

Original article:

Prevalence and isolation frequency of pathogenic strains responsible for some infections in Bechar's community (Southwest of Algeria): About 1 458 cases.

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Abstract:

Background: The microbial infections represent a real public health problem with considerable individual and economical consequences. Their surveillance has become, in recent decades, an essential element of any program to control these infections. **Aim:** In this study, we estimated the prevalence and frequencies of some microbial infections amongst the Bechar's community (Algeria). **Methods:** The microbiological analyzes of various pathological samples namely : urine and vaginal discharge, pus, nipple discharge, originally sampled from throat, otitis discharge and effusion liquid, have been carried out in the medical analysis laboratory of Bechar for seven months. **Results:** The obtained results showed that out of 1458 samples, 506 cases were positive (34.7 %), for some infections, there is a relationship correlation between the infections' frequency and their prevalence with the age and gender of patients, as factors; which the female gender presents 74.7 % of positive cases, a sex-ratio less than 1; besides, the most affected population is aged between 25 and 39 years old. The isolated microbial species, which are the most incriminated agents responsible for the studied infections, were mainly *Escherichia coli* with an isolation frequency of 87.15 %, followed by coagulase-positive *Staphylococci* CoPS (5.13 %), *Streptococcus sp* (3.16 %), fungal species *Candida albicans* (1.58 %), Coagulase-negative *Staphylococci* Co NS (1.18 %). However, less than 2 % of infectious agents were recorded for the bacterial strains *P. aeruginosa*, *K. pneumoniae* and *Enterococcus sp*. **Conclusion :** This study can provide a platform for an effective prevention strategy and can be useful for epidemiological prediction with a decision-making.

Keywords: Infections; Pathogenic strains; Isolation frequency; Occurrences' factors; Bechar (Algeria).

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Introduction

Infection is the result of complex interactions between the patient's defense mechanism, the site of intervention and the microorganisms. It is a microbial proliferation resulting in cellular, tissue or general reactions, and most often, with an inflammatory syndrome¹.

A pathogenic bacterium is the basis of many

infectious diseases for humans especially in developing countries. In 1995, infectious diseases were responsible for a third (17 million people) of deaths worldwide².

The pathogenicity of a bacterium is due to its ability to invade tissues by resisting host defenses and multiplying (virulence). It may also be due to the ability of the germ to secrete a toxin, it is a

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macromolecule endowed with a toxic action in humans (example: diphtheria toxin and tetanus), it is called toxicity³.

In 2014, British Prime Minister *David Cameron* was solemn. He said: “*If we fail, we will face a scenario where antibiotics will have no effect, referring us to the Middle Ages of medicine*”. In the process, the United Kingdom announced the creation of a 228 million euros fund. In January 2015, it was the US President *Barack Obama* who announced a doubling of federal funding for the plan to fight the bacterial resistance and to improve the research on new antibiotics.

In May 2016, the US health officials did not hide their excitement after the discovery. In a patient suffering from a urinary tract infection, of a mutant strain of *Escherichia coli* ‘*E.coli*’, which became resistant to all medications, Dr. *T. Frieden*, director of the center for disease control and prevention warned “*We risk going back to a pre-antibiotic world*”⁴.

The development of new antibiotics is a public health emergency. This is the message issued on Monday, February 27 (2017) by the World Health Organization (WHO) that, for the first time, issued a “priority pathogens” list with antibiotic resistance.

This group includes the following bacteria: *Acinetobacter*, *Pseudomonas* and various *Enterobacteriaceae* (including *Klebsiella*, *E. coli*, *Serratia* and *Proteus*). The WHO says “They can cause severe infections, often fatal, such as blood infection and pneumonia”. The organization states that these bacteria have become resistant to many antibiotics; including those (third generation of carbapenems and cephalosporins) intended to treat multidrug-resistant bacteria.

The other two groups in the list include other bacteria becoming more resistant, causing more common diseases such as: gonorrhoea or food poisoning by salmonella⁵. This WHO initiative was discussed at a meeting of G20 health experts in March 2017 in Berlin⁴.

To this end, this study was conducted at the level of Dr. Benchaib's medical analysis laboratory, which aims to estimate the isolation frequency of pathogenic microorganisms responsible for different infections in Bechar's community (Algeria).

Materials and Methods

This experimental study aimed to specify the nature of microorganisms responsible for different

infections in *Bechar* community; their prevalence and isolation frequency, and how they related to the patients' gender and age as factors. A total of 1458 pathological specimens were analyzed at the level of Dr. Benchaib's medical analysis laboratory in *Bechar* (Algeria), during seven months, for the period from January 1st to July 31st (2017). The infections' occurrences are also reported in the studied community.

