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Original Research Article

Antimicrobial susceptibility profile of aerobic bacterial strains isolated from periodontal lesions

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ABSTRACT

Background: Periodontitis or gum disease is a serious gum disease that damages soft tissue and without treatment can destroy the bone that support the teeth. It is because of consequences of specific or non-specific bacterial infections. Drug resistance is a major concern in treatment of periodontitis also. So this study was planned to isolate, identify aerobic bacteria and to characterize the antibiotic sensitivity pattern of isolate from periodontal lesions and to analyze host/patient related factors.
Methods: Patients coming to dental OPD with signs and symptoms of periodontitis were selected for the study. Samples were collected from periodontal pockets using bent swab stick and were processed for aerobic culture and antibiotic susceptibility test. Demographic data, results of culture, isolated pathogens and its antimicrobial sensitivity related data were entered in Excel sheet and analyzed using SPSS software version 23.
Results: Total 71 aerobic isolates were found out of 100 aerobic culture. In *E.coli* isolates 83% resistance was found for ceftriaxone and ceftazidime. *K.oxytoca* isolates show 75% resistance to ceftazidime. K.pneumonaie shows 75% resistance to ceftazidime, 50% to amoxiclav, 29% to minocyclin. *S.viridans* shows 22% resistance to azithromycin, 19% to tetracycline and erythromycin. In present study higher drug resistance was found in *K.pneumonaie* followed by *E.coli*, *K.oxytoca*, and *S.viridans*. There is significant association between dental hygiene and the culture result (chi-square=20.771, df=8. $p<0.01$) and no significant association between age and the culture result (chi-square=44.032, df=40, p=0.305) were found.
Conclusions: Periodontitis patients shows drug resistant to commonly used antibiotics for the same. Also, there is an association between dental hygiene and culture positivity. So proper dental hygiene can prevent periodontitis.

Keywords: Periodontitis, Aerobic culture, Drug resistance

INTRODUCTION

Periodontitis or gum disease is a serious gum disease that damages soft tissue and without treatment can destroy the bone that support the teeth. It is because of consequences of specific or non specific bacterial infections by either aerobic or anaerobic bacteria.[1-3] Periodontitis being the result of both direct degradative action and inflammatory process mediated by the presence of microbial cells and virulence factors of microorganisms affecting the periodontal integrity. In present era, emergence and evolution of antimicrobial resistance represent important cause of therapeutic failure of periodontal disease.[4] Moreover, the production of biofilm produced by periodontopathogenic bacteria makes the antibiotic ineffective.[4-6] There are also other host factor like proper oral hygiene, cessation of smoking etc, immunological status of the host, comorbid conditions of the patients have role in oral health periodontitis.[7-10] So this study was planned to isolate, identify aerobic bacteria and to

characterize the antibiotic sensitivity pattern of this isolate from periodontal lesions and to analyze host/patient related factors.

METHODS

Sample collection

It was a prospective study done in the Government Institute of South Gujarat Region which was started after ethical approval for the same from Human Research Ethics Committee of Government Medical College. Patients who came to dental OPD with signs and symptoms (swollen gum, bright red, dusky red gums, pus between gum and teeth, complain of pink –tinged tooth brush after brushing, gums that bleeds easily etc.) of periodontitis during mid 2022 were included in the study. Patients already diagnosed and treated for periodontitis or having other dental or oral problems were excluded from the study. Patients were given information sheet in the language they Comprehend regarding the study, its objectives, methodology, its benefits, harmful effects etc. Only patients who were voluntarily willing to participate in the study were included and informed consent was taken from them. Samples were collected from periodontal lesions' pocket using bent swab stick and transported to Microbiology laboratory in cold chain at 2-4º for further process.

Isolation and identification bacterial strains and its antimicrobial susceptibility test

As facility for only aerobic culture is available at Microbiology department, samples were processed only for aerobic culture, for which culture on blood agar, Mac Conkey agar and Chocolate agar was done and incubated in an incubator at 37ºC±2ºC for 18-24 hours. Simultaneously gram staining was performed from swab to know to the gram reaction of organism if present. After incubation of culture, microbiological identification was performed based on culture positivity, colony characteristic, different biochemical reactions, motility etc. From culture, antibiotic susceptibility test was done using Kirby–Bauer disc diffusion method and interpretation of results was done based on CLSI M100 2022. Demographic data, results of culture, isolated pathogens and its antimicrobial sensitivity related data were entered in Excel sheet and analyzed using SPSS software version 23.

