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Comparison of Oral Microbial Flora in Tobacco Smokers and Non-Smokers

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Abstract:-

Aim

The present study was undertaken to evaluate oral microbial flora in samples from three different sites that is dental plaque, saliva and tongue smear in smokers and compare the results with equal number of age matched non-smokers.

Materials and Methods

This case-control study comprised of 35 subjects with history of tobacco smoking and equal number of age matched non-smokers, with age range between 20 to 30 years. All the subjects included were male patients as smoking in this geographic region is more prevalent in males. The samples from subjects were taken by aseptic method from three different oral sites and take to the laboratory, to assess the microbial count.

Results

The culture reports of samples of the study subjects clearly indicated a higher amount of mean value of microbial flora count that is 5.64 X 10^5 C.F.U. / ml as compared to that of control subjects which was 4.77 X 10^5 C.F.U. / ml. The salivary samples of smokers and non-smokers showed a p value less then 0.05, with respect to Moraxella catarrhalis and Corynebacteria, which was statistically significant using Wilcoxon rank test.

Conclusion

The study shows that there is a definite increase in the amount of microbial flora in tobacco smokers as compared to non-smokers. These increased microbes may ultimately increase the chances for oral diseases and impaired wound healing thus affecting the treatment outcome of oral health.

Keywords:- *Oral Microbial Flora, Tobacco Smoking, Oral Health.*

I. INTRODUCTION

"Cigarette smoke
is a custom loathsome to the eye,
hateful to the nose, harmful to the brain,
dangerous to the lungs and in the black, stinking fume thereof
nearest resembling
the horrible Stygian smoke of the pit
that is bottomless."

By James I of England, King of Great Britain

The oral cavity is one of the site, with varied microbes among those found in the body. The most common species, which have been isolated in oral cavity are Streptococcus, Staphylococcus, Lactobacilli, Actinomyces, Moraxella catarrhalis, Corynebacteria, Bacteroids etc. The favorable conditions required for the bacterial growth like temperature, humidity and nutrients are present in the oral cavity [1].

In oral health a balance exists between three factors: the host, the environment and the microorganisms, whereas any imbalance in these factors causes disease. The oral cavity constitutes distinct ecosystems for microbial colonization and growth like buccal mucosa, dorsum of the tongue, teeth surfaces and crevicular epithelium. Teeth provide non-shedding surfaces that allow accumulation of dental plaque in retentive areas. The dorsal surface of the tongue is papillated, thus retains more microbes [1,2].

Cigarettes are chemical cocktail containing more than 4000 harmful chemicals. The burning cigarette works as a miniature blast furnace, yielding odorless, colorless deadly gas carbon monoxide, increased levels of acetaldehyde, arecoline, formaldehyde etc that may have an effect on pathogenesis, leading to progression of many diseases. In fact, seemingly harmless sounding ingredients, such as chocolate, licorice and sugars do contribute to increased carcinogenic and additive effects [4].

During the past few decades a number of studies have found that smoking, either alone or in combination with other factors, appears to be an important predisposing factor for a change in oral microbial flora although the exact pathogenic influence of smoking is yet to be resolved. The results of the previous studies are conflicting and inconclusive [5].

Hence, this case control study had been undertaken to examine the change in microbial flora in samples taken from dental plaque, saliva and tongue smear in systemically healthy, young, male smokers and non-smokers.

II. MATERIALS AND METHODS

Patients reporting to the Department of Oral medicine and Radiology at K.L.E.S.'S Institute of Dental Sciences, Belgaum were included in the study. All the subjects included were male patients as smoking in this geographic region is more prevalent in males. The study included two groups:
Group 1 = STUDY GROUP (35 smokers)
Group 2 = CONTROL GROUP (35 non-smokers)

Selection criteria

1) **Group 1: Study group**

Inclusion criteria:

- Patients with a history of smoking, at least for one year.
- Patients in the age group of 20-30 years were considered.

Exclusion criteria:

- Patients on drug therapy, local or systemic.
- Patients having an ulcer, infection in oral cavity.
- Patients suffering from any known systemic disease.

Group 2: Control group

Inclusion criteria

- Patients who were non-smokers.
- Patients in the age group of 20-30 years were considered.

Exclusion criteria

- Patients on drug therapy, local or systemic.
- Patients having an ulcer, infection in oral cavity.
- Patients suffering from any known systemic disease

After explaining about the study to the subjects, a detailed history with thorough clinical examination was carried out and the findings were recorded in the case history proforma for all the patients. The patients were asked to sign the consent form. From all the subjects, samples were taken from tongue smear, teeth (dental plaque) and saliva.

Collection of the Microbial Samples:

Group 1: Three samples were collected from each subject.

- Site A = Dental plaque sample from study group
- Site B = Saliva sample from study group
- Site C = Tongue smear sample from study group

Group 2: Three samples were collected from each subject.

- Site D = Dental plaque sample from control group
- Site E = Saliva sample from control group
- Site F = Tongue smear sample from control group

Procedure for microbial sample collection:

a) Samples from tongue were taken, using a sterile cotton swab. The swab was rolled on the dorsal surface of tongue to collect the smear. The sample was transferred to a sterile bottle containing 2 ml thioglycollate transport media.
b) Samples from teeth were taken using a periodontal scaler and then transferred to a sterile cotton swab. Later the swab was transferred to a sterile bottle containing 2 ml thioglycollate transport media.
c) Samples of saliva were collected by using sterile cotton swab in contact with floor of the mouth for a minute. Later the swab was transferred to a sterile bottle containing 2 ml thioglycollate transport media.

These media containing bottles were then taken to a microbiology laboratory to assess the microbial content in the samples.

Laboratory procedure

In the laboratory, the swabs were taken out from the bottles and about 10 microlitre of sample was inoculated on equally divided Blood agar, Mitis Salivarius agar and MacConkey agar culture media plates. These culture plates were incubated at 37 degree Celsius for 48 hours.

