Decreased Susceptibility to Commonly Used Antimicrobial Agents in Bacterial Pathogens Isolated from Urinary Tract Infections in Rwanda: Need for New Antimicrobial Guidelines

Claude Mambo Muvunyi,* Florence Masaisa, Claude Bayingana, Léon Mutesa, André Musemakweri, Grégoire Muhirwa, and Geert (W.) Claeys

Department of Clinical Chemistry, Microbiology and Immunology, Ghent University Hospital, De Pintelaan 185, Ghent, Belgium;

Department of Clinical Biology, Centre Hospitalier Universitaire-Butare, National University of Rwanda, Butare, Rwanda;

Department of Internal Medicine, Centre Hospitalier Universitaire-Butare, National University of Rwanda, Butare, Rwanda

Abstract. The aim of this study was to obtain data on susceptibility patterns of pathogens responsible for both community and hospital urinary tract infections (UTIs); and analyzed risk factors for infection caused by ciprofloxacin-resistant *Escherichia coli* and extended-spectrum β -lactamase (ESBL)-producing strains in Rwanda. Of 1,012 urine cultures prospectively studied, a total of 196 (19.3%) yielded significant growth of a single organism. The most common isolate (60.7%) was *Escherichia coli*. The antibiotics commonly used in UTIs are less effective except Fosfomycin-trometamol and imipenem. The use of ciprofloxacin in the previous 6 months (odds ratio [OR] = 7.59 [1.75–32.74]), use of other antibiotics in the previous 6 months (OR = 1.02 [1.02–2.34]), and production of ESBL (OR = 19.32 [2.62–142.16]) were found to be associated with ciprofloxacin resistance among the *E. coli* isolates. Risk factors for ESBL positivity were the use of ciprofloxacin and third-generation cephalosporin in the preceding 6 months (OR = 3.05 [1.42–6.58] and OR = 9.78 [2.71–35.25], respectively); and being an inpatient (OR = 2.27 [1.79–2.89]). Fosfomycin-trometamol could be included as a reasonable alternative for the therapy of uncomplicated UTI in Rwanda.

INTRODUCTION

Urinary tract infections (UTIs) are among the most common bacterial infections both in the community and hospital setting. In the majority of cases, antibiotics are given empirically before the final bacteriology results are available. Therefore, area-specific monitoring studies to document the microorganisms causing UTIs and their antimicrobial susceptibility is mandatory for helping the selection of an effective empirical treatment.¹ Rwanda is among the poorest countries in the world and most people can only afford generic drugs. The most commonly used antibiotics include amoxicillin, nitrofurantoin, and trimethoprim/sulfamethoxazole, and more recently ciprofloxacin was approved to treat UTIs and became available at a low price.^{2,3}

An increasing rate of antibiotic resistance among pathogens responsible for UTIs has caused growing concern worldwide. A number of studies in Europe and in the United States showed a steady increase of the resistance rate of uropathogens to commonly prescribed antibiotics (amoxicillin, trimethoprim-sulfamethoxazole), reducing therapeutic possibilities.^{4–6} In some countries high levels of resistance to ciprofloxacin, one of the current drugs of choice for empiric therapy has been reported in recent years.^{7–9}

Mechanisms of resistance against β -lactam antibiotics in gram-negative bacilli include production of TEM- and AmpC β -lactamases, porin deficiency, and efflux mechanisms and, more recently, extended-spectrum β -lactamase (ESBL). The ESBLs are defined as β -lactamases capable of hydrolyzing oxymino-cephalosporins and are inhibited by β -lactamase inhibitors.¹⁰ Microorganisms responsible for UTIs such as *Escherichia coli* and *Klebsiella* spp. remain the major ESBL-producing organisms isolated worldwide, but these enzymes have also been identified in several other members of the Enterobacteriaceae family and in certain non-fermenters.¹¹

Levels of antibiotic consumption, including the use of fluoroquinolones, show great variations.⁹ As the emergence of resistance is associated with high antibiotic consumption,¹² it is not surprising that resistance to ciprofloxacin in *E. coli* shows great geographical variations as well, reaching high levels in some developing countries.¹³ In addition to monitoring of resistance patterns, identification of risk factors for resistance may contribute to improved empirical treatment. No data on antimicrobial resistance and the prevalence of ESBL producers in UTIs in Rwanda have been published to date.

The aim of this prospective study was to obtain data on susceptibility patterns of pathogens responsible for both community and hospital UTIs in Rwanda to antimicrobials agents currently used to treat UTIs. In addition, we analyzed risk factors for infection caused by ciprofloxacin-resistant *E. coli* and, for the first time, the prevalence and risk factors of ESBL-producing strains in Rwanda are described in this study.

