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Effects of conventional & Non-conventional probiotic formulations on the gingival health status of undergraduate dental students in Kanpur City, India- A Randomized Controlled Trial

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Abstract

Aim: To evaluate the effects of conventional probiotic powder & non-conventional probiotic (yogurt) in plaque reduction and gingival status of undergraduate dental students.

Objectives

1. To compare the efficacy of the probiotic conventional product (powder) and non-conventional product (yogurt).
2. To compare the efficacy of the probiotic conventional products with the control group and non- conventional product with the control group
3. To compare the plaque scores in both the groups
4. To assess the gingival status in both the groups

Material and methods: In Group A, subjects consumed probiotic powder (1gm once daily) with 20 ml of water in a measuring jar using a stirrer as demonstrated. Subjects in group B were given a product similar in taste and colour. And they also followed the same procedure.

In Group C participants consumed probiotic yogurt (100gms once daily), while subjects in the control group D consumed plain yogurt (100gms once daily) for 14 days.

The gingival and plaque index was recorded. Statistical analysis was done using SPSS version 21.

Results: The results of the present study showed that statistically significant difference was found in the gingival and plaque scores on the 14th day amongst probiotic powder and yogurt groups as compared to the control groups.

Conclusion: Probiotics are useful in promoting oral health and in prevention of dental caries and periodontal diseases.

Keywords: Oral health, Probiotics, Probiotic powder, Probiotic curd, Plaque index

Introduction

Health is a valuable positive aspect of life; henceforth support of health is extremely important for a productive life. Oral health is an indispensable part of general health which can be accomplished by having an excellent oral hygiene. Lactobacilli assumes an essential role in maintaining the health by enhancing the immunity, creating a balance with the microflora and having an interaction with the other members of the flora. For therapeutic purposes, one of the strongest emerging field is the use of health promoting bacteria. Time has come to move the world's view of the treatment from specific elimination of bacteria to alterations in the bacterial environment by using probiotics.[1]

The word probiotic is derived from the Greek word meaning life. In 1908 the concept of probiotics came into existence when Eli Metchnikoff, a Nobel prize winner proposed that the long life of Bulgarian peasants had resulted after the consumption of fermented milk products.[2] The Food and Agriculture Organization (FAO) and World Health Organization (WHO) defined probiotics as 'Live microorganisms which when administered in adequate amounts confer a health benefit on the host' (FAO/WHO, 2001). There has been a rapid development in the understanding and use of such microorganisms in human conditions and diseases.[3]

Probiotics have been clinically proven to have potential benefits on the oral health especially in dental caries reduction, oral candidiasis, reduction in the oral malodour and periodontal disease. Hence, the oral cavity is considered as a soft target for the administration of probiotics. These probiotics adhere to the dental tissues as a part of a biofilm that acts as a protective barrier in combating oral diseases. Further the biofilm helps the bacterial pathogens in keeping off the oral tissues by filling the space that could have served as niche for pathogens in future and competing with the cariogenic bacteria and periodontal pathogens.[3,4]

Probiotics are generally administered through dairy products such as ice cream, cheese, yoghurt, and milk that are available easily and are considered as important vehicles for administration of probiotic bacteria. Their consumption must be acceptable to the common population and children as well.[5] However the ideal administration of probiotics is still not identified but they are accepted universally.[6]

Various mechanisms have been made for the administration of probiotics that include both conventional and non-conventional products. Some of the non-conventional probiotics are yogurts, cheese, milk, chocolates, creams, meats etc. and the conventional probiotics include beads, capsules and tablets. Although, conventional probiotics are found to be more effective as they are more characterized than other food-based carrier systems. Although, both these preparations are easily available, convenient and sold to the general public with little or no regulations.[7]

Probiotics are considered to be the economical substitutes in combating various oral diseases.[8] Although, the effect of probiotics on oral health is absolutely very less

explored till date and hence there is a need to identify the ideal administration and dosages of probiotic strains. It has also been suggested that probiotic exposure in early life may facilitate permanent installation of health promoting stains.[9]

Comparative studies of probiotic products and their benefits on the oral cavity of dental students are very less. No such studies to our knowledge have been conducted in Kanpur city. Therefore, this study was conducted to determine the effects of conventional & non-conventional probiotics in plaque reduction and gingival status of undergraduate dental students of Kanpur city, India.

