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Research Article

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Bacteriological and Immunological Study of Chronic Failure Patients Undergoing for Hemodialysis

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Abstract

Kidney failure usually occurs as a result of lacking the normal function of kidney as either partly or completely. The objective of this study is to evaluate the relation between auto immunity and renal failure and to detect the types of bacteria which responsible of urinary tract infections in renal failure patients, and study their susceptibility against antibiotics. The study was carried out in dialysis unit in Tikrit Teaching Hospital in Tikrit city, at the period from the first of March until the 15th of July 2019. A total of 140 midstream urine samples were collected from chronic kidney disease patients and inoculated on blood agar and MacConkey agar and incubated aerobically at 37˚C for 18-24 hr.

Identification of bacterial isolates was done by using standard micro biological techniques. The antibiotic susceptible test of all the strains was determined by using modified Kirby Bauer disc diffusion method. 5 ml of blood samples were pulled from each patient for Antinuclear Antibody test (ANA). Out of 140 serum sample of chronic kidney disease patients 58 (41.4%) were showed positive Antinuclear Antibody test, whereas 82 (58.6%) showed negative result for ANA. Among 58 of positive Antinuclear Antibody, 26 (44.8%) were male and 32 (55.2%) were female. Out of 140 urine samples 36 (26%) were positive bacterial culture whereas 104 (74%) were negative bacterial culture. Among Gram negative isolates *E.coli* (36.1%) was found to be most predominant organism followed by *Proteus vulgaris* (16.7%), *Pseudomonas aeruginosa* (11.1%), *Morganella morgani* (8.3%), *Klebsiella penumoniae* (2.8%) and *Proteus mirabilis* (2.8%) and among Gram positive only one type of organism was isolated *Staphylococcus saprophyticus* (22.2%), and among generally used antibiotics for UTI Gram negative isolates showed (96.4%) sensitivity towards Imipeneme followed by Gentamicin (89.3%). Nitrofurantain (75%), cotrimoxazole (71.4%) and chloramphenicol (64.3%). The least effective antibiotics for Gram negative were found to be Amoxicillin. The antibiotics, Imipeneme (87.5%). Oxacillin (87.5%) and Gentamicin (75%) were found to be most effective, whereas Erythromycin and Amoxicillin were found to be least effective drug against Gram positive isolates.

Keywords: Chronic kidney disease • Hemodialysis • Urinary tract infections • ANA test

Introduction

Autoimmunity resulting in renal injury occurs as a systemic disturbance of immunity with the central feature being loss of tolerance to normal cellular and/or extracellular proteins. Some of the target auto antigens are now identified in autoimmune diseases where tissue injury includes the kidney [1]. Kidney diseases may cause kidney failure, and kidney diseases are typically classified as either acute or chronic. Acute kidney failure in which the kidneys are completely. Losses their function suddenly, which encompasses both injury (structural damage) and impairment (loss of function) [2], and chronic kidney failure, in which the kidneys are gradually losses of their functions over 3 months [3].

In renal diseases, a change in the composition of urine in anuria , oliguria , hematuria and albuminuria is observed, The resultant changes in PH, in osmolality and urinary urea have their own effects uremic toxins inhibit the antimicrobial activity of these conditions may support the development of UTI in patients with renal disease [4,5].

A urinary tract infection (UTI) is a bacterial infection that affects part of the urinary tract. When it affects the Lower urinary tract, it is known as a simple cystitis (a bladder infection) and when it affects the upper urinary tract it is known as pyelonephritis (a kidney infection) [6,7]. One million patients visit the emergency department, and 100.000 hospitals stay every year in the United States and due to UTIs. Approximately 10% of humans will have UTI at some time during their lives; of note UTIs are also the most common hospital acquired infection accounting for as among as 35% of nosocomial infection, the exact prevalence of UTIs is age and sex-dependent. During the first year of life, the incidence of UTIs among the males remains relatively low after 1 year of age and approximately 60 years of age when the enlargement of the prostate interferes. UTI is predominantly a disease of females [8].

Escherichia coli are the cause of 80-85% of urinary tract infections, with *Staphylococcus saprophyticus* being the cause in 5-10%. Rarely may they be due to viral or fungal infections. Other bacterial causes include *Klebsiella, Proteus*, *Pseudomonas* and *Enterobacter*. These are uncommon and typically related to abnormalities of the urinary system or urinary catheterization [9].