RESULTS

Total 100 patients were enrolled with periodontitis during the study period. Out of which 60 were female and 40 were male patients. 68 were educated with minimum of SSC to maximum up to graduate level and 32 were illiterate patients. 28% patients were from age group of 50-60 year followed by 21% from 40-50 year, 17% from 60-70 year, 15% were from more than 70 year, 13% were from 30-40 year and 6% from 20-30 year. Analysis of dental problems has shown that 53% have pain in gum, 16% have mobility of teeth, 15% have swollen gum, 6% have dental caries, 6% have sensitivity in gums and 4% have bleeding gums.

Oral hygiene of 96 patients were with brushing habits of 1 time in a day while 3 patients have brushing habit for two times and one patient found of no habit of any kind of dental hygiene practices. Tobacco chewing and smoking habit were found in 22 patients. Isolates found out of these 71 samples were analyzed which shows that 36 isolates were streptococcus viridians, 21 were Klebsiella pneumoniae, 6 were *E.coli*, 4 were *K.oxytoca*, and rest are *P.aeruginosa*, *Acinatobacter baumanii*, Coagulase negative *staphylococcus* and *Staphylococcus aureus*. In Table 1 and 2 shows the association between dental hygiene and culture; age group and culture respectively. In Figure 1-3 and 4 resistance percentage of isolates of *S.viridans*, *K.oxytoca*, *K.pneumoniae*, *E.coli* is shown.

Table 1: Association between dental hygiene and culture result.

Culture results	Oral health brushing frequency				Total		Chi-square	P value
	Once		Twice					
	N	%	N	%	N	%		
No growth	29	100.0	0	0.0	29	29.0	20.771	p=0.008**
Streptococcus viridans	35	97.2	1	2.8	36	36.0		
Klebsiellapneumoniae	21	100.0	0	0.0	21	21.0		
Escherichia coli	4	66.7	2	33.3	6	6.0		
Klebsiellaoxytoca	4	100.0	0	0.0	4	4.0		
Pseudomonas aeruginosa	1	100.0	0	0.0	1	1.0		
acinetobacterbaumannii	1	100.0	0	0.0	1	1.0		
Coagulase negative staphylococcus	1	100.0	0	0.0	1	1.0		
Staphylococcus aureus	1	100.0	0	0.0	1	1.0		
Total	97	97.0	3	3.0	100	100.0		

Table 2: Association between age (years) and culture result.

Culture results	Age (years)												Total		Chi-square	P value
	20-30		30-40		40-50		50-60		60-70		>70					
	N	%	N	%	N	%	N	%	N	%	N	%	N	%		
No growth	2	6.9	1	3.4	7	24.1	3	10.3	7	24.1	9	31.0	29	29	44.032	0.305, NS
Streptococcus viridans	2	5.6	6	16.7	8	22.2	12	33.3	6	16.7	2	5.6	36	36		
Klebsiellapneumoniae	1	4.8	4	19.0	5	23.8	8	38.1	1	4.8	2	9.5	21	21		
Escherichia coli	0	0.0	1	16.7	1	16.7	3	50	1	16.7	0	0.0	6	6		
Klebsiellaoxytoca	1	25	1	25	0	0.0	0	0.0	0	0.0	2	50.0	4	4		
Pseudomonas aeruginosa	0	0.0	0	0.0	0	0.0	1	100	0	0.0	0	0.0	1	1		
Acinetobacter-baumannii	0	0.0	0	0.0	0	0.0	1	100	0	0.0	0	0.0	1	1		
Coagulase negative staphylococcus	0	0.0	0	0.0	0	0.0	0	0.0	1	100	0	0.0	1	1		
Staphylococcus aureus	0	0.0	0	0.0	0	0.0	0	0.0	1	100	0	0.0	1	1		
Total	6	6.0	13	13.0	21	21.0	28	28	17	17.0	15	15	100	100		

Table 3: Resistance percentage of all isolates for commonly used drugs for periodontitis.

