Antibiotic Susceptibility of Streptococcus Pneumoniae in Karachi

Nighat Rehman,Syed lqbal Ahmad (The Laboratory, 39, Garden Road, Karachi.)

Abstract

Objective: To evaluate, record and analyse the comparative activity of a range of oral antibiotics against Streptococcus pneumoniae.
Setting: Specimen collected at a private laboratory and some received from different hospitals and clinics.
Materials: The specimen comprised of Sputum, Throat Swabs, C.S.F., Pus, Pleural fluid, Ear Swabs, Eye Swabs, Bronchial Wash, Prostatic discharge and Antral Secretions.
Methods: The isolates were tested for their antibiotic susceptibility using the E-test by the methods described in the study protocol. The groups of antibiotics tested were Amoxicillin - Clavulanate, Clarithromycin, Cefaclor, Cefuroxime, Ceftriaxone and Penicillin.
Results: A total of 116 strains of Streptococcus pneumoniae were isolated and susceptibility to AmoxicillinClavulanate was 100%, 99.1% strains were susceptible to Cefuroxime and 0.9% were intermediate. Susceptibility to Penicillin was 80.2% and 19.8% of the strains were in intermediate categoty while susceptibilty to Clarithromycin was 89.7%, 1.7% were in intermediate region and 8.6% of the strains were resistant to this antibiotic. The susceptibility pattern of Cefaclor was not calculated as there is no current NCCLS - 97 Cefaclor breakpoints available.
Conclusion: The percentage of drug resistant Streptococcus pneumoniae was negligible in Karachi during the study period, however it is important to monitor the susceptibility pattern to keep a check on an increase in the number of DRSP as reported at different centres in Turkey, Saudi Arabia, Singapore, Thailand, Hong Kong and Indonesia (JPMA 50:58, 2000).

Introduction

Bacteria are developing different means to overcome the activity of antibacterials. The production of modifying enzymes and the alteration of binding proteins in the bacterial membrane are the important mechanism by which resistance is expressed in bacteria. Most of the non-Blactamase resistance among gram positive bacteria is mediated by changes in binding proteins[1]. Streptocccus pneumoniae that are resistant to Penicillin exhibited decreased affinity of Penicillin for whole bacterial cells, membrane preparations and two major Penicillin binding proteins.
There are differences in the antibiogram of the same organism reported throughout the world, the need for current in vitro susceptibility data is therefore apparent. These differences may be caused by inappropriate self medication, over or under prescription all using oral agents. It is important to monitor these changes to provide local information for therapy. The E. test[2] method has been described as a highly accurate method for 'detection of resistance to B-Lactam drugs.

Materials and Methods

All the isolates of Streptococcus pneurnoniae were obtained from fresh material 'using routine methods and were identified using conventional methods by optochinn susceptibility. The test for antibiotic susceptibility were run in batches of 20 including 4 Q.C strains of Staph aureus ATCC - 29213, E coli ATCC 25922, 35218 and

Streptococcus pneumoniae ATCU 4961[9] on 14 cm petil dishes containing Mueller Hinton agar supplemented with 5% sheep blood. The direct colony suspension method was recommended and applied. The colonies harvested from 16-18 hr. blood agar plate emulsified in normal saline and the turbidity adjusted to match the 0.5% McFarland standard as described by Jorgensen et al3. Within 15 minutes the surface of agar (about 4 cm in thickness) was inoculated with the aid of a swab and allowed to dry for 15 minutes. All the five antibiotic E-test[4] strips (5 cm plastic strip to which an antibiotic gradient has been incorporated) were applied using the template provided. An Oxacillin disc was also applied to the agar surface at the periphery between two of the strips. The plates were incubated at 35°C within 15 minutes of application. After 18 hours of incubation the MIC reading value written on the E-test strip was noted by an elliptical zone of inhibition produced and MIC value read from the strip where the inhibition zone intersects the strip.

