



Objective

To determine the remineralization, abrasivity, biofilm regrowth, and oxidation of salivary biomolecules of two ClōSYS[®] dentifrices containing stabilized chlorine dioxide. The efficacy of ClōSYS[®] dentifrices is compared with some leading commercial products. This study evaluates effectiveness of ClōSYS[®] dentifrices in achieving good oral health.

Introduction

Oral health can be improved through the research into the link between oral health and several systemic diseases.¹ Research has shown that fluoride dentifrices contribute to a decrease in plaque and caries. However, different fluoride containing dentifrices lead to varying incidences of caries in the animal model.² Further, the performance of a given dentifrice cannot be explained solely by its fluoride content alone. Rather, the concerted and synergistic effect of the ingredients in a particular formulation on the dentin and surrounding oral microbiome.³ As oral biofilms and dental plaque regrow rapidly after a professional cleaning and debridement, the selection of a dentifrice for twice-daily home oral care, together with proper tooth brushing techniques, is one of the most crucial component for maintaining a healthy dentition and oral health. Ability to adequately cleaning the pellicle, remove biofilm, enhanced uptake of fluoride for remineralization of the enamel, reduced biofilm regrowth, lower abrasion, and oxidation of biomolecules contributing to oral malodor are some criteria of an effective dentifrice.

Methods

Materials: ClōSYS[®] Silver toothpaste contains sodium fluoride (0.21%), stabilized chlorine dioxide, and sodium myristoyl sarcosinate. ClōSYS[®] Anticavity toothpaste contains sodium fluoride (0.24%), stabilized chlorine dioxide, and sodium lauroyl sarcosinate. Other toothpastes were procured commercially.

Enamel fluoride uptake and remineralization: Artificial lesions were formed in the bovine enamel specimens by a 33-hour immersion into a solution of 0.1 M lactic acid and 0.2% Carbopol C907 which was 50% saturated with hydroxyapatite and adjusted to pH 5.0. The lesion surface hardness range was 25-45 Vickers micro-hardness (VHN; 200 gF, 15s dwell time) and average lesion depth of 70µm. The cyclic treatment regimen consisted of a 4 hours/day acid challenge in the lesion forming solution with four, one-minute dentifrice treatment periods. Then specimens were rinsed with distilled water and placed in human saliva for the remaining time (~ 20 hours). Surface Micro Hardness (SMH) assessments were conducted after 10-days and 20-days.

Abrasivity: Relative abrasion level of dentifrices to dentin (RDA) was determined following standard procedure.

Oxidation of biomolecules: Aliquots (0.60 ml) of healthy human salivary supernatants (n = 10) were treated with equivalent volumes of the toothpaste extract supernatants and then equilibrated for 30 and 60 seconds at 37°C. These mixtures were then subjected to ¹H NMR analysis (500 MHz). Corresponding experiments were also performed with buffered (pH 7.00) chemical model systems (CMSs) containing the KSBs pyruvate and L-methionine (20.0 mM).

Inhibition of the biofilm regrowth: Biofilm was grown for 24 hours on 4x4 mm bovine enamel sections in 12-well tissue culture plates using 3 ml of Brain Heart Infusion broth supplemented with Yeast Extract and Vitamin K and hemin (BHI-YE) inoculated with 50 µl of an overnight culture of a mixed species whole salivary bacterial preparation. The enamel sections were brushed with Toothpaste (3 sections/paste) for a brushing schedule similar to a 30 second brushing by human subjects, rinsed with sterile water and inserted into a fresh tissue culture plate containing 3 ml of BHI-YE to facilitate regrowth of the remaining oral biofilm on the enamel sections for 6, 12, and 24 hours. The plates were stained with a live/dead stain (BacLight Bacterial Viability Stain containing Syto9 and Propidium iodide) for confocal microscopy.