MATERIALS AND METHODS

Study population and bacterial isolates. This prospective study was conducted in both outpatients and inpatients with UTIs at the two largest tertiary teaching hospitals after obtaining approval from the Research Ethics Committee of the Faculty of Medicine (FoMREC). These hospitals were selected because they have a large number of patients and represent patients from large geographical areas. Butare University Hospital, located in the south province of Rwanda, is a 418-bed tertiary-care, teaching hospital with 7,595 patient admissions and almost 33,304 outpatient clinic and emergency room visits annually. Kigali University Hospital, located in the center and serving as a reference center for the eastern, north, and western regions in Rwanda, is a 513-bed tertiary-care with 11,602 patient admissions and almost 105,773 outpatient clinic and emergency room visits annually. Between June and November 2009 a total of 1,012 urine cultures were analyzed in the clinical microbiology laboratories of the two participating hospitals. For each patient, data were prospectively collected through an interview with the patient or the patient's family, and their medical records were checked

*Address correspondence to Claude Mambo Muvunyi, Faculty of Medicine NUR, P.O. Box 217, Huye/Rwanda. E-mail: cmuvunyi@nur.ac.rw

when necessary. Risk factors for ciprofloxacin resistance were as follows: age, sex, presence of a urinary catheter; prior UTI, prior urinary catheter, hospitalization during the previous year; and antibiotic exposure during the preceding 6 months. Each specimen was cultured using a 0.001 mL calibrated loop to inoculate blood agar and MacConkey agar plates, incubated at 37°C for 18–24 hours and the number of colonies was counted. Significant bacteriuria was defined as greater than 10⁵ colony forming units/mL of a single pathogen. Isolates were identified biochemically using Triple Sugar Iron agar, Indol, Methyl Red, Voges Proskauer, and Citrate according to standard microbiologic procedures.¹⁴

Antimicrobial susceptibility testing. Antimicrobial susceptibility testing was performed using a disk diffusion method according to the Clinical and Laboratory Standards Institute (CLSI) guidelines.¹⁴ Antibiotic disks were obtained from Oxoid (Oxoid Ltd., Cambridge, UK): amoxicillin, amoxicillin-clavulanic acid, ceftriaxone, ceftazidime, imipenem, trimethoprim-sulfamethoxazole, trimethoprim, gentamicin, amikacin, netilmicin, nalidixic acid, ciprofloxacin, norfloxacin, ofloxacin, nitrofurantoin, fosfomycin-trometamol, cefalotin, and piperacillin. Quality control was performed once weekly using test strains *E. coli* ATCC 25 922, *Staphylococcus aureus* ATCC 25923, and *Pseudomonas aeruginosa* ATCC 27853.

All isolates were tested for ESBL production by the double-disk synergy test on Mueller-Hinton agar (Sigma Aldrich, St. Louis, MO) using ceftriaxone and ceftazidime placed at a distance of 15–20 mm apart from a disk containing amoxicillin plus clavulanic acid. A clear-cut extension of the inhibition zone around either the ceftazidime or ceftriaxone disk toward the clavulanic acid-containing disk (also called “ghost zone”) was interpreted as positive for ESBL production.¹⁴

Statistical analysis. Data processing and statistical analysis were performed using SPSS software (version 16.0, SPSS, Inc., Chicago, IL). Differences in group proportions for categorical variables were assessed using χ^2 or Fisher’s exact test. Logistic regression analysis was performed to identify risk factors for acquisition of ciprofloxacin-resistant *E. coli* and ESBL production. A *P* value < 0.05 was considered statistically significant.

RESULTS

A total of 1,012 clean-catch urine samples were processed during a 6-month study period (1 June 2009 until 30 November 2009), of which 196 (19.3%) yielded significant growth of a single organism and were included in this study. The mean age of the study population was 42 with a range of 18–90, and 71.9% of the patients were female. Of the 196 patients, 102 were outpatients and 94 were hospitalized patients.

The distribution of uropathogens for outpatients and inpatients is shown in Table 1. *Escherichia coli* was the most common uropathogen, accounting for infections in 119 (60.7%) of the 196 patients; and occurred significantly more frequently in urines from outpatients (70.6%) than from inpatients (50%). The proportion of *Klebsiella* sp., *Proteus* sp., and *Citrobacter* sp. was relatively higher in inpatients (24.5%, 6.5%, 7.4%, and 6.4%) than in outpatients (13.7%, 5.9%, 2, and 1%). *Pseudomonas aeruginosa* was only isolated from inpatients (3.2%). Gram-positive microorganism grew in only 3.1% of the positive cultures and were no longer included in the study.

