ejpmr, 2016,3(5), 484-494

SJIF Impact Factor 3.628

EUROPEAN JOURNAL OF PHARMACEUTICAL AND MEDICAL RESEARCH

www.ejpmr.com

Research Article

ISSN 2394-3211

EJPMR

EVALUATION OF ANTIMICROBIAL ACTIVITY OF COMMERCIAL ANTIBIOTICS AGAINST SELECTIVE CLINICAL ISOLATES

Khulood Khalaf Jabbar[1], Ammani K.[1], Ganesh Dama[2] and Balasubramanian Sathyamurthy[3*]

[1]Department of Microbiology, Acharya Nagarjuna University, Guntur, (A.P), India.
[2]Research Scholar, Faculty of Science, Lincoln University College, Petaling Jaya, Malaysia.
[3]Associate Professor, Department of Biochemistry, M S Ramaiah College of Arts, Science and Commerce Bangalore – 560054.

***Corresponding Author: Balasubramanian Sathyamurthy**

Associate Professor, Department of Biochemistry, M S Ramaiah College of Arts, Science and Commerce Bangalore – 560054.

Article Received on 07/03/2016 **Article Revised on 28/03/2016** **Article Accepted on 19/04/2016**

ABSTRACT

In the current study, the antimicrobial activities of some of the commercially available Antibiotics were evaluated against selected clinical Isolates. The Isolates (Test organisms) used were Staphylococcus aureus, Enterobacter aerogenes, Salmonella, Streptococcus pyogenes. These bacterial cultures were grown in Trypticose soya broth & treated with drug (Test compounds) viz., Cotrimoxazole, Tetracycline, Amoxicillin, Cefixime, Erythromycin, Ciprofloxacin, Azithromycin & Gentamicin at different concentrations. After incubation, the antimicrobial activity was evaluated by measuring the inhibition zones.

KEYWORDS: Antibiotics, Clinical Isolates, MBC, MLC, Test compounds, Trypticose broth, Agar plates.

INTRODUCTION

Staphylococcus aureus is often found in the nose, respiratory tract and on the skin. Pathogenic strains cause infections by producing potent protein toxins that bind and inactivate antibodies.

Enterobacter a common Gram-negative, facultative anaerobic, rod-shaped, nonspore-forming bacteria belonging to the family Enterobacteriaceae. Two well-known species, Enterobacter aerogenes and E. cloacae have clinical significance as opportunistic bacteria.

Salmonella causes salmonellosis. Among more than 2,300 closely-related Salmonella serovars recognized, S. Typhi and Paratyphi are human pathogens and cause systemic infections and typhoid fever, whereas others such as S. Typhimurium cause gastroenteritis.[1]

Streptococcus pyogenes (GAS) is exclusively a human pathogen. A most recent review highlights the importance of GAS virulence factors for disease manifestation and pathogenesis.[2]

Cotrimoxazole is a safe, effective and low-cost combination antibiotic widely prescribed to treat a range of bacterial, parasitic and fungal infections. CTX is a drug combination consisting of trimethoprim and sulfamethoxazole which is known to inhibit dihydrofolate reductase and has been shown to act as a sulfonamide potentiator.[3]

Tetracyclines are broad-spectrum antibiotics against a wide range of gram-positive and gram-negative bacteria like chlamydia, mycoplasmas, ricketsiae and protozoan parasites.[4-6] They were the first major group of antibiotics to which the term "broad-spectrum" was ascribed.[7] Therefore, they have been extensively used in the therapy of human and animal infections, for their prophylactic purposes in animals and plants and for growth promotion in food animals.[8,9] Tetracyclines are known to inhibit bacterial protein synthesis by preventing the association of aminoacyl tRNA with the bacterial ribosome.[10,11] Association of tetracyclines with the ribosome is reversible, providing an explanation of the bacteriostatic effects of these antibiotics.[10]

Amoxicillin, also known as amoxycillin and amox, is useful for the treatment of a number of bacterial infections. It is primarily used for treatment of middle ear infections as a first line treatment. Various aminoglycoside antibiotics interactions with the 16S rRNA and their effects on the process of translation of mRNA into polypeptide have been studied extensively.[12]

Erythromycin is a useful antibiotic for the treatment of a number of bacterial infections which includes respiratory tract infections, skin infections and syphilis. It's also used during pregnancy to prevent Group B streptococcal infection in the newborn. Erythromycin exhibits bacteriostatic activity thereby inhibiting growth of

bacteria, especially at higher concentrations, but its mechanism is not fully understood. It binds to the 50s subunit of the bacterial 70s rRNA complex, protein synthesis critical for life or replication are inhibited by this.

Ciprofloxacin is a broad spectrum, bactericidal antibiotic which acts by binding two of the four topoisomerases of bacteria.[13] They are bactericidal agents that act by interfering with the enzyme DNA gyrase to inhibit bacterial DNA synthesis.[14] Ciprofloxacin is effective against most Gram negative organisms such as Pseudomonas aeruginosa, Campylobacter, Salmonella and Shigella species etc.

