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Research Article

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Inhibition and Removal of Calculus Deposits in a 3-Month Randomized Clinical Trial

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Abstract

Aim: *To evaluate the safety and efficacy of ToothWave™ a novel RF-utilizing toothbrush for inhibition and removal of calculus deposits.*

Methods: *This was a single-blind, double arm prospective study. Subjects were randomized to one of two study groups, receiving either ToothWave™ or a control sonic vibrating toothbrush (Philips Sonicare 4100 Protective Clean), and performed twice-daily brushing during a test period of 12 weeks. Calculus was assessed at Baseline and again after 3, 6 and 12 weeks using the Volpe-Manhold Index (V-MI). Results were compared within each group between the different visits and between the groups; percent change from baseline was also compared between the groups. The differences between groups were compared using Mann Whitney non-parametric model. Differences within each group over time were compared using Friedman's test followed by Dunn's test.*

Results: *A total of 87 subjects (42 in the test group and 45 in the control) completed the study, having fully evaluable data. At Baseline, the groups did not differ significantly in the efficacy measurement mean scores. While the control group exhibited a statistically significant increase in calculus levels ($p \leq 0.001$) the test group maintained the original baseline levels and exhibited a slight negative percent change in the VMI score as compared to baseline (not significant).*
Both toothbrushes were well-tolerated and no device related adverse events were reported during the study.

Conclusions: *The ToothWave™ RF-utilizing toothbrush demonstrated statistically significant calculus reductions when compared to a powered control toothbrush while maintaining the initial calculus levels and preventing the additional calculus accumulations.*

Keywords: Calculus, Powered, Radiofrequency, Toothbrush, Toothbrush

Introduction

It is well recognized that dental calculus originates from plaque through a process which involves bonding of the plaque with saliva salts to form the mineralized deposits [1-4]. The first step of dental plaque formation is the adherence of proteins and organic substance on the teeth surface, which forms a thin biofilm, coating the teeth, gums and tongue, referred to as pellicle. The dental pellicle is composed of a variety of salivary glycoproteins, crevicular fluid, bacterial and host tissue cells, and is formed and held together by charged electrostatic forces in addition to Van der Waals and hydrophobic forces [1,2]. Specifically, phosphorene residues in the charged regions of salivary proteins are believed to facilitate the electrostatic interactions between the proteins and the teeth surface during the pellicle adhesion process [1]. Once formed, the pellicle alters the charge of the tooth surface, which in turn increases the efficiency of bacterial adhesion [1,2].

The dental plaque is a gel-like material forming on the teeth pellicle, and is composed of microorganisms and an inter-microbial matrix [1-3] Amongst the microorganisms composing the plaque there are bacterial and non-bacterial species (yeasts, viruses, etc.), whereas the inter-microbial matrix includes host cells (epithelial, macrophages, leucocytes, etc.), organic compounds (polysaccharides, proteins, glycoproteins, lipids), and inorganic compounds (calcium, phosphorus, fluoride, sodium, and potassium). Once the mineral content increases, the plaque mass becomes mineralized to form calculus [1-3].

Dental calculus is mostly inorganic, consisting primarily of calcium and phosphorus, with minor components of carbonate, sodium, magnesium, silicon, iron, and fluoride, and minerals like hydroxyapatite, whitlockite, octacalcium phosphate, and brushite [1,2]. There is also an organic component to calculus (approximately 15-20%) consisting of carbohydrates, protein, and lipids [1,2].

Calcium and phosphate are two salivary ions that are "raw materials" for dental calculus formation. Theoretical research indicates that high concentrations of calcium phosphate salts in the saliva (supersaturation) is the driving force for dental plaque mineralization [1,3,4]. Calcium phosphate represents a family of materials and minerals containing calcium cations (Ca^{2+}) together with inorganic phosphate anions (PO_4^{3-}, $H_2PO_4^{-}$, and/or HPO_4^{2-}). Once the calcium-phosphate complex is formed, apatite deposition follows when sufficient calcium and phosphate ions are present in the environment [1,3,4]. One of the most abundant structure composing the dental calculus is hydroxyapatite, which is composed of calcium cations (Ca^{2+}) that are electrostatically bound to phosphate ions (PO_4^-) to form the hydroxyapatite crystal [$Ca_{10}(PO_4)_6(OH)_2$]. This structure is held together via electrostatic forces, where the positively charged calcium ion interacts with negatively charged phosphate and hydroxyl (OH^-) ions [1,3,4]. The addition of mineral salts to the plaque structure results in a hardened material that is very hard to break using brushing forces [5].

