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Research Article

ANTIMICROBIAL RESISTANCE (AMR) SURVEILLANCE IN RURAL MEDICAL COLLEGE IN MAHARASHTRA: NEED OF HOUR

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Abstract- Introduction: Antimicrobial resistance Surveillance (AMR) of frequently isolated bacterial pathogens causing severe infections is of great importance. Thus, this study aims to identify the prevalence of common bacterial isolates and β lactamases producing resistant strains and their antimicrobial susceptibility pattern of these isolates from patients attending Rural medical college and hospital which will help in framing institutional policy and better patient management. Material and methods: This study includes a total of 3675 bacterial isolates from urinary tract infections (UTIs), Lower respiratory tract infections (LRTIs), Blood stream infections (BSIs), Skin and soft tissue infections (SSIs) from January 2018 to December 2018. All the study isolates were characterised up to species level, antibiotic susceptibility pattern was determined. Gram negative bacteria were screened for β lactamases production. Results: Among 3675 isolates collected, *E.coli* (n= 1062) and *Klebsiella* spp(n= 745) are most common followed by *S.aureus* (n= 645), *Pseudomonas* spp (n= 511), *Acinetobacter* spp (n= 197) and other bacteria s(n= 515). *E.coli* and *Klebsiella* spp. were predominant pathogen isolated from UTI and LRTI respectively. *Staphylococcus aureus* was predominant pathogen isolated in BSI and SSI. Among the antimicrobials tested against Gram negative organisms, Colistin and Imipenem were the most active, followed by Amikacin and Piperacillin- Tazobactam. Moderate activity was noted for fluoroquinolones. Resistance to cephalosporins was high. ESBL Production is highest in *Klebsiella* spp(65%) and *E.coli*(60%) followed by *Pseudomonas* spp(37%) and *Acinetobacter* spp(30%) whereas *Acinetobacter* spp (51%) shows highest Ampc production followed by *E.coli* (40%), *Klebsiella* spp,(30%) and *Pseudomonas* spp(25%).The major MBL producer were *Acinetobacter* spp (28%) and *Klebsiella* spp(25%) followed by *Pseudomonas* spp(20%) and *E.coli*(12%). Among the antimicrobials tested against *S.aureus*, Vancomycin and Linezolid having no resistance, followed by Clindamycin and Gentamycin having Moderate activity. Resistance to fluoroquinolones and Cotrimoxazole was relatively high. The prevalence of Methicillin resistant *Staphylococcus aureus*(MRSA) was 39%. Conclusion: Increasing rates of β lactamases producers emphasizes the need for their early detection which can help in providing an appropriate antimicrobial therapy and in avoiding the development and the dissemination of these multidrug resistant strains. Need of hour is that every health care institute must have own AMR Surveillance data which will helps to develop antimicrobial stewardship program. Antimicrobial stewardship program along with Preventive measures like continuous surveillance of wards/ICUs and strict implementation of infection control practices can go long way in containing the menace of drug resistance.