After incubation of culture plates, 48 hours later the bacterial growth was analyzed for:

- Colony morphology
- Types of colonies
- Colony count

Bacterial identification was done on the basis of various staining techniques and biochemical characteristics.

Colony count of bacteria was done and expressed in terms of colony forming units per milliliter (C.F.U. /ml) by using the following formula:
1 swab was immersed in 2 ml of transport media.
1 ml = 1000 micro liter
Total volume of transport media = 2 ml (2000 micro liter)
10 micro liter of sample was inoculated on culture plates.
Therefore, a dilution factor is equal to 200.

Number of colony count of each type of microorganisms isolated in media was multiplied by 200 to express the colony count in colony forming units per milliliter (C.F.U. /ml).

Tabulation of results was done for study group and control group. Evaluation of results was done by calculating the measures of central tendency that is mean value and median value. Wilcoxon rank test to calculate the probability value (p value).
p value was considered in the following manner:

- Not significant = > 0.05
- Significant = < 0.05

<u>Armamentarium for Clinical Examination and Collection of Microbial Samples</u> :

1. Sterilized facemask.
2. Sterilized gloves.
3. Sterilized kidney tray.
4. Sterilized mouth mirror and probe.
5. Sterilized tweezer.
6. Sterile cotton swab.
7. Thioglycollate transport media in sterile glass bottles.

<u>Materials used for Microbial analysis</u>:

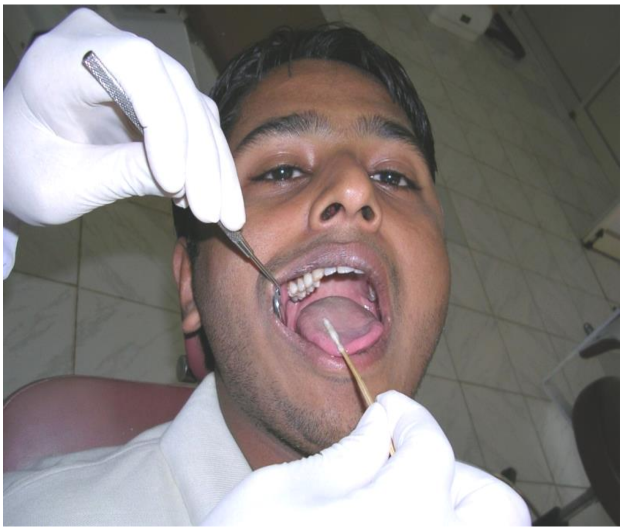
1. Spirit lamp.
2. Inoculation loop.
3. Blood agar plate.
4. MacConkey agar plate.
5. Mitis Salivarius agar plate.
6. Microscopic slides.
7. Materials for gram staining like gentian violet, iodine, ethanol, safranine etc.

Photo – 1 : Intra Oral Photograph of Patient – I

Photo – 2 : Intra Oral Photograph of Patient – II

from Teeth (dental plaque)

from Saliva



from Tongue

Photo – 3: Technique Of Collection Of Samples

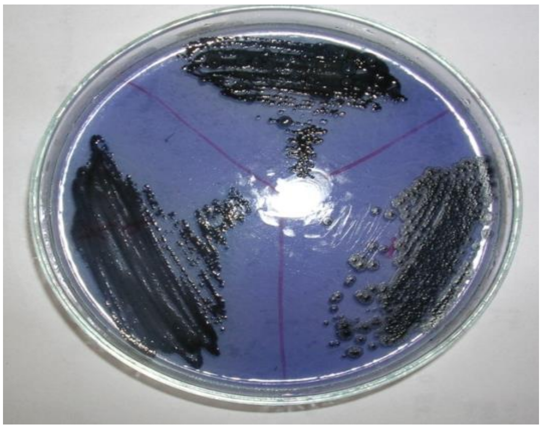
Photo – 4 : Armamentarium for clinical examination and sample collection

Photo – 5 : Armamentarium of culture technique

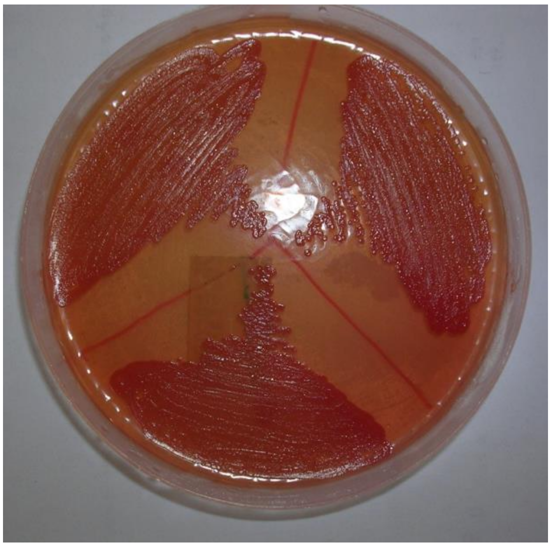
Photo – 6 :Inoculation of Sample in Culture Plates