Calculus accumulation is known as one of the major causes of gingival inflammation [6,7,8]. This rough and hardened substance provides an ideal surface for further plaque formation, which leads to calculus build-up and impairs gingival health. The understanding that dental plaque and calculus are key etiological agents in the initiation and progression of gingival inflammation has been the basis of vast dental scientific and industrial research, aiming to find the optimal daily dental hygiene procedure [9-12]. Currently available scientific publications discuss the efficacy of various powered toothbrushes in reducing plaque, gingival inflammation, and gingival bleeding, following several weeks of twice-daily brushing at home [13-15]. However, none of the published research studies shows reduction of calculus by a standard powered toothbrush. It is widely accepted that once the calculus is formed, it is firmly attached to the tooth surface and is too hard to be removed with a regular toothbrush; thus, in the conventional way, calculus build-up must be removed with ultrasonic tools or dental hand instruments (such as a dental scaler) [5].

With an aim to provide efficient reduction of dental plaque and calculus at home, and without changing the daily dental hygiene routine, Home Skinovations LTD. (Yokneam, Israel) has developed the ToothWave™ powered toothbrush. It is a novel toothbrush intended to remove effectively the impurities that are strongly attached to the tooth surface, such as stains and calculus, and thus to promote the reduction of bleeding and gingival inflammation. ToothWave™ utilizes low-power RF energy that streams between two electrodes over a silicon barrier and reaches the tooth surface during brushing (Figure 1). RF is an alternating electric current that oscillates at radio frequencies in the range of 3kHz-300GHz. It has been used in medicine for several decades for many different applications, from surgical to aesthetic, providing various effects, depending on the specific parameters of the device in use [16]. Specifically, the ToothWave™ RF technology is proposed to bring charged molecules that originate from the toothpaste to the tooth-surface to destabilize the electrostatic bonds that strongly hold together the hard impurities that are attached to the tooth (calculus, stains). In the current clinical study, the RF technology utilized by the ToothWave™ was evaluated for patient safety and efficacy for calculus reduction and inhibition compared to a standard powered toothbrush (control brush).

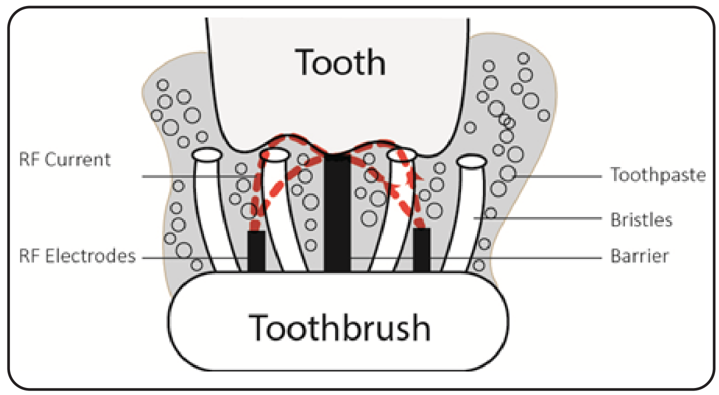

Figure 1: Schematic illustration of RF current on tooth surface

Methods

A randomized single-blind, double arm prospective study was conducted in order to evaluate the safety and efficiency of the RF-utilizing toothbrush (ToothWave™, Manufactured by Home Skinovations LTD, Yokneam, Israel) in reduction and prevention of calculus accumulation, as compared to an ADA accepted reference powered toothbrush (Philips Sonicare 4100 ProtectiveClean). The protocol and consent form were approved by the U.S.IRB (IRB#: U.S.IRB2020SRI/03), Miami, FL, USA, before study initiation, and verbal and written consent were obtained from all subjects.

Participants

Screened subjects received an oral soft tissue (OST) examination for assessment of their eligibility. Evaluation of the calculus levels during the study was performed using the Volpe-Manhold Index (V-MI) [17]. Recruited subjects were 18 years of age and older, having a minimum of 16 natural teeth, including six mandibular anterior teeth with no crowns or veneers. All subjects demonstrated a propensity for calculus formation as evidenced by at least 9 mm of calculus on the lingual surfaces of the six mandibular teeth following the 2-month run-in phase.

