Evaluation Study

THE EFFICACY OF TOOTHPASTE IN CONTROLLING DENTAL BACTERIAL LOADING

S.M. Tecchio[1], A. Zama Montagnana[1] and E. Isufaj[2]

[1]University of Ferrara, Ferrara, Italy
[2]Albanian University, Tirana, Albania

Correspondence to:
Evisa Isufaj, MD
Albanian University,
Tirana, Albania
e-mail: e.isufaj@au.edu.al

ABSTRACT

The aim of this study was to assess the anti-plaque effects of a toothpaste containing 0.3% triclosan, 2% copolymer, and 0.2% sodium fluoride (TCSF) in subjects with moderate plaque-induced gingivitis. A total of 10 patients with gingivitis were enrolled. None of these patients have previously been treated for periodontal disease and demonstrated radiographic evidence of bone loss. Inclusion characteristics included good general health, male and female subjects aged 18–70. Informed consent was obtained from all individual participants included in the study. Patients underwent professional oral hygiene (POH) and were instructed to use toothpaste TCSF at home twice daily for 2 weeks. Microbial analyses were performed before POH and at the end of the second week and statistically compared to the initial results. The statistic t-student-test was used to outline the statistically relevant results. All subjects completed the study. The results showed statistically significant reductions in total bacterial loading. The overall conclusion was that TCSF was a comprehensive dentifrice significantly reducing bacterial loading.

KEYWORDS: *toothpaste, oral hygiene, oral health, gingivitis, periodontal disease*

INTRODUCTION

Oral home-based hygiene is fundamental to ensure good health and prevent periodontal pathology. Poor dental hygiene can result in tooth decay, gingivitis, periodontitis, tooth loss, bad breath (halitosis), fungal infection, and gum diseases. Using a toothbrush is the most critical oral hygiene measure (1). Fair to poor oral hygiene increases the risk of periodontitis by two- to five-fold. This risk can be reduced by regular toothbrushing and dental visits (2). The most effective way to prevent dental disease is to control the production of dental plaque. Plaque is a soft, thin layer that deposits on teeth, gums, and all appliances fitted in the mouth. It is formed by microbial action. Dietary sugars, particularly sucrose, contribute to plaque formation, and their presence increases plaque formation and thickness rate. Removing plaque from the teeth and related areas is essential for maintaining a healthy mouth (3).

There are various tools that each patient can use: manual toothbrush, electric toothbrush, interdental brush, dental floss, and mouthwash. The literature has tried to demonstrate the most effective, but this does not alter the fact that the crucial point is to carry out home hygiene procedures. There is moderate quality evidence that powered toothbrushes provide a statistically significant benefit compared with manual toothbrushes regarding reducing short- and long-term plaque (4). On the other hand, using toothpaste to improve oral hygiene was relevant. The study aims to evaluate the

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effectiveness of toothpaste in reducing oral bacterial plaque. The investigated toothpaste is composed of the following components: Sodium Fluoride (NaF), Hydrated Silica, Sodium Lauryl Sulfate (SLS), Calcium Pyrophosphate and Tetrasodium Pyrophosphate, Arginine, Zinc Oxide and Zinc Citrate (5-7).

Sodium Fluoride (NaF) is primarily used to reduce the prevalence of caries and to enhance enamel remineralization (8, 9). The antibacterial and cariostatic effects of fluorides have been extensively accepted (10), and the widespread use of fluorides has been attributed to the decline of dental caries in Western countries in recent years (11).

Fluorides act primarily by forming fluorohydroxyapatite crystals that are more resistant to organic acids than hydroxyapatite crystals of tooth enamel (12). It has also been shown to reduce organic acid production in cariogenic bacteria such as Streptococcus mutans (13). The main ingredient of the silica used in abrasives is high-purity amorphous silicon dioxide, and there are varieties of different types whose properties vary depending on the production method. Since it has been an abrasive, silica is added for rubbing, grinding, or polishing. It removes substances that adhere to the surface of the teeth without scratching and enhances the natural shine. Silica is very suitable for toothpaste containing fluoride because no insoluble salt is formed when it reacts with fluoride. As its refractive index is lower than other abrasives, silica can be used to make transparent gel toothpaste.

