

Colgate® 360^o™

Whole Mouth Clean

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Biology of the Oral Mucosa

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The oral mucosa is composed of an outermost layer of stratified squamous epithelium. It serves both as a protective barrier for the body against environmental insults including microorganisms, viruses, toxins, antigens, and chemicals as well as an interactive interface with the environment. The epithelium of the oral mucosa is continuously being shed. Without efficient clearance, the exfoliated cells will accumulate and form a biofilm on oral surfaces. Putrefaction of the biofilm may contribute to undesirable oral conditions.

Epithelial Turnover

The dynamic nature of the oral cavity subjects the oral mucosa to friction, compression, and stretching during various oral functions, such as talking, chewing, and swallowing. Considering the surface location of epithelial tissues, they are exposed to a wide variety of environmental insults ranging from mechanical, thermal, and microbial to chemical and toxic. Therefore, the epithelial cells are vulnerable to damage, which, consequently, mandates continuous cellular replacement. Under normal physiologic conditions, the oral epithelium undergoes frequent cellular renewal (Figure 1). Cell division in the oral epithelium is five times faster than it is in the skin. Although the turnover rate of an epithelial cell is measured by days, the shedding of the superficial keratinized layer occurs in a matter of hours. The turnover time for the buccal epithelium has been estimated at 5 to 6 days, and this is probably representative of the oral mucosa as a whole.¹ This rapid epithelial turnover represents an important mucosal defense mechanism.

The oral microflora contains a broad range of microorganisms that cannot survive in the mouth without attaching to a surface. However, studies have shown that the multiplication of bacteria attached to oral epithelial cells does not proceed beyond one cell division. Therefore, rapid turnover of the surface layer is probably important in limiting and preventing microbial colonization and invasion of the mucosa by pathogenic microorganisms and opportunistic infections. Clearance of the exfoliated cells, microorganisms, and other debris is facilitated by the flushing system, which is provided by saliva; accumulation of about 2 mL of saliva triggers a swallowing reflex. However, this flushing system may not be sufficient without mechanical removal of the biofilm and employing meticulous oral hygiene. It has been estimated that there are approximately 100 bacteria attached to the epithelium per epithelial cell, and this number doubles with time.² Without efficient clearance, these exfoliated cells and bacteria will accumulate in the mouth and form a biofilm on the oral tissues, teeth, and mucosal surfaces. Such a microbial load becomes a risk factor for oral diseases, and the degradation by-products of this biofilm become a source for the bacterial production of volatile substances that can result in oral malodor. A direct relationship has been observed between the thickness of the biofilm on the oral mucosa and oral malodor.³

Oral Malodor

The oral cavity—mainly the tongue and, to a lesser extent, the gingivae—is the principal source of oral malodor. The most intense oral malodor

is exhibited after prolonged periods of reduced salivary flow and abstinence from food and drinks. Oral malodor is mainly composed of volatile sulfur compounds (VSC), such as hydrogen sulfide, methyl mercaptan, and dimethyl sulfide.⁴ Various other compounds contributing to oral malodor include volatile organic acids (such as butyric acid and propionic acid) and amines (such as cadaverine, indole, and skatole).^{5,6} These components are derived from the proteolytic breakdown by oral bacteria of peptides and amino acids derived from various salivary origins. One key source of food for oral bacteria is the desquamated epithelial cells from the cheeks and lips.⁷

Mechanical cleaning of the teeth and soft tissues is probably the most effective measure for removing the biofilm and controlling the number of oral microorganisms, including those associated with the formation of plaque, gingivitis, and oral malodor.

Dental plaque biofilm, which is composed of desquamated epithelial cells from the oral mucosa, salivary components, and microorganisms, has a strong potential for oral malodor formation.^{8,9} Tonzetich and Kestenbaum⁹ showed that incubating whole saliva or saliva sediment that contained desquamated epithelial cells produced oral malodor, whereas centrifuged salivary supernatant did not generate oral malodor. These studies indicate that desquamated epithelial cells play a key role in the production of oral malodor. Several approaches have been pursued to control oral malodor and other oral conditions. However, none of the approaches involved a means of actively reducing or removing desquamated epithelial cells from the oral mucosa. Yonezawa et al¹⁰ found a direct relationship between oral mucosal cleaning and the production of VSC in the mouth. Under normal physiologic conditions, chewing and swallowing provide a mechanism for continuous removal of loose epithelial cells, bacteria, and food debris. However, microorganisms of the biofilm are more resistant to dislodging without mechanical manipulation. Although antiseptic oral products may improve oral malodor,¹¹ they are ineffective in eliminating the cellular components of the biofilm. It is well accepted that employing diligent oral hygiene is

effective in controlling physiologic oral malodor.⁴ Therefore, mechanical cleaning of the teeth and soft tissues is probably the most effective measure for removing the biofilm and controlling the number of oral microorganisms, including those associated with the formation of plaque, gingivitis, and oral malodor.

Benefits of Oral Hygiene Practice

There is a strong correlation between poor oral hygiene and the incidence of chronic inflammatory disease and oral malodor.^{7,12-14} Consumer education and awareness has created a demand for dental products that can provide multiple benefits without significantly increasing the effort to achieve those benefits. The Colgate-Palmolive Company has developed an innovatively designed manual toothbrush that actively removes desquamated epithelial cells and microorganisms from the teeth, tongue, buccal mucosa, and lips. This toothbrush has tightly packed and tapered bristles for subgingival and interdental cleaning, a raised cleaning tip for reaching hard-to-reach places in the mouth, and soft dental-like polishing cups for the delicate removal of tooth stains. Additionally, a special feature of the toothbrush is the soft conically shaped nubs on the back of the toothbrush head for the gentle and soft cleansing of the oral mucosa lining of the tongue, cheeks, and lips.

This multifaceted toothbrush has been evaluated in several clinical studies for its ability to remove desquamated epithelial cells from the cheeks and lips, cleanse hydrogen-sulfide-form-

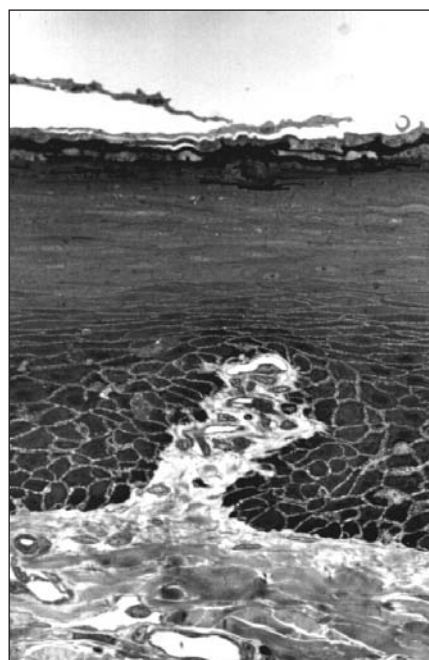


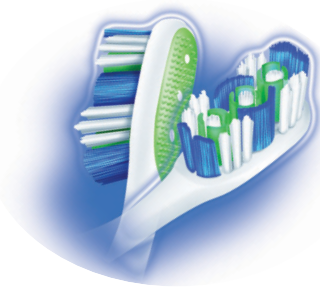
Figure 1—Light micrographs of oral mucosa depicting cells shedding from the surface. (Photo courtesy of Dr. Philius Garant, SUNY at Stony Brook, Stony Brook, New York.)

ing bacteria from the tongue, reduce VSC associated with oral malodor, decrease plaque and gingivitis, and reduce surface stain on the teeth. Compared to two commercially available manual toothbrushes and a commercially available battery-powered toothbrush, the new manual toothbrush was statistically significantly better at reducing the level of desquamated epithelial cells. Concomitantly, hydrogen-sulfide-forming bacteria on the tongue, breath VSC, plaque, gingivitis, and tooth staining also were significantly reduced after using the multifaceted toothbrush vs the comparative toothbrushes. Details of these results are presented throughout this supplement.

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Driving Toothbrush Innovation Through a Cross-functional Development Team



Abstract: A cross-functional team was assembled to take on the project of developing a toothbrush that could deliver a “whole mouth clean.” As such, this brush needed to not only demonstrate superior plaque removal from teeth, it also needed to provide a means for safe cleaning of soft oral tissues. During use, the brush needed to be comfortable both in hand and in mouth, and it required contemporary esthetics to help drive consumer acceptance. This article will take the reader through the full development cycle of the new toothbrush, identifying the proposed benefit of each brush feature along the way. Where appropriate, care was taken to identify the specific insights leading to the development of several new features not previously seen in commercial toothbrushes. This article also will attempt to demonstrate the value of forming new product development teams with representatives of multiple functional areas, including marketing, product development, advanced technology, clinical, engineering, and manufacturing.

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The first phase of the project entailed a thorough review of existing toothbrush design information. Previous research by Mintel and Crawford¹ has indicated that regardless of the design of toothbrush people use, their brushing technique will remain basically unchanged and sorely inadequate. The task, therefore, was to design an effective plaque-removing toothbrush with a bristle configuration that could adapt to any brushing style. In vitro testing of brush designs addressing this need has shown a clear benefit to alternating the heights of bristle tufts. These alternate heights allow taller tufts to act independently, uninfluenced by adjacent bristles during brushing. Once independent motion is achieved, the longer bristles can effectively reach further between the teeth.

Early consumer research showed that when people were given the choice between flat-tipped brushes and the same brush with raised bristle tufts at the tip, consumers preferred designs with raised tips. Consumers perceived that this configuration would offer improved plaque removal in hard-to-reach areas, such as behind the posterior teeth and the lingual tooth surfaces.

Part of the development team's initial research included an evaluation of the tools used by dental professionals. Of particular interest to the team was the “prophy cup,” noted for its cleaning and polishing performance in prophylaxis treatment (Figure 1). The team observed that one of the key characteristics that make the prophy cup an effective tooth-surface cleaning tool is its ability to hold the paste against the tooth surface to maximize the paste's performance. In the team's quest for improved tooth-surface cleaning, an exciting opportunity was realized: explore design configurations that mimic the “paste holding” characteristics of the prophy cup.

Initial Research: Preferred Handle Configurations

A number of studies¹⁻³ have been conducted examining toothbrushing time and motion. While handle design does not have a major impact on the



Figure 1—Typical prophy cup, noted for its paste-holding characteristics.

clinical efficacy of the toothbrush, it does impact rather significantly on the comfort and resulting compliance of the brushing experience. The angled offset design and the offset design (Figure 2) have excellent ergonomics by maintaining the point-of-bristle contact in line with the longitudinal axis of the handle during toothbrushing. This prevents the handle from becoming unstable as forces are applied to the head.

Initial Research: Soft Tissue Cleaning

The next area requiring research was soft tissue cleaning. Cleaning the gingival margin has been a subject of previous toothbrush design research by Volpenhein and Hartman.⁴ Toothbrush features that address the cleaning of this area include extended tufts of bristles along the sides of the brush head. Seeking to broaden the efficacy of this product, the team also researched the state of the art of cleaning other major oral soft tissues—the tongue and the cheeks.

Rosenberg and McCulloch⁵ reported on the benefits of tongue cleaning in the control of bad breath. The primary methods of tongue cleaning used by the general population include tongue scraping and tongue brushing. The function of both is to mechanically remove the biofilm that develops on the surface of the tongue. Research by Yaegaki and Sanada⁶ supported findings that the tongue coating is comprised of desquamated epithelial cells, blood cells, and bacteria. A very interesting hypothesis was put forth that removing these epithelial cells from the mouth by cleaning the cheeks could help minimize the number of dead cells available to settle on the tongue, thus minimizing the food source for a host of odor-causing bacteria.

Initial Research: Materials and Manufacturing

In addition to reviewing the hygienic and

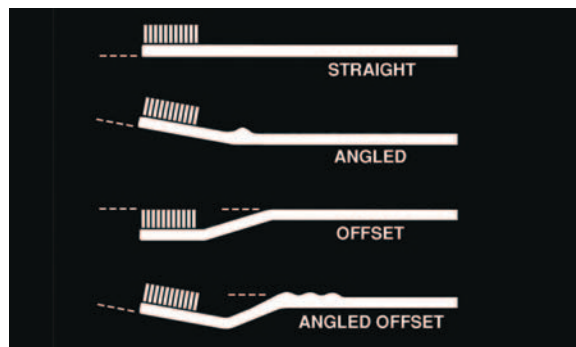


Figure 2—Four basic types of toothbrush handles.

ergonomic aspects of toothbrush design, the team also conducted a review of the latest material and manufacturing trends to be sure that it was armed with the latest opportunities these critical areas had to offer. Of particular interest were “anchorless” manufacturing technologies that could allow unique bristle-tuft shapes, unlike conventional brush-making technology, which, for all practical purposes, is limited to uniformly sized bristle tufts. Another benefit of anchorless technology is its ability to produce thinner brush heads, thereby potentially offering a lower-profile brush head for improved oral access. A material trend noted by the development team was the availability of soft, yet highly durable elastomer, which could be molded into nearly any shape for novel toothbrush-cleaning features. These synthetic materials did not contain latex, and they had been approved for many medical applications.

Initial Research: Summary

Armed with the general knowledge and insights gained through the initial research stage, a design brief was generated with specific guidelines as follows:

Brush Head

1. Explore cleaning features based on the functionality of dental prophy cups for improved broad-surface cleaning and surface-stain removal.
2. Include taller outer tufts of bristles for gingival-margin and interproximal cleaning.
3. Include a raised tip for improved cleaning behind posterior teeth as well as the lingual surfaces of teeth.
4. Explore soft tissue cleaning features (tongue and cheeks).
5. Explore all of the above with the expanded design opportunities offered by state-of-the-art toothbrush manufacturing processes and materials.

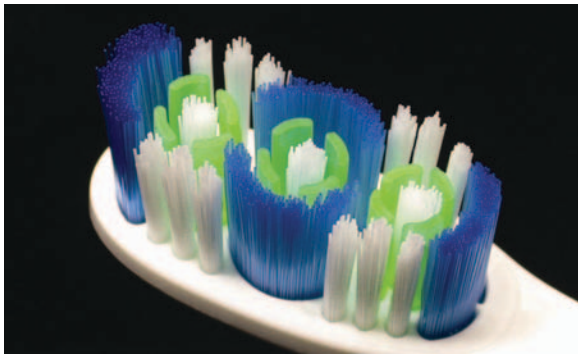


Figure 3—Close-up of three polishing cups at the center of the toothbrush head.

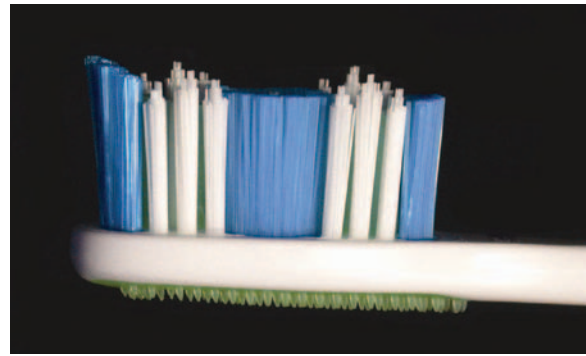


Figure 4—Side view of the pointed outer tufts.

Handle

1. Working within the context of either an offset head or an angled offset head, develop a handle that is comfortable to use. Comfortable is defined as offering ease of maneuverability and control with minimal effort.
2. The design must be esthetically attractive, considering current design trends both within the toothbrush category and beyond.

The focus of this article is, as it should be, on the development of the efficacious aspects of this toothbrush. It is, however, worth a brief discussion about one of the less tangible aspects of toothbrush design—product esthetics. This subject has drawn a fair amount of attention in recent years. Vanderbilt⁷ quotes the Norwegian Design Council regarding the Jordan Multi Action toothbrush: “its soft contemporary form and topical colors turn what is usually regarded as an annoying duty into a pleasure.” Product esthetics play a large role in consumer perception: Is it a modern design? Is it an object I *want* to use? While on the surface these perceptions may seem to belong solely in the “commercial” domain, these factors also must be acknowledged for their ability to foster patient compliance. To this end, the development team sought the assistance of an external industrial design partner. A particular interest was expressed for a European influence, so a review of potential European design partners ensued. A firm based in Milan convinced the development team they had the ability to digest the team’s insights and experiences and help deliver a leading-edge product.

Design Execution: Fulfilling the Brief

Working closely with Colgate’s experts, a winning combination of ideas began to emerge. In an effort to evaluate the function of prophy cups, a design emerged with three cups in the center of the bristle field. These cups went through a series

of evolutions to achieve the benefits of a prophy cup, while being comfortable to use. The winning combination—developed through a series of three-dimensional computer-aided design models, physical models, and working prototypes—was a soft elastomer ring surrounding a round tuft of bristles (Figure 3). It was hypothesized that this ring would help to retain dentifrice in and around these central tufts, making them more effective at broad tooth-surface cleaning and surface-stain removal, similar to its inspiration, the prophy cup. It was discovered during the prototype stage that the comfort of the configuration was significantly increased by slitting these rings into four arcs, providing greater flexibility to adapt to non-planar tooth surfaces and improving the manufacturability of the design. A further prophy-cup-inspired enhancement was added to the central cup. A pair of long, arc-shaped tufts of bristles was added to help surround the center cup to aid in maintaining the presence of dentifrice.

Previous research has indicated that regardless of the design of toothbrush people use, their brushing technique will remain basically unchanged and solely inadequate. The task, therefore, was to design an effective plaque-removing toothbrush with a bristle configuration that could adapt to any brushing style.

These long, continuous walls of bristles would be achieved easily with the aforementioned anchorless tufting technology. Another key benefit of this new brush-making technology was its ability to consistently deliver high-quality bristle-end



Figure 5—Close-up of the tongue and cheek cleaning texture on the back of the toothbrush head.

rounding. End-rounded bristles are essential to toothbrush safety with regard to gum abrasion.⁸ Conventional brush manufacturing requires features to be configured to allow sharp-edged bristles to be ground and polished as a secondary operation, which would have imposed many restrictions on design options. It would have been difficult, for example, to achieve a tall elastomer feature next to a shorter tuft of bristles, as the taller rubber feature would interfere with the polishing of the shorter bristles.

Another set of features that emerged were the tall, thin, pointed outer tufts of bristles (Figure 4). It was hypothesized that pointing these tufts would enhance their ability to reach into tight interproximal spaces and along the gingival margin. This was in addition to the previously mentioned insight of variable-height tufts to allow freedom of movement from the restriction of nearby bristles. A raised pair of bristle tufts was added to the tip as discussed, to aid cleaning behind posterior teeth.

Several intriguing ideas emerged in the attempt to address soft tissue cleaning. The decision to explore cleaning textures on the back of the brush head was made very early on in the development of the brush. Many features were considered for this purpose, with the leading option being a low-profile, bristle-like, textured soft tissue cleaner. Because the back of the brush head comes in regular contact with much of the cheek and inner lip surfaces during normal brushing, the team was looking for a cleaning surface that would be gentle yet effective on these sensitive tissues. The team hypothesized that the short bristle-like “nubs” of elastomer could be configured to have extremely thin tips, which also would provide a good cleaning effect on the irregular surfaces of the tongue (Figure 5).

Many handle designs were explored. One handle feature that quickly gained favor was the



Figure 6—Textured gel-like thumb grip.

design proposal for the thumb grip (Figure 6). The proposal was to create a large, soft, compressible gel-like structure that would adapt its shape to the thumb and forefinger under the pressure encountered during normal brushing. These general assumptions were proven, and the design optimized, through a series of working prototypes (Figure 7).

Consumer Research

A considerable amount of time and effort was needed to identify and optimize the features discussed for this toothbrush. Working closely with the design consultants, the development team was able to blend all of these disparate features into a cohesive multibenefit product. The design effort was paralleled by an extensive amount of consumer research. This research either validated the design proposals at that given time or revealed the need for further refinement. Once the design was fully validated in qualitative testing, a preliminary manufacturing evaluation was undertaken. While manufacturing was involved throughout the development process, many capability assumptions that were made during the development stages needed to be validated in a more production-like environment.