Photo – 7: Incubator with Culture Plates

Blood agar Plate

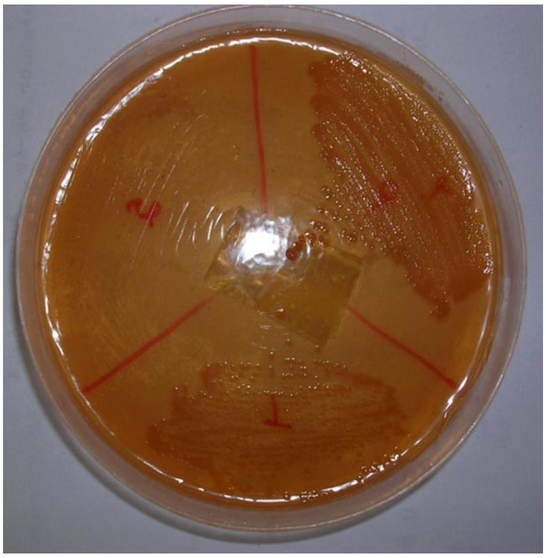


Mitis Salivarius agar Plate



Mac Conkey agar Plate

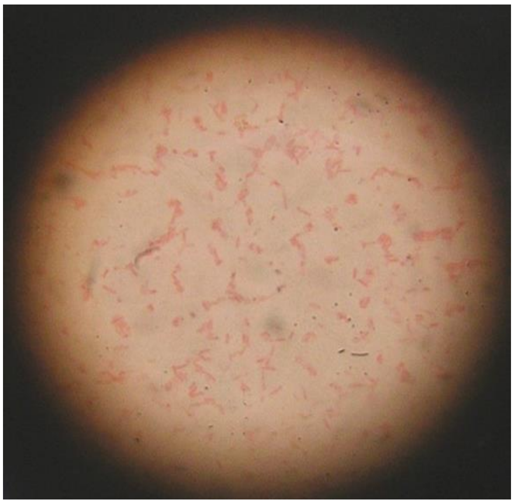
Photo – 8 : Culture Plate Photographs of Study Group with more Microbial Flora



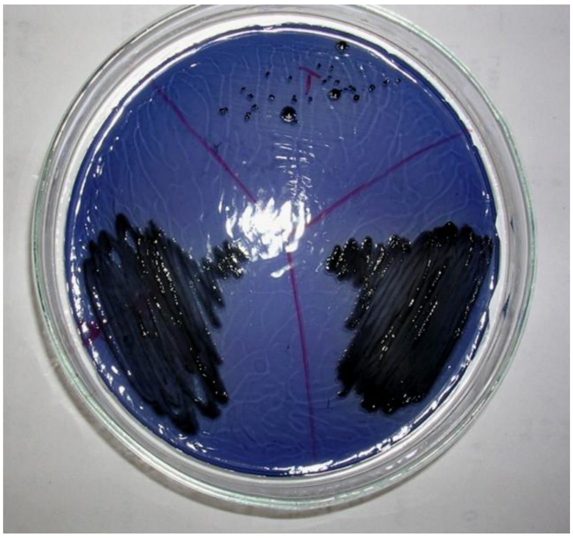
Mac Conkey agar Plate

Photo – 9 : Culture Plate Photographs of Control Group with less Microbial Flora

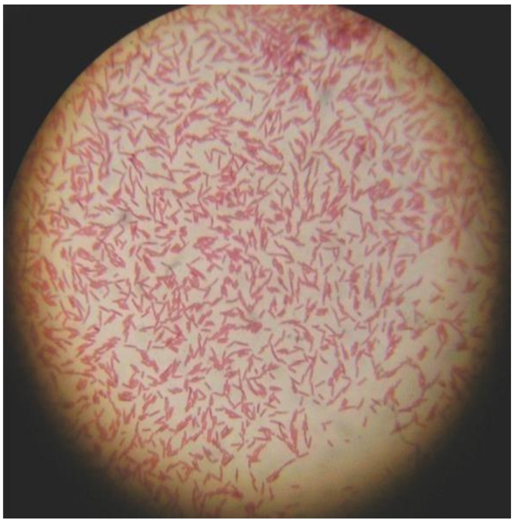
Blood agar Plate



Moraxella catarrhalis

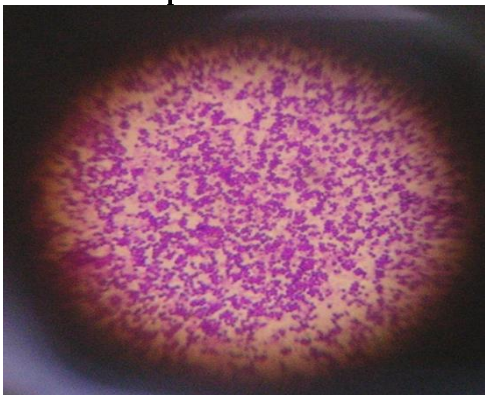


Mitis Salivarius agar Plate

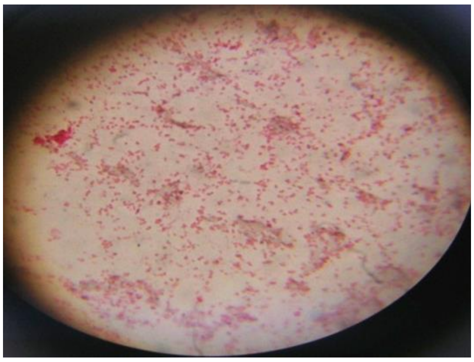


Corynebacteria

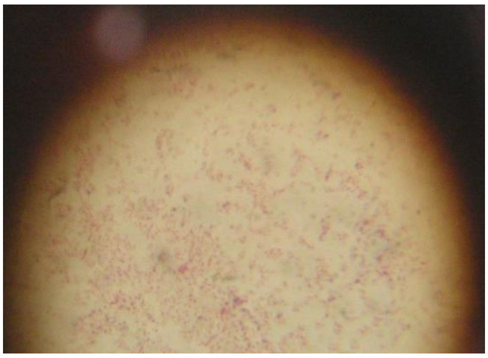
Streptococcus mutans



Staphylococcus aureus



Escherichia coli



Haemophilus

Photo – 10 : Bacterias Isolated by Culture Technique as viewed under microscope in 1000 X magnification

III. RESULTS AND OBSERVATIONS

The present study was undertaken to evaluate the microbial flora in oral cavity of tobacco smokers and to compare them with age matched non-smokers. 35 subjects were included in the study group and 35 subjects in control group after obtaining a written informed consent for participation. All the subjects were male patients with age ranging from 20 to 30 years. The samples were collected from dental plaque, saliva and tongue smear using aseptic technique and taken to laboratory for inoculation in different culture medias.

The over all results of microbial flora count from three different sites in study group and control group are shown in table 1,2,3,4,5 and 6. From the total microbial flora count as shown in table 7, the calculated mean value in study group was 5.64 X 10^{5} C.F.U./ ml, while that in control group was 4.77 X 10^{5} C.F.U./ ml, showing a higher amount of microbial flora count in study group. The median counts of microbial flora from all three different sites are shown in table 8. The median counts of study group and control group are also shown in graph number 1,2 and 3.