Material and Methods

Study design and study population

The present study was a 2-week randomized controlled trial done on sixty undergraduate dental students.

Inclusion and exclusion criteria

Inclusion criteria was subjects with mean gingival score >1, subjects with mean plaque scores >1 and subjects with no active carious lesions or signs of periodontal disease were included in the study. Whereas, subjects who had not taken any antibiotics since last month, subjects who had not undergone any topical fluoride treatments 4 weeks prior to the study, subjects who had lactose intolerance and subjects who had used any probiotic supplements were excluded from the study.

Ethical approval and informed consent

Before the start of the study, ethical approval was obtained from the Institutional Review Board (Ethical clearance no: IEC/RDCHRC/2016-17/020). A written informed consent was obtained and information sheet was given to the enrolled participants in their spoken language.

Training and Calibration

Before the commencement of the study, the investigator was standardized and calibrated in the Department of Public Health Dentistry by Faculty members to ensure uniform interpretations and understanding.

Sample Size Estimation

Sample size was calculated using the following formula:

$$N = \frac{4pq}{L^2}$$

Where, N= is the sample size, p= Prevalence (95%), q= (1- p), L= is the permissible error in the estimation of p = 0.05

N= 4*0.95*0.05/0.05*0.05

The estimated sample size was 76 which was rounded off to a sample of 80 to accommodate dropouts. There were 20 dropouts as the participants were absent on the days of study and some students did not agreed to participate in the study. Therefore, the final sample size was 60 (15 in each group).

Data collection and analysis

Data was collected by initially selecting the subjects and dividing them into 4 groups of 15 subjects each (Figure 1). Group A was conventional probiotic group**,** Group B: Control placebo group, Group C: Non- conventional probiotic group and Group D: Control placebo group.

Fifteen subjects of Group A were given powder (Gutgain ® with strains of Saccharomyces boulardii- 5 CFU & Bacillus subtilis- 1 CFU) containing probiotics and the subjects of the control placebo group were given (Tang - Lemon). In non- conventional probiotic group, 15 subjects were allowed to administer yogurt (Mother dairy b-active with strain of Lactobacillus acidophilus) and the subjects of the control placebo group were given mother dairy plain yogurt.

Subjects consuming powder (1 sachet, 1gram once daily) were directed to mix the powder with 20 ml of water in a measuring jar using a stirrer as demonstrated. Subjects consuming Tang-Lemon (1gram once daily) were also

directed to mix the powder with 20 ml of water in a measuring jar using a stirrer as demonstrated.

In Group B, 15 participants consuming probiotic yogurt (100 grams cup once daily) were advised to include it in their daily diet. While other subjects in the control placebo group consuming plain yogurt (100 grams cup once daily) were advised to include it in their daily diet.

In all the groups participants were instructed to consume these products up till 14 days. To ensure uniformity, it was assured that all the subjects were using similar oral hygiene practices (toothbrush & toothpaste at least once daily).

Gingival and plaque index was recorded as baseline data at day 1 of the 14 days study period. And then after discontinuation of the probiotic products. The gingival index of Loe H and Sillness P[10] (1963) and plaque index of Turesky and Glickman [11] was recorded.

Blinding

To ensure blinding, following procedures were done:

1.Participants did not know that in which group they were included.

2.The principal investigator who did the recording of indices, was not aware to which group the participant belonged to.

3.The analyser did not know that to which group the participants belonged

The person knowing the codes and allotting them to groups had decoded them only after the results had been obtained.

Statistical Analysis

Data were analysed using IBM SPSS Statistics-version 21 (IBM Corp. Released 2012. IBM SPSS Statistics for Windows, Version 21.0. Armonk, NY: IBM Corp.) Descriptive statistics included calculation of means, standard deviation (S.D) and percentages. Data distribution was assessed for Normality using Shapiro-Wilk test and box-plots. Wilcoxon Signed-Rank test was used for comparison of plaque and gingival index scores for all groups, before and after the study. Mann-Whitney-U test was used for comparison of the after-study plaque and gingival index scores between the probiotic powder and probiotic yogurt groups. P-value of <0.05 was considered to be statistically significant.