Safety Testing

With confidence gained in the ability to manufacture the new toothbrush design, a significant amount of physical-endurance testing needed to be conducted to ensure that this novel toothbrush would be safe to use. This testing included both static and dynamic testing to evaluate neck and head strength, bristle/tuft retention strength, and bristle end-rounding quality. All of the base materials also were tested and approved for safe use. In addition to laboratory testing, which sets pass/fail criteria based on time-tested safety factors, an “extended home-use” test also was con-

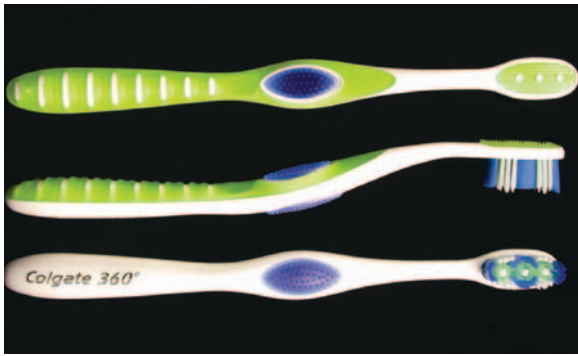


Figure 7—The new Colgate® 360™ toothbrush.

ducted. This test monitored the actual normal home use of the product by hundreds of potential users for the recommended lifetime of the brush, which is 3 months. After completion of the test, the brushes were returned for a detailed quality examination along with a survey of the user's perceptions of quality.

Clinical Validation

The next milestone would be the most critical of all. Once manufacturing validation was completed, a series of clinical studies were conducted to see if the product would fulfill its clinical expectations. Throughout the following articles of this supplement, we will report the results of in-depth clinical evaluations of the Colgate® 360™ toothbrush^a, including plaque removal, gingivitis reduction, stain removal, mouth odor, tongue bacteria, and an epithelial MTT assay.

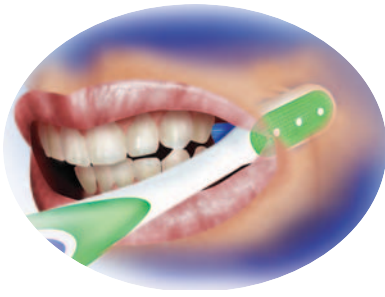
^aThe Colgate-Palmolive Company, New York, NY 10022; 800-338-8388

Conclusion

The methodical development process described herein clearly underscores the importance of cross-category interaction throughout the development of the Colgate® 360™ toothbrush, particularly during the initial “discovery” stages. The following articles will disclose the significant body of work undertaken to objectively assess the clinical performance of this new toothbrush, which was designed to deliver a “whole mouth clean.”

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Clinical Efficacy of Colgate® 360^o™ and Three Commercially Available Toothbrushes on the Removal of Desquamated Epithelial Cells

Abstract: A clinical study was done to evaluate the performance of four toothbrushes on the removal of desquamated epithelial cells after brushing according to the manufacturers' instructions for use. This randomized, crossover-design clinical study compared a new manual toothbrush (Colgate® 360^o™) to two commercially available manual toothbrushes (Oral-B® CrossAction® and Oral-B® Indicator®) and a commercially available battery-powered toothbrush (Crest® SpinBrush™ PRO). Adult men and women subjects reported to the clinical facility after a 1-week "washout" period of brushing with a regular fluoride dentifrice and a soft-bristled toothbrush. Participants reported having refrained from oral hygiene procedures, eating, and drinking that morning. After providing a baseline rinse sample, subjects brushed their teeth for 1 minute with their assigned toothbrush and a commercially available fluoride toothpaste, then returned 30 minutes later to provide postuse rinse samples. Subjects refrained from dental hygiene, eating, and drinking during the 30-minute evaluation period. To provide the samples, subjects rinsed with 10 mL of sterile phosphate-buffered saline solution for 10 seconds. Each collected sample was centrifuged, resuspended, and run in a colorimetric assay to determine the level of desquamated epithelial cells found in the rinse as measured by the absorbance at 570 nm. Twenty adults completed the study. At baseline, the mean levels of desquamated epithelial cells for the 4 treatments were 0.70 ± 0.27 , 0.63 ± 0.20 , 0.69 ± 0.30 , and 0.62 ± 0.31 for the Colgate® 360^o™, Oral-B® Indicator®, Crest® SpinBrush™ PRO, and Oral-B® CrossAction®, respectively. Posttreatment, the mean levels of epithelial cells were 0.19, 0.38, 0.42, and 0.34, respectively. All of the treatments provided a statistically significant reduction compared to their respective baseline. In addition, the Colgate® 360^o™ toothbrush was statistically significantly better than the other three toothbrushes in reducing desquamated epithelial cells. Therefore, the results of this randomized, crossover clinical study indicate that the newly designed Colgate® 360^o™ manual toothbrush, with a tongue-cleaning implement on the back of the brush head, was statistically significantly more effective than the Oral-B® Indicator®, Crest® SpinBrush™ PRO, and Oral-B® CrossAction® toothbrushes in removing desquamated epithelial cells.

Oral malodor is composed mainly of volatile sulfur compounds (VSC), especially hydrogen sulfide, methyl mercaptan, and dimethyl sulfide.¹ Various other compounds contributing to oral malodor include volatile organic acids such as butyric acid and propionic acid, and amines such as cadaverine, indole, and skatole.^{2,3} These components are derived from the proteolytic breakdown by oral bacteria of sulfur-

Table 1—Summary of Age and Gender Characteristics of Study Participants

Toothbrush	Men	Women	Total	Mean Age	Range
Colgate® 360™	6	14	20	33	23–53
Oral-B® Indicator®	6	14	20	33	23–53
Crest® SpinBrush™ PRO	6	14	20	33	23–53
Oral-B® CrossAction®	6	14	20	33	23–53

containing peptides and amino acids derived from various salivary origins. One key source of food for the bacteria is desquamated epithelial cells from the cheeks and lips.⁴ Tonzetich and Kestenbaum⁵ showed that incubating whole saliva or saliva sediment, which contained exfoliated epithelial cells, produced oral malodor, whereas centrifuged salivary supernatant did not generate oral malodor. Tonzetich and Johnson⁶ provided further evidence of the importance of cellular components in oral malodor formation. Plaque, which is composed of bacteria and salivary proteins, has a strong potential for oral malodor formation.⁵ The loose, outermost layer of plaque, often referred to as the material alba, is composed of desquamated epithelial cells and some blood cell elements.⁷ These studies indicate that desquamated epithelial cells play a key role in the production of oral malodor.

Several approaches have been pursued to control oral malodor. However, none of the approaches involved a means of actively reducing or removing desquamated epithelial cells from the primary source—cheeks and lips. Yonezawa and coworkers⁸ found a direct relationship between oral mucosal cleaning and the production of VSC in the mouth. Under normal physiologic conditions, chewing and swallowing provide a mechanism for continuous removal of loose epithelial cells, bacteria, and food debris. However, microorganisms of the biofilm are more resistant to dislodging without mechanical manipulation. Although antiseptic oral products may improve oral malodor,⁹ they are ineffective in eliminating the cellular components of the

biofilm. It is well accepted that employing diligent oral hygiene practice is effective in controlling physiologic oral malodor.¹ Therefore, mechanical cleaning of the teeth and soft tissues is probably the most effective measure for removing the biofilm and controlling the number of oral microorganisms.

The Colgate-Palmolive Company has developed a newly designed manual toothbrush with a cleaning implement on the back of the brush head for the gentle and safe cleaning of the oral mucosa lining of the tongue, cheeks, and lips. A clinical study was done to compare the newly designed manual toothbrush and three commercial toothbrushes (two manual toothbrushes and one battery-operated toothbrush) using the reduction of desquamated epithelial cells as a measurement of efficacy.

Materials and Methods

Study Subjects

A randomized, single-use clinical design with a four-treatment crossover phase compared the desquamating effect of four toothbrushes. All study participants used each of the test toothbrushes. The subject population was comprised of healthy adult men and women who were enrolled in the study based on the following criteria: (1) they were between the ages of 18 and 70 and in generally good health; (2) they possessed a minimum of 20 natural, uncrowned teeth (excluding third molars); (3) they would be available for the duration of the study; and (4) they signed an informed consent form.

Participants were excluded from the study if

Table 2—Mean Pretreatment Baseline Levels of Desquamated Epithelial Cells of Study Participants, Measured by Absorbance at 570 nm

Toothbrush	n	Baseline Levels of Epithelial Cells (A_{570})	
		Mean ± SD	Significance*
Colgate® 360™	20	0.70 ± 0.27	NS
Oral-B® Indicator®	20	0.63 ± 0.20	NS
Crest® SpinBrush™ PRO	20	0.69 ± 0.30	NS
Oral-B® CrossAction®	20	0.62 ± 0.31	NS

*Significance of analysis of variance comparison of baseline means. NS = $P \geq .05$
 A_{570} = absorbance at 570 nm; SD = standard deviation; NS = not significant.

Table 3—Mean Posttreatment Levels of Desquamated Epithelial Cells After Brushing With the Four Toothbrushes

Toothbrush	n	OD ₅₇₀ Postbrushing Mean ± SD	Percent Reduction vs Baseline*	Significance vs Baseline†
Colgate® 360™	20	0.19 ± 0.12	74.3	<i>P</i> < .05
Oral-B® Indicator®	20	0.38 ± 0.19	39.7	<i>P</i> < .05
Crest® SpinBrush™ PRO	20	0.42 ± 0.27	39.1	<i>P</i> < .05
Oral-B® CrossAction®	20	0.34 ± 0.17	45.2	<i>P</i> < .05

*Within-treatment percent reduction between baseline and posttreatment levels of epithelial cells, expressed as a percentage of the baseline levels.
†Significance of within-treatment paired *t* test comparison of the baseline levels vs postbaseline levels.
OD₅₇₀ = optical density at 570 nm; SD = standard deviation.

they had allergies to consumer products or their ingredients, had full or partial dentures, used tobacco products, or had metabolic diseases. Additionally, pregnant or lactating women and individuals participating in other panel tests or clinical studies were excluded from the study.

Test Products

Desquamated epithelial cells were evaluated after using the following toothbrushes: Colgate® 360™,^a Oral-B® Indicator®,^b Crest® SpinBrush™ PRO®, and Oral-B® CrossAction®.^b All subjects brushed with a regular fluoride toothpaste (Colgate® Cavity Protection Great Regular Flavor Fluoride Toothpaste^a) for the entire study.

Clinical Procedure

Before beginning the study, participants were required to wash out the effects of previously used oral products by brushing with a regular toothbrush and the fluoridated dentifrice. The subjects reported to the designated clinical study site on the morning of the baseline saliva collection without performing oral hygiene, eating, or drinking. The subjects provided baseline saliva samples by rinsing for 10 seconds with 10 mL of sterile phosphate-buffered saline solution. Subjects were then issued a randomly assigned toothbrush and a tube of fluoride toothpaste and given instructions to brush their teeth for 1 minute per manufacturer's suggested instructions for use. For subjects using the Colgate® 360™ toothbrush, the 1-minute brushing was followed by sweeping the tongue with the implement on the back of the brush head for 10 seconds. After the treatment, subjects refrained from eating and drinking for 30 minutes, at which time they provided a postbrushing saliva sample. After a 1-

week washout period of brushing with a regular fluoridated dentifrice and a soft-bristled, flat-trimmed toothbrush, the treatment regimen was repeated with subjects using the other products.

Analysis of Desquamated Epithelial Cells

Samples collected at baseline and 30 minutes after brushing were analyzed for epithelial cells. Samples were mixed with 1 mL of 10X phosphate-buffered saline and vigorously vortexed for 10 seconds. Duplicates of each sample were centrifuged for 15 minutes at 3,000 rpm. After removing the supernatant, the cells were resuspended in 2.5 mL of phosphate-buffered saline solution, and 2.5 mL of MTT reagent^d (tetrazolium (3-(4, 5-dimethylthiazolyl-2)-2, 5-diphenyl-tetrazolium bromide) was added to the tubes and then incubated in a gently shaking water bath for 2 hours at 37°C. The samples were then centrifuged for 15 minutes at 3,000 rpm. The supernatant was removed and 3 mL of acid isopropanol was added to the tube. The absorbance at 570 nm was measured. For each treatment, the absorbance of the posttreatment values was compared to the absorbance values at baseline.

Statistical Methods

The absorbance (*A*₅₇₀) measurement of duplicate samples was averaged and the reduction from baseline was calculated. The data were analyzed as follows:

- For each product, a paired *t* test was done to compare postuse to baseline.
- For postbrushing time points, analysis of covariance (ANCOVA) was performed on the scores, with the corresponding baseline as covariate and subject and product as effects.

Treatments were declared statistically significantly different if *P* < .05.

^aThe Colgate-Palmolive Company, New York, NY 10022; 800-338-8388

^bOral-B Laboratories, Belmont, CA 94002; 800-44ORALB

^cThe Procter & Gamble Company, Cincinnati, OH 45202; 800-492-7378

^dSigma-Aldrich Inc, St. Louis, MO 63103; 800-325-3010

Results

A summary of the age and gender characteristics of the study population is presented in Table 1. All participants completed the study and showed satisfactory signs of compliance to the instructions. There were no adverse reactions observed or reported by the participants when questioned about adverse effects.

Baseline Data

Table 2 is a summary of the baseline desquamated-cell levels for the four toothbrush treatment groups. The desquamated-cell levels measured by the absorbance at 570 nm (A_{570}) were 0.70, 0.63, 0.69, and 0.62 for the Colgate® 360^o, Oral-B® Indicator®, Crest® SpinBrush™ PRO, and Oral-B® CrossAction® toothbrush groups, respectively. There was no statistically significant difference at baseline in desquamated-cell levels for the 4 study groups ($P > .05$).

Postuse Results

Subjects brushed their teeth as specified by the toothbrush manufacturers, with at least a 2-day washout period between the use of each toothbrush. The posttreatment levels of desquamated epithelial cells after brushing with the four toothbrushes are shown in Table 3. The final mean level of desquamated epithelial cells for the 4 treatment groups was statistically significantly different from baseline ($P < .05$). The greatest reduction was observed after brushing with the Colgate® 360^o toothbrush, which provided a 74.3% reduction compared to baseline (Table 3, Figure 1). Interestingly, when the post-treatment desquamated epithelial-cell levels

were compared using ANCOVA, the Colgate® 360^o provided statistically significantly greater reduction than the Oral-B® Indicator®, Crest® SpinBrush™ PRO, and Oral-B® CrossAction® toothbrushes, which were not statistically significantly different from each other. The adjusted treatment means for the postuse levels of desquamated epithelial cells are given below, using the lines method to display.

Colgate® 360 ^o	Oral-B® Indicator®	Crest® SpinBrush™ PRO	Oral-B® CrossAction®
0.19	0.38	0.42	0.34

Discussion

There is a strong correlation between poor oral hygiene and the common dental infections, namely caries and periodontal diseases, as well as other oral conditions such as oral malodor.^{4,10-13} Consumer education and awareness have created a demand for dental products that can provide multiple benefits. A new manual toothbrush, the Colgate® 360^o toothbrush, was recently developed. This toothbrush has tightly packed and tapered bristles for subgingival and interproximal cleaning; a raised cleaning tip for accessing hard-to-reach places in the mouth; and soft, dental-like polishing cups for the delicate removal of tooth stains. Additionally, a special feature of the toothbrush is the soft, conically shaped nubs on the back of the brush head for cleaning the tongue, cheeks, and lips. The Colgate® 360^o toothbrush has been shown to reduce plaque, gingivitis, and breath VSC responsible for oral malodor, as well as remove hydrogen-sulfide-forming bacteria on the tongue.¹⁴⁻¹⁷

Although there is no ideal single procedure that can objectively assess and quantify the extent of oral cleanliness, we can use information from direct clinical methods along with indirect methods to build a profile for a product's cleaning effect. Plaque, gingivitis, and mouth-odor assessments by trained practitioners are among the most notable direct clinical methods. In-

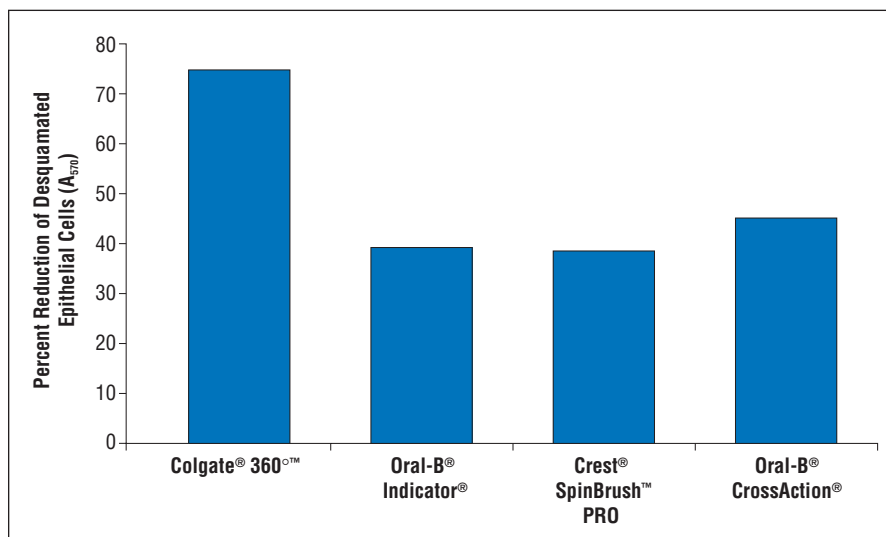


Figure 1—The percent reduction of the levels of desquamated epithelial cells after brushing with the four toothbrushes.

direct methods—such as bacterial cultures that assess oral-malodor potential by quantifying putative microorganisms believed to produce various bacterial by-products—also are helpful in determining product efficacy. However, these methods cannot measure the ability of a product to exfoliate or clean the surfaces of the oral soft tissue of desquamated epithelial cells that can serve as a major source of food for bacteria. An assay that uses the reduction of tetrazolium (3-(4, 5-dimethylthiazolyl-2)-2, 5-diphenyltetrazolium bromide) salts by metabolically active cells to form intracellular purple dye appears to be an effective method to measure and quantify desquamated epithelial cells. This article presents the findings of a clinical study that compared the Colgate® 360°™, Oral-B® Indicator®, Crest® SpinBrush™ PRO, and Oral-B® CrossAction® toothbrushes for their ability to reduce the levels of desquamated epithelial cells. The Colgate® 360°™ toothbrush was statistically significantly more effective than the three commercially available toothbrushes in reducing the levels of desquamated epithelial cells. All subjects used the same toothpaste. Therefore, the increase in the removal of desquamated epithelial cells observed after using the Colgate® 360°™ toothbrush was likely because of the implement on the back of the brush head. It was shown that the use of this toothbrush led to the reduction of plaque, gingivitis, VSC, and hydrogen-sulfide-forming bacteria.¹⁴⁻¹⁷ A likely mechanism of action is the reduction of the desquamated epithelial cells. These cells contain proteins that are rich in sulfur-containing amino acids and that accumulate in tongue biofilm and dental plaque—sites of oral malodor production.^{1,18} Given that the severity of oral malodor is mainly influenced by the bacterial plaque level and substrate availability, reducing one or both of these variables would have a significant effect on oral malodor.

Conclusion

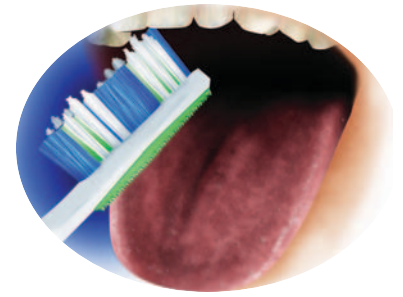
The results of this randomized, crossover clinical study indicate that the Colgate® 360°™ toothbrush, with its unique features, provides significant reduction of desquamated epithelial cells that serve as a key source of food for bacteria responsible for the formation of oral malodor. This toothbrush was statistically significantly

better than three commercially available toothbrushes in reducing such cells when the toothbrushes were used as intended by the manufacturers. Subjects using the Colgate® 360°™ toothbrush also brushed their tongue with the implement on the back of the brush head. Because of its unique features, the Colgate® 360°™ toothbrush stands to provide consumers with a safe and convenient means to achieve a healthy, clean mouth.