The antimicrobial susceptibility results of the most frequent uropathogens in the two hospitals are summarized in the

TABLE 1
Bacterial isolates from urinary tract infections, in in- and outpatients

Organisms	No. of isolates (%)		
	Outpatients	Inpatients	Total
<i>Escherichia coli</i>	72 (70.6)	47 (50)	119 (60.7)
<i>Klebsiella</i> spp.	14 (13.7)	23 (24.5)	37 (18.9)
<i>Proteus</i> spp.	6 (5.9)	6 (6.4)	12 (6.1)
<i>Enterobacter</i> spp.	2 (2)	7 (7.4)	9 (4.6)
<i>Citrobacter</i> spp.	1 (1)	6 (6.4)	7 (3.6)
<i>Acinetobacter</i> spp.	2 (2)	1 (1.1)	3 (1.5)
<i>Pseudomonas aeruginosa</i>	0 (0)	3 (3.2)	3 (1.5)
Gram-positive	5 (4.9)	1 (1.1)	6 (3.1)
Total	102 (52)	94 (48)	196 (100)

Table 2. In general, isolates from inpatients were significantly more resistant to antimicrobials than outpatients. One hundred seventy-seven (90.3%) of isolates were susceptible to fosfomycin, and 99.5% of isolates were susceptible to imipenem, which were the most effective drugs. Conversely, high resistance rates were detected among isolates from inpatients of Butare and Kigali university hospitals for amoxicillin (92.9% versus 100%), amoxicillin-clavulanic acid (50% versus 76.3%), trimethoprim-sulfamethoxazole (93.8% versus 71.4%), ciprofloxacin (61.3% versus 35.7%), ceftriaxone (57.1% versus 53.8%), ceftazidime (50% versus 50%), nalidixic acid (64.3% versus 77.5%), and nitrofurantoin (78.6% versus 48.8%). For outpatients the corresponding resistance rates were 73.8% versus 85%, 40.5% versus 51.7%, 71.4% versus 78.3%, 21.4% versus 30%, 9.5% versus 13.3%, 11.9% versus 8.3%, 45.2% versus 41.6%, and 38.1% versus 36.7%. Strains of *E. coli*, isolated from inpatients were more resistant to ciprofloxacin and to third-generation cephalosporin when compared with *E. coli* from outpatient strains and to other species from inpatients as well (Table 3).

The ESBLs were detected in 38.3% (36 of 94) of the strains from inpatients (13.8% of *E. coli*) and 5.9% (6 of 102) of all strains from outpatients (1.9% of *E. coli*).

The univariate and multivariate analysis of risk factors for ciprofloxacin-resistant *E. coli* are shown in Table 4. In univariate analysis, use of ciprofloxacin and other antibiotics in the previous 6 months, history of UTI in the past 12 months, presence of current or previous urinary catheter, being an inpatient or previous hospitalization, and ESBL production were found to be significantly associated with ciprofloxacin resistance among *E. coli* isolates. However, in a forward stepwise multivariate model, only the use of ciprofloxacin in the previous 6 months (odds ratio [OR] = 7.59; 95% confidence interval [CI] = 1.75–32.74; *P* < 0.001), but also the use of other antibiotics in the previous 6 months (OR = 1.02; 95% CI = 1.02–2.34; *P* = 0.005), and production of ESBL (OR = 19.32; 95% CI = 2.62–142.16; *P* < 0.001) were found to be associated with ciprofloxacin resistance among the *E. coli* isolates.

The univariate and multivariate analysis of risk factors for ESBL positivity are presented in Table 5. In univariate analysis, previous hospitalization, prior or presence of urinary tract catheterization, use of ciprofloxacin, and third-generation cephalosporin in the preceding 6 months and being an inpatient were significantly associated with ESBL production. In multivariate only the use of ciprofloxacin and third-generation cephalosporin in the preceding 6 months (OR = 3.05; 95% CI = 1.42–6.58; *P* = 0.04 and OR = 9.78; 95% CI = 2.71–35.25; *P* = 0.01, respectively); and being an inpatient (OR = 2.27; 95%

TABLE 2
Resistance rates to antimicrobial agents tested, in- and outpatients in the two hospitals

Antimicrobial agent	Overall (%) N = 196	Butare University Hospital		Kigali University Hospital	
		Outpatients (%)	Inpatients (%)	Outpatients (%)	Inpatients (%)
		N = 42	N = 14	N = 60	N = 80
Amoxicillin	175 (89.3)	31 (73.8)	13 (92.9)	51 (85)	80 (73.8)
Amoxicillin/clavulanic acid	116 (59.2)	17 (40.5)	8 (50)	31 (51.7)	61 (76.3)
Cefalotin	158 (80.6)	7 (16.7)	4 (28.6)	18 (30)	42 (52.5)
Ceftriaxone	63 (32.1)	4 (9.5)	8 (57.1)	8 (13.3)	43 (53.8)
Ceftazidime	57 (29.1)	5 (11.9)	7 (50)	5 (8.3)	40 (50)
Ciprofloxacin	81 (41.3)	9 (21.1)	5 (35.7)	18 (30)	49 (61.3)
Norfloxacin	78 (39.8)	11 (26.2)	6 (42.9)	16 (26.7)	45 (56.3)
Ofloxacin	71 (36.2)	7 (16.7)	4 (28.6)	18 (30)	42 (52.5)
Nalidixic acid*	86 (56.2)	19 (45.2)	9 (64.3)	20 (41.6)	38 (77.5)
Nitrofurantoin	88 (44.9)	16 (38.1)	11 (78.6)	22 (36.7)	39 (48.8)
Trimethopim/sulfamethoxazole	162 (82.7)	30 (71.4)	10 (71.4)	47 (78.3)	75 (93.8)
Gentamicin	91 (46.4)	12 (28.6)	7 (50)	21 (35)	51 (63.8)
Amikacin	81 (41.3)	20 (47.6)	3 (21.4)	20 (33.3)	38 (47.5)
Piperacillin	152 (77.6)	26 (61.9)	11 (78.6)	39 (65)	76 (95)
Fosfomycin/trometamol	19 (9.7)	3 (7.1)	5 (35.7)	5 (8.3)	6 (7.5)
Imipinem	1 (0.5)	0 (0)	0 (0)	0 (0)	1 (1.3)