Materials and Methods

A cross sectional study was conducted in the period from the first of March until the 15th of July 2019 at Dialysis unit in Tikrit Teaching Hospital in Tikrit city. A total of 140 midstream urine samples were collected from chronic kidney disease patients and inoculated in blood agar and MacConkey agar and incubated aerobically at 37˚C for 18-24 hour. Macroscopic (turbidity, odor and color), and microscopic (WBC and RBC) were done. Identification of the bacterial isolates was done by using standard microbiological techniques which involve the morphological appearance of the colonies. Gram's staining reaction and biochemical properties like catalase production test oxidase test and coagulase test, in dole production test. Methyl red test, Voges-Proskaure (VP) test, Citrate Utilization test, Triple Sugar Iron (TSI) agar test and Urea Hydrolysis test. The antibiotic susceptibility pattern of all the strains was determined by using modified Kirby Bauer disc diffusion method. 5 ml of blood samples were pulled from each patient by using sterile syringes, the blood were

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left at room temperature about 10-15 min. Fresh serum were collected and kept in deep freeze (-20°C) for serological test of Antinuclear Antibody test (ANA), by using the manufacturer's steps.

Results

Out of 140 serum samples of chronic kidney disease patients 58(41.4%) were showed positive anti-nuclear antibody test, whereas 82 (58.6%) showed negative results (Table 1).

Among 58 of positive Antinuclear Antibody test, 26(44.8%) were male and 32(55.2%) were female (Table 2).

Out of 140 urine samples of chronic kidney disease patients 36(26%) were showed bacterial culture positive result, whereas 104 (74%) showed culture negative result (Table 3).

Both Gram negative as well as Gram positive organisms was found to be responsible for urinary tract infection. Among Gram negative isolates *E.coli* (36.1%) was found to be most predominant organisms followed by *Proteus vulgaris* (16.7%) Pseudomonas *aeruginosa* (11.1%), *Morganella morgani* (8.3%), *Klebsiella pneumoniae* (2.8%), and *Proteus mirabilis* (2.8%), and among Gram positive only one type of organism was isolated *Staphylococcus saprophyticus* (22.2%) (Table 4).

Among generally used antibiotics for UTI Gram negative isolates showed (96.4%) sensitivity towards Imipeneme followed by Gentamicin (89.3%), Nitrofurantain (75%), cotrimoxazole (71.4%) and chloramphenicol (64.3%). The least effective antibiotics for Gram negative were found to be Amoxicillin (10.7%) (Table 5).

The antibiotics Imipeneme (87.5%), Oxacillin(87.5%) and Gentamicin (75%) were found to be most effective whereas Erythromycin and Amoxicillin were found to be least effective drug against Gram positive isolates (Table 6).

Table 1. The association between Antinuclear Antibody test and chronic kidney disease.

Chronic kidney disease patients	ANA +ve		ANA -ve		Total	
	No.	%	No.	%	No.	%
	58	41.4	82	58.6	140	100

Table 2. Distribution of positive ANA patients according to gender.

Gender	Male		Female		Total	
Positive ANA patients	No.	%	No.	%	No.	%
	26	44.8	32	55.2	58	100

Table 3. Growth pattern of bacteria.

Growth	No. of samples	Percentage of samples (%)
Culture positive	36	26
Culture negative	104	74
Total	140	100

Table 4. Bacterial isolates frequency.

Bacterial isolates	Frequency	Percentage (%)
Escherichia coli	13	36.1
Klebsiella pneumonia	1	2.8
Proteus mirabilis	1	2.8
Proteus vulgaris	6	16.7
Pseudomonas aeruginosa	4	11.1
Morganella morgani	3	8.3
Staphylococcus saprophyticus	8	22.2
Total	36	100

Table 5. Antibiotic Susceptibility Pattern of Gram negative bacterial isolates.

Antibiotic used	Antibiotic susceptibility pattern		
	Sensitive No. (%)	Resistant No. (%)	Total No.
Amoxycillin	3(10.7)	25(89.3)	28
Cefuriazone	12(42.9)	16(57.1)	28
Cephalexin	13(46.4)	15(53.6)	28
Chloramphenicol	18(64.3)	10(35.7)	28

Ciprofloxacin	16(57.1)	12(42.9)	28
Cotrimoxazole	20(71.4)	8(28.6)	28
Gentamicin	25(89.3)	3(10.7)	28
Imipeneme	27(96.4)	1(3.6)	28
Nitrofurantain	21(75)	7(25)	28
Norofloxacin	15(53.6)	13(46.4)	28

Table 6. Antibiotic Susceptibility Pattern of Gram positive bacterial isolates.