Subjects who had a medical condition requiring antibiotic premedication prior to dental procedures, regular users of a chlorhexidine mouth rinse, or subjects with any oral condition or pathogenesis that could interfere with study compliance or examination procedures were excluded. Subjects with current or history of oral cavity cancer or oropharyngeal cancer, pacemaker or internal defibrillator, or any other active electrical implant anywhere in the body, pregnant or nursing subjects, or subjects that do not brush regularly were also excluded.

Study Procedures

Eligible subjects underwent a dental prophylaxis, to render the lower anterior teeth calculus free, and entered a two-month run-in phase by the end of which a V-MI calculus examination was performed. Subjects who formed at least 9 mm of calculus on the lingual surface of the mandibular anterior teeth and were eligible to continue participation according to the eligibility criteria were randomly assigned to either the ToothWave™ or the control brush (Philips Sonicare 4100 Protective Clean) group. Subjects were stratified according to calculus levels, age, gender, and ethnic group. The study test phase included a total of 168 treatment sessions and 5 clinic visits over a period of 12 weeks. Participating subjects were instructed to brush twice daily (morning and evening), unsupervised, in a regular manner, during a three-month test period using a standard fluoride toothpaste (Crest® Cavity Protection Cool Mint Gel, 0.243% Sodium Fluoride). Subjects returned to the clinic at weeks 3, 6 and 12 for safety (OST) and V-MI examinations, and additionally at week 9, during which a supervised brushing was conducted. Supervised brushing sessions were performed also at Baseline and week 3 in order to verify brushing was conducted according to the instructions. Participants recorded each brushing in a treatment diary that was provided to them, and was checked at each visit to assess compliance.

Assessments

For clinical efficacy, the supragingival calculus present on the lingual surfaces of six mandibular anterior teeth was evaluated according to the V-MI[17] at visits 3, 4 and 6 (following 3, 6 and 12 weeks of brushing, respectively).

After drying the teeth with a stream of air and using a standard periodontal probe graduated in millimeters, the examiner placed the instrument on the most inferior border of the visible calculus, and measurements were obtained on the following three planes:

1. Bisecting the center of the lingual surface;
2. Diagonally through the mesial-incisal point angle of the tooth through the area of greatest calculus height; and
3. Diagonally through the distal point angle of the tooth through the area of the greatest calculus height. The examiner assigned a score to each measurement plane, with measurements made in 0.5 mm increments starting at 0.5. A score of zero (0) denoted that there was no calculus present at a measurable site. The V-MI was calculated for each subject by summing the millimetre scores over all sites graded.

Safety

For safety, a thorough evaluation of the oral soft tissues was conducted at each visit, by way of a visual examination of the oral cavity, including the Lips/Labial Mucosa, Bucal Mucosa, Mucobuccal Fold, Sublingual Mucosa, Gingiva (free and attached), Tongue, Hard/Soft Palate, Uvula/Oropharynx, Teeth and Dental Restorations. A trained dental evaluator performed intraoral examinations during the study visits as required according to the study protocol. In case an adverse event occurred (AE) it was recorded and monitored throughout the study. Any observed abnormalities noted during the OST examination were transcribed beginning at the screening visit until 5 days after the final use of study product. The investigator determined the causal relationship of each AE using their clinical experience and selected the appropriate severity descriptor as mild, moderate, or severe. Treatment-emergent AEs were reported for the safety population, which included all randomized participants who received study product.

Data Analysis

A sufficient number of participants were to be screened in order to randomize at least 90 participants (approximately 45 to the Test, and 45 to the Control groups) to ensure 84 evaluable participants completed the entire study. The sample size in this study provided 80% power to detect a significant difference in the score improvements with type 1 error of 5%. Safety analyses were carried out on a modified intent-to-treat (ITT) population, defined as all randomized participants who conducted at least one treatment. Efficacy analysis was conducted on the per-protocol (PP) populaiton, defined as subjects who had full evaluable data and conducted all treatment sessions according to the study protocol.

Summary statistics (e.g., count, mean & SD, minimum and maximum) of the demographic characteristics and the efficacy measurements were calculated for each group and study visit.

Categorical variables (race & gender) were presented in contingency table and compared using Fisher exact test. Normality distribution of measures was evaluated using Shapiro & Wilk test; as the majority of measures deviate from normal distribution, non-parametric approach was implemented. To evaluate the improvement after 3, 6, and 12 weeks compared to baseline, the difference was calculated as follows:

$$Delta = Score_{Weeki} - Score_{Baseline}$$

The percent of change from baseline was calculated as follows:

$$Percent\ change = \frac{(Score_{Week_i} - Score_{Baseline})x100}{Score_{Baseline}}$$

The differences between groups were compared using the Mann Whitney test. Differences within group over time were compared using Friedman's test followed by Dunn's test. A logistic regression model was applied to identify factors that are related to improvements in calculus scores. Significance level was defined as α=0.05. Analyses were carried out using SPSS version 25.0.

