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Influence of fluoroquinolone consumption in inpatients and outpatients on ciprofloxacin-resistant *Escherichia coli* in a university hospital

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Background: The increase in fluoroquinolone-resistant *Escherichia coli* has raised the issue of treatment failure in common infections. Few studies have investigated the possible relationship between outpatient fluoroquinolone consumption and resistance in hospital.

Objective: To investigate the relationship between inpatient and outpatient fluoroquinolone use and ciprofloxacin-resistant *E. coli* in a teaching hospital.

Methods: An ecological study was conducted in Toulouse University Hospital and its surrounding area, the Midi-Pyrénées region (south-western France), in 2004–07. Dynamic regression models were built to study how the hospital resistance rate was linearly related to current and past values of fluoroquinolone consumption. Resistance forecasts for 2008 were then calculated and compared with actual rates for the first 5 months of the year.

Results: Mean resistance rate was 13.7% and mean fluoroquinolone use was 89.9 defined daily doses (DDDs)/1000 inpatient days in hospital and 2.6 DDDs/1000 inhabitants/day in the region. Taking into account past values of fluoroquinolone consumption in hospital and in outpatients, only levofloxacin use in the community remained significantly associated with resistance in hospital, with a lag of 12 months. This model explained 50% of the resistance variability.

Conclusions: This ecological analysis, conducted on a teaching hospital scale, suggests that ciprofloxacin resistance in *E. coli* in hospital is linked to consumption of fluoroquinolones within the hospital and its surrounding community. Among all fluoroquinolones, levofloxacin use was found to be the most important factor. Consumption in outpatients appears to be a relevant determinant to consider in designing interventions to reduce resistance in hospitals.

Keywords: antimicrobials, resistance, drug use, time-series analysis, dynamic regression, linear transfer function

Introduction

At the introduction of fluoroquinolones to the pharmaceutical market in the 1980s, it was believed, by some people, that fluoroquinolones were effective drugs that should not encounter bacterial resistance because of their complex mechanism of action and their entirely synthetic origin.[1,2] However, the emergence and dramatic increase in isolates of fluoroquinolone-resistant *Escherichia coli* (FQ-R *E. coli*)[3–5] have raised the issue of treatment failure in very common infections, such as urinary tract infections.

Antibiotic consumption is widely recognized as the main cause of this resistance. Several studies have focused on the relationship between fluoroquinolone use and the emergence or dissemination of FQ-R *E. coli*, but few used adequate methods.[6–8] Among them, studies adopting ecological designs are relevant to the assessment of this relationship because they allow a better understanding of bacterial ecology as a

whole. Besides geographic correlation studies,[9–14] some authors focused on correlation over time,[15–19] but few reported appropriate statistical methods. Indeed, the correlation between data collected over time must be taken into account by resorting to time-series analysis. Lately, these methods have been applied to the field of microbiology.[20–23]

Antimicrobial use in the community is also believed to be an important determinant of resistance in hospitals, although very few studies have investigated the influence of outpatient consumption of fluoroquinolones on hospital FQ-R *E. coli*.[10,24] As outpatient quinolone use keeps increasing despite a national campaign to reduce antibiotic use in France,[25] it is of special interest to focus on the relationship between outpatient use and hospital resistance.

The objectives of the present study were to investigate the temporal relationship between inpatient and outpatient fluoroquinolone use and FQ-R *E. coli* in a university hospital and to forecast resistance rates.

Methods

Design

This was an ecological study.

Setting

The study was set in the Midi-Pyrénées region (south-western France), with 2 777 000 inhabitants in 2006, essentially living in rural communities within a radius of 200 km around the main town, Toulouse (which was home to 850 900 inhabitants in 2006). Toulouse University Hospital (TUH) is the sole tertiary hospital of the Midi-Pyrénées region, with 2848 beds, including 101 intensive care beds, 120 critical care beds and 155 long-term beds. Around 90% of patients admitted to TUH in 2006 were living in the Midi-Pyrénées region.

Methods

The monthly FQ-R *E. coli* rate and fluoroquinolone use were retrospectively collected from existing sources from January 2004 to December 2007.

FQ-R E. coli

Every biological specimen that had yielded *E. coli* in TUH's bacteriology laboratory was systematically tested for ciprofloxacin susceptibility (except in children, fluoroquinolones not being indicated in paediatric patients). Ciprofloxacin susceptibility was used to determine the percentage of FQ-R *E. coli*, defined as the proportion of ciprofloxacin-resistant strains or of intermediate susceptibility to ciprofloxacin among all *E. coli* isolates tested for ciprofloxacin susceptibility every month. Duplicate isolates were deleted; only the first isolate of *E. coli* for each patient remained in the database. Susceptibility testing was performed by the disc diffusion method on Mueller–Hinton agar (BioMérieux® agar, Bio-Rad® discs) or by an automated technique (Vitek®2, BioMérieux®), according to the guidelines of the CLSI (formerly NCCLS).[26] Isolates were collected from both TUH inpatients and outpatients (emergency departments, outpatient clinics).