Drugs commonly used for periodontitis	*S.viridans* (36 isolates) (%)	*K.pneumoniae* (21isolates) (%)	*K.oxytoca* (4isolates) (%)	*E.coli* (6 isolates) (%)	*Acinatobacter baumanii* (1 isolate) (%)	*P.aeruginosa* (1 isolate) (%)	*S.aureus* (1 isolate) (%)	*Coagulase negative staphylococcus aureus* (1 isolate) (%)
Tetracycline	19	24	0	50	0	0	0	0
Doxycycline	-	24	0	50	0	-	0	0
Minocycline	-	29	50	50	0	-	0	0
Amoxiclav	-	50	50	-	-	-	0	0
Azithromycin	22	-	-	-	-	-	0	100
Erythro-mycin	19	-	-	-	-	-	0	100
Ceftriaxone	11	0	0	83	-	-	0	0
Ceftazidime	14	75	75s	83	100	100	0	0

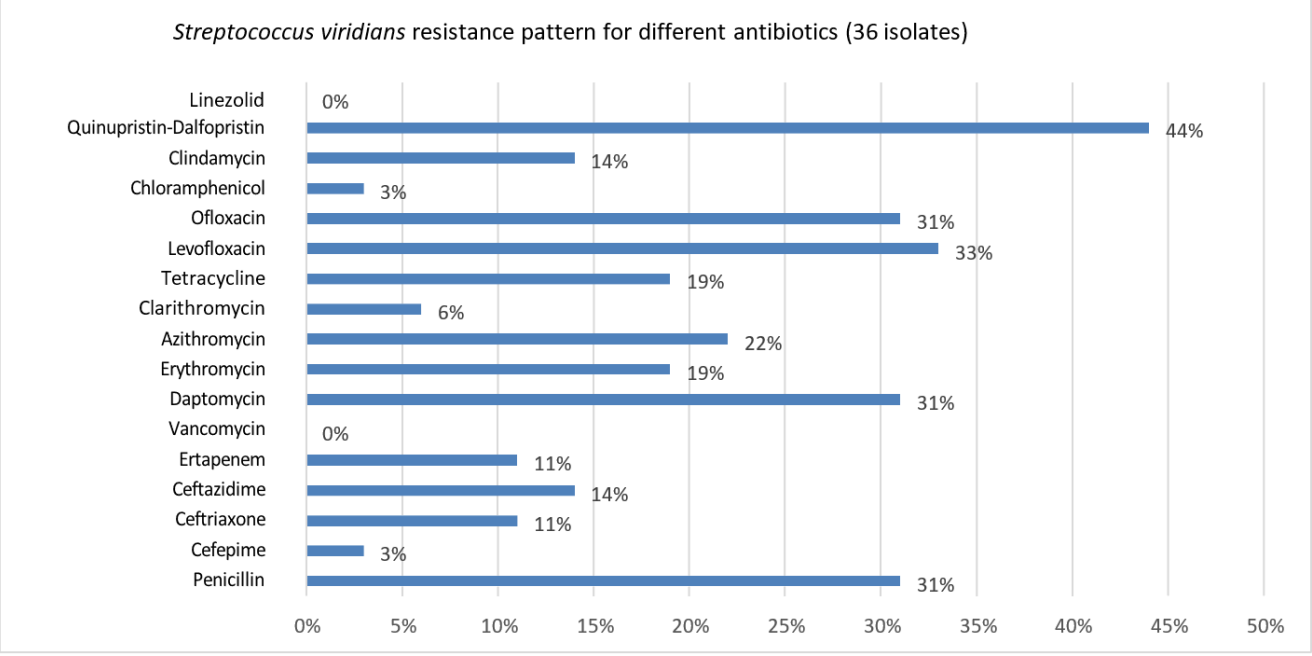

Figure 1: *S.viridans* resistance pattern.