Specimen collection and microbiological analysis

The sampling concerns the following pathological products: urine and vaginal discharge, pus, nipple discharge, originally sampled from throat, otitis discharge and effusion liquid.

All samples were microbiologically screened by the isolation method on agar medium⁶. The selective isolation and enrichment culture media used (Lioflichem Diagnostici, Italy) are as follows: Mannitol Salt Agar, Hektoen Enteric agar medium, Blood agar base, Sabouraud-Chloramphenicol agar medium, Esculin azide bile agar, Cetrimide agar, King A & King B agar, nutrient broth, and sterile physiological water (0.9% NaCl) as diluents. Identification of the isolates was performed according to standard microbiological methods⁷. Firstly, identification by macroscopic examination of colonies' phenotypic and microscopic characterization by fresh observation between slide and coverslip, and after differential Gram staining; secondly, identification of biochemical characteristics by means of Classical biochemical gallery on Triple Sugar Iron agar, Mannitol-Mobility test medium, Meat-Liver glucose 0.6% agar and Simmons Citrate agar, IMViC test led to distinguish four (4) characters used to differentiate within coliforms (*Escherichia coli* and *Klebsiella pneumoniae*), Cytochrom C Oxidase assay, catalase assay, Staphylocoagulase assay, and miniaturized biochemical test system using API 20 E, API Staph (BioMerieux, France).

Statistical analysis

The statistical analysis was performed by calculating the means and percentages relating to the isolations frequencies of microorganisms using Excel software, which are translated into graphs in the form of histograms and circles.

Ethical clearance

This study meets the ethical requirements given by the national ethics council on health sciences and the local ethics committee of the Faculty of life and

Natural Science ‘Mohammed TAHRI University of Bechar (Algeria)’, according to which, the anonymity and confidentiality were respected for all information revealing the patient’s identity. The declared information was only limited by patient’s age and gender. Informed consent was obtained from all patients before participation

Results

Characterization of patients

Number of positive / negative cases

According to the obtained results, there were 1 458 examinations carried out at the laboratory level, 506 samples were positive and 952 samples were negative, representing a rate of 34.7 and 65.3 % respectively (Figure 1).

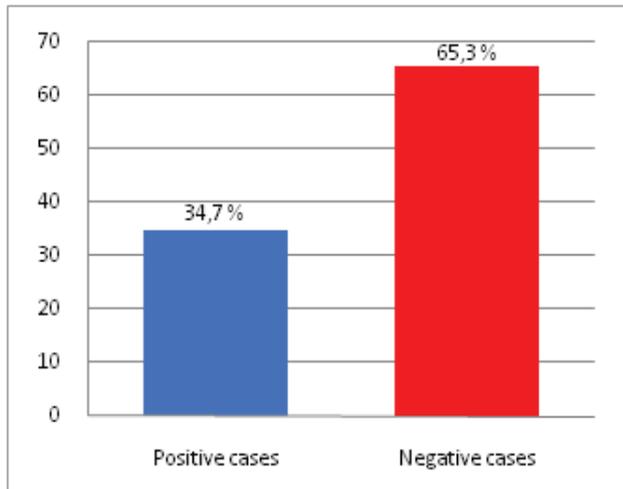


Figure 1: Percentage of positive and negative bacteriological tests.

Patients’ age distribution

The analysis of the results shows that 33 % of patients (n: 167) who aged between 25 and 39 years old are the most affected by infections compared to other ages as shown in the figure below (Figure 2).

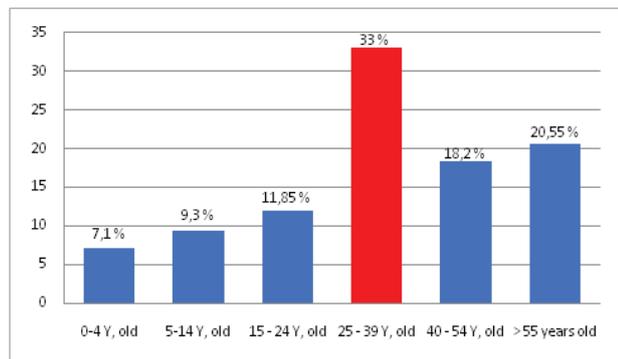


Figure 2: Patients’ distribution by age group.

