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Ciprofloxacin dosage and emergence of resistance in human commensal bacteria

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Abstract

Background

Although optimization of fluoroquinolone dosage increases the efficacy of this class of drugs against bacterial infections, its impact on the emergence of resistance in commensal bacteria is unknown.

Methods

Six different dosing regimens of oral ciprofloxacin for 14 days were randomly assigned to 48 healthy volunteers. Individual pharmacokinetic and pharmacodynamic parameters combining antibiotic exposure in plasma, saliva and stool and MIC and MPC of ciprofloxacin against viridans group streptococci in the pharyngeal flora and Escherichia coli in the fecal flora were estimated. Their links with the emergence of resistance to nalidixic acid or ciprofloxacin in the fecal flora and to levofloxacin in the pharyngeal flora, at day 7, 14 or 42, were investigated.

Results

Resistance emerged in 25% and 33% of the subjects in the fecal flora and the pharyngeal flora, respectively, mainly when local concentrations of ciprofloxacin were below the MIC. No variable integrating pharmacokinetic data and pharmacodynamic parameters were found to differ significantly between the subjects in whom resistance emerged or not. Probabilities of emergence of resistance were not significantly different whatever the antibiotic exposure.

Conclusions

Selection of resistant commensals during ciprofloxacin therapy is a frequent ecological side effect which is not preventable by optimizing dosing regimen.

MESH Keywords Adult ; Anti-Bacterial Agents ; administration & dosage ; pharmacokinetics ; pharmacology ; Ciprofloxacin ; administration & dosage ; pharmacokinetics ; pharmacology ; Drug Resistance, Bacterial ; Escherichia coli ; drug effects ; Feces ; microbiology ; Female ; Humans ; Male ; Microbial Sensitivity Tests ; Pharynx ; microbiology ; Saliva ; metabolism ; Viridans Streptococci ; drug effects ; Young Adult

Bacterial resistance to antibiotics is a growing therapeutic problem, both in the community and the hospital, involving all antibiotics including fluoroquinolones [1]. Decreased susceptibility to fluoroquinolones arises mainly by single-step mutations in the gyrA and parC genes, which encode the fluoroquinolones targets, the topoisomerase enzymes [2], conferring cross-resistance to all fluoroquinolones [3]. Accumulation of multiple mutations in several genes confers increasing level of resistance associated with clinical failure [4 , 5]. However, even low-level resistance can generate therapeutic failure [6].

Resistance to fluoroquinolones can result from direct selection at the site of infection or from selection in commensal bacteria followed by horizontal gene transfer to pathogens [7], which might even be more frequent because there are many more bacteria among commensal flora than within any infectious focus [8]. In addition, differences in local antibiotic concentrations, as compared with plasma, can affect selection of resistant bacteria in different sites [9 , 10]. Also, resistant commensal bacteria may be selected after any antibiotic treatment, whereas resistant pathogens can emerge only in actually infected patients [11].

Pharmacokinetic and pharmacodynamic parameters give rationale to antibacterial dosing [12 , 13]. The minimal inhibitory concentration (MIC) and more recently the mutant prevention concentration (MPC) are parameters used to investigate the relationship between antibiotic exposure and efficacy or prevention of resistance development [14]. The ratio of the area under the concentration-time curve (AUC) to the MIC and the ratio of the peak concentration to the MIC for the infecting bacteria have been linked to treatment success with fluoroquinolones [15 –18], but not to emergence of resistant pathogens, probably because the inocula at the foci of infection are limited and emergence of resistance is uncommon [15 ,19]. To investigate whether optimization of dosing regimens might prevent the emergence of resistance in commensal flora, we studied the relationship between antibiotic pharmacokinetic and pharmacodynamic parameters in plasma, saliva and feces and the emergence of resistant strains in the pharyngeal and fecal flora in healthy volunteers receiving variable dosing regimens of ciprofloxacin, the reference fluoroquinolone.