Fluoroquinolone consumption

Fluoroquinolone [identified by the code 'J01MA' in the anatomical, therapeutic and chemical (ATC) classification] use was expressed in defined daily doses (DDDs), according to the WHO manual, version 2007.[27] DDD is the assumed average maintenance dose per day for a drug used for its main indication in adults. It allows the standardization of the consumption of different drugs whatever their differences in the mean prescribed daily dose.[27] A DDD of 0.4 mg was assigned to lomefloxacin (as no DDD was officially assessed). Consumption of first-generation quinolones (such as nalidixic acid and pipemidic acid) was not recorded.

Inpatient fluoroquinolone use was collected from dispensing data supplied by TUH's pharmacy department (number of tablets, intravenous bottles, etc.). The data were converted into DDDs/1000 inpatient-days.

Community fluoroquinolone use was approximated by reimbursement data collected from the computerized database of the major regional healthcare scheme. This database takes an inventory of all reimbursement applications for drugs for around 65% of the Midi-Pyrénées region population (not including reimbursements for farmers, self-employed people, students, etc.). Reimbursed quantities for the whole region's population were inferred and expressed in DDDs/1000 inhabitants/day. As drug packages are usually designed to provide a treatment course for the most common infections, when calculating DDDs we assumed that the entire drug package was consumed.

Statistical analysis

We discounted pefloxacin consumption because of its very low use, and pooled consumptions of norfloxacin, enoxacin and lomefloxacin (not available in TUH) as urinary fluoroquinolone use.

Following the method developed by Box and Jenkins,[28] we generated autoregressive integrated moving average (ARIMA) models for inpatient and outpatient use of fluoroquinolones. We computed 12 month-forecasts for these series in order to introduce them as inputs in dynamic regression (DR) models or linear transfer function models. For an introduction to time-series analysis in clinical microbiology, refer to Monnet *et al.*[22]

A DR model states how an output (resistance rate) is linearly related to current and past values of one or more inputs (fluoroquinolone use). It can be written as:

$$Y_t = C + f_1(X_{1,t}) + f_2(X_{2,t}) + \cdots + f_n(X_{n,t}) + N_t$$

where Y_t is the hospital resistance rate at month t, C is a constant, f_1 *to* f_n are n linear transfer functions that can be written for each fluoroquinolone i from 1 to n as: $f_i(X_{i,t}) = \nu_{o,i}X_{i,t} + \nu_{1,i}X_{i,t-1} + \nu_{2,i}X_{i,t-2} + \cdots + \nu_{k,i}X_{i,t-k}$ and N_t is a disturbance series (what is left unexplained of Y_t after taking inputs into account); in a dynamic regression model, N_t is autocorrelated and is introduced as an ARIMA model to account for autocorrelation.

We followed the method developed by Pankratz[29] to build a DR model. As a preliminary step, we ruled out feedback from output to inputs, estimating the following model for each i of the n fluoroquinolones: $X_{i,t} = C' + b_{1,1}X_{1,t-1} + \cdots + b_{1,k}X_{1,t-k} + \cdots + b_{i,1}X_{i,t-1} + \cdots + b_{i,k}X_{i,t-k} + \cdots + b_{n,1}X_{n,t-1} + \cdots + b_{n,k}X_{n,t-k} + d_1Y_{t-1} + \cdots + d_kY_{t-k} + \varepsilon_t$. To rule out feedback, we checked that none of the d_k coefficients was statistically significant, using individual Wald t-tests and a Fisher joint test.

We then identified linear transfer functions and ARIMA disturbance series using Pankratz's identification rules. Model parameters were estimated using the method of maximum likelihood. Finally, we checked model adequacy on the residuals of N_t: no residual autocorrelation, normal distribution of residuals and residuals not greater than twice their standard deviation. We selected the best model as the one fulfilling all of the checking stages and with the smallest Akaike Information

Criterion (AIC). We built three DR models stating how hospital resistance is linked, respectively, to consumption of fluoroquinolones in TUH, the community and in both areas.

Lastly, we performed one step ahead resistance forecasts for the year 2008 using the ARIMA predictions obtained for the input series. The resistance forecasts were compared with the actual rates for the first five months of 2008.

Statistical analysis was performed with SAS/ETS© 'proc arima' (version 9.1, SAS Institute, Cary, NC, USA).

Results

Descriptive analysis

On average, 458 strains of *E. coli* were isolated every month in TUH. Most of the isolates were cultured from urine (62%), genital fluids (9%) or pus (8%).

The mean ciprofloxacin-resistant *E. coli* percentage in TUH increased during the period, from 12.3% in 2004 to 15.2% in 2007 (Table 1). The chronological series, presented in Figure 1, showed some seasonal patterns for the years 2004 and 2005 that were not seen in the subsequent years.

During the period of the study, mean fluoroquinolone use was 82.9 DDDs/1000 inpatient days in TUH and 2.6 DDDs/1000 inhabitants/day in the community (Table 1). Inpatient fluoroquinolone use in TUH (Figure 2) showed a decrease in ofloxacin and pefloxacin, and an increase in the latest fluoroquinolones to be marketed—moxifloxacin and especially levofloxacin—since 2006. A slight decline in fluoroquinolone use was found in the community between 2004 and 2007 (Figure 3). This decrease was observed for all fluoroquinolones except levofloxacin and lomefloxacin. The strong seasonal patterns observed in the community were due to winter peaks in consumption of levofloxacin and moxifloxacin, which are used against *Streptococcus pneumoniae* in particular.

