

Multiple Antibiotics Resistance Index of both Gram-positive and Gram- Negative Bacteria Isolated from Diabetic patients attending General hospital Minna, Niger State, Nigeria

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multiple Antibiotics Resistance Index of both Gram-positive and Gram- Negative Bacteria Isolated from Diabetic patients attending General hospital Minna, Niger State, Nigeria.

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ABSTRACT

A hospital based cross sectional study was conducted at General Hospital Minna, Niger State from January, 2025 to November, 2025. A total of 400 Diabetic patients with asymptomatic and symptomatic urinary tract infection were investigated for common uropathogenic microorganisms. Clean catch mid-stream urine specimens were collected from each participant. Uropathogens were isolated and identified using conventional standard techniques, the samples were cultured on Cystine Lactose Electrolyte Deficient and MacConkey agar. Antimicrobial susceptibility patterns of the uropathogens isolated were determined using disc diffusion method for bacteria pathogens and MAR index was calculated as the ratio of the number of antibiotics to which an organism is resistance to the total number of antibiotics to which the organism is exposed. which is calculated as a/b , where a = number of antibiotics isolate was resistance to, b = the total number of antibiotics used. The overall prevalence of uropathogens in patients was (71.00%) with prevalence of (60.5%) bacteriuria. *Escherichia coli* (30.31%), *Staphylococcus aureus* (15.33%), *Proteus mirabilis* (11.49%), *Klebsiella pneumoniae* (9.41%), *Pseudomonas aeruginosa* (9.41%) and Coagulase Negative Staphylococci (8.36%) were the common uropathogens obtained. Both Gram-positive and Gram-negative bacteria showed high level of resistance to antibiotics such as Amoxicillin, ceftazidime, Erythromycin, Gentamycin, Levofloxacin and Rifampicin. Gender, age and type of diabetes were significant risk factors influencing the distribution of uropathogens among the patients. High prevalence of multidrug resistant uropathogens and high resistance index among the isolates indicated rising burden of drug resistant uropathogenic infection among patients. Therefore, routine antimicrobial susceptibility testing should be incorporated into clinical practice to guide appropriate therapy.

KEYWORDS: Multi Drug Resistance, Antimicrobial Susceptibility, Gram-positive, Gram-negative, Uropathogens

1.0 Introduction

The World Health Organization (WHO) recognizes urinary tract infections as a prevalent infection and a significant global health challenge in terms of morbidity, healthcare cost and antimicrobial resistance rates. The infections are among the most widespread infectious diseases, affecting millions worldwide annually (Babaoye and Ogala 2021).

Moreover, the Centers for Disease Control and Prevention, recognizing the depth of this issue, has identified urinary tract infections as a leading cause of healthcare-associated infections (HAIs) and a major driver of antibiotic consumption and misuse (World Health Organization, 2021). Also, the World Health Organization (WHO) reports that 80% of antibiotics are used in the community, with 20—50% being administered improperly (Opinta *et al.* 2018). In recent years, the prevalence of multidrug-resistant organisms (MDRO), or called multidrug-resistant (MDR) bacteria, which are both pathogenic and opportunistic, has risen. MDR urinary tract infections are infections that are not susceptible to at least one antimicrobial agent from three or more antibiotics. Globally, notably MDR bacteria include Gram-negative bacteria, such as members of enterobacteriaceae producing plasmid-mediated, extended-spectrum β -lactamase (ESBL), and Gram-positive bacteria such as MRSA and Vancomycin-resistant Enterococci (VRE). The emergence of MDR urinary tract infections is a significant public health concern, demanding appropriate measures to prevent their spread and manage affected patients. (Craig 2018).

The readily available over-the-counter drugs, illiteracy, and fake and cheap substandard drugs are also part of the reasons that promote the misuse and abuse of drugs. In Ghana, effective treatment of urinary tract infections is often hampered by inadequate facilities for isolation and antimicrobial susceptibility testing. This can sometimes lead to urologic complications due to untreated, undetected and improperly treated urinary tract infections. Northern Ghana is a region of limited resources in term of health care. Microbiological services (Culture and susceptibility test) are occasionally available at the only teaching hospital in Tamale, the Capital. The published data on urinary tract infections are generally scarce and are mostly

reported from other regions of the country, revealing striking resistance among isolates to commonly prescribed antibiotics (Opinta *et al.*, 2018). Several years ago, South Africa suggested including urinary tract infection identification in IMCi based on dipstick urinalysis for leucocytes and nitrites but this approach has been largely discarded. Oman, with basic laboratory facilities at primary health care level decided to include urinary tract infection management in its IMCi adaptation through screening febrile or symptomatic children with urine microscopy and referring those with urine white cells of 20 per cubic mm or more for pediatric Consultation (personal communication) (Banapurmath and Jayamony, 2018). The studies included in that research was identified by a search on Medicine and pub medicine of the relevant scientific literature published in English from 1966 to the present. There urinary tract infections in children were linked with the following keywords; developing countries, IMCi, epidemiology, etiology, risk factors, diagnosis, symptoms, signs, urinalysis, treatment, prevention, prognosis, urinary tract abnormalities, vesico-ureteric reflux, acute pyelonephritis, renal scarring, hypertension, renal imaging, chronic renal failure, reflux nephropathy and end stage renal failure (Tansril *et al*, 2019).

