ANTIMICROBIAL RESISTANCE PATTERNS OF *PROTEUS* ISOLATES FROM CLINICAL SPECIMENS

Saleh A. Bahashwan
Pharmacology and Toxicology Department, College of Pharmacy, Taibah University, Al Madinah Al Munawara, KSA
Hatem M. El Shafey
Respiratory Therapy Department, College of Medical Rehabilitation Sciences, Taibah University, Madinah Munawarah, KSA
Microbiology Department, National Center for Radiation Research and Technology, Nasr City, Cairo, Egypt

Abstract

Total of 6840 clinical samples were collected from King Fahd Hospital at Medina, Kingdom of Saudi Arabia (KSA). Clinical samples were screened for *Proteus* spp. It is found that *Proteus* spp isolates representing about 3% of all positive samples isolated from different clinical sources. Males were found to be more vulnerable than females in acquiring *Proteus* infections. Results showed that the greatest number of *Proteus* spp. isolates from clinical specimens were isolated from wound and sputum swabs specimens representing about 88% of all clinical specimens.

Thirteen different antibiotics representing different families of antibiotics were tested on *Proteus* spp. specimens for studying its antimicrobial sensitivity pattern. Results of antimicrobial sensitivity tests revealed that imipenem (IMP) antibiotic was the most effective antibiotic against *Proteus* spp. with 91% of antimicrobial sensitivity. Imipenem (Imp) was followed by amikacin (AK) with 61% of antimicrobial sensitivity.

Results are recommending prescribing of imipenem (IMP) antibiotic in the treatment of *Proteus* spp infections. It is recommended also to prescribe amikacin (AK) in the case of sensitivity to imipenem (IMP). In acute cases, *Proteus* infections could require use of combined antimicrobial therapy (imipenem- amikacin). Results also demonstrated that percentage of *Proteus* spp. infections was highest during summer season representing about 40% of infections all year along. Summer season was followed by winter season with about 32% of infections.

Keywords: *Proteus*, Enterobacteriaceae, Antimicrobial susceptibility, antibiotic resistance, multi-drug-resistance

Introduction

Proteus is a genus of Gram-negative bacteria belonging to the family of *Enterobactericeae. Proteus* species are distinguishable from most other genera by their ability to swarm across an agar surface (Jacobsen et al., 2008). *Proteus* is widespread in the environment and makes up part of the normal flora of the human gastrointestinal tract. *Proteus* ranks third as the cause of hospital-acquired infections (Stamm, 1999*).* Three species: *P. vulgaris, P. mirabilis,* and *P. penneri* are opportunistic human pathogens (Guentzel, 1996).

Proteus species are the major cause of diseases acquired outside the hospital, where many of these diseases eventually require hospitalization (De Champs et al, 2000). *P. mirabilis* causes 90% of *Proteus* infections. *Proteus species*, particularly *P. Mirabilis,* is believed to be the most common cause of infection-related kidney stone, one of the most serious complications of unresolved or recurrent bacteruria (Coker et al., 2000).

P. mirabilis has been implicated in meningitis, empyema, osteomyelitis and gastroenteritis. Also, it frequently causes nosocomial infections of the urinary tract (46%), surgical wounds (24%) and lower respiratory tract (30%). Less frequently, *proteus species* cause bacteraemia (17%), most often in elderly patients (Mansy, 2001).

The phenomenal evolution and increase of multidrug-resistance of many bacterial pathogens is increasing and representing a growing public health problem in the world.

Evolution and spread of a multidrug-resistant *Proteus mirabilis* clone with chromosomal AmpC-type beta-lactamase was reported in Europe (Luzzaro et al., 2009; D'Andrea et al., 2011)

Multidrug-resistance of *Proteus* spp. calls for regular review of antimicrobial sensitivity pattern among clinically isolated *Proteus* spp. in order to be able to decide which antibiotic to be prescribed.

Materials and methods

Specimens' collection

Different clinical samples such as sputum, wound swab, cerebrospinal fluid (CSF), tracheal aspirate (Tr. asp.), throat aspirate (Th. asp.), catheter Tip, pus, abdominal abscess (Abd. ab.), ear swab, bed sores, peritoneal wound swab (Peri. w.s.), pleural fluid (Pler. fluid), were collected from 6840 patients suspected of bacterial infection at King Fahd Hospital at Medina, Kingdom of Saudi Arabia (KSA). Clinical samples were cultured to isolate the organisms. Demographic data such as sex of the patients was recorded prior to sample collection.

Cultivation and Identification

The clinical samples collected were aseptically inoculated on plates of Blood agar, Chocolate agar Cystine-Lactose-Electrolyte-Deficient (CLED) agar and MacConkey agar (Oxoid Cambridge, UK) and incubated at 37ºC for 24 h. The morphological characteristics of the colonies including size, shape, colour, pigmentation and haemolytic nature were recorded. Suspected *Proteus* colonies were isolated and identified through biochemical tests according to Barrow and Felthan:[9] based on whether they were positive for nitrate reduction; H2S gas production; methyl-red and urease reactions; and negative for lactose fermentation.