Gentamicin or Garamycin (brand name) is used to treat many types of bacterial infections including bone infections, pelvic inflammatory disease, pneumonia, urinary tract infections, meningitis and sepsis. Gentamicin is a type of aminoglycoside that works by stopping the bacteria from making protein thereby killing the bacteria.

Cefixime is used in the treatment of a number of bacterial infections. It is a third generation cephalosporin. It is in the list of World Health Organization's of Essential Medicines. Cefixime acts by inhibiting cell wall synthesis of bacteria by binding to one of the penicillin binding proteins (PBPs) inhibiting the final transpeptidation in the bacterial cell wall peptidoglycan synthesis, thus arresting the cell wall assembly, resulting in cell death.

Azithromycin is used for treatment of a number of bacterial infections such as middle ear infections, strep throat, pneumonia etc. The precise mechanism of protein synthesis inhibition by macrolides depends on the specific chemical structure of the drug molecule. It functions by the following ways : 1) By inhibiting the progression of the nascent peptide chain during early period of translation.[15,16]; 2) By Promoting dissociation of peptidyl tRNA from the ribosome[17]; 3) By Inhibiting formation of peptide bond[231]; and 4) By Interference with 50S subunit assembly.[18]

MATERIALS AND METHODOLOGY

MBC or MLC determination against Bacteria by micro broth dilution technique as per NCCLS method.

Test organisms (Bacteria)

Staphylococcus aureus MTCC 7443, Enterobacter aerogenes, MTCC 7325, Salmonella typhi, MTCC 733, Streptococcus pyogenes MTCC 442.

Test compounds

Co-trimoxazole (25mcg), Tetracycline (30 mcg), Amoxicillin (30 mcg), Cefixime (5 mcg), Erythromycin (15 mcg), Ciprofloxacin (5 mcg), Azithromycin (15 mcg), Gentamicin (10 mcg).

Inoculum

Bacterial cell cultures adjusted to 1-2 x 10^5Cell suspension / mL grown on Tyrpticose soya broth was used as inoculum.

Antibiotics concentrations

1)1- 64µg/ml of test antibiotics (Two fold dilutions) in Trypticose soya broth.
2) Control: Trypticose soya broth inoculated with culture and without drug.

Procedure

Mix 90 µl drug / test compounds of different test concentration with 10 µl Inoculum in 96 well plates in triplicate. Control: Mix 90 µl Trypticose soya broth without drug with 10 µl Inoculum. Treated bacterial cultures are incubated at 22^{o}C and 35^{0}C. The bacterial test plates were observed after 24-48 hrs and O.D @ 600 nm is measured in Tecan plate reader. After determining MIC, each concentration in the wells were serially diluted and each diluted concentrations were plated in Muller-Hinton agar plates and are incubated at 22^{o}C and 35^{0}C. Agar plates were observed after 24 hrs and colonies were counted. The lowest antibiotic concentration at which no bacterial growth was observed on the plates is defined as the minimal bactericidal concentration (MBC). The minimal concentration which reduced the number of CFUs to 1/1000 that in the original inoculum was defined as minimal lethal concentration.

Minimum Bactericidal concentration of test compounds

Table 1: Minimum Bactericidal concentration of test compounds against Enterobacter aerogenes.

Name of antibiotics	MBC concentration (µg/ml) plated
Co-trimoxazole	16
Tetracycline	32
Amoxicillin	64
Cefixime	8
Erythromycin	16
Ciprofloxacin	4
Azithromycin	32
Gentamicin	16

Table 2: Minimum Bactericidal concentration of test compounds against Staphylococcus aureus.

Name of antibiotics	MBC concentration (µg/ml) plated
Co-trimoxazole	16
Tetracycline	32
Amoxicillin	8
Cefixime	32
Erythromycin	16
Ciprofloxacin	4
Azithromycin	16
Gentamicin	16

Table 3: Minimum Bactericidal concentration of test compounds against Salmonella typhi

Name of antibiotics	MBC concentration (µg/ml) plated
Co-trimoxazole	32
Tetracycline	64
Amoxicillin	64
Cefixime	16
Erythromycin	32
Ciprofloxacin	8
Azithromycin	64
Gentamicin	16

Table 4: Minimum Bactericidal concentration of test compounds against Streptococcus pyogenes

Name of antibiotics	MBC concentration (µg/ml) plated
Co-trimoxazole	64
Tetracycline	16
Amoxicillin	64
Cefixime	8
Erythromycin	8
Ciprofloxacin	4
Azithromycin	32
Gentamicin	32

RESULTS AND DISCUSSION

5.1 RESULTS

Table 5: Determination of Minimum Inhibitory concentration of test compounds against Enterobacter aerogenes