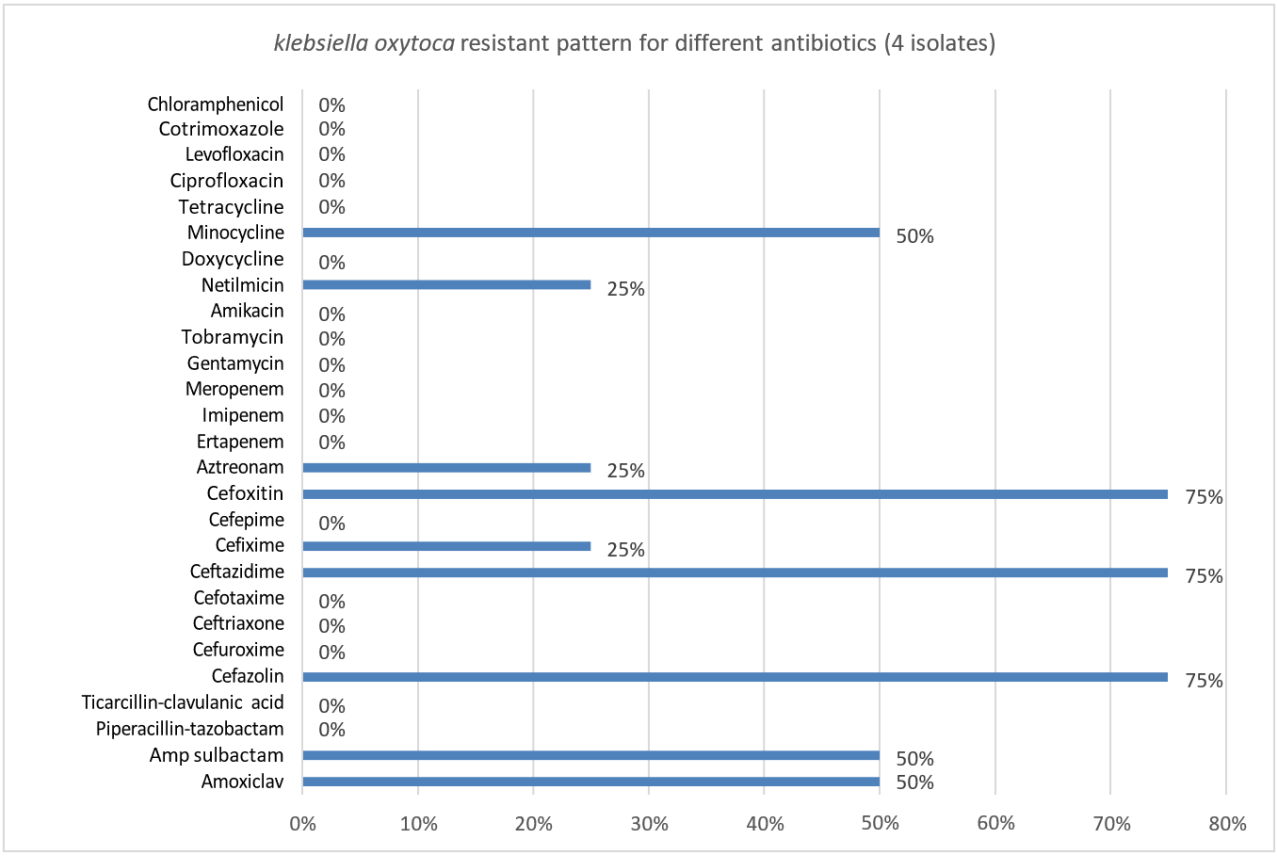

Figure 2: *K.oxytoca* resistance pattern.