The results have been evaluated using Wilcoxon rank test to calculate the probability (p) value of all the microbes and shown in table 9. The results suggest that in the salivary samples the gram – ve cocci, Moraxella catarrhalis has a p value of 0.047 ($p < 0.05$) and the gram + ve bacilli, Corynebacteria has a p value of 0.027 ($p<0.05$); which are statistically significant, whereas the p value for other microbes were not statistically significant in study group and control group.

TABLE 1: Microbial flora count from Dental Plaque: STUDY GROUP (X 10^5 C.F.U. /ml)

S. No	Age	Cocci					Bacilli						
		Gram + ve				Gram - ve	Gram + ve	Gram - ve					
		S.mutans	S.mitis/ salivaris	Other Strep	Staph aure	M. catarrhalis	Corynebac	Klebsiella	Citrobacter	E. coli	Proteus	Pseudomonas	Haemophilus
1	22			2.0	2.0								
2	30			1.0									
3	20	1.0										2.0	
4	22	0.014		1.0		0.08							
5	21	0.1		1.0		0.4		0.004					
6	25	0.04		1.0	0.5								
7	26		0.028	0.2									0.6
8	25		0.04	2.0			0.02			0.02			
9	30	0.14		2.0	0.3								
10	23	0.014		1.0		0.01							
11	24	0.2		0.15		0.006							
12	30	0.4		0.5	0.2								
13	28	0.2		0.2									
14	22		0.014	0.2	0.5								
15	26		0.04	0.4		0.1							
16	21	0.016		2.0		0.05			0.014				
17	21	0.3		0.2									
18	22	0.024		1.0		0.2							
19	28		0.02	0.5		0.5							
20	25		1.0	1.0		0.2							
21	20	1.0		0.3							0.4		
22	21	1.0		1.0									1.0
23	21	0.04		1.0			0.2						0.5
24	30	1.0				2.0		2.0					
25	24		1.0	1.0	0.5								
26	25	0.2		1.0		0.4							
27	23	0.5		0.3									
28	25	0.1		0.1									
29	22	0.04	0.1	0.5								2.0	
30	30			2.0									2.0
31	21		0.012	2.0									
32	21		0.2	0.4									
33	22								0.024				
34	24	0.1		1.0		0.02							
35	21	0.014		1.0		0.06							

TABLE 2: Microbial flora count from Dental Plaque: CONTROL GROUP (X 10^5 C.F.U. /ml)

S. No	**Age**	**Cocci**					**Bacilli**						
		Gram + ve				**Gram - ve**	**Gram + ve**	**Gram - ve**					
		S.mutans	**S.mitis/ salivaris**	**Other Strep**	**Staph aure**	**M. catarrhalis**	**Corynebac**	**Klebsiella**	**Citrobacter**	**E. coli**	**Proteus**	**Pseudomonas**	**Haemophilus**
1	23	0.2		0.2				2.0					
2	24							2.0					
3	24			1.0	1.0			2.0				0.4	
4	24	1.0		2.0						2.0			
5	24			2.0				2.0					
6	21									2.0			
7	21				0.24	0.04							
8	25		0.06										0.2
9	21		0.04	2.0	1.0					0.06			
10	21			0.2					0.014				
11	23	0.018		0.1	0.06								
12	23	0.008		1.0									
13	22		0.04	0.5	0.5								0.6
14	22		0.08	2.0									
15	21		0.002	0.8	0.1								
16	22			0.2		0.02							
17	25		0.012	0.6									
18	25		0.15	1.0	0.2								
19	26		0.006	0.3									
20	22	0.02		1.0		0.1			0.01				
21	21			1.0									
22	23			1.0									
23	25	0.1	0.04	1.0					0.008				
24	30		0.01	1.0			0.1						
25	23			0.2	0.1								
26	24	0.3		2.0		0.2							
27	24			1.0		0.4							
28	22		0.2	2.0		0.5							
29	21			1.0		0.3				0.01			
30	22			1.0		0.04							
31	22		0.004	0.2									
32	21			1.0									
33	23			1.0		0.3							
34	21		0.18	0.2						0.008			
35	24		0.06	1.0			0.4						

TABLE 3: Microbial flora count from Saliva: STUDY GROUP (X 10^5 C.F.U. /ml)

S. No	Age	Cocci					Bacilli						
		Gram + ve				Gram - ve	Gram + ve	Gram - ve					
		S.mutans	S.mitis/ salivaris	Other Strep	Staph aure	M. catarrhalis	Corynebac	Klebsiella	Citrobacter	E. coli	Proteus	Pseudomonas	Haemophilus
1	22			0.016	0.4								
2	30			2.0									
3	20		0.2									2.0	
4	22	0.05		1.0		0.2							1.0
5	21	0.2		2.0			0.2						
6	25	0.2		2.0									
7	26	0.056		0.04		0.1		0.016					
8	25		0.12	2.0	1.0								
9	30	0.04		1.0	0.2								
10	23	0.08		0.2		0.05							
11	24	0.5		2.0									0.4
12	30	1.0		0.4	2.0					0.002			
13	28	1.0		2.0			0.3						
14	22		0.2	0.5	0.5								
15	26	0.056		0.1		0.1							
16	21	0.2		2.0									
17	21	0.1		0.1									
18	22	0.1		0.05									
19	28		0.5	1.0		0.4							
20	25		0.1	0.9		0.3							
21	20		0.01	1.0	0.02						0.1		
22	21	1.0		1.0									1.0
23	21	1.0		1.0			1.0						
24	30	1.0				2.0		2.0					
25	24	1.0		1.0	0.1								
26	25	0.1		1.0		0.3							
27	23	0.04		0.04									
28	25	1.0		0.2		1.0							
29	22	0.1	0.4	1.0									0.5
30	30		0.014	1.0		0.1							
31	21	0.1		1.0		1.0							
32	21		0.2	0.2									
33	22	0.5		1.0		0.5							
34	24	0.2		1.0			0.3						
35	21	0.1		1.0									