Conc. (µg/ml)	COT % Inhibition	TE % Inhibition	AMX % Inhibition	CFM % Inhibition	E % Inhibition	CIP % Inhibition	AZM % Inhibition	GEN % Inhibition
0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
1.00	21.88	19.05	9.67	39.16	25.28	51.84	13.83	10.02
2.00	39.76	25.86	15.09	60.16	34.16	63.20	21.97	18.48
4.00	52.19	40.48	21.73	71.59	45.73	79.16	39.89	25.16
8.00	69.24	55.19	38.65	84.46	71.25	85.29	62.03	43.98
16.00	81.09	79.24	59.82	90.12	83.67	89.72	75.46	51.27
32.00	93.28	87.16	69.34	95.62	95.48	94.63	83.19	68.19
64.00	94.10	93.72	82.77	96.37	95.81	97.45	88.24	75.82
MIC	4.00	8.00	16.00	2.00	8.00	1.00	8.00	16.00
Class	S	I	S	S	R	S	-	S

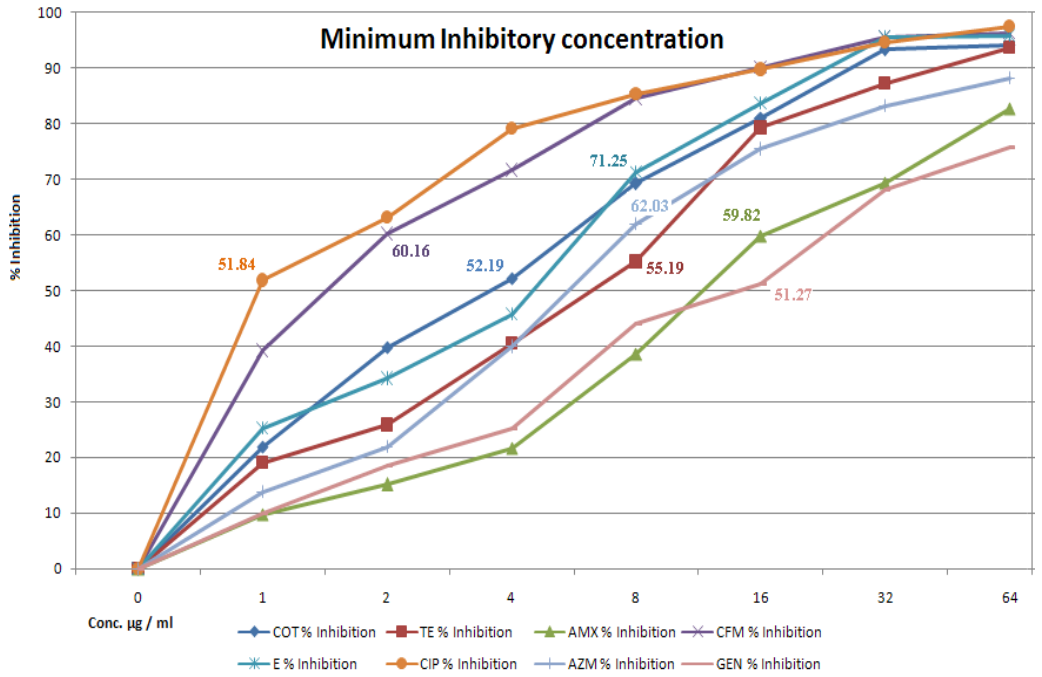


Fig. 1: Determination of Minimum Inhibitory concentration of test compounds against Enterobacter aerogenes

Table.6: Determination of Minimum Inhibitory concentration of test compounds against Staphylococcus aureus

Conc. (µg/ml)	COT % Inhibition	TE % Inhibition	AMX % Inhibition	CFM % Inhibition	E % Inhibition	CIP % Inhibition	AZM % Inhibition	GEN % Inhibition
0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
1.00	22.16	9.71	37.81	21.10	16.86	65.59	48.55	24.25
2.00	43.19	17.85	53.48	29.98	29.13	77.15	59.91	55.11
4.00	54.93	35.77	69.55	41.55	57.98	82.54	75.87	65.56
8.00	62.10	57.91	76.13	67.07	70.19	86.76	82.69	71.61
16.00	75.02	71.34	89.28	79.49	85.56	90.29	86.43	83.46
32.00	81.99	79.07	95.13	91.28	91.22	96.11	91.34	95.65
64.00	90.81	84.12	97.15	91.63	94.70	98.4	94.16	96.46
MIC	4.00	8.00	2.00	8.00	4.00	1.00	2.00	2.00
Class	-	R	S	-	R	I	R	S