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Clinical Comparison of a New Manual Toothbrush on the Level of Hydrogen-sulfide-forming Bacteria on the Tongue



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Abstract: *The objective of this randomized, crossover study was to compare the effectiveness of a newly designed manual toothbrush (Colgate® 360^{OTM}) to two commercially available manual toothbrushes (Oral-B® Indicator® and Oral-B® CrossAction®) and a battery-powered toothbrush (Crest® SpinBrush™ PRO) for their ability to reduce hydrogen-sulfide-forming bacteria on the tongue. After a washout period, subjects arrived at the clinical site for baseline sampling without performing dental hygiene, eating, or drinking. Subjects sampled the left side of their tongue with a cotton swab. Subjects brushed for 1 minute with the assigned test toothbrush and regular fluoride toothpaste. Those using the Colgate® 360^{OTM} toothbrush were instructed to clean their tongue with the implement on the back of the brush head for 10 seconds. After 2 hours, the subjects returned to the clinical site having refrained from dental hygiene, eating, and drinking for posttreatment sampling, this time sampling the right side of their tongue. After a minimum 2-day washout period, subjects repeated the same regimen using the other toothbrushes. Collected tongue samples were dispersed in sterile water, serially diluted in sterile phosphate-buffered saline, and plated in duplicate onto lead acetate agar. When plated on this medium, bacteria that produce hydrogen sulfide appear as dark-pigmented colonies. After 72 hours of incubation, the dark colonies were counted, expressed as log colony-forming units/mL, and reduction from baseline was calculated. Thirty-one adult men and women completed the clinical study. There was no significant difference between baseline hydrogen-sulfide-forming bacteria levels. Posttreatment, the log reduction of bacteria was 0.80, 0.41, 0.33, and 0.44 for the Colgate® 360^{OTM}, Oral-B® Indicator®, Crest® SpinBrush™ PRO, and Oral-B® CrossAction®, respectively. Statistical analysis indicated that the Colgate® 360^{OTM} toothbrush was statistically significantly better ($P < .05$) than the 3 commercial toothbrushes in reducing the levels of hydrogen-sulfide-forming bacteria on the tongue.*

At any given time—and especially after waking up in the morning—the majority of the adult population is afflicted with unpleasant or bad breath, a condition clinically referred to as halitosis.¹⁻³ In a healthy individual with no systemic disorders, the mouth is the main contributor of bad breath.⁴ People suffering from bad breath are very sensitive about it and tend to avoid social situations. The problem results when proteins from food and saliva debris are broken down by bacteria.⁵⁻⁷

The tongue, with its fissures, crypts, and high mucosal papillae, retains considerable quantities of debris that support and protect a large bacterial population, including bacteria that have been strongly implicated in causing oral malodor.⁸ Under anaerobic conditions, the tongue's bacterial population forms waste products such as volatile sulfur compounds (VSC),

Table 1—Inclusion and Exclusion Criteria of Study Participants

Inclusion Characteristics
Must be a man or woman between 18 and 66 years of age
Must be in good general health based on self-assessment
Must be in good oral health based on self-assessment
Must be available during the treatment phase of this study for baseline and 2-hour appointments
Should have a minimum of 20 natural, uncrowned teeth (excluding third molars)
Must give written informed consent
Exclusion Characteristics
Participation in any other oral clinical studies for the duration of this study
Full or partial (upper or lower) dentures
Pregnant or lactating (breast-feeding)
Use of tobacco products
History of allergy to common dentifrice ingredients
Use of phenol-flavored products, such as mint-flavored candies and chewing gum, the morning of the study and during the sampling periods
Immunocompromised individuals (HIV, AIDS, immunosuppressive drug therapy)
Individuals who, because of medical conditions, cannot go without eating or drinking for the postuse treatment-evaluation time points (2 hours + overnight)

organic acids, and amines.⁹ Bacterial species that produce such malodorous compounds include *Treponema denticola*, *Porphyromonas gingivalis*, *Prevotella intermedia*, *Fusobacterium nucleatum*, the *Eubacterium* species, and not-yet-cultivated species.^{6,10-12} Attempts to treat oral malodor involve the use of different medicaments, many of which merely mask the problem, while others—like Colgate® Total®^a, a toothpaste containing triclosan, copolymer, and fluoride—control the problem at the source by controlling the bacteria.¹³⁻¹⁵ It has been well established that an effective means of alleviating oral malodor is through the active removal of bacteria from the tongue.^{1,3,16-18} Bladed tongue scrapers have been used.¹⁹ These devices, usually made of hard plastic or metal, can lead to abrasion of the tongue when used vigorously. The Colgate-Palmolive Company has developed a newly designed manual toothbrush with a cleaning implement on the back of the brush head for the safe and soft cleansing of oral soft tissue, such as the tongue and cheeks. A clinical study was done to compare this newly designed manual toothbrush and three commercial toothbrushes (two manual toothbrushes and a battery-operated toothbrush) using the reduction of hydrogen-sulfide-forming

bacteria on the tongue as the measurement of efficacy.

Materials and Methods

Test Products

The effect on breath VSC was evaluated after using the following four toothbrushes: Colgate® 360™ toothbrush^a, Oral-B® Indicator® toothbrush^b, Crest® SpinBrush™ PRO toothbrush^c, and Oral-B® CrossAction® toothbrush^b. All subjects brushed with fluoridated toothpaste (Colgate® Cavity Protection Great Regular Flavor Fluoride Toothpaste^a) for the entire study.

Clinical Methods

The study employed a crossover, four-treatment design in which all study participants used each of the test toothbrushes. Subjects who met the entrance criteria (Table 1) and signed the informed consent form were entered into the study.

After a 7-day pretreatment washout period in which subjects used a Colgate® Plus toothbrush^a and a regular fluoride toothpaste, subjects reported to the clinical site in the morning for baseline tongue bacteria sampling without performing oral hygiene, eating, or drinking. Tongue samples

^aOral-B Laboratories, Belmont, CA 94002; 800-44ORALB

^cThe Procter & Gamble Company, Cincinnati, OH 45202; 800-492-7378

^aThe Colgate-Palmolive Company, New York, NY 10022; 800-338-8388

Table 2—Summary of Age and Gender Characteristics of Study Participants

Toothbrush	Men	Women	Total	Age Range
Colgate® 360™	13	18	31	23-54
Oral-B® Indicator®	13	18	31	23-54
Crest® SpinBrush™ PRO	13	18	31	23-54
Oral-B® CrossAction®	13	18	31	23-54

were collected by swabbing the left side of the tongue six times, rotating the swab as it was brought forward. The subjects were given a test toothbrush with a full ribbon of toothpaste and instructed to brush their teeth for 1 minute. Subjects brushing with the Colgate® 360°™ toothbrush were instructed to follow up the brushing with a 10-second sweep of the tongue surface with the tongue-and-cheek cleaner located on the back of the toothbrush head. Subjects were instructed to refrain from eating, drinking, and performing oral hygiene for 2 hours, after which they returned to the testing facility for postbrushing tongue sampling from the right side of the tongue. There was a minimum 2-day washout period during which subjects used a Colgate® Plus toothbrush and the regular fluoride toothpaste.

Tongue Sample Analysis

The tongue samples collected at baseline and 2 hours after brushing were dispersed in sterilized water by vigorously vortexing them for 10 seconds. Each sample was serially diluted in phosphate-buffered saline and plated, in duplicate, onto lead acetate agar. The plates were incubated anaerobically for a minimum of 72 hours at 37°C. Colony-forming units (CFU) indicated by dark colonies were counted and converted to log units. The reduction in hydrogen-sulfide-forming bacteria vs baseline was calculated for each of the products.

Statistical Methods

The numbers of hydrogen-sulfide-forming bacteria were expressed as log CFU/mL. The mean and standard deviation of the log CFU data were calculated for each treatment at the baseline and 2-hour postuse time points. The following analyses were performed:

- For each toothbrush, a paired *t* test was done to compare the 2-hour postuse time points to baseline.
- For the 2-hour postbrushing time point, analysis of covariance (ANCOVA) was performed on the scores, with the corresponding baseline as covariate and with subject and product as effects.

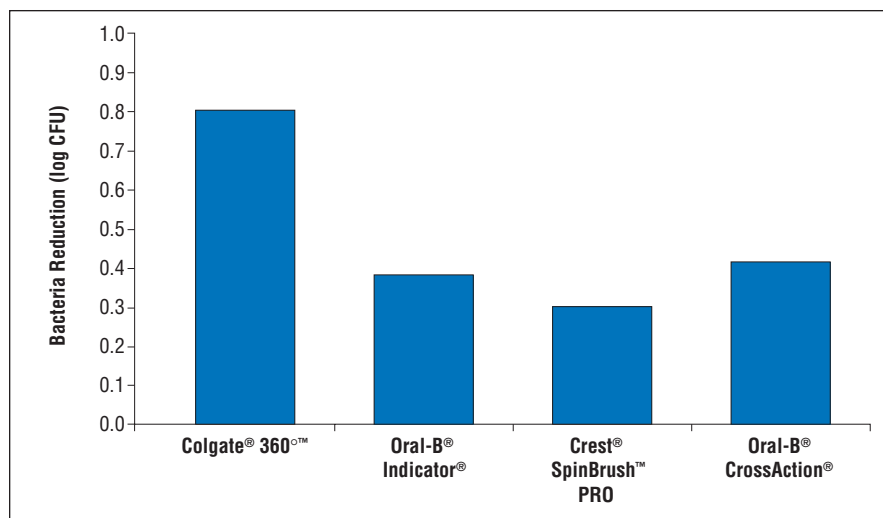


Figure 1—Reduction in tongue bacteria for the postuse time point. CFU = colony-forming units.

Results

A summary of the characteristics of the subjects who participated in the study is presented in Table 2. Thirty-one adults qualified and completed the study. All subjects who completed the study showed satisfactory signs of compliance to the instructions, and there were no reported side effects.

Baseline

Table 3 presents a summary of the baseline hydrogen-sulfide-forming bacteria levels on the tongue for the four treatment groups. The mean CFU of odor-producing bacterial levels for the Colgate® 360°™, Oral-B® Indicator®, Crest® SpinBrush™ PRO, and Oral-B® CrossAction® toothbrush groups were 5.72, 5.62, 5.58, and 5.53, respectively. No statistically significant difference was observed between the bacteria CFU associated with the 4 study treatments at baseline ($P > .05$).

Posttreatment Data

The posttreatment levels of hydrogen-sulfide-producing bacteria on the tongue after using the test toothbrushes are summarized in Table 4. The mean CFU of hydrogen-sulfide-forming bacteria levels for the Colgate® 360°™, Oral-B® Indicator®, Crest® SpinBrush™ PRO, and Oral-B® CrossAction® toothbrushes were 4.92, 5.21, 5.25, and 5.09, respectively. The respective reductions of these bacteria, represented by dark-pigmented spots on the lead acetate agar, were 0.80, 0.41, 0.33, and 0.44 compared to baseline (Figure 1). Compared to baseline, the level of bacteria on the tongue was statistically significantly reduced after using the four test toothbrushes (Table 4).

Table 3—Mean Pretreatment Baseline Breath Hydrogen-sulfide-forming Bacteria Levels of Study Participants

Toothbrush	n	Baseline Bacteria Levels (CFU)—Mean ± SD	Significance*
Colgate® 360™	31	5.72 ± 0.43	NS
Oral-B® Indicator®	31	5.62 ± 0.44	NS
Crest® SpinBrush™ PRO	31	5.58 ± 0.39	NS
Oral-B® CrossAction®	31	5.53 ± 0.44	NS

*Significance of analysis of variance comparison of baseline means. NS = $P \geq .05$.
CFU = colony-forming units; SD = standard deviation; NS = not significant.

However, when the posttreatment odor-causing bacteria levels for the 4 test toothbrushes were compared, the ANCOVA analysis showed that the Colgate® 360™ toothbrush was statistically significantly better ($P < .05$) than the Oral-B® Indicator®, Crest® SpinBrush™ PRO, and Oral-B® CrossAction® toothbrushes, which were not statistically significantly different from each other. The adjusted treatment means for the postuse levels of hydrogen-sulfide-forming bacteria are given below, using the lines method to display.

Colgate® 360™	Oral-B® Indicator®	Crest® SpinBrush™ PRO	Oral-B® CrossAction®
0.80	0.41	0.33	0.44

Discussion

Several studies have shown that gram-negative anaerobic bacteria, especially on the tongue dorsum, are mainly responsible for oral malodor formation.^{2,3,20-22} Therefore, to control oral malodor, the focus can be on the reduction of the bacteria on the tongue, especially those that are responsible for the production of volatilized odiferous compounds such as hydrogen sulfide. For this, mechanical cleaning of the tongue has been strongly emphasized as a means of controlling oral malodor. Studies have shown that cleaning the tongue reduced the number of microorganisms in saliva, in plaque, and on the tongue.^{8,22,23}

Despite this evidence, patients do not readily clean their tongue during daily oral hygiene procedures. Providing patients with an oral hygiene implement that will help or encourage cleaning the tongue during regular oral hygiene procedures could lead to improvement in oral hygiene. Studies have shown that merely improving oral hygiene procedures reduces the level of oral malodor.^{3,8,17,24}

Conventionally, oral hygiene has focused on toothbrushing using, for example, a toothbrush and a toothpaste containing fluoride. For this, manufacturers have developed various toothbrushes that have been documented to help in the reduction of dental plaque, gingivitis, and tooth staining.²⁵⁻²⁹ A high-performance toothbrush, the Colgate® 360™, was developed by the Colgate-Palmolive Company. The toothbrush has tightly packed and tapered bristles for subgingival and interdental cleaning, a raised cleaning tip for reaching hard-to-reach places in the mouth, and soft dental-like polishing cups for the delicate removal of tooth stains. An additional special feature of the toothbrush is the soft, conically shaped nubs on the back of the toothbrush head for cleaning the tongue, cheeks, and lips. This toothbrush has been documented to reduce plaque and gingivitis, clean desquamated epithelial cells from the cheeks and lips, and reduce VSC, which are a significant component of the oral malodor bouquet.³⁰⁻³³

Table 4—Summary of Hydrogen-sulfide Levels (in CFU) After Brushing With the Four Toothbrushes

Toothbrush	n	Postbrushing Bacteria Levels (CFU) Mean ± SD	Percent Reduction vs Baseline*	Significance Compared to Baseline†
Colgate® 360™	31	4.92 ± 0.40	84.2	$P < .05$
Oral-B® Indicator®	31	5.21 ± 0.41	54.0	$P < .05$
Crest® SpinBrush™ PRO	31	5.25 ± 0.53	53.2	$P < .05$
Oral-B® CrossAction®	31	5.09 ± 0.44	63.7	$P < .05$

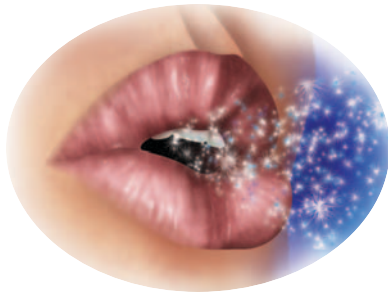
*Within-treatment percent reduction between baseline and postbaseline levels, expressed as a percentage of the baseline levels. Percent reduction was calculated using the antilog of the mean values at baseline and postbrushing.
†Significance of analysis of covariance comparison of baseline-adjusted mean level of bacteria.
CFU = colony-forming units; SD = standard deviation.

Conclusion

Reported in this article are the results of a study that compared the new Colgate® 360°™ toothbrush to three commercially available toothbrushes (Oral-B® Indicator®, Crest® SpinBrush™ PRO, and Oral-B® CrossAction®) for their ability to reduce the level of hydrogen-sulfide-producing bacteria on the tongue. The subjects used the toothbrushes as intended by the manufacturers. In addition to brushing their teeth, subjects using the Colgate® 360°™ toothbrush brushed their tongue with the implement on the back of the brush head. The subjects using the Colgate® 360°™ toothbrush had statistically significantly lower levels of hydrogen-sulfide-forming bacteria on the tongue than the groups using the three commercially available toothbrushes. The reduction of bacteria was corroborated by the concurrent reduction of VSC.³³ There were no reports of negative side effects associated with using the toothbrush. The results of this study support the use of the new Colgate® 360°™ toothbrush as part of an everyday oral hygiene procedure to help reduce oral malodor of intraoral origin.

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Clinical Comparison of a New Manual Toothbrush on Breath Volatile Sulfur Compounds

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Abstract: The objective of this randomized, crossover study was to compare the effectiveness of a newly designed manual toothbrush (Colgate® 360^{OTM}) to two commercially available manual toothbrushes (Oral-B® Indicator® and Oral-B® CrossAction®) and a battery-powered toothbrush (Crest® SpinBrush™ PRO) for their ability to reduce overnight volatile sulfur compounds (VSC) associated with oral malodor. The study followed a four-period crossover design. Following a washout period, prospective subjects arrived at the testing facility without eating, drinking, or performing oral hygiene for baseline evaluation of breath VSC levels. For each phase of the study, subjects were given one of the test toothbrushes and a tube of regular toothpaste to take home, and they were instructed to brush their teeth in their customary manner for 1 minute. When using the Colgate® 360^{OTM} toothbrush, subjects were instructed to clean their tongue with the implement on the back of the brush head for 10 seconds. The following morning, subjects reported to the testing facility, again without performing oral hygiene, eating, or drinking, for the overnight evaluation. After a minimum 2-day washout period, subjects repeated the same regimen using the other toothbrushes. The levels of breath VSC were evaluated instrumentally using a gas chromatograph equipped with a flame photometric detector. Measurements were taken in duplicate and then averaged. The levels of VSC were expressed as parts per billion (ppb) in mouth air. Sixteen men and women completed the study. At baseline, the mean levels of VSC in mouth air for the 4 toothbrushes were 719.8 ppb ± 318.4 ppb, 592.8 ppb ± 264.6 ppb, 673.8 ppb ± 405.9 ppb, and 656.2 ppb ± 310.2 ppb for the Colgate® 360^{OTM}, Oral-B® Indicator®, Crest® SpinBrush™ PRO, and Oral-B® CrossAction®, respectively. Overnight, the mean breath VSC levels after using the four toothbrushes were lower than those observed at baseline. The respective mean levels of breath VSC were 266.5 ppb ± 269.9 ppb, 545.2 ppb ± 346.1 ppb, 567 ppb ± 335.7 ppb, and 554.6 ppb ± 398.4 ppb. Only the Colgate® 360^{OTM} toothbrush provided a statistically significant reduction ($P < .05$) in breath VSC vs baseline. Additionally, the Colgate® 360^{OTM} toothbrush was statistically significantly better ($P < .05$) than the three commercial toothbrushes in reducing breath VSC. Therefore, the results of this randomized, crossover clinical study indicate that a newly designed manual toothbrush with a tongue-cleaning implement on the back of the brush head was significantly more effective than three commercially available toothbrushes in reducing morning breath VSC associated with oral malodor.

Writings on oral malodor (*fetor oris*) date back to ancient Greek and Roman times, indicating that this disaffecting condition has been a long-held concern of humans for thousands of years.¹ Oral malodor not only causes embarrassment to people during social engagements but also creates a barrier in people's personal lives. Oral malodor is a common phenomenon that results from the bacterial metabolism of pro-

teinaceous materials from the food we eat, or salivary particles such as desquamated epithelial cells and blood cells.² Studies have shown that gram-negative bacteria are mainly responsible for oral malodor formation, with the tongue being the primary site for such activity.³⁻⁷ These bacteria produce volatile sulfur compounds (VSC) through the anaerobic digestion of sulfur-containing amino acids. About 85% of bad breath consists of VSC such as hydrogen sulfide, methyl mercaptan, and dimethyl sulfide.⁸

Attempts to treat oral malodor involve the use of different medicaments, many of which merely mask the problem, while others—like Colgate® Total®^a, a toothpaste containing triclosan, a copolymer, and fluoride—control the problem at the source by controlling the bacteria.⁹⁻¹¹ Additionally, bladed tongue scrapers have been used in the past.¹² However, these devices, usually made of hard plastic or metal, when used vigorously to clean the tongue can lead to abrasion of the tongue. The Colgate-Palmolive Company has developed a newly designed manual toothbrush with a cleaning implement on the back of the brush head for the soft and safe cleaning of oral soft tissues, such as the tongue and cheeks. A clinical study was done to compare this newly designed manual toothbrush and three commercial toothbrushes (two manual toothbrushes and one battery-operated toothbrush) using the reduction of breath VSC as the measurement of efficacy.

^aThe Colgate-Palmolive Company, New York, NY 10022; 800-338-8388

Materials and Methods

Test Products

The effect on breath VSC was evaluated after using the following four toothbrushes: Colgate® 360^o™,^a Oral-B® Indicator®^b, Crest® SpinBrush™ PRO^c, and Oral-B® CrossAction®^b. All subjects brushed with a regular fluoride toothpaste (Colgate® Cavity Protection Great Regular Flavor Fluoride Toothpaste^a) for the entire study.

Clinical Methods

The study employed a randomized, crossover, four-treatment design in which all study participants used each of the test toothbrushes. Subjects who met the entrance criteria (Table 1) and signed the informed consent form were entered into the study.