Methods

Subjects

Eighteen to 45 year-old healthy volunteers were selected on the basis of normal findings in a thorough general examination (interview and physical examination), normal intestinal transit (one formed stool a day), normal ECG findings with a QTC < 450 ms, and normal results in a biological work-up (blood count, blood biochemistry, liver tests, urinalysis, serological tests for hepatitis viruses and HIV) [20] . Women of child-bering age were included if they were using effective contraception and had a negative pre-inclusion pregnancy test. The volunteers had no known allergy to a fluoroquinolone, had taken no antibacterial or antifungal drug, theophylline, steroids, vitamin K antagonists or barrier antacids during the three months before inclusion. Subjects unlikely to adhere to the study protocol, who had given blood (> 500 ml) or who had participated in another study within the previous three months were not included. Caffeine consumption was limited and stable during the two-week treatment period. Volunteers were advised to avoid exposure to sunlight throughout the treatment period. The study design was approved by the local ethics committee of Paris Bichat-Cergy Pontoise. All the participants gave their written informed consent before entering the study.

Regimens

Volunteers were randomly assigned into 6 groups of 8 individuals each, receiving either 250 mg every 12h, 500 mg every 24 h, 500 mg every 12 h, 750 mg every 24 h, 750 mg every 12h, or 1000 mg every 24 h of oral ciprofloxacin for a total of 14 days. These regimens were chosen in order to generate pharmacokinetic variability within range of clinically relevant total daily doses. Each intake was observed, and its time recorded.

Outcome and follow-up

Microbiological study

Samples

Fecal and pharyngeal samples were collected before initiation of treatment and on days 7, 14 and 42, stored at −80°C and blinded until analysis [20]. Analysis focused on viridans group streptococci (VGS) in the pharyngeal flora and Escherichia coli in the fecal flora for several reasons: these bacterial species are present in all subjects, they are involved in various clinical infections (bacteremia, endocarditis, urinary tract infections), and are recognized sources of horizontal gene transfer within the commensal flora. We determined for each of these target species the susceptibility to quinolones of the global population (dominant flora) and we also detected the emergence of quinolone-resistant subpopulations (subdominant flora).

Suceptibility to ciprofloxacin of the dominant flora

We used a procedure specifically designed to estimate susceptibility to fluoroquinolones of the dominant flora as a whole. Saliva samples were inoculated on colimycin-nalidixic acid (CNA) blood agar (Biomérieux, Craponne, France) and fecal samples on Drigalski agar. After growth, we isolated ten separate colonies from each plate, identified as VGS and E. coli using standard techniques, in order to obtain a representative sample of the dominant flora. These ten colonies were mixed, and susceptibility to fluoroquinolones of the mixture was tested using minimum inhibitory concentration (MIC) in duplicate by the agar dilution method [21] and mutant prevention concentration (MPC) in triplicate [14 , 22], as described. Geometric means of these replicates were used in the analysis.

Detection of resistance in the subdominant flora

Resistance in the subdominant flora of each target bacterial species was detected by plating the fecal samples on Drigalski agar with 16 mg/L of nalidixic acid to detect first step mutant in E. coli , or containing 1 mg/L of ciprofloxacin to detect mutants resistant to ciprofloxacin. Pharyngeal samples were plated on CNA blood agar with 2 mg/L of levofloxacin because VGS are naturally resistant to ciprofloxacin but susceptible to levofloxacin [23]. MICs of the colonies growing on selective media were determined by the agar dilution method [21].

Endpoints

Resistance to nalidixic acid and ciprofloxacin in E. coli from the fecal flora and resistance to levofloxacin in VGS from the pharyngeal flora were determined according to CLSI breakpoints [24]. Emergence of resistance was defined by the detection of resistant strains at day 7, 14 or 42 in subjects in whom only susceptible strains were detected and resistant strains were not detected before treatment.

Pharmacokinetic study

Serum and saliva samples were taken from each volunteer before and 1, 3, 6 and 12 hours after the first ciprofloxacin intake, at trough on day 8 and day 14, and again 1, 3, 6 and 12 hours after the last intake [10]. Stool samples were collected on days 0, 7, 14 and 42. All samples were blinded and stored at −80°C until analysis. Ciprofloxacin concentrations were determined by liquid chromatography with fluorimetric detection after deproteinization or stool extraction in acidic medium, as described [25].

