

This Clinical Study conducted by the University of Milano clearly demonstrates the effectiveness of the Silver Care System in fighting Bacterial colonization of the tooth brush between use. The reduction in the colonization of the Streptococcus mutans is particularly addressed.

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BACTERIAL CONTAMINATION OF TOOTHBRUSHES

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SUMMARY: Bacteria Contamination of Toothbrush The toothbrush, the most common and important device in the prevention of plaque-related diseases, is subject to constant contamination by saprophyte and pathogenic microorganisms, found in the oral cavity, which gives it an infective charge for the next use. The aim of this study was to evaluate the residual contamination of *Streptococcus mutans* on a conventional toothbrush compared to a new model with a silver-plated head. Even though the antimicrobe activity of the silver-plating that covers the replaceable head had already been confirmed by laboratory tests, it was necessary to compare further data with uncontained samples of *Streptococcus mutans*. A clinical randomized trial was performed on 20 patients (average age 24 ± 0.3) assigned to an experimental group ($n=10$) and a controlled group ($n=10$). After using the toothbrush at home for 3 days, the toothbrushes were returned to the laboratory and kept in a humid atmosphere to prevent alteration of the bacterial flora. Cultures were lifted from various tufts of each toothbrush head at the end of the clinical test and again 6 hours later. These were placed on MSB agar plates and incubated. The data obtained demonstrates that the toothbrush with the silver-plated head has significantly higher reduction of the colonization of *Streptococcus mutans* as apposed to the conventional toothbrush. These results show two things; one being the colonization of bacteria on the surface of the toothbrush, the other, the antibacterial activity of the silver ions on the replaceable head after normal domestic use.

KEY WORDS: prevention, silver, *Streptococcus mutans*, toothbrush

Caries prevention is based on the control and elimination of bacterial plaque from tooth surface. The toothbrush is the most common and important device in the prevention of plaque-related diseases. The Italian market offers a wide range of models that differ in shape, material and technical characteristics; relevant differences in efficiency of plaque removal and abrasiveness on the hard or soft oral tissues can be demonstrated. The residual microbial contamination of toothbrushes is a problem which has become evident in recent years. According to British Standards Institution specifications all toothbrushes are made-up of different parts,

each of which is specifically defined:

- ◊ brush (the part of the toothbrush that comprises the tufts);
- ◊ tuft (the aggregate of filaments that are fixed in one hole in the stock);
- ◊ stock (the extension of the handle which supports the tufts);
- ◊ head (the stock and the group of tufts);
- ◊ handle (the part of the toothbrush which is not defined as the head).

The head is the part of the toothbrush which comes into direct contact with the oral cavity, where it is subject to contamination by saprophyte, and in certain cases pathogenic microorganisms. Besides the infective load which the

head is in contact with, the residual contamination depends on some structural characteristics. The head retains water amongst the bristles and thus preserves viability of the contaminating microorganisms after use¹⁻⁵. Experimental studies have demonstrated that the microorganism load retained and preserved viable also depends on the rinsing and drying procedures of the toothbrush after use⁶.

Oral saprophyte species are the microorganisms that the toothbrush comes into contact with during normal use. Toothbrushes can become carriers of pathogenic microorganisms when used by subjects affected with infectious diseases su-

HYGIENE AND PREVENTION

TTEST RESULTS

	Diff mean	DF	t	p	
Control, Silvercare	0,007	18	0,103	0,9194	
	N	Mean	Variance	SD	SE
Control	10	1,699	0,026	0,162	0,051
Silvercare	10	1,692	0,016	0,125	0,039

TABLE 1

VARIANCE ANALYSIS

	DF	Sum of squares	Quadratic mean	F	p	λ	Power
Toothbrush	1	0,096	0,096	5,779	0,0272	5,779	0,621
Subject (Group)	18	0,300	0,017				
Category for Time	1	1,215	1,215	69,679	<0,0001	69,679	1,000
Category for Time* Toothbrush	1	0,084	0,084	4,799	0,0419	4,799	0,537
Category for Time* Subject (Group)	18	0,314	0,017				
	N	Mean	SD	SE			
Control	20	1,570	0,193	0,043			
Silvercare	20	1,472	0,251	0,056			

TABLE 2

ch as Hepatitis or AIDS^{1,3,5,6}. It must be underlined that *Streptococcus mutans*, the most important cariogenic microorganism, may easily be transferred from the oral cavity of one subject to that of another by the use of the same toothbrush. This microorganism, in fact, can survive for a relatively long period of time on the toothbrush, re-infect the oral cavity of the person who uses the toothbrush next². This phenomenon is particularly important in longitudinal transmission (mother-child) of *Streptococcus mutans*.

Toothbrushes that claim to have an antibacterial action and instruments to reduce the level of residual microbial contamination are available

on the Italian and international markets. Companies sensitive to the hygienic needs expressed by society specifically design them.

The aim of the experiment is the comparative evaluation of residual *Streptococcus mutans* contamination of two toothbrushes: a standard and a silver-plated head toothbrush, after three-days use.

MATERIALS AND METHODS

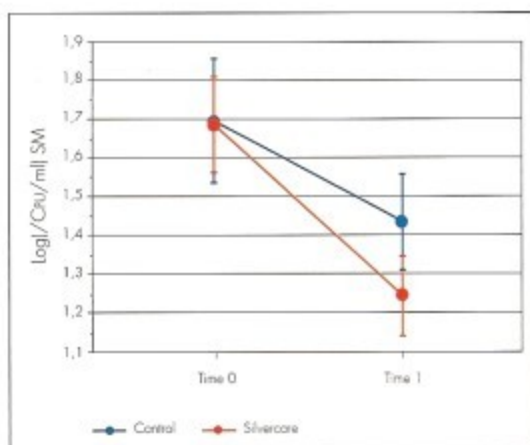
Twenty subjects coming from the patients of Dental Clinic of the Department of Medicine, Surgery and Dentistry S.Paolo were selected

(mean age 24±0,3), with a minimum of 24 teeth.