TABLE 4: Microbial flora count from Saliva: CONTROL GROUP (X 10^5 C.F.U. /ml)

S. No	**Age**	**Cocci**					**Bacilli**						
		Gram + ve				**Gram - ve**	**Gram + ve**	**Gram – ve**					
		S.mutans	**S.mitis/ salivaris**	**Other Strep**	**Staph aure**	**M. catarrhalis**	**Corynebac**	**Klebsiella**	**Citrobacter**	**E. coli**	**Proteus**	**Pseudomonas**	**Haemophilus**
1	23	0.1		0.2		0.2							
2	24	1.0		0.1		0.2							2.0
3	24			0.4							2.0		
4	24	2.0	0.3	1.0						2.0			1.0
5	24			0.4				2.0					
6	21							2.0					
7	21		0.24			0.04							
8	25	0.1		0.5									
9	21		0.08	1.0	1.0								
10	21			2.0					0.04				
11	23	0.024		0.35									
12	23	0.3		2.0	0.5								
13	22		0.1	0.6	0.3								
14	22	0.4		2.0									
15	21		0.07	2.0	0.2								
16	22			2.0									
17	25	0.4		0.4									
18	25	0.6		1.0	0.4								
19	26	0.01		0.02									
20	22	0.20		1.0									
21	21			0.5								0.08	
22	23	0.2		1.0									
23	25	0.05	0.05	1.0									
24	30			1.0			0.04						
25	23			0.1			0.02						
26	24		0.12	0.3		0.2							
27	24			2.0									0.2
28	22	2.0		2.0		0.1							0.6
29	21			2.0		0.04							
30	22	0.4		0.2		0.05							
31	22		0.004	0.3									
32	21	0.04		1.0									
33	23		0.014	0.3									
34	21	1.0		1.0		0.3							
35	24	0.1		1.0			0.2						

TABLE 5: Microbial flora count from Tongue: STUDY GROUP (X 10^5 C.F.U. /ml)

S. No	Age	Cocci					Bacilli						
		Gram + ve				Gram - ve	Gram + ve	Gram – ve					
		S.mutans	S.mitis/ salivaris	Other Strep	Staph aure	M. catarrhalis	Corynebac	Klebsiella	Citrobacter	E. coli	Proteus	Pseudomonas	Haemophilus
1	22	1.0			2.0	1.0	0.2						
2	30			1.0									
3	20											2.0	
4	22	1.0		0.5		1.0							0.2
5	21	0.4		1.0			1.0						
6	25			1.0	0.04								
7	26	1.0		2.0			1.0	0.006					0.4
8	25		1.0	2.0	1.0								
9	30	1.0		2.0	0.2			0.02					
10	23	1.0		1.0		0.2				0.04			
11	24	0.4		2.0									1.0
12	30	1.0								0.012			1.0
13	28	1.0		1.0			0.6						
14	22		0.1	0.2	0.3								1.0
15	26	0.5		1.0		0.5							
16	21		0.2	1.0					0.09				
17	21	0.1		1.0									
18	22	1.0		0.4		1.0		0.004					
19	28		1.0	0.5			0.2						
20	25		1.0	1.0				1.0					
21	20		0.1	0.3							1.0		0.3
22	21	0.4		1.0	0.5					0.004			1.0
23	21	1.0		1.0			0.2						
24	30	2.0		2.0		2.0		2.0					
25	24	1.0		1.0									
26	25	0.6		1.0		0.4							
27	23	1.0		0.2									
28	25	1.0		0.3		0.4	0.4						
29	22		0.4	0.4								1.0	
30	30	0.5	0.03	1.0		0.04		0.004					2.0
31	21	0.3		1.0		0.2		0.01					
32	21	0.1		0.2	0.5								0.5
33	22	0.5		0.5		0.1			0.006				
34	24	0.5		2.0									
35	21	1.0		1.0		0.04							

TABLE 6: Microbial flora count from Tongue: CONTROL GROUP (X 10^5 C.F.U. /ml)

S. No	**Age**	**Cocci**					**Bacilli**						
		Gram + ve				**Gram - ve**	**Gram + ve**	**Gram - ve**					
		S.mutans	**S.mitis/ salivaris**	**Other Strep**	**Staph aure**	**M. catarrhalis**	**Corynebac**	**Klebsiella**	**Citrobacter**	**E. coli**	**Proteus**	**Pseudomonas**	**Haemophilus**
1	23							2.0					
2	24	0.1		0.1		1.0							
3	24	0.2		0.2								0.4	
4	24	2.0		2.0	0.2					2.0			
5	24			2.0				2.0					
6	21							2.0					
7	21		2.0			0.5							
8	25	1.0		1.0									1.0
9	21	1.0		2.0	1.0					0.06			
10	21	1.0		1.0					0.06				0.7
11	23	0.5		1.0	0.1								
12	23	0.04		2.0	0.5								
13	22		0.1	2.0	0.2								
14	22	1.0	0.04	0.3			0.04			0.004			1.0
15	21		0.04	1.0	1.0								
16	22		0.12	2.0		0.6							
17	25			0.5						0.004			1.0
18	25	1.0		1.0	0.5			0.02					
19	26	1.0		1.0			0.2						
20	22	1.0		1.0				0.026					
21	21		0.2	1.0									
22	23		0.1	1.0	0.2								0.2
23	25	0.1		0.5			0.2						
24	30			1.0			1.0						
25	23	0.4		1.0	0.3								
26	24	0.6		0.2		0.4							
27	24	0.2		2.0		0.08							
28	22	2.0		2.0		0.4							
29	21		0.4	0.2		0.2							0.2
30	22	0.3		2.0		1.0							
31	22	0.5		0.2		0.2							
32	21	0.2		0.5							0.5		
33	23			1.0									0.3
34	21	1.0		1.0			0.5						
35	24	0.6		1.0			0.5						