Analysis of the influence of fluoroquinolone consumption on ciprofloxacin resistance

Influence of hospital fluoroquinolone use

Consumption levels of ciprofloxacin, ofloxacin and levofloxacin were positively associated with resistance trends, taking into account past values of resistance. The influence of fluoroquinolones contributed with a lag of 2 to 4 months. Moxifloxacin and urinary fluoroquinolone uses were not statistically significant. Estimated parameters of the identified model are shown in Table 2 (model A). This DR model fulfilled checking controls and explained 46% of the observed variations in the ciprofloxacin resistance series. One can interpret these findings, for instance, by saying an increase of 1 DDD/1000 inpatient days in ciprofloxacin use was related, with an average lag of 4 months, to an increase of 0.13% in ciprofloxacin resistance, taking into account past values of resistance and consumption of other fluoroquinolones.

Influence of community fluoroquinolone use

Parameters of the identified model are presented in Table 2 (model B). Only the previous year's values of levofloxacin use

Table 1. Evolution of fluoroquinolone-resistant *E. coli* and fluoroquinolone use during 2004–07 in TUH and the Midi-Pyrénées region

	2004	2005	2006	2007	Change 2004–07 (%)
Hospital ciprofloxacin-resistant *E. coli* (mean %)					
FQ-R *E. coli*	12.345	12.539	14.789	15.190	+23.1
Hospital FQ use (mean DDDs/1000 inpatient days)					
total FQs	81.220	93.368	89.237	92.841	+14.3
ofloxacin	39.885	39.176	35.523	27.652	−30.7
ciprofloxacin	33.866	36.140	31.167	33.529	−1.0
levofloxacin	2.986	7.435	12.729	24.104	+707.1
enoxacin	2.924	0.919	0	0	
norfloxacin	0	7.305	7.752	6.031	
moxifloxacin	1.131	2.051	1.756	1.415	+25.1
pefloxacin	0.429	0.342	0.309	0.110	−74.2
Community FQ use (mean DDDs/1000 inhabitants/day)					
total FQs	2.621	2.765	2.746	2.451	−6.5
norfloxacin	0.913	0.928	0.882	0.716	−21.5
ciprofloxacin	0.590	0.598	0.610	0.545	−7.7
ofloxacin	0.417	0.455	0.453	0.394	−5.4
moxifloxacin	0.323	0.363	0.321	0.315	−2.6
levofloxacin	0.283	0.328	0.372	0.374	+32.3
lomefloxacin	0.052	0.053	0.075	0.080	+54.6
enoxacin	0.031	0.028	0.024	0.020	−34.7
pefloxacin	0.010	0.011	0.009	0.006	−51.2

FQ, fluoroquinolone; FQ-R, fluoroquinolone-resistant; DDDs, defined daily doses; TUH, Toulouse University Hospital.

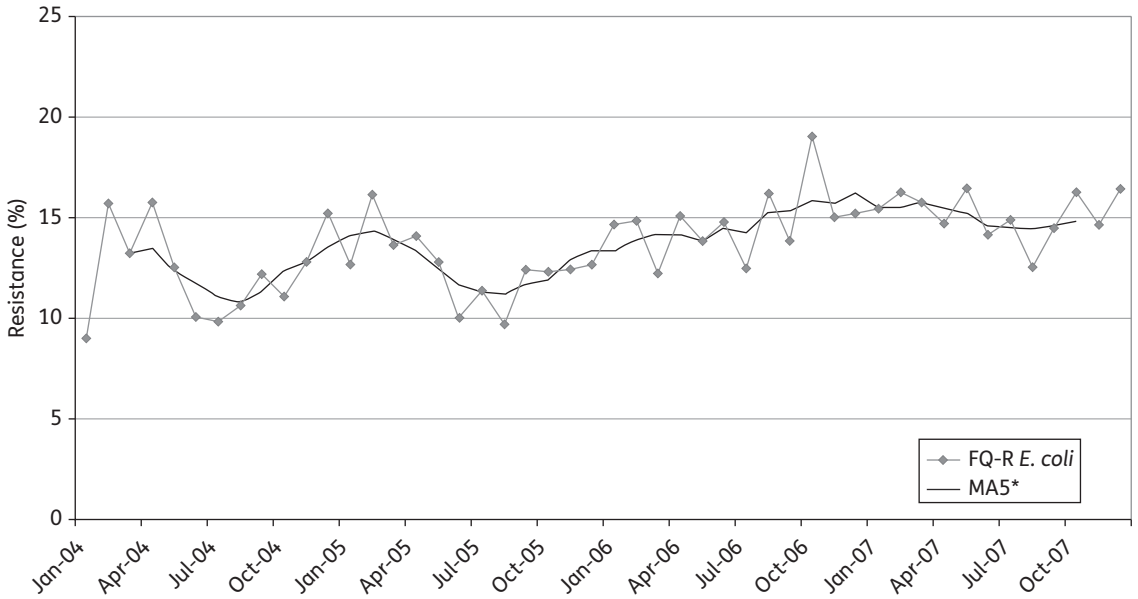

Figure 1. Changes in ciprofloxacin-resistant *E. coli* in TUH (2004–07). MA5*, 5 month-moving average series.

