

Analysis of the In-vitro Resistance developed by Uropathogenic Enterobacteria towards Antibiotics commonly used against the Urinary Infections in the City of Lubumbashi (DRC)

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Abstract

At present, the increased resistance shown by uropathogenic enterobacteria towards antibiotics commonly used against the urinary infections is looked at as public health issue of great concern in the city of Lubumbashi. Indeed, the loss in efficiency shown by a number of antibiotics is leading to the growth in the morbidity and mortality recrudescence among the population. As a result, the health's expenses have increased on both the individual and the community level. This increased resistance shown by the uropathogenic bacteria is thought to arise from the self-medication and the misuse of antibiotics in the take in charge of the urinary infections. It has caused the experts involved in the health sector to wonder on how the empirical treatment scheme could be

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amended. The present survey aims at isolating and describing bacteria strains acknowledged to be responsible for urinary infections in view evaluating their sensitivity towards antibiotics that are commonly prescribed to the patients by the physicians. It is based on the data from a cross sectional study (1222 patients monitored) carried out during two years at the University Clinics of Lubumbashi and tests of sensitivity of the bacterial strains towards a set of the commonly used antibiotics as recommended by the French Society of Microbiology Antibiogram Committee (FSM-AC). It is expected that the findings of the present survey could enable adapting the therapeutic scheme based on the best choice of antibiotics in view minimizing the increase in resistance shown by the uropathogenic bacteria.

Keywords: In-vitro resistance, Antibiotics, Uropathogenic enterobacteria, Urinary infections

1 Introduction

The urinary infections constitute the most recurrent pathologies encountered in the communities and hospitals [1, 2]. As the matter of fact, they occupy the second position after the respiratory infections [3], and are yearly affecting about 10% of the population. In the city of Lubumbashi, the urinary infections are generally healed using antibiotics. At the University Clinics of Lubumbashi, the physicians have observed that the uropathogenic enterobacteria are being less sensitive to the commonly prescribed antibiotics. One thinks that the bacterial strains responsible for the urinary infections have gained in resistance and tends to use the more efficient bactericidal drugs. In spite of the legendary sensitivity of the uropathogenic enterobacteria towards antibiotics, the increasing resistance, which characterise them in some circumstances, remains an issue of great concern on the health public standpoint [4, 5]. This issue is extensively studied in the European, Asian and African countries [2, 6]. In the city of Lubumbashi, one thinks that the increased resistance shown by the uropathogenic bacteria towards the commonly used antibiotics is related to their misuse of drugs and the self-medication. Some experts involved in health issues encourage the physicians to improve their therapeutic scheme when dealing with the urinary infections based on the achievement antibiograms in order to combat the misuse of antibiotics in spite of the difficult access to medical materials experienced by laboratories due to their scarcity and high prices on the local market. The incentive of the present survey is to provide the best description of the bacterial strains implicated in the urinary infections and evaluate their sensitivities towards a set of antibiotics available in the drugstores. As the matter of fact, the best medical diagnosis can be done when the therapeutic choice is based on the correct evaluation of the resistance of the bacteria strains of interest towards a large set of antibiotics.

2 Material and methods

The present survey has been carried out at the Medical Laboratory, the Unit of Microbiology of the university clinics of Lubumbashi. It consists in a cross sectional study achieved from January 2008 to December 2010 and has been oriented towards a

bacterial population made of the non-redundant strains. The latter have been isolated from the patients (1222) based on the biologic diagnosis of the urinary infection (UI) using the KASS criteria: a leucocyturia superior or equal to 10000/ml along with a bacteriuria superior or equal to 100 000/ml. The isolated bacterial strains have been identified on the cultural and biochemical standpoint and based on their usual bacteriological characters. As recommended by the French Society of Microbiology Antibigram Committee (FSM-AC), the resistance of enterobacteria towards a set of antibiotics (Ampicillin, Amoxicillin, Augmentin, Cefotaxime, Cefuroxime, Meropenem, Chloramphenicol, Nalidixic acid, Ciprofloxacin, pefloxacin, Gentamicine, Amikacine and Cotrimoxazole) has been evaluated at laboratory through the achievement of antibiograms by the disc diffusion method of Kirby and Bauer. The strains producing betalactamase with a broadened spectrum (BLSE) have been analysed using a double synergy test between a central disk of amoxicillin + clavulamic acid and the C3G disks. The ATCC25922 *Escherichia Coli* strain has been chosen as reference for the internal quality control of enterobacteria. The characteristics of the patients concerned by the urinary infections have been described based on the data provided by the Microbiology Unity at the University Clinics of Lubumbashi. These data and the results given the bioassays have been processed by means of the Epi Info 3.5.3 software.