Patients’ distribution by gender

The results show a female predominance (n: 378) that appears to be the most affected portion by infections where the calculated sex-ratio was 0.33 (Figure 3).

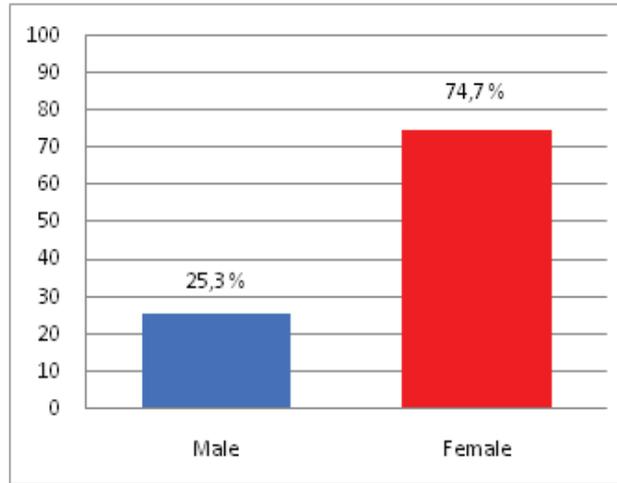


Figure 3: Patients’ distribution by gender.

Characterization of isolated microorganisms

Isolates’ frequency of microbial strains

Escherichia coli was the most isolated species, and likely to be the most incriminated germ in different types of infections with a percentage of 87.15 % (n: 441 strains), then *Staphylococcus aureus*, *Streptococcus sp* and *Candida albicans* with an isolation rate of 5.13; 3.16 and 1.58 % respectively (Figure 4).

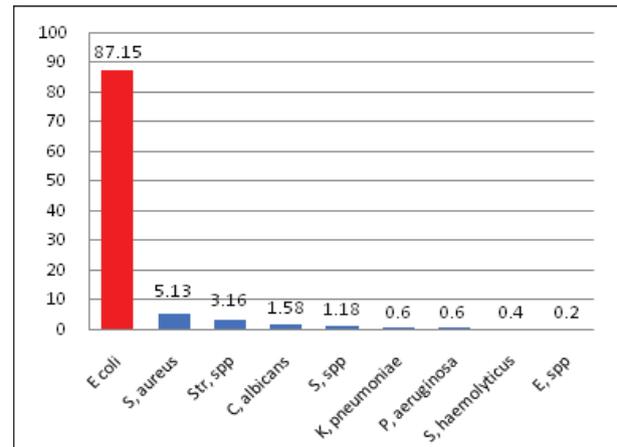


Figure 4: Isolation frequency of pathogenic microorganisms.

Germs’ distribution by microbial groups

Gram-negative bacilli represents more than three-quarters (n: 447 strains) of all isolated microorganisms, 88.35 %, while Gram-positive Cocci were 10.07 % (n: 51 strains). *Candida albicans* as fungal strain represents 1.58 % of isolated microbial species (Figure 5).

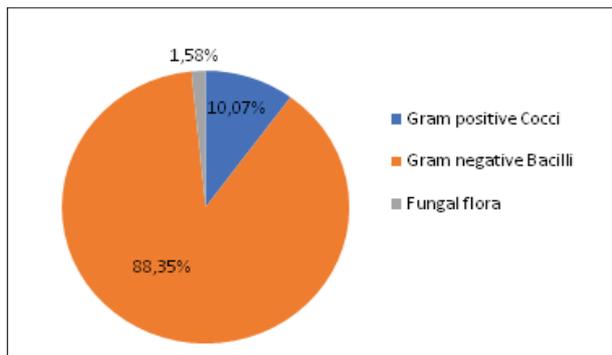


Figure 5: Distribution of isolated microorganisms' according to the microbial groups

Infection types and pathological samples analysis
Urinary tract infection

There is a female dominance in patients with urinary tract infection 'UTI'. Out of 372 urine cytobacteriological exam 'UCBE', 270 samples were positive for female patients (72.6 %) (Figure 6). The calculated sex ratio was 0.37.

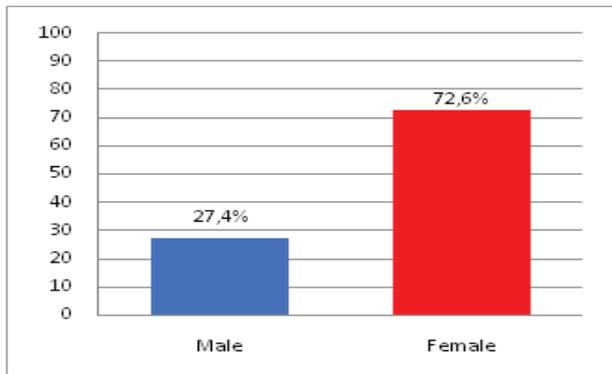


Figure 6: UTI patients' distribution by gender.

