ANTIMICROBIAL SUSCEPTIBILITY PATTERN OF BACTERIAL ISOLATES FROM CLINICAL SPECIMEN AMONG CHILDREN IN JIMMA UNIVERSITY SPECIALIZED HOSPITAL

BY KUSSIA AYANO (MD)

ARESEARCH PAPER TO BE SUBMITTED TO JIMMA UNIVERSITY COLLEGE OF PUBLIC HEALTH AND MEDICAL SCIENCES SCHOOL OF POSTGRAGUATE AND DEPARTMENT OF PEDIATRICS AND CHILD HEALTH FOR PARTIAL FULFULLIMENT OF CERTIFICATE IN PEDIATRICS AND CHILD HEALTH SPECIALITY

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ABSTRACT

Objective: the aim of this study is to assess antimicrobial susceptibility pattern of bacteria isolated from clinical specimen among children visiting Jimma university specialized hospital.

Methods and Materials:

Clinical samples were collected from children presented to Jimma university specialized hospital from April 1, 2014-June 30, 2014 for infections in different systems. Culture and antimicrobial susceptibility pattern of clinical isolates for commonly used antibiotics was done using standard microbiologic techniques. Data were entered and analyzed by using SPSS version 20 soft ware and p value of < o.o5 was considered statistically significant.

Results

A total of 230 clinical specimen including: soft tissue abscess, eye discharge and body fluids were analyzed and 72 bacteria were isolated(31.3%) with the highest percentage recovered from soft tissue abscess (40.2%) followed by ear discharge (26.4%).From the total bacterial isolates 56(78%) were gram positive and staphylococcus aureus was the predominant isolate 47(65.3%) followed by streptococci pneumonia 7(9.7%). Majority of isolates from soft tissue abscess were staphylococcus aureus(86%).Staphylococcus aureus was the commonest organism cultured from ear discharge 11(57.9%) followed by proteus 4(21.1%),klebsiella ,2(10.5%) and E.coli 1(5.3%).

Staphylococcus aureus showed high level of resistance for commonly used antibiotics: ampicilline (89.4%) , chloramphenicole(89.4%) ,gentamicin (74.5%),cloxacilline (55.3%) and ceftriaxone(44.7%). Resistance rate for klepsella spp.was high for all antibiotics for which it was tested i.e; 83 % for ampicilline,chloramphenicole,gentamicin and ceftriaxone. Streptococcus pneumonia showed better susceptibility for most of the antibiotics for which it was tested: chloramphenicole(14.3%),ampicilline(28.6%) and ceftriaxone (14.3%). There was no statistically significant difference in staphylococcus aures susceptibility pattern between in-patients and out patient isolates (P -value.838) nor between naïve and antibiotic exposed isolates (P -value.838).No statistically significant difference was found in staphylococcus aureus susceptibility pattern among well nourished and malnourished children in this study.

Conclusion

High level of resistance was found for most of the commonly used antibiotics which mandates culture and susceptibility before prescribing any antibiotics whenever possible.

Key words: antimicrobial susceptibility pattern, antimicrobial resistance, bacteria ,culture

LIST OF TABLES

ABBREVATIONS

OPD	Out patient department
S.aureu	Staphylococcus aureus
MRSA	Methicillin resisistant staphylococcus aureus
WHO	World health organization
HIV	Human immune deficiency virus
UK	United kingdom
USA	United states of America
CDC	Center for disease control
Spp	Species
Strept.	Streptococcus
JUSH	Jimma university specialized hospital

I. INTRODUCTION

1.1.*Back ground*

Antibiotics are one of the most important therapeutic discoveries in medical history. They have revolutionized the way we treat patients with bacterial infections and have contributed to reducing the mortality and morbidity from infectious diseases. Unfortunately, antibiotics have been liable to misuse. They are often unnecessarily prescribed for viral infections, against which they have no effect. Misuse of antibiotics leads to the emergence and selection of resistant bacteria . The decreasing effectiveness of antibiotics in treating common infections has quickened in recent years, and with the arrival of untreatable strains , we are at the dawn of a postantibiotic era. The selection pressure caused by the use of antibiotics over the past 75 years since antibiotics were introduced has made almost all disease-causing bacteria resistant to antibiotics commonly used to treat them.[4]

Antimicrobial resistance among nosocomial pathogens is a significant problem in many countries with severe consequences including increased medical costs ,morbidity and mortality of patients. Since with the emergence of s.aureus strains with resistance to penicillin and methicillin ,in 1948 and 1961 respectively it has become a well known etiologic agent of a wide variety of infections and has assumed increasing importance internationally as a cause of both nosocomial and community acquired infections.[1]

