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EVALUATION OF MOUTHWASH CONTAINING LACTOBACILLUS IN LOCALIZED PERIODONTAL POCKET AS AN ADJUNCT TO NON-SURGICAL PERIODONTAL THERAPY IN CHRONIC PERIODONTITIS - A CLINICAL & MICROBIOLOGICAL STUDY

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ABSTRACT

Background: The study was to evaluate the effect on clinical and microbiological analysis of Lactobacillus containing probiotic supplementation adjunctive to initial periodontal therapy in patients with chronic periodontitis. **Materials and Methods:** 20 sites were selected having diagnosed with Chronic Periodontitis from the Department of Periodontology, I.T.S Dental College, Muradnagar, Ghaziabad. Patients were equally and randomly divided into two groups. Each patient with minimum of pocket probing depth of 4 mm and gingival index of ±2.The test group received scaling and root planing (SRP) and pocket irrigation with probiotic suspension, the control group received (SRP) and irrigation with placebo. The clinical and microbiological parameters such as plaque index, gingival index, bleeding on probing, probing pocket depth, clinical attachment level, colony forming unit (CFU/ml) were recorded for each patient at baseline and 21 days. **Results**: There was significant improvement in the gingival index, pocket probing depth and total bacterial count in test group as compare to control group after 21 days. There was no significant difference seen in plaque index in test and control group. **Conclusion**: Probiotic containing suspension may be a useful adjuvant agent to slow re-colonization and improve clinical outcomes of chronic periodontitis. Further studies are required to the optimal dosage of the lozenges and time period for that they should be given.

KEYWORDS: Chronic Periodontitis; Probiotic; Mouthwash.

1. INTRODUCTION

There are various microbial model used in treatment of periodontal disease. With the threat of widespread antibiotic resistance rendering many antibiotics are useless against many diseases; there is an increased necessity not only to minimize antibiotic use and develop novel non antibiotic based treatments, but also to raise the profile of disease prevention. Probiotics may be a promising area of research in periodontal therapy. Probiotics are defined as "live microorganisms that when administered in adequate amounts confer health benefits on the host."[1] An easy and simple way to define probiotics is "those supplements of food which contain microbial flora that has the capacity to effect human health in a beneficial way.".[2]

Many studies have been done in the late 90's on this subject.[1,2] There are certain beneficial bacteria that keep the harmful pathogens at bay. In fact, beneficial microbes represent the future of medicine. Despite the difficulties inherent in characterizing the microbiology of periodontal diseases, a small group of pathogens is recognized because of their close relationship with periodontal disease. The organisms in our oral cavity are a mixture of commensals and pathogens. A commensal microorganism is defined as one that lives on or within the host, but does not cause any apparent disease.[3]

Mechanical removal of supragingival plaque is the most effective tool to prevent gingivitis (Loe et al. 1965) but most individuals do not adequately control plaque accumulation and gingivitis continues to be prevalent. To overcome this hindrance, antimicrobial products in the form of dentifrices or mouthwashes have been tested for their adjunctive efficacy in reducing plaque and gingivitis. Among them, chlorhexidine is regarded as a gold standard in dentistry for the prevention of dental plaque. Though very effective, it has certain side effects such as brown discoloration of teeth, oral mucosal erosion and bitter taste.[4]

Probiotics are living microorganisms, principally bacteria that are safe for human consumption and when ingested in sufficient quantities, have beneficial effects on human health, beyond basic nutrition.[5] So, the aim of the study was to evaluate the effect on clinical and microbiological analysis of Lactobacillus containing probiotic supplementation adjunctive to initial periodontal therapy in patients with chronic periodontitis.

2. MATERIALS AND METHORDS

2.1 Patient's selection criteria

The presented clinical study was performed at the Department of Periodontology and Oral Implantology, I.T.S Centre for Dental Studies & Research, Ghaziabad, India. The research protocol was reviewed and approved by the Ethical Committee of the Institution. All patients were educated about the study and then written consent was acquired before enrolment in the study.