A mixture of sodium alkyl sulfates consisting mainly of sodium dodecyl sulfate. It is a white or pale-yellow powder or crystals with a slight characteristic odor. Freely soluble in water; partly soluble in alcohol. The adverse effects of SLS have resulted in the development of toothpaste and mouthwashes with alternative surfactants such as sodium lauryl sarcosinate.

Calcium Pyrophosphate and Tetrasodium Pyrophosphate supplementation in toothpaste or mouth rinse will increase the concentration of these ions in the oral cavity. In this way, they improve remineralization and increase fluoride uptake. Pyrophosphates prevent the formation of calcium phosphate, a calcified inorganic material that makes up about 75% of the tartar, preventing its deposit on the teeth.

Arginine is a prebiotic amino acid that has been shown to affect oral biofilm ecology. Within the limitations of the in vitro study, past research concluded that the incorporation of 2% arginine in NaF toothpaste significantly enhances the antimicrobial effect against caries-generating bacteria (S. mutans) when compared to NaF (alone). In comparison, 4% and 8% arginine in NaF toothpaste were ineffective in enhancing the antimicrobial effect of NaF. The 2% Arg-NaF toothpaste might maintain better ecological homeostasis by upregulating the non-mutant streptococci (S. sanguis and S. gordonii).

Zinc is added to toothpaste and mouthwashes as zinc chloride or citrate. Zinc is a relatively non-toxic, noncumulative essential trace element. Zinc inhibits the glucose uptake pathway by Streptococcus mutans, Streptococcus sanguis, Actinomyces naeslundii, and glucose metabolism to lactic acid.

The present study aimed to assess the effects of a toothpaste containing 0.3% triclosan, 2% copolymer, and 0.2% sodium fluoride (TCSF) in subjects with moderate plaque-induced gingivitis.

MATERIAL AND METHODS

This study involved the selection of 10 healthy patients who presented gingivitis. Inclusion characteristics included good general health, male and female subjects aged 18–70. Informed consent was obtained from all individual participants included in the study. Patients underwent professional oral hygiene (POH) and were instructed to use toothpaste at home twice daily for 2 weeks. Microbial analyses were performed before POH and at the end of the second week and statistically compared to the initial results. The deepest periodontal pocket site of the oral cavity was used for microbiological analysis. The sites were isolated using cotton rolls. Sterile absorbable paper points (size 60) were used to collect subgingival samples and were immediately transferred to the microbiological lab for processing with Real-Time Polymerase Chain Reaction (PCR). *Aggregatibacter actinomycetemcomitans* (AA), *Porphyromonas gingivalis* (PG), *Tannerella forsythia* (TF), *Treponema denticola* (TD), *Fusobacterium Nucleatum* (FN), *Campylobacter rectus* (CR) and Total Bacterial Loading (CBT) were evaluated.

Real-time polymerase chain reaction (PCR)

Probes oligonucleotides were designed based on 16S rRNA gene sequences of the Human Oral Microbiome Database (HOMD 16S rRNA RefSeq Version 10.1), counting 845 entries. All the sequences were aligned to find either consensus sequences or less conservative spots. Two real-time polymerase chain reaction (PCR) runs were performed for each sample. The first reaction quantified the total amount of bacteria using two degenerate primers and a single probe matching a highly conservated 16S ribosomal RNA gene sequence. The second reaction detected and quantified all selected bacteria in two multiplex PCR. This reaction included twelve primers and six particular probes for each species.