Antimicrobial susceptibility test

Susceptibility to antimicrobial agents was determined by using the disk diffusion method (Oqunshe, 2006). The following antimicrobial agents (obtained from BDH London, UK) were used: ampicillin (AP), augmentin (AUG), gentamycin (GM), cefoxitin (FOX), cephalothin (KF), cotrimoxazole (TS), amikacin (AK), ceftazidime (CAZ), aztreonam (AZT), piperacilline (PRL), imipenem (IMP), ciprofloxacin (CIP), cefpiramide (CPM)**.**

The inocula were prepared by growing the various *Proteus* species on separate agar plates and colonies from the plate were transferred with inoculating loop into 3 ml of normal saline in a test tube. The density of these suspensions was adjusted to 0.5 McFarland standards. The surface of Muller-Hinton agar (Oxoid Cambridge, UK) plate was evenly inoculated with the organisms using a sterile swab. The swab was dipped into the suspension and pressed against the side of the test tube to remove excess fluid. The wet swab was then used to inoculate the Muller-Hinton agar by evenly streaking across the surface. By means of Disc Dispenser (Oxoid Cambridge, UK), the antibiotic discs were applied to the surface of the inoculated agar and the plates were incubated overnight at 37ºC. The diameter of zone of growth-inhibition observed was measured and compared to the chart provided by National Committee for Clinical Laboratory Standards (NCCLS).

Results and discussion

Total 6840 clinical samples were collected in the period from 22/6/2011 to 22/8/2012 (14 months) from King Fahd Hospital at Medina, Kingdom of Saudi Arabia (KSA). Clinical samples were screened for *Proteus* spp. It is found that 193 isolates were identified as *Proteus* spp. representing about 3% of all positive samples isolated from sputum, wound swab, cerebrospinal fluid (CSF), tracheal aspirate (Tr. asp.), throat aspirate (Th. asp.), catheter Tip, pus, abdominal abscess (Abd. ab.), ear swab, bed sores, peritoneal wound swab (Peri. w.s.), pleural fluid (Pler. fluid); (**Figure**

1). Results showed completely absence of isolates from many sources such as urine, blood, semen, ascetic fluid, nasal swabs, axilla, perineum, etc…

The calculated percentage of *Proteus* spp. in relation to other clinically isolates were not far away from other studies reported similar results. O'Hara et al, (2000) reported that *P. mirabilis* accounts for approximately 3% of nosocomial infections in the United States and is commonly isolated in clinical microbiology laboratories, where it is probably responsible for 6000,000 patients each year. In the same trend Sekowska et al. (2004) reported that *P. mirabilis* strains were isolated from 4.3% of all positive samples isolated from urine, wounds and ulcers. Also, Feglo et al. (2010) reported a prevalence rate of 8.4 % of *Proteus* species collected from clinical samples.

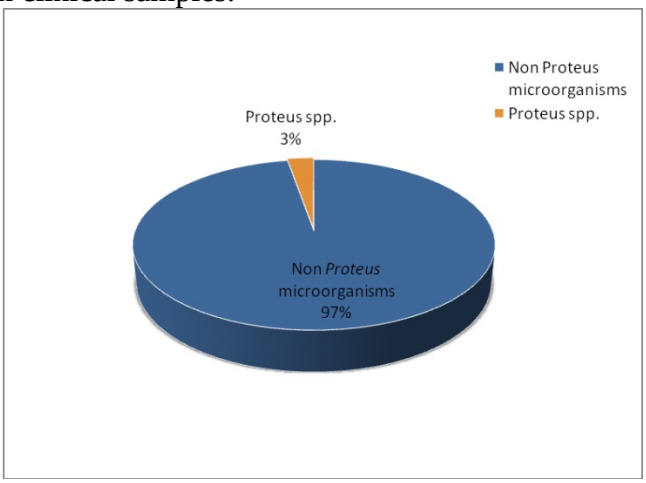

Figure (1): Percentage of *Proteus* spp. isolates to other clinical isolates

It is found that one hundred forty four samples representing 75% of positive *Proteus* isolates were taken from male patients, while forty nine samples representing 25% of positive Proteus isolates were taken from female patients; (**Figure 2**). Males were found to be more vulnerable than females in acquiring *Proteus* infections.

We have no adequate studies concerned with gender distribution of *Proteus* infections. This gender distribution was different from that reported by Feglo et al. (2010), who reported that 43 % of *Proteus* species collected from clinical samples were taken from male patients while 57 % from females.

Our results with male patient predominance are most likely due to the fact that male exposure is greater as they are representing the majority of workforce in Kingdom of Saudi Arabia, so they are exposed more to acquiring infectious diseases, to road traffic accidents and more to metallic fixation of the bone fractures and other operations.