Results

The present study was a 2-week randomized controlled trial done on sixty undergraduate dental students.

Table 1 shows the mean age of the study participants in all the four groups. The mean age of participants in Group A & group B was 22.8 years and 22.73 years respectively, whereas in Group C and Group D, it was 22.33 years and 23.80 years.

Table 2 shows the gender of the study participants in all the four groups. In group A and B, majority of the participants were females (86.7%) than the males (13.3%). However, in group C and D, females were more (60%) than the males (40%).

Table 3 shows the baseline and 14th day gingival scores of the study participants. There was reduction in the mean scores in group A at baseline and 14th day (From 1.078 to 0.999). This was found to statistically significant ($p<0.001$). However, no change was seen in Group B. Statistically significant differences ($p<0.001$) were observed in Group C from baseline (1.082) to 14th day (0.989).

Table 4 shows the baseline and 14th day plaque scores of the study participants. There was reduction in the mean scores in group A at baseline and 14th day (From 0.906 to 0.863). This was found to statistically significant ($p<0.001$). However, no change was seen in Group B. Statistically significant differences ($p<0.001$) were observed in Group C from baseline (1.025) to 14th day (0.900).

Table 5 shows the comparison between the conventional and non-conventional probiotic group. There were significant reductions (p= 0.244 & p= 0.604 respectively) found in the mean scores of both Group A and Group C amongst the gingival and plaque scores when compared from baseline to 14th day.

Discussion

Probiotics are living microorganisms, mainly bacteria which are considered safe for human consumption and have beneficial effects on oral and general health.[12] There are certain standards that are considered to be essential in order to accept probiotic products as legitimate for consumption. An ideal preparation of a probiotic product must have following features:

- High cell viability
- Adherence to gut epithelium to cancel the flushing effects of peristalsis
- Ability to persist in the intestine even if the strain cannot colonize the gut
- Must be of human origin
- Non-pathogenic in nature
- Resistant
- Able to influence local metabolism

The objective of administering a probiotic is to create a balance enteric microbiota that will have a major impact on the individual's health. In the present study, Saccharomyces boulardii and Lactobacillus acidophilus strains were administered.[13]

Lactobacillus acidophilus are weak lactic bacteria incapable of producing lactic acid. These grow at a temperature of 20°C and 48°C. However, the optimal growth temperature is 37°C. This strain helps in the stimulation of immune system responses, preservation of intestinal integrity during radiotherapy, increase in the availability of iron etc.[14]

Saccharomyces boulardii is a type of yeast that is used in the prevention of severe gastrointestinal illness. It is effective against the inflammatory bowel diseases and bacterial infections. In addition, this strain is anti-pathogenic, has anti-inflammatory effects on the mucosa of intestine and is anti-secretor in nature. It is also believed that when this strain is administered orally on a daily basis, the microorganisms do not reside in the gastrointestinal tract but creates a viable form in a stable level of concentration from the third day of administration.[15]

The oral ecosystem might be affected by the probiotic use specifically in the prevention of adherence by other bacteria and by modification in the protein composition of salivary pellicle. This is done by 2 methods i.e. degrading the salivary proteins and binding to.[16] The probiotics lowers the pH so that microorganisms fail to form dental plaque and calculus which predisposes to inflammation.[1] There are hypothetical mechanisms of action of probiotics in the oral cavity that includes: direct interaction of dental plaque and its formation, competitive exclusion and modulation of host immune response.[17, 18]

As we know that probiotics have number of potential benefits like reduction in the susceptibility to infections, allergies, lactose intolerance as well as lowered blood pressure and cholesterol values. According to dental literature on lactobacillus strains and Saccharomyces boulardii, there are mixed results on the oral microorganisms.[19]

The purpose of our study was to analyze the effects of conventional and non- conventional probiotic formulations on the gingival health status of undergraduate dental students.