TABLE 7: Total Microbial Flora Count

Sl.No	Study Group		**Control Group**	
	Age	**Count (X 10^5 C.F.U./ml)**	**Age**	**Count (X 10^5 C.F.U./ml)**
1	22	8.616	23	4.90
2	30	4.00	24	6.50
3	20	7.20	24	7.60
4	22	6.044	24	17.50
5	21	6.304	24	10.40
6	25	4.78	21	6.00
7	26	5.446	21	2.81
8	25	9.20	25	3.86
9	30	6.90	21	9.24
10	23	3.594	21	5.014
11	24	6.656	23	2.148
12	30	6.514	23	6.348
13	28	6.30	22	4.94
14	22	5.514	22	6.864
15	26	2.796	21	5.212
16	21	5.57	22	4.94
17	21	1.80	25	2.916
18	22	3.778	25	5.87
19	28	4.62	26	2.536
20	25	6.50	22	4.536
21	20	4.53	21	2.70
22	21	8.904	23	3.70
23	21	6.94	25	3.048
24	30	18.00	30	4.15
25	24	6.60	23	2.12
26	25	5.00	24	4.32
27	23	2.08	24	5.88
28	25	4.50	22	11.80
29	22	6.44	21	4.35
30	30	8.688	22	4.99
31	21	5.622	22	0.908
32	21	2.30	21	3.24
33	22	3.13	23	2.914
34	24	5.12	21	5.188
35	21	4.214	24	4.86

TABLE 8: Median values of microorganism from three different sites

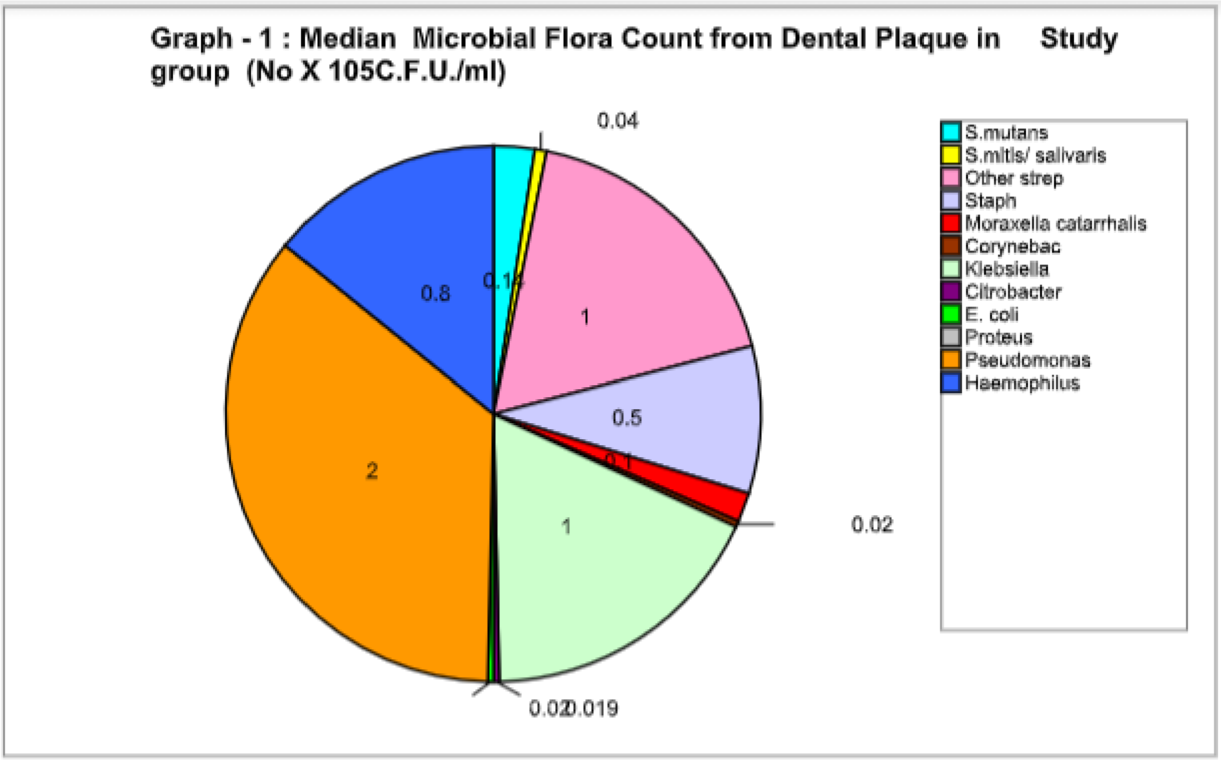
S. No	*Microorganisms*	**SITE**					
		Dental Plaque		**Saliva**		**Tongue**	
		S.G.	**C.G.**	**S.G.**	**C.G.**	**S.G.**	**C.G.**
1.	S. mutans	0.14	0.10	0.20	0.20	1.00	0.60
2.	S. mitis/salivaris	0.04	0.04	0.20	0.08	0.30	0.11
3.	Other Strep	1.00	1.00	1.00	1.00	1.00	1.00
4.	Staphylococcus aureus	0.50	0.22	0.40	0.40	0.50	0.30
5.	M. catarrhalis	0.10	0.20	0.30	0.15	0.40	0.40
6.	Corynebactria	0.02	0.25	0.30	0.03	0.40	0.35
7.	Klebsiella	1.00	2.00	1.00	2.00	0.01	2.00
8.	Citrobacter	0.019	0.01	0	0	0.048	0.06
9.	Escherichia coli	0.02	0.06	0.002	2.00	0.012	0.032
10.	Proteus	0	0	0.10	2.00	1.00	0.50
11.	Pseudomonas	2.00	0.04	2.00	0.08	1.50	0.40
12.	Haemophilus	0.80	0.40	0.75	0.80	1.00	0.70