Results

Abrasivity

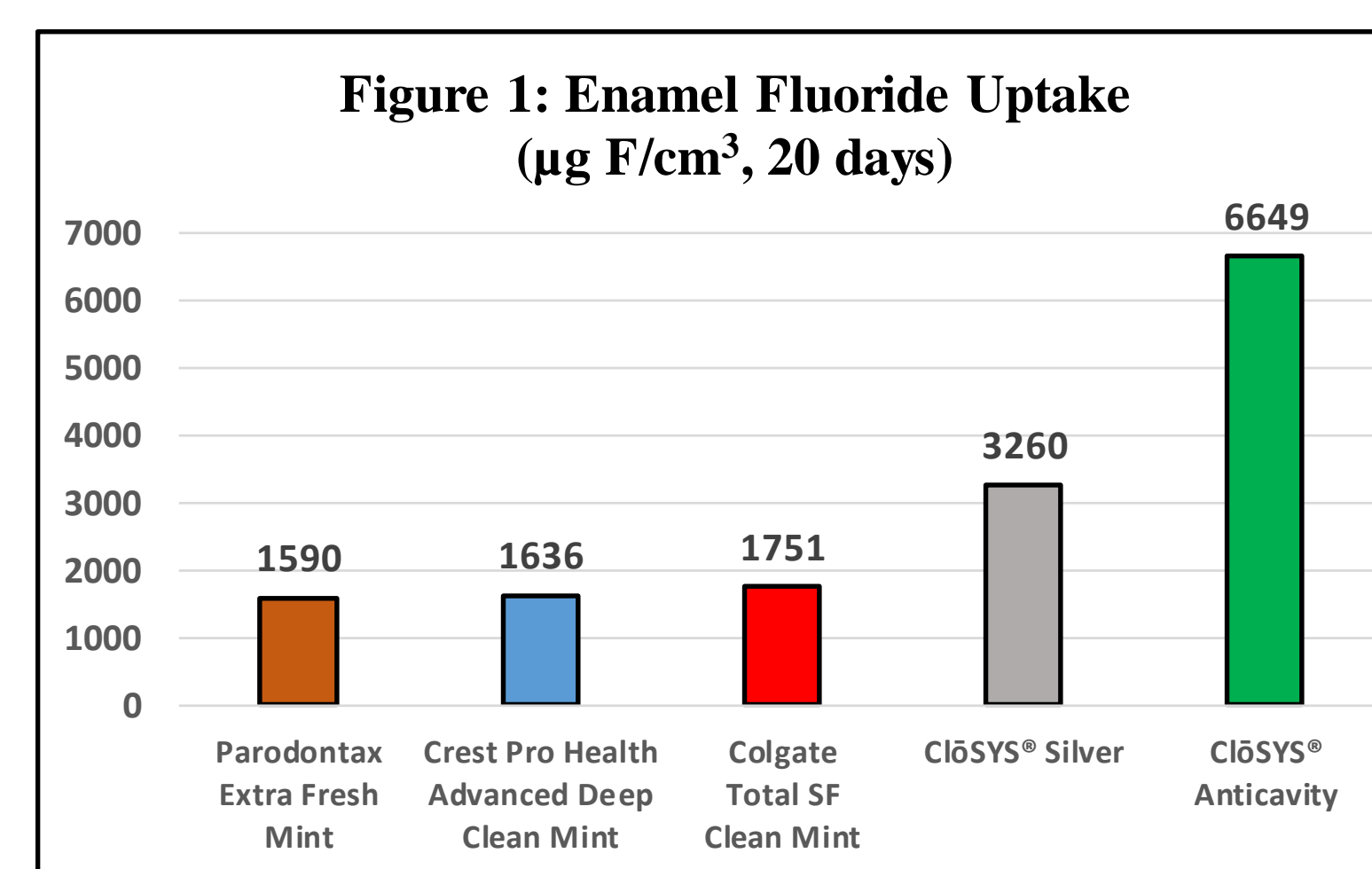
Table 1: Abrasivity of ClōSYS[®] Toothpastes

Toothpaste (n = 8)	RDA Value ±SEM
ClōSYS [®] Anticavity Toothpaste	207.55 ± 4.86
ClōSYS [®] Silver Toothpaste	142.03 ± 2.50

The RDA value of Silver toothpaste was 65% lower; than Anticavity toothpaste. However, both toothpastes contain same quantity of silica, an abrasive agent. Lower RDA for Silver toothpaste is attributed to the synergy within the other ingredients. RDA value of both ClōSYS[®] toothpastes was below the 250 ceiling recommended by ANSI/ADA.