A flow diagram for complete methodology is presented in Figure 1. Twenty (20) patients diagnosed with the problem of **localized** gingivitis were included, having Periodontal Pockets (3-4 mm). The patients were between the age group of 18-45 years. The exclusion criteria was patient with allergy to the medicament used in the study, medically compromised, lactating mother.

2.2. Clinical evaluation and indices

Prior to the therapy, all patients received thorough supragingival and subgingival scaling and root planing, Oral hygiene instructions were given to the patients and asked to perform tooth brushing twice daily using modified bass technique. Following clinical parameters were assessed at baseline and 21Days.

- Plaque Index (Turesky – Gilmore- Glickman modification of Quingley-Hein Plaque Index).[6]
- Gingival Index (loe & silness, 1963).[7]
- Pocket Probing Depth (PPD) in mm.
- Total Colony count (CFU/ml)

2.3 Patient's allocation & procedures

Patients were assigned equally but randomly to one of the two treatment groups (N = 20). Envelopes containing identifications for treatment groups were enclosed, mixed and then numbered. Each participant was randomly selected to one of the two following groups. In control group scaling and root planing along with placebo irrigation in periodontal pocket would be performed on selected sites. In test group scaling root planing was given along with probiotic irrigation in periodontal pocket. Microbiological samples would be collected using paper points from the subgingival plaque samples will be collected using curettes from the pocket, at baseline and 21 days and send for microbiological lab for bacterial count (Figure 2). The patients in Test Group A received solution irrigation with probiotic suspension that was prepared in Lab of pharmacology department of ITS-CDSR Murad Nagar Ghaziabad U.P containing Lactobacillus acidophilus, Lactobacillus sporogenes, Bifidobacterium longum Lactobacillus rhamnosus and Saccharomyces boulardii.

2.4. Statistical analysis

Statistical analysis was carried out using Statistical Package for Social Sciences (SPSS Inc., Chicago, IL, version 16.0 for windows). Test for qualitative variables, mean and standard deviation were calculated. To test the significance of difference of mean rank in GI, PI, PD, CFU of two groups was test by Mann-Whitney *U* test.

1. RESULTS

The mean Plaque index (PI) scores at baseline in control and test group were 2.50 ± 0.527 and 2.30 ± 0.483 respectively. At 21st day the mean PI scores were found to be 2.00 ± 0.00 and 1.50 ± 0.527 in the control and test group respectively. There was no significant difference in the mean PI score at 21st day in both test and control groups (TABLE 1). The unpaired t test was also used to compare the mean PI scores between that there is no significant difference in PI score at baseline and 21st Days ($p>0.05$). The mean Gingival index (GI) scores at baseline in control and test group were 2.20 ± 0.422 and 2.20 ± 0.422 respectively. At 21st Day the mean GI scores were found to be 2.00 ± 0.00 and 1.10 ± 0.316 in the control and test group respectively. There was highly significant difference seen in the mean PI score at 21st Day in both test and control groups (TABLE 2). The mean pocket probing depth (PPD) scores at baseline in control and test group were 4.60 ± 0.699 and 4.40 ± 0.699 respectively. At 21st Day the mean PPD scores were found to be 4.10 ± 0.316 and 2.90 ± 0.738 in the control and test group respectively. There was highly significant difference seen in the mean PPD score at 21st Day test and control groups (TABLE 3). The mean Colony forming unit (CFU/ml) scores at baseline in control and test group were 373.60 ± 43.295 and 362.80 ± 42.166 respectively. At 21st Day the mean CFU/ml scores were found to be 347.60 ± 44.061 and 272.80 ± 52.917 in the control and test group respectively. There was significant difference found in the mean CFU/ml score at 21st Day in both test and control groups (TABLE 4). The unpaired t test was used to compare the mean CFU/ml scores between that there is significant difference in CFU/ml score at baseline and 21st Days ($p\leq0.05$).

