بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

قال الله تعالى

يَرْفَعِ اللَّهُ الَّذِينَ آمَنُوا مِنْكُمْ وَالَّذِينَ أُوتُوا الْعِلْمَ دَرَجَاتٍ وَاللَّهُ بِمَا تَعْمَلُونَ خَبِيرٌ

صدق الله العظيم

سورة المجادلة الآية 11

Dedication

To my parent
To my brothers and
my sister
To my extended

family

Acknowledgement

First of all unlimited thanks to ALLAH for giving me strength and power to complete this study, after that I would like to thank my supervisor Dr. Humodi Ahmed Saeed, Dean college of Medical Laboratory Science, Sudan University of Science and Technology for his patience. Then give my great thank to my colleagues in bacteriology department in National Health Laboratory especially Mohammed Abdelrahman "Garja" the senior technologist in bacteriology department for his help and support. Also I thank the staff of Microbiology lab in Sudan University of Science and Technology for their help.

Abstract

This study had been carried out in Khartoum State during period between August 2005 to October 2005. The study evaluate the potency of third generation cephalosporins (Ceftazidime, Ceftraixone, Cefotaxime, cefoprazone) against infection caused by *Pseudomonas aerugninosa*.

Specimens were collected from different hospitals, include Khartoum Teaching hospital 113 (38%), Ear Nose Throat hospital 96 (33%), Military hospital 58 (20%) and National Health laboratory 26 (9%).

Different types of bacteria isolated including *Staphylococcus aureus* 78(28%), *Pseudomonas aeruginosa* 68(24%), *Escherichia coli* 49(18%), *Klebsiella spp* 40 (14%), *Proteus mirabilis* 38(13%), *Citrobacter spp* 5(2%), *Proteus vulgaris* 2(1%).

Pseudomonas aeruginosa, isolates were recovered from infected wound 36/68 (53%), infected ear 24/68(35%), and urine 8/68(12%).

The study showed that ceftazidime was most effective (91%) followed by cefoprazone (47%), ceftriaxone (7%) and cefotaxime (3%).

The results also indicated that the activity of the antibiotics under test against organisms isolated from males and females were 36% and 39% respectively.

ملخص الأطيروحة

تم تنفيذ هذه الدراسة في ولاية الخرطوم في الفترة من أغسطس 2005م وحتى أكتوبر 2005م. تذاولت الدراسة تقييم فاعلية المجموعة الثالثة من مضادات السفالوس بورينات (سفتازديم، السفتريكسون، السفوتاكسيم والسفوبيرازون) لعلاج الإلتهابات المسببة بواسطة باكتيريا الزائفة الزنجارية. تم جمع عينات طبية من عدة مستشفيات شملت مستشفى الخرطوم 113 عينة وتمثل 38%، مستشفى الأذن والانف والحنجرة 96 عينة وتمثل 38% ومستشفى السلاح الطبي 58 عينة وتمثل 20% والمعمل القومي الصحي 20 عينة وتمثل 90 عينة وتمثل 90 عينة وتمثل 90%.

تم عزل وتحديد أنواع مختلفة من الباكتيريا شملت العن قودية الذهبية 78 سلالة وتمثل 24% والإشريكية سلالة وتمثل 24% والإشريكية الا قولونية 49 سلالة وتمثل 14%، أنواع الكبسيلا 40 سلالة وتمثل 14%، المت قلبة الرائعة 38 سلالة وتمثل 13%، أنواع الستروباكتر 5 سلالات وتمثل 2% وأخيراً المت قلبة الإعتيادية سلالتين وتمثل 1%.

مثلت باكتيريا الزائفة الزنجارية المعزولة من مسحات الجروح الملتهبة 36 سلالة (53%)، مسحات الآذان الملتهبة 24 سلالة وتمثل 35%، وعينات البول 8 سلالات وتمثل 12%.

النتائج المستخلصة أثبتت أن السيفتازديم هو أكثر المضادات الحيودة كفاءة (91%) يتبعها السيفوبيرازون (47%)، السيفترايكسون (7%) أخيراً السيفوتاكسيم (3%). النتائج أيضاً أشارت إلى أن نشاط المضادات الحيودة تحت الإختبار ضد الميكروب المعزول من الذكور والإذات هي 36% و 39% على التوالى.

