Antimicrobial activity against gram negative bacilli from Yaounde Central Hospital, Cameroon

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Abstract

Background: Antimicrobial resistance among bacteria pathogens is a world-wide issue. The antimicrobial susceptibility patterns of common pathogenic bacteria are essential to guide empirical and pathogen-specific therapy; unfortunately, these data are scarse in Cameroon.

Objective: To determine the antimicrobial susceptibility patterns of Gram-negative bacilli isolated in Yaounde Central Hospital Laboratory of Bacteriology.

Methods: Gram-negative bacilli isolates (n = 505), obtained from a wide range of clinical specimens (urine, pus and blood) in Yaoundé Central Hospital Laboratory of Bacteriology between March 1995 and April 1998, were evaluated for resistance to antibiotics (amoxicillin, amoxicillin/clavulanate, piperacillin, cefazolin, cefoxitin, cefotaxime, ceftazidime, aztreonam, imipenem, gentamicin, tobramicin, ofloxacin and trimethoprim/sulfamethoxazole) by the Kirby-Bauer disk diffusion method.

Results: High rates of resistance were found in most of the bacteria studied. Resistance to all isolates was mostly observed for amoxicillin (87%), piperacillin (74%) and trimethoprim/sulfamethoxazole (73%). Susceptibilities to third generation cephalosporins (cefotaxime, ceftazidime) and monobactame (aztreonam) were = 91% for *Escherichia coli*, = 71% for *Klebsiella* spp., = 98% for *Proteus mirabilis*, = 50% for *Enterobacter* spp. and *Citrobacter* spp. *Pseudomonas aeruginosa* was less susceptible to cefotaxime (2%) and aztreonam (33%), and highly susceptible to ceftazidime (72%) whereas *Acinetobacter baumannii* was highly resistant to aztreonam (100%), to cefotaxime (96%) and cetazidime (62%). Imipenem (98%) was the most active antibiotic followed by the ofloxacine (88%). Susceptibility of all isolates to gentamicin was 67%.

Conclusion: These results indicate that surveillance to antimicrobial resistance in Cameroon is necessary to monitor microbial trends, antimicrobial resistance pattern, and provide information for choosing empirical or direct therapy to physicians.

Key words: antimicrobial agents, resistance, Gram-negative bacilli, bacteria susceptibility testing, Cameroon.

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Introduction

Gram-negative bacilli are the most important bacterial pathogen, and are generally resistant to antibiotics [1, 2, 3]. Monitoring for antimicrobial resistance in this group is important because resistance has been reported to be associated with increased patient morbidity and mortality, and contributed to escalating healthcare cost [4]. Antimicrobial resistance is increasing in many bacteria and is a worldwide problem [5, 6].

The antimicrobial susceptibility patterns of common pathogenic bacteria are essential to guide empirical and pathogen-specific therapy. This information is also important for rational policies against antimicrobial resistance. Unfortunately, in many developing countries, these data are scarce because of dwindling resources. In Cameroon, data on antimicrobial resistance among bacterial pathogens are sparse. In an effort to determine the extent of antimicrobial resistance amongst Gram-negative bacilli isolated in Yaounde Central Hospital Laboratory of Bacteriology, we conducted a survey of 505 Gram-negative bacilli and measured their in vitro susceptibility to antimicrobial agents.

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Materials and Methods

Between April 1995 and March 1998, all the aerobic Gram-negative bacilli isolated in the laboratory of bacteriology of Yaounde Central Hospital were collected prospectively. The isolates studied were confined to unrelated first isolates from different patients, and did

not include multiple isolates from the same patient. Isolates were recovered from urine, pus and blood cultures and identified by standard laboratory technique methods [7] and confirmed by Api 20E (BioMerieux, France).

The antimicrobial susceptibility test was determined by the Kirby-Bauer disk diffusion method following the National Committee of Clinical Laboratory Standards (NCCLS) for agar diffusion tests [8]. The antibiotics tested were amoxicillin (30μg), amoxicillin/clavulanate (20/10 μg), piperacillin (100 μg), imipenem (10 μg), cefazolin (30 μg), cefoxitin (30 μg), cefotaxime (30 μg), ceftazidime (30 μg), aztreonam (30 μg), gentamicin (10 μg), tobramycin (10 μg), ofloxacin (5 μg) and trimethoprim/sulfamethoxazole (1.25/23.75 μg). The following American Type Culture Collection (ATCC) microorganisms were tested each time susceptibility testing was performed: *Escherichia coli* ATCC 25922 and *Pseudomonas aeruginosa* ATCC 27853. Test results were only validated in the cases where inhibition zone diameters of the control strains were within performance ranges [8].

Data were analyzed using Whonet 4 (World Health Organization, Geneva, Switzerland) and resistance included combined, intermediary and resistance results.

Results

A total of 505 aerobic isolates were collected, identified, and tested. Enterobacteriaceae (*E. coli*, *Klebsiella* spp., *Proteus* spp., *Enterobacter* spp., and *Citrobacter* spp.) represented 79.8% of the isolated strains. Non fermentative Gram negative bacilli (*Pseudomonas* spp. and *Acinetobacter baumannii*) represented 20.2%. The sources of the isolates are shown in table 1.

Table 2 summarizes the results of susceptibility tests of Gram-negative bacilli studied against antimicrobial agents. Imipenem (98% susceptible (S)) was the most active agent against all pathogens tested followed by ofloxacine (88% S) and ceftazidime (86% S). Amoxicillin (13% S), piperacillin (26% S), trimethoprim/sulfamethoxazole (27% S), cefazolin (30% S) and amoxicillin/clavulanate (37% S) were the least active agents.