MRSA is of concern not only because of its resistance to methicillin but also many of these MRSA isolates are becoming multidrug resistant and are susceptible only to glycolipides antibiotics such as vancomycin. Indeed low level resistance even to vancomycin is emerging at present [2]

The emergence of poly antimicrobial resistant strains of hospital pathogens has also presented a challenge in the provision of good quality in-patient care .The battle between bacteria and their susceptibility to drugs is yet problematic among public, researchers, clinicians and drug companies who are looking for effective drugs.[3] Crises has been building up over decades ,so that today many common and life threatening infections are becoming difficult or even impossible to treat turning a common infection in to a life threatening one.[5]

Although the resistance pattern and magnitude varies from place to place; antimicrobial resistance is the challenge worldwide. Diseases caused by resistance organisms increase morbidity, mortality and health costs. Therefore assessment of antimicrobial resistance trend is crucial especially in developing countries like Ethiopia where infectious disease burden is high.

1.2 OBJECTIVES

1.2.1 *General objective*

To determine antimicrobial susceptibility pattern of bacteria isolated from clinical specimen among children in JUSH

1.2.2 Specific objectives

- To determine common bacterial isolates from different clinical specimen
- To determine antimicrobial susceptibility pattern of bacterial isolates for commonly used antimicrobials
- To assess antimicrobial susceptibility pattern in relation to patient hospitalization
- To assess antimicrobial susceptibility pattern in relation to patient nutritional status
- To assess antimicrobial susceptibility pattern in relation to antibiotics exposure

II. METHODS

2.1 *Study area and period*

The study was conducted at outpatient and pediatric ward of Jimma university specialized hospital, Jimma zone Oromia region south west Ethiopia which is located 350 km from capital city ,Addis Ababa. Data was collected over three months from April 1 to June 30, 2014

2.2 *Study design*

Prospective cross-sectional study design was employed.

A total of 230 samples including soft tissue abscess ,ear discharge, wound swab ,eye discharge ,urine and body fluids were collected from pediatric OPD and ward of Jimma university specialized hospital from April 1, 2014-June 30, 2014.All children aged 0-15yrs were included. The specimen were collected as part of routine clinical management of patients with various clinical profiles by pediatric residents under possible aseptic clinical procedures and handled to clinical microbiology laboratory aseptically.

Specimen were inoculated on sheep blood agar, nutrient agar, manitol salt agar and Maconky agar and incubated at 37°c for 48 -72 hrs. Colonial morphology, Gram`s rxn and biochemical tests like catalase ,slide coagulase were done for isolation of bacteria.

All the isolates were inoculated on to Muller Hilton agar at 35°c and modified Kirby Bauer disc diffusion method was used for antimicrobial susceptibility test.

Susceptibility test was done for commonly used antibiotics including ampicilline ,cloxacilline, gentamicin, cholramphenicole, ceftriaxone and ciprofloxacin and interpreted based on recommendations of the clinical and laboratory standards institute(CLSI).

2.3 Variables

2.3.1 Dependent variable

Antimicrobial susceptibility and resistance

2.3.2 Independent variables

Age, sex , nutritional status(anthropometry),hospital admission , previous antibiotic use ,bacteria isolates ,previous hospitalization, type of antibiotics given ,chronic illness

2.3.3. Inclusion criteria-All patients aged 0-15 yrs for whom clinical specimen were required as part of routine clinical assessment and diagnosis.

2.3.4 Study size-All patient aged 0-15 visiting Jimma university specialized hospital during the study period and satisfying the inclusion criteria were included

Statistical methods - .Data were entered and analyzed by using SPSS version 20 soft ware ,chi-square test was employed to assess association and p value of < 0.05 was considered statistically significant.

2.3.5 Ethical considerations

Institutional ethical approval was secured from the research ethics committee of Jimma University.

III. RESULTS

A total of 230 samples were collected and analyzed from 160 male children and 70 female children with M:F of 2.2:1. A total of 72 bacteria were isolated (31.3%) with the highest percentage recovered from soft tissue abscess (40.2%) followed by ear discharges (26.4%),eye discharge 5(6.9%) and the least percentage was recovered from pleural fluid and ascetic fluid 2(2.8%) each. The highest number of bacteria were recovered from samples taken from male children 50(69.4%). In 8 patients more than 1 bacteria were isolated (table 1, 2). 42(58.3%) bacteria were isolated from in-patients while 30 (41.7%) bacteria were cultured from OPD samples. Out of total samples with positive isolates, 35% of children have received antibiotics for current illness before specimen was collected and 26% of them received more than one antibiotics. Majority of children who received antibiotics were aged 6 to 15 yrs 15(60%) where as only 2(8%) children less than one year of age received antibiotics .