Keywords- *AMR, β lactamase, Rural, Surveillance, Infection control*

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Introduction

Sepsis, respiratory tract infections (RTIs), intra-abdominal infections (IAI), skin and soft tissue infections (SSI) and urinary tract infections (UTI) are the most commonly encountered infections [1]. The pattern of bacteria causing these infections and their antibiogram vary widely from one country to another as well as from one hospital to other and even among the Wards/ICUs within one hospital [2]. β-lactam antibiotics are among the most commonly prescribed antibiotics, because of their efficacy, broad spectra and low toxicity. The selective pressures which are generated by the indiscriminate use of the β lactam antibiotics have led to the selection of a variety of mutated forms of β lactamases such as the ESBLs, Ampc β lactamases and Metallo- β lactamases which have emerged as the most worrisome resistance mechanism which poses a therapeutic challenge to the health care settings[3].They are of significant concern because they restrict the therapeutic options, cause treatment failures and are increasing in occurrence worldwide[4].The problem with resistant strains is further worsened by the fact that they are not recognized in the routine laboratory testing as they may appear falsely susceptible leading to patients receiving ineffective antibiotics[5]. Knowledge of etiological agents of infections along with prevalence of such β lactamases strains and their sensitivities to available drugs is of immense value to the rational selection and use of antimicrobial agents and to the development of appropriate antibiotic policy and better management of patients [2,5]. since the pipeline of new antibiotic development is nearly dry, surveillance of the resistance and judicious use of available antibiotics is necessary [6]. Antimicrobial resistance Surveillance (AMR) of frequently isolated bacterial pathogens causing severe infections is of great importance. Thus, this study aims to bridge the gap in knowledge and provide the clinician with the tools to provide safe and effective empirical therapy by identifying the prevalence of common bacterial isolates and β lactamases producing resistant strains and their antimicrobial susceptibility pattern of these isolates causing UTIs, LRTIs ,Blood stream infections(BSIs),SSIs from patients attending Rural medical college and hospital.

Materials and Methods

This Prospective observational study was done in Department of Microbiology, Swami Ramanand Teerth Rural Govt. Medical College, Ambajogai. Our study includes a total of 3675 clinical bacterial isolates from patients with UTIs, LRTIs, BSIs, SSIs from January 2018 to December 2018. Identification of bacterial isolates upto species level was done by their colonial morphology, Gram staining, and standard biochemical reactions [7,8].

Table-1 *Clinical pathogens in Clinical infective syndromes*

	E.coli	*Klebsiella*spp	*Pseudomonas spp*	*Acinetobacter spp*	*S. aureus*	other bacterias	Total
UTIs	742	305	176	35	97	115	1470(40%)
LRTIs	78	175	92	61	57	52	515(14%)
BSIs	85	97	45	12	256	75	570(15.51%)
SSIs	157	168	198	89	235	273	1120(30.47%)
Total	1062	745	511	197	645	515	3675

Table-2 *Antimicrobial resistance pattern of Gram Negative Organisms in percentage (%)*

	E.coli	*klebsiella spp.*	*Pseudomonas spp.*	*Acinetobacter spp.*
Colistin	0%	0%	0%	0%
Imipenem	16%	24%	25%	75%
Amikacin	28%	56%	40%	80%
piperacillin- tazobactam	28%	40%	30%	81%
Fluoroquinolones	65%	75%	60%	90%
Cefepime	52%	68%	60%	90%
Third generation cephalosporins	74%	80%	80%	92%

Antibiotic sensitivity testing was done by Kirby Bauer disc diffusion method according to Clinical Laboratory Standard Institute (CLSI)[9]. HiMedia's antibiotic discs were used. *For S.aureus, the antibiotics tested and reported were as follows:* Erythromycin (15µg), Clindamycin (2µg), Gentamicin (10mcg, Ciprofloxacin (5 µg), Linezolid(30 µg), and Cotrimoxazole (1.25/23.75 µg).E strips were used for determining susceptibility (MIC) of Vancomycin. Cefoxitin (30 µg) was used for detection of Methicillin resistant *Staphylococcus aureus* (MRSA). For Gram-negative organisms, the antibiotics were chosen from the following: Ciprofloxacin (5 µg), Levofloxacin (5 µg), Norfloxacin (5 µg), Amikacin (30 µg), Cefotaxime (30 µg), Ceftazidime (30 µg), Ceftazidime Clavulanate(30/10 µg), Cefepime (30 µg), Piperacillin - Tazobactam (100/10 mcg), Imipenem (10 µg), Imipenem-EDTA(10/750ug), Cefoxitin (30ug), Cefoxitin-Cloxacillin(30/200ug). E strips were used for determining susceptibility (MIC) of colistin. Extended spectrum β lactamases (ESBLs) production was detected by CLSI Phenotypic confirmatory test (disk Potentiation test) using Ceftazidime (30 µg), Ceftazidime – Clavulanate (30/10 µg) discs [9]. Ampc β Lactamase production detected by Cloxacillin combined disc diffusion test using Cefoxitin (30ug), Cefoxitin-Cloxacillin(30/200ug). 10Metallo β Lactamases (MBL) production was detected by combined disc test (disk Potentiation test) using Imipenem (10 µg), Imipenem-EDTA (10/750 ug) discs[10]. *S. aureus* ATCC 25923, *E. coli* ATCC 25922, and *P. aeruginosa* ATCC 27853, *K. Pneumoniae* ATCC 700603 (ESBL-positive control) were used for quality controls.