3 Results

The results discussed in the present survey are related both to the description of the patients' characteristics and the isolated bacterial strains versus their morphological characters. They are also related to tests of the sensitivity of the uropathogenic enterobacteria towards a set of antibiotics of our choice.

3.1 Characteristics of the Population concerned by the Urinary Infections

The processing of the data provided by the Service of Microbiology has given the results depicted in Figure 1.

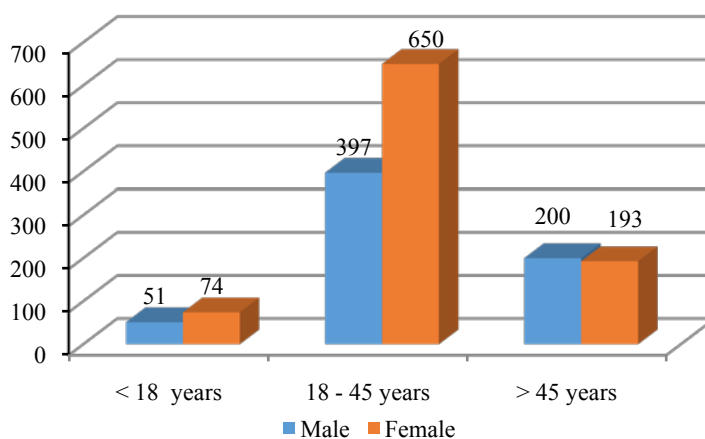


Figure 1: Characteristics of the population concerned by the urinary infections

With the highest occurrence of 59% among the target population, the urinary infections are pathologies that meanly affect the women aged between 18 and 40 years. At first instance, their great occurrence among the younger women can be associated to the beginning of their sexually active period next to puberty. It can also be the consequence their more sexually active period of life leading to menopause. This high occurrence of the urinary infections among women may also arise from prostitution, which unfortunately remains a problem of great concerns on the public health standpoint in the city of Lubumbashi along with the use of water of contaminated from the rivers and wells by the dwellers for the bodily care.

3.2 Bacterial Strains Description versus their Morphologic Characters

A set of antibiotics commonly used in the city of Lubumbashi to treat the urinary infections has been tests against the bacterial strains of our focus in view evaluating their efficacy. The morphological characteristics of the studied bacteria are given in Table 1.

Table 1: Bacterial strains versus the morphologic character

Bacterial strains	Number (Frequency)	Proportion (%)
Enterobacteria	1222	78.1
<i>Escherichia coli</i>	701	57.4
<i>Klebsiella sp.</i>	272	22.3
<i>Klebsiella pneumoniae</i>	114	9.3
<i>Proteus sp.</i>	70	5.7
<i>Proteus mirabilis</i>	33	2.7
Other enterobacteria	32	2.6
<i>Pseudomonas aeruginosa</i>	233	14.9
Positive gram Cocci	110	7.1
<i>Staphylococcus aureus</i>	70	4.5
<i>Enterococcus faecalis</i>	40	2.6
Total	1565	100

Enterobacteria are the predominant strains responsible for the urinary infections since they represent about 78% of 1222 uropathogenic bacteria isolated through urinoculture from 3694 patients. *Escherichia Coli* obviously is the main bacterial strain (57.4%) among the isolated uropathogenic microbes. Based on the high (65-85%) isolation frequency of *Escherichia Coli* reported by many researches [7, 2, 3, 8, 9, 10]; and our results, it can be stated that *Escherichia Coli* is undeniably the bacteria more implicated in the urinary infections. It is followed by *Klebsiella sp* (22.3%), which belongs to the second important subgroup of bacteria responsible for urinary infections with a varying isolation frequency [9]. *Klebsiella pneumoniae* (9.3%), *Proteus sp* (5.7%) and *Proteus mirabilis* (2.7%) belong to the less predominant subgroups of bacterial strains provoking the urinary infections. However, it is worth stressing on the fact that in the present survey, *Proteus sp.* ranks in the third position. Our results are supported the paper of Mahamat et al. [4] where an isolation frequency varying between the third and sixth ranks depending on the type of populations has been suggested. As for the gram-positive bacteria observed among the isolated bacterial strains, they are due to the ascending physiopathology of the

urinary infections. As the matter fact, it starts from the urethral flora due to colonisation of the perinea by enterobacteria of the digestive origin along with the presence of specific factors of uropathogenicity namely: adhesines and urease [2, 5].