The species of *Escherichia coli* was the most isolated strain with a frequency rate of 97.6 % (n: 363 strains), whereas the microbial strains *Candida albicans*, *Staphylococcus sp* and *Klebsiella pneumoniae* represent a rate inferior to 2.5 % (Figure 7).

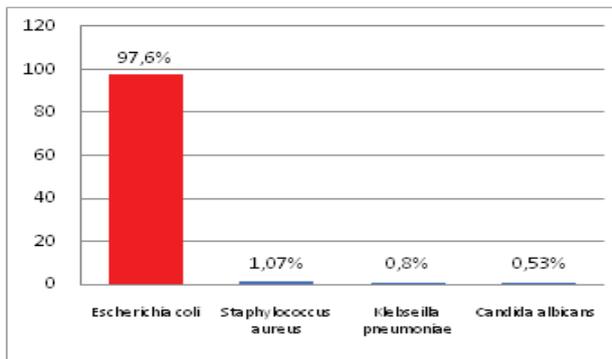


Figure 7: Isolation frequency of strains responsible for UTI.

Nipple discharge

The species of *Staphylococcus aureus* was the only strain isolated by microbiological exam of nipple discharge 'breast infection'.

Effusion liquids

The microbiological exam of the effusion fluids was represented by only three cases of male patients where *Escherichia coli* strain was the only isolated strain.

Microbiological exam of otitis

The otitis exam results are very close for both genders, where the sex ratio was 1.16 (Figure 8).

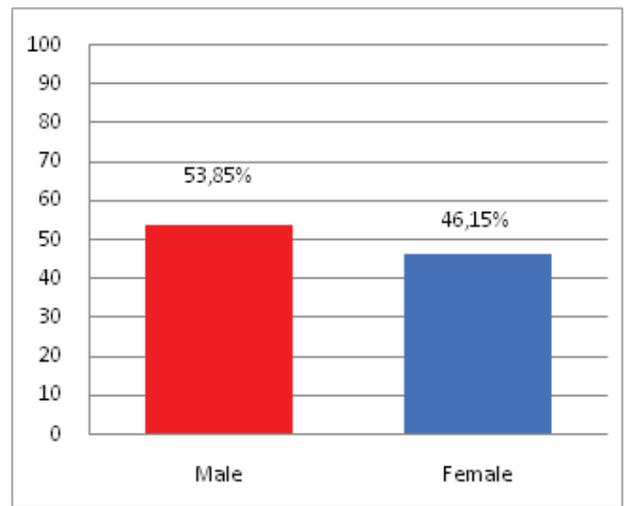


Figure 8: Otitis patients' distribution by gender.

The strains of *Staphylococcus aureus* were predominantly isolated with a rate of 76.92 %, followed by *Pseudomonas aeruginosa* and *Streptococcus sp* with an isolation rate of 15.35 and 7.7 % respectively (Figure 9).

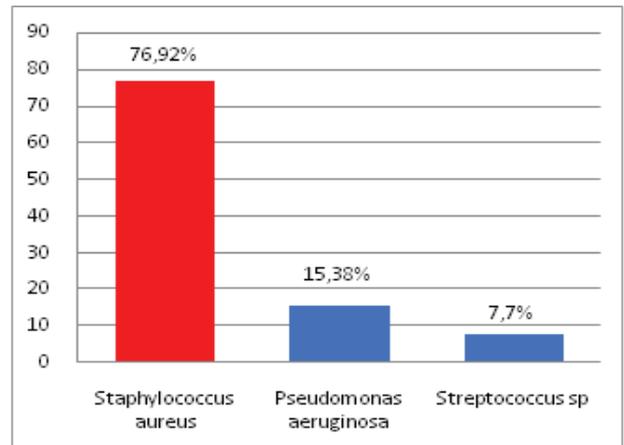


Figure 9: Isolation frequency of microorganisms responsible for otitis.

Analysis of throat specimens

For the throat examination, we observe that the female gender represents the majority of the analyzed cases (3/4), in which the sex-ratio was 0.33 (Figure 10).