Antibiotic used	Antibiotic susceptibility pattern		
	Sensitive No. (%)	Resistant No. (%)	Total No.
Amoxycillin	0	8(100)	8
Ceftrixone	3(37.5)	5(62.5)	8
Chloramphenicol	4(50)	4(50)	8
Ciprofloxacin	2(25)	6(75)	8
Cotrimoxazole	5(62.5)	3(37.5)	8
Erythromycin	0	8(100)	8
Gentamicin	6(75)	2(25)	8
Imipeneme	7(87.5)	1(12.5)	8
Nitrofurantain	4(50)	4(50)	8
Norofloxacin	3(37.5)	5(62.5)	8
Oxacillin	7(87.5)	1(12.5)	8

Discussion

The present study was carried out in Dialysis Unit in Tikrit Teaching Hospital in Titkrit City. Out of 140 serum samples of chronic kidney disease patients 58(41.4%) were positive for Anti-Nuclear Antibody (ANA) test, whereas 82(58.6%) were negative. Among 58 of positive ANA test, 26 (44.8%) were male and 32 (55.2%) were female. Inflammatory renal disease in the autoimmunity occurs because the kidney is targeted by effector responses [10]. Within the kidney, the local response of resident cells plays an important role in determining the severity of inflammation. If severe and/or unlimited, these events may lead to fibrosis and organ failure, the intensity and severity of inflammation and fibrosis are also influenced by genetic factors (e.g., that determine the fibro genic response) [11]. For most autoimmune diseases there is a clear sex difference in prevalence. Where by females are generally more frequently affected than males, this may be due to pregnancy and sex hormones [12,13]. Out of the 140 urine samples of chronic kidney disease patients 36(26%) were positive bacterial culture, whereas 104 (74%) were negative. A study which was done in Iran in 2011 showed the isolation rate to be 35.5% [14]. A similar study which was done in Baghdad showed a higher positive growth rate of 62% [15]. The reason behind obtaining low rate of positive bacterial culture in this study may be due to the fact that the patients are undergoing for various treatments, or due to show growing organisms or those organisms that were not able to grow on the routine media we used, or the reason of chronic kidney infection.

Among Gram negative isolates *E.coli* (36.1%) was found to be most predominant organisms followed by *Proteus vulgaris* (16.7%). Pseudomonas *aeruginosa* (11.1%), Morganella morgana (8.3%) *Klebsiella pneumoniae* (2.8%), and *Proteus mirabilis* (2.8%), and among Gram positive isolate only one, type of organism was isolated *Staphylococcus saprophyticus* (22.2%). Among antibiotics used for urinary tract infection, Gram negative isolates showed (96.4%) sensitivity towards Imipeneme followed by Gentamicin (89.3%). Nitrofurantain (75%). Cotrimoxazole (71.4) and chloramphenicol (64.3%). The least effective antibiotics for Gram negative were found to be Amoxicillin (10.7%). The antibiotics Imipeneme (87.5%), Oxacillin (87.5%) and Gentamicin (75%) were found to be most effective whereas Erythromycin and Amoxicillin were found to be least effective drug against Gram positive isolates. Other study by Muhsinin Iraq who found that *E.coli* isolates were resistant to ceftriaxone and ceftazidim in 100%. A study by Mirzarzi reported that among *E.coli* isolates the highest antibiotic resistance was related to nalidixic acid and trimethoprim-sulfamethoxzcole, and K.pneumoniae isolates were the most antibiotic resistant to trimethoprim-sulfamethoxazole, ciprofloxacin, and nalidixic acid [16]. In another study, it was determined that *E.coli* was the most frequent isolate with resistance to ampicillin, with 69.5% and that a high or increasing resistance to trimethoprim-sulfamethoxazole [17,18].

Conclusion

The antibiotic susceptible test of all the strains was determined through Antinuclear Antibody test and examined the results. It was determined that *E.coli* was the most frequent isolate with resistance to ampicillin, with a high or increasing resistance to trimethoprim-sulfamethoxazole. The antibiotics such as Imipeneme, Oxacillin and Gentamicin were found to be most effective whereas Erythromycin and Amoxicillin were found to be least effective drug against Gram positive isolates. The low rate of positive bacterial culture in this study could be due to the fact that the patients are undergoing various treatments, or it could be due to the presence of growing organisms or organisms that were unable to grow, or it could be due to chronic kidney infection.

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