after the consumption in order to make sure they did not miss the consumption schedule.

Figure 1. Shows Yakult probiotic yoghurt containing *Lactobacillus casei* Shirota.

Patients were instructed to drink the yogurt after keeping in the mouth for few minutes Patients were asked to brush twice daily for 3 min; this was shown by the operator. They were instructed to not intake any food or beverage 30 min before and after having the probiotic and avoid chewing gums, mouthwashes, and medications during the study. The yogurts were to be administered to the patients from day 1 after the first unstimulated saliva sample and continued until day 15. Saliva samples were to be again taken and evaluated at the end of 15 days. At next appointment, unstimulated saliva samples of the patient were collected in a sterile container and immediately transferred to the laboratory.

The saliva was then streaked onto *S. mutans* agar and inoculated for 1 day at 37°C in *S. mutans* agar as shown in **Figure 2** and the colony count of *S. mutans* was counted. After incubation the count of *S. mutans* colonies were counted in a digital colony counter. The counts were recorded in the colony forming units/ml (cfu/ml). Preliminary confirmation of the colonies was done by gram staining and biochemical characterization assays.

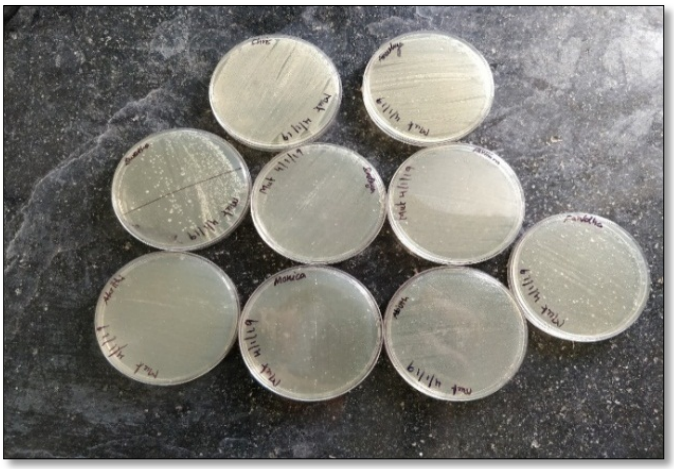

Figure 2. Shows petri dish containing saliva streaked onto *S. mutans* agar.

RESULTS

The data was processed using Statistical Package for Social Sciences (SPSS). Pre and post treatment values were compared. The following tables are those showing the paired T-test and descriptive statistics of the collected data. Significance level was established at $P<0.05$.

Table 1 show that there is positive correlation between consumption of yoghurt and reduce in *S. mutans* count. **Table 2** shows that the mean CFU of *S. mutans* reduces by about 20 CFU. There is decrease in the *S. mutans* count after consumption of yogurt containing probiotic Lactobacillus and this is evident from the data we have collected. **Table 3** represents Kurtosis descriptive statistics of the data.

Table 1. Paired samples correlations.

	N	Correlation	Sig.
Pair 1	One and Two	20	0.856	0.000

Table 2. Paired samples statistics.

		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	One	55.5500	20	44.42199	9.93306
	Two	38.7000	20	39.30997	8.78998

Table 3. Kurtosis descriptive statistics.

Descriptive Statistics							
	N	Minimum	Maximum	Mean	Std. Deviation	Kurtosis	
	Statistic	Statistic	Statistic	Statistic	Statistic	Statistic	Std. Error
One	20	6.00	130.00	55.5500	44.42199	-1.465	0.992
Two	20	3.00	116.00	38.7000	39.30997	-0.567	0.992
Valid N (list wise)	20						

DISCUSSION

In this study, the consumption of yogurt containing probiotic bacteria and the levels of *S. mutans* in the unstimulated saliva of fixed orthodontic patients was reduced to a significant level. This study was focused on patients with fixed orthodontic appliance as they have an increased risk to develop caries and white spot lesions. Brackets play a vital role in this, as they are bonded to the teeth, making it difficult for the patient to cleanse and enamel surface is etched for this bracket to be bonded. Bacteria adhere to the pathogenic plaque that adheres to the orthodontic bracket and then there is change in the oral environment and saliva in patients who are in fixed orthodontic appliances. Metallic brackets have high critical surface tension and hence and increased risk of enamel demineralization [7]. At least some of the probiotic bacteria used in various probiotic products may colonize the oral cavity during the time they are in use; thus, the effects of probiotic bacteria in the oral cavity are important to understand. Probiotic bacteria seem to affect both oral microbiota and immune responses. L.GG (*Lactobacillus rhamnosus* GG, a well-studied strain, has been shown to be able to colonise the oral cavity for at least two weeks after discontinuation of consumption of the yoghurt. Despite great efforts, our understanding of the underlying mechanisms of probiotic behavior is still inadequate, however. As mixed-species biofilms are undoubtedly the dominant form in nature and the oral cavity, there are pressing needs to discover behaviors of bacteria and yeasts in a more complex system [8]. As mixed-species biofilms are undoubtedly the dominant form in nature and the oral cavity, there are pressing needs to discover behaviors of bacteria and yeasts in a more complex system [9]. Probiotic can be provided in the form of fruit juices, prebiotic fibres, milk based food like yogurt, cheese, kefir, bio drink and in the form of supplements like powder, capsule, gelatin tablets [1]. Consumption of fruit yogurt containing *Bifidobaterium animalis* subsp. *lactis* has been studied to reduce the levels of *S mutans* in orthodontic patients with fixed appliance [4]. The positive effects of the lozenges containing same probiotic strain (*L. brevis* CD2) on gingivitis, periodontitis, and other oral lesions as well. One recent study found effective reduction in *Streptococcus mutans* concentration in orthodontic patients in response to probiotic curd and toothpaste [10]. Short-term ingestion of a *Lactobacilli brevis* derived probiotic through a lozenge tablet could reduce the levels of *S. mutans* in plaque around orthodontic brackets. Probiotic lozenges have shown to have a potential therapeutic value for the prevention of demineralization and white spot lesions formation during orthodontic treatment [1]. Probiotic strains administered for oral care are microorganisms mainly used to obtain gastrointestinal benefits, so they might not be ideal for the oral environment, quite different from the intestinal habitat. The effect of probiotics on dental caries and its related risk factors has been evaluated in several experimental studies using different strains; *Lactobacillus rhamnosus* GG, *L. casei*, *L. reuteri*, *L. plantarum*, *L. brevis* CD2, *Bifidobacterium* spp. etc. were proposed and used to obtain caries incidence reduction, mutans streptococci and lactobacilli count change, plaque pH control and root caries lesions reversal [11-15].

Caries promoting bacteria are *S. mutans*, these bacteria are both acidogenic and aciduric. Association of mutans streptococci with the initiation and prevalence of caries has been clearly established in several epidemiological studies [12-18]. The level of *Streptococcus mutans* in saliva has been shown to correlate with both past caries experience and future caries activity [17].

CONCLUSION

According to our data; the oral use of probiotic yogurt (*L. brevis* CD2) had significant effect on the *S. mutans* levels in the saliva of orthodontic patients. There was a significant decrease in the *S. mutans* count after consuming probiotic yogurt. Therefore it can be said that probiotics can be used to prevent white spot lesions and dental caries. Further studies with close follow up and long-time consumption of the probiotic can be done in a larger sample size for conclusive results and to determine its exact mechanism.

CONFLICTS OF INTEREST

There are no conflicts of interest.

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