Statistical analysis

A population pharmacokinetic analysis with, as previously described [26 –28] a one compartment model with first order absorption was used to analyze plasma and saliva concentrations and estimate the maximal concentration (peak) and AUC from 0 to 24 h at steady-state for each volunteer, taking into account the dosing schedule. AUC/MIC, AUC/MPC, peak/MIC, peak/MPC, AUC above MIC, AUC above MPC, AUC between MIC and MPC, time above MIC, time above MPC, and time between MIC and MPC were determined using individual MIC and MPC of ciprofloxacin. In feces, concentration/MIC and concentration/MPC ratios were determined using the average of the concentrations measured at days 7 and 14.

Confidence intervals for proportion of emergence of resistance were estimated using the binomial distribution. For each target flora, the volunteers were divided into two groups (regardless of the dosing schedule) according to the emergence or not of resistance. Variables integrating pharmacokinetic and pharmacodynamic parameters were compared between the two groups using the Wilcoxon-Mann and Whitney non parametric test. Logistic regression analysis was also performed in each flora to model the link between the probability of emergence of resistance and the logarithm of AUC/MIC.

The number of subjects were determined expecting emergence of quinolone resistance in the fecal flora in one third of the volunteers [29 –31]. With this proportion and the mean AUC/MIC ratios reported previously to be significantly associated with respect to emergence of resistance at the site of infection [32], the power of this study was 90% with a type I error of 5%.

Results

Subjects

Eighty subjects were screened and 48 subjects (28 males and 20 females) who fulfilled the predefined criteria for healthy subjects were selected. Median age was 28.9 y [range 19.5–43.9], and median weight was 65 kg [47–93] (71 kg [53–93] in males and 55 kg [47–85] in females). One subject who developed tendinitis at day 8 while receiving the 750 mg bid regimen and stopped therapy was excluded from the microbiological and pharmacokinetic follow-up analysis. The remaining 47 subjects had no treatment related side effects.

Microbiology study (Figure 1)

Before treatment, median [range] MIC and MPC of ciprofloxacin against the dominant flora were 0.016 mg/L [0.005–0.5] and 0.25 mg/L [0.06–2.8] in the fecal flora and 2 mg/L [0.71–45.25] and 22.6 mg/L [8–512] in the pharyngeal flora, respectively. In the fecal flora, one subject had no detectable E. coli while six (13%) initially had strains resistant to nalidixic acid, including one with resistance to both ciprofloxacin (MIC = 32 mg/L) and nalidixic acid (MIC > 1024 mg/L) and five with resistance to nalidixic acid only (MICs: 64 to > 1024 mg/L and MICs to ciprofloxacin: 0.01 to 0.5 mg/L). All the strains resistant to ciprofloxacin were also resistant to nalidixic acid. Before treatment, in the pharyngeal flora, one subject had VGS strains resistant to levofloxacin (MIC = 16 mg/L) and for one subject there was no pharyngeal sample.

During treatment, susceptible E. coli were not detected in the fecal flora from any volunteers. Resistance to ciprofloxacin was detected in three subjects (6%) at day 7 and 14. By contrast, at day 42, 14 subjects (30%) were colonized by strains resistant to nalidixic acid (MICs: 64 to >1024 mg/L), including four with resistance to ciprofloxacin (MICs: 32–64 mg/L). Overall, resistance to nalidixic acid or ciprofloxacin that was not initially detected before therapy emerged in 10/40 subjects (25%, 95%CI: 13–40%), during or after therapy.

In the pharyngeal flora, VGS resistant to levofloxacin were detected in eight subjects (17%) at day 7, ten (21%) at day 14, and four (10 %) at day 42. MICs of levofloxacin against the resistant strains ranged between 4 and 64 mg/L. Overall, resistance to levofloxacin that was not initially detected before therapy emerged in 15/45 volunteers (33%, 95%CI: 20–49%), during or after ciprofloxacin therapy.