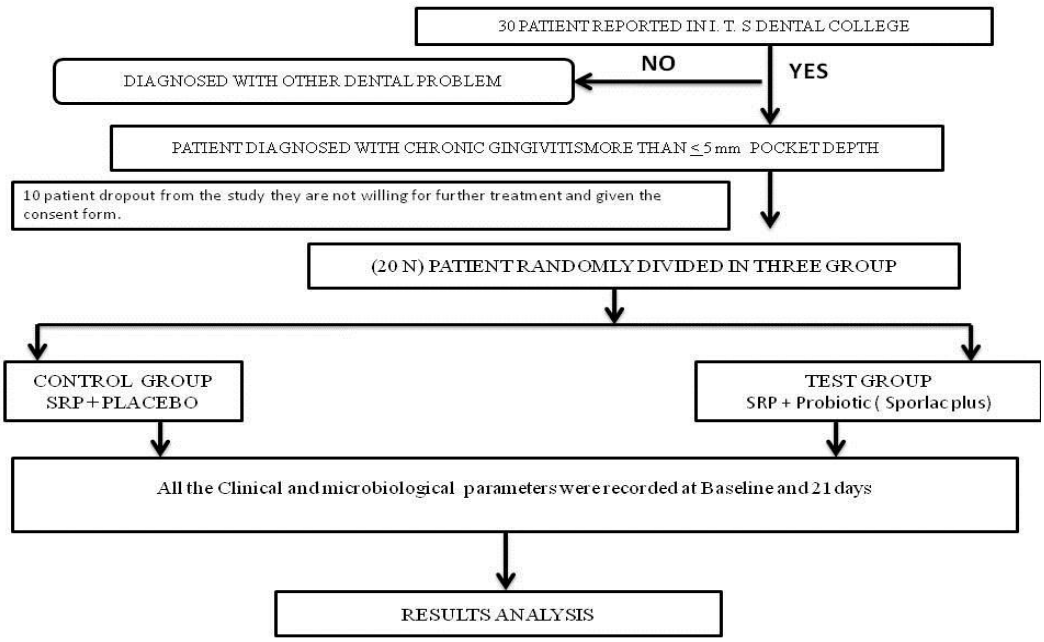
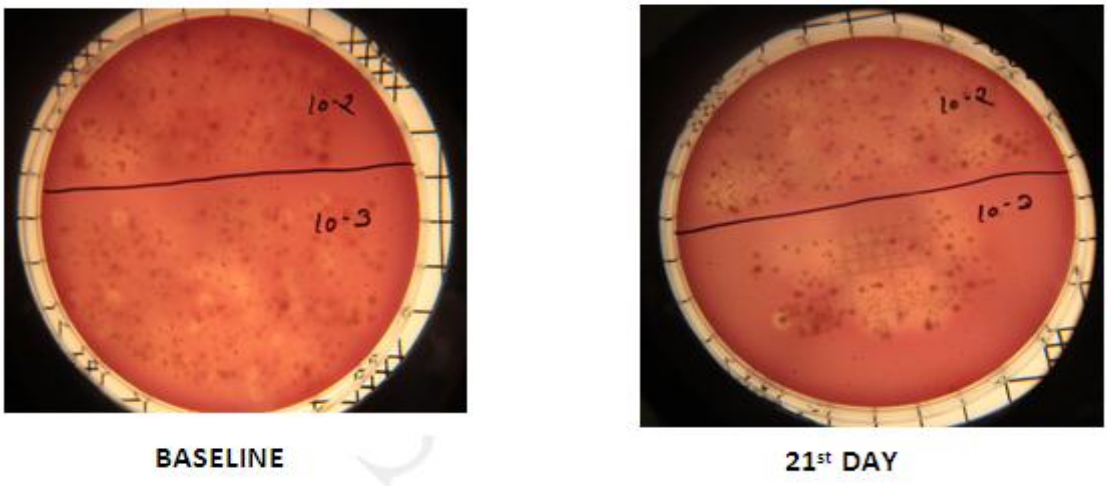