Extensive research has been conducted in developed countries on the epidemiology, risk factors, etiology, diagnosis, treatment, prognosis and prevent of urinary tract infections in children and all demonstrated bacteriuria in asymptomatic children of all ages from premature infant to school age children. It is also now accepted that asymptomatic bacteriuria does not present a risk to a child of any age and screening for bacteriuria in the asymptomatic child is not indicated (Sun *et al.*, 2021). Therefore, this study aims to determine the multi drug resistance index among uropathogenic bacteria isolated and their implications for clinical practice, infection control policies, and the development of effective treatment strategies for UTIs within the study area and beyond. (Seifu *et al.*, 2018).

3.0 MATERIALS AND METHODS

3.1 Study Area

Minna lies between Latitudes 9°33 and 9°40' north of the Equator and Longitudes 6°29 and 6°35' East of the Greenwich Meridian (Fig. 3.1).

The town spanned along the main spine road that separates the city into west and east. This road is from Chanchaga in the south to Maikunkele in the North, covering a distance of about 20 km. The west-East pattern, spanned from Gidan- Kwano along Bida axis in the west, to Maitumbi to Gwada axis, in the East, over a distance of 15km (Idowu *et al.*2020,). It has a land area of about 79.87 km², with a current metro area population of 496,000. The major occupations of the populace are agriculture, mining, leather production and metal processing. General Hospital Minna is one of the major hospitals attended by the populace in Minna. It was established in 1946 and was initially under the control of regional and native authorities as well as missionaries. The hospital has since then undergone various transformations from a Railway Staff Medical Centre to General Hospital offering primary, secondary and some specialist services. It has 762 bed capacities and the hospital is one of the main referral public health facilities owned by Niger State Government.

3.2 Study Design

A cross-sectional study was conducted at General Hospital Minna, Niger State. A total of Four Hundred (400) patients attending hospital for follow up were included. patient from age of 18-65 years of age and above was included in the study regardless of the presence or absence of urinary tract infection symptoms. All these patients have been diagnosed according to the World Health Organization (WHO) 2023 and American Diabetes Association. All the participants also meet the criteria with symptoms of diabetes plus fasting blood glucose level equal or more than 126g/l.

3.3 Sample Size Determination

Single population proportion using the provenance reported by Joshi *et al*, (2016) was used to determine the sample size. The sample size was calculated using equation 3.1

$$n = \frac{Z^2 P (1-P)}{d^2}$$

Where n is sample size,

P= prevalence rate = 1.6 % by Olowo-Okere

Z= 1.9 for confidence level at 95%

D= marginal tolerable error at 0.5%

$$n = \frac{(1.96)^2 \times 0.146 \times (1-0.146)}{(0.05)^2}$$

$$n = \frac{0.479}{0.0025} = 191.6$$

Total number of samples = 192 x2 = 384

Enrollment = 400 sample size

3.4 Inclusion and Exclusion Criteria

3.4.1 Inclusion Criteria

Participants included are patient between the ages of 18 - 65 years old and above and willing to participate in the study. Also, Participants considered for the study fall in the strata of diabetes including male and female and are non-pregnant females.

3.4 .2 Exclusion criteria

. Those that did not consent to participating in this study were also excluded.

3.5 Media Preparation

The media used include CLED, MacConkey and Nutrient agar. These media were prepared according to the manufacture's instruction and autoclave at 121°C for 15 minutes.

3.6 Sample Collection

Freshly voided 10ml of clean catch midstream urine specimen was collected using a sterile plastic container. All patients were instructed on how to collect mid-stream urine sample to avoid contamination and further analysis was carried out immediately at the laboratory through inoculation and culture of the samples on appropriate media. This is to ensure that the pathogenic organisms present in urine sample are isolated and identified and did not overpopulate.

In cases where the collected samples were not completely analyses at the particular period of collection, the samples were stored in a refrigerator at 4 °C.

3.6.1 Isolation of Uropathogenic Microorganisms

The urine sample was inoculated onto CLED agar, and MacConkey agar (Parkin USA0) using sterile calibrated wire loop (0.001). After 24 hours aerobic incubation at 37°C, the plate was examined macroscopically for morphological appearance as presumptive identification. The colonies were subcultured to obtain pure culture of each isolate. The isolate was stored on nutrient agar slant for further analysis.

3.6.2 Identification and Characterization of Bacterial Isolate

Identification of the bacterial isolates was carried using the colonial morphology on culture media, Gram's reaction and biochemical test's reaction.

3.7 Gram staining procedure

A drop of normal saline solution or phosphate buffer saline was placed on the middle of a slide using sterile loop; a colony was picked and placed on a slide to make a smear. The smear was air dried and fixed by passing the slide over the flame of a Bunsen burner. After heat fixing, the smear was stained with crystal violet for 1 minute; it was washed with water, Lugol's iodine was applied for 1 minute, rinsed with water and then decolorized with acetone or alcohol briefly, and washed with water. Lastly, it was counter-stained with safranin for 1 minute, it was washed with water, air dried and examined microscopically under X100 objective lens. Gram positive organisms appeared purple coloured, while Gram negative organisms appeared red.