* Overall N = 153, outpatient N = 48, and inpatient N = 49 in Kigali University Hospital.

CI = 1.79–2.89; $P < 0.001$) were independently associated with ESBL production.

DISCUSSION

Antimicrobial resistance of bacteria is considered an increasing global concern. The majority of UTIs are treated empirically, especially in developing countries where patients often cannot afford consulting a doctor or having laboratory tests done. Local susceptibility patterns of uropathogens should be available to prescribe appropriate antibiotics. To our knowledge, this is the first study on susceptibilities of bacteria causing UTI and the risk factors for resistance and ESBL positivity in Rwanda.

This study confirms that *E. coli* is still the most common uropathogen isolated from both in- and outpatients, but that various other bacteria cause infection, especially among inpatients (Table 1). These findings are in agreement with similar surveillance studies.^{15–18} Some studies show a decline of *E. coli*,

being replaced by other members of the Enterobacteriaceae. Our hospitalized patients had fewer *E. coli* and more *Klebsiella* spp. The low percentage of *E. coli* among hospital isolates in our study corresponded to that obtained by other investigators.^{15,17}

The UTIs are more frequent in women than in men, which corresponds to our findings because 71.9% of our patients were female.¹⁹

As can be expected, and as reported in other studies, the antimicrobial resistance of hospital isolates was higher than

TABLE 4

Univariate and multivariate analysis of clinical and demographic parameters for resistance to ciprofloxacin in *Escherichia coli* isolates*

Risk factors	Total, N = 119	Ciprofloxacin resistance	P value
Univariate analysis			
Age over 50	32	16 (50)	0.194
Male gender	27	3 (11)	0.972
Diabetes mellitus	10	0 (0)	0.247
Pregnancy	10	1 (10)	0.834
Cardiovascular disease	6	0 (0)	0.379
Pulmonary disease	6	1 (14)	0.769
Oncological disease	8	2 (25)	0.186
Benign prostatic hypertrophy	2	2 (100)	< 0.001
Hospitalized (inpatient)	47	27 (57)	0.005
Hospitalization in last 12 months	39	22 (56)	0.022
UTI in last 12 months	21	13 (62)	0.037
Urinary catheter	14	11 (78)	0.004
Urinary catheter in last 12 months	19	13 (68)	0.037
Ciprofloxacin use in the previous 6 months	13	11 (84)	0.001
Use of other antibiotics in the previous 6 months	51	27 (53)	0.028
ESBL positive	15	14 (93)	< 0.001
Multivariate analysis			
	Odds ratio	Confidence interval	P value
Use other antibiotics in the previous 6 months	1.02	1.02–2.344	0.005
Ciprofloxacin use in the previous 6 months	7.59	1.75–32.74	< 0.001
ESBL positive	19.32	2.62–142.16	< 0.001

* UTI = urinary tract infection; ESBL = extended-spectrum β -lactamase.

TABLE 3
Antimicrobial resistance rates in *Escherichia coli* and other microorganisms, in in- and outpatients

Antimicrobial agent	% Resistance to antibiotic <i>E. coli</i>		Other microorganisms	
	Outpatients	Inpatients	Outpatients	Inpatients
Amoxicillin	86	100	66.7	97.9
Amoxicillin/clavulanic acid	50	70.2	40	74
Cefalotin	79.2	91.5	50	91.5
Ceftriaxone	6.9	38.3	23.3	70
Ceftazidime	4.2	31.9	23.3	68.1
Ciprofloxacin	31.9	57.4	13.3	57.4
Norfloxacin	30.6	57.4	16.7	51.1
Ofloxacin	30	57.4	10	40
Nalidixic acid	45.1	77.4	39.3	78.1
Nitrofurantoin	26.4	29.8	63.3	76.6
Trimethopim/sulfamethoxazole	80.6	95.7	63.3	85.1
Gentamicin	36.1	46.8	23.3	76.6
Amikacin	45.8	46.8	23.3	40.4
Piperacillin	73.6	97.9	40	87.2
Fosfomycin/trometamol	6.9	6.4	10	17
Imipinem	0	0	0	2.1

TABLE 5

Univariate and multivariate analysis of risk factors for ESBL positivity in UTI isolates*