After a 7-day pretreatment washout period in which subjects used a regular flat-trimmed toothbrush (Colgate® Plus toothbrush^a) and a regular fluoride toothpaste, subjects reported to the testing facility on the morning of the pretreatment sampling without eating, drinking, or performing oral hygiene for baseline breath VSC evaluation. Each subject's breath odor was evaluated using a Wasson-ECE custom-built breath-sampling gas chromatography system equipped with a flame photometric detector (Agilent Technologies Model 6890 gas chromatograph^d). This method was described initially by Tonzetich,⁸ and later by Niles and Gaffar.¹³ Qualifying subjects were ran-

^aOral-B Laboratories, Belmont, CA 94002; 800-44ORALB

^bThe Procter & Gamble Company, Cincinnati, OH 45202; 800-492-7378

^dAgilent Technologies, Inc, Palo Alto, CA 94306; 877-424-4536

Table 1—Inclusion and Exclusion Criteria

Inclusion Characteristics
Must be a man or woman between 18 and 65 years of age
Must be in good general health based on self-assessment
Must be in good oral health based on self-assessment
Must be available during the treatment phase of this study for baseline and 2-hour appointments
Should have a minimum of 20 natural, uncrowned teeth (excluding third molars)
Must give written informed consent
Must have no known history of allergy to personal care/consumer products or their ingredients
Must have an average baseline breath volatile sulfur compounds level ≥ 300 ppb, as measured by gas chromatograph
Exclusion Characteristics
Participation in any other oral clinical studies for the duration of this study
Full or partial (upper or lower) dentures
Pregnant or lactating (breast-feeding)
Use of tobacco products
History of allergy to common dentifrice ingredients
Use of phenol-flavored products, such as mint-flavored candies and chewing gum, the morning of the study and during the sampling periods
Immunocompromised individuals (HIV, AIDS, immunosuppressive drug therapy)
Individuals who, because of medical conditions, cannot go without eating or drinking for the postuse treatment-evaluation time points (2 hours + overnight)

Table 2—Summary of Age and Gender Characteristics of Study Participants

Toothbrush	Men	Women	Total	Age Range
Colgate® 360™	9	7	16	21–48
Oral-B® Indicator®	9	7	16	21–48
Crest® SpinBrush™ PRO	9	7	16	21–48
Oral-B® CrossAction®	9	7	16	21–48

domized, then assigned to one of the four treatment sequences. This sequence specified which of the study treatments were to be used by the subject during each of the four experimental periods.

After baseline evaluation, qualified subjects brushed their teeth with the assigned test toothbrush and the regular fluoride toothpaste for 1 minute. Subjects using the Colgate® 360™ toothbrush were instructed to clean their tongue with the cleaning implement on the back of the toothbrush head for 10 seconds after the 1-minute brushing cycle. Subjects were given the assigned test toothbrush and toothpaste to take home and were instructed to brush their teeth before going to bed, following the same treatment procedure as they had earlier that morning. The next day, subjects reported to the testing facility, again without eating, drinking, or performing oral hygiene, for posttreatment overnight evaluation. Subjects then went on a minimum 2-day washout period using a regular flat-head toothbrush and regular fluoride toothpaste.

The entire sampling and measurement process was performed in duplicate at each evaluation, and the resulting two scores were averaged. Subjects were instructed to sit with their mouths closed for 10 minutes before sampling. Air was pulled from the subject's mouth directly into the inlet of the gas chromatograph through the gas-sampling loop, which was used to inject 2 mL of

the sample into the column for analysis. The duplicate samples were analyzed for three VSC gases (hydrogen sulfide, methyl mercaptan, and dimethyl sulfide) commonly found in mouth air.⁸

Statistical Methods

For each product, each subject had baseline and overnight breath VSC levels recorded. For all products, paired *t* tests were performed to compare overnight breath VSC levels to baseline VSC levels. Analysis of covariance (ANCOVA) was performed on the overnight results, with subject and products as effects and baseline as the covariate. Treatments were declared statistically and significantly different if $P \leq .05$.

Results

Eighteen adult subjects qualified for the study and 16 subjects completed it. The age and gender characteristics of the study participants are presented in Table 2. Sixteen subjects showed satisfactory signs of compliance to the instructions. Two subjects were removed from the study for lack of compliance with the study instructions. There was no reported irritation or other side effects after using any of the toothbrushes. However, subjects reported that the Colgate® 360™ toothbrush provided invigorating stimulation of the cheeks and lips.

Baseline

Table 3 presents a summary of the baseline breath VSC levels for the four treatment groups. The mean breath VSC level for groups using the Colgate® 360™, Oral-B® Indicator®, Crest® SpinBrush™ PRO, and Oral-B® CrossAction® toothbrushes were 719.8 ppb, 592.8 ppb, 673.8 ppb, and 656.2 ppb, respectively. These levels of VSC were in the range corresponding to

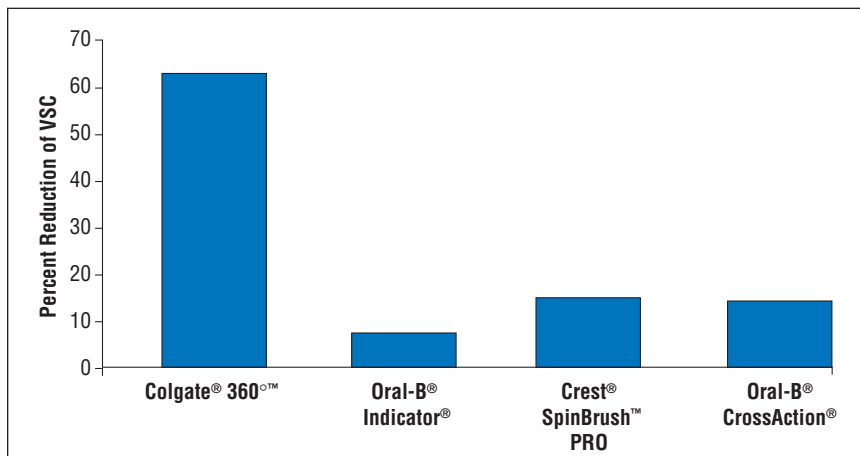


Figure 1—Percent reduction of morning breath VSC after using the four test toothbrushes. VSC = volatile sulfur compounds.

Table 3—Mean Pretreatment Baseline Breath VSC Scores of Study Participants

Toothbrush	n	Baseline VSC Levels (ppb)—Mean ± SD	Significance*
Colgate® 360™	16	719.8 ± 318.4	NS
Oral-B® Indicator®	16	592.8 ± 264.6	NS
Crest® SpinBrush™ PRO	16	673.8 ± 405.9	NS
Oral-B® CrossAction®	16	656.2 ± 310.2	NS

*Significance of analysis of variance comparison of baseline means. NS = $P \geq .05$.
VSC = volatile sulfur compounds; ppb = parts per billion; SD = standard deviation; NS = not significant.

moderate to overwhelming mouth odor, as indicated by the benchmark reported by Hunter et al.¹⁴ No statistically significant difference was observed between the breath VSC levels associated with the 4 study groups at the pretreatment baseline ($P > .05$).

Overnight Data

Subjects used the 4 toothbrushes in a sequential manner with at least a 2-day washout period between the use of each toothbrush. Toothbrushes were used as specified by the manufacturers. A summary of the breath VSC levels for the 4 toothbrush groups measured at the overnight posttreatment time point is presented in Table 4 and Figure 1. The final mean VSC level for the Colgate® 360™ toothbrush was 266.5 ppb, representing a 63% reduction in breath malodor compared to baseline. The change in overnight breath VSC level after using the Colgate® 360™ toothbrush was significantly different from that measured at baseline. The use of the Oral-B® Indicator® toothbrush resulted in a final mean VSC level of 545.2 ppb, representing an 8.1% reduction compared to baseline. This change was not significantly different from the reading at baseline. For the Crest® SpinBrush™ PRO, the final mean overnight breath VSC level was 567 ppb, representing a reduction of 15.8% compared to baseline. This change in the level of overnight breath VSC was not statistically significantly different from that measured at baseline. After using

the Oral-B® CrossAction® toothbrush, panelists had a final mean VSC level of 554.6 ppb, representing a 15.5% reduction in breath malodor compared to baseline. This change in overnight breath VSC level was not statistically significantly different from that measured at baseline. Therefore, the Colgate® 360™ toothbrush was the only toothbrush to provide statistically significant reduction in overnight breath VSC compared to baseline. Additionally, when the overnight end points for the 4 toothbrush groups were compared, the ANCOVA analysis showed that the Colgate® 360™ toothbrush was statistically significantly better ($P < .05$) than the Oral-B® Indicator®, Crest® SpinBrush™ PRO, and Oral-B® CrossAction® toothbrushes in promoting nonoffensive breath odor (Table 5).

Discussion

Poor oral hygiene can lead to various oral infections, such as caries and periodontal diseases, as well as socially embarrassing oral conditions, such as oral malodor. The central focus of good oral hygiene has been brushing teeth using, for example, a toothbrush and a toothpaste containing fluoride. Over the years, manufacturers have developed various toothbrushes that have been documented to help in the reduction of dental plaque, gingivitis, and tooth staining.^{15,19} The Colgate® 360™, a multiple-feature toothbrush, has tightly packed and tapered bristles for subgingival and interdental cleaning, a raised tip

Table 4—Mean Overnight Posttreatment VSC Scores After Brushing With the Four Toothbrushes

Toothbrush	n	Overnight VSC Levels (ppb) Mean ± SD	Percent Reduction from Baseline*	Significance Compared to Baseline†
Colgate® 360™	16	266.5 ± 269.9	63.0	$P < .01$
Oral-B® Indicator®	16	545.2 ± 346.1	8.1	$P = .1489$
Crest® SpinBrush™ PRO	16	567.0 ± 335.7	15.8	$P = .2341$
Oral-B® CrossAction®	16	554.6 ± 398.4	15.5	$P = .1786$

*Within-treatment percent reduction between baseline and postbaseline levels, expressed as a percentage of the baseline levels.
†Significance of analysis of covariance comparison of baseline-adjusted mean breath VSC levels.
VSC = volatile sulfur compounds; ppb = parts per billion; SD = standard deviation.

Table 5—Statistical Comparison of the Colgate® 360™ Toothbrush to the Commercially Available Toothbrushes

	Statistical Significance* vs Oral-B® Indicator®	Statistical Significance* vs Crest® SpinBrush™ PRO	Statistical Significance* vs Oral-B® CrossAction®
Colgate® 360™	$P < .0321$	$P < .0145$	$P < .0227$

*Significance of analysis of covariance comparison of baseline-adjusted mean breath volatile sulfur compound levels.

for cleaning hard-to-reach places in the mouth, and soft dental-like polishing cups for the delicate removal of tooth stains. Additionally, a special feature of the toothbrush is the soft conical-shaped nubs on the back of the toothbrush head for cleaning the tongue, cheeks, and lips. Cleaning of the cheeks and lips is done almost by accident because of the design of the toothbrush. This toothbrush has been documented to reduce plaque and gingivitis,^{20,21} decrease the levels of desquamated epithelial cells from the cheeks and lips,²² as well as remove hydrogen-sulfide-forming bacteria on the tongue.²³

Previously, Hunter et al¹⁴ showed that breath VSC levels above 400 ppb corresponded to subjects with moderate to overwhelming breath odor, while breath VSC levels below 300 ppb corresponded to subjects having none to slight oral malodor. The baseline mean levels of breath VSC for all four treatment groups were in the range corresponding to moderate to overwhelming breath odor. However, only the group using the Colgate® 360™ toothbrush had a mean morning breath odor level that was in the range corresponding to none to slight malodor. These results indicate that the Colgate® 360™ toothbrush was effective in promoting the reduction of overnight breath VSC associated with morning bad breath.

Conclusion

A randomized, crossover clinical study to evaluate the ability of the multiple-feature Colgate® 360™ toothbrush compared with three other commercially available toothbrushes (Oral-B® Indicator®, the battery-operated Crest® SpinBrush™ PRO, and Oral-B® CrossAction®) to promote the overnight reduction of VSC associated with bad breath has been completed. Subjects using the Colgate® 360™ toothbrush were observed to have a statistically significantly lower mean level of breath VSC compared to baseline pretreatment levels. In contrast, the mean levels of breath VSC for the groups using the three commercial toothbrushes were not statistically significantly different from VSC levels at baseline. More importantly, the subjects using the Colgate® 360™ toothbrush provided statistically

significantly lower levels of breath VSC compared to subjects using the three commercially available toothbrushes. Because all of the subjects in the study used the same toothpaste, the effect observed on breath VSC was likely because of the cleaning of the tongue with the back of the new toothbrush and the reduction of the desquamated epithelial cells.²² These cells contain proteins that are rich in sulfur-containing amino acids and that accumulate in tongue biofilm and dental plaque—sites of oral malodor production.^{8,24}

Although these results and other findings elsewhere support tongue cleaning or a combination of tooth and tongue brushing as a way to help alleviate oral malodor, consumers do not readily perform these procedures as part of their daily home oral hygiene procedures. The new high-performance, multiple-feature Colgate® 360™ toothbrush is likely to encourage consumers to brush their teeth and clean their tongue regularly, thus helping to improve their overall oral condition.

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Comparison of the Clinical Efficacy of a New Manual Toothbrush on Gingivitis Reduction and Plaque Removal

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Abstract: *The objective of this controlled, examiner-blind, 4-week clinical study was to evaluate and compare the safety and efficacy of a newly designed manual toothbrush, the Colgate® 360™ toothbrush, to the Oral-B® Indicator® toothbrush for the control of supragingival plaque and gingivitis. A total of 82 subjects from the northern New Jersey area reported to the clinical facility for a baseline plaque and gingivitis examination after having refrained from all oral hygiene procedures for 12 hours and from eating, drinking, or smoking for 4 hours. The population was comprised of healthy adult men and women 30 to 68 years of age. After the baseline examinations, qualifying subjects were randomized into two groups and assigned to one of the two test toothbrushes. All subjects were instructed to brush their teeth for 1 minute under supervision, after which they were again examined for supragingival plaque. They were then instructed to brush their teeth twice a day for 1 minute with their assigned toothbrush and a commercially available toothpaste (Colgate® Cavity Protection Great Regular Flavor Fluoride Toothpaste) for the next 4 weeks. After 4 weeks, subjects returned to the clinical facility for a final gingivitis and plaque examination. Eighty-one subjects complied with the protocol and completed the 4-week clinical study. The results of the study indicated that the new manual toothbrush was statistically significantly effective in reducing gingivitis after 4 weeks and in removing plaque after a single toothbrushing and after 4 weeks of use. Also, the new manual toothbrush exhibited a statistically significant greater reduction in gingivitis and in gingivitis-related bleeding sites after 4 weeks of use as well as statistically significant greater plaque removal after a single toothbrushing and after 4 weeks of use, as compared to the Oral-B® Indicator® toothbrush. This superior plaque-removal performance was found in separate analyses of the whole mouth, at interproximal surfaces, and at the gumline.*

The central role of dental plaque in the development of gingivitis and caries is well established.¹ The American Dental Hygienists' Association estimates that 70% of Americans have some form of periodontal disease and that only 50% are receiving regular oral care.² Regular and complete plaque removal is the best way to prevent these diseases, and toothbrushing is the most widely used and accepted form of daily oral hygiene. Although there is conflicting evidence concerning the comparative efficacy of powered and manual toothbrushes,³ the majority of toothbrushes sold on the market today are manual toothbrushes. Mechanical removal of plaque depends on many factors and is, therefore, variable and inconsistent in the general population. Thus, the design of toothbrushes is important for facilitating proper brushing technique and encouraging regular toothbrush-

ing, and manufacturers have expended considerable resources in developing toothbrushes that combine ergonomic design and cleaning efficacy.

The objective of this controlled, examiner-blind, 4-week clinical study was to compare the safety and efficacy of a newly designed manual toothbrush, the Colgate® 360™ toothbrush^a (Figure 1), with that of the commercially available Oral-B® Indicator® toothbrush^b for the control of supragingival plaque and gingivitis. The study was approved by an accredited institutional review board.

Materials and Methods

Adult men and women from the northern New Jersey area were enrolled into the study based on the following criteria:

- Subjects had to be between the ages of 18 and 70, in generally good health, with no history of allergies to dentifrice ingredients, and possess a minimum of 20 scoreable teeth;
- Subjects needed to be available for the duration of the study, and were required to sign an informed consent form;
- Subjects were required to present, at the pre-brushing examination, a mean Rustogi Modification of the Navy Plaque Index score of 0.6 or greater;
- Subjects were required to present, at the pre-brushing examination, a mean Löe-Silness



Figure 1—The Colgate® 360™ toothbrush.

Gingival Index score of 1.0 or greater.

- Subjects were excluded from the study if they had orthodontic appliances or removable prostheses, tumors of the soft and hard oral tissues, advanced periodontal disease, 5 or more carious lesions requiring restorative treatment, had participated in any other clinical study or test panel within 4 weeks before entry into the study, had received a dental prophylaxis within the 2 weeks before entry into the study, or if they had received antibiotic therapy during the 1-month period before entry into the study. Pregnant or lactating women also were excluded.

Before the baseline examination, potential study subjects were required to refrain from any oral hygiene procedures for 12 hours, and to refrain from eating, drinking, or smoking for 4 hours.

After baseline (prebrushing) screenings for

^aThe Colgate-Palmolive Company, New York, NY 10022; 800-338-8388
^bOral-B Laboratories, Belmont, CA 94002; 800-44ORALB

Gingivitis Index Calculation

$$\frac{\text{Sum of all scores for all surfaces}}{\text{Total number of surfaces scored}} = \frac{\quad}{\quad} = \boxed{\quad} \text{ Gingivitis Index per Surface}$$

The Löe-Silness Gingival Index criteria are as follows:

0 = Absence of inflammation.

1 = Mild inflammation: slight change in color and little change in texture.

2 = Moderate inflammation: moderate glazing, redness, edema, and hypertrophy; tendency to bleed on probing.

3 = Severe inflammation: marked redness and hypertrophy; tendency to bleed spontaneously.

Tooth	32	31	
Facial Surface Score	X		⇐ Mesial Gingivitis Score
			⇐ Middle Gingivitis Score
			⇐ Distal Gingivitis Score

Figure 2—Gingivitis index calculation using the Löe-Silness Gingival Index criteria.

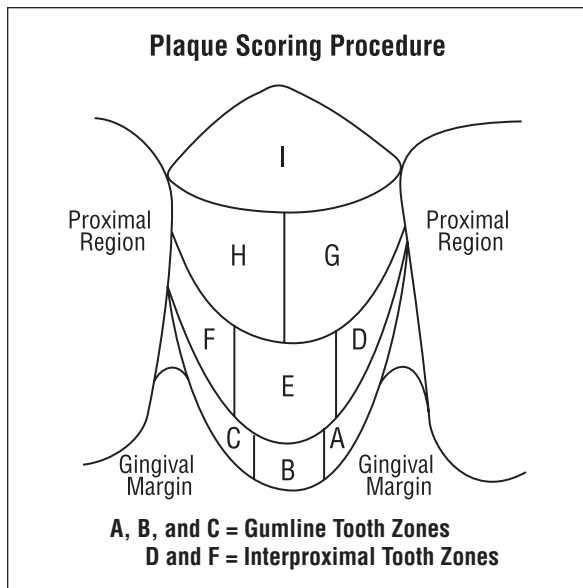


Figure 3—Plaque scoring procedure using the Rustogi Modification of the Navy Plaque Index.

plaque and gingivitis, qualifying subjects were randomized into two balanced groups and then randomly assigned to one of the test toothbrushes (Colgate® 360™ toothbrush or Oral-B® Indicator® toothbrush) and a commercially available toothpaste (Colgate® Cavity Protection Great Regular Flavor Fluoride Toothpaste^a). They were instructed to brush their teeth for 1 minute under supervision in their usual manner. Subjects were again examined for supragingival plaque (postbrushing). All subjects were then instructed to brush their teeth with the toothbrush and toothpaste twice a day (mornings and evenings) for 1 minute for the next 4 weeks, after which they were once again evaluated for gingivitis and supragingival plaque.

Clinical Scoring Procedures

Gingivitis

Gingivitis was scored according to the Löe-Silness Gingival Index as modified by Talbott et al^{4,5} (Figure 2). Each tooth was scored in six areas—mesiofacial, midfacial, distofacial, mesiolingual, midlingual, and distolingual—as follows:

0 = Absence of inflammation.

1 = Mild inflammation: slight change in color and little change in texture.

2 = Moderate inflammation: moderate glazing, redness, edema, hypertrophy; tendency to bleed on probing.

3 = Severe inflammation: marked redness and hypertrophy; tendency to bleed spontaneously.

Third molars and teeth with cervical restorations or prosthetic crowns were excluded from the scoring procedure. Whole-mouth mean scores were obtained by averaging the values recorded from all scoreable tooth surfaces.