In the present study, statistically significant reductions were found in the gingival scores on the 14th day of the participants who consumed probiotic powder (p <0.001).

Similar results were found in studies conducted by Nagashima IA [13] et al. 2013 and Yosuf F [8] et al. 2017. In addition, the study revealed a clear explanation regarding the efficacy of probiotic powder and it was also considered as the best probiotic formulation.

Studies conducted by Harini PM at al. 2010, Thakkar et al. 2013 and Nandkerny et al. 2015 showed that after using probiotic as a mouthrinse (powder form) statistically significant reduction in gingival and plaque scores was found amongst the probiotic group. These results were not in accordance with the present study as the delivery of probiotic formulations were totally different from each other. [20, 21, 22]

Karuppaiah R [1] et al. 2013 and Bhalla M [6] et al. 2015 conducted studies on probiotic yogurt where statistically significant reductions were found in the gingival and plaque scores after 30 days and 7 days of study. These results were similar to the present study conducted as significant reductions in gingival and plaque scores were found amongst the probiotic yogurt group on the 14th day. ($p<0.001$)

In the present study conducted there was no significance found in the gingival and plaque scores on the 14th day when intergroup comparisons were made. In addition, no such studies were found that showed the intergroup comparisons between probiotic powder and yogurt formulations and their efficacy levels.

To some extent valid comparisons were done between our study and other studies reported in dental literature. However, due to wider variations observed in respect to the selected participants, age groups, applied indices, strains of probiotics, vehicles and dosage of administration, the comparisons cannot be considered to be exactly valid. Although, a very sincere attempt has been made in order to compare and assess the effects of probiotics.

To conclude, irrespective of probiotic strains and vehicles used for administration; significant reductions in the gingival and plaque scores were found at the 14th day of the study amongst the probiotic powder and yogurt group when compared to the respective control groups.

Limitations

The study had following limitations as follows:

- In the present study, only short-term administration of probiotics was assessed.
- The results cannot be generalized to the other populations as the study was conducted on dental students so it was believed that they had better oral hygiene practices.

Conclusion

The results of the present study showed that statistically significant reduction was found in the gingival and plaque scores on the 14th day amongst probiotic powder and yogurt groups as compared to the respective control groups. Hence, Probiotics are useful in promoting oral health and in prevention of dental caries and periodontal diseases.

References

1. Karuppaiah RM, Shankar S, Raj SK, Ramesh K, Prakash R, Kruthika M. Evaluation of the efficacy of probiotics in plaque reduction and gingival health maintenance among school children – A Randomized Control Trial. J Int Oral Health. 2013; 5(5):33-7.
2. Brown AC, Valiere A. Probiotics and medical nutrition therapy. Nutr Clin Care. 2004; 7:56-68.
3. Dhawan R, Dhawan S. Role of probiotics on oral health: A randomized, double-blind, placebo-controlled study. J Interdiscip Dentistry .2013; 3:71-8.
4. Mishra R, Tandon S, Rathore M, Banerjee M. Antimicrobial and plaque inhibitory potential of herbal and probiotic oral rinses in children: A

randomized clinical trial. Indian J Dent Res. 2014; 25:485-92.