3.3 Sensitivity of the Uropathogenic Enterobacteria towards Antibiotics

The results given by tests of sensitivity of the bacterial strains implicated in urinary infections are depicted in Figure 2.

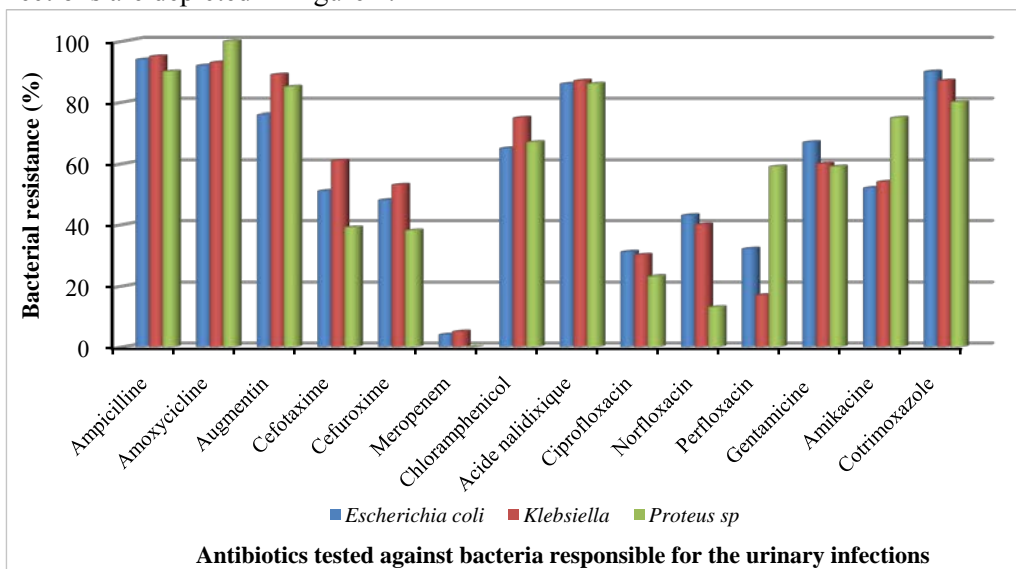


Figure 2: Sensitivity of bacterial strains towards the tested antibiotics

Concerning the resistance towards the tested antibiotics, the studied bacterial strains classify as follows: *Klebsiella* sp, *Escherichia Coli* and *Proteus* sp. Indeed, the highest resistance (> 85%) shown by *Klebsiella* sp is the highest of all in 50% of the cases. This resistance is mainly oriented towards Ampicillin (95%), Amoxicillin (93%), Augmentin (89%), Nalidixic acid - C₁₂H₁₂N₂O₃ (87%) and Cotrimodazole (87%). *Escherichia Coli* and *Proteus* sp have strongly resisted against the tested antibiotics respectively in nearly 29 and 21% of the cases. The highest resistance shown by *Escherichia Coli* is mainly oriented towards Ampicillin (94 and 90%), Amoxicillin (92 and 100%), Cotrimodazole (90 and 80%) and Nalidixic acid - C₁₂H₁₂N₂O₃ (86%). The more sensitive bacterial strain towards the tested antibiotics is *Proteus* sp since it has showed a poor resistance in nearly 57% of the cases especially towards Meropenem (0%), Norfloxacin (13%), Ciprofloxacin (23%), Cefuroxime (38%) and Cefotaxime (39%). It is followed by *Escherichia Coli* due to its resistances in 29% of the cases towards Meropenem (4%), Ciprofloxacin (31%), Perfloxacin (32%) and Cefuroxime (48%).

On average, the resistance showed by the studied bacterial strains towards the tested antibiotics varies around 59%. *Escherichia Coli* and *Proteus* sp are ordinary sensitive towards amoxicillin, which is a beta-lactamin. However, this antibiotic cannot be recommended for the urinary infections considering the high resistance (92%) shown by *Escherichia coli* and *Proteus* sp in the present survey comparatively to the threshold given by multicentre studies achieved in Tunisia (62.5%) [11], America (37%) [12] and Europe (53- 60%) [3, 6, 9, 13, 14, 15]. Based on our results, Meropenem appears to be the most efficient antibiotic against the urinary infections. Meropenem can be prescribed instead of

amoxicillin due to the lowest resistance (3%) shown by *Escherichia Coli*, *Klebsiella* and *Proteus sp.* On the efficiency standpoint, Meropenem is followed by Ciprofloxacin, Norfloxacin, Perfloxacin, Cefuroxime and Cefotaxime. The average resistance shown by the studied bacterial strains towards these antibiotics ranges from 28 to 50%. Ciprofloxacin, Norfloxacin and Cefuroxime are efficient when used against *Proteus sp* contrarily to Perfloxacin, which is more efficient against *Klebsiella*. However, it is worth mentioning that the resistance towards Ciprofloxacin averages 28% and surpasses the threshold published in the papers from Tunisia (14%) and Europe (3- 12%) [3, 8, 11, 13, 14].