The following biochemical tests were carried out for further identification of microorganisms; These includes, Catalase test, Coagulase test, Indole test, Citrate utilization test, Sugar fermentation test, Methyl red test, Vogues Proskauer test, Hydrogen sulfide gas production test and Oxidase test.

3.8 Antimicrobial Susceptibility Testing

3.8.1 Antibiotic Susceptibility Test

Antibiotic susceptibility test was carried on each isolated bacteria using Kirby Bauer disc diffusion method according to the Clinical and Laboratory Standard Institute (CLSI; M100-S22) guideline. Bacterial suspension was prepared by emulsifying pure colonies in nutrient broth and adjusted to 0.5 McFarland standard. A sterile cotton swab was dipped into the suspension

and swabbed on surface of Muller -Hinton agar plate (Oxoid). The following standard antibiotic disc (Ceftazidime 30mg, Cefuroxime 30mg, Gentamycin 10mg, Streptomycin 30mg, Amoxycillin 500mg, Azithromycin 10mg, Ciprofloxacin 10mg, Erythromycin 30mg, Levofloxacin 20mg and Rifampicin 20mg) was placed aseptically on the inoculated Mueller - Hinton agar plate and incubated at 37°C for 18 hours. After incubation the diameters of the zones of complete inhibition were measured using caliper. The isolated zone of inhibition was reported based on CLS, M100-S 22 standard as Sensitive, Intermediate and Resistance.

3.9 Quality Control

Strict measures were followed from the beginning till end of this study. The questionnaire was pretested in many patients. Quality control of each agar plate in the prepared biochemical for microbe identification.

3.10 Multi-Drug Resistance Index (MDRI)

Based on the susceptibility pattern of the isolated bacteria, bacteria resistant to ≥ 3 classes of anti-biotic were considered as multi drug resistant. Bacterial susceptibility to antimicrobial agent was determined according to the Clinical Laboratory Standard Institute (CLSI, 2022)

3.10.1 Multiple Antibiotic Resistance Index

MARI was calculated using the formula (Equation 3.1)

$$\text{MARI} = \frac{\text{Number of antibiotics isolate is resisted to}}{\text{Total number of antibiotics tested}}$$

3.11 Data Analysis

All data generated during the research work was subjected to one way analysis of the variance. (ANOVA) using Statistical Package for Social Science (SPSS 2020). The data obtained was analyzed using descriptive statistics where frequencies and percentages were presented on the tables. Statistical level of significant was determined at p- value < 0.05. In addition, Chi -square test was employed to test the association between the Socio-Demographic Data of diabetic patients and growth of uropathogenic microorganisms.

RESULTS AND DISCUSSION

4.1 Results

The bacterial isolated were identified as *Escherichia coli*, *Klebsiella pneumoniae*, *Proteus mirabilis*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Coagulase negative staphylococci*, using their morphological appearance on differential media and reaction to various biochemical tests as presented in (Table 1). Out of 400 samples collected the most common uropathogen isolated are *E. coli* 87 (30.31%), *Klebsiella pneumoniae* 27(9.41%), *proteus mirabilis* 33(11.49%), *Pseudomonas aeruginosa* 27(9.41%), *Staphylococcus aureus* 44 (15.33%) and Coagulase negative staphylococci (CoNs) 24 (8.36%) were isolated from the culture positive samples as presented in (Table 2). In female the most common uropathogens isolated were *E. coli*, *Staphylococcus aureus* and *Klebsiella pneumoniae*. Among males the most common uropathogens isolated are *E. coli* and *Staphylococcus aureus* (Table 3).

More uropathogens were seen in females than in male's patients and age groups between 40, 55 years and above had high frequency of pathogens. (Table 4).

There is high prevalence of uropathogens among patients with blood glucose level between 126/ml and above. The patients are considered as diabetics. One hundred and twenty-six gram per litre (126/ml) sugar level patients has 36.24% and those with sugar level above 126g/km has 63.28% which gives non-diabetic total prevalence of 37.28% and diabetic patient 62.72% has shown in (Table 5). In this study, the overall prevalence of uropathogens in patient was 71.75 % with prevalence of 56.25% in significant bacteria (Table 6).

Four hundred (400) patients were enrolled in this research work. Out of 400 participants 241 were females between age of 18 to 65 years and above and 159 are males Meaning greater than half of participants are females which is $\frac{241}{400}$ (60.25%) and $\frac{159}{400}$ (39.75%) respectively. Two hundred and eighty $\frac{280}{400}$ (70%) participants had type I and 120 participant $\frac{120}{400}$ (30%) had type II mellitus. More than 45% of total participant have less than 10 years history of diabetes. The blood glucose level of the study participant was 126mg/dL in $\frac{99}{400}$ (24.756) and ≥ 126 mg/dL in 301 (75.256). Majority of the participant are urban dweller $\frac{224}{400}$, (56%), married, $\frac{266}{400}$ (66.56) and did not attend any level of education (illiteracy) $\frac{223}{400}$ (55.75%).

And from the result of diagnosis of the participant, majority of them have previous urinary tract infections $^{288}/_{400}$ (50%) and history of antibiotic used $^{294}/_{400}$ (73.5%) (Table 7).