Results

In present study, a total of 3675 isolates obtained from UTIs, SSIs, BSIs and LRTIs. UTI n= 1470(40%) were most common infections ,followed by SSI n=1120 (30.47%),BSI n= 570(15.51%)and LRTI n=514 (14%). Among these 3675 isolates collected, *E.coli* (n= 1062) and *Klebsiella* spp (n= 745)are most common followed by *S.aureus* (n= 645), *Pseudomonas* spp (n= 511), *Acinetobacter* spp (n= 197) and other bacteria s(n= 515). Detailed distribution of these clinical pathogens in various above mentioned clinical infective Syndrome studied shown in [Fig-1], [Table-1] and [Graph-1].

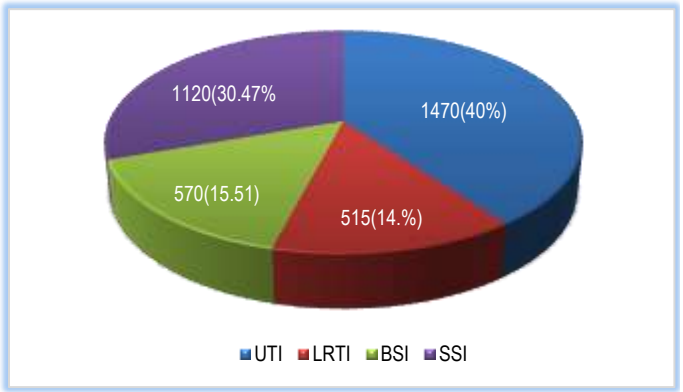

Fig-1 Pie diagram showing number(n) and percentage(%) of Isolates in Clinical infective Syndromes

Among the antimicrobials tested against Gram negative organisms, Colistin and Imipenem were the most active, followed by Amikacin and Piperacillin-Tazobactam. Moderate activity was noted for Fluoroquinolones. Resistance to Cephalosporins was high though the activity of Cefepime was found superior than third generation Cephalosporins. Antimicrobial resistance pattern of Gram-Negative Organisms shown in [Table-2]. Notably, antimicrobial susceptibility profile revealed the increasing rates of β lactamase production in Gram Negative organisms studied.