	Total, N = 196	ESBL positivity, N = 42 (%)	P value
Univariate analysis			
Age over 50	56	14 (25)	0.441
Hospitalized (inpatient)	94	36 (38)	< 0.001
Hospitalization in the last 12 months	70	26 (37)	< 0.001
UTI in last 12 months	37	10 (27)	0.357
Urinary catheter	40	20 (50)	< 0.001
Urinary catheter in last 12 months	43	21 (49)	< 0.001
Use of other Antibiotics in the previous 6 months	88	22 (25)	0.271
Ciprofloxacin use in the previous 6 months	22	10 (45)	0.004
Third-generation Cephalosporin use in the previous 6 months	11	8 (73)	< 0.001
Multivariate analysis			
Ciprofloxacin use in the previous 6 months			0.04
Third-generation Cephalosporin use in the previous 6 months			0.01
Hospitalized (inpatient)			< 0.001

* ESBL = extended-spectrum β -lactamase; UTI = urinary tract infection.

in outpatients.^{20,21} In our study, more than 70% of the outpatients isolate and more than 90% of the hospitalized isolates were resistant to Trimethoprim-sulfamethoxazole. Several other studies from the United States and worldwide indicate the emergence of trimethoprim-sulfamethoxazole resistance in a significant percentage (> 20%) of community-acquired UTI isolates.^{17,22} In a study of UTI from outpatients in Canada, Zhanel and others⁵ found resistance to amoxicillin and nitrofurantoin at rates of 41.0% and 0.1%, respectively. Corresponding average values in the two hospitals in our study were 79.4% and 37.4%. In this study, the higher proportion of outpatient and inpatient *E. coli* strains resistant to amoxicillin (86% versus 100%), nitrofurantoin (26.4% versus 29.8%), nalidixic acid (45.1% versus 77.4%), amoxicillin-clavulanic acid (56% versus 70.2%), and gentamycin (36.1% versus 46.8%), is similar to what was reported by Aboderin and coworkers.²³

We documented a remarkably high resistance to ciprofloxacin, which is of great concern because fluoroquinolones are the drugs of choice for first-line empiric treatment of both community and hospital acquired UTI in settings where resistance to trimethoprim/sulfamethoxazole exceeds 20%, and they have become more commonly prescribed as first-line antibiotic therapy in the last few years.^{24,25} Resistance rates for ciprofloxacin against uncomplicated and complicated UTI strains were reported as 8.5% and 19.5%, respectively, by Alos and coworkers.⁷ Recently, Arslan and coworkers reported 17% and 38% resistance rates for the uncomplicated and complicated UTI strains, respectively.²¹ Our rates were found to be much higher: average of 25.7% for the outpatient UTI strains and 48.5% for the inpatient UTI strains for the two hospitals.

Resistance rates in isolates of outpatients were also higher compared with studies from other countries for the majority of antimicrobials except third-generation cephalosporin ceftriaxone and ceftazidime. However, the resistance to third-generation cephalosporins was significantly higher in strains from inpatients (Table 2). Randrianirina and coworkers¹⁶ have

reported an increasing resistance rate to the third-generation cephalosporin in their study in patients 65 years of age. The findings of this study indicate that β -lactams, trimethoprim/sulfamethoxazole, nitrofurantoin, and ciprofloxacin should no longer be used as empirical treatments of UTI in Rwanda, because of their high rate of resistance. Alternatives must be recommended, especially for empirical treatments of uncomplicated UTI (cystitis) in outpatients.

Fosfomycin-trometamol and imipenem were found to be the most effective antimicrobials, 99% and 93% *E. coli* isolates tested being susceptible respectively. As a result, fosfomycin-trometamol and imipenem may be the drugs of choice for empirical therapy of UTIs based on the *in vitro* data. Previous studies have reported fosfomycin-trometamol resistance rates of 0.3% in 288 and no resistance in 100 *E. coli* strains.²⁶ Ullah and coworkers²⁷ in Pakistan have recorded 86.9% of isolates being susceptible to imipenem. Fosfomycin-trometamol has been recommended as a reliable empirical treatment of uncomplicated UTI because of its easy use (single dose), its good tolerance, and its efficacy.^{26,28} Unfortunately, this antibiotic is not yet available in Rwanda. The high susceptibility to imipenem observed in our study is a clear indication that carbapenem resistance is still almost absent in Enterobacteriaceae isolated from UTI in the region. This can be explained by the infrequent use of this antibiotic in the developing world because of its cost and limited availability.