Antibiotic Susceptibility Pattern of Gram-positive Bacterial Isolates

The two Gram positive bacterial isolates were observed to be resistance to Amoxicillin, Ceftazidime, Erythromycin, and Rifampicin. But moderates' resistance to cefuroxime and Levofloxacin. Among Gram negative organism *Staphylococcus aureus* showed 44/44 (100%) resistance to Amoxicillin and Rifampicin but 40/44 (80%) sensitive to Azithromycin and also showed 25/44 (55%) sensitive to cefuroxime. Also, *Staphylococcus aureus* showed moderate sensitive to ciprofloxacin 40/44 (9.0) and intermediate resistant's Gentamycin 40/44 (90.1) respectively. Coagulase negative *Staphylococcus* (CONS) showed high level of sensitivity to Azithromycin and Ciprofloxacin and moderate resistant to Gentamycin and Streptomycin as shown in (Table 8).

Most Gram-negative bacterial isolates were also observed to be resistance to Amoxicillin except for *Klebsiella pneumoniae* which 20/27 (74.16%) sensitive to Amoxicillin. *E. coli* showed 86/87 (98.9%) resistance to ceftazidime, 70/78 (80.0%) sensitive to Rifampicin. *Klebsiella* also showed 11/27 (35.0%) resistant to Erythromycin and 20/27 (74%) sensitive to Gentamycin. *Proteus* showed 21/33 (63.6%) sensitive to ceftazidime, 30/33 (90.4%) sensitivity to Ciprofloxacin, 30/33 (90.1%) sensitivity to Rifampicin 30/33 (90.8%) sensitivity to Streptomycin and 33/33 (100%) sensitive to both Erythromycin and Levofloxacin respectively, but 30/33 (90.9%) resistance to Cefuroxime. *Pseudomonas aeruginosa* showed 27/27 (100%) sensitivity to Ciprofloxacin, Gentamycin and Streptomycin, also showed 20/27 (74.1%) sensitive to Levofloxacin, Rifampicin and ceftazidime. It also showed 21/27 (78%) sensitivity to Cefuroxime, moderate resistance to Azithromycin and 27/27 (100%) resistance to Erythromycin (Table 9).

Drug resistance for three or more drugs was observed in 48/68 (70.63%) and 102/174 (58.62%) of Gram positive and gram-negative bacteria respectively. Many *E. coli* 62/87 (71.2%) and *Klebsiella Pneumoniae* 16/27 (59.3%) among Gram-negative isolate and 32/44 (47.1%) of

Staphylococcus aureus in Gram- positive bacteria shows resistance to more than five antimicrobials. Both *proteus mirabilis* and *pseudomonas aeruginosa* 13/33 (39.39%) and 11/27 (40.7%) shows moderate resistance to antimicrobial in this study. Also, *Coagulase negative Staphylococcus* shows intermediate level of 16/24 (66.7.%) antimicrobial resistance in this study. The overall prevalence of multi drug resistance in both groups was remarkable high in all isolates in this study (Table10 and 11).

4.2 Discussion

The 56.25% prevalence of bacteria obtained in this study was contrary with what had been previously reported in Ethiopia. However, this finding is in accordance and similar with the results reported from India (32%), Iraq (49.1%) and Pakistan (51%). The reason for these differences in rate of bacterial urinary tract infection (UTI) etiologies may be due to the differences in methodology used, the environmental condition, social habits of the community, health status of individual, the standard of personal hygiene and educational level in each country (Woldermariam *et al*,2019).

In this study, *Escherichia coli* was the most predominant bacterial isolate and its prevalence was 30.31 %, this is lower than the finding in Gondar, Ethiopia, where 31.7% was reported and however, it also lower than the rate reported by Geerlings *et al*, (2020), and Awwardik *et al*, (2019) in Khartum, Sudan and Ago-Iwoye, Ogun State, Nigeria respectively (Wiles *et al*.,2018).

The second most common bacteria reported in this study were *Staphylococcus aureus* 15.33 %, followed by *Proteus mirabilis*, 11.49%, *Klebsella pneumoniae* 9.41%, *Pseudomonas aeruginosa* 9.41% and Coagulase negative *Staphylococcus aureus* (CONs) 8.36 %. From similar findings reported by Saleem *et al*., (2022) in Hail, Kingdom of Saudi Arabia started that the high isolation rate of CONs is due to contamination during specimen collection or processing and changes in pattern of infection in patients . The reason is that females are more prone to UTIs due to several reason including anatomy based in gender that is, shorter urethra, the promixity of the urethra to the anus in females as well as entry of pathogens promoted by

sexual intercourse among females with relationship status in sexually active age group and other factors like estrogen deficiency (Table 4.3). Also, recently discovered showed that Coagulase negative staphylococci are regarded as the important agent of hospital infections and also opportunist infections. Therefore, there prevalence in these studies maybe due to the lack of proper medication of patients involved in the study (Rinkin and ponaam,2016).

In different previous studies, diabetes related risk factors have been proposed as an enhance6d factor for urinary tract infection in patients with diabetes. In this study among the variables as diabetes related risk factor for urinary tract infection, a significant association was observed for uropathogens and being type 1diabetic patients with prior study from South African other neighbouring Gulf countries such as Kuwait, India, Oman and elsewhere (Gyansa-lutterodt.,2022).