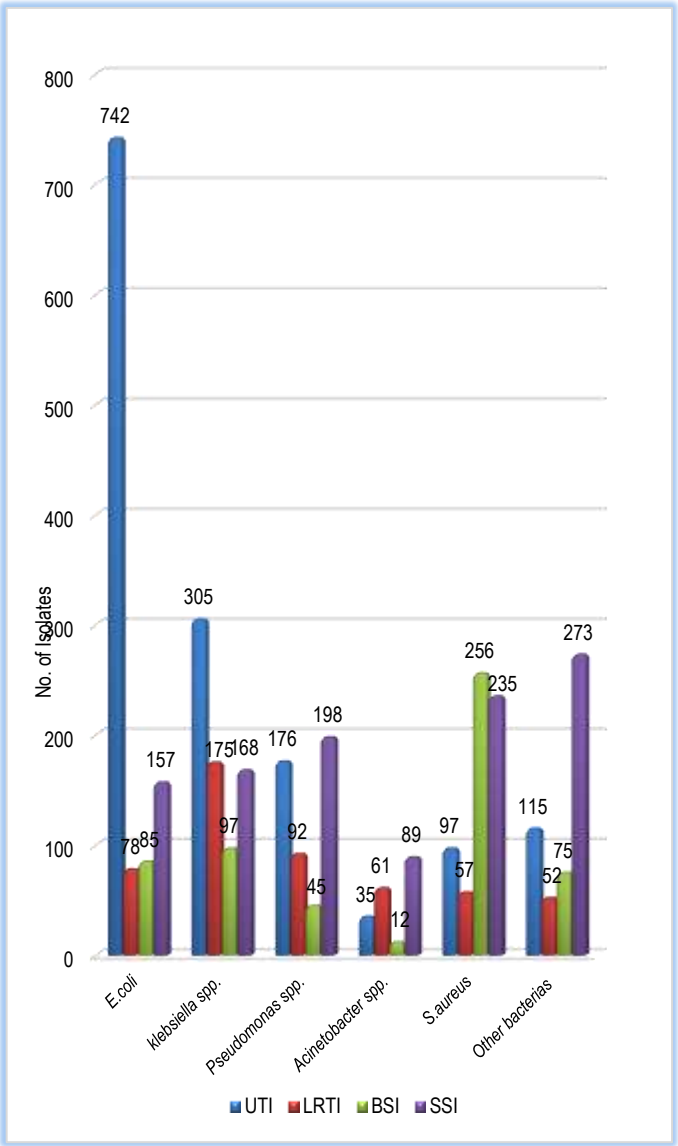

Fig-2 Clinical pathogens in Clinical infective syndromes

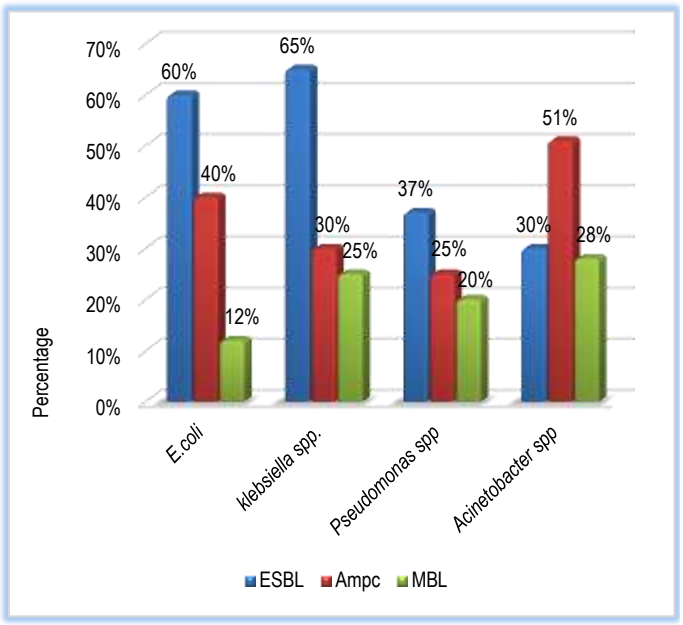

Fig-3 Distribution of β lactamases in clinical isolates

ESBL Production is highest in *Klebsiella* spp(65%) and *E.coli*(60%) followed by *Pseudomonas* spp (37%) and *Acinetobacter* spp(30%) whereas *Acinetobacter* spp (51%) shows highest Ampc production followed by *E.coli* (40%), *Klebsiella* spp, (30%) and *Pseudomonas* spp (25%). The major MBL producer were *Acinetobacter* spp (28%) and *Klebsiella* spp (25%) followed by *Pseudomonas* spp (20%) and *E. coli* (12%). Distribution of β lactamases in clinical isolates shown in [Fig-3]. Among the antimicrobials tested against *S.aureus*, Vancomycin and Linezolid having no resistance, followed by Clindamycin and Gentamycin having Moderate activity. Resistance to Ciprofloxacin and Cotrimoxazole was relatively high. Antimicrobial susceptibility profile has shown prevalence of Methicillin resistant *Staphylococcus aureus* (MRSA) is 39%. Antimicrobial resistance pattern of *S.aureus* shown in [Table-3].