Table of contents

		Page
	Acknowledgement	i
	Abstract	ii
	ملخص الأطروحة	iii
	Table of contents	iv
	List of tables	ix
	List of plates	xi
	List of figures	xii
	Chapter One: Introduction and Literature Revie	W
1.	Introduction and Literature Review	1
1.1.	Antibiotic	1
1.1.1	Definition	1
1.1.2	Cephalosporins	2
1.1.2.1	Mode of action	2 2 2 3
1.1.2.2	Phamacokinetics	2
1.1.2.3	Adverse effects	3
1.1.2.4	Classification	3
1.1.2.4.1	First generation	3
1.1.2.4.2	Second generation	4
1.1.2.4.3	Third generation	4
1.1.2.4.4	Fourth generation	6
1.2.	Pseudomonas aeruginosa	7
1.2.1	Definition	7
1.2.2	Normal habitat	7
1.2.3	Antigenic structure	7
1.2.4	Pathogensis	8
1.2.5	Clinical finding	9
1.2.6	Laboratory diagnosis	9
1.2.6.1	Specimen	9
1.2.6.2	Morphology and staining	9
1.2.6.3	Culture	9
1.2.6.4	Biochemical reaction	10
1.2.7	Treatment	10
1.3	Other Pseudomonas species	11
1.3.1	Ps. pseudomallei	11
1.4	Microbial resistance	12
1.5	Mechanism of resistance	13
1.6	Resistance of <i>Ps. areuginosa</i> to cephalosporins	14
1.6.1	Resistance of <i>Ps. Aeruginosa</i> to ceftriaxone	15
162	Resistance of Ps. <i>aeruainosa</i> to cefotaxime	16

1.6.3	Resistance of <i>Ps. aeruginosa</i> to cefoperazone	16
1.6.4	Resistance of Ps. aeruginosa to ceftazidime	16
1.7	Problem of the research	18
1.8	Objectives	18
1.8.1	General objective	18
1.8.2.	Specific objectives	18
	Chapter Two: Materials and Methods	
2.	Materials and Methods	19
2.1	Study area	19
2.2	Subjects	19
2.3	Sample size	19
2.4	Site of collection	19
2.5	Age group	19
2.6	Sterilization	19
2.6.1	Glass ware	20
2.6.2.	Media	20
2.6.2.1	MacConkey agar	20
2.6.2.2	Blood agar	20
2.6.2.3	Nutrient agar	20
2.6.2.4	Mueller- Hinton	20
2.6.2.5	Cystine lactose electrolyte deficient (CLED)	20
2.6.2.6	Kligler iron agar	20
2.6.2.7	Simmon's citrate agar	20
2.6.2.8	Urea agar	20
2.6.2.9	Peptone water	21
2.7	Types of stains	21
2.7.1	A set of gram stain	21
2.8	Type of reagents	21
2.8.1	Oxidase reagent	21
2.8.2	Kovacs reagent	21
2.8.3	Sodium chloride (Normal saline '0.85% w/v').	21
2.8.4	MacFarland standard	21
2.8.5	Hydrogen peroxide	21
2.8.6	Plasma	21
2.9	Standard strains	21
2.10	Antimicrobial Discs	21
2.11	Experimental work	22
2.11.1	Collection of specimens	22
2.11.2	Inoculation	22
2.11.3	Incubation	22
2.11.4	Examination of growth	22

2.11.5	Identification	22
2.11.5.1	Gram stain	22
2.11.5.2	Biochemical tests	23
2.11.5.2.1	Oxidase test	23
2.11.5.2.2	Citrate utilization	23
2.11.5.2.3	Fermentation of sugars, production of gas and acid	23
2.11.5.2.4	Indole production	24
2.11.5.2.5	Urease test	24
2.11.5.2.6	Catalase test	24
2.11.5.2.7	Coagulase test	24
2.11.6	Susceptibility test	25
2.11.6.1	Preparation of inocula	25
2.11.6.2	Inoculation of Mueller -Hinton agar	25
2.11.6.3	Antimicrobial disc application	25
2.11.6.4	Incubation	25
2.11.6.5	Reading and interpretation	25
	Chapter Three: Results	
3.	Results	26
3.1	Clinical specimens	26
3.2	Identification of Ps. aeruginosa	26
3.3	Susceptibility test	39
3.3.1	Ceftazidime	43
3.3.2	Cefoperazone	44
3.3.3	Cefotaxime	45
3.3.4	Ceftriaxone	46
3.3.5	Quality control	47
	Chapter Four: Discussion	
4	Discussion	55

	Chapter Five: Conclusion and Recommendations		
5.	Conclusion and recommendations	58	
5.1	Conclusion	58	
5.2	Recommendations	59	
	References	60	
	Questionnaire	65	
	Table 2 NCCLS		
	Table 3 NCCLS		