Antibiotic concentrations and pharmacokinetic studies (Figure 2)

Median peak concentrations of ciprofloxacin in plasma at steady state ranged from 1.35 mg/L [range: 1.13–1.77] for the 250 mg twice daily regimen to 4.26 mg/L [2.89–5.66] for the 1000 mg once daily regimen; median AUC ranged from 11.77 mg/L.h [11.56–12.08] for the 250 mg bid regimen to 36.27 mg/L.h [33.23–45.49] for the 750 mg bid one. Thus, median concentrations of ciprofloxacin in plasma were above median MIC against the fecal dominant flora during most of the time for all regimens, and above median MPC for all twice daily regimens. For the pharyngeal flora, median ciprofloxacin concentrations in plasma were far below median MPC and just above median MIC at peak, except for the 250 mg twice daily regimen.

Median concentrations of ciprofloxacin in stools at steady state ranged from 845 mg/L [455–1265] for the 250 mg twice daily regimen to 1938 mg/L [1100–3235] for the 500 mg twice daily regimen, far above the median MIC and MPC of ciprofloxacin against the fecal dominant flora. Ciprofloxacin concentrations were undetectable in stools at day 42 in all subjects.

Median peak concentrations of ciprofloxacin in saliva at steady state ranged from 0.48 mg/L [0.32–0.84] for the 250 mg twice daily regimen to 1.79 mg/L [1.05–2.35] for the 1000 mg once daily one, never reaching the median MIC level of ciprofloxacin against the pharyngeal dominant flora, and far below its median MPC. Ciprofloxacin concentrations were undetectable in saliva at day 42 in all subjects.

Relationship between antibiotic exposure and emergence of bacterial resistance

Distribution of AUC/MIC of ciprofloxacin in plasma, as well as ciprofloxacin concentrations/MIC ratios in feces, were not significantly different in subjects in whom resistance to nalidixic acid or ciprofloxacin emerged or not in fecal flora (Table 1 and Figure 3). Other variables integrating plasma pharmacokinetic and pharmacodynamic parameters of ciprofloxacin failed to show any significant difference (Table 2). Logistic regression analysis did not evidence any significant link between probability of emergence of resistance in fecal flora and antibiotic exposure as measured by AUC/MIC for ciprofloxacin in plasma or by ciprofloxacin concentrations in feces/MIC (Table 1).

Similar results were obtained in pharyngeal flora. Distribution of plasma AUC/MIC of ciprofloxacin in plasma or saliva did not significantly differ in subjects in whom resistance to levofloxacin emerged or not in the pharyngeal flora (Table 1 and Figure 3), nor were any other variables integrating pharmacokinetic data in plasma or saliva and pharmacodynamic parameters of ciprofloxacin (Table 3). Probability of emergence of resistance in pharyngeal flora was not significantly linked to antibiotic exposure measured as AUC/MIC ratio of ciprofloxacin in plasma or in saliva (Table 1).

Discussion

We found a high rate of emergence of resistance to quinolones in commensal flora from healthy volunteers during of after a 14-day course of oral ciprofloxacin, both in fecal E. coli and pharyngeal VGS. Because this was observed in healthy volunteers, without recent antibiotic exposure, these rates are probably the minimal to be expected in patients. Indeed, higher rates of ciprofloxacin resistance, from 32 to 40%, have already been reported in fecal Enterobacteriaceae in cancer or leukemia patients exposed to fluoroquinolones [28 –29]. This can be explained by two reasons: i) antibiotic exposure of the commensal flora is more frequent in patients than in healthy volunteers and may accumulate resistance over time; ii) interpatient pharmacokinetic variability is expected to be greater in heavier, sicker and older patients from the medical wards than observed in healthy volunteers.

Emergence of resistance was mainly observed for fecal E. coli against nalidixic acid and for pharyngeal VGS against levofloxacin. Few patients developed E. coli resistance to ciprofloxacin in the fecal flora, and our study was underpowered to specifically examine differences in ciprofloxacin resistance.

Although not specifically designated to analyze time-effect relationship, our study showed that prevalences of resistance were not different between day 7 and day 14, suggesting that impact on commensal flora was already achieved at day 7 (Figure 1).