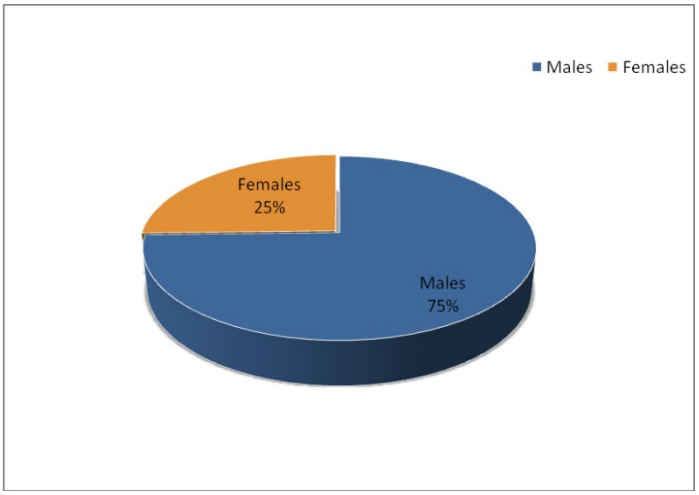

Figure 2: Percentage of males and females positive *Proteus* spp. Isolates

Table (1) showing numbers of males and females specimens isolated from different sources. Seventy four isolates were isolated from sputum specimens (sixty isolates from males and fourteen isolates from females); seventy six isolates were isolated from wound swabs specimens (fifty two isolates from males and twenty four isolates from females); three isolates were isolated from cerebrospinal fluid (CSF) specimens (three isolates were from males); eleven isolates were isolated from tracheal aspirate (Tr. asp.) specimens (nine isolates from males and two isolates from females); twenty isolates were isolated from catheter tip specimens (thirteen isolates from males and seven isolates from females); only one isolate was isolated from throat aspirate (Th. asp.), pus, abdominal abscess (Abd. ab.), and ear swabs specimens (isolated from men); two isolates were isolated from bed sores specimens (one isolate from each gender); two isolates were isolated from peritoneal wound swab (Peri. w.s.) specimens (isolated from men); one isolate was isolated from pleural fluid (Pler. Fluid) specimens (isolated from a woman).

Table 1: Numbers of males and females specimens isolated from different sources:

Sex	Source of specimens											
	Sputum	Wound swab	CSF	Tr. asp.	Th. asp.	Catheter Tip	Pus	Abd. ab.	Ear swab	Bed sores	Peri. w.s.	Pler. fluid
Males	60	52	3	9	1	13	1	1	1	1	2	0
Females	14	24	0	2	0	7	0	0	0	1	0	1
Total	74	76	3	11	1	20	1	1	1	2	2	1

CSF: Cerebrospinal fluid, Tr. asp.: Tracheal aspirate, Th. asp.: Throat aspirate, Abd. ab.: Abdominal abscess, Peri. w.s.: Peritoneal wound swab, Pler. fluid: Pleural fluid

The calculated percentages of positive *Proteus* spp. specimens isolated from different clinical samples are shown in **Figure (3)**. It is found that positive *Proteus* spp. specimens isolated from sputum, wound swabs, cerebrospinal fluid (CSF), tracheal aspirate (Tr. asp.), throat aspirate (Th. asp.), catheter tip, , pus, abdominal abscess (Abd. ab.), ear swabs, bed sores, peritoneal wound swab (Peri. w.s.), and pleural fluid (Pler. Fluid) specimens were representing 38.34, 49.74, 1.55, 5.67, 0.52, 10.36, 0.52, 0.52, 0.52, 1.36, 1.36, and 0.52%, respectively.

Results showed that the greatest number of *Proteus* spp. isolates from clinical specimens were isolated from wound and sputum swabs specimens representing about 88% of all clinical specimens, while the percentage of *Proteus* spp. isolates from throat aspirate (Th. asp.), pus, abdominal abscess (Abd. ab.), ear swabs, and pleural fluid (Pler. Fluid) specimens were the least representing no more than 0.52%.

Being wound isolates were the highest (49.47%) are in the same trend with many results. This result agrees with similar studies conducted in England, Wales and Northern Ireland. (Chow et al., 1979; Jones et al., 2003). Wounds recorded the highest percentage of *Proteus* isolates (64.5 %) and this confirmed the findings of Newman *et al.* (2006) in Ghana; Yah *et al.* (2001) in Nigeria; and Feglo et al. (2010) in Ghana. *Proteus* seems to be a common cause of wound infections in West Africa , in contrast with those from Europe and Asia (Reslinski et al., 2005; Orett, 1999) which showed *Proteus* species to be more commonly encountered in urine than in other clinical specimens.

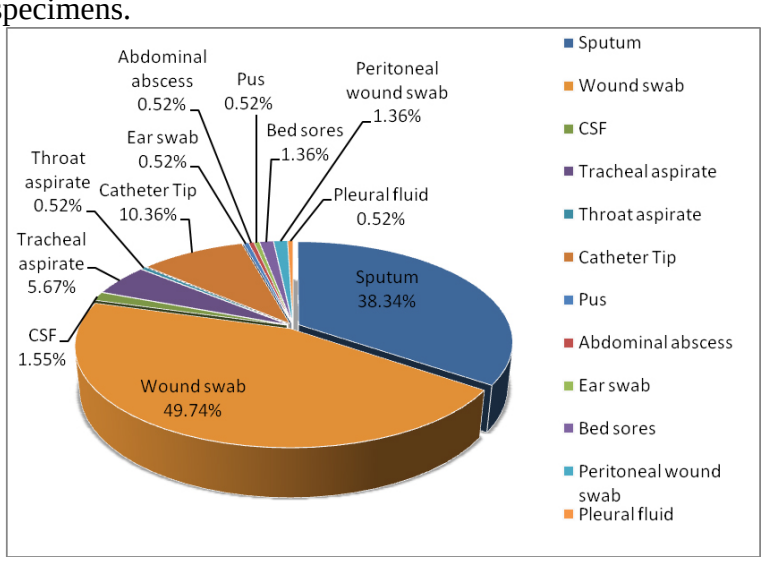

Figure 3: Percentage of positive *Proteus* spp. specimens isolated from different clinical samples

The calculated percentages of males and females specimens isolated from different sources are shown in **Table (2) and Figure (4)**. Results

showed that about 80% of positive *Proteus* spp. isolates from sputum and tracheal aspirate (Tr. asp.) were collected from males, while about 65-70% of isolates from wound swabs and catheter tips were isolated from males. Percentages of other sources of specimens are not indicative for comparing males to females percentage seen to the low number of specimens. In conclusion results showed that males were found to be more vulnerable than females in acquiring *Proteus* infections from all types of considered clinical specimens. male patient predominance is elucidated previously on the base of the fact that male exposure to infections is greater.