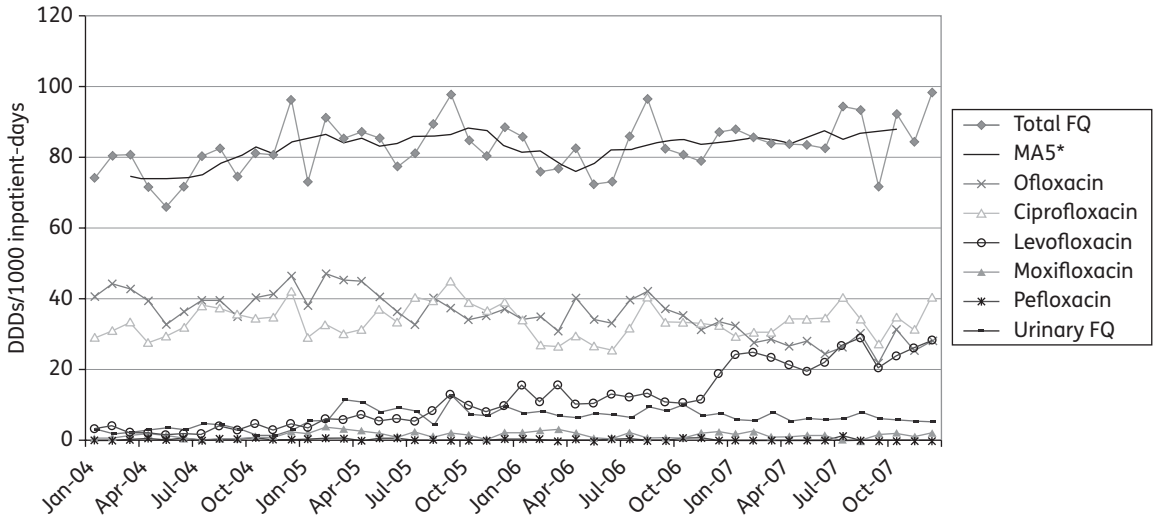

Figure 2. Changes in inpatient fluoroquinolone use in TUH (2004–07). FQ, fluoroquinolones; MA5*, 5 month-moving average series for total fluoroquinolone use. Urinary fluoroquinolones = norfloxacin + enoxacin + lomefloxacin.

were significantly associated with resistance evolution. An increase of 1 DDD/1000 inhabitants/day in levofloxacin use was associated with a rise of 6% in the resistance rate the next year.

Influence of hospital and community fluoroquinolone use

Taking into account both hospital and community fluoroquinolone consumption, only the use of levofloxacin in the community 12 months ago remained statistically associated with an increase in resistance, taking into account past values of resistance. As the use of levofloxacin in TUH 4 months ago was nearing the statistical boundary and may contribute more directly to the generation of resistant strains in the hospital, we decided to include this parameter in the final model. The data are presented in Table 2 (model C). Model C was the best-fitting model and allowed us to explain around 50% of the resistance variability.

Predictions

Using parameters identified in model C and ARIMA forecasts for inputs for the year 2008, we predicted the resistance rate for a 12 month-period (Figure 4). Forecasts ranged between 14.9% and 16.2%. Actual resistance rates were available for the first 5 months of 2008 and were, respectively, 17.8%, 16.0%, 13.3%, 16.0% and 16.0%. All actual rates fell within the 95% confidence interval. The difference between actual and forecast rates divided by its standard error was not larger than 0.15.