In our study, > 50 years of age seems to be unrelated to ciprofloxacin resistance in *E. coli* strains by univariate analysis. Interestingly, in a recent study, Nicoletti and coworkers⁹ were not able to show a correlation between age (50 years and 65 years or older, respectively) and ciprofloxacin resistance. In contrast, several previous studies have identified > 50 years of age to be associated with high ciprofloxacin rates.^{7,21,29} Our failure to find this association could have been caused by a small number of patients in our study. In our multivariate model, we observed that ciprofloxacin and the use of other antibiotics in the previous 6 months are independent risk factors contributing to ciprofloxacin resistance, as previously reported in other studies.^{4,21,29,30}

For the first time in our laboratory, we performed phenotypic confirmation tests for ESBL production. The prevalence of ESBLs both in outpatients and inpatients has been investigated and varies among countries in several studies.^{31,32} In all of these, *E. coli* and *Klebsiella* spp. accounted for the vast majority of isolates. Our study showed a higher percentage of ESBL positivity among *E. coli* isolates when compared with reports from Canada (0.26%), Europe (1.3%), and Tunisia (2.7%).^{33–35} In contrast, in agreement with our data, Russian, Cameroonian, and Korean studies revealed that overall prevalence of ESBLs among *E. coli* isolates was 15.8%, 14.3%, and 9.3%, respectively.^{36–38} Studies conducted by Akram and others¹⁷ showed a higher percentage, than our study, of *E. coli* producing ESBL. Prior antibiotics, especially ciprofloxacin, third-generation cephalosporins, and stay in hospital; major risk factors retained in our multivariate analysis, were also found to be independent risk factors for ESBL positivity in previous studies.^{39,40}

In conclusion, Gram-negative organisms, especially *E. coli* were the most common organisms isolated. We found that antibiotics commonly used for the treatment of UTI in Rwanda are far from effective. The ESBL producers are, as in many countries, frequent in Enterobacteriaceae in Rwanda. On the

basis of our findings, we suggest that an antimicrobial agent such as fosfomycin-trometamol could be alternative therapy for uncomplicated UTI, and should be introduced in the national guidelines. Antibiotics not tested in this study, such as tigecycline and temocillin (only parenteral available), should be investigated for their potential in the future. Urine culture and antimicrobial susceptibility testing are recommended in Rwanda for UTI patients. Further studies with a larger number of isolates involving a laboratory from a primary care setting in various part of the country are required. Moreover, this high resistance is probably of relevance for the treatment of other infections, and resistance in Gram-positive organisms should also be studied.

Received January 28, 2011. Accepted for publication February 23, 2011.

Acknowledgments: We thank the staff in the Bacteriology unit at Kigali and Butare Teaching Hospitals for their technical assistance. In particular, we thank Carine Uwera, Innocent Nzabahimana, and Jean Baptiste Mugema for their excellent technical support in data collection and processing.

Financial support: This study was financially supported by a PhD grant from the Ghent University, Ghent, Belgium through its VLIR (Flemish Interuniversity Council) own initiative project number VLIR-UOS ZEIN2007PR342-19878.

Disclaimer: This study was approved by the Research Ethics Committee of the Faculty of Medicine (code: 04/FoMREC/09).

Disclosure: Part of this manuscript was presented at the 50th Interscience Conference on Antimicrobial Agents and Chemotherapy in Boston, MA in September 2010 (control/tracking number: 2010-A-1286-ASM-ICAAC).

Authors' addresses: Claude Mambo Muvunyi, Florence Masaisa, Claude Bayingana, Léon Mutesa, André Musemakweri, and Grégoire Muhirwa, Faculty of Medicine NUR, Huye/Rwanda, E-mails: cmuvunyi@nur.ac.rw, kabasius@yahoo.fr, cbayingana@nur.ac.rw, lmutesa@nur.ac.rw, amusemakweri@nur.ac.rw, and gmuhirwa@nur.ac.rw. Geert (W.) Claey's, Department of Clinical Chemistry, Microbiology and Immunology, Ghent University Hospital, De Pintelaan 185, Ghent, Belgium, E-mail: geert.claeys@ugent.be.