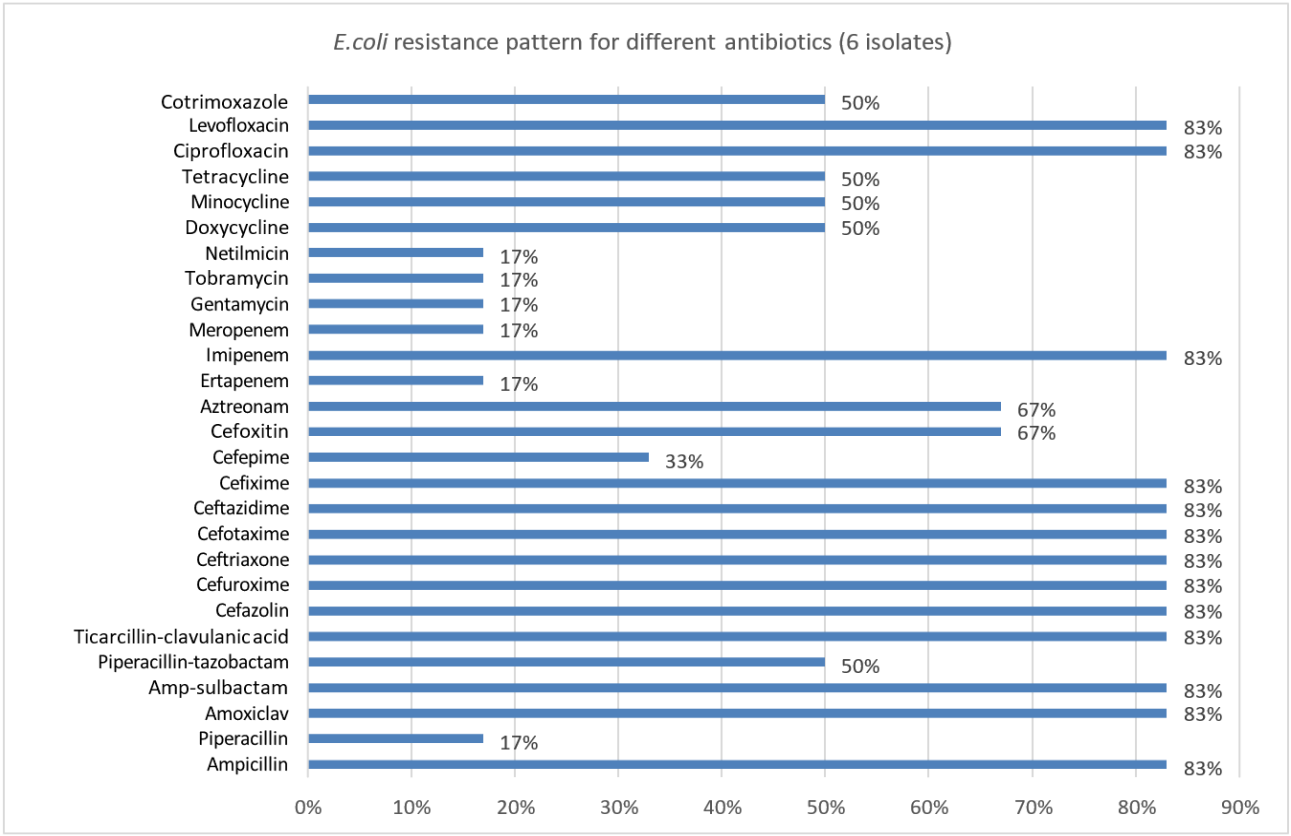

Figure 3: *E.coli* resistance pattern.