Results

Out of 116 isolates of Streptococcus pneumoniae 67 (57.75%) were sputum/respiratory in origin. 12 (10.34%) Ear swabs, 17 (14.65%) Throat swabs, 6 (5.17%) Eye swabs, 2 (1.72%) Pus swabs, 8 (6.89%) CSF and others including Prostatic discharge 1 (0.86%) and 2 (1.75%) antral secretions. The MIC of different antibiotics were taken according to the NCCLS criteria (Table 1).

Table 1. Criteria of MIC reading for Streptococcus Pneumoniae.

	Susceptible	Intermediate	Resistant
Amoxycillin-Clavulanate	<0.5/0.25	1/0.5	>2/1
Cefaclor	NA	NA	NA
Ceftriaxone	<0.5	1	>2
Clarithromycin	<0.25	0.5	>1
Cefuroxime	<0.5	1	>2
Penicillin	<0.06	0.1-1	>2

Strains that have MIC <0.06 ug/ml are designated as susceptible to Penicillin. Strains that have MIC between 0.12-1.0 mg/ml are considered relatively resistant to Penicillin or interniediate. Those with MIC >1.0 ug/ml are designated as resistant.

The percentage of susceptibility for different antibiotics were as follows (Table 2).

Table 2.

Antibiotic	Susceptible		Intermediate		Resistant	
	n	%	n	%	n	%
Amoxycillin-Clavulanate	116	100.0	0	0.0	0	0.0
Penicillin	93	80.2	23	19.8	0	0.0
Ceftriaxone	116	100.0	0	0.0	0	0.0
Clarithromycin	104	89.7	2	1.7	10	8.6
Cefuroxime	115	99.1	1	0.9	0	0.0

All the strains showed (n=l 16) 100% susceptibility (M1C <0.5-0.25) to Amoxycillin-Clavulanate and Ceftriaxone (MIC <0.5). For Ccfuroxime strains showed (n=l 15) 99.1% susceptibility i.e., (MIC <0.5) and (n=l) 0.9% were in intermediate zone (MIC-l).

Strains of Streptococcus pneumoniae showing susceptibility (MIC <0.25) to Clarithromycin (n=104) was 89.7% and (n2) 1 .7% were in intermediate category while (n=l0) 8.6% showed definite resistance (MIC >1) to Clarithromycin.

Penicillin susceptibility (n=93) was 80.2% and (n=23) 19.8% of the strains were in intermediate category. No strain was found to be resistant to Penicillin in this study.

Discussion

Penicillin has been the drug of choice for pneumococcal infection for many years and susceptibility testing were not required. The emergence of strains that are resistant to multiple antibiotics including Penicillin has changed this conception[5].

The disc difftision test does not differentiate strains that are relatively resistant (MIC of 0.12-1.0 ug/mI) from strains that are resistant (MIC >1.0 ug/mI). The E-test[6] strips having antibiotic gradient incorporated is a sensitive method for the detection of resistant strains. The NCCLS recommends a 1 mg disc of Oxacillin for detection of Penicillin resistance and strains having a zone diameter less than or equal to 19mm should be reported as provisionally resistant to Penicillin as Oxacillin is a better predictor of Penicillin resistance.

The prevalence of Penicillin resistant pneumococci is being reported recently[7], there have been reports of an increase in the percentage of DRSP (Drug Resistant Strept. Pneumoniae) strains in the Far East. Saudi Arabia. Thailand and Turkey. In a study in Denver[8], 7% of patients with pneumoccal infections (bacteraem ia or meningitis) were thfected by relatively resistant strains.

A study conducted in Bangladesh in the year 1993-97 on antimicrobial resistance of Streptococcus pneumoniae showed that 11 .6% of the strains were in intermediate zone (MIC <0.1 ug/ml) and only 1.1% showed complete resistance (MIC >2.0 jig/mi) to Penicillin[9].