Results

A total of 107 subjects provided informed consent and were screened for potential enrollment in the study. Ninety (90) of these subjects were enrolled to the study, all of them met the entrance criteria and were randomized at baseline to receive either ToothWave™ or the control powered toothbrush. One subject in the control group discontinued study participation prior to study end, and 2 subjects in the test group were not included in the per-protocol population due to incomplete follow-up. Eighty-seven (87) subjects (96.67%) completed and deemed fully evaluable at the trial's conclusion (Figure 2).

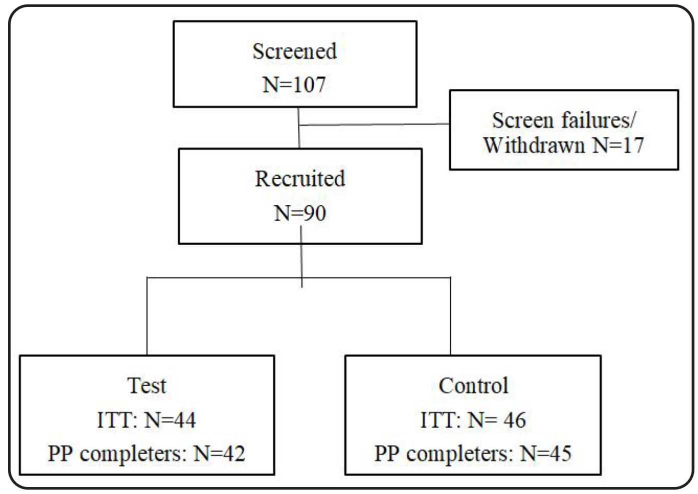

Figure 2: Study flowchart

The baseline demographic characteristics are exemplified in Table 1, indicating that the study population was well-balanced with respect to all baseline demographic variables (p≥0.171).

Table 1: Baseline Demographic Characteristics

	Characteristic	Test N=42	Control N=45	Overall N=90	P value
Age	Mean age (SD)	52.5 (13. 8)	52.5 (13.1)	52.5 (13.3)	0.782
	Age range	21-76	23-74	21-76	
Gender	Male (%)	14 (33.3%)	15 (33.3%)	29 (33.3%)	1.000
	Female (%)	28 (66.7%)	30 (66.7%)	58 (66.7%)	
Ethnicity	Caucasian (%)	38 (90.5%)	36 (80%)	74 (85.1%)	0.171*
	Other (%)	4 (33.3%)	9 (20%)	13 (14.9%)	
*Representing significance level comparing the distribution of Caucasian and other ethnic populations between the study groups.					

Efficacy

The average V-MI scores at baseline, 3, 6 and 12 weeks are shown in Tables 3, indicating a statistically significant increase in calculus levels in the control group during 12 weeks of brushing, while no significant differences were found in the calculus scores of the test group over time (p=0.793). In addition, no statistically significant differences were found between the groups in the V-MI baseline scores (p=0.737).

Figure 3 exemplifies the percent changes from baseline in V-MI scores following 3, 6 and 12 weeks of brushing. Positive percent changes in the control group represent the significant increase in calculus levels, while negative percent changes in the test group represent calculus reduction (not significant). The percent changes in the test group at 6 and 12 weeks were found to be statistically significantly different when compared to those of the control (*p =0.040, **p=0.014).

The logistic regression results are shown in table 3, verifying that there is no significant influence of subject age, gender or baseline V-MI scores on the improvement rate in calculus levels. However, the study group (test vs control) had a significant impact on improvement rate, where subjects in the test group had 3.4 times more chance to improve during 12 weeks, when compared to the control (Table 3).

Safety

Both toothbrushes were well-tolerated and no device-related adverse events or any side effects were reported during the study. There were no serious AEs, no medical devise incidents, and no participants with AEs that led to discontinuation of treatment or withdrawal from the study.