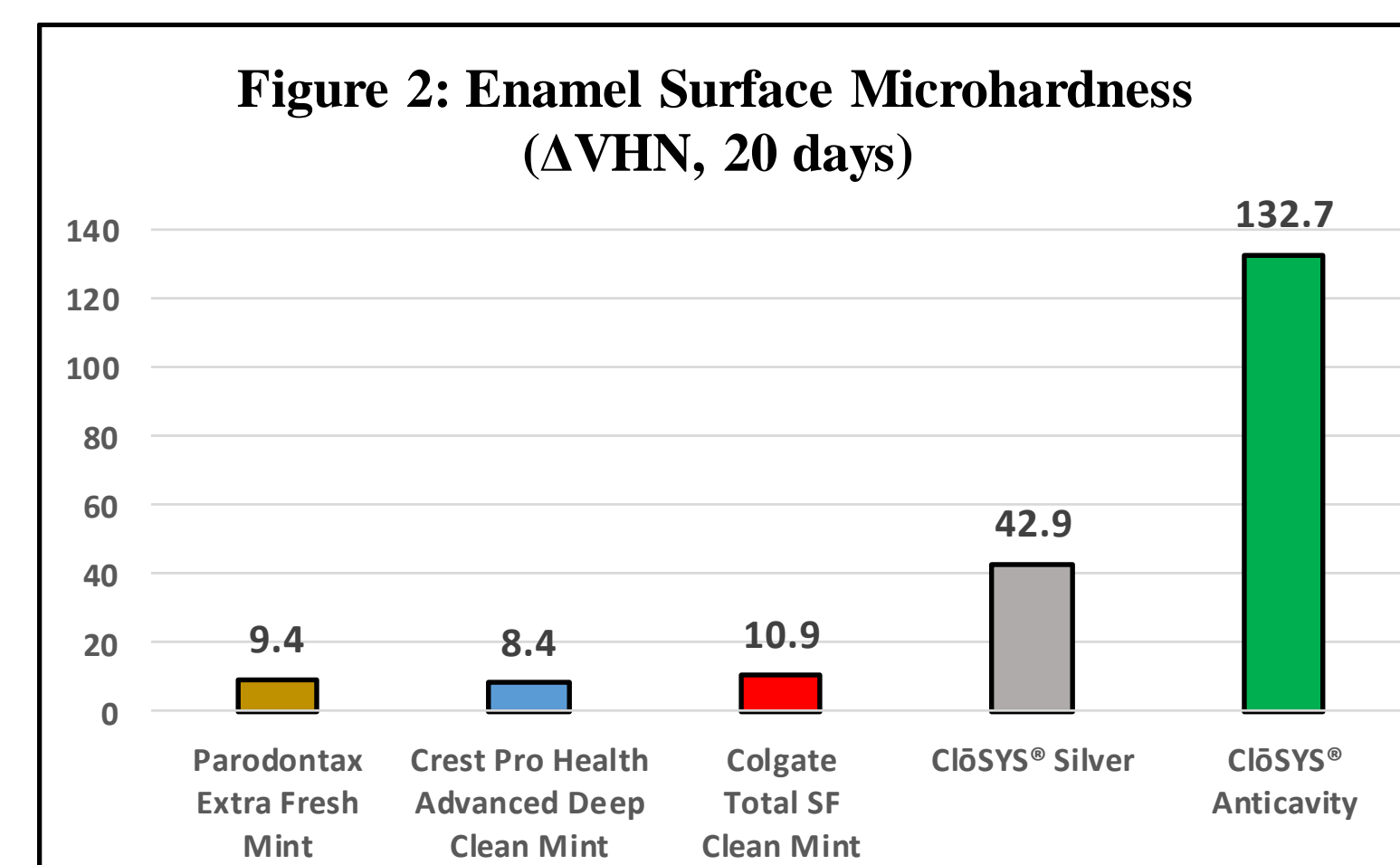


Fluoride Uptake



Fluoride uptake into incipient lesioned enamel after 20 days of treatment by ClōSYS[®] Silver and ClōSYS[®] Anticavity toothpastes was **86 - 100% and 270 - 300% higher** than Parodontax Extra Fresh Mint, Crest Pro Health Advanced Deep Clean Mint, and Colgate Total SF Clean Mint toothpastes, respectively.

Remineralization



The remineralization effect of ClōSYS[®] Silver and ClōSYS[®] Anticavity toothpastes was **4- to 5-fold and 12- to 15-fold greater** than Parodontax Extra Fresh Mint, Crest Pro Health Advanced Deep Clean Mint, and Colgate Total SF Clean Mint toothpastes, respectively, as determined by SMH after 20-days.