Table 2: Percentage of males and females specimens isolated from different sources:

Sex (%)	Source of specimens											
	Sputum	Wound swab	CSF	Tr. asp.	Th. asp.	Catheter Tip	Pus	Abd. ab.	Ear swab	Bed sores	Peri. w.s.	Pler. fluid
Males	81.08	68.42	100	81.82	100	65	100	100	100	50	100	0
Females	18.92	31.58	0	18.18	0	35	0	0	0	50	0	100

CSF: Cerebrospinal fluid, Tr. asp.: Tracheal aspirate, Th. asp.: Throat aspirate, Abd. ab.: Abdominal abscess, Peri. w.s.: Peritoneal wound swab, Pler. fluid: Pleural fluid

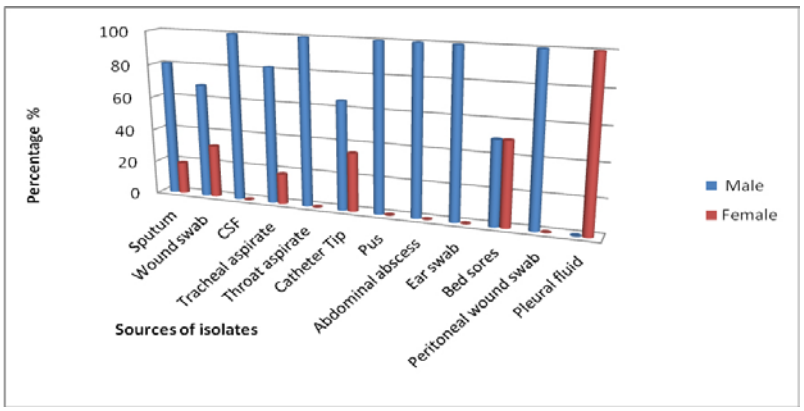

Figure 4: Percentage of males and females specimens isolated from different sources

Thirteen different antibiotics (ampicillin (AP), augmentin (AUG), gentamycin (GM), cefoxitin (FOX), cephalothin (KF), cotrimoxazole (TS), amikacin (AK), ceftazidime (CAZ), aztreonam (AZT), piperacilline (PRL), imipenem (IMP), ciprofloxacin (CIP), cefpiramide (CPM)) representing different families of antibiotics were tested on *Proteus* spp. specimens for studying its antimicrobial sensitivity pattern. Results of antimicrobial sensitivity tests **(Table 3, 4 and Figure 5)** revealed that imipenem (IMP) antibiotic **(Figure 6)** was the most effective antibiotic against *Proteus* spp. with percentage (%) of antimicrobial sensitivity of 91% of *Proteus* spp.

Imipenem (Imp) was followed by amikacin (AK) **(Figure 7)** with percentage (%) of antimicrobial sensitivity of 61% of *Proteus* spp.

Imipenem (Imp) and amikacin (AK) were followed by cefoxitin (FOX), aztreonam (AZT), and piperacilline (PRL) with percentage (%) of antimicrobial sensitivity of 48.2, 47.7, and 44.1, respectively. The rest of the tried antibiotics had less than 40 % of antimicrobial sensitivity pattern.

Results also showed that resistance of *Proteus* spp. was evident to the following antibiotics cotrimoxazole (TS), cefpiramide (CPM), ampicillin (AP), cephalothin (KF) with percentage (%) of antimicrobial resistance reaching higher than 80% *Proteus* spp. Four antibiotics (augmentin, amikacin, piperacilline, imipenem) yielded intermediate reactions ranging from 0.5 to 3.1%.

Results are recommending prescribing of imipenem (IMP) antibiotic in the treatment of *Proteus* spp. because it is the most effective antibiotic against *Proteus* spp in vitro. Although *Proteus* spp. are less sensitive to amikacin (AK) antibiotic, it is recommended to be prescribed in the case of sensitivity to imipenem (IMP). In acute cases *Proteus* infections could require use of synergic combined antimicrobial therapy (imipenem-amikacin).

Attention should be paid to the increasing resistance of *Proteus* spp. to many antibiotics especially augmentin, amikacin, piperacilline, and imipenem. So, those antibiotics should not be prescribed to treat *Proteus* infection cases, as a multidrug resistance would be produced.

The antimicrobial susceptibility testing carried out by El-Tahawy (2000) showed that imipenem was the most effective against gram-negative organisms.

The study carried out by Makled and Alghamdi (2006) showed that aminoglycosides commonly used in treatment of infections caused by *Pr. mirabilis* isolates are still effective.

Resistance of 77-85% of *Proteus* spp. against ampicillin, co-trimoxazole, tetracycline, and chloramphenicol was reported by Feglo et al. (2010). Similar results were reported by Newman et al. (2006).