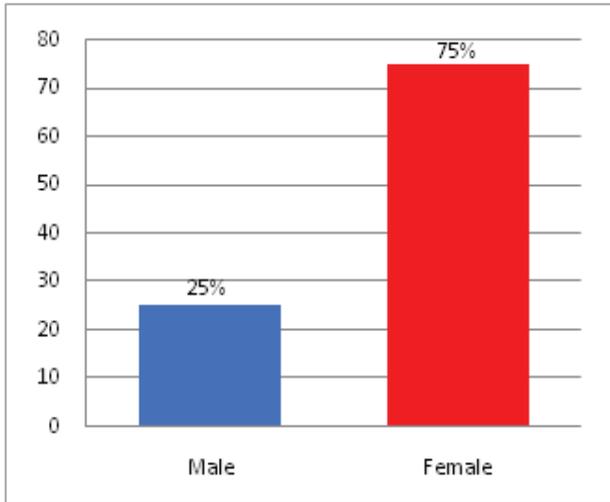


Figure 10: Patients' distribution by gender for throat specimens' analysis.

The microbiological analysis allowed isolating *Streptococcus sp* and *Staphylococcus haemolyticus* with an isolation rate of 87.5 and 12.5 % respectively (Figure 11).

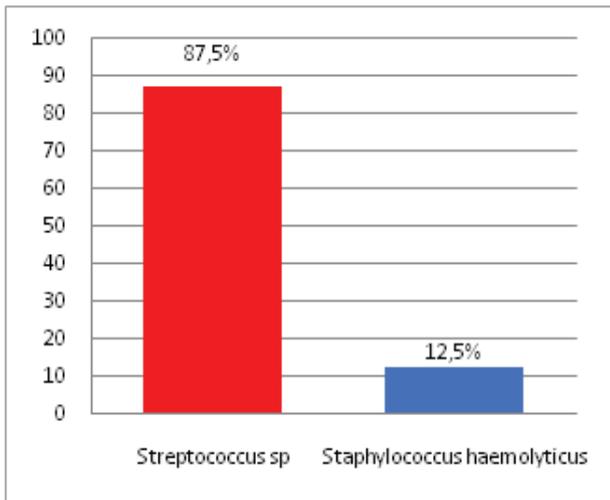


Figure 11: Isolation frequency of throat microbial pathogens

Pus samples analysis

For the bacteriological exam of pus, with a set of 14 cases, the male gender was dominant over the female where the calculated sex ratio was 1.8 (Figure 12).

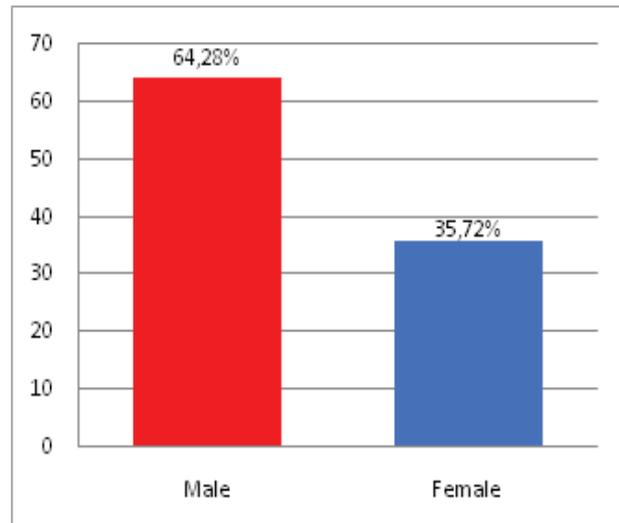


Figure 12: Patients' distribution by gender for pus analysis.

For isolated species, *Staphylococcus aureus* was the dominant strain with an isolation rate of 64.28 %, followed by *Escherichia coli* 28.57 %, then *Pseudomonas aeruginosa* at 7.15 % (Figure 13).