Different kinetic patterns of emergence of the resistant bacteria were observed in the two commensal flora. Resistant E. coli strains were detected mainly 4 weeks after the end of therapy in the fecal flora while resistant VGS were primarily selected during the two weeks of therapy in the pharyngeal flora (Figure 1). Pharmacokinetic and pharmacodynamic parameters could account for these differences. In the feces, ciprofloxacin concentrations were very high, indeed several thousand times greater than the initial MPC on the dominant flora, explaining that E. coli virtually disappeared from the stool during therapy and that selection of resistance was unlikely. Such disappearance has previously been reported with different quinolones [33]. It has also been shown that ciprofloxacin persists in the feces of volunteers for several days after oral treatment ends [34]. Therefore, resistance was probably selected in the fecal flora when ciprofloxacin concentrations decreased below the MPC and the MIC [14 , 19]. This occurred between day 14 and day 42, when no ciprofloxacin could be detected in the feces anymore. In contrast, pharyngeal concentrations of ciprofloxacin during therapy were close to - but below - initial MIC level against VGS during the entire therapy, whatever the dosing regimens, explaining why resistance could be selected during treatment, and why it then decreased as antibiotic selective pressure vanished (Figure 1). Indeed, subinhibitory concentrations of

fluoroquinolones favor selection of resistance and induce genetic transformability increasing the rate of mutation and genetic exchange in response to antibiotics [19 , 35].

Depending on the target flora, we based the detection of quinolone resistance on different surrogate markers, according to their clinical relevance. Resistance to nalidixic acid, a first generation quinolone that can be prescribed for the treatment of uncomplicated urinary tract infections, was used in E. coli since it is indicative of a single first step mutation in the target gene gyrA and associated with reduced activity of fluoroquinolones [29 –31]. Resistance to levofloxacin was used to detect emergence of resistance in pharyngeal VGS since VGS are naturally resistant to ciprofloxacin and therefore no MIC breakpoints exist. In addition, levofloxacin is widely recommended for the treatment of respiratory infections [23]. Emergence of resistance to levofloxacin in pharyngeal VGS obviates the selection of cross-resistance among fluorquinolones, as shown for Streptococcus pneumoniae [3].

A major issue in the use of fluoroquinolone is the role of dosing regimen to optimize efficacy. We did not identify any significant difference in antibiotic exposure in plasma, saliva and stool between subjects in whom resistance was selected or not. Similarly, the probability of emergence of resistance was comparable whatever the ciprofloxacin exposure, expressed as the ratio of the AUC to the MIC against the dominant flora. This was observed despite the fact that large ranges of ciprofloxacin dosing regimens were investigated, from the minimum to the maximum total daily dose that can be administered therapeutically. Lack of power is here unlikely and duration of treatment was sufficient to allow the selection of resistant mutants. Therefore, these results indicate that, whereas optimizing dosing regimen of ciprofloxacin is useful to increase efficacy at the focus of infection [15 –18], it is not likely to be helpful to decrease the risk for selection of resistant strains in commensal flora. This may be explained by the different pharmacokinetic patterns of ciprofloxacin in saliva and feces as compared with the one in plasma and by the fact that variations in dosing regimens had little impact on local ciprofloxacin concentrations in saliva and in feces as compared with the respective MIC and MPC of ciprofloxacin against the dominant flora at these sites (Figure 2). Indeed, for all ciprofloxacin regimen tested, concentrations of ciprofloxacin were always far above the MPC for Enterobactericeae in feces and below the MIC for VGS in saliva (Figure 2). Overall, the comparable rates of emergence of resistance in the fecal and pharyngeal flora whatever the antibiotic exposure suggest a random phenomenon occuring at subinhibitory concentrations (Table 1). Thus, selection of resistant commensals during ciprofloxacin therapy should be considered as an ecological side effect which is not preventable by optimizing antibiotic dosing. Other strategies to prevent such an event are warranted.

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Footnotes:

For all authors: no conflict of interest.

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Potential conflicts of interest. All authors: no conflicts.