The highest number of bacteria were isolated from children 1 to 5 years of age 30(41.7%).Gram positive bacteria accounted for 56(78%) of total isolates and gram negative 16(22 %).The most commonly identified organisms were staphylococcus aureus 47(65.3%),streptococcus pneumonia 7(9.7%),klepsella species 6(8.3%),E.coli 6(8.3%) ,proteus species 4(5.6%) and coagulase negative staphylococcus 2(2.8%). The majority of staphylococcus aureus were isolated from soft tissue abscess 25(53.2%),followed by ear discharge 11(23.4%). Staphylococcus aureus was the commonest organism recovered from ear discharge 11(57.9%) followed by proteus 4(21.1%),klebsella ,2(10.5%) and E.coli 1(5.3%). Staphylococcus aureus was the only organism isolated from eye discharge 5(100%) where as all the proteus species were recovered from ear discharges 4(100%) .E.coli was the predominant organism recovered from urine samples (75%) followed by klepsella spp.(25%).Majority of organisms isolated from CSF were streptococcus pneumonia 2(50%) followed by klepsella spp and E.coli 1(25%) each.

Staphylococcus aureus showed high level of resistance for commonly used antibiotics: ampicilline (89.4%) , chloramphenicole(89.4%) ,gentamicin (74.5%),cloxacilline (55.3%) and ceftriaxone(44.7%).Resistance rate for klepsella spp.was high for all antibiotics for which susceptibility test was done : 83 % for ampicilline,chlorampenicole,gentamicine and ceftriaxone..Resistance rate for ciprofloxacilline was also high except for staphylococcus aureus and proteus ;E.coli(50%),coagulase negative staphaylococcus (50%),staphaphylococcus aureus(19%) and proteus spp.(0%) . Streptococcus pneumonia has better susceptibility for most of the tested antibiotics:Ampicilline (28%),chloramphenicole (14%)and ceftriaxone (14%). .(table 3)

There is no statistically significant difference in staphylococcus aures susceptibility pattern between in-patients and out patient isolate (P value.838) nor between naïve and antibiotic exposure before presentation.(p value.228).No statistically significant difference was found in staphylococcus aureus susceptibility pattern among well nourished and malnourished children.

Table 1.Age sex distribution of bacterial isolates

type of bacteria isolated			sex of the child		Total
			female	male	
staph.aureus	age of the child	age less than 1 year	8	10	18
		age 1 to 5 years	7	9	16
		age 6 to 18 years	3	10	13
	Total		18	29	47
strept.pneumonia	age of the child	age less than 1 year	0	1	1
		age 1 to 5 years	0	2	2
		age 6 to 18 years	1	3	4
	Total		1	6	7
Klebsella	age of the child	age less than 1 year	0	1	1
		age 1 to 5 years	1	4	5
	Total		1	5	6
E.colli	age of the child	age less than 1 year	0	1	1
		age 1 to 5 years	1	2	3
		age 6 to 18 years	1	1	2
	Total		2	4	6
Proteus	age of the child	age 1 to 5 years		4	4
	Total			4	4
coagulase -ve staph	age of the child	age 6 to 18 years		2	2
	Total			2	2

Table 2. bacterial isolates by type of sample

Type of sample	type of bacteria isolated						Total
	staph.aureus	strept.pneumonia	klebsella	E.colli	proteus	coagulase -ve staph	
soft tissue abscess	25	2	0	1	0	1	29
wound swab	3	0	0	0	0	0	3
joint aspirate	3	1	0	0	0	0	4
ear discharge	11	0	2	1	4	1	19
eye discharge	5	0	0	0	0	0	5
CSF	0	2	1	1	0	0	4
pleural fluid	0	1	1	0	0	0	2
ascitic fluid	0	1	1	0	0	0	2
urine	0	0	1	3	0	0	4
Total	47	7	6	6	4	2	72

Table 3.antimicrobial susceptibility pattern of bacterial isolates

T=Total number tested

R=Number of resistant strains

Antimicrobial agent	Staph.aureus		Strept.pneumonia		Proteus spp		E.coli		Klepsella spp		Coagulase -ve staph	
	T	R	T	R	T	R	T	R	T	R	T	R
Ampicilline	47	42(89.4)	7	2(28.6)	4	4(100)	6	3(50)	6	5(83)	0	0(0)
chloramphenicole	47	42(89.4)	7	1(14.3)	4	1(25)	6	2(33)	6	5(83)	0	0(0)
cloxacilline	47	26(55.3)	0		0		0		0		2	2(100)
gentamicine	47	35(74.5)	0		4	2(50)	6	2(33)	6	5(83)	2	2(100)
ceftriaxone	47	21(44.7)	7	1(14.3)	4	0(0)	6	1(16.7)	6	5(83)	2	0(0)
ciprofloxacine	47	9(19)	0		4	0(0)	4	2(50)	0		2	1(50)

IV. DISCUSSION

Antimicrobial resistance becomes increasingly a challenge for clinicians who care for infectious diseases world wide. Infection is one of the most common reasons for out patient department visits and in patient admissions in our hospital which implies that antimicrobial susceptibility testing is one of the priority health research.