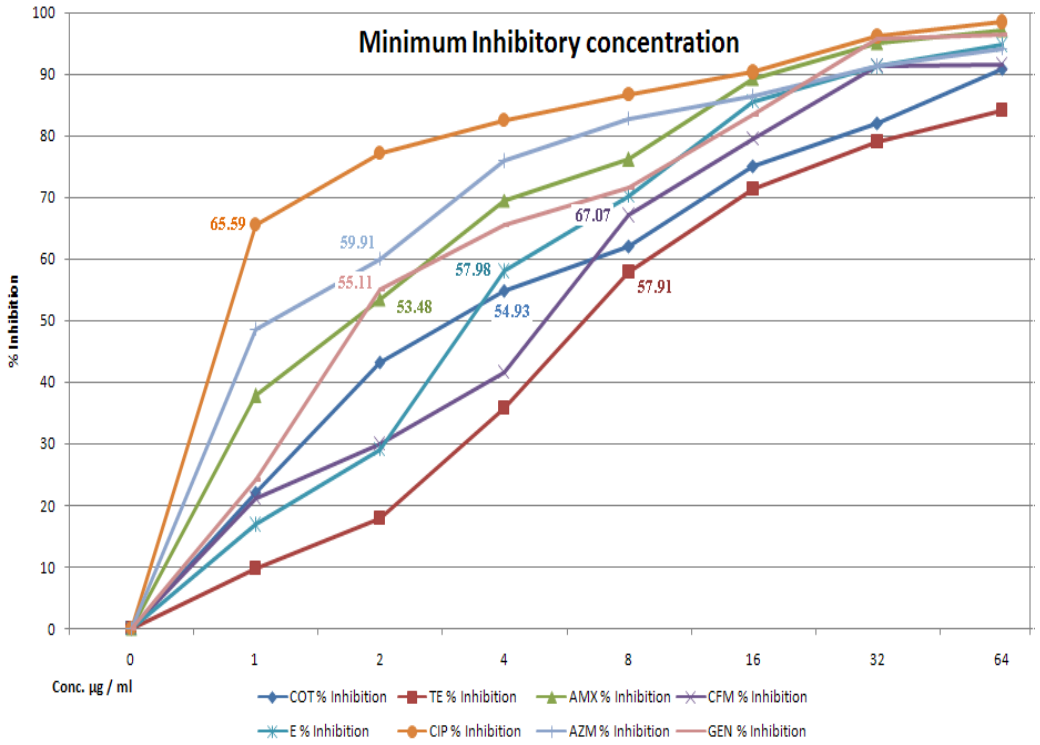


Fig.2: Determination of Minimum Inhibitory concentration of test compounds against Staphylococcus aureus

Table.7: Determination of Minimum Inhibitory concentration of test compounds against Salmonella typhi

Conc. (µg/ml)	COT % Inhibition	TE % Inhibition	AMX % Inhibition	CFM % Inhibition	E % Inhibition	CIP % Inhibition	AZM % Inhibition	GEN % Inhibition
0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
1.00	14.13	6.78	4.89	23.05	8.19	45.13	5.11	14.59
2.00	22.46	11.29	8.14	41.83	19.22	**71.54**	10.27	43.57
4.00	35.11	23.44	15.98	**65.43**	31.09	79.57	17.43	**61.22**
8.00	**51.83**	35.19	27.43	77.12	**52.05**	85.01	29.15	69.09
16.00	67.96	**61.50**	48.86	86.94	77.81	91.69	**53.99**	75.16
32.00	72.49	75.38	**65.19**	92.47	85.46	91.73	68.05	81.24
64.00	79.56	81.66	77.54	95.53	87.29	92.09	81.71	89.93
MIC	**8.00**	**16.00**	**32.00**	**4.00**	**8.00**	**2.00**	**16.00**	**4.00**
Class	-	**I**	**I**	-	**S**	**S**	**S**	**S**