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Figure 1

Number of subjects harbouring strains of Escherichia coli in fecal flora that were resistant to nalidixic acid or ciprofloxacin (top left) or strains of viridans group streptococci in pharyngeal flora that were resistant to levofloxacin (bottom left) and their corresponding MIC values (mg/L) for nalidixic acid (Nal, white circles) and ciprofloxacin (Cip, black circles) (top right) and levofloxacin (bottom right) in healthy subjects receiving various dosing regimens of ciprofloxacin from day 1 to day 14. Strains that were resistant to ciprofloxacin also appear among strains resistant to nalidixic acid. There were 48 subjects at the start of therapy and 47 subjects later because of one subject discontinuing therapy. In addition, there was no pharyngeal sample to perform measurement at day 0 for one patient.

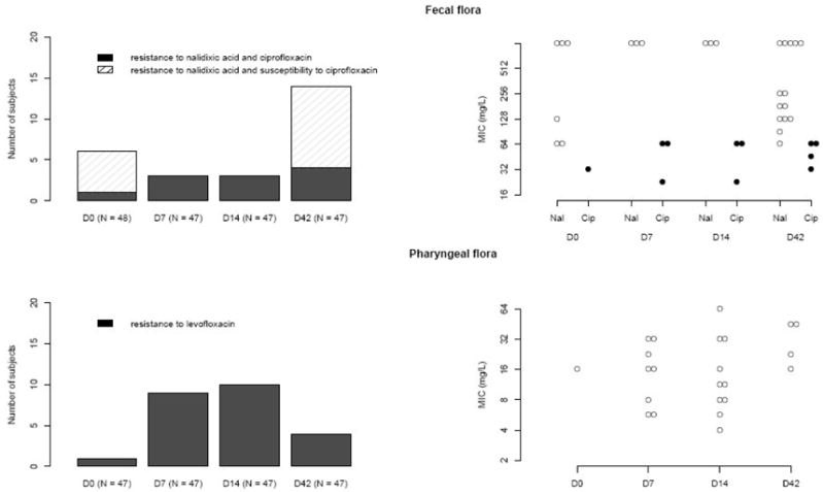

Figure 2

Mean concentration profiles of ciprofloxacin at steady state in plasma and saliva from healthy subjects receiving ciprofloxacin from day 1 to day 14, according to dosing regimens, and boxplot of the distribution of the fecal concentration at day 7 and day 14. Horizontal lines represent median MIC (mg/L) (full line) and MPC (mg/L) (dotted line) of ciprofloxacin against the dominant flora in each commensal flora.

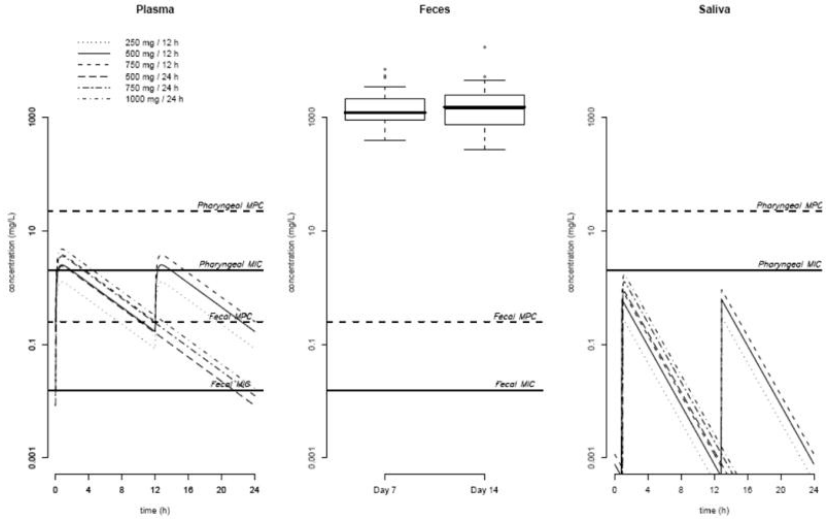

Figure 3

Distribution of the area under the ciprofloxacin concentration-time curve from 0 to 24 h (AUC)/MIC ratio in plasma (top and bottom left) and saliva (bottom right) and of the fecal concentrations of ciprofloxacin/MIC ratio (top right) in healthy subjects receiving various regimens of ciprofloxacin from day 1 to day 14 and in whom no resistant strains were detected at day 0. Results are presented according to the emergence or not of strains of Escherichia coli resistant to nalidixic acid or ciprofloxacin in fecal flora (top, n=40) or of strains of viridans group streptococci that were resistant to levofloxacin in pharyngeal flora (bottom, n=45), from day 7 to day 42. The solid line represents the median in each group. Each circle represents a subject.

