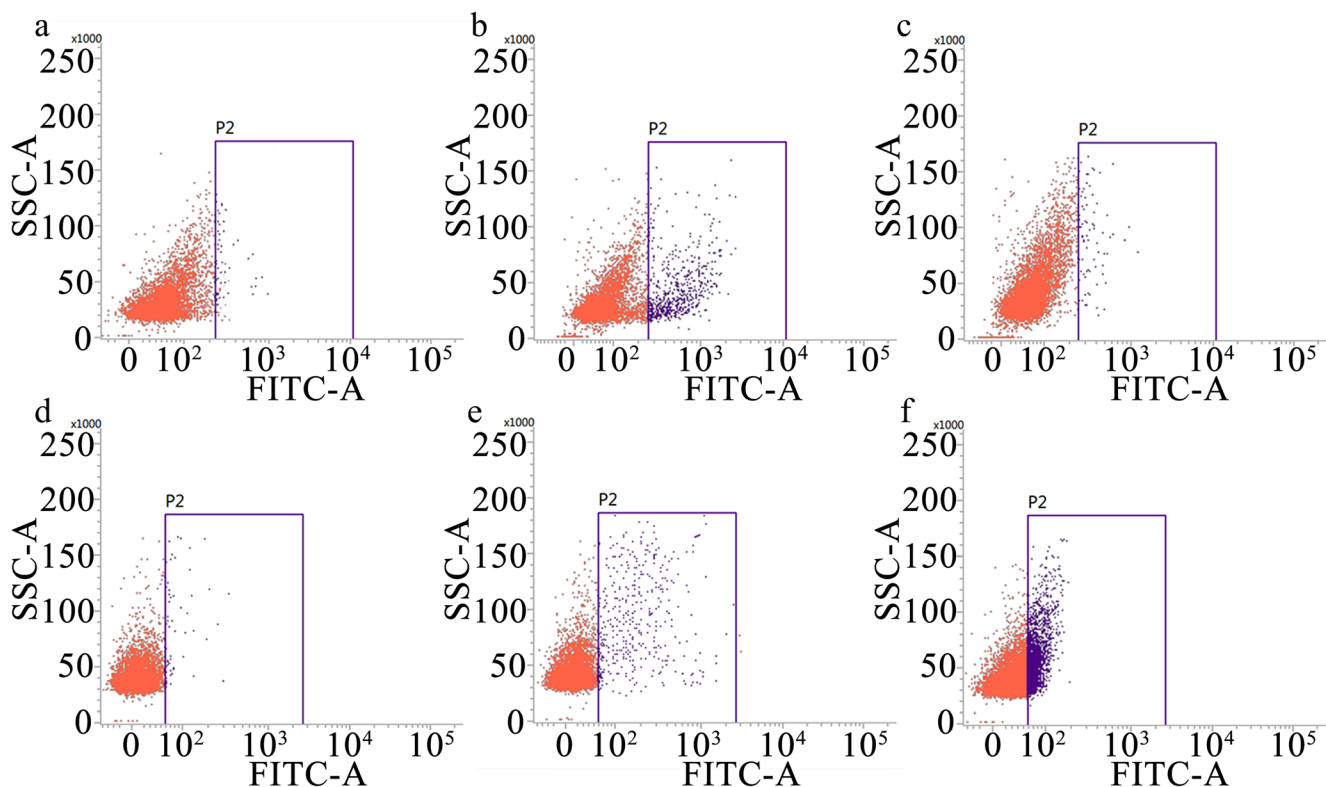

Fig. 5 The changes of ROS production of donor (**a**, **b**, **c**) and recipient (**d**, **e**, **f**) cells under the influence of QAC. **a**, **d** Blank control group. **b**, **e** The addition of QAC concentration of 10^{-1} mg/L. **c**, **f**, The addition of QAC concentration of 100 mg/L. P2 areas: DCFH-DA-positive cells (with increased ROS production). Outside of the P2 areas: negative signal (normal cells)

bacterial cell membrane was the main influencing factor in the conjugation transfer of the resistant plasmid (Lopatkin et al. 2016; Zhang et al. 2017). Therefore, QAC enhanced the transfer of drug-resistant plasmid between bacteria by promoting the intercellular association of bacteria and permeability of bacterial cell membrane (Tabata et al. 2003).

Conclusions

QACs could promote the evolution of bacterial resistance to multiple antibiotics. In addition, different QAC concentrations could affect the conjugation transfer efficiency. Five QACs were found to promote the transfer of RP4 resistant plasmids in *E. coli* at a low concentration. The optimal concentrations of QACs enhancing transconjugative efficiency were similar to environmental concentrations, suggesting that residual QACs in the environment can exacerbate the problem of environmental resistance. Copious amounts of bactericides are currently being used in surface active agent, emulsifier, fabric softener, disinfectant, insecticide, preservative, and personal care products. This study emphasizes the risk of QACs and appeals to the community to pay more attention to the influence of QACs on ARGs transfer. The environmental residues of these compounds, as well as factors that promote or restrict AMR generation and exchange between environmental microbiota, human commensals, and particularly pathogens, are worthy of further investigation.

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