The high antibiotic resistance of *Proteus* spp. may be an indication of the resistance levels among the enterobacteriaceae and perhaps salmonellae since indiscriminate ingestion of antibiotics provides selective pressure, leading to a higher prevalence of resistant bacteria **(**Barrow and Felthan, 2003**).** Not only are these species potential causes of infections but also potential reservoirs of resistance genes that could be transferred to other bacterial pathogens. The high levels of β-lactamase production and multi-drug resistance of the isolates are indications of an increase in the resistance menace reported by many studies (Feglo et al., 2010).

Results of Fass et al. (1995) reported decreasing susceptibility of *P. stuartii* to ciprofloxacin from 100 to 46% over a 6-year period in their institution; are in the same trend with our results reporting 33% susceptibility of *Proteus* spp. isolates.

Swenson et al, (1999) showed that virtually all *Proteus vulgaris* and *Proteus penneri* strains are capable of producing inducible b-lactamases that will hydrolyze primary and extended-spectrum penicillins and cephalosporins (Swenson et al., 1999). For these reasons, the susceptibility of *Proteus* isolates needs to be monitored.

The different *Proteus* species differs in their susceptibility pattern to different antibiotics. For example, the indole-negative *P. mirabilis* strains are generally more susceptible to antimicrobials than are *P. vulgaris*, *P. penneri*, and *P. hauseri. P. mirabilis* has intrinsic resistance to nitrofurantoin and tetracycline but is generally susceptible to the amino- and ureido-penicillins (ampicillin, amoxicillin, and piperacillin), cephalosporins (cefazolin, cefoxitin, cefuroxime, cefotaxime, ceftazidime, ceftriaxone, ceftizoxime, and cefepime), aminoglycosides (amikacin, gentamicin, and tobramycin), imipenem, ciprofloxacin, and trimethoprim-sulfamethoxazole (Fuchs et al., 1996; Ronald, 1994; Thornsberry and Yee, 1996; Yao and Moellering, 1999). However, high levels of ciprofloxacin resistance have been reported for *P. mirabilis* in hospitals where use of this agent is unrestricted (Thomson et al., 1994). In 1979, Chow et al. reported an outbreak of *P. mirabilis* that was resistant to ampicillin, cephalothin, tetracycline, chloramphenicol, carbenicillin, colistin, trimethoprim-sulfamethoxazole, streptomycin, and the aminoglycosides (Chow et al., 1979). An outbreak of *P. mirabilis* that was resistant to both gentamicin and the antiseptic chlorhexidine, as well as seven other antimicrobial agents, was reported as the cause of urinary tract infections in 90 patients in England in 1987 (Dance et al., 1997). The source of the outbreak was linked to the introduction of a catheter care policy involving chlorhexidine. In an unusual outbreak in a hospital nursery, the strain of *P. mirabilis* that was responsible for bacteremias and meningitis in newborns was tetracycline susceptible. This very unusual antimicrobial pattern was used as a marker to trace the epidemiology (Burke et al., 1971). *P. penneri* is generally more resistant to penicillin than is *P.vulgaris*, and its susceptibility pattern more closely reflects that of *M. morganii* than that of *P. vulgaris*. These *Proteeae* are generally susceptible to cefoxitin, broad-spectrum cephalosporins (cefotaxime, ceftriaxone, ceftizoxime, and ceftazidime), cefepime, aztreonam, aminoglycosides, ciprofloxacin, tazobactam, and imipenem (Fuchs et al., 1996; Yao and Moellering, 1999) and may be resistant to cefazolin, cefprozil, cefuroxime, cefamandol, cefdinir, cefoperazone,

loracarbef, ampicillin, and the ureidopenicillins (Biedenbach and Jones, 1994).

Table 3: Antimicrobial sensitivity pattern of *Proteus* specimens to different antibiotics.

Antibiotic	Abbreviation	Sensitive	Resistant	Intermediate
Ampicillin	AP	27	166	0
Augmentin	AUG	64	128	1
Gentamycin	GM	71	120	2
Cefoxitin	FOX	93	100	0
Cephalothin	KF	33	160	0
Cotrimoxazole	TS	18	175	0
Amikacin	AK	118	74	1
Ceftazidime	CAZ	73	120	0
Aztreonam	AZT	92	101	0
Piperacilline	PRL	85	107	1
Imipenem	IMP	175	12	6
Ciprofloxacin	CIP	64	129	0
Cefpiramide	CPM	22	171	0

Decreasing susceptibility of *Proteus* group and the potential for emerging resistance illustrated in this group needs for routine susceptibility tests (Fass et al., 1995). This study is therefore a step towards the generation of national data on the prevalence of antimicrobial resistance patterns of *Proteus* spp.

Table 4: Percentage (%) of antimicrobial sensitivity pattern of *Proteus* specimens to different antibiotics.