In this study intermediate to low level resistance for one or more antimicrobial agents was observed for both Gram negative and Gram-positive bacteria excerpt for Amoxycillin that show high level of resistance. Comparatively similar findings have reported in previous studies in Nigeria, Sudan Ethiopia and Saudi Arabia. (Wagenlehner *et al.*, 2018).

The highest level of resistance to Amoxycillin may be due to the easily availability, low cost of the antibiotic and use of drugs without prescription.

Multi drug resistance was observed in more than 59.3% of the isolated bacteria in this study. This is higher than the previous study finding in Addis Ababa and Gondar in Ethiopia. This study and other studies carried out about uropathogenic microorganism exhibited that the multi-drug resistance was found to be very high to the commonly used antibiotics drug in patient. Antibiotic resistance has been recognized as the consequence of inappropriate use of antibiotic. Therefore, the reasons for this alarming event might be improper and incorrect administration of antimicrobial agents as empiric therapy (Aldecoa, *et al.*,2022).

5.0 Conclusion

The overall prevalence of urinary tract infections was high among the participants and the incidence varies mainly with ages and sexes and predominantly caused by bacterial mostly *E.*

coli followed by *Staphylococcus aureus*. The risk of urinary tract infection was increased in diabetics patients having previous urinary tract infection history in this study. This study showed a large number of isolate resistances to Amoxicillin except in *Klebsiella pneumoniae*. It's advisable to enhance sensitization work for any nosocomial setting to have regular antibacterial susceptibility investigation equipment and should be carrying out regularly. because, antibacterial susceptibility testing can be beneficial beyond the selection of an antibacterial agent for individual patient therapy. The rate of drug resistance that are obtained from antibacterial data are valuable on both a small scale (institutional) and large scale (regional/international continental). Institutionally, rates of antimicrobial resistance are an important part of epidemiological studies used to determine susceptibility profiles and empirical therapy for "local" bacterial pathogens. This can be used to establish a health Care centres or hospitals and in turn set institutional antibacterial stewardship. On a large scale, the surveillance and monitoring of rates of antibacterial drug resistance are important components of developing robust data sets that will ultimately be used to define or redefine breakpoints or epidemiological cutoff value.

Tables 1: Biochemical and Morphological Characteristics of uropathogenic microorganism isolated from patients.

												Suspected Organisms	
Morphology	Hydrogen sulfide gas production test	Vp test	Mr test	Oxide test	Sugar fermentation Test	Citrate Utilization Test	Indole test	Coagulase Test	Catalase Test	Motility	Shape		Gram reaction
Pinkish	-	-	+	-	+	-	+	-	+	+	Rod	-	<i>Escherichia coli</i>
Yellow	+	+	+	+	+	+	+	-	-	-	Rod	-	<i>Klebsiella pneumoniae</i>
Blue/ Translucent	+	-	+	-	-	+	-	-	-	+	Rod	-	<i>Proteus mirabilis</i>
Greenish blue	-	-	-	+	-	-	-	-	+	+	Rod	-	<i>Pseudomana aeruginosa</i>
Opaque/ Golden yellow	-	-	-	-	+	-	+	+	+	+	Cocci	+	<i>Staphylococcus aureus</i>
Golden yellow	-	-	-	-	-	-	+	-	+	-	Cocci	+	<i>Coagulase negative staphylococci (CONS)</i>

KEY = - Negative + Positive-

Table ;2 Prevalence of Uropathogenic microorganisms isolated from patients attending General hospital, Minna

Organism	Number of Isolates	Percentage%
<i>Escherichia coli</i>	87	30.31
<i>Klebsiella pnemoniae</i>	27	9.41
<i>Proteus mirabilis</i>	33	11.49
<i>Pseudomonas aeruginosa</i>	27	9.41
<i>Staphylococcus aureus</i>	44	15.33
<i>CONs</i>	24	8.36
Total:	287	100%

CONs, Coagulase negative staphylococci

Table 3: Distribution of Uropathogenic microorganisms in relation to the Gender of patients

Uropathogens Isolated	Species	Numbers		percentage %	p. value
		Male	Female		
Bacterial	<i>Escherichia coli</i>	24(8.36)	63(21.95)	30.31	0.005
	<i>Klebsella pneumoniae</i>	9(3.14)	18(6.27)	9.41	
	<i>Proteus mirabilis</i>	10(3.48)	23(8.01)	11.49	
	<i>Pseudomonas aeruginosa</i>	15(5.23)	12(4.18)	9.41	
	<i>Staphylococcus aureus</i>	20(6.97)	24(8.36)	15.33	
	<i>Coagulase negative Staphylococci</i> (CONS)	6(2.10)	18(6.27)	8.37	
Total		93(32.40%)	194(67.60%)	287 (100%)	

The subject, gender is a significant risk factor influencing bacterial infection in patient at $p \leq 0.05$

Table 4 Distribution of Uropathogenic Microorganisms in Respect to The Age of Patient