A marked increase in the overall prevalence of resistance to Penicillin (69.1%) was found in a study in

Hong Kong in the year 1998 as compared to rates from previous studies in 1993 (18%) and 1995 (28.9%)[10]. This high rate of resistance has only been reported in several Asian countries including Korea (73.4%), Taiwan (71%), Japan (67.7%) and Thailand (63.1%). High rates of cross resistance to Cephalosporins among the Penicillin intermediate or resistant isolates were also found.

In this study 116 strains of Streptococcus pneumoniae isolated from different sites showed no drug resistance to Cefuroxime, Ceftriaxone and AmoxycillinClavulanate, Intermediate sensitivity or relative resistance to Clarithromycin and Cefuroxime was l.7% and 0.9% respectively. Definite resistance to Clarithromycin was 8.6% and no strain was found to be resistant to Cefuroxime.

The percentage of strains depicting intermediate sensitivity or relative resistance to Penicillin was 19.8% whether some of these strains will develop definite resistance or not is a matter of time. Definite resistance to Penicillin was not found in our study. Pneumococci with intennediate sensitivity to Penicillin generally will respond to higher doses of Penicillin therapy". However it is important to monitor the drug susceptibility of Streptococcus pneumoniae in our community, to see if there are differences in the susceptibility pattern of the species as has been observed in other countries.

References

1.Hakenheck R, Tarpay M. Tomasz A. Multiple changes of penicillin-binding proteins in penicillin resisiniil isolates of Streptococcus pnenmoniae. Antim icrob. Agents eltemother., 1980:17:364-71.

2.Brown DEJ, Brown 1., Evaluation ofulie P test, A iuovel method ofquantifving antimicrobial activity. J. antimicrob. chemother., 1991:27:1 85-90.

3.Jorgensen JH, Howell AW. Matter LA Quantitative antimicrobial susceptibility testing of Haemophilus influenzae and Streptococcus pneumonmae by using E.test 3. Clin. Mierohiol., 1991:29:109-14.

4.Jorgensen JH, Ferraro Mi, Me-Elmeel Ml.. ci al, Delcction of penicillin and extended spectrum Cephalosprin resistance among Streptococcus pneumoniae clinical isolates by use of P. test. J. Cl in.. Microbial., 1994:32: 159-63.

5.Friedland lR, Mc Gahen GIl. Jr. Management of infections caused by antibiotic resistant Streptococcus pneumon mac. N. EngI. J. Med., 1 994;33l :337-38.

6.Tenover PC, Baker CN, Swenson 3M. Evaluation of cominercial methods for determining anlimicrobial suseeptihi lily of Streptococcus pneumoniae. J. Clin. Microbiol., 1996:34:10-14.

7.IIaglausd LA. Invasive pmmeuniococcal disease in Central Oklahoma: Emergence ofhiglt level penicillin resistance tmd multiple antibiotic J. Infect. Dis., 1993:168:1532.

8.Istre CR, Humphreys iT, Albrechl KI), et al. Clmloramiiphenieol and iienmeillin resistance in pneumococci isolated from blood aid Cerebro spiital fluid. A prevalence study in metropolitan Denver. J. Clin. Microbiol. 1983:17:472-75.

9.Samir KS, Rikitomi N, Rtuhulam in M, ci . Antimicrobial. Resistttnce laud serotype distribution of Streptococcus pneumoniac straimm coveting childhood infection in Bangladesh 1993 to 1997. J. Clin. Microbiol., 1999:38:798-800.

10.Pak WH. Tak-huim Q. Dominic N, et al. Eummergence of Flnoroquinolone Resistance amnong Mtultiple resittauti strains of Slreptococcns pneumimac in Hong Kong. Antiniicrob. agents chemother., 1999:64:1310-13.

11.Pallares It, Gudiol F, Linares J, et aI. Risk limciors mid response to antibiotic therapy in adults with bacteremic pncumonine catmsed by penicillin resistant pneuunococci-N. EngI. J. Med., 1987:317:18-22.