Table-3 *Antimicrobial resistance pattern of S.aureus*

Antimicrobial Agent	Resistance in percentages
Cefoxitin	39%
Clindamycin	30%
Cotrimoxazole	61%
Erythromycin	60%
Ciprofloxacin	68%
Gentamycin	32%
Linezolid	0%
Vancomycin	0%

Discussion

Since our hospital is 518 bedded rural medical college and hospital, the study was undertaken to evaluate the developing bacterial trends and their susceptibility patterns to understand the prevalent resistance patterns along with β-lactamase production and to determine the effectiveness of prescribed drugs for treatment of infections. In our study we found that *E.coli* and *Klebsiella* spp. were predominant pathogen isolated from UTI and LRTI respectively. Staphylococcus aureus was predominant pathogen isolated in BSI and SSI. Similar trend seen in Veeraraghavan *et al,* Kotgire *et al,* Sundararajan *et al*., Bhave PP *et al*. and Arun Kumar *et al*. Studies [1, 11-14]. Among the Gram negative organisms, Resistance to almost all classes of antimicrobials tested such as Cephalosporins, Fluoroquinolones, aminoglycosides and carbapenems was high except colistin has been observed. In present study *Acinetobacter* spp was the most resistant among gram negative organisms showing higher degree of resistance against third generation Cephalosporin (92%), Cefepime (90%), Fluoroquinolones (90%) followed by Piperacillin-Tazobactam (81%) and Amikacin (80%). Even resistance Imipenem was much higher (75%). Nazneen *et al*, Moolchandani K *et.al*, AMR Surveillance had reported similar resistance pattern for *Acinetobacter* spp[2,15,16]. However, we found no colistin resistance which is supported by other Indian studies[5,16,17].So to be reserved in multi drug resistance (MDR) cases. In present study we noted *Pseudomonas* spp. also showing high degree resistance against to several classes of antimicrobials tested, but lower compared to *Acinetobacter* spp. Resistance of *Pseudomonas* spp. Against Imipenem, Piperacillin-Tazobactam and Amikacin was found to be 25%, 30% and 40% respectively whereas Fluoroquinolones resistance was around 60%. Resistance to third generation Cephalosporin and cefepime was 80% and 60% respectively. Our findings were in concordance with several other Indian studies in the recent past [1,2,15,16]. Among *E.coli* and *Klebsiella* spp.,we noted much higher resistance against third generation Cephalosporins (74%- 80%), Fluoroquinolones(65%-75%) and Cefepime (52%-68%). Resistance was slightly lower against Piperacillin-Tazobactam (28%-40%) and aminoglycosides (28%-56%). Even Significant resistance to carbapenems (16%-24%) was also seen. We noted *Klebsiella* spp. were more resistant as compared to Escherichia coli. Similar findings were observed by Nazneen *et al,* Moolchandani K *et al*. AMR. Surveillance [2,15,16]. Initially β-lactamase enzymes were found in the *Klebsiella* spp and *E.coli* but now all members of Enterobacteriaceae and other gram negative bacilli also producing these enzymes[4]. In our Study, of major concern was the increasing rates of β lactamases in gram negative organisms. About 60%*E.coli*, 65% *Klebsiella* spp,37% *Pseudomonas* spp., and 30% *Acinetobacter* spp were ESBL producers . Whereas Ampc β lactamases production was seen in 40% in *E.coli*, 30%in *Klebsiella* spp., 25%in *Pseudomonas* spp.,51% in *Acinetobacter* spp. In present study Metallo β lactamases production highest observed in *Acinetobacter* spp. (28%) followed by *Pseudomonas* spp. (20%), *Klebsiella* spp. (18%) and *E.coli*(12%).Other studies different parts of India have observed ESBL production ranging from 12%-81%, Ampc production ranging from 2-64%,and MBL production ranges from 11-54% [1,2,6,11-21]. Thus, frequency of β- lactamase production may vary depending on the geographical location and time. Method of assessing β- lactamase detection could also affect its frequency (6]. These observations suggest that ESBLs which were usually widespread among members of enterobacteriaceae are now increasing in *Pseudomonas* spp., *Acinetobacter* spp. Emergence. Spread of MBL-Mediated resistance is of serious concern as they would restrict the therapeutic options [22]. The colistin remains as the last option for treating infections due to MBL [1]. The low resistance to colistin, may be because of their recent introduction in the healthcare sector. Higher cost of these drugs is also responsible for their restricted use. Although, antibiotic resistance is a worldwide problem, it is the first and foremost a local problem. Selection for and amplification of resistant species which are present in each hospitals and communities, which then helps to spread it worldwide [17]. Resistance pattern of *S. aureus* which observed in our study found to be 68% against Ciprofloxacin, 61% against Cotrimoxazole, 60% against Erythromycin,32% against Gentamycin, 30% against Clindamycin. Our sensitivity pattern was in concordance with studies carried out by many other researchers [2,11,15]. However, we found no resistance to Vancomycin and Linezolid. Similar findings observed by other Indian studies [11,16,23]. In present study, prevalence of MRSA found to be 39%, similar findings documented by other Indian studies Kotgire *et al* (38.06%), Moolchandani K. *et al* (40.6%), Sangeeta *et al*(41%) studies[11,15,23]. The increasing incidence of MRSA is alarming as no βlactams drug would work in this situation and increase use of Vancomycin opens the possibility of emergence of Vancomycin resistance in *S.aureus* in near future[15]. Use of Vancomycin and Linezolid only in MRSA cases should be encouraged [23].