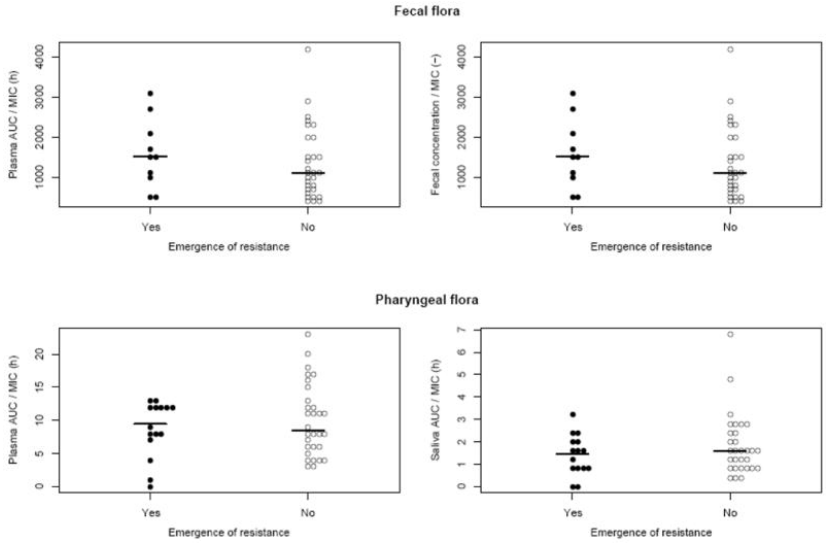

Table 1

Observed percentage of emergence of resistance in Escherichia coli from the fecal flora and in viridans group streptococci from the pharyngeal flora in healthy volunteers according to the importance of ciprofloxacin exposure expressed as quartile of the distribution. P values of the nonparametric test comparing exposure in patients with or without emergence or resistance. P values of the link between exposure and probability of emergence of resistance in logistic regression

Parameter of ciprofloxacin exposure	No of subjects at risk	Emergence of resistance		P values	
		Number	Percent	Group comparison	Logistic regression
Fecal flora					
AUC/MIC in plasma (h)				0.35	0.36
361 – 721	10	2	20%		
722 – 1112	10	2	20%		
1113 – 2004	10	3	30%		
2005 – 4236	10	3	30%		
Total	40	10	25%		
Concentration/MIC in stool				0.93	0.86
20 – 49 × 10^3	10	2	20%		
50 – 77 × 10^3	10	2	30%		
77 – 117 × 10^3	10	3	30%		
118 – 358 × 10^3	10	2	20%		
Total	40	10	25%		
Pharyngeal flora					
AUC/MIC in plasma (h)				0.86	0.27
0.37 – 5.80	11	3	27%		
5.81 – 8.54	11	4	36%		
8.55 – 12.22	11	4	36%		
12.24 – 22.79	12	4	33%		
Total	45	15	33%		
AUC/MIC in saliva (h)				0.45	0.16
0.06 –0.87	11	4	36%		
0.88 – 1.47	11	4	36%		
1.48 – 2.21	11	4	36%		
2.22 – 6.82	12	3	25%		
Total	45	15	33%		

Table 2

Comparison of variables integrating ciprofloxacin pharmacokinetic parameters in plasma or concentrations in stool and pharmacodynamic parameters of ciprofloxacin against Escherichia coli from the dominant fecal flora between groups of subjects in whom resistant Escherichia coli emerged or not from day 7 to day 42.