Gingivitis Severity

In addition to calculating a mean Löe-Silness Gingival Index score for each subject, a mean Gingivitis Severity Index⁶ also was calculated for each subject. This index allows for a comparison of the gingival sites that received the highest possible Löe-Silness Gingival Index scores, ie, scores of 2 or 3 (bleeding sites). The mean Gingivitis Severity Index was calculated for each subject by dividing the total number of gingival sites scored 2 or 3 by the total number of teeth scored in the mouth.

Supragingival Dental Plaque

Supragingival plaque was scored according to the Rustogi Modification of the Navy Plaque Index.⁷ Supragingival plaque was disclosed and recorded as present or absent on 9 different areas of the tooth (Figure 3). Third molars were excluded from the scoring procedure. From these sitewise scores, a score was determined for each subject by calculating the proportion of sites in the mouth at which plaque was present. Three parameters were evaluated as follows:

Whole-mouth scores—A, B, C, D, E, F, G, H, I

Gumline scores—A, B, C

Interproximal scores—D, F

Dr. Mankodi has been calibrated and well trained and is highly experienced with regard to the clinical scoring procedures used in this study.

Statistical Methods

Statistical analyses were performed separately for the gingival index and the plaque index. Comparisons of the treatment groups with respect to baseline scores were performed using analyses of variance.

Table 1—Summary of Gender and Age Characteristics for Subjects Who Completed the 4-Week Clinical Study

Treatment	Men	Women	Total	Mean Age	Range
Colgate® 360™	13	26	39	48.5	30–66
Oral-B® Indicator®	5	37	42	48.2	30–68

Table 2—Summary of the Baseline Gingival Index Scores for Subjects Who Completed the 4-Week Clinical Study

Parameter	Treatment	n	Baseline Summary*	Significance [†]
Gingival Index	Colgate® 360™	39	1.071 ± 0.047	NS
	Oral-B® Indicator®	42	1.080 ± 0.064	
Gingivitis Severity Index	Colgate® 360™	39	0.091 ± 0.079	NS
	Oral-B® Indicator®	42	0.086 ± 0.063	

*Mean ± standard deviation.
[†]Significance of analysis of variance comparison of baseline mean scores. NS = $P > .05$.
 NS = not significant.

For plaque index scores, within-treatment comparisons of the pre- vs postbrushing scores were performed using paired *t* tests. Comparisons of treatment groups with respect to pre- to postbrushing reductions were performed using analysis of covariance (ANCOVA), in which the prebrushing scores were employed as a covariable.

For gingival index and plaque index scores, within-treatment comparisons of the scores obtained at the 4-week examination vs baseline were performed using paired *t* tests. Comparisons of the treatment groups with respect to baseline-adjusted scores at the 4-week examination were performed using ANCOVA.

All statistical tests of hypotheses were 2-sided and employed a level of significance of $\alpha = 0.05$.

Results

Eighty-one subjects completed the study. A summary of the age and gender of the study population is presented in Table 1. Throughout the study, no adverse effects on the oral hard or soft tissues were observed by the dental examiner or reported by the participants when questioned.

Gingivitis Reduction—Overall

Tables 2 and 3 and Figure 4 present a comparison of the Löe-Silness Gingival Index scores at baseline and after 4 weeks of toothbrush use for each toothbrush group. The baseline (prebrushing) mean gingival index scores for the Colgate® 360™ toothbrush group and the Oral-B® Indicator® toothbrush group were 1.071 and 1.080, respectively, indicating no statistically significant difference between gingival index scores for the 2 toothbrush groups.

After 4 weeks of toothbrush use, the mean gingival index score for subjects who used the new Colgate® 360™ toothbrush was 0.964, indicating a statistically significant 10% reduction in gingival index scores from baseline ($P < .05$). The mean gingival index score for subjects who used the Oral-B® Indicator® toothbrush was 1.070, indicating no statistically significant difference in gingival index scores from baseline ($P > .05$).

A comparison of the mean gingival index scores of the 2 toothbrush groups showed that the Colgate® 360™ toothbrush provided a statistically significant 9.9% greater reduction in gingivitis

Table 3—Summary of the 4-Week Gingival Index Scores for Subjects Who Completed the 4-Week Clinical Study

Parameter	Treatment	n	4-week Summary*	Within-treatment Analysis		Between-treatment Analysis	
				Percent Reduction [†]	Significance [‡]	Percent Difference [§]	Significance
Gingival Index	Colgate® 360™	39	0.964 ± 0.067	10.0	$P < .05$	9.9	$P < .05$
	Oral-B® Indicator®	42	1.070 ± 0.058	0.9	NS		
Gingivitis Severity Index	Colgate® 360™	39	0.030 ± 0.029	67.0	$P < .05$	72.0	$P < .05$
	Oral-B® Indicator®	42	0.107 ± 0.141	-24.4	NS		

*Mean ± standard deviation.
[†]Reduction between the baseline and 4-week mean scores, expressed as a percentage of the baseline mean score. Positive values indicate lower gingivitis scores at the 4-week examination than at the baseline examination.
[‡]Significance of paired *t* test comparing the baseline and 4-week examinations. NS = $P > .05$.
[§]Difference between the 4-week mean scores, expressed as a percentage of the 4-week mean score for the Oral-B® Indicator®. Positive values indicate lower gingivitis scores for the Colgate® 360™.
^{||}Significance of postanalysis of covariance comparison of baseline-adjusted mean scores.
 NS = not significant.

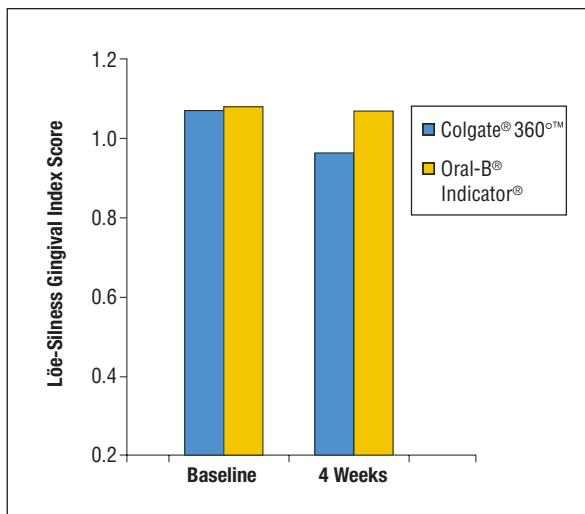


Figure 4—Overall gingivitis scores.

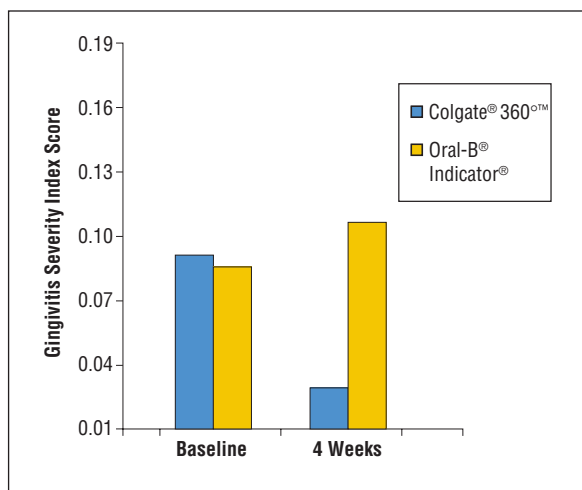


Figure 5—Gingivitis severity scores.

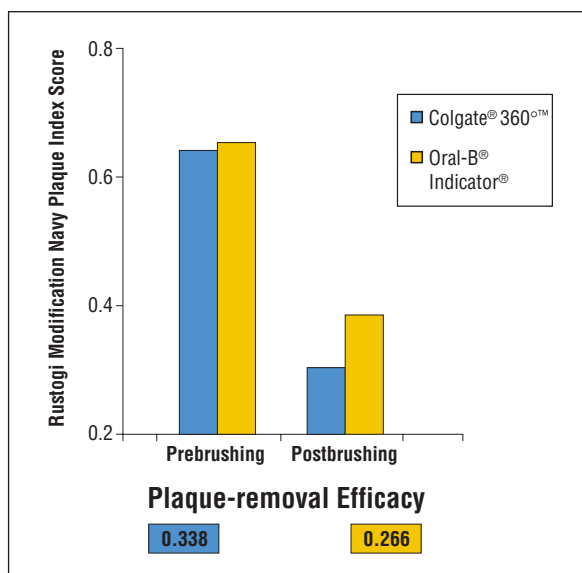


Figure 6—Whole-mouth mean plaque index scores and comparative plaque-removal efficacy.

as compared to the Oral-B® Indicator® toothbrush ($P < .05$) after 4 weeks of daily use.

Gingivitis Severity

Tables 2 and 3 and Figure 5 present a comparison of the Gingivitis Severity Index scores at baseline and after 4 weeks of daily use. The baseline (prebrushing) mean Gingivitis Severity Index scores for the Colgate® 360™ toothbrush group and the Oral-B® Indicator® toothbrush group were 0.091 and 0.086, respectively, indicating no statistically significant difference between Gingivitis Severity Index scores for the 2 toothbrush groups.

After 4 weeks of toothbrush use, the mean Gingivitis Severity Index score for subjects who used the new Colgate® 360™ toothbrush was 0.030, indicating a statistically significant 67% reduction in Gingivitis Severity Index scores from baseline ($P < .05$). The mean Gingivitis Severity Index score for subjects who used the Oral-B® Indicator® toothbrush was 0.107, indicating no statistically significant difference in Gingivitis Severity Index scores from baseline ($P > .05$).

A comparison of the mean Gingivitis Severity Index scores of the 2 toothbrush groups showed that the Colgate® 360™ toothbrush provided a 72% greater reduction in gingivitis-related bleeding sites as compared to the Oral-B® Indicator® toothbrush ($P < .05$) after 4 weeks of daily use.

Removal of Supragingival Plaque Whole Mouth—After Single Toothbrushing

Table 4 and Figure 6 present a comparison of the whole-mouth Rustogi Modification of the Navy Plaque Index scores at baseline (prebrushing scores), after 1 minute of toothbrushing (postbrushing scores), as well as the differences between the 2 scores. The baseline (prebrushing) mean plaque index scores for the Colgate® 360™ toothbrush group and the Oral-B® Indicator® toothbrush group were 0.641 and 0.652, respectively, indicating no statistically significant difference between plaque index scores for the 2 toothbrush groups.

The postbrushing (after 1 minute of toothbrushing) mean plaque index score for subjects who used the new Colgate® 360™ toothbrush was 0.303, indicating a difference in plaque index scores of 0.338 ($P < .05$) and a 52.7% reduction in plaque from prebrushing (baseline). The mean postbrushing plaque index score for subjects who used the Oral-B® Indicator® toothbrush was 0.386, indicating a difference in plaque index scores of 0.266 ($P < .05$) and a 40.8% reduction in plaque

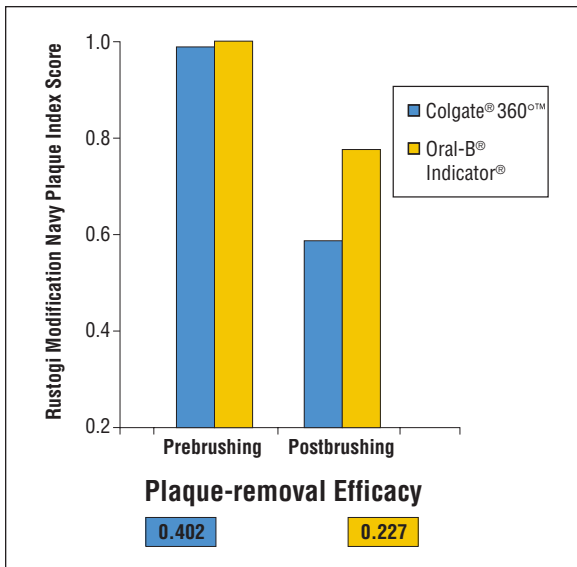


Figure 7—Interproximal mean plaque index scores and comparative plaque-removal efficacy.

from prebrushing (baseline).

These differences indicate that after a single, 1-minute toothbrushing, the new Colgate® 360™ toothbrush removed statistically significantly 27.1% more plaque as compared to the Oral-B® Indicator® toothbrush.

Interproximal Surfaces—After Single Toothbrushing

Table 4 and Figure 7 present a comparison of the interproximal Rustogi Modification of the

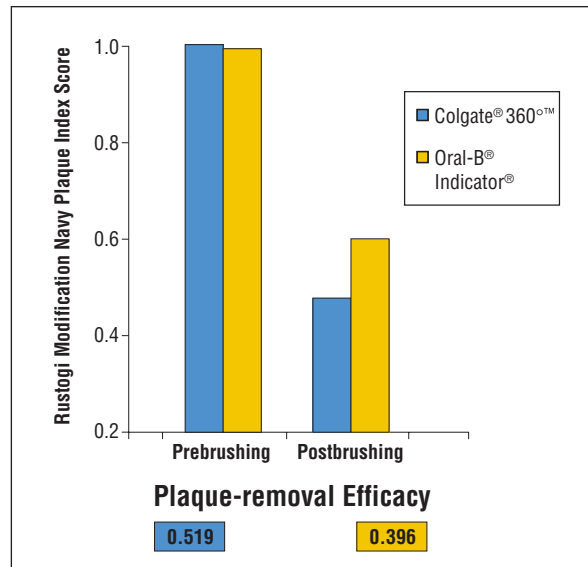


Figure 8—Gumline mean plaque index scores and comparative plaque-removal efficacy.

Navy Plaque Index scores at baseline (prebrushing scores), after 1 minute of toothbrushing (postbrushing scores), as well as the differences between the 2 scores. The baseline (prebrushing) mean plaque index scores for the Colgate® 360™ toothbrush group and the Oral-B® Indicator® toothbrush group were 0.986 and 0.998, respectively, indicating no statistically significant difference between plaque index scores for the 2 toothbrush groups.

The postbrushing (after 1 minute of toothbrushing) mean plaque index score for subjects

Table 4—Summary of the Pre- and Postbrushing Plaque Index Scores for Subjects Examined After a Single Toothbrushing

Parameter	Treatment	n	Prebrushing Summary*	Postbrushing Summary*	Pre- to Postbrushing Reductions in Plaque Index Between-treatment Analysis			
					Difference Mean ± SD	Percent Reduction†	Percent Difference‡	Significance§
Whole-mouth Plaque Index	Colgate® 360™	39	0.641 ± 0.032	0.303 ± 0.090	0.338 ± 0.084	52.7	27.1	P < .05
	Oral-B® Indicator®	42	0.652 ± 0.050	0.386 ± 0.066	0.266 ± 0.068	40.8		
Plaque Index on Interproximal Sites	Colgate® 360™	39	0.986 ± 0.044	0.584 ± 0.221	0.402 ± 0.218	40.8	77.1	P < .05
	Oral-B® Indicator®	42	0.998 ± 0.008	0.771 ± 0.194	0.227 ± 0.195	22.7		
Plaque Index on Gumline Sites	Colgate® 360™	39	0.998 ± 0.012	0.479 ± 0.180	0.519 ± 0.178	52.0	31.1	P < .05
	Oral-B® Indicator®	42	0.996 ± 0.015	0.600 ± 0.144	0.396 ± 0.148	39.8		

*Mean ± standard deviation.

†Reduction between the prebrushing and postbrushing scores, expressed as a percentage of the prebrushing score.

‡Difference between the pre- and postbrushing reductions in plaque, expressed as a percentage of the reduction for the Oral-B® Indicator®. Positive values indicate greater plaque reductions for the Colgate® 360™.

§Significance of postanalysis of covariance comparison of mean plaque reductions, adjusted for prebrushing plaque scores.

||Statistically significant reduction from pre- to postbrushing measurements.

SD = standard deviation.

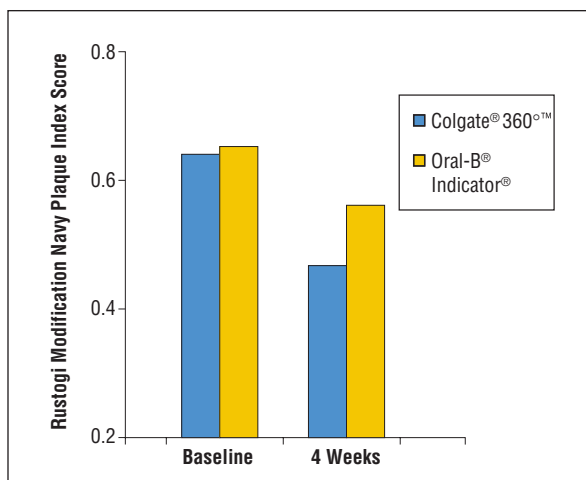


Figure 9—Whole-mouth mean plaque index scores at 4 weeks.

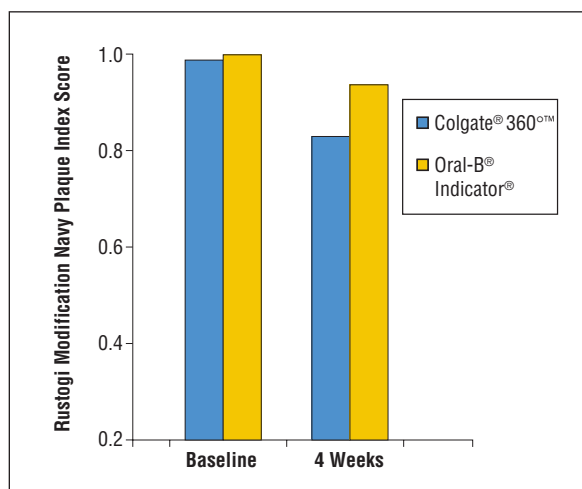


Figure 10—Interproximal mean plaque index scores at 4 weeks.

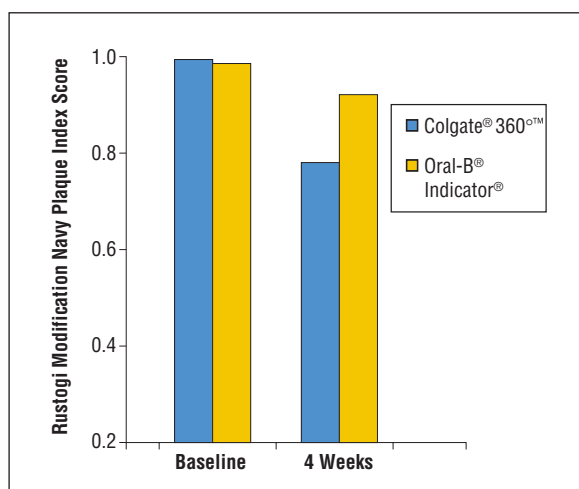


Figure 11—Gumline mean plaque index scores at 4 weeks.

who used the new Colgate® 360™ toothbrush was 0.584, indicating a difference in plaque index scores of 0.402 ($P < .05$) and a 40.8% reduction in plaque from prebrushing (baseline). The mean plaque index score for subjects who used the Oral-B® Indicator® toothbrush was 0.771, indicat-

ing a difference in plaque index scores of 0.227 ($P < .05$) and a 22.7% reduction in plaque from prebrushing (baseline).

These differences indicate that after a single, 1-minute toothbrushing, the new Colgate® 360™ toothbrush removed statistically significantly 77.1% more plaque at interproximal sites as compared to the Oral-B® Indicator® toothbrush.

Gumline Surfaces—After Single Toothbrushing

Table 4 and Figure 8 present a comparison of the gumline Rustogi Modification of the Navy Plaque Index scores at baseline (prebrushing scores), after 1 minute of toothbrushing (postbrushing scores), as well as the differences between the 2 scores. The baseline (prebrushing) mean plaque index scores for the Colgate® 360™ toothbrush group and the Oral-B® Indicator® toothbrush group were 0.998 and 0.996, respectively, indicating no statistically significant difference between plaque index scores for the 2 toothbrush groups.

The postbrushing (after 1 minute of toothbrushing) mean plaque index score for subjects who used the new Colgate® 360™ toothbrush was 0.479, indicating a difference in plaque index scores of 0.519 ($P < .05$) and a 52% reduction in plaque from prebrushing (baseline). The mean plaque index score for subjects who used the Oral-B® Indicator® toothbrush was 0.600, indicating a difference in plaque index scores of 0.396 ($P < .05$) and a 39.8% reduction in plaque from prebrushing (baseline).

These differences indicate that after a single, 1-minute toothbrushing, the new Colgate® 360™ toothbrush removed statistically significantly 31.1% more plaque at the gumline as compared to the Oral-B® Indicator® toothbrush.