Conclusion

Even in Rural medical college, we have been noted increasing rates of β lactamases in Gram Negative bacteria, leaves us with limited therapeutic options like colistin which is alarming, needs much attention. It emphasizes the need for an early detection by simple screening methods which can help in providing an appropriate antimicrobial therapy and in avoiding the development and the dissemination of these multidrug resistant strains. Higher Prevalence of MRSA was observed with Vancomycin and Linezolid continue to remain the mainstay for treatment against MRSA. Need of hour is that every health care institute must have own AMR Surveillance data which will helps to develop antimicrobial stewardship program.

Antimicrobial stewardship program along with Preventive measures like continuous surveillance of wards/ICUs and strict implementation of infection control practices can go long way in containing the menace of drug resistance.

Application of Research: To assess the changing trend of antimicrobial resistance of the commonly isolated pathogens and implementation of that for forming the hospital antimicrobial stewardship.

Research Category: Antimicrobial Resistance (AMR).

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Research Project Name: Clinical Study.

Author Contributions: All authors equally contributed

Author statement: All authors read, reviewed, agreed and approved the final manuscript. Note-All authors agreed that- Written informed consent was obtained from all participants prior to publish / enrolment

Study area / Sample Collection: SRT Rural Govt Medical College, Ambajogai, 431517

Species name: *E.coli, Klebsiella* spp, *S. aureus, Pseudomonas* spp, *Acinetobacter* spp

Conflict of Interest: None declared

Ethical approval: Ethical approval taken from SRT Rural Govt. Medical College, Ambajogai, 431517. MUHS, Nashik.422004, Maharashtra, India.
Ethical Committee Approval Number: SRTRGMC/Phar/IEC/232/17.