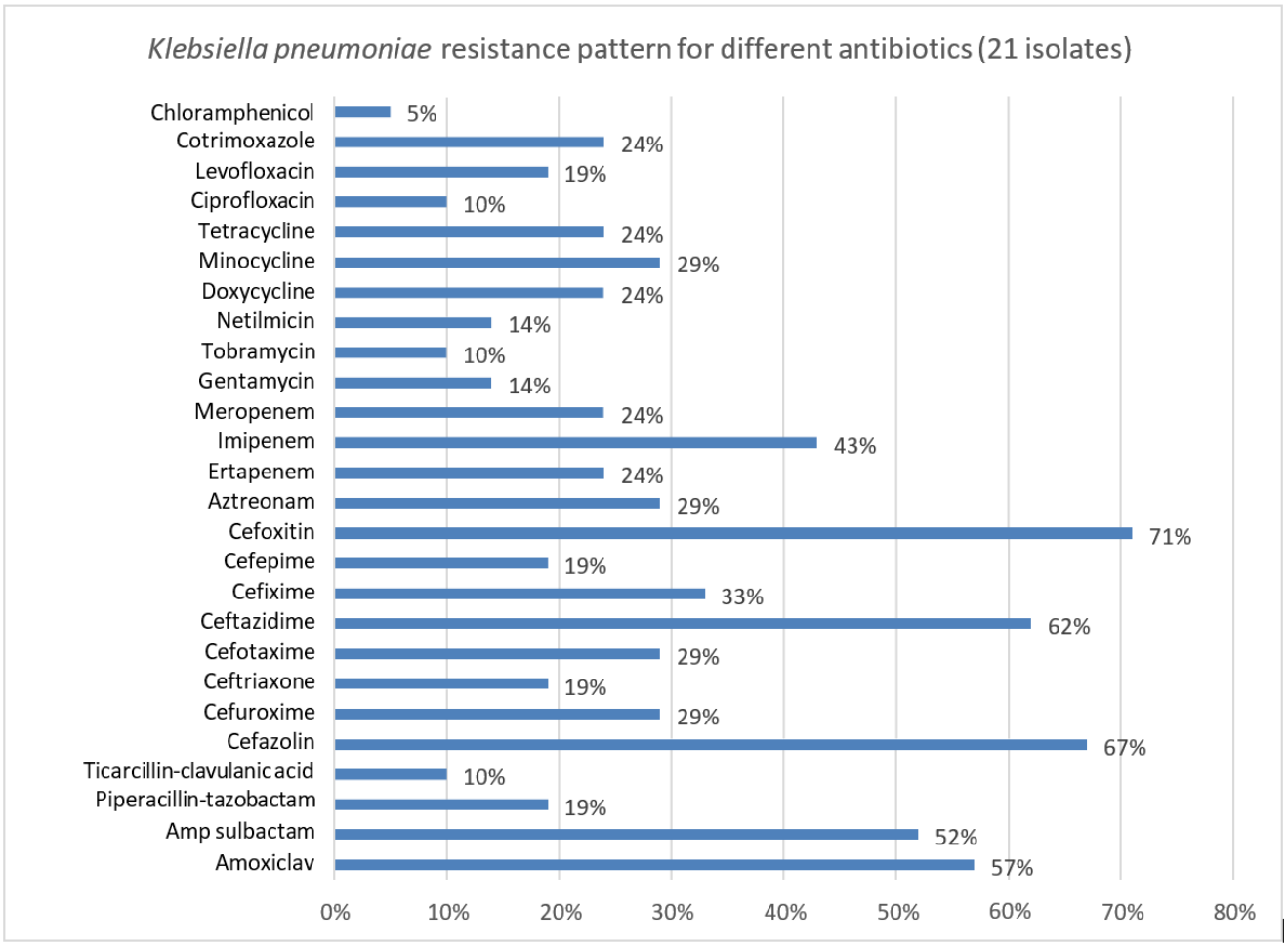

Figure 4: *K.pneumoniae* resistance pattern.

Oral samples from all 100 patients were processed for gram stain and aerobic culture out of which 71 samples were positive by aerobic culture. In Table 3 resistance percentage of all isolates for commonly used drugs for periodontitis is shown.

DISCUSSION

100 new patients with periodontitis were enrolled from dental OPD area. 28% patients belonged to age group 51-60 followed by 21% from 41-50 age group. The result of chi-square test for association between attributes indicates that there is no significant association between age and the culture result (chi-square=44.032, df=40, p=0.305). The result of chi-square test for association between attributes indicates that there is significant association between dental hygiene (brushing of teeth 2 times a day) and the culture result (chi-square=20.771, df=8. p<0.01).

68% patients were literate. Maximum patients presentation was found with pain in gum (53%) followed by mobility of teeth (16%), swollen gum (15%), sensitive teeth (6%), dental carries (6%) and bleeding gum (4%).

Out of 100 patients 29% was negative in aerobic culture while 71% were positive in culture with diverse group of organisms like *S.viridans* (36%), *K.pneumoniae* (21%), *E.coli* (6%), *K.oxytoca* (4%), *P.aeruginosa* (1%), *A.baumanii*(1%), *CONS* (1%), *S.aureus* (1%).

Commonly used antibiotics foe periodontitis are tertacyclines, doxycyclin, minocyclin, amoxiclav, azithromycin, erythromycin, ceftraixone, ceftazidime, metronidazole. In present study higher drug resistance was found in *K.pneumonaie* followed by *E.coli*, *K.oxytoca*, and *S.viridans* which is much higher as compared to the study by Sakthivel et al and study by Dascalu et al.[11,12]

In *E.coli* isolates 83% resistance was found for ceftriaxone and ceftazidime followed by 50% resistance to tetracycline, doxycyclin and minocyclin. *K.oxytoca* solates show 75% resistance to ceftazidime followed by 50% resistance to minocycline and amoxiclave. *K.pneumonaie* shows 75% resistance to ceftazidime, 50% to amoxiclav, 29% to minocyclin and 24% to tetraxyclin and doxycyclin. *S.viridans* shows 22% resistance to azithromycin, 19% to tetracycline and erythromycin, 14%

to ceftazidime and 11% to ceftriaxone, 14% to clindamycin.

CONCLUSION

Periodontitis patients shows drug resistant to commonly used antibiotics for the same. Also, there is an association between dental hygiene and culture positivity. So proper dental hygiene can prevent periodontitis.

Limitation

As we have only facility for aerobic cultures, we were not able to isolates anaerobic organisms which are also main culprit for periodontitis. We can have larger study, multi center studies to analyze drug resistance pattern as Antimicrobial resistance is an global issue.

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