Whole Mouth—After 4 Weeks

Tables 5 and 6 and Figure 9 present a comparison of the whole-mouth Rustogi Modification of the Navy Plaque Index scores at baseline (prebrushing scores) and after 4 weeks. The baseline (prebrushing) mean plaque index scores for the Colgate® 360™ toothbrush group and the Oral-B® Indicator® toothbrush group were 0.641 and 0.652, respectively, indicating no statistically significant difference between plaque index scores for the 2 toothbrush groups.

The 4-week mean plaque index score for subjects who used the new Colgate® 360™ toothbrush was 0.466, while the 4-week mean plaque

index score for subjects who used the Oral-B® Indicator® toothbrush was 0.558, indicating statistically significantly 16.5% ($P < .05$) greater plaque removal for the Colgate® 360°™ toothbrush.

This difference indicates that after 4 weeks, the new Colgate® 360°™ toothbrush was statistically significantly more effective in removing plaque as compared to the Oral-B® Indicator® toothbrush.

Interproximal Surfaces—After 4 Weeks

Tables 5 and 6 and Figure 10 present a comparison of the interproximal Rustogi Modification of the Navy Plaque Index scores at baseline (pre-brushing scores) and after 4 weeks. The baseline (prebrushing) mean plaque index scores for the Colgate® 360°™ toothbrush group and the Oral-B® Indicator® toothbrush group were 0.986 and 0.998, respectively, indicating no statistically significant difference between plaque index scores for the 2 toothbrush groups.

The 4-week mean plaque index score for sub-

jects who used the new Colgate® 360°™ toothbrush was 0.828 and the 4-week mean plaque index score for subjects who used the Oral-B® Indicator® toothbrush was 0.934, indicating statistically significantly 11.3% ($P < .05$) greater plaque removal at interproximal sites for the Colgate® 360°™ toothbrush.

This difference indicates that after 4 weeks, the new Colgate® 360°™ toothbrush was statistically significantly more effective in removing plaque at interproximal sites as compared to the Oral-B® Indicator® toothbrush.

Gumline—After 4 Weeks

Tables 5 and 6 and Figure 11 present a comparison of the gumline Rustogi Modification of the Navy Plaque Index scores at baseline (pre-brushing scores) and after 4 weeks. The baseline (prebrushing) mean plaque index scores for the Colgate® 360°™ toothbrush group and the Oral-B® Indicator® toothbrush group were 0.998

Table 5—Summary of the Baseline Prebrushing Plaque Index Scores for Subjects Who Completed the 4-Week Clinical Study

Parameter	Treatment	n	Baseline Summary*	Significance†
Whole-mouth Plaque Index	Colgate® 360°™	39	0.641 ± 0.032	NS
	Oral-B® Indicator®	42	0.652 ± 0.050	
Plaque Index on Interproximal Sites	Colgate® 360°™	39	0.986 ± 0.044	NS
	Oral-B® Indicator®	42	0.998 ± 0.008	
Plaque Index on Gumline Sites	Colgate® 360°™	39	0.998 ± 0.012	NS
	Oral-B® Indicator®	42	0.996 ± 0.015	

*Mean ± standard deviation.

†Significance of analysis of variance comparison of baseline mean scores. NS = $P > .05$.

NS = not significant.

Table 6—Summary of the 4-Week Plaque Index Scores for Subjects Who Completed the 4-Week Clinical Study

Parameter	Treatment	n	4-Week Summary*	Within-treatment Analysis		Between-treatment Analysis	
				Percent Reduction†	Significance‡	Percent Difference§	Significance
Whole-mouth Plaque Index	Colgate® 360°™	39	0.466 ± 0.078	27.3	$P < .05$	16.5	$P < .05$
	Oral-B® Indicator®	42	0.558 ± 0.071	14.4	$P < .05$		
Plaque Index on Interproximal Sites	Colgate® 360°™	39	0.828 ± 0.190	16.0	$P < .05$	11.3	$P < .05$
	Oral-B® Indicator®	42	0.934 ± 0.096	6.4	$P < .05$		
Plaque Index on Gumline Sites	Colgate® 360°™	39	0.781 ± 0.137	21.7	$P < .05$	15.4	$P < .05$
	Oral-B® Indicator®	42	0.923 ± 0.076	7.3	$P < .05$		

*Mean ± standard deviation.

†Reduction between the baseline and 4-week mean scores, expressed as a percentage of the baseline mean. Positive values indicate lower plaque scores at the 4-week examination than at the baseline examination.

‡Significance of paired t test comparing the baseline and 4-week examinations.

§Difference between the 4-week mean scores, expressed as a percentage of the 4-week mean score for the Oral-B® Indicator®. Positive values indicate lower plaque scores for the Colgate® 360°™.

||Significance of postanalysis of covariance comparison of baseline-adjusted means.

and 0.996, respectively, indicating no statistically significant difference between plaque index scores for the 2 toothbrush groups.

The 4-week mean plaque index score for subjects who used the new Colgate® 360°™ toothbrush was 0.781 and the 4-week mean plaque index score for subjects who used the Oral-B® Indicator® toothbrush was 0.923, indicating statistically significantly 15.4% ($P < .05$) greater plaque removal for the Colgate® 360°™ toothbrush.

This difference indicates that after 4 weeks, the new Colgate® 360°™ toothbrush was statistically significantly more effective in removing plaque at the gumline as compared to the Oral-B® Indicator® toothbrush.

Discussion

Mechanical plaque removal with a manual toothbrush remains the primary method of maintaining good oral hygiene for the majority of the population. Although efficient brushing technique is a major determinant of plaque reduction, product design also can influence plaque removal. Manual toothbrushes differ in their ability to remove plaque from interproximal surfaces, and comprehensive plaque control requires use of other cleaning aids such as dental floss.⁸ Because of the generally low compliance with flossing instructions,⁹ a toothbrush designed to reach areas of plaque buildup should be beneficial to patients. The Colgate® 360°™ toothbrush contains tightly packed and tapered bristles that facilitate interproximal and gumline cleaning and a raised cleaning tip to access back teeth.

Our study shows that the Colgate® 360°™ toothbrush was more effective than the Oral-B® Indicator® toothbrush for both removing plaque and reducing gingivitis. Prebrushing data showed no statistically significant difference between baseline plaque index or gingivitis scores between the two groups. After a single use, both toothbrushes reduced plaque from prebrushing levels, but the reduction with the Colgate® 360°™ toothbrush was significantly greater than that with the Oral-B® Indicator® toothbrush for the whole mouth (52.7% vs 40.8%), interproximal area (40.8% vs 22.7%), and gumline sites (52% vs 39.8%). Similarly, after 4 weeks of use, although both toothbrushes reduced plaque at all sites, the Colgate® 360°™ toothbrush was more effective than the Oral-B® Indicator® toothbrush, and differences between the 2 brushes were significant at all sites (whole mouth: 27.3% vs 14.4%; interproximal sites: 16% vs 6.4%; gumline sites: 21.7%

vs 7.3%). The increased control of plaque with the Colgate® 360°™ toothbrush also translated into improved gingival health. Thus, 4 weeks of use of the Colgate® 360°™ toothbrush significantly reduced both the extent (10%) and the severity (67%) of gingivitis, whereas with the Oral-B® Indicator® toothbrush, the gingival index was lowered by only 0.9% and the severity of gingivitis increased by 24.4%.

Conclusion

Eighty-one subjects complied with the protocol and completed the 4-week, examiner-blind, clinical study. Under the conditions of this clinical study, the new Colgate® 360°™ toothbrush removed statistically significantly more plaque than the Oral-B® Indicator® toothbrush. Superior plaque-removal performance was found in separate analyses of whole-mouth, interproximal, and gumline areas. The results of the study also indicated that the new manual toothbrush, the Colgate® 360°™ toothbrush, also was statistically significantly more effective in reducing moderate and severe (bleeding sites) gingivitis compared with the Oral-B® Indicator® toothbrush after 4 weeks. In conclusion, in this 4-week study, the Colgate® 360°™ toothbrush proved superior to the Oral-B® Indicator® toothbrush in reducing plaque and reversing gingivitis. Based on its clinically proven superiority, dental practitioners can recommend the use of a clinically proven, superior manual toothbrush such as the Colgate® 360°™ toothbrush as a means of improving their patients' oral health.

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A Clinical Comparison of the Gingivitis Reduction and Plaque-removal Efficacy of a New Manual Toothbrush



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Abstract: *The objective of this controlled, examiner-blind, 4-week clinical study was to evaluate and compare the safety and efficacy of a newly designed manual toothbrush (Colgate® 360™ toothbrush) to the Oral-B® CrossAction® toothbrush for the control of supragingival plaque and gingivitis. A total of 80 subjects from the central New Jersey area reported to the clinical facility for a baseline plaque and gingivitis examination after having refrained from all oral hygiene procedures for 12 hours and from eating, drinking, or smoking for 4 hours. The population was comprised of healthy adult men and women 18 to 67 years of age. After the baseline examinations, qualifying subjects were randomized into two groups and assigned to one of the two test toothbrushes. All subjects were instructed to brush their teeth for 1 minute under supervision, after which they were again examined for supragingival plaque. They were then instructed to brush their teeth twice a day for 1 minute with their assigned toothbrush and a commercially available toothpaste (Colgate® Cavity Protection Great Regular Flavor Fluoride Toothpaste) for the next 4 weeks. After 4 weeks of use, subjects returned to the clinical facility for a final gingivitis and plaque examination. Seventy-eight subjects complied with the protocol and completed the 4-week clinical study. The results of the study indicated that the new manual toothbrush was statistically significantly effective in reducing gingivitis after 4 weeks and in removing plaque after a single toothbrushing and after 4 weeks of use. Also, the group using the new manual toothbrush exhibited a statistically significant greater reduction in plaque of up to 40% and no statistically significant difference in gingivitis reduction after 4 weeks of use, as compared to the Oral-B® CrossAction® toothbrush.*

Destructive periodontal disease is one of the most common diseases affecting people worldwide. Dental plaque plays a central role in the development of gingivitis, an inflammation of the gums that is the most common type of periodontal disease.¹ Plaque is essentially a biofilm that forms on hard tissue, primarily enamel; it is a complex blend of bacteria, extracellular matrix, and fluid channels that, when left undisturbed, grows and increases the risk of dental and gingival disease.² Entry of plaque products such as toxins and cell wall remnants into gingival tissue induces gingivitis. Mechanical debridement is currently the best way to break through the impermeable protective coating of biofilm; regular toothbrushing diminishes or eliminates plaque buildup and is associated with improved gingival health.^{1,3} Toothbrushing with toothpaste is the most common oral dental-hygiene habit in developed countries.⁴ Because of the importance of toothbrushing to oral health, good toothbrush design must consider not only plaque-removal efficacy but also ease of use to enhance compliance.

This article reports the results of a study on the efficacy of a new toothbrush designed to improve plaque removal in difficult areas such as interproximal sites, the gingival margin, and posterior areas of the mouth. The objective of this controlled, examiner-blind, 4-week clinical study was to compare the safety and efficacy of a newly designed manual toothbrush (Colgate® 360™ toothbrush^a) (Figure 1) to the commercially available Oral-B® CrossAction® toothbrush^b for the control of supragingival plaque and gingivitis. This study was approved by an accredited institutional review board.

Materials and Methods

Adult men and women from the central New Jersey area were enrolled into the study based on the following criteria:

- Subjects had to be between the ages of 18 and 70, in generally good health, with no history of allergies to dentifrice ingredients, and possess a minimum of 20 scoreable teeth.
- Subjects needed to be available for the duration of the study, and to sign an informed consent form.
- Subjects were required to present, at the prebrushing examination, a mean Rustogi Modification of the Navy Plaque Index score of 0.6 or greater.
- Subjects were required to present, at the pre-



Figure 1—The Colgate® 360™ toothbrush.

brushing examination, a mean Löe-Silness Gingival Index score of 1.0 or greater.

- Subjects were excluded from the study if they had orthodontic appliances or removable prostheses, tumors of the soft and hard oral tissues, advanced periodontal disease, 5 or more carious lesions requiring restorative treatment, had participated in any other clinical study or test panel within 4 weeks before entry into the study, had received antibiotic therapy during the 1-month period before entry into the study, or if they had received a dental prophylaxis within the 2 weeks before entry into the study. Pregnant or lactating women also were excluded.

Before the baseline examination, potential study subjects were required to refrain from any oral hygiene procedures for 12 hours as well as refrain from eating, drinking, or smoking for 4 hours. After baseline (prebrushing) screenings for gingivitis and plaque, qualifying subjects were

^aThe Colgate-Palmolive Company, New York, NY 10022; 800-338-8388

^bOral-B Laboratories, Belmont, CA 94002; 800-44ORALB

Gingivitis Index Calculation

$$\frac{\text{Sum of all scores for all surfaces}}{\text{Total number of surfaces scored}} = \frac{\quad}{\quad} = \boxed{\quad} \quad \text{Gingivitis Index per Surface}$$

The Löe-Silness Gingival Index criteria are as follows:

0 = Absence of inflammation.

1 = Mild inflammation: slight change in color and little change in texture.

2 = Moderate inflammation: moderate glazing, redness, edema, and hypertrophy; tendency to bleed on probing.

3 = Severe inflammation: marked redness and hypertrophy; tendency to bleed spontaneously.

Tooth	32	31	
Facial Surface Score	X		⇐ Mesial Gingivitis Score
			⇐ Middle Gingivitis Score
			⇐ Distal Gingivitis Score

Figure 2—Gingivitis index calculation using the Löe-Silness Gingival Index criteria.

randomized into two balanced groups and then randomly assigned to one of the test toothbrushes (Colgate® 360°™ toothbrush or Oral-B® CrossAction® toothbrush) and a commercially available toothpaste (Colgate® Cavity Protection Great Regular Flavor Fluoride Toothpaste^a). They were instructed to brush their teeth for 1 minute under supervision in their usual manner. Subjects were again examined for supragingival plaque (postbrushing). All subjects were then instructed to brush their teeth with the toothbrush and toothpaste twice a day (mornings and evenings) for 1 minute for the next 4 weeks, after which they were once again evaluated for gingivitis and supragingival plaque.

Clinical Scoring Procedures

Gingivitis

Gingivitis was scored according to the Löe-Silness Gingival Index as modified by Talbott and colleagues^{5,6} (Figure 2). Each tooth was scored in six areas—mesiofacial, midfacial, distofacial, mesiolingual, midlingual, and distolingual—as follows:

- 0 = Absence of inflammation.
- 1 = Mild inflammation: slight change in color and little change in texture.
- 2 = Moderate inflammation: moderate glazing, redness, edema, hypertrophy; tendency to bleed on probing.
- 3 = Severe inflammation: marked redness and hypertrophy; tendency to bleed spontaneously.

Third molars and teeth with cervical restorations or prosthetic crowns were excluded from the scoring procedure. Whole-mouth mean scores were obtained by averaging the values recorded from all scoreable tooth surfaces.

Gingivitis Severity

In addition to calculating a mean Löe-Silness Gingival Index score for each subject, a mean Gingivitis Severity Index⁷ also was calculated for each subject. This index allows for a comparison of the gingival sites that received the highest possible Löe-Silness Gingival Index scores, ie, scores of 2 or 3 (bleeding sites). The mean Gingivitis Severity Index was calculated for each subject by dividing the total number of gingival sites scored 2 or 3 by the total number of teeth scored in the mouth.

Supragingival Dental Plaque

Supragingival plaque was scored according to the Rustogi Modification of the Navy Plaque

Index.⁸ Supragingival plaque was disclosed and recorded as present or absent on 9 different areas of the tooth (Figure 3). Third molars were excluded from the scoring procedure. From these sitewise scores, a score was determined for each subject by calculating the proportion of sites in the mouth at which plaque was present. Three parameters were evaluated as follows:

Whole-mouth scores—A, B, C, D, E, F, G, H, I

Gumline scores—A, B, C

Interproximal scores—D, F

Dr. Nathoo has been calibrated and well trained and is highly experienced with regard to the clinical scoring procedures used in this study.

Statistical Methods

Statistical analyses were performed separately for the gingival index and the plaque index. Comparisons of the treatment groups with respect to baseline scores were performed using analyses of variance.

For plaque index scores, within-treatment comparisons of the pre- vs postbrushing scores were performed using paired *t* tests. Comparisons of treatment groups with respect to pre- to postbrushing reductions were performed using analysis of covariance (ANCOVA), in which the prebrushing scores were used as a covariable.

For gingival index and plaque index scores, within-treatment comparisons of the scores obtained at the 4-week examination vs baseline were performed using paired *t* tests. Comparisons of the treatment groups with respect to baseline-adjusted scores at the 4-week examination were performed using ANCOVA.

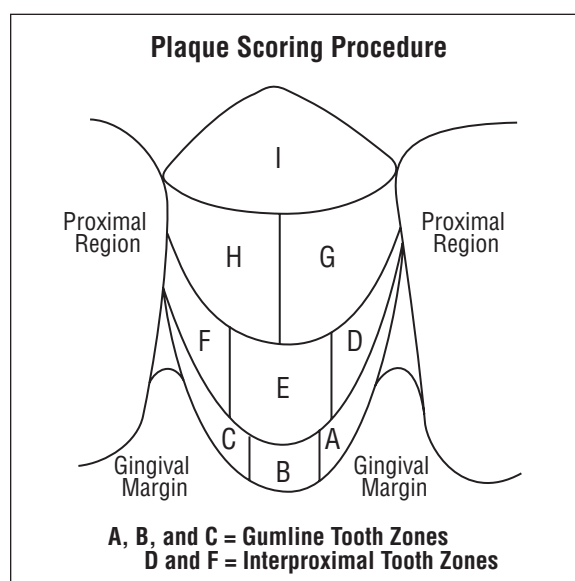


Figure 3—Plaque scoring procedure using the Rustogi Modification of the Navy Plaque Index.

Table 1—Summary of Gender and Age Characteristics for Subjects Who Completed the 4-Week Clinical Study

Treatment	Men	Women	Total	Mean Age	Range
Colgate® 360™	15	24	39	38.3	20–67
Oral-B® CrossAction®	16	23	39	33.7	18–58

All statistical tests of hypotheses were 2-sided and employed a level of significance of $\alpha = 0.05$.

Results

Seventy-eight subjects completed the 4-week study. A summary of the age and gender of this study population is presented in Table 1. (Eighty subjects were evaluated for prebrushing and post-brushing scores. The 2 subjects who did not complete the study did so for reasons unrelated to the use of the test toothbrushes.) Throughout the study, no adverse effects on the oral hard or soft tissues were observed by the dental examiner or reported by the participants when questioned.

Gingivitis Reduction—Overall

Tables 2 and 3 and Figure 4 present a comparison of the Löe-Silness Gingival Index scores at baseline and after 4 weeks of toothbrush use for each toothbrush group. The baseline (prebrushing) mean gingival index scores for the Colgate® 360™ toothbrush group and the Oral-B® CrossAction® toothbrush group were 1.464 and 1.471, respectively, indicating no statistically significant difference between gingival index scores for the 2 toothbrush groups.

After 4 weeks of use, the mean gingival index score for subjects who used the new Colgate® 360™ toothbrush was 1.321, indicating a statistically significant 9.8% reduction in gingival index scores from baseline ($P < .05$). The mean gingival index score for subjects who used the Oral-B® CrossAction® toothbrush was 1.347, indicating an 8.4% statistically significant difference in gingival index scores from baseline ($P > .05$).

A comparison of the mean gingival index scores of the 2 toothbrush groups showed that

there was no statistically significant difference in gingivitis scores between the Colgate® 360™ toothbrush and the Oral-B® CrossAction® toothbrush after 4 weeks of daily use.

Gingivitis Severity

Tables 2 and 3 and Figure 5 present a comparison of the Gingivitis Severity Index scores at baseline and after 4 weeks of daily use. The baseline (prebrushing) mean Gingivitis Severity Index scores for the Colgate® 360™ toothbrush group and the Oral-B® CrossAction® toothbrush group were 0.484 and 0.467, respectively, indicating no statistically significant difference between Gingivitis Severity Index scores for the 2 toothbrush groups.

After 4 weeks of use, the mean Gingivitis Severity Index score for subjects who used the new Colgate® 360™ toothbrush was 0.359, indicating a statistically significant 25.8% reduction in Gingivitis Severity Index scores from baseline ($P < .05$). The mean Gingivitis Severity Index score for subjects who used the Oral-B® CrossAction® toothbrush was 0.369, indicating a 21% statistically significant difference in Gingivitis Severity Index scores from baseline ($P > .05$).