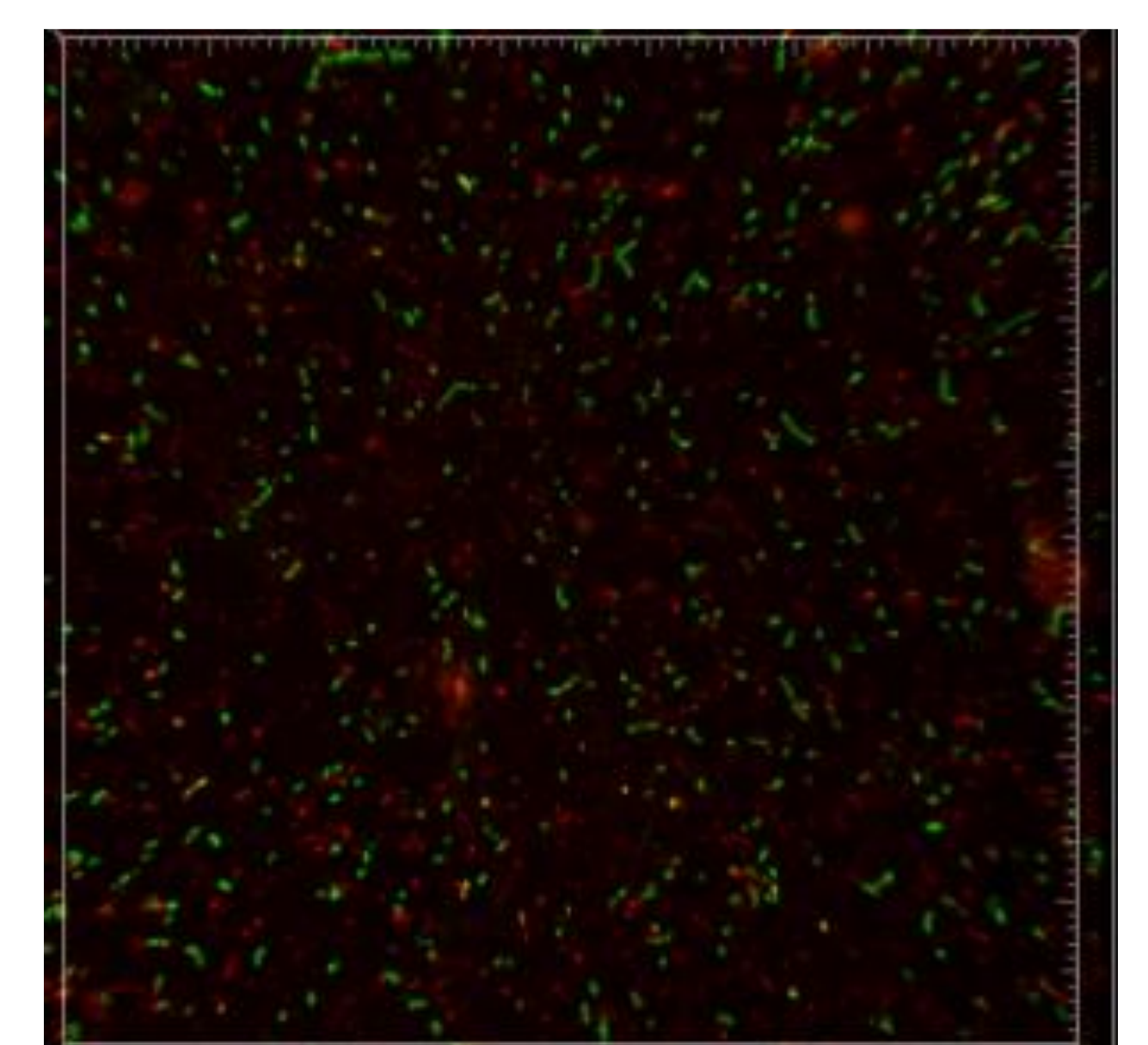
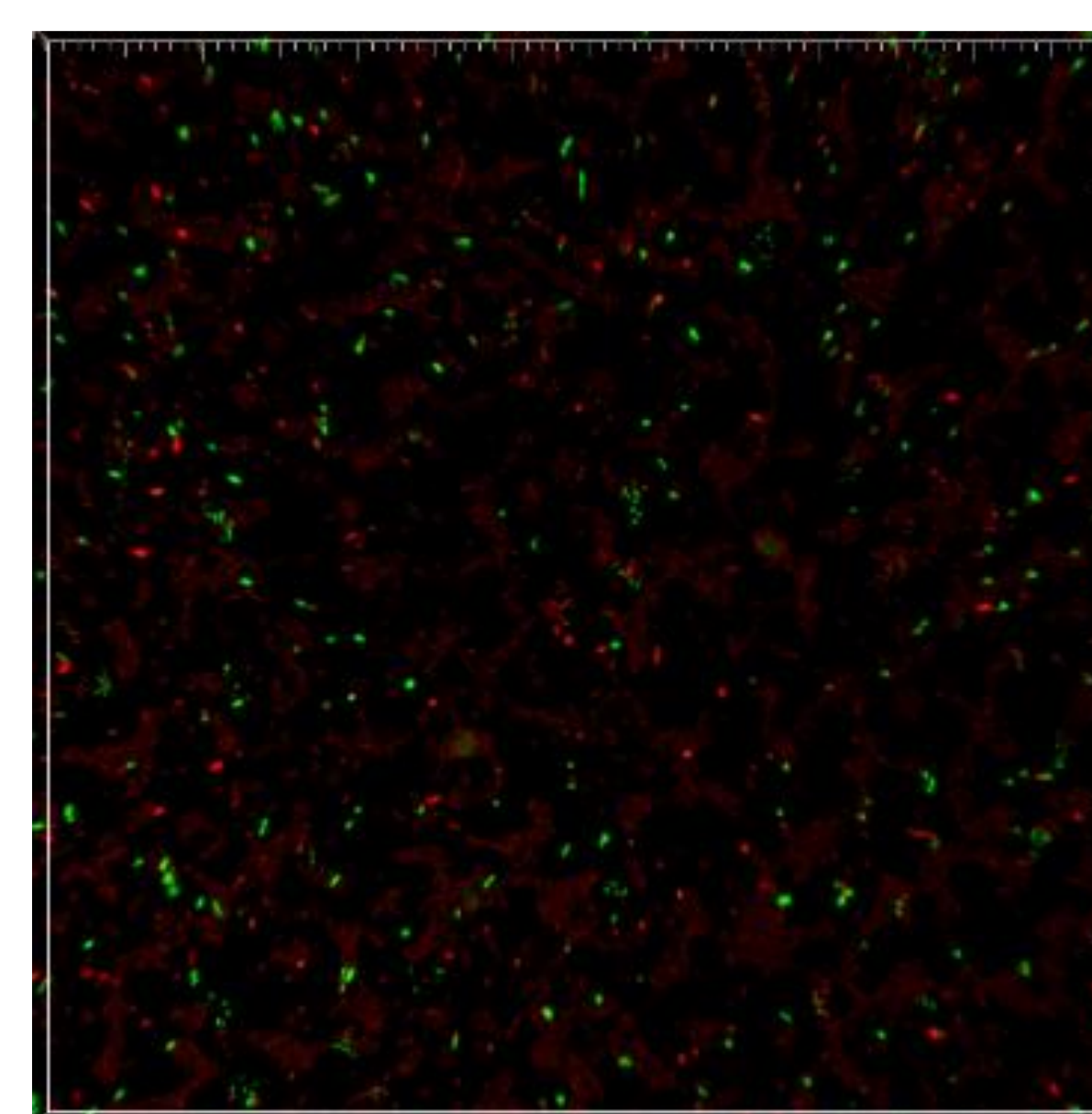
Oxidation of Salivary Biomolecules