Uropathorogens	Age (Years)					P-value	
	18-25	26-35	36-45	46-60	60 and Above	Total	
<i>Escherichia coli</i>	7 (2.4)	11 (3.8)	17 (5.9)	32 (11.1)	20 (6.9)	89 (30.1)	
<i>Klebsiella pneumoniae</i>	2(0.7)	5(1.7)	6(2.1)	9(3.1)	5(1.7)	27(9.3)	
<i>Proteus mirabilis</i>	5(1.7)	6(2.1)	4(1.4)	8(2.8)	10(3.5)	33(11.5)	0.05
<i>Pseudomonas aeruginosa</i>	3(1.4)	5(1.7)	4(1.4)	8(2.8)	7(2.4)	27(9.7)	
<i>Staphylococcus aureus</i>	8(2.7)	6(2.1)	10(3.5)	12(4.5)	8(2.8)	27 (9.7)	
<i>Coagulase negative</i>							
<i>staphylococci (cons)</i>	2 (0.7)	3 (1.0)	1 (0.3)	10 (3.5)	8 (2.7)	27 (9.7)	5 (1.7)
Total	32 (11.1)	44 (15.33)	54 (18.81)	93 (32.4)	64 (22.3)	287 (100%)	

The subject age is a significant risk factor influencing bacterial infections in patients at $p \leq 0.05$

Table 5 Distribution of Uropathogenic Microorganism in Relation to the Blood Glucose Level and Types of Diabetes

Uropathogen	Glucose Level		Types of Patients		Total
	126g/ml	Above126g/ml	Non-Diabetic	Diabetic	
<i>E. coli</i>	36 (12.5)	51 (17.8)	40 (13.9)	47(16.4)	87
<i>Klebsiella pneumoniae</i>	11(3.8)	16 (5.6)	7 (2.4)	20 (6.9)	27
<i>Proteus mirabilis</i>	11 (3.8)	22 (7.7)	14 (4.9)	19 (6.6)	33
<i>Pseudomonas aeruginosa</i>	10 (3.5)	17 (5.9)	9 (3.1)	18 (6.3)	27
<i>Staphylococcus aureus</i>	14 (4.9)	30 (10.5)	12 (3.4)	32 (11.2)	44
CONs	24 (1.4)	20 (6.9)	10 (3.4)	14 (4.9)	24
<i>Total</i>	104 (36.24)	183 (63.76%)	107 (37.28%)	180 (62.72%)	287(100%)

The subject glucose level and types of diabetes are significant factors influencing urinary tract infections in patients

Table 6: Frequency of Occurrence of Uropathogenic Microorrganism with Respect to the Marital Status of Patient.

Uropathogens	MARITAL STATUS			Total P-- Value
	Single	Married	W/Divorced	
<i>Escherichia coli</i>	15 (5.2)	52 (18.11)	20 (6.9)	87(30.3)
<i>Klebsiella preumoniae</i>	4 (1.4)	18 (6.3)	5 (1.7)	27 (9.4)
<i>Proteus mirabilis</i>	3 (1.0)	22 (7.7)	8 (2.8)	33 (11.5)
<i>Pseudomona aeruginosa</i>	7 (2.4)	15 (5.2)	5 (1.7)	27 (9.3)
<i>Staphylococcus aureus</i>	8 (2.8)	28 (9.8)	18 (2.8)	44 (15.4)
<i>Coagulase negative</i>				
<i>Staphylococci (CONs)</i>	2 (1.0)	14 (4.9)	8 (2.8)	24 (8.7)
<i>Total</i>	45 (15.68%)	187 (64.11%)	58 (20.20%)	287 (100%)

The subject marital status is a significant risk factor influencing urinary tract infection in patients

Table 7: Socio-Demographic characteristics of patients investigated their uropathogens (400)

Socio- Demographic characteristics	Classification of variables	Frequency No (%)
1. Age (year)	18 – 21	40 (10)
	22 – 30	60 (15)
	31 – 45	82 (20.5)
	46 – 60	126 (31.5)
	61 and above	92 (23)
2. Sex (Gender)	Male	159 (39.75)
	Female	241 (60.25)
3. Address	Urban	224 (56)
	Rural	176 (44)
4. Marital Status	Single	72 (18)
	Married	266 (66.5)
	Widowed/ Divorced	62 (15.5)
5. Literacy Level	Illiterate	223 (55.75)
	Primary School	80 (20)
	Secondary School	50 (12.5)
	Higher Education	47 (11.75)
6. Types of diabetes	Type I	280 (70)
	Type II	120 (30)
7. Duration of (years)	< 5	80 (20)
	5 – 10	122 (30.5)
	≥ 10	198 (49.5)
8. Blood glucose level	126glml	99 (24.75)
	<126glml	301 (75.25)
9. History of previous UTI	Yes	288 (72)
	No	112 (28)
10. History of previous antibiotics	Yes	294 (73.5)
	No	106 (26.5)

Table 8 Antibiotic susceptibility pattern of Gram positive bacterial isolated from urine culture of patients (n=68)