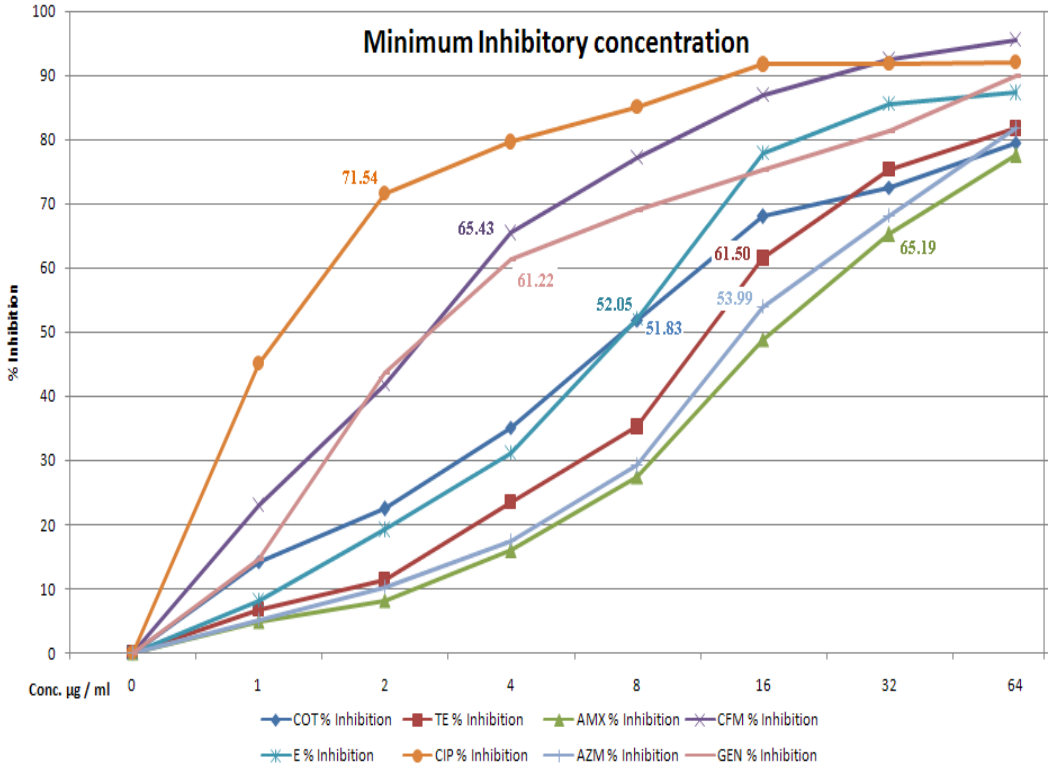


Fig.3: Determination of Minimum Inhibitory concentration of test compounds against Salmonella typhi

Table.8: Determination of Minimum Inhibitory concentration of test compounds against Streptococcus pyogenes

Conc. (µg/ml)	COT % Inhibition	TE % Inhibition	AMX % Inhibition	CFM % Inhibition	E % Inhibition	CIP % Inhibition	AZM % Inhibition	GEN % Inhibition
0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
1.00	2.95	15.58	4.50	38.11	26.19	53.95	9.57	11.89
2.00	5.52	37.19	9.71	61.73	52.08	69.08	20.83	27.13
4.00	11.93	50.43	22.04	70.09	62.75	75.16	35.24	41.87
8.00	20.87	57.91	41.93	78.86	75.43	89.28	61.59	55.17
16.00	55.09	64.15	58.11	85.43	79.16	92.43	68.17	71.23
32.00	64.13	77.08	73.83	92.47	87.79	95.79	81.73	84.91
64.00	79.72	83.13	84.17	95.80	91.08	98.46	92.47	88.02
MIC	16.00	4.00	16.00	2.00	2.00	1.00	8.00	8.00
Class	-	I	R	-	I	S	I	S

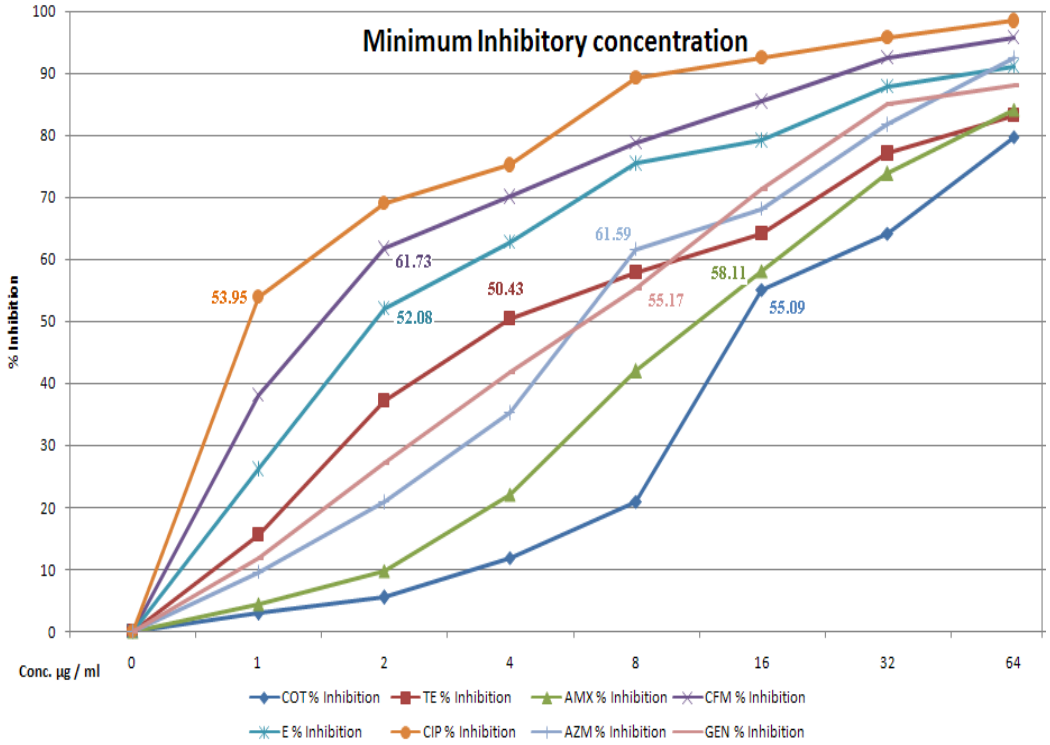


Fig. 4: Determination of Minimum Inhibitory concentration of test compounds against Streptococcus pyogenes