A comparison of the mean Gingivitis Severity Index scores of the 2 toothbrush groups showed that there was no statistically significant difference in Gingivitis Severity Index scores (bleeding sites) between the Colgate® 360™ toothbrush and the Oral-B® CrossAction® toothbrush after 4 weeks of daily use.

Removal of Supragingival Plaque

Whole Mouth—After Single Toothbrushing

Table 4 and Figure 6 present a comparison of

Table 2—Summary of the Baseline Gingival Index Scores for Subjects Who Completed the 4-Week Clinical Study

Parameter	Treatment	n	Baseline Summary*	Significance†
Gingival Index	Colgate® 360™	39	1.464 ± 0.288	NS
	Oral-B® CrossAction®	39	1.471 ± 0.309	
Gingivitis Severity Index	Colgate® 360™	39	0.484 ± 0.285	NS
	Oral-B® CrossAction®	39	0.467 ± 0.309	

*Mean ± standard deviation.

†Significance of analysis of variance comparison of baseline mean scores. NS = $P > .05$.

NS = not significant.

Table 3—Summary of the 4-Week Gingival Index Scores for Subjects Who Completed the 4-Week Clinical Study

Parameter	Treatment	n	4-week Summary*	Within-treatment Analysis		Between-treatment Analysis	
				Percent Reduction [†]	Significance [‡]	Percent Difference [§]	Significance
Gingival Index	Colgate® 360™	39	1.321 ± 0.292	9.8	<i>P</i> < .05	1.9	NS
	Oral-B® CrossAction®	39	1.347 ± 0.291	8.4	<i>P</i> < .05		
Gingivitis Severity Index	Colgate® 360™	39	0.359 ± 0.245	25.8	<i>P</i> < .05	2.7	NS
	Oral-B® CrossAction®	39	0.369 ± 0.260	21.0	<i>P</i> < .05		

*Mean ± standard deviation.
[†]Reduction between the baseline and 4-week mean scores, expressed as a percentage of the baseline mean score. Positive values indicate lower gingivitis scores at the 4-week examination than at the baseline examination.
[‡]Significance of paired *t* test comparing the baseline and 4-week examinations.
[§]Difference between the 4-week mean scores, expressed as a percentage of the 4-week mean score for the Oral-B® CrossAction®. Positive values indicate lower gingivitis scores for the Colgate® 360™.
^{||}Significance of postanalysis of covariance comparison of baseline-adjusted mean scores. NS = *P* > .05.
 NS = not significant.

whole-mouth Rustogi Modification of the Navy Plaque Index scores at baseline (prebrushing scores), after 1 minute of toothbrushing (postbrushing scores), as well as the differences between the 2 scores. The baseline (prebrushing) mean plaque index scores for the Colgate® 360™ toothbrush group and the Oral-B® CrossAction® toothbrush group were 0.710 and 0.714, respectively, indicating no statistically significant difference between plaque index scores for the 2 toothbrush groups.

The postbrushing (after 1 minute of toothbrushing) mean plaque index score for subjects who used the new Colgate® 360™ toothbrush was 0.436, indicating a difference in plaque index scores of 0.274 (*P* < .05) and a 38.6% reduction in plaque from prebrushing (baseline). The mean plaque index score for subjects who used the Oral-B® CrossAction® toothbrush was 0.476, indicating a difference in plaque index scores of 0.238 (*P* < .05) and a 33.3% reduction in plaque

from prebrushing (baseline).

These differences indicate that after a single, 1-minute toothbrushing, the new Colgate® 360™ toothbrush removed statistically significantly 15.1% more plaque as compared to the Oral-B® CrossAction® toothbrush.

Interproximal Surfaces—After Single Toothbrushing

Table 4 and Figure 7 present a comparison of the interproximal Rustogi Modification of the Navy Plaque Index scores at baseline (prebrushing scores), after 1 minute of toothbrushing (postbrushing scores), as well as the differences between the 2 scores. The baseline (prebrushing) mean plaque index scores for the Colgate® 360™ toothbrush group and the Oral-B® CrossAction® toothbrush group were 0.945 and 0.961, respectively, indicating no statistically significant difference between plaque index scores for the 2 toothbrush groups.

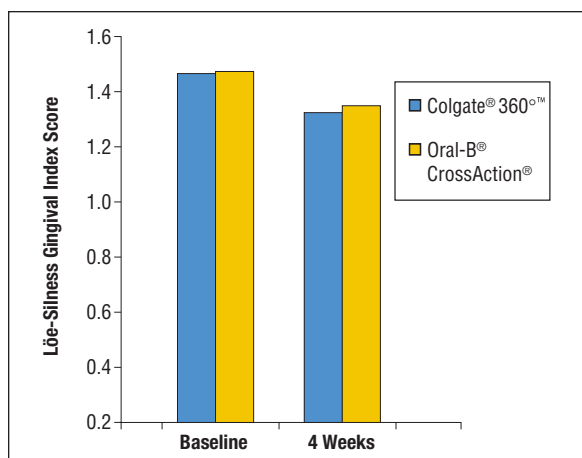


Figure 4—Overall gingivitis scores.

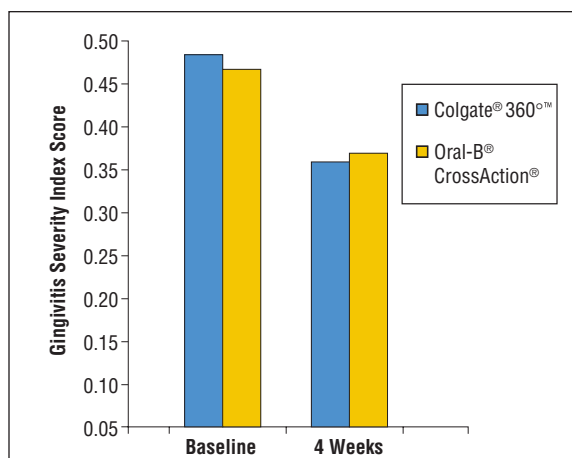


Figure 5—Gingivitis severity scores.

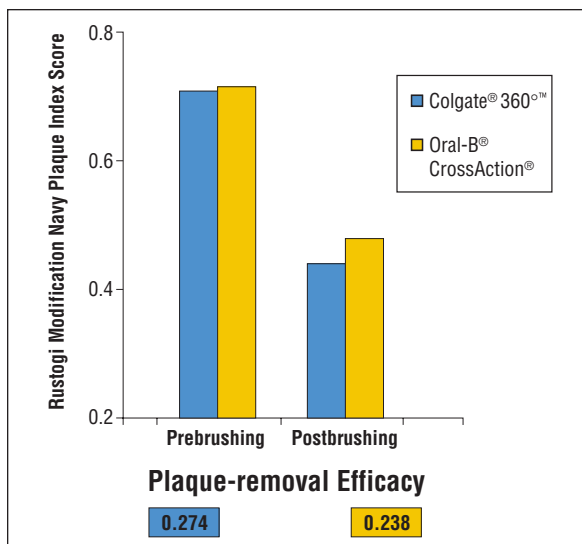


Figure 6—Whole-mouth mean plaque index scores and comparative plaque-removal efficacy.

The postbrushing (after 1 minute of toothbrushing) mean plaque index score for subjects who used the new Colgate® 360™ toothbrush was 0.487, indicating a difference in plaque index scores of 0.458 ($P < .05$) and a 48.5% reduction in plaque from prebrushing (baseline). The mean plaque index score for subjects who used the Oral-B® CrossAction® toothbrush was 0.577, indicating a difference in plaque index scores of 0.384 ($P < .05$) and a 40% reduction in plaque from prebrushing (baseline).

These differences indicate that after a single, 1-minute toothbrushing, the new Colgate® 360™

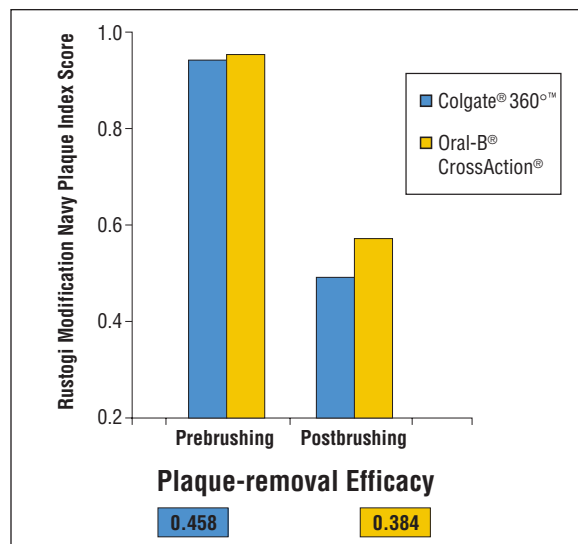


Figure 7—Interproximal mean plaque index scores and comparative plaque-removal efficacy.

toothbrush removed statistically significantly 19.3% more plaque from the interproximal sites in the mouth as compared to the Oral-B® CrossAction® toothbrush.

Gumline Surfaces—After Single Toothbrushing

Table 4 and Figure 8 present a comparison of the gumline Rustogi Modification of the Navy Plaque Index scores at baseline (prebrushing scores), after 1 minute of toothbrushing (postbrushing scores), as well as the differences between the 2 scores. The baseline (prebrushing) mean

Table 4—Summary of the Pre- and Postbrushing Plaque Index Scores for Subjects Examined After a Single Toothbrushing

Parameter	Treatment	n	Prebrushing Summary*	Postbrushing Summary*	Pre- to Postbrushing Reductions in Plaque Index			
					Difference Mean ± SD	Percent Reduction [†]	Between-treatment Analysis	
						Percent Difference [‡]	Significance [§]	
Whole-mouth Plaque Index	Colgate® 360™	40	0.710 ± 0.065	0.436 ± 0.066	0.274 ± 0.074	38.6	15.1	$P < .05$
	Oral-B® CrossAction®	40	0.714 ± 0.056	0.476 ± 0.064	0.238 ± 0.062	33.3		
Plaque Index on Interproximal Sites	Colgate® 360™	40	0.945 ± 0.092	0.487 ± 0.181	0.458 ± 0.176	48.5	19.3	$P < .05$
	Oral-B® CrossAction®	40	0.961 ± 0.067	0.577 ± 0.173	0.384 ± 0.183	40.0		
Plaque Index on Gumline Sites	Colgate® 360™	40	1.000 ± 0.000	0.934 ± 0.067	0.066 ± 0.067	6.6	29.4	NS
	Oral-B® CrossAction®	40	1.000 ± 0.000	0.949 ± 0.065	0.051 ± 0.065	5.1		

*Mean ± standard deviation.
[†]Reduction between the prebrushing and postbrushing scores, expressed as a percentage of the prebrushing score.
[‡]Difference between the pre- and postbrushing reductions in plaque, expressed as a percentage of the reduction for the Oral-B® CrossAction®. Positive values indicate greater plaque reductions for the Colgate® 360™.
[§]Significance of postanalysis of covariance comparison of mean plaque reductions, adjusted for prebrushing plaque scores. NS = $P > .05$.
^{||}Statistically significant reduction from pre- to postbrushing measurements.
SD = standard deviation; NS = not significant.

plaque index scores for the Colgate® 360™ toothbrush group and the Oral-B® CrossAction® toothbrush group were 1.000 for both groups, indicating no statistically significant difference between plaque index scores for the 2 toothbrush groups.

The postbrushing (after 1 minute of toothbrushing) mean plaque index score for subjects who used the new Colgate® 360™ toothbrush was 0.934, indicating a difference in plaque index scores of 0.066 ($P < .05$) and a 6.6% reduction in plaque from prebrushing (baseline). The mean plaque index score for subjects who used the Oral-B® CrossAction® toothbrush was 0.949, indicating a difference in plaque index scores of 0.051 ($P < .05$) and a 5.1% reduction in plaque from prebrushing (baseline).

These differences indicate that after a single, 1-minute toothbrushing, there was no statistically significant difference in plaque index gumline scores between the new Colgate® 360™ toothbrush and the Oral-B® CrossAction® toothbrush.

Whole Mouth—After 4 Weeks

Tables 5 and 6 and Figure 9 present a comparison of the whole-mouth Rustogi Modification of the Navy Plaque Index scores at baseline (prebrushing scores) and after 4 weeks. The baseline (prebrushing) mean plaque index scores for the Colgate® 360™ toothbrush group and the Oral-B® CrossAction® toothbrush group were 0.708 and 0.713, respectively, indicating no statistically significant difference between plaque index scores for the 2 toothbrush groups.

The 4-week mean plaque index score for subjects who used the new Colgate® 360™ toothbrush

was 0.325, while the 4-week mean plaque index score for subjects who used the Oral-B® CrossAction® toothbrush was 0.391, indicating 16.9% statistically significantly ($P < .05$) greater plaque removal for the Colgate® 360™ toothbrush.

This difference indicates that after 4 weeks, the new Colgate® 360™ toothbrush was statistically significantly more effective in removing plaque in the whole mouth as compared to the Oral-B® CrossAction® toothbrush.

Interproximal Surfaces—After 4 Weeks

Tables 5 and 6 and Figure 10 present a comparison of the interproximal Rustogi Modification of the Navy Plaque Index scores at baseline (prebrushing scores) and after 4 weeks. The baseline (prebrushing) mean plaque index scores for the Colgate® 360™ toothbrush group and the Oral-B® CrossAction® toothbrush group were 0.944 and 0.960, respectively, indicating no statistically significant difference between plaque index scores for the 2 toothbrush groups.

The 4-week mean plaque index score for subjects who used the new Colgate® 360™ toothbrush was 0.246 and the 4-week mean plaque index score for subjects who used the Oral-B® CrossAction® toothbrush was 0.409, indicating 39.9% statistically significantly ($P < .05$) greater plaque removal for the Colgate® 360™ toothbrush.

This difference indicates that after 4 weeks, the new Colgate® 360™ toothbrush was statistically significantly more effective in removing plaque at interproximal sites as compared to the Oral-B® CrossAction® toothbrush.

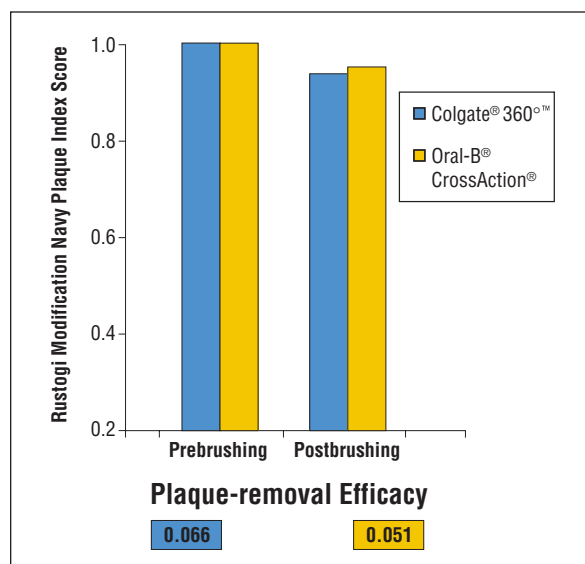


Figure 8—Gumline mean plaque index scores and comparative plaque-removal efficacy.

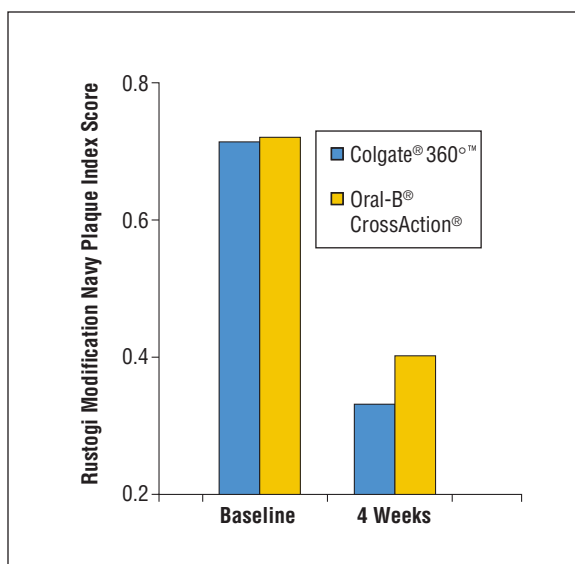


Figure 9—Whole-mouth mean plaque index scores at 4 weeks.

Gumline—After 4 Weeks

Tables 5 and 6 and Figure 11 present a comparison of the gumline Rustogi Modification of the Navy Plaque Index scores at baseline (prebrushing scores) and after 4 weeks. The baseline (prebrushing) mean plaque index scores for the Colgate® 360°™ toothbrush group and the Oral-B® CrossAction® toothbrush group were both 1.000, indicating no statistically significant difference between plaque index scores for the 2 toothbrush groups.

The 4-week mean plaque index score for subjects who used the new Colgate® 360°™ toothbrush was 0.797 and the 4-week mean plaque index score for subjects who used the Oral-B® CrossAction® toothbrush was 0.874, indicating 8.8% statistically significantly ($P < .05$) greater plaque removal for the Colgate® 360°™ toothbrush.

This difference indicates that after 4 weeks, the new Colgate® 360°™ toothbrush was statistically significantly more effective in removing

plaque at the gumline as compared to the Oral-B® CrossAction® toothbrush.

Discussion

Plaque removal has been a major focus in the prevention and control of dental diseases, particularly periodontal diseases. This study shows that after both single use and extended use, the Colgate® 360°™ toothbrush and the Oral-B® CrossAction® toothbrush effectively removed plaque at all sites when compared with prebrushing baseline values. However, after a single brushing, significantly more plaque was removed by the Colgate® 360°™ toothbrush than the Oral-B® CrossAction® toothbrush for the whole mouth (reduction of 38.6% vs 33.3%) and for interproximal surfaces (reduction of 48.5% vs 40%), although no differences were noted for gumline sites (reduction of 6.6% vs 5.1%). This plaque-removal superiority of the Colgate® 360°™ tooth-

Table 5—Summary of the Baseline Prebrushing Plaque Index Scores for Subjects Who Completed the 4-Week Clinical Study

Parameter	Treatment	n	Baseline Summary*	Significance†
Whole-mouth Plaque Index	Colgate® 360°™	39	0.708 ± 0.065	NS
	Oral-B® CrossAction®	39	0.713 ± 0.056	
Plaque Index on Interproximal Sites	Colgate® 360°™	39	0.944 ± 0.092	NS
	Oral-B® CrossAction®	39	0.960 ± 0.068	
Plaque Index on Gumline Sites	Colgate® 360°™	39	1.000 ± 0.000	NS
	Oral-B® CrossAction®	39	1.000 ± 0.000	

*Mean ± standard deviation.
†Significance of analysis of variance comparison of baseline mean scores. NS = $P > .05$.
NS = not significant.

Table 6—Summary of the 4-Week Plaque Index Scores for Subjects Who Completed the 4-Week Clinical Study

Parameter	Treatment	n	4-week Summary*	Within-treatment Analysis		Between-treatment Analysis	
				Percent Reduction†	Significance‡	Percent Difference§	Significance¶
Whole-mouth Plaque Index	Colgate® 360°™	39	0.325 ± 0.057	54.1	$P < .05$	16.9	$P < .05$
	Oral-B® CrossAction®	39	0.391 ± 0.062	45.2	$P < .05$		
Plaque Index on Interproximal Sites	Colgate® 360°™	39	0.246 ± 0.165	73.9	$P < .05$	39.9	$P < .05$
	Oral-B® CrossAction®	39	0.409 ± 0.179	57.4	$P < .05$		
Plaque Index on Gumline Sites	Colgate® 360°™	39	0.797 ± 0.080	20.3	$P < .05$	8.8	$P < .05$
	Oral-B® CrossAction®	39	0.874 ± 0.075	12.6	$P < .05$		

*Mean ± standard deviation.
†Reduction between the baseline and 4-week mean scores, expressed as a percentage of the baseline mean. Positive values indicate lower plaque scores at the 4-week examination than at the baseline examination.
‡Significance of paired t test comparing the baseline and 4-week examinations.
§Difference between the 4-week mean scores, expressed as a percentage of the 4-week mean score for the Oral-B® CrossAction®. Positive values indicate lower plaque scores for the Colgate® 360°™.
¶Significance of postanalysis of covariance comparison of baseline-adjusted means.