Bacterial isolates	Pattern	AMX	AZI	CRX	CFT	CXM	ERY	GEN	LEV	RMP	STP
Staphylococcus aureus (44)	S	0(0)	40(90.9)	4(9.0)	0(0)	24(54.5)	0(0)	4(9.0)	4(9.1)	0(0)	20(45.5)
	I	0(0)	49.0)	40(91)	0(0)	5(11.4)	0(0)	40(9.1)	0(0)	0(0)	4(9)
	R	44(100)	0(0)	0(0)	44(100)	15(34.1)	44(100)	0(0)	40(90.9	4(100)	20(45.5)
Coagulase negative staphylococci (CONS) (24)	S	0(0)	21(87.5)	20(83.3)	0(0)	2(8.3)	0(0)	7(29.2)	44(16.7)	0(0)	0(0)
	I	7(29.2)	3(12.5)	2(87.5)	24(100)	3(12.5)	5(20.8)	17(70.8)	6(25)	4(16.7)	14(58.3)
	R	17(70.8)	0(0)	2(8.3)	0(0)	19(79.1)	19(79.1)	0(0)	14(58.3)	20(83.3)	10(4.1)
Total = (68)	S	0(0)	61(89.7)	24(35.3)	0(0)	26(38.3)	(0)	11(16.2)	8(11.8)	0(0)	20(29.4)
	I	7(10.3)	7(10.3)	42(1.8)	24(35.3)	8(11.7)	6(7.4)	57(83.8)	6(8.8)	4(5.9)	4(5.9)
	R	61(89.7)	0(0)	2(2.9)	44(64.7)	34(50)	63(92.6)	0(0)	54(79.4)	64(94.1)	30(44)

Abbreviation: R =Resistance, S = Sensitive, I = Intermediate, AMX: Amoxicillin, AZI: Azithromycin, CPX; Ciprofloxacin, CFT: Ceftazidine, CXM: Cefuroxime, ERY: Erythromycin, GEN: Gentamycin, LEV: Levofloxacin, RFP: Rifampicin, STP: Streptomycin.

Table 9: Antibiotic susceptibility pattern of Gram negative bacterial isolated from urine culture of patients (n=174)

Bacterial isolates	Pattern	AMP NO	AZI NO	CRX NO	CFT NO	CXM NO	ERY NO	GEN NO	LEV NO	RMP NO	STP NO
<i>Escherichia coli</i> (87)	S	0(0)	50(57.5)	0(0)	0(0)	20(22.9)	50(57)	60(69)	60(69)	70(80)	40(46)
	I	2(2.3)	7(8)	82(94.3)	1(1.1)	31(35)	0(0)	20(23)	27(31)	15(17)	30(34)
	R	85(97.7)	30(34.5)	5(5.7)	86(98.9)	36(41)	37(43)	7(8)	0(0)	2(2.3)	17(20)
<i>Klebsiella pneumoniae</i> (27)	S	20(74.1)	10(37)	5(18.5)	10(37)	6(22.2)	0(0)	20(74)	15(55)	10(35)	8(30)
	I	0(0)	12(44.4)	5(18.5)	10(37)	4(14.8)	0(0)	0(0)	5(18)	6(22.2)	3(11.1)
	R	7(25.9)	5(18.5)	17(63)	7(26)	17(63)	27(100)	7(36)	7(36)	11(40.7)	16(59.3)
<i>Proteus mirabilis</i> (33)	S	0(0)	3(9.1)	30(90.1)	21(63.6)	0(0)	33(100)	11(33.3)	33(100)	30(90.8)	30(90.8)
	I	3(9.1)	15(46.3)	3(9.1)	1(3.0)	0(0)	0(0)	21(63.6)	0(0)	3(9.1)	3(9.1)
	R	30(90.9)	15(45.5)	0(0)	11(33.3)	33(100)	0(0)	0(0)	0(0)	0(0)	0(0)
<i>Pseudomonas aeruginosa</i> (27)	S	0(0)	0(0)	27(100)	20(74.4)	21(78)	0(0)	27(100)	20(74)	20(74)	27(100)
	I	7(26)	27(100)	0(0)	7(26)	6(22)	0(0)	0(0)	3(11.2)	2(7.4)	0(0)
	R	20(74)	0(0)	0(0)	0(0)	0(0)	27(100)	0(0)	4(15)	5(18.5)	0(0)
TOTAL =174	S	20(12)	63(36)	62(36)	51(29)	47(27)	83(48)	118(68)	128(74)	130(75)	105(60)
	I	12(7)	61(35)	90(52)	19(21)	56(32)	0(0)	31(18)	35(20)	26(15)	36(21)
	R	142(81)	50(29)	22(13)	104(60)	86(49)	91(52)	14(8)	11(6)	18(10)	33(19)

Abbreviation: R Resistance, S Sensitive, I intermediate, AMX Amoxicillin, AZI Azithromycin, CPX Ciprofloxacin, CFT Ceftazidine, CXM Ceuroxine, ERY Erythromycin, GEN Gentamycin, LEV Levofloxacin, RFP Rifampicin, STP Streptomycin.