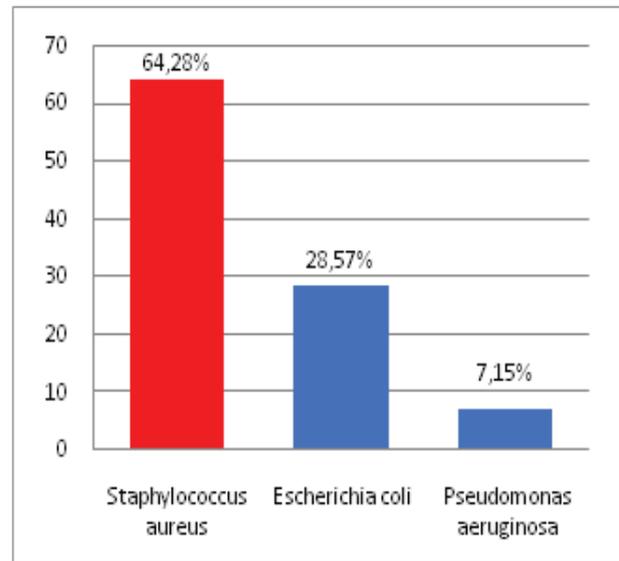


Figure 13: Frequency of isolated strains from pus specimens

Bacteriological exam of vaginal discharge

The analysis showed that *Escherichia coli* strains were the most isolated microbial strains with an isolation rate of 87.65 %, followed by fungal strain *Candida albicans* 7.41 %, *Staphylococcus sp* 2.46 %. Then, only 1.24 % for *Streptococcus sp* and *Enterococcus sp* strains respectively (Figure 14)

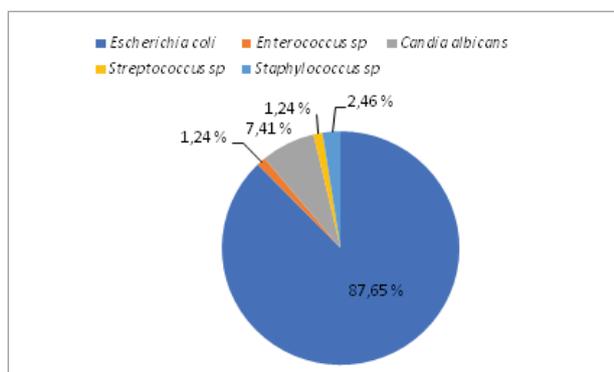


Figure 14: Isolation frequency of strains responsible for women's genital infection.

Occurrence rate of infections

Urinary tract infection ranks first among the various infections, with an occurrence rate of (73.51 %), followed by genital infection (16.6 %), otitis (5.73 %), and skin infection (2.76 %). Finally, breast and serous infection does not exceed (1.5 %) (Figure15).

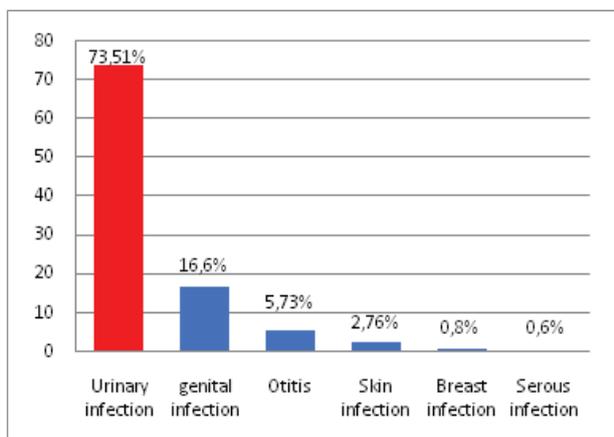


Figure 15: Prevalence rate of infections in Bechar's community.

Discussions

Based on our results, urinary tract infection is among the most common bacterial infections with an occurrence rate of 73.51 % of the infections cases analyzed; it is caused mainly by *Enterobacteriaceae*, where *Escherichia coli* represent from 70 to 80 % of isolated microorganisms.

It is noted that women are most likely to capture these infections, especially women over 25 years of age. This high urinary infection rate in women can be explained by two factors that play a major role; one at the beginning of the sexual activity and the other in the post menopausal period. These identifications occur more frequently in women than in men due to anatomical and physiological factors specifically

favoring the installation of the pathogenic germs (Urethra short, pregnancy ...), and secondly, by the fact that the latter are more enumerated by the number of urine cyto-bacteriological exam requested from women⁸⁻⁹.

Genital infections take second place in this study, being (16.6 %) of the analyzed cases, where most cases were dominated by female gender, followed by otitis with an occurrence rate of 5.73 %, and finally, skin infection, breast infection and serous infection with an occurrence rate inferior to 5 %. Thus, that the most affected population is aged between 25 and 39 years old.