Variables	Emergence of resistance		P value
	Yes (n=10)	No (n=30)	
	Median (Min - Max)	Median (Min - Max)	
Plasma			
AUC/MIC (h)	1516.0 (524.8 – 3137.0)	1094.0 (360.5 – 4236.0)	0.35
AUC/MPC (h)	66.9 (22.0 – 375.9)	89.5 (32.2 – 374.2)	0.41
Peak/MIC	186.9 (61.6 – 443.2)	148.4 (29.5 – 715.0)	0.38
Peak/MPC	9.3 (3.9 – 46.3)	13.3 (3.8 – 42.2)	0.40
AUC > MIC (mg/L.h)	13.5 (11.0 – 34.3)	21.1 (10.3 – 45.2)	0.38
AUC > MPC (mg/L.h)	8.4 (5.5 – 31.1)	16.3 (5.4 – 39.6)	0.18
MIC<AUCMPC (mg/L.h)	4.2 (1.7 – 8.0)	3.2 (0.4 – 7.1)	0.57
Time > MIC (h)	24.0 (22.7 – 24.0)	24.0 (13.3 – 24.0)	0.15
Time > MPC (h)	15.0 (7.4 – 24.0)	16.0 (7.8 – 24.0)	0.64
MIC< Time <MPC (h)	8.9 (0.0 – 16.6)	7.4 (0.0 – 14.1)	0.16
Stool			
Concentration/MIC	80×10^3 (22×10^3 – 358×10^3)	75×10^3 (20×10^3 – 340×10^3)	0.93
Concentration/MPC	48×10^2 (8×10^2 – 223×10^2)	7×10^3 (1×10^3 – 46×10^3)	0.30

Table 3

Comparison of variables integrating ciprofloxacin pharmacokinetic parameters in plasma or saliva and pharmacodynamic parameters of ciprofloxacin against viridans group streptococci (VGS) from the dominant pharyngeal flora between groups of subjects in whom resistant VGS emerged or not from day 7 to day 42.

Variables	Emergence of resistance		P value
	Yes (n=15)	No (n=30)	
	Median (Min – Max)	Median (Min – Max)	
Plasma			
AUC/MIC (h)	9.47 (0.37 – 12.79)	8.47 (2.95 – 22.79)	0.86
AUC/MPC (h)	0.76 (0.10 – 2.98)	0.55 (0.10 – 2.01)	0.53
Peak/MIC	1.45 (0.04 – 3.90)	1.29 (0.34 – 4.00)	0.97
Peak/MPC	0.10 (0.01 – 0.46)	0.09 (0.01 – 0.80)	0.58
AUC > MIC (mg/L.h)	2.30 (0.00 – 8.46)	0.71 (0.00 – 12.52)	0.78
AUC > MPC (mg/L.h)	0.00 (0.00 – 0.00)	0.00 (0.00 – 0.00)	NA
MIC<AUC <MPC (mg/L.h)	2.30 (0.00 – 8.46)	0.71 (0.00 – 12.52)	0.78
Time > MIC (h)	3.96 (0.00 – 4.82)	2.22 (0.00 – 9.26)	0.72
Time > MPC (h)	0.00 (0.00 – 0.00)	0.00 (0.00 – 0.00)	NA
MIC<Time<MPC (h)	3.96 (0.00 – 4.82)	2.22 (0.00 – 9.26)	0.72
Saliva			
AUC/MIC (h)	1.46 (0.063 – 3.01)	1.57 (0.53 – 6.82)	0.45
AUC/MPC (h)	0.11 (0.01 – 0.53)	0.09 (0.01 – 0.60)	0.77
Peak/MIC	0.39 (0.02 – 1.12)	0.46 (0.14 – 1.66)	0.71
Peak/MPC	0.03 (0.00 – 0.23)	0.03 (0.00 – 0.10)	0.73
AUC > MIC (mg/L.h)	0.00 (0.00 – 0.06)	0.00 (0.00 – 0.42)	0.70
AUC > MPC (mg/L.h)	0.00 (0.00 – 0.00)	0.00 (0.00 – 0.00)	NA
MIC<AUC<MPC (mg/L.h)	0.00 (0.00 – 0.06)	0.00 (0.00 – 0.42)	0.70
Time > MIC (h)	0.00 (0.00 – 0.41)	0.00 (0.00 – 0.94)	0.68
Time > MPC (h)	0.00 (0.00 – 0.00)	0.00 (0.00 – 0.00)	NA
MIC<Time<MPC (h)	0.00 (0.00 – 0.41)	0.00 (0.00 – 0.94)	0.68