5. Mahantesha T, Parveen KM, Praveen NH, Asha N, Ashwin D, Vinutna B. Comparative study of probiotic ice cream and probiotic drink on salivary Streptococcus mutans levels in 6-12 years age group children. J Int Oral Health. 2015;7(9):47-50.
6. Bhalla M, Ingle NA, Kaur N, Yadav P. Mutans streptococci estimation in saliva before and after consumption of probiotic curd among school children. J Int Soc Prevent Communit Dent. 2015; 5:31-4.
7. Govender M, Choonara Y, Kumar P, du Toit L, van Vuuren S, Pillay V. A Review of the Advancements in Probiotic Delivery: Conventional vs. Non-conventional Formulations for Intestinal Flora Supplementation. AAPS PharmSciTech. 2013;15(1):29-43.
8. Yousuf A, Sidiq M, Ganta S, Nagaraj A, Vishnani P, Jan I. Effect of freeze dried powdered probiotics on gingival status and plaque inhibition: A randomized, double-blind, parallel study. Contemp Clin Dent. 2017; 8:116-21.
9. Siddiqui M, Singh C, Masih U, Chaudhry K, Hegde DY, Gojanur S. Evaluation of Streptococcus mutans levels in saliva before and after consumption of probiotic milk: A clinical study. J Int Oral Health. 2016;8(2):195-198.
10. Loe H, Silness J. Periodontal Disease in Pregnancy. Acta Odontologica Scandinavica 1963; 21(6): 533-51.
11. Turesky S, Gilmore ND, Glickman I. Reduced plaque formation by the chloromethyl analogue of vitamin C. J Periodontol. 1970; 41:41-3.
12. Shetty SM, Shah DS, Goyal G, Kathuria NS, Abraham J, Bhatia V. A study to and the status of probiotics in New Delhi, India and review of strains of bacteria used as probiotics. J Int Soc Prevent Communit Dent. 2014;4: S18-22.
13. Nagashima A, Pansiera P, Baracat M, Gómez R. Development of effervescent products, in powder and tablet form, supplemented with probiotics Lactobacillus acidophilus and Saccharomyces boulardii. Food Sci Technol, Campinas. 2013;33(4):605-11.
14. Mountzouris KC. et al. Effects of Lactobacillus acidophilus on gut microflora metabolic biomarkers in fed and fasted rats. Clinical Nutrition. 2009; 28: 318-24.
15. Martins FS. et al. Inhibition of tissue inflammation and bacterial translocation as one of the protective mechanisms of Sacharomyces boulardii against Salmonella infection in mice. Microbes and Infection. 2013; 15:270-79.
16. Polonskaya MS. Antibiotic from Acidophilus. Microbiologiya. 1952; 21:303-10.
17. George VT, Varghese MM, Vaseem MS, Thomas A, Ittycheria PG, Sreejith CK. The promising future of probiotics: A new era in periodontal therapy. J Int Oral Health. 2016;8(3):404-08.
18. Koll Klais P, Mandan R. Oral lactobacilli in Chronic periodontitis and Periodontal health: Species composition and anti-microbial activity. Oral Microbiol Immunol .2005;20;354-61.
19. Devine DA, Marsh PD. Prospects for the development of probiotics and prebiotics for oral applications. J Oral Microbiol. 2009;1.
20. Harini P, Anegundi R. Efficacy of a probiotic and chlorhexidine mouth rinses: A short-term clinical study. J Indian Soc Pedod Prev Dent. 2010;28(3):179-82.
21. Thakkar PK, Imranulla M, Naveen Kumar PG, Prashant GM, Sakeenabi B, Sushanth VH. Effect of

probiotic mouthrinse on dental plaque accumulation: A randomized controlled trial. Dent Med Res.2013; 1:7-12.

22. Nadkerny PV, Ravishankar PL, Pramod V, Agarwal LA, Bhandari S. A comparative evaluation of the efficacy of probiotic and chlorhexidine mouthrinses on clinical inflammatory parameters of gingivitis: A randomized controlled clinical study. J Indian Soc Periodontol. 2015; 19:633-9.

Tables

Table 1: Showing the mean age of the study participants

S.no	Groups	No of participants	Mean age ±SD
1.	Group A (Probiotic powder)	15	22.8 ± 1.82
2.	Group B (Non-probiotic powder)	15	22.73 ± 0.88
3.	Group C (Probiotic yogurt)	15	22.33 ± 1.44
4.	Group D (Non-probiotic yogurt)	15	23.80 ± 1.01

Table 2: Showing the gender of the study participants

S.N.	Groups	Total No. of Participants	Males		Females		Total %
			No.	%	No.	%	
1.	Group A (Probiotic powder)	15	2	13.3	13	86.7	100
2.	Group B (Non-probiotic powder)	15	2	13.3	13	86.7	100
3.	Group C (Probiotic yogurt)	15	6	40.0	9	60.0	100
4.	Group D (Non-probiotic yogurt)	15	6	40.0	9	60.0	100