In the literature, several studies argue the dominance of *E. coli*, *Staphylococcus sp* and *Streptococcus sp* as the most frequently isolated species responsible for urogenital infection (UGI), namely the study of Lemort et al.¹⁰ which shows the relationship of this bacterium with the physiology of urinary tract infection that is usually ascending, and there is a strong colonization of the perineum by digestive origin enterobacteria, and in particular *E. coli*. They added specifically uropathogenicity factors, for example, *Escherichia coli* which has adhesins, capable of binding the bacterium to the urinary epithelium and preventing its elimination by bladder emptying¹¹.

The obtained results are different from those obtained in a previous study on the same region showing 111 patients with UGI which the species *Staphylococcus aureus* was the agent responsible for more than 36 % (40 cases) of UGI, followed by coagulase-negative staphylococci (CoNS) and *Candida albicans* with a rate of 23.42 % (26 cases) and 21.62 % (24 cases) respectively. A rate of almost 10 % (11 cases) was revealed for *Escherichia coli*, less than 3 % (4 cases) for *Streptococcus agalactiae* and *Trichomonas vaginalis*. However, the other infectious agents had a lower level (< 2 %) where the isolated species were "*Pseudomonas aeruginosa*, *Serratia sp* and *Citrobacter sp*"¹²

A retrospective study conducted by Chedi et al.¹³ at Aminu Kano Teaching Hospital, northwestern Nigeria on 123 patients with positive cyto-bacteriological exam of urine between December 2004 and July 2005 where 58 % of cases were females. They found that *E. coli* was the predominant cause of urinary tract infection 39.8 %, followed by *Proteus sp* (26 %), *Klebsiella sp* (21.1 %), *Enterococcus faecalis* (7.3 %), *Citrobacter sp* (3.3 %), *Staphylococcus sp*

(1.5 %) and *Pseudomonas sp* (0.8 %).

The retrospective study of Sangare¹⁴ at the Gyneco-Obstetrics Department of the Gabriel Touré Teaching Hospital of Bamako from January 1st, 2006 to December 31st, 2006, on 106 patients with a positive cyto-bacteriological exam of urine on pregnant women showed a predominance by *E. coli* (34.9 %), followed by *Klebsiella pneumoniae* (24.5 %), *Staphylococcus aureus* (20.7 %), *Streptococcus pneumoniae* (4.7 %), *Serratia sp* and *Enterobacter cloacae*, *Citrobacter freundii* and *Pseudomonas aeruginosa* (2.8 %), *Acinetobacter baumannii*, *Klebsiella oxytoca*, Coagulase positive *Staphylococci* and Group A of *Streptococci* (0.9 %).

The study of Chemlal et al.¹⁵ conducted in the nephrology department at Oujda regional hospital in eastern Morocco on 43 patients (40 adults and 3 children); 60.4 % were females. The distribution of isolated organisms was dominated by *E. coli* (58.3 %), *Klebsiella sp* (29.2 %). *Candida albicans* (4.2 %), and *Streptococcus sp* (4.2 %) of cases.

According to the study conducted by Kpoda et al.¹⁶ on the susceptibility of extended spectrum beta-lactamase producing *Enterobacteriaceae* causing urinary tract infections in Ouagadougou, Burkina Faso, a total of 324 isolates of *Enterobacteriaceae* were identified during the study period, including 211 (65 %) *E. coli*, 75 (23 %) *Klebsiella spp.*, 18 (6 %) *Enterobacter spp.*, 11 (3 %) *Proteus spp.*, 5 (2 %) *Citrobacter spp.*, and 3 (1 %) for *Serratia spp.*

Urinary tract infection is the most common nosocomial infection. It represents up to 40 % of all infections acquired in the hospital¹⁷, where the risk factors for nosocomial UI were the female gender; the urinary catheterization and the historical antecedent of repeated UTIs. However, the study conducted in Nigeria by Onifade et al.¹⁸ looked into profile of bacterial pathogens associated with nosocomial infections in hospitals within Makurdi metropolis where 71 bacterial pathogens were encountered from 240 analyzed samples showed that nosocomial pathogenic bacteria, in particular *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Staphylococcus spp* and enteric bacteria, *Escherichia coli* and *Klebsiella spp* are the predominant pathogens associated with infection acquired in hospital environment.