Table 2: Oxidation of Salivary Biomolecules (¹NMR Analysis)

Time	Pyruvate [Acetate] : [Pyruvate]	L-Methionine [Methionine Sulfoxide] : [Methionine]
0 Seconds	0.15 x 10 ⁻³	1.69 x 10 ⁻³
30 Seconds	9.57 x 10 ⁻³	13.0 x 10 ⁻³
60 Seconds	11.3 x 10 ⁻³	14.0 x 10 ⁻³

ClōSYS[®] Anticavity toothpaste effectively oxidized salivary biomolecules within 30 seconds of its contact with saliva i.e. soon after administration in the oral cavity. L-methionine represents precursors for generating compounds leading to oral malodor. Pyruvate represents organic acids that result in acidic mouth.

Biofilm Regrowth



ClōSYS Silver Toothpaste Brushed Sample

Water Brushed Sample

Figure 3: Confocal microscopy images after 6 hours of brushing.

ClōSYS[®] Silver toothpaste significantly (p = 0.0388) inhibited the mass of the live bacterial biofilm (green stained) 6 hours after the biofilm was brushed when compared to the water brushed specimens. Images in Silver toothpaste brushed specimen exhibited lesser number of live cells (green stained) compared to water brushed sample, demonstrating the reduced regrowth of polymicrobial biofilm as a result of effective removal of biofilm during brushing (Figure 3).

Conclusions

- ClōSYS[®] Anticavity and ClōSYS[®] Silver toothpastes achieve much higher enamel fluoride uptake and greater remineralization compared to leading brands containing stannous fluoride.
- ClōSYS[®] Anticavity toothpaste efficiently oxidize harmful salivary biomolecules within 30 seconds of its administration.
- ClōSYS[®] Silver toothpaste effectively removed the dental biofilm thereby reducing its regrowth.
- Abrasivity of ClōSYS[®] Anticavity and ClōSYS[®] Silver toothpastes was below the limits recommended by ANSI/ADA.
- **ClōSYS[®] Anticavity and ClōSYS[®] Silver toothpastes are multifunctional dentifrices achieving superior oral health.**

Financial Disclosure

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