Table.9: Minimum Bactericidal concentration of test compounds against Enterobacter aerogenes, Staphylococcus aureus, Salmonella typhi and Streptococcus pyogenes

MBC concentration(µg/ml) plated				
Name of antibiotics	**Enterobacter aerogenes**	**Staphylococcus aureus**	**Salmonella typhi**	**Streptococcus pyogenes**
Co-trimoxazole	16	16	32	64
Tetracycline	32	32	64	16
Amoxicillin	64	8	64	64
Cefixime	8	32	16	8
Erythromycin	16	16	32	8
Ciprofloxacin	4	4	8	4
Azithromycin	32	16	64	32
Gentamicin	16	16	16	32

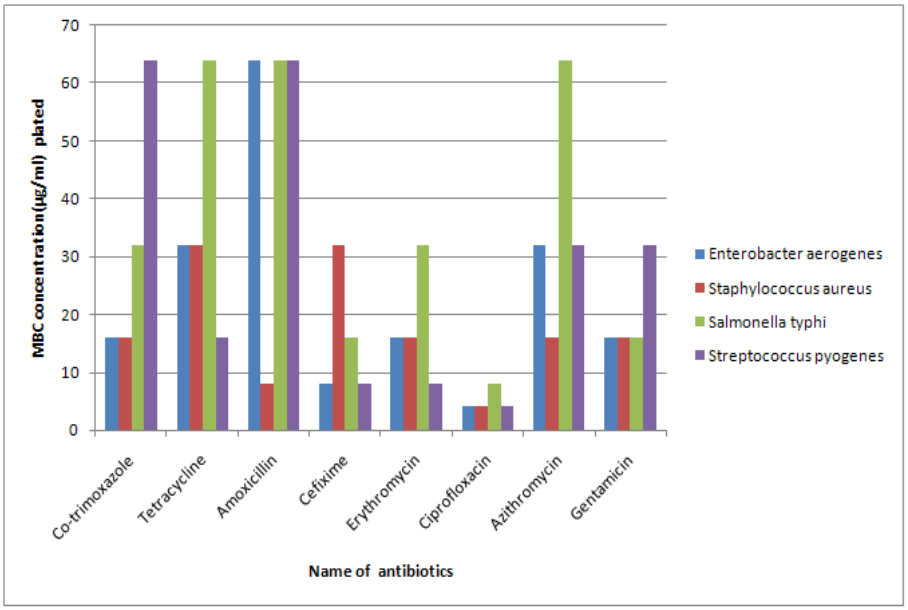
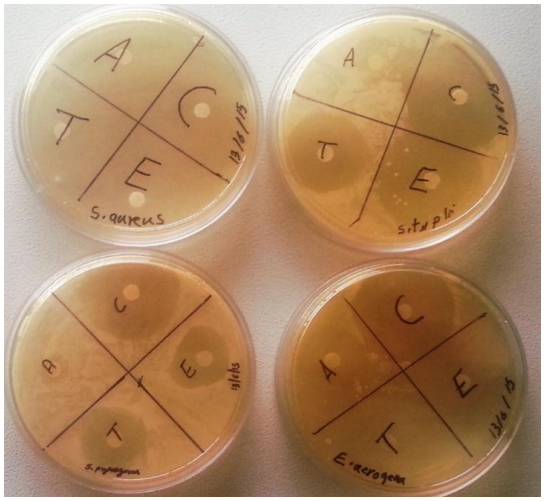


Fig.5: Minimum Bactericidal concentration of test compounds against Enterobacter aerogenes, Staphylococcus aureus, Salmonella typhi and Streptococcus pyogenes

Table 10: Antibiotic sensitivity against human pathogens

Microorganism	**Zone of inhibition (mm) & sensitivity**							
	TE 30mcg	**AMX 30mcg**	**E 15mcg**	**CIP 5mcg**	**COT 25mcg**	**CFM 5mcg**	**AZM 15mcg**	**GEN 10mcg**
Enterobacter aerogenes	20mm (I)	25mm (S)	13mm (R)	35mm (S)	20mm (S)	20mm (S)	-	23mm (S)
Salmonella typhi	17mm (I)	16mm (I)	35mm (S)	35mm (S)	-	-	19mm (S)	24mm (S)
Staphylococcus aureus	10mm (R)	-	5mm (R)	20mm (I)	-	-	12mm (R)	23mm (S)
Streptococcus pyogenes	20mm (I)	10mm (R)	20mm (I)	30mm (I)	-	-	15mm (I)	25mm (S)

Note: TE – Tetracycline, AMX – Amoxicillin, E – Erythromycin, CIP – Ciprofloxacin, COT – Co – trimoxazole, CFM – Cefixime,
AZM – Azithromycin, GEN – Gentamicin.
S – Sensitive, I – Intermediate, R – Resistant.