FIGURE 1: FLOW CHART INDICATING THE METHODOLOGY IMPLEMENTED IN THE PRESENT CLINICAL STUDY

TEST GROUP



CONTROL GROUP

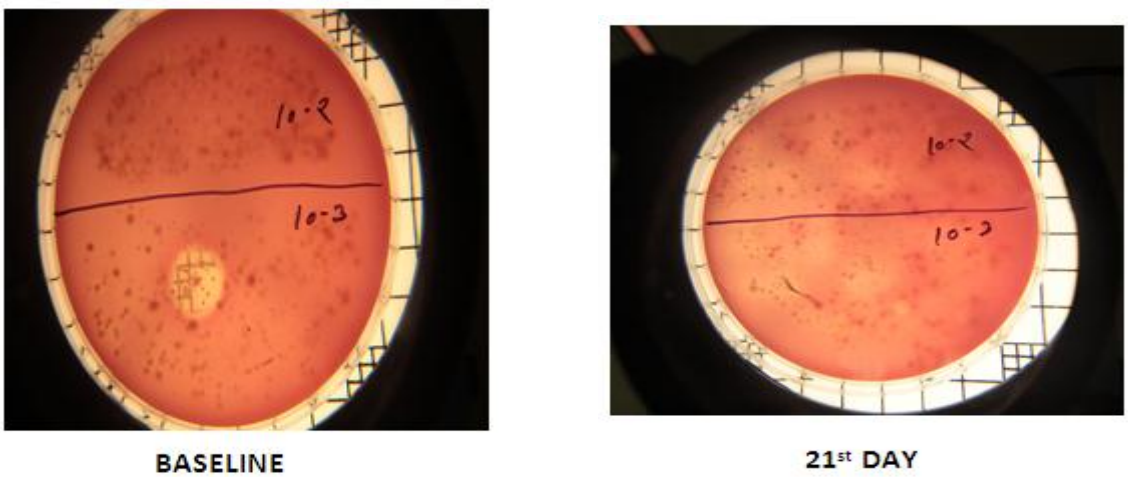


FIGURE 2: CFU ON BLOOD AGAR IN TEST AND CONTROL GROUP AT DIFFERENT TIME INTERVAL

TABLE 1: COMPARISON OF PLAQUE INDEX BETWEEN TEST AND CONTROL GROUPS

PI	Groups	Mean	Std. Deviation	t-test value	P-value	Mean Difference
Baseline	CONTROL GROUP	2.50	.527	.374	-.890	.200
	TEST GROUP	2.30	.483			
At 21 DAYS	CONTROL GROUP	2.00	.000	**.012#**	-2.517	.500
	TEST GROUP	1.50	.527			
Unpaired t-test ***Non-significant difference (p-value<0.05)**						

TABLE 2: COMPARISON OF GINGIVAL INDEX BETWEEN TEST AND CONTROL GROUPS

GI	Groups	Mean	Std. Deviation	t-test value	P-value	Mean Difference
Baseline	CONTROL GROUP	2.20	.422	1.000	.000	.000
	TEST GROUP	2.20	.422			
At 21 DAYS	CONTROL GROUP	2.00	.000	**.000***	-3.943	.900
	TEST GROUP	1.10	.316			
Unpaired t-test ***significant difference (p-value<0.05)**						

TABLE 3: COMPARISON OF POCKET PROBING DEPTH BETWEEN TEST AND CONTROL GROUPS

PD	Groups	Mean (mm)	Std. Deviation	t-test value	P-value	Mean Difference
Baseline	CONTROL GROUP	4.60	.699	.435	-.781	.200
	TEST GROUP	4.40	.699			
At 21 DAYS	CONTROL GROUP	4.10	.316	**.001***	-3.430	1.200
	TEST GROUP	2.90	.738			
Unpaired t-test ***significant difference (p-value<0.05)**						