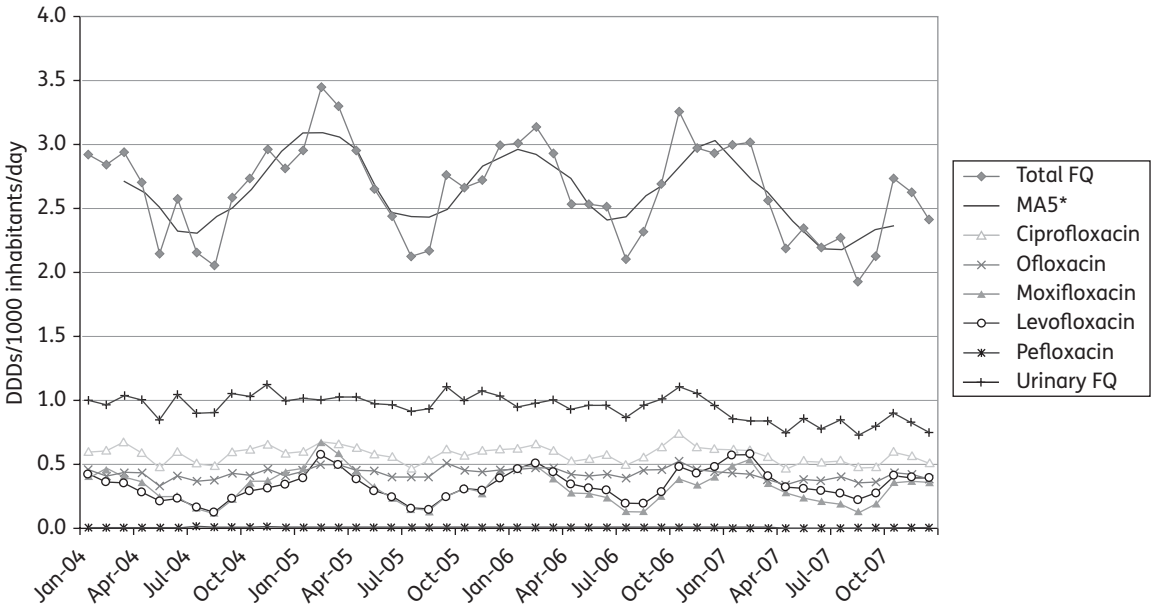

Figure 3. Evolution of community fluoroquinolone use in the Midi-Pyrénées region (2004–07). FQ, fluoroquinolones; MA5*, 5 month-moving average series for total fluoroquinolone use. Urinary fluoroquinolones = norfloxacin + enoxacin + lomefloxacin.

Table 2. Dynamic regression models exploring the relationships between hospital fluoroquinolone-resistant *Escherichia coli* and hospital (model A), community (model B) and overall (model C) fluoroquinolone use

	Order[a]	Parameter (SE)	*P*	Variance	R^2	AIC
Model A: influence of hospital fluoroquinolone use				2.518	46.5	169.6
resistance—autoregressive	2	0.41 (0.11)	0.0050			
hospital ciprofloxacin use	4	0.13 (0.04)	0.0006			
hospital ofloxacin use	2	0.19 (0.03)	$<10^{-4}$			
hospital levofloxacin use	2	0.22 (0.04)	$<10^{-4}$			
Model B: influence of community fluoroquinolone use				2.110	44.8	132.7
constant	0	12.07 (0.94)	$<10^{-4}$			
resistance—autoregressive	2	0.58 (0.14)	$<10^{-4}$			
community levofloxacin use	12	6.48 (2.27)	0.0043			
Model C: influence of hospital and community fluoroquinolone use				1.974	50.1	131.1
constant	0	10.85 (1.08)	$<10^{-4}$			
resistance—autoregressive	2	0.53 (0.15)	0.0003			
hospital levofloxacin use	4	0.09 (0.05)	0.0583			
community levofloxacin use	12	6.70 (2.22)	0.0025			

SE, standard error; *P*, Wald *t*-test; R^2, determination coefficient; AIC, Akaike Information Criterion.
[a]In months.

Discussion

Ciprofloxacin-resistant *E. coli* in TUH increased strongly during the first years of the millennium [on average from 8% in 2003 (data not shown) to 16% in 2008]. This trend is consistent with data collected by the European Antibiotic Resistance Surveillance System (EARSS). In 2007, FQ-R *E. coli* rates ranged from 7% in Norway to 53% in Turkey.[30] In the French EARSS collection, 14.8% of *E. coli* isolates were resistant to fluoroquinolones and 2.5% were of intermediate susceptibility. In our study, the resistance series displayed seasonal variations in 2004–05 that declined in later years. Unfortunately, we have not been able to compare these patterns with other series as resistance is reported yearly in the literature.

France is well known for its high antibiotic consumption. In 2002, France was ranked first for inpatient use[31] and seventh for outpatient use of fluoroquinolones in Europe.[9,32]