Oligonucleotide concentrations and PCR conditions were optimized to ensure sensitivity, specificity, and no inhibitions in case of unbalanced target amounts. Absolute quantification assays were performed using the Applied Biosystems 7500 Sequence Detection System. The amplification profile was initiated by a 10 min incubation period at 95°C to activate polymerase, followed by a two-step amplification of 15 s at 95°C and 60 s at 57°C for 40 cycles. All these experiments, including nontemplate controls, were performed to exclude contamination of reagents.

Plasmids containing synthetic DNA target sequences (Eurofins MWG Operon, Ebersberg Germany) were standard for the quantitative analysis. Standard curves for each target were constructed in two triplex reactions using a mix of the same plasmids in serial dilutions ranging from 101 to 107 copies. There was a linear relationship between the threshold cycle values plotted against the copy number log over the entire range of dilutions (data not shown). The copy numbers for individual plasmid preparations were estimated using the Thermo NanoDrop spectrophotometer.

The absolute quantification of total bacterial genome copies in samples allowed for calculating the relative number of red complex species. Plasmid purification and handling were performed in a separate laboratory with dedicated pipettes to prevent contamination of samples and polymerase chain reactions. The SPSS program and paired simple statistic T-test were used for the statistical analysis to detect statistically significant differences.

RESULTS

Both clinical and microbiological parameters showed improvements. After 15 days of toothpaste, TCSF microbiological analysis significantly reduced total bacterial loading (Table I).

Table I. *Paired sample test.*

		pairwise differences					t	Df	Sig. (2-code)
		Mean	Standard deviation	Mean Error	95% confidence interval for the difference				
					inferior	superior			
Couple 1	AA1-AA2	401639.66667	1253679.07356	417893.02452	-562023.37595	1365302.70928	.961	9	.365
Couple 2	PG1-PG2	230.10000	924.99147	292.50798	-431.59903	891.79903	.787	9	.452
Couple 3	TF1-TF2	1622099.80000	2812431.86976	889369.04725	-389792.76060	3633992.36060	1.824	9	.101
Couple 4	TD1-TD2	1231941.50000	1526348.88168	482673.89702	140057.28657	2323825.71343	2.552	9	.031
Couple 5	FN1-FN2	5417714.70000	4361539.98644	1379240.04631	2297656.95003	8537772.44997	3.928	9	.003
Couple 6	CR1-CR1	20809.60000	25403.34994	8033.24460	2637.13819	38982.06181	2.590	9	.029
Couple 2	TBL1-TBL2	34725019.60000	37150995.83389	11748176.41785	8148798.16654	61301241.03346	2.956	9	.01

DISCUSSION

Dental plaque is a major etiological factor in the causation of dental caries and plaque-induced gingival diseases. Mechanical removal using toothbrushes, toothpaste, and mouth rinses helps check pathogenic plaque build-up, thereby preventing these diseases. Effective and therapeutic plaque control is an essential aspect of personal hygiene. Appropriate toothpaste has been documented as an effective tool among plaque control measures.

The positive results obtained in this study cannot be completely related to the introduction of TCSF toothpaste during the treatment since a comparison with subjects treated with toothpaste only was not performed. Nevertheless, in the case of acute pathologies, such as gingivitis, the results provisionally demonstrate the most significant effect of using TCSF toothpaste as an adjuvant treatment is primarily related to its ability to reduce bacterial loading post-treatment. TCSF toothpaste has been shown to reduce plaque and gingivitis. Most studies on TCSF toothpaste are related to periodontitis, a highly prevalent, chronic, non-specific, and immunologically devastating disease of periodontal tissues caused by microbial infection.

It may be speculated that TCSF toothpaste's ability to promote antibacterial activity most likely contributes to an overall improvement in the patient's oral hygiene.

CONCLUSIONS

In our opinion, the result of this clinical trial is very promising with regards to the benefits of using TCSF toothpaste as an adjuvant in the standard treatment of gingivitis to reduce bacterial loading, contributing to an overall improvement in the oral health of the patient. Further studies with a larger sample size are required to confirm the results obtained.

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