(A – Amoxicillin, C – Ciprofloxacin, E – Erythromycin, T – Tetracycline)

A – Azithromycin, C – Cefixime, G – Gentamicin, CT – Co-trimoxazole

Fig. 6: Antibiotic sensitivity against human pathogens

DISCUSSION

Minimum Inhibitory concentration of test compounds against Enterobacter aerogenes

From the Table – 5 and Fig – 1 the Minimum Inhibitory concentration of test compounds determined against Enterobacter aerogenesis for all eight commercial antibiotics are as follows:

Ciprofloxacin is the lowest and found be at 1 µg/ml Conc. the % Inhibition is found to be 51.84 followed by Cefixime is at 2 µg/ml Conc. the % Inhibition is found to be 60.16. Co-trimoxazole is at 4 µg/ml Conc. the % Inhibition is found to be 52.19. Tetracycline, Erythromycin and Azithromycin has at 8 µg/ml Conc. the % Inhibition is found to be 55.19, 71.25 and 62.03. Amoxicillin and Gentamicin has at 16 µg/ml Conc. the % Inhibition is found to be 59.82 and 51.27

Minimum Inhibitory concentration of test compounds against Staphylococcus aureus:

From the Table – 6 and Fig – 2 the Minimum Inhibitory concentration of test compounds determined against Staphylococcus aureus for all eight commercial antibiotics are as follows:

Ciprofloxacin is the lowest and found be at 1 µg/ml Conc. the % Inhibition is found to be 65.59 followed by Amoxicillin, Azithromycin and Gentamicin has at 2 µg/ml Conc. the % Inhibition are found to be 53.48, 59.91 and 55.11. Co-trimoxazole and Erythromycin has at 4 µg/ml Conc. the % Inhibition are found to be 54.93 and 57.98. Tetracycline and Cefixime has at 8 µg/ml Conc. the % Inhibition is found to be 57.91 and 67.07

Minimum Inhibitory concentration of test compounds against Salmonella typhi

From the Table – 7 and Fig – 3 the Minimum Inhibitory concentration of test compounds determined against Salmonella typhi for all eight commercial antibiotics are as follows:

Ciprofloxacin is the lowest and found be at 2 µg/ml Conc. the % Inhibition is found to be 71.54 followed by Cefixime and Gentamicin has at 4 µg/ml Conc. the % Inhibition are found to be 65.43 and 61.22. Co-trimoxazole and Erythromycin has at 8 µg/ml Conc. the % Inhibition are found to be 51.83 and 52.05. Tetracycline and Azithromycin has at 16 µg/ml Conc. the % Inhibition is found to be 61.50 and 53.99. Amoxicillin is the highest and found be at 32µg/ml Conc. the % Inhibition is found to be 65.19

Minimum Inhibitory concentration of test compounds against Streptococcus pyogenes:

From the Table – 8 and Fig – 4 the Minimum Inhibitory concentration of test compounds determined against Streptococcus pyogenes for all eight commercial antibiotics are as follows:

Ciprofloxacin is the lowest and found be at 1 µg/ml Conc. the % Inhibition is found to be 53.95 rouded off to 54% followed by Cefixime and Erythromycin has at 2 µg/ml Conc. the % Inhibition are found to be 61.73 and 52.08. Tetracycline is at 4 µg/ml Conc. the % Inhibition are found to be 50.43. Azithromycin and Gentamicin has at 8 µg/ml Conc. the % Inhibition are found to be 61.59 and 55.17. Co-trimoxazole and Amoxicillin has at 16 µg/ml Conc. the % Inhibition is found to be 55.09 and 58.11

Minimum Bactericidal concentration of test compounds against Enterobacter aerogenes, Staphylococcus aureus, Salmonella typhi and Streptococcus pyogenes

Minimum Bactericidal concentration of test compounds against the clinical isolates from Table – 9 and Fig – 5 are as follows:

For Enterobacter aerogenes, Ciprofloxacin has the lowest which is at conc. 4µg/ml and Cefixime is at 8µg/ml followed by Co-trimoxazole, Erythromycin and Gentamicin whose conc are at 16µg/ml. Tetracycline and

Azithromycin are found at 32µg/ml. Amoxicillin is the highest of all which is found to be 64µg/ml.

For Staphylococcus aureus, Ciprofloxacin has the lowest which is at conc. 4µg/ml and Amoxicillin is at 8µg/ml followed by Co-trimoxazole, Erythromycin, Azithromycin and Gentamicin whose conc are at 16µg/ml. Tetracycline and Cefixime are found to be highest which are at 32µg/ml.

For Salmonella typhi, Ciprofloxacin has the lowest which is at conc. 8µg/ml followed by Cefixime and Gentamicin whose conc are at 16µg/ml. Co-trimoxazole and Erythromycin are found at 32µg/ml. Tetracycline, Amoxicillin and Azithromycin are the highest of all which are found to be 64µg/ml.

For Streptococcus pyogenes, Ciprofloxacin has the lowest which is at conc. 4µg/ml followed by Cefixime and Erythromycin is at 8µg/ml. Tetracycline is found to have at 16µg/ml. Azithromycin and Gentamicin are found at 32µg/ml. Co-trimoxazole and Amoxicillin are the highest of all which are found to be 64µg/ml.