Antimicrobial resistance is described as an intuitive event in which microbes develop their ability to resist against any threat, including antimicrobials,

so to survive. According to Haque¹⁹, where a recent study revealed that antibiotic consumption had been increased around 39 % in 76 countries from 15 years' time (2000-2015), especially observed in low and middle-income countries. According to Gharamaleki et al²⁰, nationality could be regarded as a risk factor of multidrug resistance (MDR)

An earlier study on urogenital infections within the same community, the resistance of strains involved in urogenital infection against antibiotics is relatively high for some molecules; including the penicillin family (penicillin, oxacillin, ampicillin and amoxicillin + clavulanic acid), sulfamide (cotrimoxazole) and Macrolides (Erythromycin) for *E. coli* and *S. aureus* species. The last strain is a urinary pathogen, and once colonized the urinary tract, could be a source of future staphylococci infection. However, carbapenems, cephalosporins third-generation (C3G), aminoglycosides (amikacin, gentamycin), glycopeptides (vancomycin), phenicol (chloramphenicol), fosfomycin, amoxicillin and other molecules have an antimicrobial activity switched between moderate and good effect²¹.

However, through many published studies warned that there was not much time left to observe *Escherichia coli* and that *Klebsiella pneumoniae* would be resistant to third-generation cephalosporins and carbapenems, all over the planet. Probably, one of the involved major causes in the antimicrobial resistance is the unnecessary use of antibiotics either self-prescribed, or prescribed for in- or out-patients or for patient satisfaction. An antibiotic management program, regulatory measures and promotion of safe use of the antimicrobial molecules in health professionals could helpful to minimize the development of resistant microorganisms¹⁹.

As an Oto-rhino-laryngology infection at the 3rd place behind urinary and genital infections, otitis has an occurrence rate higher than 5 %. According to Carroll and Reimer²², the etiological agents responsible for the cases' majority of acute otitis media are the most common; *Streptococcus pneumoniae*, untyped *Haemophilus influenzae*, *Moraxella catarrhalis*, Group A *Streptococci* and *Staphylococcus aureus*. These data corroborate the obtained results in this study.

The examination of nipple flow has shown a predominance of *Staphylococci* strain whose low exam frequency limits the diversity of isolates. These accords with the literature, where the most

commonly implicated organisms are *Staphylococcus aureus*, *Streptococci*, anaerobes (*B. fragilis*, *Peptostreptococcus*) and enterobacteria²³.

According to Le Floch et al.²⁴, many infections are cutaneous. If the surface's bacteria are destroyed at the time of the burn, those located at depth (cutaneous appendages) are at least partly respected. The burn, sterile in the first hours, is rapidly colonized, initially (within 48 hours) by cutaneous bacteria (mainly Gram-positive cocci) and then (at the end of the first week) by bacteria that may be of digestive, Oto-rhino-laryngology or environmental origin as well as fungi.

According to the various consulted studies, we reveal a model that is repeated and a almost similarity concerning the obtained results, an infectious dominance that affects the female gender with a defined age range for uro-genital infections, and then come the other studied infections. The microbial dominance is revealed especially for *Staphylococcus spp* and *Pseudomonas aeruginosa*, the enteric bacteria *Escherichia coli* and *Klebsiella spp* as pathogenic strains responsible for different types of infections.

Conclusion

This work contributed to the study of the frequency and occurrence of infections in Bechar's community by 1458 pathological samples' analysis during seven months for the period from January 1st to July 31st (2017), which 506 cases were positive representing a rate of 34.7 %.

The case of identified infections including those of urogenital origin occur more frequently in women than in men with a rate of 74.7 %, where the most isolated species was *E. coli* with an isolation rate of 87.15 %.

Knowing that the power of these microorganisms to develop resistance to the antimicrobial agents (antibiotics), measures to prevent and control the consequences of this resistance in hospitals must act on the factors that determine its emergence and evolution.

Finally, we can say that uro-genital infection is the

most common infection in Bechar's community. Factors such as age and gender favor the spread of different infection types, where the most affected population are between 25 to 39 years old. The prevention is essential, but if the individual is infected, the patient must consult a doctor promptly for treatment in order to avoid unwanted complications. In addition, prevention of cross-transmission and reduction of selection pressure through rational use of antibiotics could be the two essential components that constitute a public health priority. Unfortunately, according to several authors, despite being the rational or irrational antimicrobials drug consumption, the microbial resistance cannot be avoided. However, the obtained results could help medical teams and pharmacists in both public and private sectors as well as decision-makers to understand the infections distribution, especially in Bechar's community, and therefore to improve the local health program.

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Declaration of interests

The authors declare having no conflicts of interest, financial or otherwise in relation to this article.

Authors' contributions

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Data gathering: E. Benyagoub, S.M Benchaib

Writing and submitting manuscript: E. Benyagoub

Editing and approval of final draft: E. Benyagoub, F. Bendada, S.M Benchaib, A. Zaalán

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