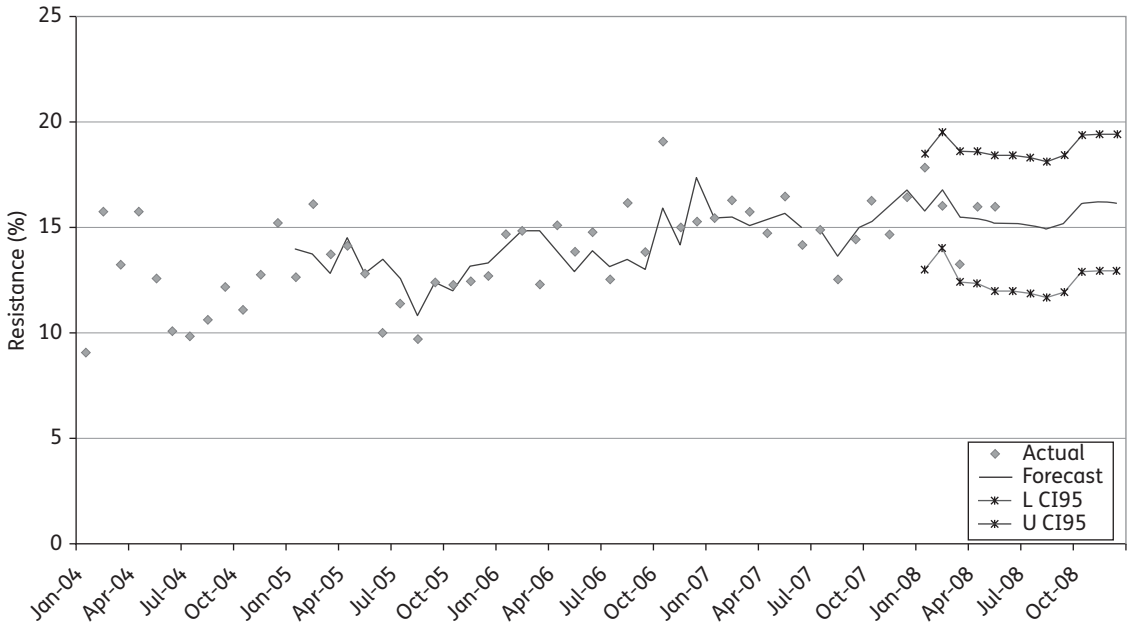

Figure 4. Influence of hospital and community fluoroquinolone use on hospital resistance (ARIMA predictions for consumption were used for inputs). L CI95, lower boundary of the 95% confidence interval for forecast; U CI95, upper boundary of the 95% confidence interval for forecast.

In 2007, consumption of quinolones represented around 7% of the total consumption of antibiotics in France (A. Gallini and R. Bourrel, personal data). During our study period, outpatient consumption in the Midi-Pyrénées region was higher than the mean reported by the European Surveillance of Antimicrobial Consumption (ESAC) group for France in 2003: 2.6 versus 2.0 DDDs/1000 inhabitants/day (wholesalers' data, using the 2004 new DDD for levofloxacin).[32] This difference can be attributed to variations in consumption among French regions. In 2007, the Midi-Pyrénées region ranked first out of the 22 French regions for quinolone consumption in the community, taking into account regional differences in age structures (A. Gallini and R. Bourrel, personal data).

The use of available data is one of the strengths of our study. It demonstrates that this modelling may be achieved on a routine basis. However, the use of data not specifically designed for our objectives raises several issues, which are discussed below.

Dispensing data or reimbursement claims were used as proxies for fluoroquinolone consumption. This may have led to overestimation of fluoroquinolone use (non-consumption of dispensed antibiotics, non-consumption of the entire drug package in the community). Less likely, some situations may underestimate consumption in the community, such as the use of a left-over fluoroquinolone treatment; dispensing without a medical prescription; or dispensing without a reimbursement claim (these last two situations are thought to be very unusual in France). However, these limitations did not affect the time-series evolution that was of interest in this study.

Taking into account the changes in resistance and past values of fluoroquinolone consumption allowed us to explain around 50% of the variation in resistances, suggesting that fluoroquinolone use is the main factor involved in bacterial resistance to ciprofloxacin. A dynamic regression model can handle the use of several antimicrobials, quantify the effect of antimicrobial consumption on resistance and estimate the delay between variations in use and subsequent variations in resistance.[22] Additionally, it is possible to make predictions of future resistance levels. Nonetheless, as this was an ecological study it was more sensitive to bias than individual-level studies[33] and did not address individual risk factors for resistance.[34] In particular, it was impossible to study the effects of some of the factors already known to affect the emergence or dissemination of bacterial resistance, such as inappropriate antibacterial use (i.e. inappropriate choice of antibiotic, dose or length of treatment), hygiene practices (i.e. poor hand-washing or sanitation practices) or individual factors identified in case–control studies (i.e. age, comorbidities, immunosuppression, the presence of urinary catheters, nosocomial infections or hospitalization in an intensive care unit).[4,8,35–39] In addition, some studies found that high levels of consumption of other antibiotics were associated with high levels of resistance to fluoroquinolones in *E. coli*,[24] most probably because of cross-resistance to antimicrobials.[2,13,40,41] We have not been able to examine this relationship.

Interestingly, the influence of fluoroquinolone use in the community seemed to have a stronger influence on hospital ciprofloxacin resistance than inpatient fluoroquinolone consumption. To our knowledge, this is the first published study to have used time-series methods, focusing both on inpatient and outpatient use in relation to FQ-R *E. coli* in hospital. Previously, only two ecological studies had assessed this relationship, but they used different methods and had discordant findings. Bergman *et al.*[24] studied the influence of consumption of various antibiotics in the community on the susceptibility of *E. coli* to various antibiotics between 1997 and 2005 in 20 hospital districts in Finland. Resistance to fluoroquinolones (ciprofloxacin or levofloxacin) was found to be related to amoxicillin consumption, but there was no significant association with fluoroquinolone

use. In this Finnish area, fluoroquinolone use was much lower than in our study, as was the resistance rate (3.5% in 2005). The other study, by MacDougall *et al.*,[10] did not report a significant correlation between hospital fluoroquinolone use and resistance in 17 hospitals in 2000 in the USA, but found a positive correlation between community fluoroquinolone use and hospital resistance. One possible explanation is that the greater exposure to fluoroquinolones in the community may lead to a larger reservoir of resistant organisms in outpatients, spreading in the community and detected in hospitals. In our study, 22% of the *E. coli* isolates tested in TUH's bacteriology laboratory came directly from outpatients through the emergency departments or outpatient clinics. Levofloxacin consumption in the community was one of the strongest predictors of resistance to ciprofloxacin in our hospital. This finding is consistent with MacDougall's results, which suggested that population density may appear as a confounder in this relationship as population density was both correlated to consumption of levofloxacin in the community and to resistance in hospitals. Unfortunately, we were not able to investigate this hypothesis in our context.