Antibiotic sensitivity against human pathogens

From Table – 10 and Fig – 6 are as follows the selected antibiotic sensitivity against Human pathogens is as follows:

Enterobacter aerogenes was found to be susceptible to most of the antibiotics namely Ciprofloxacin, Co-trimoxazole, Cefixime, Gentamicin.

Salmonella typhi also exhibited susceptibility towards a wide range of Antibiotics viz., Erythromycin, Ciprofloxacin, Azithromycin, Gentamicin.

Staphylococcus aureus & Streptococcus pyogenes both were found to be susceptible to Gentamicin. Gentamicin therefore exhibited to be a broad spectrum Antibiotic effective against all the clinical isolates selected for the study.

CONCLUSION

Micro broth dilution technique as per CLSI standards offers rapid method for characterization of microbial pathogens for antibiotic susceptibility. In the present study 4 common bacterial pathogens were tested against 8 standard antibiotics that were commercially available. MIC and MBC of microbial community from isolated cultures may be helpful in selection and prescription of antibiotic dosage.

ACKNOWLEDGEMENTS

We wish to acknowlege Skanda Life sciences Pvt Limited, Bangalore and Ms. Ghousia Begum, ITC, Bangalore for their support to complete this project.

REFERENCES

1. McClelland M, Sanderson KE, Spieth J, et al. Complete genome sequence of Salmonella enterica serovar Typhimurium LT2. Nature. 2001; 413: 852-856.
2. Walker, M. J., Barnett, T. C., McArthur, J. D., Cole, J. N., Gillen, C. M., Henningham, A., et al. Disease manifestations and pathogenic mechanisms of group a Streptococcus. Clin. Microbiol. Rev., 2014; 27: 264–301.
3. Bushby SR, Hitchings GH (1968) Trimethoprim, a sulphonamide potentiator. Br J Pharmacol Chemother, 33: 72–90.
4. Katiyar S.K., Elend T.D. (1991): Enhanced antiparasitic activity of lipophilic tetracyclines: role of uptake. Antimicrob. Agents Chemother., 35, 2075–2080.
5. Chopra I., Hawkey P.M., Hinton M. (1992): Tetracyclines, molecular and clinical aspects. J. Antimicrob. Chemother, 29, 245–277.
6. Roberts M.C. Tetracycline resistance determinants: mechanisms of action, regulation of expression, genetic mobility, and distribution. FEMS Microbiol. Rev., 1996; 19, 1–24.
7. Roberts M.C. (1997): Genetic mobility and distribution of tetracycline resistance determinants. In: Chadwick D.J, Goode J. (eds.): Antibiotic Resistance: Origins, Evolution, Selection and Spread. Wiley, Chichester (Ciba Foundation Symposium, 1997; 207: 206–222.
8. Levy S.B. The Antibiotic Paradox: How the Miracle Drugs Are Destroying the Miracle. Plenum Press, N.Y, 1992.
9. IOM (Institute of Medicine) Division of Health Promotion and Disease Prevention) Report of a study: Human health risks with the sub therapeutic use of penicillin or tetracyclines in animal feed. National Academy Press, Washington, D.C, 1998.
10. Chopra, I., P. M. Hawkey, and M. Hinton. Tetracyclines, molecular and clinical aspects. J. Antimicrob. Chemother, 1992; 29:245–277.
11. Schnappinger, D., and W. Hillen. Tetracyclines: antibiotic action, uptake, and resistance mechanisms. Arch. Microbiol, 1996; 165: 359–369.
12. Noller, H. F. Ribosomal RNA and translation. Annu. Rev. Biochem, 1991; 60: 191–227.
13. Gendrel D, Chalumeau M, Moulin F, et al. Fluoroquinolones in paediatrics: a risk for the patient or for the community? Lancet Infect Dis., 2003; 3: 537– 46.
14. Fluoroquinolones. In: Olin BR, ed. Drug Facts and Comparisons. St. Louis; Facts and Comparisons, Inc., 1998; 340d - 340.
15. Mao, J.C.-H.; Robishaw, E.E. Erythromycin, a peptidyl transferase effector. Biochem, 1972; 11: 48644872.
16. Vazquez, D. The Macrolide Antibiotics. In Antibiotics III. Mechanism of action of antimicrobial and antitumor agents; Corcoran, J.W.;

Hahn, F.E. Eds.; Springer-Verlag: New York, Heidelberg, Berlin, 1975; 459-479.
17. Menninger, J.R.; Otto, D.P. Erythromycin, carbomycin, and spiramycin inhibit protein synthesis by stimulating the dissociation of peptidyl-tRNA from ribosomes. Antimicrob. Agents Chemother, 1982; 21: 810-818.
18. Champney, W.S.; Burdine, R. Macrolide antibiotics inhibit 50S ribosomal subunit assembly in Bacillus subtilis and Staphylococcus aureus. Antimicrob. Agents Chemother, 1995; 39: 2141-2144.