Antibiotic	Abbreviation	Sensitive (%)	Resistant (%)	Intermediate (%)
Ampicillin	AP	14	86	0
Augmentin	AUG	33.2	66.3	0.5
Gentamycin	GM	36.8	62.2	1
Cefoxitin	FOX	48.2	51.8	0
Cephalothin	KF	17.1	82.9	0
Cotrimoxazole	TS	9.3	90.7	0
Amikacin	AK	61.1	38.4	0.5
Ceftazidime	CAZ	37.8	62.2	0
Aztreonam	AZT	47.7	52.3	0
Piperacilline	PRL	44.1	55.4	0.5
Imipenem	IMP	90.7	6.2	3.1
Ciprofloxacin	CIP	33.2	66.8	0
Cefpiramide	CPM	11.4	88.6	0

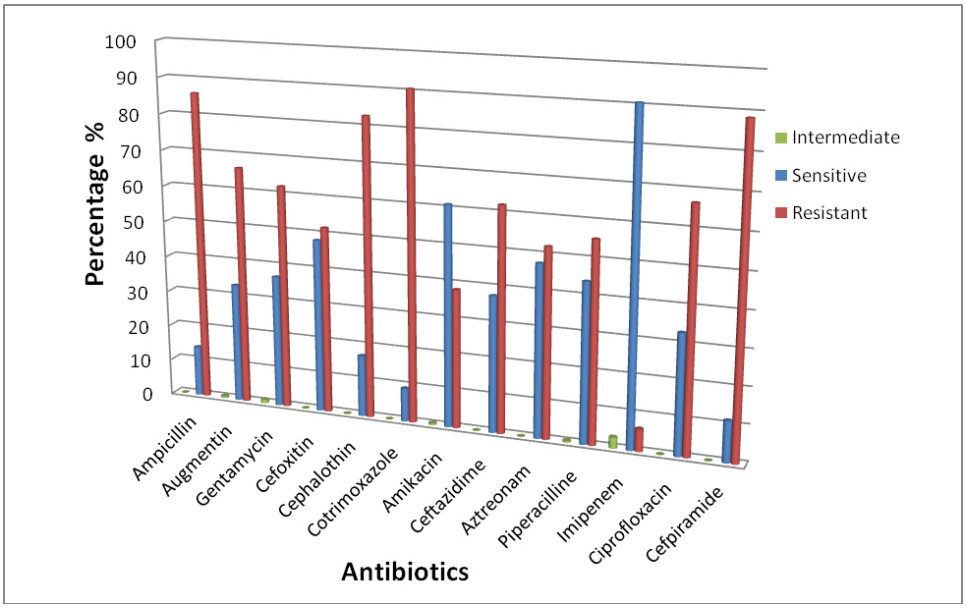

Figure 5: Percentage (%) of antimicrobial sensitivity pattern of *Proteus* specimens to different antibiotics

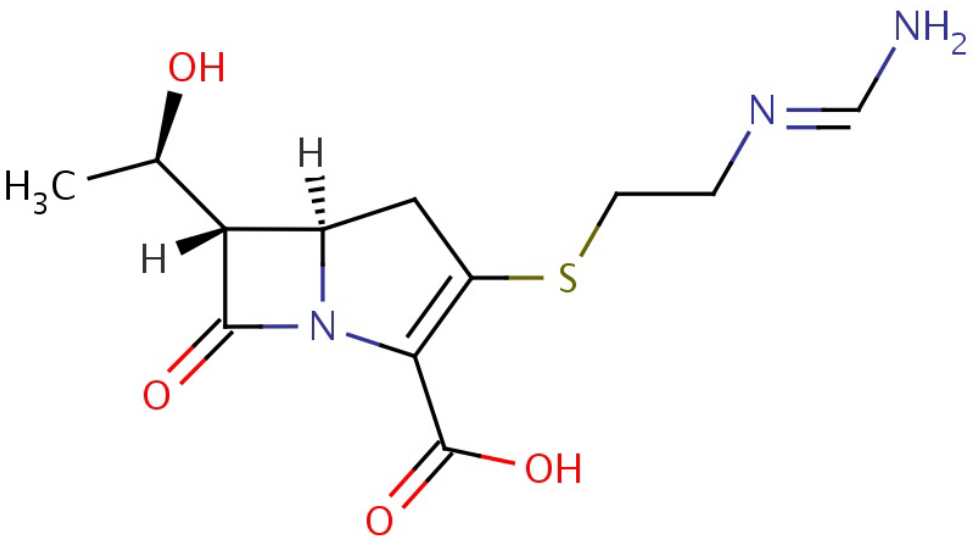

Figure 6: Chemical structure of imipenem antibiotic
(Chemical formula: C12H17N3O4S, Drugbank ID: DB01598)

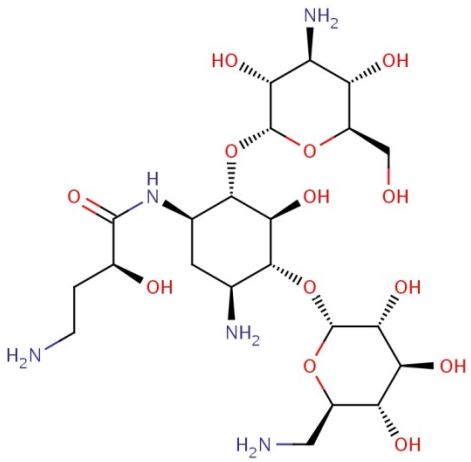

Figure 7: Chemical structure of amikacin antibiotic
(Chemical formula: C22H43N5O13, Drugbank ID: DB00479)

Results (**Table 5 and Figure 8**) demonstrated that percentage of *Proteus* spp. infections was highest during summer season representing about 40% of infections all year along. Summer season was followed by winter season with about 32% of infections, and autumn season (which coincided with the season of pilgrimage) with about 22% of infections. While spring season has the least recorded percentage of *Proteus* infections with only 6% of infections.