Against *E. coli*, susceptibility rates range from 15% (amoxicillin, piperacillin) to 99% (imipenem). The least active agents against *Klebsiella* spp. were amoxicillin (0% S) followed by piperacillin (10% S) and trimethoprim/sulfamethoxazole (15% S). The most active agents were imipenem (93% S) and ofloxacin (98% S). *Proteus mirabilis* was the most sensitive pathogen of all microorganisms. The susceptibility rate ranged from 40 % (amoxicillin) to 100% (imipenem, ceftazidime and aztreonam). Concerning *Enterobacter* spp., only two antimicrobial agents had activity rate >80% ((ofloxacin (98%) and imipenem (97%)). For *Citrobacter* spp. the most active agents were imipenem (96% S) followed by ceftazidime (76% S) and ofloxacin (73% S). Only 94% of *P. aeruginosa* isolates were susceptible to imipenem, as compared to 100% observed for *A. baumannii*. However, 72% of *P. aeruginosa* were susceptible to ceftazidime as compared to only 38 % of the *A. baumannii*. All the *A. baumannii* isolates tested were resistant to aztreonam.

Table 1. Distribution (%) of bacterial species by clinical specimen

Organism	Clinical specimen		
	Pus	Urine	Blood
Escherichia coli	33.7	65.0	1.3
Klebsiella spp.	43.3	44.3	12.4
Proteus mirabilis	58.0	42.0	0.0
Enterobacter spp.	47.5	42.5	10.0
Citrobacter spp.	50.0	50.0	0.0
Indole positif *Proteus* spp.	50.0	43.8	6.2
Pseudomonas aeruginosa.	71.6	22.4	6.0
Acinetobacter baumannii	54.2	25.0	20.8
Pseudomonas spp.	70.0	10.0	20.0

Table 2. Susceptibility (%) of Gram negative bacilli studied

	All isolates	*E. coli*	*Klebsiella* spp.	*P. mirabilis*	*Enterobacter* spp.	*Citrobacter* spp.	Indole positif *Proteus* spp.	*P. aeruginosa*	*A. baumannii*	*Pseudomonas* spp.
	n=505	n=163	n=97	n=62	n=40	n=26	n=16	n=67	n=24	n=10
Isolates										
Antibiotic	%S	%S	%S	%S	%S	%S	%S	%S	%S	%S
Amoxicillin	13	15	0	40	0	0	0	/	/	/
Amoxicillin / clavulanate	37	43	35	69	0	40	0	/	/	/
Piperacillin	26	15	10	53	35	19	56	46	4	50
Cefazolin	30	41	24	48	0	4	0	/	/	/
Cefoxitin	74	91	84	97	5	8	31	/	/	/
Cefotaxime	67	91	71	98	60	50	94	2	4	30
Ceftazidime	86	95	88	100	80	76	94	72	38	70
Aztreonam	73	92	75	100	65	69	88	33	0	20
Imipenem	98	99	99	100	97	96	94	94	100	100
Gentamicin	67	84	65	68	60	58	50	49	58	50
Tobramycin	57	/[a]	/	/	/	/	/	56	69	50
Ofloxacine	88	91	98	98	98	73	82	66	70	90
Trimethoprim /sulfamethoxazole	27	22	15	55	43	19	6	/	/	/

[a] not tested

Discussion

This study represents an extensive examination of the susceptibility patterns of Gram negative bacilli isolated in the laboratory of bacteriology of Yaounde Central Hospital. Our isolates represente both nosocomial and community acquired-pathogens, and were collected from April 1995 to March 1998. The results of this study show that in general, high rate of resistance occurs among Gram-negative bacilli to commonly used antibiotics (penicillins, first generation cephalosporin and trimethoprim/sulfamethoxazole).

For *E. coli* and *Klebsiella* spp., the rate of resistance to third generation cephalosporins and other â-lactam antibiotics can be explained by the high production of penicillinase and the production of extended spectrum â-lactamase [3, 9]. However, the rate of this resistance is high compared to those reported in developed countries [10, 11].

Enterobacter spp. and *Citrobacter* spp. were highly resistant to ceftazidime, cefotaxime and aztreonam. Similar results were observed in Cairo (Egypt) by El Kholy *et al.*, 2003 [12] and in developed countries [10, 13]. This resistance could be explained by the high production of cephalosporinase and the production of extended spectrum â-lactamase by these strains [3, 9].

Our data showed that *P. mirabilis* is the most sensitive species. The susceptibility rates of this species to all antibiotics tested are compared to those observed in developed countries [10, 11].

Non-â-lactam antibiotics resistance rates among the species of the *Enterobacteriaceae* family studied were comparable to the reported rates in other parts of developing countries [14, 15], but higher than those reported in developed countries [10, 13]. The combination of trimethoprime/sulfamethoxazole is extensively used in Africa owing to its antimicrobial spectrum of activity, and its low cost [16]. In addition, extended spectrum â-lactamase production is usually associated with resistance to non-â-lactam antibiotics such as aminoglycosides, fluoroquinolones and trimethoprim/sulfamethoxazole [17].

The susceptibility rates of isolates of *P. aeruginosa* and *A. baumannii* in this study to all antibiotics tested except for imipenem, were low compared to those reported in developed countries [10, 18, 19] and similar to those observed in Egypt [12] and in West Africa [15].

In conclusion, our study suggests the high rates of antimicrobial resistance among Gram-negative bacilli. The presence of *E. coli* and *Klebsiella* spp. isolates resistant to third generation cephalosporin suggests the importance of monitoring this phenotype. Particularly alarming is the appearance of low level imipenem resistance among different species of Gram negative bacilli studied. The results of this study indicate that more resources should be allocated to encourage good antibiotics utilization and practice. In addition, to provide information for choosing either empirical or direct therapy to physicians, surveillance to antimicrobial resistance is necessary.

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