REFERENCES

- Smith RD, Coast J, 2002. Antimicrobial resistance: a global response. *Bull World Health Organ* 80: 126–133.
- Warren JW, Abrutyn E, Hebel JR, Johnson JR, Schaeffer AJ, Stamm WE, 1999. Guidelines for antimicrobial treatment of uncomplicated acute bacterial cystitis and acute pyelonephritis in women. Infectious Diseases Society of America (IDSA). *Clin Infect Dis* 29: 745–758.
- Echols RM, Tosiello RL, Haverstock DC, Tice AD, 1999. Demographic, clinical, and treatment parameters influencing the outcome of acute cystitis. *Clin Infect Dis* 29: 113–119.
- Kahlmeter G, Munday P, 2003. Cross-resistance and associated resistance in 2478 *Escherichia coli* isolates from the Pan-European ECO.SENS Project surveying the antimicrobial susceptibility of pathogens from uncomplicated urinary tract infections. *J Antimicrob Chemother* 52: 128–131.
- Zhanell GG, Hisanaga TL, Laing NM, DeCorby MR, Nichol KA, Weshnowski B, Johnson J, Noreddin A, Low DE, Karlowsky JA, Hoban DJ, 2006. Antibiotic resistance in *Escherichia coli* outpatient urinary isolates: final results from the North American Urinary Tract Infection Collaborative Alliance (NAUTICA). *Int J Antimicrob Agents* 27: 468–475.
- Gupta K, Scholes D, Stamm WE, 1999. Increasing prevalence of antimicrobial resistance among uropathogens causing acute uncomplicated cystitis in women. *JAMA* 281: 736–738.
- Alos JI, Serrano MG, Gomez-Garcés JL, Perianes J, 2005. Antibiotic resistance of *Escherichia coli* from community-acquired urinary tract infections in relation to demographic and clinical data. *Clin Microbiol Infect* 11: 199–203.
- Kahlmeter G, 2003. An international survey of the antimicrobial susceptibility of pathogens from uncomplicated urinary tract infections: the ECO.SENS Project. *J Antimicrob Chemother* 51: 69–76.
- Nicoletti J, Kuster SP, Sulser T, Zbinden R, Ruef C, Ledergerber B, Weber R, 2010. Risk factors for urinary tract infections due to ciprofloxacin-resistant *Escherichia coli* in a tertiary care urology department in Switzerland. *Swiss Med Wkly* 140: w13059.
- Bush K, Jacoby GA, Medeiros AA, 1995. A functional classification scheme for beta-lactamases and its correlation with molecular structure. *Antimicrob Agents Chemother* 39: 1211–1233.
- Jacoby GA, Munoz-Price LS, 2005. The new beta-lactamases. *N Engl J Med* 352: 380–391.
- Goossens H, Ferech M, Vander Stichele R, Elseviers M, 2005. Outpatient antibiotic use in Europe and association with resistance: a cross-national database study. *Lancet* 365: 579–587.
- Dromigny JA, Nabeth P, Juergens-Behr A, Perrier-Gros-Claude JD, 2005. Risk factors for antibiotic-resistant *Escherichia coli* isolated from community-acquired urinary tract infections in Dakar, Senegal. *J Antimicrob Chemother* 56: 236–239.
- Clinical and Laboratory Standards Institute, 2008. *Performance Standards for Antimicrobial Susceptibility Testing: Eighteenth Information Supplement 100-S18*. Wayne, PA: Clinical and Laboratory Standards Institute.
- Raka L, Mulliqi-Osmani G, Berisha L, Begolli L, Omeragiq S, Parsons L, Salfinger M, Jaka A, Kurti A, Jakupi X, 2004. Etiology and susceptibility of urinary tract isolates in Kosovo. *Int J Antimicrob Agents* 23 (Suppl 1): S2–S5.
- Randrianirina F, Soares JL, Carod JF, Ratsima E, Thonnier V, Combe P, Grosjean P, Talarmin A, 2007. Antimicrobial resistance among uropathogens that cause community-acquired urinary tract infections in Antananarivo, Madagascar. *J Antimicrob Chemother* 59: 309–312.
- Akram M, Shahid M, Khan AU, 2007. Etiology and antibiotic resistance patterns of community-acquired urinary tract infections in J N M C Hospital Aligarh, India. *Ann Clin Microbiol Antimicrob* 6: 4.
- Andrade SS, Sader HS, Jones RN, Pereira AS, Pignatari AC, Gales AC, 2006. Increased resistance to first-line agents among bacterial pathogens isolated from urinary tract infections in Latin America: time for local guidelines? *Mem Inst Oswaldo Cruz* 101: 741–748.
- Schaeffer AJ, Rajan N, Cao Q, Anderson BE, Pruden DL, Sensibar J, Duncan JL, 2001. Host pathogenesis in urinary tract infections. *Int J Antimicrob Agents* 17: 245–251.
- Hryniewicz K, Szczypa K, Sulikowska A, Jankowski K, Betlejewska K, Hryniewicz W, 2001. Antibiotic susceptibility of bacterial strains isolated from urinary tract infections in Poland. *J Antimicrob Chemother* 47: 773–780.
- Arsilan H, Azap OK, Ergonul O, Timurkaynak F, 2005. Risk factors for ciprofloxacin resistance among *Escherichia coli* strains isolated from community-acquired urinary tract infections in Turkey. *J Antimicrob Chemother* 56: 914–918.
- Karlowsky JA, Kelly LJ, Thornsberry C, Jones ME, Sahm DF, 2002. Trends in antimicrobial resistance among urinary tract infection isolates of *Escherichia coli* from female outpatients in the United States. *Antimicrob Agents Chemother* 46: 2540–2545.
- Aboderin OA, Abdu AR, Odetoyin BW, Lamikanra A, 2009. Antimicrobial resistance in *Escherichia coli* strains from urinary tract infections. *J Natl Med Assoc* 101: 1268–1273.
- Goettsch W, van Pelt W, Nagelkerke N, Hendrix MG, Buiting AG, Petit PL, Sabbe LJ, van Griethuysen AJ, de Neeling AJ, 2000. Increasing resistance to fluoroquinolones in *Escherichia coli* from urinary tract infections in The Netherlands. *J Antimicrob Chemother* 46: 223–228.
- Hooton TM, 2003. Fluoroquinolones and resistance in the treatment of uncomplicated urinary tract infection. *Int J Antimicrob Agents* 22 (Suppl 2): 65–72.
- Schito GC, 2003. Why fosfomycin trometamol as first line therapy for uncomplicated UTI? *Int J Antimicrob Agents* 22 (Suppl 2): 79–83.
- Ullah F, Malik SA, Ahmed J, 2009. Antimicrobial susceptibility and ESBP prevalence in *Pseudomonas aeruginosa* isolated