It is known long time ago that bacterial infections peak during summer season. Winter season also a candidate season for bacterial infections especially those concerned with respiratory system. Many studies reported incidence of seasons on increasing *numbers of* bacterial infections (O'Hara et al, 2000; Smith and Hogan, 2008; Bryan, 2011).

It was clear that autumn season coinciding with the season of pilgrimage characterizing by the large number of delegations of pilgrims to Medina did not reported to be one of the highest seasons with infections (summer and winter) which could be attributed to success of control infections strategies taken by the health authorities which give special attention to fighting bacterial infections during pilgrims season to avoid epidemics.

Table 5: Percentage (%) of *Proteus* infections pattern during different seasons:

Season	Percentage (%) of *Proteus* infections
Summer (22 June -22 September)	40
Autumn (23 September -21 December): Pilgrimage season	22
Winter (22 December -30 Mars)	32
Spring (21 Mars-21 June)	6

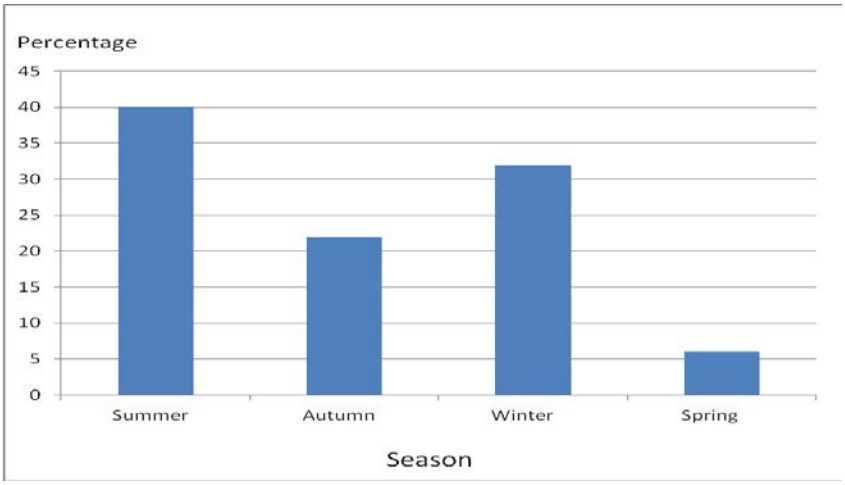

Figure 8: Percentage (%) of *Proteus* infections pattern during different seasons

References:

Barrow G.I. and R.K.A. Felthan (2003) Cowan and Steel's Manual for the Identification of Medical Bacteria. 3rd Ed. Cambridge University Press. Cambridge UK. 351-353.

Biedenbach, D. J., and R. N. Jones. (1994). Predictive accuracy of disk diffusion test for *Proteus vulgaris* and *Providencia* species against five newer orally administered cephalosporins, cefdinir, cefetamet, cefprozil, cefuroxime, and loracarbef. J. Clin. Microbiol. 32:559–562.

Burke, J. P., D. Ingall, J. O. Klein, H. M. Gezon, and M. Finland. (1971). *Proteus mirabilis* infections in a hospital nursery traced to a human carrier. N. Engl. J. Med. 284:115–121.

Charles Bryan. infectious disease, chapter seven, urinary tract infections. copyright (2011), The Board of Trustees of the University of South Carolina.

Chow A.W., Taylor P.R., Yoshikawa T.T., and L.B. Guze (1979). A nosocomial outbreak of infection due to multiple strains of resistant *Proteus mirabilis*: Role of intestinal colonization as a major reservoir. J. Infect. Dis. 139:621–627.

Orett (1999) Prevalence of *Proteus species* in urinary tract infections in a regional hospital in Trinidad. Zhonghua Yi Xue Za Zhi (Taipei), 62:438-442.

Coker, C.; Bakare, O.O. and H.L.T. Mobley (2000): H-NS Is a repressor of the *Pr. mirabilis* urease transcriptional activator gene ureR. J. Bacteriol. 128 (9): 2649-2553.

Dance, D. A. B., A. D. Pearson, D. V. Seal, and J. A. Lowes. (1987). A hospital outbreak caused by a chlorhexidine- and antibiotic-resistant *Proteus mirabilis*. J. Hosp. Infect. 10:10–16.

D'Andrea M.M., Literacka E., Zioga A., Giani T., Baraniak A., Fiett J., Sadowy E., Tassios P.T., Rossolini G.M., Gniadkowski M., and V.

Miriagou. (2011). Evolution and spread of a multidrug-resistant *Proteus mirabilis* clone with chromosomal AmpC-type cephalosporinases in Europe. Antimicrob Agents Chemother. 55(6):2735-42.

De Champs, C., Bonnet, R., Sirot, D., Chanal, C. and J. Sirot (2000). Clinical relevance of *Pr. mirabilis* in hospital patients: A two year survey. J Antimicrob. Chemoth. 45: 537-*539*

El-Tahawy A.T (2000). Bacteriology of diabetic foot infections. Saudi Medical Journal; 21 (4): 344-347

Fass, R. J., J. Barnishan, and L. W. Ayers. (1995). Emergence of bacterial resistance to imipenem and ciprofloxacin in a university hospital. J. Antimicrob. Chemother. 36:343–353.