In this study the highest number of bacteria isolates are gram positive 56(78%) predominantly staphylococcus aureus 47(65.3%) ,similar finding was reported by Gebreselasse S ,gram positive 60.5% and staphylococcus aures 24% of the total isolates.(28). Staphylococcus aureus and proteus were the dominant organisms isolated from ear discharge ; similar finding was also noted by Bayeh Abera .and Dagnachew M .(26,27) .Staphylococcus aureus was the only organism isolated from eye discharge 5(100%) which is consistent with the result reported by Muluye D ,Wondimneh Y where coagulase negative staphylococcus(27.4%) and staphylococcus aureus(21%) were the predominant isolates from eye discharge in children. Similar findings was also reported by Dagnachew M (32).

E.coli was the predominant organism cultured from urine samples (75%) followed by klepsella spp.(25%) .Mulgeta K and Bayeh A also reported the highest prevalence of E.coli (65%) followed by klepsella(8.5%)from urine samples.(33)

Data in the present study shows high rate of antimicrobial resistance to common antibiotics. . Staph. aureus resistance rate for ampicilline(89.4%),chloramphenicole(89.4%),cloxacilline (55.3%),gentamicin (74.5%) and ceftriaxone(44.7%). High level of resistance for these antibiotics were reported in Ethiopia and elsewhere.(16,17,18,20,24,27,29,30). Methicillin resistance rate of staphylococcus aureus reported by Gebresilasse S was relatively low (8.3%) that could be explained by the progressively increasing antimicrobial resistance as this study was done 10 yrs from now (2003-2004) (28)..Resistance rate for klepsella spp.was high for all tested antibiotics : 83 % for ampicilline,chlorampenicole,gentamicine and ceftriaxone which is comparable with the finding reported by Dagnachew Muluye**,** Yitayih Wondimeneh ;83.3% resistance for ampicilline (27) and william D.Chey ,71% reistance for gentamicin isolated from neonatal samples(8).Another study by Ahmed S showed 100% of klepsella pneumonia were resistant for ampicilline.(34) Resistance rate for ciprofloxacilline was also high except for staphylococcus aureus (19%) and proteus (0%) . Bayeh Abera and Mulugeta Kibre also reported,87-100% sensitivity for staphaphylococcus aureus, proteus and pseudomonas.(26) In study done by Gebresilasse S staphylococcus aureus,coagulase negative staphylococcus and proteus were 100% susceptible for ciprofloxacilline.(28). Streptococcus pneumonia showed better susceptibility for most of the antibiotics for which it was tested: chloramphenicole(14.3%),ampicilline(28.6%) and ceftriaxone (14.3%) .Comparable finding was reported by Stephen G Jenkins,21.3% resistance for penicillin and 0.9% for ciprofloxacilline.

There was no statistically significant difference in staphylococcus aures susceptibility pattern between in-patients and out patient isolates (P value.838) nor between naïve and antibiotic exposure before presentation.(p value.228).No statistically significant difference was found in staphylococcus aureus susceptibility pattern among well nourished and malnourished children in this study. These findings are not in agreement with other studies: Lennox A has documented statistically significant difference in antimicrobial resistance among out patient vs hospitalized patiaents.(35). Alem A also reported 100% resistance rate of staphylococcus aureus for methicilline among severely malnourished children.(30).This dissimilar findings may be explained by the small sample size in the present study.

Limitations of the study

Small sample size

Some of the commonly used antibiotics were not tested for susceptibility like cotrimoxazole ,amoxicilline, crystalline penicillin ,vancomycine etc.

Anaerobic media was not used for growth of anaerobic bacteria

Conclusion

Gram positive bacteria were predominantly isolated from clinical specimen the majority of which was staphylococcus aureus.

Staphylococcus aureus and proteus were the predominant organisms isolated from ear discharge

Staphylococcus aureus was the only organism cultured from eye discharge

High level of resistance was found for most of the commonly used antibiotics

Streptococcus pneumonia has better susceptibility for commonly used antibiotics

No statistically significant difference in staphylococcus aureus susceptibility pattern between OPD and in-patients , previous antibiotic use and nutritional status was seen in this study

Recommendations

Treatment of suspected bacterial infection should always be guided by culture and susceptibility whenever possible

Prescribes should follow principles of rational use of antimicrobials to delay antimicrobial resistance

Since antimicrobial resistance is increasing**,** large scale antimicrobial susceptibility assessment is recommended in our hospital

Generalizability- Because of the small sample size and other limitations it is difficult to draw a generalized conclusion from this study

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