- from burn patients in the north west of Pakistan. *Burns* 35: 1020–1025.
28. Lobel B, 2003. Short term therapy for uncomplicated urinary tract infection today. Clinical outcome upholds the theories. *Int J Antimicrob Agents* 22 (Suppl 2): 85–87.
 29. Sire JM, Nabeth P, Perrier-Gros-Claude JD, Bahsoun I, Siby T, Macondo EA, Gaye-Diallo A, Guyomard S, Seck A, Breurec S, Garin B, 2007. Antimicrobial resistance in outpatient *Escherichia coli* urinary isolates in Dakar, Senegal. *J Infect Dev Ctries* 1: 263–268.
 30. Howard AJ, Magee JT, Fitzgerald KA, Dunstan FD, 2001. Factors associated with antibiotic resistance in coliform organisms from community urinary tract infection in Wales. *J Antimicrob Chemother* 47: 305–313.
 31. Paterson DL, Bonomo RA, 2005. Extended-spectrum beta-lactamases: a clinical update. *Clin Microbiol Rev* 18: 657–686.
 32. Reinert RR, Low DE, Rossi F, Zhang X, Wattal C, Dowzicky MJ, 2007. Antimicrobial susceptibility among organisms from the Asia/Pacific Rim, Europe and Latin and North America collected as part of TEST and the *in vitro* activity of tigecycline. *J Antimicrob Chemother* 60: 1018–1029.
 33. Mulvey MR, Bryce E, Boyd D, Ofner-Agostini M, Christianson S, Simor AE, Paton S, 2004. Ambler class A extended-spectrum beta-lactamase-producing *Escherichia coli* and *Klebsiella* spp. in Canadian hospitals. *Antimicrob Agents Chemother* 48: 1204–1214.
 34. Nijssen S, Florijn A, Bonten MJ, Schmitz FJ, Verhoef J, Fluit AC, 2004. Beta-lactam susceptibilities and prevalence of ESBL-producing isolates among more than 5000 European Enterobacteriaceae isolates. *Int J Antimicrob Agents* 24: 585–591.
 35. Smaoui H, Mahjoubi F, Boutiba I, Jouaihia W, Thabet L, Znazen A, Kammoun A, Mezghanni S, Triki O, Hammami A, Ben Hassen A, Kechrid A, Ben Redjeb S, 2003. Antibiotic resistance among *E. coli* isolates from urinary tract infections (1999–2000): a multicenter study. *Tunis Med* 81: 390–394.
 36. Edelstein M, Pimkin M, Palagin I, Edelstein I, Strachounski L, 2003. Prevalence and molecular epidemiology of CTX-M extended-spectrum beta-lactamase-producing *Escherichia coli* and *Klebsiella pneumoniae* in Russian hospitals. *Antimicrob Agents Chemother* 47: 3724–3732.
 37. Gangoue-Pieboji J, Bedenic B, Koulla-Shiro S, Randegger C, Adiogo D, Ngassam P, Ndumbe P, Hachler H, 2005. Extended-spectrum-beta-lactamase-producing Enterobacteriaceae in Yaounde, Cameroon. *J Clin Microbiol* 43: 3273–3277.
 38. Ryoo NH, Kim EC, Hong SG, Park YJ, Lee K, Bae IK, Song EH, Jeong SH, 2005. Dissemination of SHV-12 and CTX-M-type extended-spectrum beta-lactamases among clinical isolates of *Escherichia coli* and *Klebsiella pneumoniae* and emergence of GES-3 in Korea. *J Antimicrob Chemother* 56: 698–702.
 39. Skippen I, Shemko M, Turton J, Kaufmann ME, Palmer C, Shetty N, 2006. Epidemiology of infections caused by extended-spectrum beta-lactamase-producing *Escherichia coli* and *Klebsiella* spp.: a nested case-control study from a tertiary hospital in London. *J Hosp Infect* 64: 115–123.
 40. Yilmaz E, Akalin H, Ozbey S, Kordan Y, Sinirtas M, Gurcuoglu E, Ozakin C, Heper Y, Mistik R, Helvacı S, 2008. Risk factors in community-acquired/onset urinary tract infections due to extended-spectrum beta-lactamase-producing *Escherichia coli* and *Klebsiella pneumoniae*. *J Chemother* 20: 581–585.