Table 3: Showing the baseline and 14th day gingival scores of the study participants

Group	N	Baseline		14th day		z*	p-value**
		Mean	SD	Mean	SD		
A	15	1.078	0.199	0.999	0.125	-3.187	0.001
B	15	1.044	0.151	1.044	0.151	0.000	-
C	15	1.082	0.134	0.989	0.089	-3.416	0.001
D	15	1.133	0.280	1.131	0.281	-1.342	0.180

*Wilcoxon signed rank test, ** P-value <0.05

Table 4: Showing the baseline and 14th day plaque scores of the study participants

Group	N	Baseline		14th day		z*	p-value**
		Mean	SD	Mean	SD		
A	15	0.906	0.154	0.863	0.160	-3.420	0.001
B	15	1.011	0.14	1.011	0.14	0.000	-
C	15	1.025	0.126	0.900	0.115	-3.412	0.001
D	15	1.078	0.270	1.077	0.272	-1.000	0.317

*Wilcoxon signed rank test, ** P-value <0.05

Table 5: Showing the comparison between the conventional and non-conventional probiotic group

	Group A (Conventional probiotic group)	Group C (Non-conventional probiotic group)	z*	p-value
Gingival score (At 14th day)Mean ± SD	0.999 ± 0.125	0.989 ± 0.089	-1.165	0.244
Plaque score (At 14th day)Mean ± SD	0.863 ± 0.160	0.900 ± 0.115	-0.519	0.604

* Mann Whitney U test, ** P-value <0.05

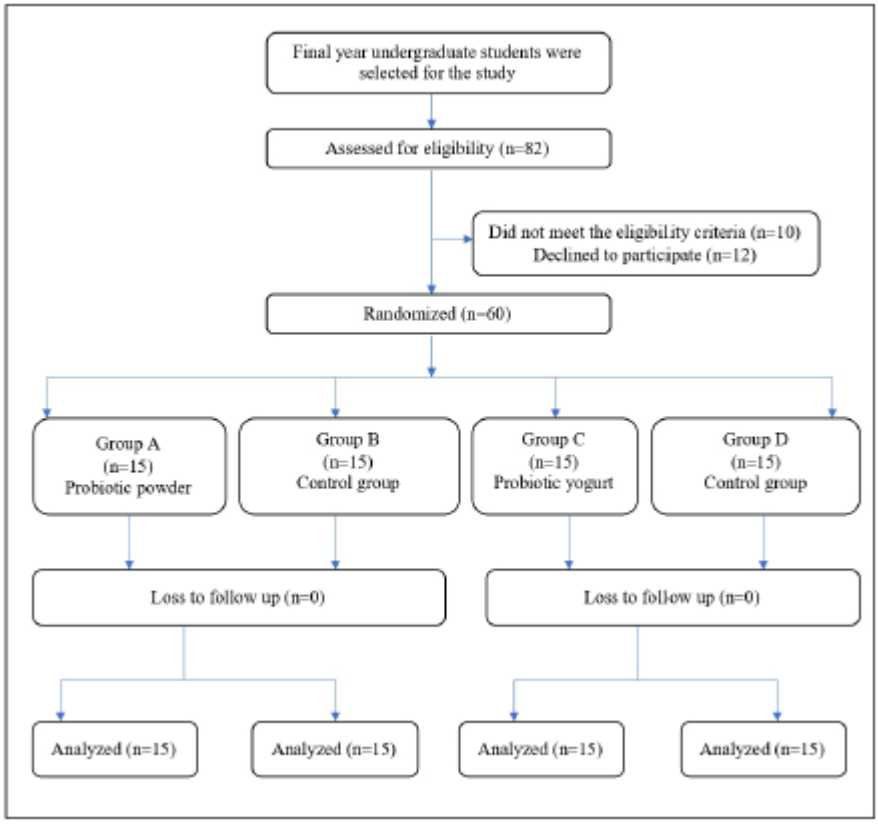

Fig 1: Flowchart of the study participants (CONSORT GUIDELINES)