References

[1] Veeraraghavan B., Jesudason M.R., Prakasah J.A., Anandan S., Sahni R.D., Pragasam A.K., *et al.* (2018) *Indian J Med Microbiol*, 36(1), 32-6.
[2] Nazneen S., Mukta K., Santosh C., Borde A. (2016) *Indian J Cancer*, 53(4), 583-6.
[3] Deshmukh D.G., Damle A.S., Bajaj J.K., Bhakre J.B. (2011) *Journal of Laboratory physicians*,3(2), 93-7.
[4] Oberoi L., Singh N., Sharma P., Aggarwal A. (2013) *Journal of Clinical and Diagnostic Research*,7(1),70-3.
[5] Nidhi Goel, Parul Umachaudhary (2017) *International Journal of Contemporary Medical Research*, 4(10), 2091-5.
[6] Nanoty V.V., Agrwal G.N., Tankhiwale S.S. (2018) *Journal of Clinical and Basic Research*, 2(1)1-5.
[7] Koneman E.W., Allen S.D., Janda W.M., Schreckember P.C., Winn WC .Koneman's (2016) *Colour Atlas and Textbook of Diagnostic Microbiology. 6th edNew York, Lippincott*, 97-9.
[8] Forbes B.A., Sahm D.F., Weissfeld A.S. Bailey and Scott's (2007) *Diagnostic Microbiology. 12th ed. Missouri: Mosby Elsevier*, 779.
[9] Clinical and Laboratory Standards Institute (2018) Performance Standards for Antimicrobial Susceptibility Testing. Twentyeight Informational Supplement. CLSI Document M100-S28.Wayne, PA, USA: Clinical and Laboratory Standards Institute
[10] Department of Microbiology, Institute of medical sciences, Banaras Hindu University, Varanasi (2011) *Manual of workshop on detection of β-lactamase: phenotypic and genotypic methods*.
[11] Kotgire S.A, Hatkar S.S, Siddique S., Deshmukh A.B., Afreen U., Mariya S. (2016) *Ann Pathol Lab Med*,4(3)357-61.
[12] Sundararajan Thangavel, Gomathi Maniyan S., Vijaya C. Venkateswaran ((2017) *International Journal of Bioassays*, 6.03, 5317-20.
[13] Bhave P.P., Kartikeyan S., Ramteerthakar M.N., Patil N.R. (2016) *Int J Res Med Sci*, 4(7), 2630-5.
[14] Arun Kumar P., Chougale R.A., Pandian M. (2018) *IOSR Journal of Dental and Medical Sciences*, 17(10), 54-8.
[15] Moolchandani K., Sastry A.S., Deepashree R., Sistla S., Harish B.N., Mandal Jharna (2017) *Journal of Clinical and Diagnostic Research*,11(2), DC01-DC07.
[16] Indian Council of Medical Research. Annual report Antimicrobial Resistance surveillance Network Indian Council of Medical Research.
[17] Kulkarni Vishal, Kinikar Anagha, Bhalerao Deepika, Deorukhkar Sachin and Roushani Shahriar. (2017) *Int. J. Curr. Microbiol. App. Sci*, 6(9),1103-11.
[18] Chatterjee S.S., Karmacharya R., Madhup S.K., Gautam V., Das A., Ray P. (2010) *Indian J Med Microbiol* 28 (3), 267-8.
[19] Choudhary Vinita, Pal Nita and Hooja Saroj (2018) *International Journal of Current Medical and Pharmaceutical Research*, 04(12), 3902-6.
[20] Kolhapure R.M., Kumar A., Rajkumar H.R.V. (2015) *Int.J Res Med Sci.*, 3, 2698-703.
[21] Shaikh A., Gaikwad A.A., Iravane J.A., Harbade, Damle A.S. (2018) *Int. J. Curr. Microbiol. App.Sci*., 7(02),1662-7.
[22] Pramodhini S., Umadevi S., Seetha K.S. (2015) *Int. J. Microbiol. App. Sci*., 4(12),718-26.
[23] Sangeeta Joshi, Pallab Ray, Vikas Manchanda, Jyoti Bajaj, D.S.Chitnis, Vikas Gautam *et al*. (2013) *IndianJ.Med.Res*,137,363-9.