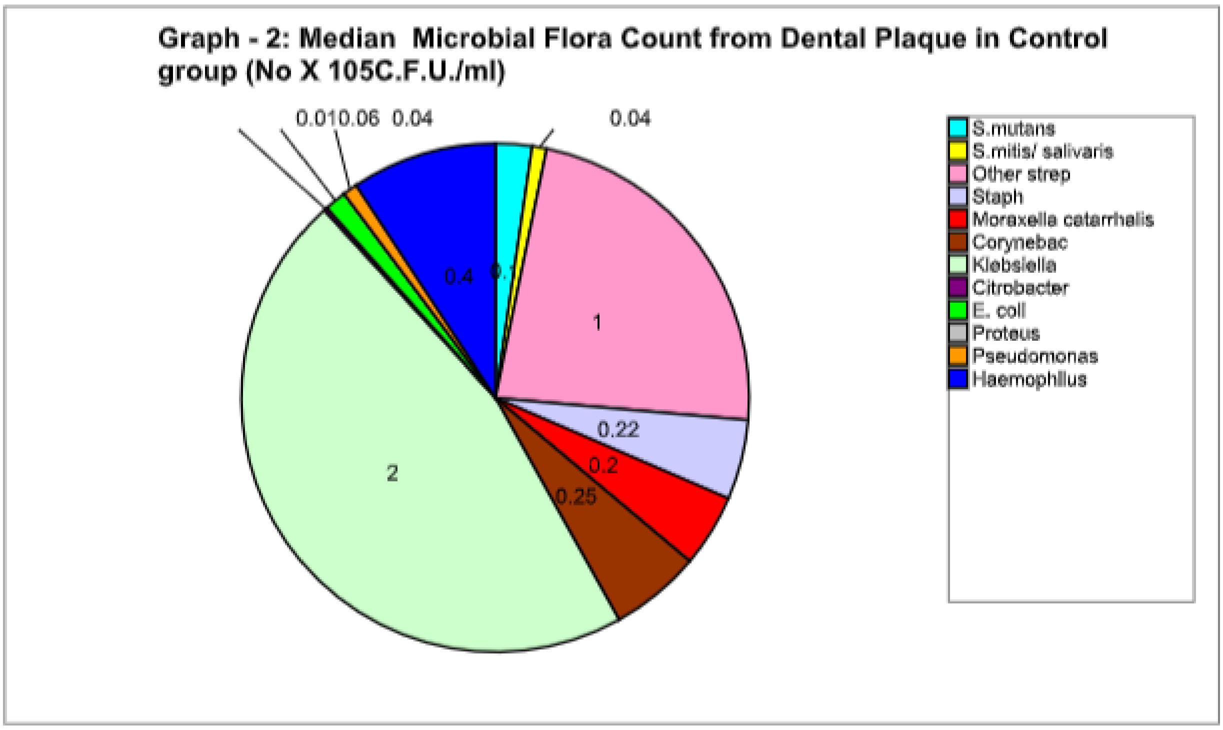
S.G. = Study group
C.G. = Control group
S. mutans = Streptococcus mutans
S. mitis/salivaris = Streptococcus mitis/salivaris
Others Strep = Other Streptococci
Staph aure = Staphylococcus aureus
M. catarrhalis = Moraxellacatarrhalis

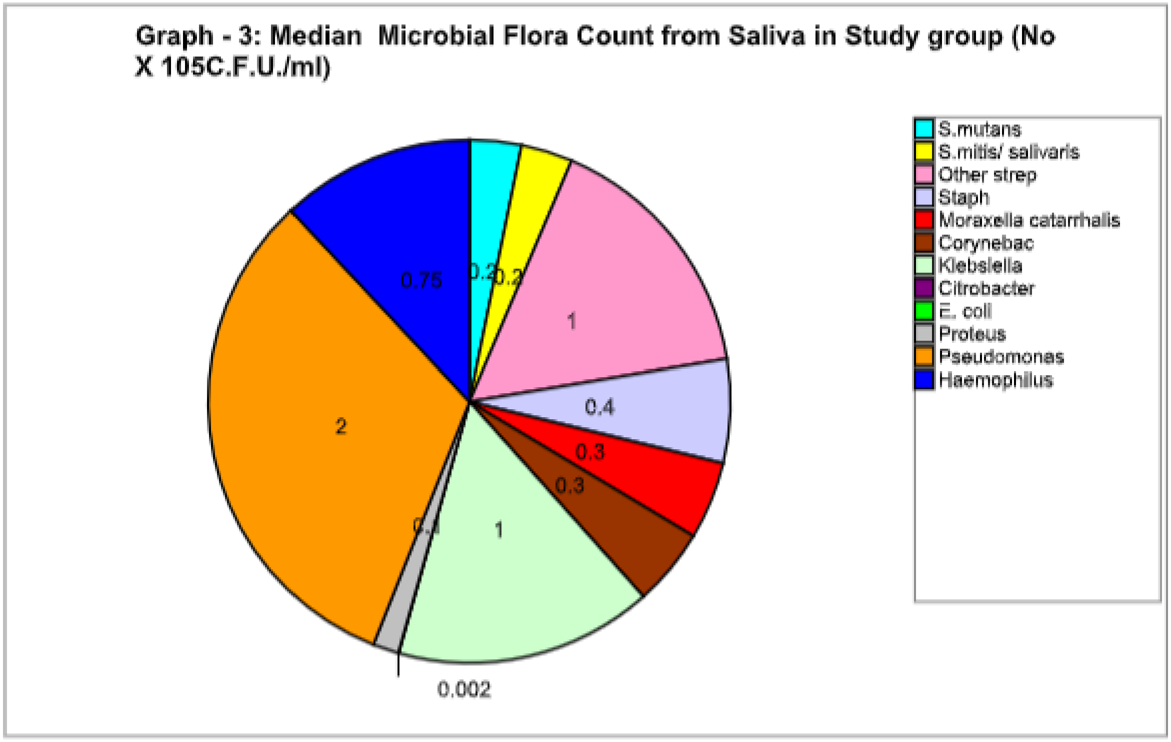
TABLE 9: Probability values of microorganism fromthree different sites