Table 2: Endpoints of Baseline, 3, 6, and 12 weeks of VMI

Group	Time (SD)	Mean	95%CI	Median	[P25, P75]¥	P-value
Control§	Baseline	13.922 (4.013)	[12.717, 15.128]	13	[11, 16]	<0.001*
	3 Weeks	14.589 (3.822)	[13.441, 15.737]	13.5	[12, 17]	
	6 Weeks	14.822 (4.162)	[13.572, 16.073]	14	[11.5, 17]	
	12 Weeks	15.311 (4.397)	[13.990, 16.118]	14.5	[12, 17.5]	
Test£	Baseline	14.524 (4.891)	[13.003, 16.118]	13.5	[10.5, 17]	0.793
	3 Weeks	14.651 (5.808)	[12.767, 16.423]	14	[11, 17.5]	
	6 Weeks	14.419 (5.884)	[12.450, 16.121]	13.5	[9.5, 18.5]	
	12 Weeks	14.593 (5.967)	[12.570, 16.263]	13.5	[9.5, 18.5]	
N£=42, N§=45						
*Representing significant increase in calculus levels over time, each of the endpoints in the control group was found to be statistically significantly different when compared to baseline. ¥representing percentiles 25 and 75, respectively.						

Table 3: Logistic regression for Improvement in VMI following 12 Weeks

Measure	P-value	OR	95% CI for OR
Test vs Control	0.009*	3.430	[1.366, 8.613]
Subject age	0.337	1.017	[0.982, 1.054]
Female vs Male	0.611	0.772	[0.285, 2.089]
Baseline-VMI	0.792	1.014	[0.914, 1.125]
* Representing a significant impact of the study group on the improvement rate in calculus levels.			

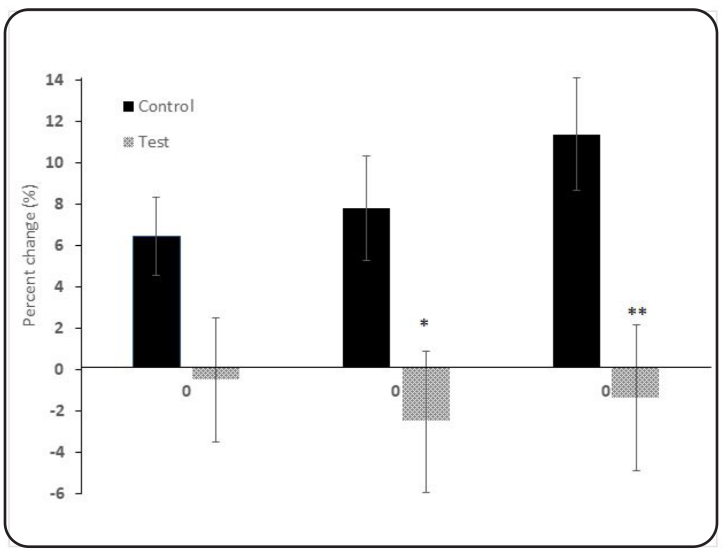

Figure 3: Percent change from baseline in V-MI scores following 3, 6 and 12 weeks of brushing

Statistically significant differences were found between the test and control groups at 6 and 12 weeks (* p =0.040, ** p=0.014).

Discussion

Calculus build-up on the tooth surface includes an initial stage of pellicle and plaque formation and a final mineralization (calcification) process. This process relies on the addition of minerals to the soft plaque material, via electrostatic interactions, to form the final multicomponent structure of calculus [1]. The electrostatic interactions involved in this process facilitate the formation of the hardened material and its strong attachment to the teeth surface, and the final calculus structure is held together by these electrostatic forces [1,2].

The calculus attached to the teeth surface, can rarely be removed by regular brushing, either with a manual or powered toothbrush; thus, while brushing with standard toothbrushes, calculus is gradually accumulated over time, and is removed periodically by mechanical dental scaling [5]. As expected, the control group in our study exhibited statistically significant calculus accumulation over the course of 12 weeks. On the other hand, ToothWave™ was shown to prevent calculus accumulation as the Baseline calculus levels were maintained during the study test phase, exhibiting significantly greater percent reduction in V-MI scores when compared to the control. Logistic regression analysis conducted on the study results demonstrated that the change in calculus level is strongly affected by the test group (test vs control), moreover, other parameters such as subject age, gender, and Baseline V-MI score had no significant effect on the results. Additionally, the Baseline calculus levels (after the 2-month pre-trial) of each subject represented the specific calculus accumulation rate and was expected that the Baseline calculus levels would be correlated with the ability of

ToothWave™ to reduce calculus and affect its buildup rate. The fact that no such correlation was seen indicates that there might be a stronger influencing factor such as the calculus composition, which varies between subjects on the ability of ToothWave™ to affect the calculus buildup process and reduce calculus levels [1].