TABLE 4: COMPARISON OF COLONY FROMING UNIT BETWEEN TEST AND CONTROL GROUPS

CFU	Groups	Mean	Std. Deviation	t-test value	P-value	Mean Difference
Baseline	CONTROL GROUP	373.60	43.295	.820	-.228	10.800
	TEST GROUP	362.80	42.166			
At 21 DAYS	CONTROL GROUP	347.60	44.061	**.002***	-3.031	74.800
	TEST GROUP	272.80	52.917			
Unpaired t-test ***significant difference (p-value<0.05)**						

2. DISCUSSION

The plaque scores were assessed to monitor the patient's oral hygiene and its effects on the soft tissues. There is conclusive evidence regarding the importance of plaque control on the outcome of surgical procedures in general as well as regenerative attempts. *Nyman et al*[8] demonstrated that poor plaque control and recall compliance resulted in a loss of periodontal support following periodontal surgery. Good plaque control and maintenance of oral hygiene has been cited as an important factor in treatment outcome. These are also in accordance with *Noordin and Kamin*[9] who conducted a study on school children and assigned them into placebo, chlorhexidine and probiotic groups; and plaque scores were recorded at different intervals they found that probiotic mouthrinse group was more effective for inhibition of dental plaque accumulation after 14 days of intervention and also after 3 weeks of discontinuation of intervention. (Chx study?).

The *gingival index* was assessed to monitor the patient's gingival health and condition. The results of our study are consistent with the study done by *Harini and Anegundi*[10] evaluated clinically the efficacy of a probiotic and chlorhexidine mouthrinses on plaque and gingival accumulation in children for 14 days and concluded that the probiotic mouthrinse was found effective in reducing plaque accumulation and gingival

inflammation. This is attributable to good oral hygiene maintenance by the patients during the study. Inflammed gingiva which has tendency to bleed tends to reflect sites of active destruction and recurrence of the disease process. However, the low gingival scores reflect that the healing progressed uneventfully and periodontal destruction was arrested in the treated sites. This also reflects the resolution of disease process and maintenance of healthy periodontium in the previously diseased sites. They suggested that improved gingival condition could be due to lower plaque scores as well as proper maintenance of oral hygiene by the patient after non surgical periodontal therapy.

Reduction in periodontal pocket depths eliminates an environment that is hospitable for virulent periodontal pathogens and is considered as a definitive goal of periodontal therapy. The findings of our study are also in consistent with study by *Teughels et al.* in a randomized placebo controlled clinical trial evaluated the effects of L. reuteri containing probiotic lozenges and placebos as an adjunct to SRP in 30 patients with chronic periodontitis, monitored clinically and microbiologically at baseline, 3, 6, 9 and 12 weeks after therapy. Significant improvement in all clinical parameters reduced P. gingivalis levels, more pocket depth reduction and attachment gain in moderate and deep pockets was observed in the SRP + probiotic group.[11] Our results indicate that probiotics could be useful in the improvement/maintenance of oral health in subjects at a high risk of periodontal disease and add to the body of data supporting the effectiveness of both chlorhexidine and probiotic as antiplaque/antigingivitis agents. The advantages of using a probiotic mouthrinse are that as it contains friendly commensals, there is no issue of antibiotic resistance and there are no known/proven toxicities caused due to their use.[11]

However, we would like to state that the major limitation of our probiotic preparation is that it needs to be used immediately once prepared and cannot be stored. Thus, we would recommend that a proper vehicle is needed for delivering probiotics so that patient compliance can be improved. Longitudinal studies involving probiotics and further microbiological evaluation are also essential when prescribing them in place of antiseptics and antimicrobials.

3. CONCLUSION

In the recent times, when organisms are developing resistance to antibiotics, the emergence of probiotics appears to be a boon for the treatment of diseases. Researchers have confirmed that diseases of the periodontium are not confined to the oral cavity but have strong systemic effects. Hence, probiotics offer a natural and promising option to establish both a good oral and systemic health. In the present study, the probiotic sub gingival irrigation tested was effective in reducing plaque accumulation and gingival inflammation. Therefore, probiotic irrigation has a potential therapeutic value and further longterm studies are recommended to determine its efficacy.

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