This emerging resistance is thought to arise from a first intention probabilistic treatment using a fluoroquinolone-type antibiotic along with self-medication. As for Cefotaxime, it remains the more active antibiotic used against enterobacteria in spite of the increased resistance shown by *Escherichia Coli* (51%) and *Klebsiella* (61%) when compared to figures (3.7 and 13.8%) released by studies achieved in Tunisia [2, 11] and Europe [13, 15]. The loss in efficiency of Cefotaxime observed in the present study may be explained by the continuously increasing frequency of strains producing the betalactamase. Additionally, *Escherichia Coli* has shown a great resistance (86%) towards Nalidixic acid, which is an association of Amoxicillin and Clavulanic acid. This resistance surpasses the one observed (7-14%) in studies achieved in Europe [8, 13, 15]; and is due to the large use of these antibiotics as well as their determinism genetic which gives them a great power of dissemination. The above-mentioned resistance is also associated to the emergence and to extension of mechanisms of resistance relying on the secretion of beta-lactamins, which hydrolyze the betalactame molecules nucleus [15]. Other researchers have shown that the resistance against Nalidixic acid is frequently accompanied by the decreased sensitivity towards the fluoroquinolone-type antibiotics. The bacterial strains concerned by this behavior are mutants with low resistance that can develop into strains more resisting towards Ciprofloxacin and Nalidixic acid under the stress of selection exerted by fluoroquinolones [2, 5, 16]. That is why the surveillance of the more resisting bacterial strains is based on detection of resistance towards Nalidixic acid. In the present study, the strains of *Escherichia Coli*, *Klebsiella* and *Proteus sp* that have resisted towards Nalidixic acid on average at 86% were found to be also sensitive to Ciprofloxacin in 54 up to 63% of the cases. These results incite us reinforcing the surveillance and sensitization. Additionally, the studies achieved in France with regard to the increased incidence towards Fluoroquinolones have related it to the misuse along with their use in prophylaxis [6, 17, 18, 19]. Based on our results, it can be stated this phenomenon arises from the clinical customs of prescription, which is based in our region on a large use of Fluoroquinolones. Considering the efficiency of the studied antibiotics, one can classify them as follows: Meropenem, Ciprofloxacin, Norfloxacin, Perfloxacin, Cefuroxime, Cefotaxime, Amikacine, Gentamicine, Chloramphenicol, Augmentin, Cotrimoxazole, Acid nalidixic, Ampicilline and Amoxyciline.

5 Conclusion

Considering that *Escherichia Coli*, *Klebsiella sp* and *Proteus sp.* are responsible for the urinary infections on the one hand and their sensitivity towards the tested antibiotics on the other, it can be recommended to physicians to prescribe to the patients Cefotaxime or Cefuroxime in the early stage of the illness. In the case of resistance, they can use the

more efficient antibiotics namely Perfloracin, Norfloxacin and Meropenem depending on the resistance opposed by bacterial strains involved in the urinary infections based on antibiograms. As for Amikacin, Gentamicin, Chloramphenicol, Augmentin, Cotrimoxazole, Ampicilline and Amoxycycline, they cannot be recommended in the case of the urinary infections due to their low efficiencies (< 50%) against the studied bacterial strains which appear to be more resisting. These increased resistances shown by the uropathogenic enterobacteria towards a number of antibiotics commonly used in the city of Lubumbashi are related to the practice of auto-medication and the misuse of drugs by the patients. As the matter of fact, the use of the same type of drugs is inducing a pressure of selection leading to the emergence of the resistances of the enterobacteria responsible for urinary infection towards antibiotics. The increasing doorsteps of resistance opposed by the uropathogenic enterobacteria towards the commonly used the antibiotics requires a strengthened surveillance, to amend the prescriptions by the physician and sensitization of the community. The sensitivity of the pathogenic microbes towards antibiotics needs to be interestedly studied. The adoption of stringent steps to discourage to anarchic trade of antibiotics and the establishing of the guidelines for their prescriptions by the physicians in the city of Lubumbashi are suggested among strategies that can provide the best solutions to the relevant issue of the increased resistance shown by bacteria responsible for urinary infection towards the commonly used antibiotics.

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