List of tables

NO	Tables	Page
Table 1	Distribution of specimens according to Hospitals	26
Table 2	Distribution of specimens according to sex	27
Table 3	Distribution of specimens according to age groups	27
Table 4	Distribution of specimens according to the sites of	27
	collection from both males and females	
Table 5	Percentage of significant and insignificant growth	28
Table 6	Colony size after primary isolation	28
Table 7	Lactose fermentation pattern on MacConkey's agar	28
	after overnight incubation	
Table 8	Haemolytic activity	28
Table 9	Gram reaction and cell morphology	29
Table 10	Organisms isolated	29
Table 11	Biochemical test for Gram +ve	30
Table 12	Biochemical test for Gram –ve	32
Table 13	Distribution of <i>Ps. aeruginosa</i> according to site of	37
	infection	
Table 14	Distribution of <i>Ps. aeruginosa</i> according to the sex	37
Table 15	Distribution of Ps. aeruginosa according to site of	37
	infection among males and females	
Table 16	Distribution of <i>Ps. aeruginosa</i> according to hospitals	38
Table 17	Distribution of <i>Ps. aeruginosa</i> according to age	38
	group	
Table 18	Diameters of zones inhibition of antimicrobial	39
	against ps. aeruginosa.	
Table 19	Susceptibility of <i>Ps. aeruginosa</i> to Ceftazidime,	41
	Cefoperazone, Cefotaxime and Ceftriaxone	
	according to Table 2NCCL	
Table 20	Susceptibility of <i>Ps. aeruginosa</i> (n = 68) to	41
	Ceftazidime, Cefoperazone, Cefotaxime and	
	Ceftriaxone	
Table 21	Activity of ceftazidime against <i>Ps.aeruginosa</i> (n=68)	43
	according to site of infection	
Table 22	Activity of ceftazidime against <i>Ps.aeruginosa</i> (n=68)	43
	according to age group	

Table 23	Activity of ceftazidime against <i>Ps.aeruginosa</i> (n=68)	44
	according to sex	
Table 24	Activit of cefoperazone against <i>Ps.aeruginosa</i> (n=68)	44
Table 25	according to site of infection Activity of cefoperazone against <i>Ps.aeruginosa</i> (n=68)	44
Table 26	According to age group Activityof cefoperazone against <i>Ps.aeruginosa</i> (n=68)	45
Table 27	according to sex Activity of cefotaxime against <i>Ps.aeruginosa</i> (n=68)	45
Table 28	according to site of infection Activity of cefotaxime against <i>Ps.aeruginosa</i> (n=68)	45
Table 29	according to age group Activity of cefotaxime against <i>Ps.aeruginosa</i> (n=68)	46
	according to sex	
Table 30	Activity of ceftriaxone against $Ps.aeruginosa(n=68)$	46
Table 31	according to site of infection Activity of ceftriaxone against <i>Ps.aeruginosa</i> (n=68)	46
Table 32	according to age group Activity of ceftriaxone against <i>Ps.aeruginosa</i> (n=68)	47
Table 33	according to sex Quality control	47

List of plates

NO	Plates	Page
Plate (1)	Growth of Ps. aeruginosa on MacConkey agar	48
Plate (2)	Growth of <i>Ps. aeruginosa</i> on CLED	48
Plate (3)	Growth of <i>Ps. aeruginosa</i> on nutrient agar showed	49
Plate (4)	green pigment Growth of <i>Ps. aeruginosa</i> on Blood agar	49
Plate (5)	Growth of <i>Ps. aeruginosa</i> on nutrient agar showed	50
1 late (5)		50
D1 (6)	yellow-red pigment	
Plate (6)	Growth of <i>Ps. aeruginosa</i> on nutrient agar after 48	50
	hours the red pigment overlapped	
Plate (7)	Positive oxidase test for <i>Ps. aeruginosa</i>	51
Plate (8)	Citrate utilization test, left positive test of <i>Ps</i> .	51
Plate (9)	aeruginosa, right negative test of <i>E. coli</i> KIA test, left, red slope, red butt of <i>Ps. aeruginosa</i> ,	52
	middle yellow slope, yellow butt of <i>E. coli</i> , right red	
Plate (10)	slope, yellow butt, H_2S production of $Ps.$ mirabilis Susceptibility testing of standard strain of $Ps.$	52
Plate (11)	aeruginosa ATCC 27853 Susceptibility testing of <i>Ps. aeruginosa</i> on Mueller-	53
Plate (12)	Hinton agar with green yellow pigment Susceptibility testing of <i>Ps. aeruginosa</i> showed	53
Plate (13)	ceftazidime sensitive Susceptibility testing of <i>Ps. aeruginosa</i> showed	54
	multidrug resistance with yellow- red pigment	

List of Figures

Figure (1)	Susceptibility of $Ps.$ aeruginosa (n = 68) to	42
	ceftazidime, cefoperazone, cefotaxime and	
	ceftriaxone	