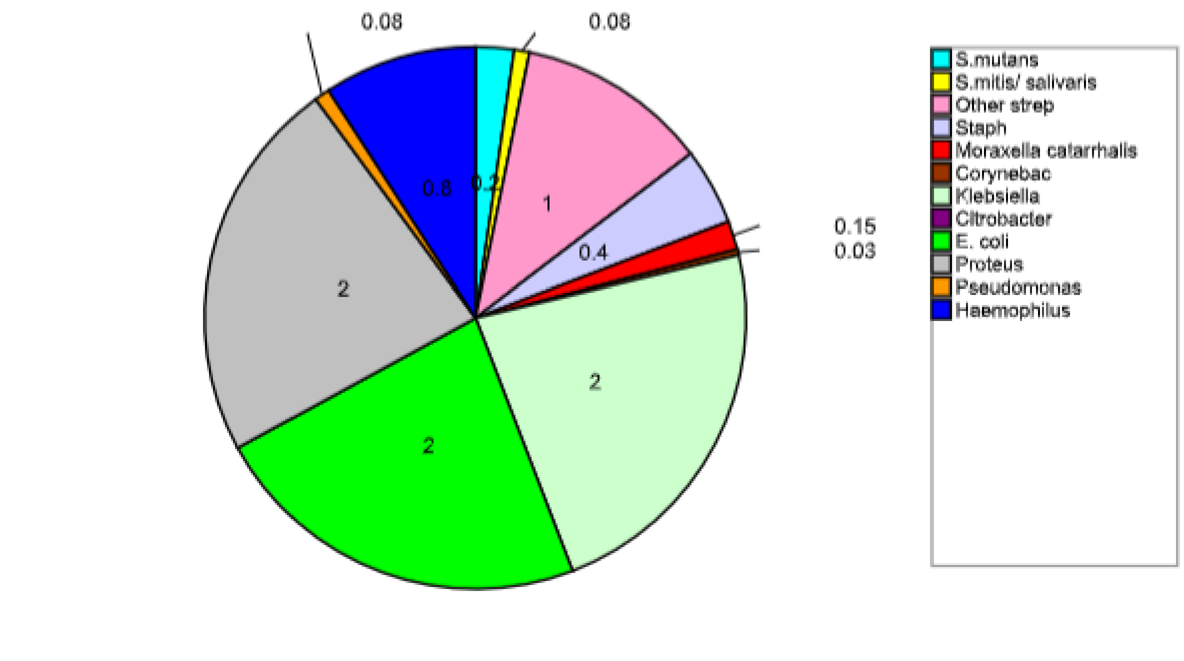
S. No	**Microorganisms**	***SITE***		
		Dental Plaque	**Saliva**	**Tongue**
1.	S. mutans	0.557	0.950	0.447
2.	S. mitis/salivaris	0.377	0.249	0.457
3.	Other Strep	0.773	0.726	0.602
4.	Staphylococcus aureus	0.238	0.806	0.306
5.	M. catarrhalis	0.789	**0.047** (S)	0.857
6.	Corynebactria	0.102	**0.027** (S)	0.554
7.	Klebsiella	0.157	0.317	0.056
8.	Citrobacter	0.139	N.A	1.00
9.	Escherichia coli	0.766	0.317	0.714
10.	Proteus	N.A	0.317	0.317
11.	Pseudomonas	0.157	0.317	0.221
12.	Haemophilus	0.240	0.767	0.437

N.A = Not assessed
S = Statistically significant
S. mutans = Streptococcus mutans
S. mitis/salivaris = Streptococcus mitis/salivaris
Others Strep = Other Streptococci
Staph aure = Staphylococcus aureus
M. catarrhalis = Moraxella catarrhalis


Graph - 1 : Median Microbial Flora Count from Dental Plaque in Study group (No X 105C.F.U./ml)


Graph - 2: Median Microbial Flora Count from Dental Plaque in Control group (No X 105C.F.U./ml)


Graph - 3: Median Microbial Flora Count from Saliva in Study group (No X 105C.F.U./ml)



Graph - 4: Median Microbial Flora Count from Saliva in Control group (No X 105C.F.U./ml)

IV. DISCUSSION

Various studies in the past have reported evidence about the impact of smoking on oral microbiology. Smoking is an established risk factor for various diseases such as oral cancer, leukoplakia, smoker's palate, poor wound healing, acute necrotizing ulcerative gingivitis etc [3,4].

Hence, this case control study had been undertaken to examine the change in microbial flora in samples taken from dental plaque, saliva and tongue smear in systemically healthy young adult smokers and non-smokers. Among the different modalities available to analyze the oral microbial flora, in this study culture media technique was used to isolate them from samples collected.

The present study clearly showed an increased amount of microbial flora in smokers as compared to non-smokers. The mean value of total microbial flora isolated from smokers is more (5.64 X 10^5 C.F.U. / ml) than in non-smokers (4.77 X 10^5 C.F.U. / ml). The salivary samples of the study group and control group, with respect to Moraxella catarrhalis and Corynebacteria showed p value less then 0.05, which is statistically significant. Thus suggesting a greater extent of colonization by pathogenic microbes in smokers as compared to non-smokers, thus increasing the chances for oral diseases.

Donna L. M et al studied the effect of smoking and periodontitis on the microbial flora of oral mucous membranes and saliva in systemically healthy subjects. Their study suggests that in smokers, higher proportions of Porphyromonas nigrescens, Fusobacterium and Actinomyces species were present. Notably, 81 % of the smoking group had periodontal diseases, compared with non-smokers [19].

V. CONCLUSION

The present study, which includes 35 male smokers and equal number of non-smokers. The study shows that there is a definite increase in the amount of microbial flora in tobacco smokers as compared to non-smokers. These increased microbes may ultimately increase the chances for oral diseases and impaired wound healing thus affecting the treatment outcome of oral health. This increased number of bacterias may translocate through the damaged mucosa, thereby increasing the risk of local and systemic infections in smokers.

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