Subjects included in the experimental group had not had antibiotic treatment or any other pharmacological treatment (including chlorhexidine or topical fluorides) affecting oral flora 30 days prior to the beginning of the study. They did not have dental prosthesis, implants or orthodontic appliances.

Two models of toothbrushes available on the Italian market were used as vehicles for this experiment: one with a silver-plated head (*Silver Care*, Spazzolificio Piave, Onara di Tombolo) and, as control, a standard toothbrush, similar in the number of tufts and the size of the head. 10





1 Analysis of variance results. The reduction trend of the infection load is significantly different in the two groups

samples of each model were used. On the first day of the experiment the subjects were asked to sign an informed consent to participate in the research.

The subjects were then randomly assigned to the Silver Care group (n=10) and to the control group (n=10) and were given the corresponding toothbrush.

The subjects were asked to use the toothbrush twice a day (in the morning and in the evening), using the modified Bass technique, for a total of 3 minutes, abstaining from the use of any kind of toothpaste.

After a 3-day period of use, the toothbrushes were re-consigned to the laboratory immediately after the last brushing, which was carried out just before the beginning of the microbiological procedures. Under a laminar flow cabinet the head of each toothbrush was cut and placed in sterile tubes containing 0,5 ml of

sterile PBS (Sigma Chemicals, St. Louis, MO, USA), to prevent the dehydration of the samples. Each head was sampled twice, respectively at the end of the clinical procedure (T 0) and after 6 hours (T 1), by inserting 10 µl of sterile saline into the center of a tuft of each head with the aid of a micropipette.

The suspension was recovered and diluted in 1ml of sterile saline and 20 µl were then placed on a MSB agar and incubated for 48 hours at 37 °C.

At the end of the incubation the colony count was performed.

STATISTICAL ANALYSES

Statview 5.0 for Windows (SAS Institute Inc, Cary, North Carolina, USA) was used for the statistical analyses.

A paired Student t-test was carried

out to evaluate the homogeneity of the two groups at T 0.

An ANOVA for repeated measures was used to evaluate the reduction of the concentration over the experimental period. The significance level of was set at $p < 0,05$.

RESULTS

A Student t-test was used to evaluate the homogeneity of the two groups at T 0 (table 1). Data show that the mean concentration in both groups are not significantly different ($p=0,919$) at the beginning of the experimental period.

Results of ANOVA (table 2) show that, although the reduction of the microbial concentration in the tufts mainly depends on time ($p < 0,0001$), there is a statistically significant difference in the results obtained from the two toothbrushes ($p=0,027$) (figure 1).

DISCUSSION AND CONCLUSION

Caries decline in industrialized countries and preventive dentistry efforts has changed the target of caries prevention. Recently a new objective has been identified with the aim of optimizing the cost-benefit ratio of prevention: the identification of «high risk subjects». These individuals demonstrate caries prevalence significantly higher than the norm.

Microbiological research has identified *Streptococcus mutans* (Sm) as the principal etiological agent of the disease. Sm produces the acid meta-



bolites that dissolve dental enamel causing the development of white spots.

Nevertheless, in general terms, other bacterial species also have cariogenic potential⁷.

High risk subject treatment is based on the application of preventive measures aimed at reducing the «infection load» of cariogenic bacteria. In recent years with the appearance of transmittable pathologies such as AIDS and infection by HBV and HCV, the concept of longitudinal transmission of caries⁸ has become widely accepted.

Infection control measures for these systemic diseases caused a marked reduction of the use at a community level, and therefore the sharing of, oral hygiene instruments.

While prevention of longitudinal transmission of Sm during pregnancy⁹, is quite easily achieved, it is more difficult to prevent Sm horizontal transmission.

With the aim of reducing residual bacterial contamination of toothbrushes, for the prevention of cross-infection amongst subjects in a community context, (be it the family context or the wider scholastic community) industry has tried to satisfy these health needs, by researching different techniques for decontamination of toothbrushes. The use of metals with bactericidal activity goes back to the end of the nineteenth century when the first experimental observations were made. These were then confirmed at the start of the twentieth century by a more precise oligodynamic classification of metallic ions¹⁰. Colloidal metal solutions have different antibacterial mechanisms that are

mainly based on the interference with the metabolism of cell wall and with activity of some of the enzymatic proteins¹¹.

The antimicrobial activity of the silver that covers the replaceable head of the Silver Care toothbrush, has already been confirmed by in vitro experiments¹⁰.

However, further information about in vivo Sm contamination was needed. Experimental results show that this microorganism is able to colonize the surface of any instrument used during daily domestic oral hygiene.

Results also demonstrated that the activity of the silver ions released by Silver Care is able to significantly reduce the concentration of Sm after a period of time equivalent to the normal interval between brushing.

Although our data is specific to the colonization on the bristle tufts, these results are partially in agreement with in vitro studies results. The variability observed in the data is quite large. This is probably due to the action of confounding factors as other principles with bactericidal activity.

Available literature provides little information on the effect of toothpaste² on the bacterial colonization pattern of the toothbrush. We have to take into account that the adherence of oral microorganisms can markedly be affected by the presence in the oral cavity of different compounds with antibacterial action. The use of toothpaste containing these active principles may influence the decontamination pattern that we have found, if their concentration is sufficient.

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