The results in this study are in agreement with the findings of a previous comparative single-blind 6-weeks study, on the effect of calculus, plaque and gingival inflammation by ToothWave™ [18]. In a very similar manner to the current study, these previous results showed that the RF test group demonstrated a statistically significant reduction in calculus levels as compared to the control. Two additional comparative single-blind, 6-weeks studies tested the effect of ToothWave™ on teeth shade and dental stains, showing a significantly greater extrinsic dental stain reduction and improvement in tooth whitening as compared to a control ADA-accepted powered toothbrush [19]. These studies further support the claims that the RF energy in ToothWave that streams on the teeth surface during brushing is capable of removing hard substances and surface stains that are strongly attached to it.

Apart from ToothWave, there is another type of toothbrush which relies on electric current to achieve its effect ("electronic toothbrushes" or "ionic toothbrushes"), and is the closest in technology and mechanism of action to ToothWave™, but with several key technological differences. First, the ionic toothbrushes produce a low-level direct electrical current that streams from the brush head into the oral cavity, and secondly, they use a power source (battery or solar) and a metal rod conductor [20-27]. The scientific data available on these ionic toothbrushes is highly inconsistent, as some in-vitro and clinical studies indicate their beneficial effect on plaque and gingival inflammation, while others conclude that the performance of an ionic toothbrush does not differ from that of a conventional power toothbrush [20-24,26-28].

When the ToothWave™ effect is compared with that of ionic toothbrushes, the differences can be explained by the technological variances between the two toothbrush types. The ionic toothbrush utilizes a direct electrical current (DC), which runs from the brush into the oral cavity through the body and arm, back to the brush-handle [22]. Instead, Toothwave™ utilizes RF energy (dentalRF), which is an alternating electrical current (AC) that streams back and forth between two electrodes, providing a localized effect that is limited to the tooth surface. The high frequency of the alternating current that is set by the RF parameters allows to safely increase the electrical power as opposed to DC current, and thus achieve significant results [29]. Therefore, the difference in the effect of ToothWave™ when compared to ionic toothbrushes, results from the type of current that is utilized (alternating vs direct) and its intensity. Moreover, the RF current tends to flow along the surfaces of electrical conductors, what is known as the "skin effect", and thus directing the current towards the tooth surface [30]. The electro-mechanical silicon barrier, which is located between the ToothWave™ electrodes additionally contributes to its increased efficacy. Furthermore, when compared to a standard power toothbrush, the electric current is theorized to reach hard-to-reach areas (i.e. between teeth) as these areas and surfaces would otherwise be chronically missed using traditional mechanical means (i.e. bristles).

Despite their technological differences, both ToothWave and the ionic toothbrushes share the same mechanism of action, which is based on the principle of polarity that every element in nature has a positive or negative charge and attach to other charged substances or form complex structures via electrostatic bonds [28]. The ionic toothbrushes induce an electric charge, which is postulated to damage electrostatic bonding of plaque proteins to tooth surfaces; thus, enhancing plaque removal [23]. Similarly, since the RF alternating current streams close to the tooth, it brings the charged molecules that are present in the toothpaste close to the tooth surface and changes the chemical environment around it. Once these molecules accumulate near the tooth surface the chemical balance is shifted towards the removal of compounds that are electrostatically attached, replacing them by other, charged substances, which might have greater affinity to the tooth surface (for instance fluoride). In our suggested mechanism of action, the electrically charged toothpaste ingredients take part in the process that occurs on the tooth surfaces. Toothpastes are water-based complex mixtures of abrasives and surfactants, humectants, binders, and other active ingredients, and as such contain charged molecular compounds that once the RF is activated, act as electrolytes in the medium, carry the charges along the tooth surface, and achieve the desired effect. However, since the technology of the sonic vibrating TB is one of the most popular power toothbrushes amongst the commercially available toothbrushes, we chose to use the Philips Sonicare 4100 ProtectiveClean toothbrush as our control rather than an ionic toothbrush.

Conclusions

The present clinical trial demonstrated that The ToothWave™ RF-utilizing toothbrush provided statistically significant calculus reductions when compared to a powered control toothbrush (Philips Sonicare 4100) while maintaining the initial calculus levels and preventing the additional calculus accumulations. The oral health improvements from this human clinical study provides positive evidence to support the RF mechanism as a beneficial feature that is uniquely utilized by the ToothWave™ power toothbrush.

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