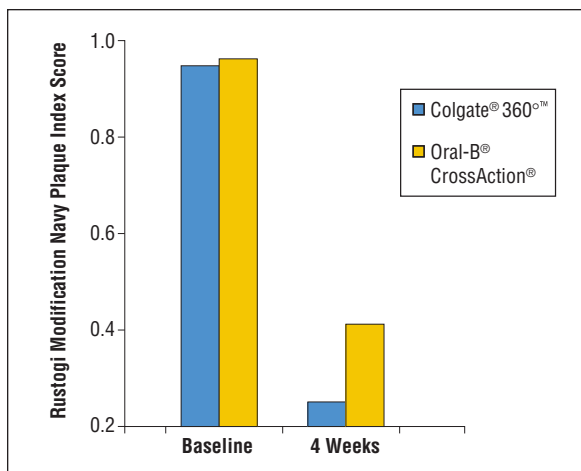


Figure 10—Interproximal mean plaque index scores at 4 weeks.

brush was sustained over an extended period of toothbrushing, and at 4 weeks, significant differences in favor of the Colgate® 360™ toothbrush were again noted when compared with the Oral-B® CrossAction® toothbrush for removing plaque from the whole mouth (54.1% vs 45.2%), interproximal sites (73.9% vs 57.4%), and the gumline (20.3% vs 12.6%). The results from this study also indicate that the Colgate® 360™ toothbrush was comparable to the Oral-B® CrossAction® toothbrush in reducing moderate and severe gingivitis after 4 weeks of daily use.

Although the efficacy in plaque removal is central to the prevention of periodontal diseases, oral health also is influenced by patient compliance in establishing an effective home-care regimen. In this context, a patient's overall perception of the cleaning efficacy of a toothbrush could be an important motivator for maintaining good oral hygiene.⁹ In addition to the tightly packed and tapered bristles that enhance interproximal and gumline cleaning, the Colgate® 360™ toothbrush includes additional features, such as polishing cups and textured tongue-and-cheek cleaners, which extend cleaning beyond the dentition to include other areas of the mouth. Thus, the Colgate® 360™ toothbrush provides cleaning of the whole mouth and may increase oral hygiene practice.

The results of this 4-week study support the conclusion that the Colgate® 360™ toothbrush provides clinically superior control of supragingival plaque as compared with the Oral-B® CrossAction® toothbrush. Both toothbrushes were equally effective in reducing the severity of gingivitis.

Conclusion

Seventy-eight subjects complied with the pro-

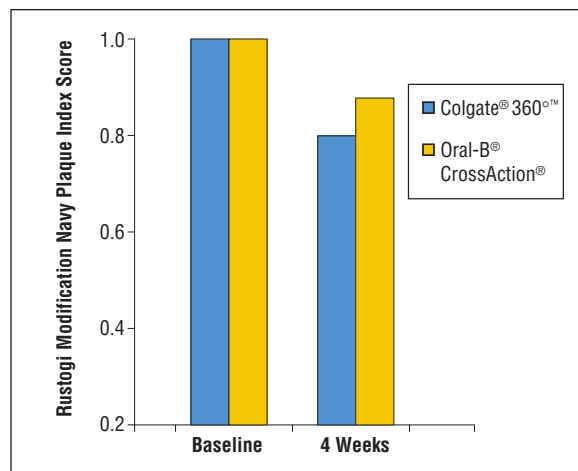
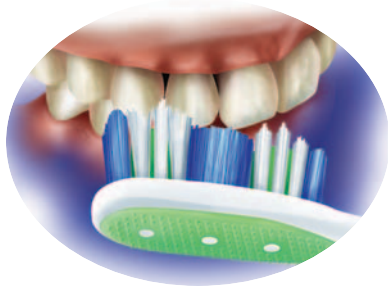


Figure 11—Gumline mean plaque index scores at 4 weeks.

tolocol and completed the 4-week, examiner-blind, clinical study. Under the conditions of this clinical study, the new Colgate® 360™ toothbrush removed statistically significantly more plaque than the Oral-B® CrossAction® toothbrush. Superior plaque-removal performance was found in separate analyses of the whole-mouth, interproximal, and gumline areas. The results of the study also indicated that the new manual toothbrush, the Colgate® 360™ toothbrush, was comparable in reducing moderate and severe (bleeding sites) gingivitis as compared with the Oral-B® CrossAction® toothbrush after 4 weeks.

References

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Laboratory Investigation of Colgate® 360°™ Toothbrush and Oral-B® Indicator® Toothbrush for the Removal of Dental Stains

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Abstract: The objective of this *in vitro* study was to evaluate the stain-removal efficacy of a newly designed manual toothbrush, the Colgate® 360°™, relative to a commercially available toothbrush, the Oral-B® Indicator®. A modification of Stookey et al was used to evaluate the stain-removal effects of toothbrushes instead of dentifrice on bovine teeth. A V-8 mechanical cross-brushing machine equipped with the test toothbrushes and adjusted to 500g to enamel surfaces evaluated stain removal using a dentifrice slurry and water after 800 double strokes. The overall results of this laboratory investigation indicate that the Colgate® 360°™ toothbrush is more effective, $P < .05$, than the commercial Oral-B® Indicator® toothbrush in removing dental stain and brightening teeth using a standard toothpaste or water.

Removing extrinsic stains from teeth for cosmetic, therapeutic, or psychological reasons is a steadily growing trend among consumers and dental professionals.¹ The demand for whiter teeth among consumers is evidenced by the expanding plethora of products targeted to this goal.² Within this demand for teeth brightening products, toothbrushes have become a recognizable tool in maintaining or enhancing levels of tooth brightness by inhibiting extrinsic-stain affluence.³ The purpose of this laboratory investigation was to evaluate the Colgate® 360°™ toothbrush^a for its ability to remove various types of extrinsic stains from teeth when used with a conventional dentifrice and water, compared to a commercial toothbrush, the Oral-B® Indicator®.^b

Materials and Methods

The laboratory method in this investigation is a modification and improvement of that described by Stookey et al,⁴ which has been shown to correlate with the cleaning and whitening properties of dentifrices in clinical trials. The Stookey method was modified to evaluate the stain-removal effects of toothbrushes—full-head soft variants in this case—instead of dentifrices.

Squares of dental enamel 4 mm on each side were cut, using a diamond-cutting disk, from permanent bovine incisors. Using a mold, the enamel squares were embedded in clear polyester casting resin to provide 1.5-cm square blocks with the labial surface exposed and raised about 4 mm above the acrylic block surface. The labial surface of the enamel squares was made flat by a dental model trimmer so that color readings could be taken with a

^aThe Colgate-Palmolive Company, New York, NY 10022; 800-338-8388

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^cKonica Minolta Holdings, Inc, Ramsey, NJ 07446; 201-825-4000

Minolta® CM-503i Spectrophotometer^c with diffuse illumination/8° viewing angle and 3-mm aperture. The surface was then smoothed by hand sanding on 400-grit emery paper using water as the lubricant until all grinding marks were removed. Finally, the labial surface was hand-polished to a mirror finish using a water slurry of GK1072 calcined kaolin (median particle size = 1.2 μm) on a cotton cloth. The finished specimens were examined under a dissecting microscope and discarded if surface imperfections were observed.

Removing extrinsic stains from teeth for cosmetic, therapeutic, or psychological reasons is a steadily growing trend among consumers and dental professionals.

In preparation for the formation of stained pellicle on the enamel, the specimens were etched for 60 seconds in 0.2 mL hydrochloric acid followed by a 30-second immersion in a saturated solution of sodium carbonate. A final etch was performed with 1% phytic acid for 60 seconds, then the specimens were rinsed with deionized water and attached to the staining apparatus.

The tooth-staining apparatus was designed to provide alternate immersion into the staining broth and subsequent air-drying of the specimens. The apparatus consisted of an aluminum platform base that supported a Teflon rod (0.75 inch in diameter) connected to an electric motor. By means of a speed reduction box, the motor rotated the rod at a constant rate of 1.5 rpm. Threaded screw holes were spaced at regular intervals along the length of the rod. The tooth specimens were attached to the rod by first gluing the head of a plastic screw to the back of the specimen, and then screwing the tooth onto the rod. Beneath the rod was a removable, 300-mL capacity trough

that held the tooth-staining broth.

The multiple-stain broth was prepared by adding 1.02 g of instant coffee, 1.02 g of instant tea, 10 mL of red wine, and 0.75 g of gastric mucin to 250 mL of sterilized trypticase soy broth. Approximately 50 mL of a 24-hour *Micrococcus luteus* stain-forming bacterial culture also was added to the stain broth. The apparatus, with the enamel specimens attached and the staining broth in the trough, was then placed in an incubator at 37°C, with the specimens rotating continuously through the staining broth and air. The staining broth was replaced once every 24 hours until the stained pellicle film on the specimens was sufficiently dark. With each broth change, the trough and specimens were rinsed and brushed with deionized water to remove any loose deposits. When staining was complete, the specimens were removed from the staining broth, brushed thoroughly with deionized water, placed in a humidior, and refrigerated until used.

The amount of stain on the teeth was measured by taking color readings with a Minolta® spectrophotometer. A spectrophotometer obtains reflectance measurements indicated in International Commission on Illumination (CIE) tristimulus values X, Y, and Z, from which CIELAB L*a*b* values are calculated.⁵ The CIELAB color

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scale quantifies color according to three parameters: L* (light-dark), a* (red-green), and b* (yellow-blue). The stained enamel specimens were allowed to air-dry at room temperature for 30 minutes before color measurements were made. Measurements were conducted by aligning the flat center of the 4-mm square segment of stained enamel directly over the 3-mm diameter targeting aperture of the Minolta® spectrophotometer. The averages of three color readings were taken for each specimen.

In preparation for treatment, the stained teeth were stratified into equal groups of 16 specimens, with each group having balanced average baseline L*a*b* stain scores. The specimens were positioned on a V-8 mechanical cross-brushing

$$CR_x = \sqrt{(C - 1) \left(CR_x^2 \div \frac{v_1}{v_2} \right)} = CR_x \pm \frac{L}{2}$$

- C = Confidence interval
- R_x = Estimate of the true ratio
- v = Variance
- L = v₁ - v₂

Figure 1—Bliss Modification of the Fieller Confidence Interval.

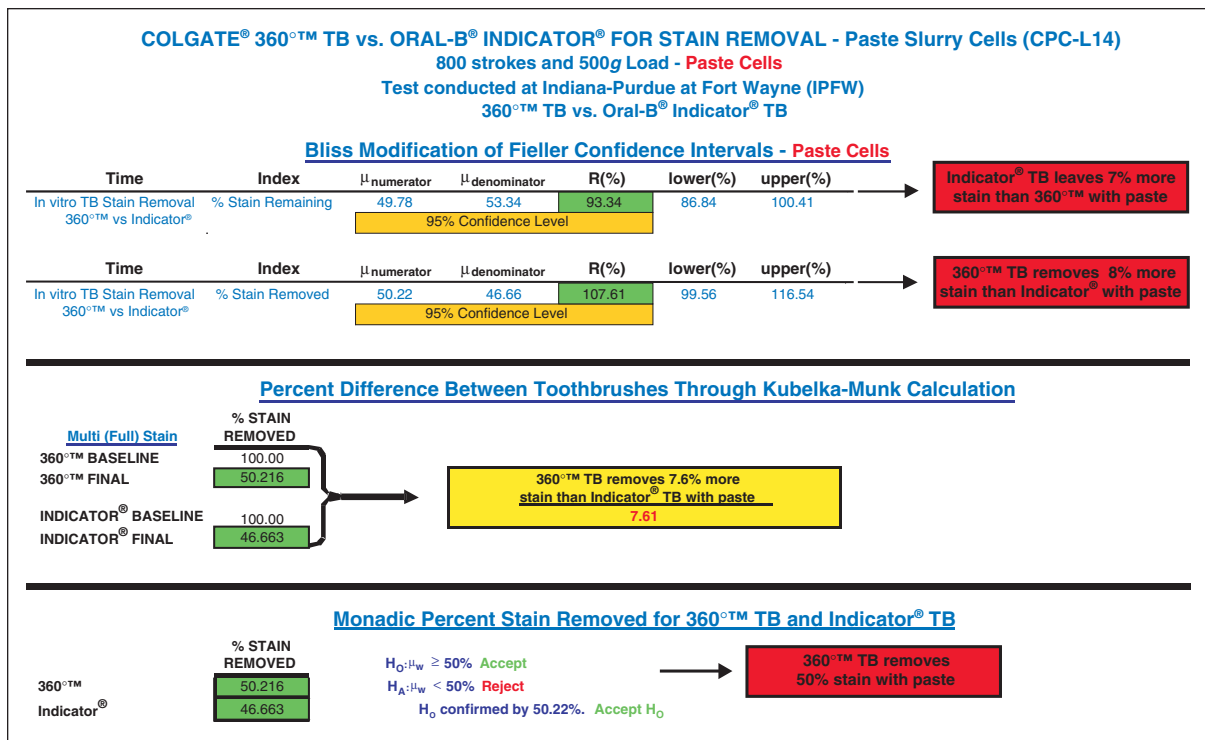


Figure 2—Summary of stain results with paste slurry.

machine⁶ equipped with the test toothbrushes adjusted to 500g on the enamel surfaces. Brushing was conducted with distilled water or a standard dentifrice (Colgate® Cavity Protection Great Regular Flavor Fluoride Toothpaste^a) for 800 double strokes. The dentifrice was used as a slurry to simulate salivary dilution in the mouth by mixing 25 g of dentifrice with 40 g of deionized water. To minimize mechanical variables, enamel specimens for different groups were brushed during each run, and the test brushes were randomly assigned to each brush station until all of the products had been tested twice at all 8 stations. Also, close attention was paid to the alignment of enamel specimens in all planes to ensure uniformity of brushing patterns. After brushing, the specimens were rinsed, allowed to dry for 30 minutes, and then color readings were made. Next, the specimens were pumiced using a dental hand-piece to clean all residual stain off of the teeth, and then color readings were taken again. This final cleaning procedure provided a value for each specimen that represented the maximum amount of stain potentially removed by a test toothbrush.⁴

The test-product group assignment and identification of the test toothbrushes were as follows:

- Group 1—Colgate® 360°™ toothbrush with Colgate® dentifrice slurry;
- Group 2—Colgate® 360°™ toothbrush with water;
- Group 3—Oral-B® Indicator® toothbrush with Colgate® dentifrice slurry; and
- Group 4—Oral-B® Indicator® toothbrush with water.

Data were tabulated using a spreadsheet program and statistically analyzed by a conventional statistics program and the Kubelka-Munk calculation.

Calculations

The overall change in the color of the stained teeth was calculated using the Kubelka-Munk⁷ equation:

$$\frac{K}{S} = \frac{(1 - R_{\infty})^2}{2R_{\infty}}$$

The K/S value is used to measure, in a linear fashion, the concentration of colorants or stains. The K/S value is obtained by analyzing the CIE tristimulus Y value, which is a broadband reflectance measurement and an indication of lightness.⁷ Use of the K/S function allows one to determine the ability of the test toothbrush to remove stain and brighten teeth. The data were calculated and are defined as follows:

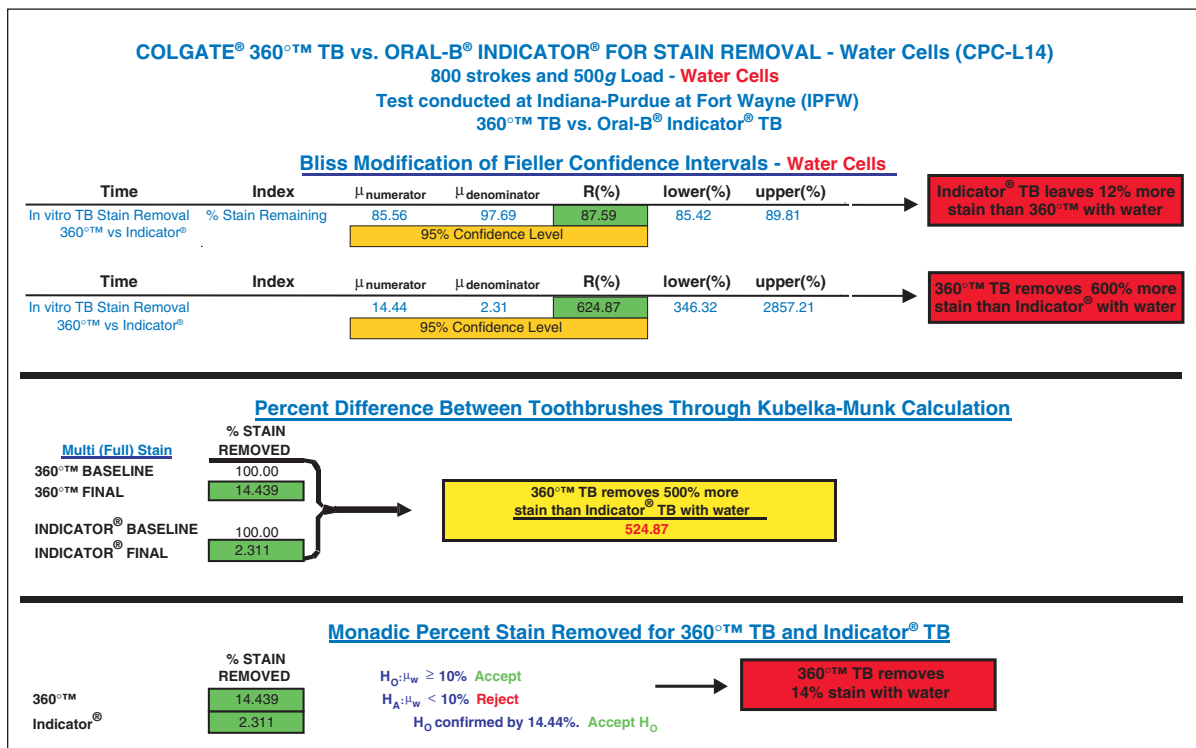


Figure 3—Summary of stain results with water.

R_∞ = Reflectance of stain-film thickness;
 K = Absorption coefficients;
 S = Scattering coefficients;
 Stain Removed = K/S score after treatment;
 Total Stain Available = Y score pre- and posttreatment and pumicing; and
 % Stain Removed = “Stain Removed” divided by “Total Stain Available” x 100%.

Data were tabulated using a spreadsheet program and statistically analyzed by a conventional statistics program and the Kubelka-Munk calculation. Comparisons of the resultant Kubelka-Munk data were evaluated using the Bliss Modification of the Fieller Confidence Interval (Figure 1).⁸

Results

Statistical analysis of the $L^*a^*b^*$ color scores for the multiple extrinsic stains on the teeth at baseline showed that the test groups were well balanced for each color factor before treatment. The data show that the Colgate® 360™ toothbrush was more effective, $P < .05$, than the Oral-B® Indicator® in removing multiple dental stains when brushing with a Colgate® dentifrice. To calculate the percentage of stain removed by the toothbrushes, all of the remaining stain on the test teeth was removed by pumicing them until they were free of stain. The average maximum K/S scores were obtained after complete stain removal. By comparing the maximum K/S values

with those produced by the respective toothbrushes, the percent reduction in stain was calculated for each toothbrush and is shown in Figures 2 and 3. When used with a Colgate® dentifrice, the Colgate® 360™ toothbrush removed 50% of the stain compared to a 47% reduction for the Oral-B® Indicator®. With water, the Colgate® 360™ toothbrush removed 14% of the stain compared to a 2% reduction for the Oral-B® Indicator®. The Colgate® 360™ toothbrush was statistically more effective, $P < .05$, in removing stain and brightening teeth than the Oral-B® Indicator®.

1. The Colgate® 360™ toothbrush removed 50.22% of multiple stains compared with 46.66% for the Oral-B® Indicator® when using monadic results obtained through the Kubelka-Munk calculation.
2. The Colgate® 360™ toothbrush left 7% (using R%) less stain than the Oral-B® Indicator® with paste slurry.
3. The Colgate® 360™ toothbrush removed 8% (using R%) more stain compared to the Oral-B® Indicator® with paste slurry.
4. The Colgate® 360™ toothbrush removed 14.44% of multiple stains compared with 2.3% for the Oral-B® Indicator® with water when using monadic results obtained through the Kubelka-Munk calculation.
5. The Colgate® 360™ toothbrush left 12%

- (using R%) less stain than the Oral-B® Indicator® with water.
6. The Colgate® 360°™ toothbrush removed 600% (using R%) more stain compared to the Oral-B® Indicator® with water.

Conclusion

The Colgate® 360°™ toothbrush was more effective than the Oral-B® Indicator® in removing dental stains from teeth when evaluated in vitro using a standard dentifrice slurry. The Colgate® 360°™ toothbrush also was more effective than the Oral-B® Indicator® in removing dental stains from teeth when evaluated in vitro using water. The overall results of this laboratory investigation indicate that the Colgate® 360°™ toothbrush is more effective than the commercial Oral-B® Indicator® in clinically removing dental stain and brightening teeth using toothpaste or water.

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