Conclusions

This ecological analysis, conducted on a university hospital scale and in a region of high fluoroquinolone consumption, suggests that hospital *E. coli* ciprofloxacin resistance may be linked to the consumption of fluoroquinolones within the hospital and its surrounding community. It also brings out two interesting findings: first, the important influence of levofloxacin consumption compared with the other fluoroquinolones on ciprofloxacin-resistant *E. coli*; second, the important impact of community consumption on hospital resistance. Thus, antimicrobial consumption in the community appears to be a relevant determinant to consider in designing interventions to reduce resistance in hospitals.

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Transparency declarations

None to declare.

References

1 Neu HC. Ciprofloxacin: an overview and prospective appraisal. *Am J Med* 1987; **82**: 395–404.

2 Robicsek A, Jacoby GA, Hooper DC. The worldwide emergence of plasmid-mediated quinolone resistance. *Lancet Infect Dis* 2006; **6**: 629–40.

3 Lautenbach E, Strom BL, Nachamkin I *et al*. Longitudinal trends in fluoroquinolone resistance among Enterobacteriaceae isolates from inpatients and outpatients, 1989–2000: differences in the emergence and epidemiology of resistance across organisms. *Clin Infect Dis* 2004; **38**: 655–62.

4 Garau J, Xercavins M, Rodríguez-Carballeira M *et al*. Emergence and dissemination of quinolone-resistant *Escherichia coli* in the community. *Antimicrob Agents Chemother* 1999; **43**: 2736–41.

5 Karlowsky JA, Thornsberry C, Jones ME *et al*. Susceptibility of antimicrobial-resistant urinary *Escherichia coli* isolates to fluoroquinolones and nitrofurantoin. *Clin Infect Dis* 2003; **36**: 183–7.

6 Mahamat A, Daurès JP, Sotto A. Evaluation of the relation between consumption of fluoroquinolones and emergence of resistance among *Escherichia coli*: contribution of observational and quasi-experimental studies. *Med Mal Infect* 2005; **35**: 543–8.

7 Bolon MK, Wright SB, Gold HS *et al*. The magnitude of the association between fluoroquinolone use and quinolone-resistant *Escherichia coli* and *Klebsiella pneumoniae* may be lower than previously reported. *Antimicrob Agents Chemother* 2004; **48**: 1934–40.

8 Harris AD, Samore MH, Carmeli Y. Control group selection is an important but neglected issue in studies of antibiotic resistance. *Ann Intern Med* 2000; **133**: 159.

9 Goossens H, Ferech M, Vander Stichele R *et al*. Outpatient antibiotic use in Europe and association with resistance: a cross-national database study. *Lancet* 2005; **365**: 579–87.

10 MacDougall C, Powell JP, Johnson CK *et al*. Hospital and community fluoroquinolone use and resistance in *Staphylococcus aureus* and *Escherichia coli* in 17 US hospitals. *Clin Infect Dis* 2005; **41**: 435–40.

11 Zervos MJ, Hershberger E, Nicolau DP *et al*. Relationship between fluoroquinolone use and changes in susceptibility to fluoroquinolones of selected pathogens in 10 United States teaching hospitals, 1991–2000. *Clin Infect Dis* 2003; **37**: 1643–8.

12 Kern WV, Steib-Bauert M, de With K *et al*. Fluoroquinolone consumption and resistance in haematology-oncology patients: ecological analysis in two university hospitals 1999–2002. *J Antimicrob Chemother* 2005; **55**: 57–60.

13 Neuhauser MM, Weinstein RA, Rydman R *et al*. Antibiotic resistance among Gram-negative bacilli in US intensive care units: implications for fluoroquinolone use. *JAMA* 2003; **289**: 885–8.

14 van de Sande-Bruinsma N, Grundmann H, Verloo D *et al*. Antimicrobial drug use and resistance in Europe. *Emerging Infect Dis* 2008; **14**: 1722–30.

15 Harbarth S, Harris AD, Carmeli Y *et al*. Parallel analysis of individual and aggregated data on antibiotic exposure and resistance in Gram-negative bacilli. *Clin Infect Dis* 2001; **33**: 1462–8.

16 Cizman M, Orazem A, Krizan-Hergouth V *et al*. Correlation between increased consumption of fluoroquinolones in outpatients and resistance of *Escherichia coli* from urinary tract infections. *J Antimicrob Chemother* 2001; **47**: 502.

17 Friedrich LV, White RL, Bosso JA. Impact of use of multiple antimicrobials on changes in susceptibility of Gram-negative aerobes. *Clin Infect Dis* 1999; **28**: 1017–24.