Table 10: Multi-Drug Resistance Pattern of Gram-Positive Bacterial isolated from urine culture of patients attending General Hospital

Resistance pattern	S aureus	(CONS)	Total	MDRI
AMX/CRX/AZI	5 (15.6)	1 (1.5)	6	0.3
AMX/CFT/CXM	1 (3.1)	1 (1.5)	2	0.3
AMX/ERY/LEV	4 (2.5)	1 (1.5)	5	0.3
AMX/AZI/CRX/CFQ/CXM	2 (6.3)	- (0)	2	0.5
AMX/AZI/CRX/CFT/CXM/ERY	3 (9.4)	- (0)	3	0.6
CPX/CFT/ERY	1 (3.1)	- (0)	1	0.3
CPX/LEV/ERY/RMP	1(3.1)	2 (2.9)	3	0.4
AZI/CPX/CFT/ERY	2 (6.3)	1 (1.5)	3	0.4
ERY/GEN/LEV	3 (9.4)	3 (4.4)	6	0.3
ERY/RMP/LEV/AZI	3 (9.4)	2 (2.9)	5	0.4
GEN/RMP/STP	2 (6.3)	1 (1.5)	3	0.3
CXM/ERY/RMP/STP	5(15.6)	2 (2.9)	7	0.4
Total	32 (47.1%)	16 (23.53)	48(70.6%)	

Abbreviation: R Resistance, S Sensitive, I intermediate, AMX Amoxicillin, AZI Azithromycin, CPX Ciprofloxacin, CFT Ceftazidine, CXM Ceuroxine, ERY Erythromycin, GEN Gentamycin, LEV Levofloxacin, RFP Rifampicin, STP Streptomycin.

Table 11: Multi-Drug Resistance Pattern of Gram-Negative bacterial isolates from urine culture of patients attending General Hospital

Resistance Pattern	<i>E. coli</i>	<i>K. pneumoniae</i>	<i>P.mirabilis</i>	<i>P.aeruginosa</i>	Total	MDRI
AMX/AZI/CXM	10(11.5)	2(7.4%)	1(3.0%)	1(3.7%)	14	0.3
AMX/CPX/AZI/CFT	5(5.8)	1(3.7%)	1(3.0%)	1(3.7%)	8	0.4
AMX/CFT/ERY	7(8.1)	2(7.4%)	1(3.0%)	1(3.7%)	11	0.3
AMX/CXM/LEV	1 (1.2)	1(3.7%)	1(3.0%)	0(0%)	3	0.33
AMX/ERY/GEN/STP	7(8.1)	1(3.7%)	0(0%)	2(7.4%)	10	0.4
AMX/AZI/CPX/LEV/RMP	1 (1.2)	1(3.7%)	0(0%)	0(0%)	2	0.5
CFT/AZI/GEN/RMP	2(2.3)	1(3.7%)	1(3.0%)	0(0%)	4	0.4
AMX/CPX/CFT/CXM/ERY	5(5.8)	0(0%)	2(6.1%)	1(3.7%)	8	0.5
CPX/ERY/GEN	5(.8)	1(3.7%)	2(6.1%)	1(3.7%)	9	0.3
CFT/ERY/GEN/LEV	1.(1.2)	0(0%)	3(9.1%)	2(7.4%)	6	0.4
ERY/GEN/STP	7(8.1)	0(0%)	0 (0%)	1(3.7%)	8	0.3
ERY/RMP/STP	2(2.3)	1(3.7%)	1(3.0%)	0(0%)	4	0.3
GEN/AZI/RMP	2(2.3)	2(7.4%)	0 (0%)	1(3.7%)	5	0.3
AMX/CPX/CXM/STP/AZI	5(5.8)	2(7.4%)	0 (0%)	0(0%)	7	0.5
RMP/STP/RMP/ERY	2(2.3)	1(3.7%)	0 (0%)	0(0%)	3	0.4
Total	62(71.3%)	16(59.3%)	13(39.39%)	11 (40.74%)	102(5.30%)	

Abbreviation: R Resistance, S Sensitive, I intermediate, AMX Amoxicillin, AZI Azithromycin, CPX Ciprofloxacin, CFT Ceftazidine, CXM Cefuroxime, ERY Erythromycin, GEN Gentamycin, LEV Levofloxacin, RFP Rifampicin, STP Streptomycin

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Authors Contributions

US collected the samples, analysed the data and write the main manuscript, NUA, supervised the main manuscript AH conducted laboratory analysis and TGM designed the study.

Declaration of Conflicting interest

The authors declared no any conflict with respect to this research

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Ethical Approval

Ethical approval was obtained from Niger State Ministry of Health Ethics Committee and from the Research, Ethics and publication committee of General Hospital, Minna, Niger State, Nigeria

Consent to participate

The patient's consent was sought by filling the necessary form. Hence, the participation of the patient was voluntary.

1. Find the values:

To find the values chi-square must be applied to see the association between the current urinary tract infections status and growth of uropathogenic microorganisms among the study populations.

$$\text{Chi-Square} = x^2 = \sum \frac{(c - E)^2}{E}$$

Where C= Observed value or observed frequency

E= Expected value or expected frequency

Σ = *summation or summarizing of total*

Calculated Value = 1269.9

df= (r-1) (7-1) (99-1) = 6 x 98 = 588

df = 588

Tabulated Value = 12.6

P = 0.05 > tabulated value 12.6

Calculated value which is 1269.9 is greater than tabulated value which is 12.6, so there is great association between the current urinary tract infection and growth of uropathogenic microorganisms.

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Figures

Image not available with this version

Figure 1

3.1 Legend not included with this version