Feglo P.K., Gbedema S.Y., Quay S.N.A., Adu-Sarkodie Y., and C. Opoku-Okrah (2010). Occurrence, species distribution and antibiotic resistance of *Proteus* isolates: A case study at the Komfo Anokye Teaching Hospital (KATH) in Ghana. International Journal of Pharma Sciences and Research (IJPSR), 1(9): 347-352.

Fuchs, P. C., A. L. Barry, S. D. Brown, and the AST Surveillance Group. (1996). Survey of antimicrobial activity of four commonly used third generation cephalosporins tested against recent bacterial isolates from ten American medical centers, and assessment of disk diffusion test performance. Diagn. Microbiol. Infect. Dis. 24:213–219.

Guentzel M.N. (1996). *Escherichia, Klebsiella, Enterobacter, Serratia, Citrobacter,* and *Proteus.* In: *Barron's Medical Microbiology* (Barron 's *et al.*, eds.) (4th ed.). Univ of Texas Medical Branch.

Jones R., Vincent B.A., and W.B. Saunders (2003). Bacteraemia, England, Wales and Northern Ireland: Commun Dis Rep CDR. Wkly

K. Larry Smith and J.S. Hogan. (2008). Environmental Mastitis: Know Your Opponent. NMC Regional Meeting Proceedings.

Luzzaro F., Brigante G., D'Andrea M.M., Pini B., Giani T., Mantengoli E., Rossolini G.M., Toniolo A. (2009). Spread of multidrug-resistant *Proteus mirabilis* isolates producing an AmpC-type beta-lactamase: epidemiology and clinical management. Int. J. Antimicrob. Agents. 33(4):328-33.

Makled A. and A. Alghamdi. (2006). Surveillance of Aminoglycosides Resistance Among *Proteus mirabilis* Isolates From Different Units in Jeddah Hospitals, Saudi Arabia. Egypt. J. Med. Microbiol., 15 (2), 33 7-351.

Mansy, M.S.M. (2001): Genomic fingerprinting using random amplified polymorphic DNA for discrimination between *Pr. mirabilis* strains. Egypt. J. Biotech. (9):67-79.

Newman M.J., Frimpong E., Asamoah-Adu A., and E. Sampane-Donkor (2006) Resistance to Antimicrbial Drugs in Ghana. The Ghanaian –Dutch collaboration for Health Research and Development, 1-6.

O'Hara C.M., Brenner F.W., and J.M. Miller. (2000). Classification, Identification, and Clinical Significance of *Proteus*, *Providencia*, and *Morganella.* Clinical Microbiology Reviews, 13 (4): 534–546.

Oqunshe, A. A. O. (2006): In vitro phenotypic antibiotic resistance in bacterial flora of some indigenous consumed herbal medications in Nigeria. J. Ru. and Tr. Pub. Health. 5: 9-15.

Reslinski A, Gospodarek E, Mikucha A. (2005) Prevalence of multi-drug resistant *Proteus species* in clinical specimens and their susceptibility to antibiotics, Med. Dosw. Micribial, 57(2):175-184.

Ronald, A. R. (1994). Urethritis and cystitis, p. 564–570. *In* P. D. Hoeprich, M. C. Jordan, and A. R. Ronald (ed.), Infectious disease: a treatise of infectious processes, 5th ed. J. Lippincott, Philadelphia, Pa.

Jacobsen, S. M., Stickler, D. J. Mobley, H. L. T. and M. E. Shirtliff (2008). Complicated Catheter-Associated Urinary Tract Infections Due to *Escherichia coli* and *Proteus mirabilis.* Clinical Microbiology Reviews, 21 (1): 26–59.

Sekowska A., Janicka G., Wróblewska J., and E. Kruszyńska (2004). Prevalence of *Proteus mirabilis* strains in clinical specimens and evaluation of their resistance to selected antibiotics. Pol Merkur Lekarski.17(101):538-40.

Stamm, W.E. (1999): Urinary Tract Infections. In Clinical Infectious Disease: A practical approach, Root, K. (ed). P: 649-656. Oxford University Press, Inc, New York.

Swenson, J. M., J. A. Hindler, and L. R. Peterson. (1999). Special phenotypic methods for detecting antibacterial resistance, p. 1563–1577. *In* P. R. Murray et al. (ed.), Manual of clinical microbiology, 7th ed. American Society for Microbiology, Washington, D.C.

Thomson, K. S., W. E. Sanders, and C. C. Sanders. (1994). USA resistance patterns among UTI pathogens. J. Antimicrob. Chemother. 33(Suppl. A): 9–15.

Thornsberry, C., and C. Yee. (1996). Comparative activity of eight antimicrobial agents against clinical bacterial isolates from the United States, measured by two methods. Am. J. Med. 100 (Suppl. 6A):26S–38S.

Yah S.C., Egbanfona N.O., Oranusi S., and A.M. Abouo (2001) Widespread plasmid resistance genes among *Proteus species* in diabetic wounds of patients in Ahmadu Bello University Teaching Hospital (ABUTH) Zaria, Afr. J. Biotechnol. 6(15):1757-1762.

Yao, J. D. C., and R. C. Moellering, Jr. (1999). Antibacterial agents, p. 1474–1504. *In* P. R. Murray et al. (ed.), Manual of clinical microbiology, 7th ed. American Society for Microbiology, Washington, D.C.