18 Goettsch W, van Pelt W, Nagelkerke N *et al*. Increasing resistance to fluoroquinolones in *Escherichia coli* from urinary tract infections in the Netherlands. *J Antimicrob Chemother* 2000; **46**: 223–8.

19 Urbánek K, Kolár M, Strojil J *et al*. Utilization of fluoroquinolones and *Escherichia coli* resistance in urinary tract infection: inpatients and outpatients. *Pharmacoepidemiol Drug Saf* 2005; **14**: 741–5.

20 López-Lozano JM, Monnet DL, Yagüe A *et al*. Modelling and forecasting antimicrobial resistance and its dynamic relationship to antimicrobial use: a time series analysis. *Int J Antimicrob Agents* 2000; **14**: 21–31.

21 Mahamat A, Lavigne JP, Fabbro-Peray P *et al*. Evolution of fluoroquinolone resistance among *Escherichia coli* urinary tract isolates from a French university hospital: application of the dynamic regression model. *Clin Microbiol Infect* 2005; **11**: 301–6.

22 Monnet DL, López-Lozano JM, Campillos P *et al*. Making sense of antimicrobial use and resistance surveillance data: application of ARIMA and transfer function models. *Clin Microbiol Infect* 2001; **7** Suppl 5: 29–36.

23 Monnet DL, López-Lozano JM. Relationship between antibiotic consumption and resistance in European hospitals. *Med Mal Infect* 2005; **35** Suppl 2: S127–8.

24 Bergman M, Nyberg ST, Huovinen P *et al*. Association between antimicrobial consumption and resistance in *Escherichia coli*. *Antimicrob Agents Chemother* 2009; **53**: 912–7.

25 Sabuncu E, David J, Bernède-Bauduin C *et al*. Significant reduction of antibiotic use in the community after a nationwide campaign in France, 2002–2007. *PLoS Med* 2009; **6**: e1000084.

26 National Committee for Clinical Laboratory Standards. *Performance Standards for Antimicrobial Disc Susceptibility Tests-Sixth Edition: Approved Standard M2-A6*. NCCLS, Wayne, PA, USA, 1997.

27 WHO Collaborating Centre for Drugs Statistics Methodology. www.whocc.no/atcddd/ (31 January 2010, date last accessed).

28 Box G, Jenkins G. *Time Series Analysis: Forecasting and Control*. San Francisco: Holden Day, 1976.

29 Pankratz A. *Forecasting with Dynamic Regression Models*. New York: Wiley, 1991.

30 European Antimicrobial Resistance Surveillance System. *EARSS Annual Report 2007*. http://www.rivm.nl/earss/Images/EARSS%202007_FINAL_tcm61-55933.pdf (31 January 2010, date last accessed).

31 Vander Stichele RH, Elseviers MM, Ferech M *et al*. Hospital consumption of antibiotics in 15 European countries: results of the ESAC Retrospective Data Collection (1997–2002). *J Antimicrob Chemother* 2006; **58**: 159–67.

32 Ferech M, Coenen S, Malhotra-Kumar S *et al*. European Surveillance of Antimicrobial Consumption (ESAC): outpatient quinolone use in Europe. *J Antimicrob Chemother* 2006; **58**: 423–7.

33 Nurminen M. Linkage failures in ecological studies. *World Health Stat Q* 1995; **48**: 78–84.

34 Greenland S, Morgenstern H. Ecological bias, confounding, and effect modification. *Int J Epidemiol* 1989; **18**: 269–74.

35 Ena J, López-Perezagua MM, Martínez-Peinado C *et al*. Emergence of ciprofloxacin resistance in *Escherichia coli* isolates after widespread use of fluoroquinolones. *Diagn Microbiol Infect Dis* 1998; **30**: 103–7.

36 Gimber EA, Shields MD, Canawati HN *et al*. Bacteriuria with *Escherichia coli* resistant to ciprofloxacin in patients with spinal-cord injury. *Infect Control Hosp Epidemiol* 1998; **19**: 85–6.

37 Lepelletier D, Caroff N, Reynaud A *et al*. *Escherichia coli*: epidemiology and analysis of risk factors for infections caused by resistant strains. *Clin Infect Dis* 1999; **29**: 548–52.

38 Peña C, Albareda JM, Pallares R *et al*. Relationship between quinolone use and emergence of ciprofloxacin-resistant *Escherichia coli* in bloodstream infections. *Antimicrob Agents Chemother* 1995; **39**: 520–4.

39 McDonald LC, Chen FJ, Lo HJ *et al*. Emergence of reduced susceptibility and resistance to fluoroquinolones in *Escherichia coli* in Taiwan and contributions of distinct selective pressures. *Antimicrob Agents Chemother* 2001; **45**: 3084–91.

40 Poirel L, Villa L, Bertini A *et al*. Expanded-spectrum β-lactamase and plasmid-mediated quinolone resistance. *Emerging Infect Dis* 2007; **13**: 803–5.

41 Lautenbach E, Strom BL, Bilker WB *et al*. Epidemiological investigation of fluoroquinolone resistance in infections due to extended-spectrum β-lactamase-producing *Escherichia coli* and *Klebsiella pneumoniae*. *Clin Infect Dis* 2001; **33**: 1288–94.