

**EFFICACY OF VARIOUS TOOTHPASTES USED IN KINGDOM OF SAUDI
ARABIA AGAINST DENTAL CARIES CAUSING STREPTOCOCCUS
MUTANS**

Yousuf A Ali,

Associate Professor, College of Applied Medical Sciences, Jazan University,
Jazan, Kingdom of Saudi Arabia.

Ahmed Abdulhaq,

Assistant Professor and Vice-Dean College of Applied Medical Sciences, Jazan University,
Jazan, Kingdom of Saudi Arabia.

Noha E Eldin,

Lecturer (Microbiology) College of Applied Medical Sciences, Jazan University,
Jazan, Kingdom of Saudi Arabia.

Shah Alam,

Lecturer (Microbiology) College of Applied Medical Sciences, Jazan University,
Jazan, Kingdom of Saudi Arabia.

ABSTRACT

One of the most important part of the body is the mouth, where the digestion process begins. For this the individual should have the healthy teeth structure to masticate the food properly to help digestion. The teeth are the toughest part of the body and cannot be destroyed easily. However there are bacteria specially Streptococcus mutans which is most important caries causing organism which can cause dental decay. Streptococcus mutans secrete dextran a complex polysaccharide which sticks to the smooth surfaces of gums and invite other bacteria viz Lactobacillus spp., Veillonella spp. and Actinomyces spp to form dental plaque. The acid secretion in this microenvironment leads to decay of tooth. Once the tooth is decayed it is

difficult to restore it. Therefore earlier the scientists have suggested various means to prevent the decay of tooth specially by natural means of brushing the teeth by neem or miswak twigs. However the recent advances in dentistry lead to the development of various tooth pastes which can easily prevent the growth of caries causing bacteria. In our present study we have concentrated on various tooth pastes which are available in Kingdom of Saudi Arabia and its effect on Streptococcus mutans the most predominant caries causing bacteria. Among all others Closeup was found to be most effective, showing highest zone of inhibition against Streptococcus mutans on Mitis Salivaris agar for the total nine isolates. However the consistency of results was of Colgate which was also found to be very effective.

Key Words:

Toothpastes, *Streptococcus mutans*, Plaque, dental decay, Dextran, Caries.

Introduction:

The teeth are one of the toughest structure in human body, which also is not spared by the attack of the bacteria. The dissolution of tooth enamel by the secretion of acid in microenvironment of plaque leads to dental caries (7).

The most common disease in the world today is Dental caries. Until recently almost everyone had experienced tooth decay in their lifetime. However, today many people are caries free and there has been a 40-60 % reduction in the incidence of tooth decay around the Western world (6). The disease which was once considered to be pandemic has been declined over a period of time and is now only endemic. This is because of general awareness among people and children in particular, of how to maintain the oral hygiene and use of different tooth pastes and mouth washes.

Teeth are composed of a thin layer (1-2mm) of dental enamel which forms the hard protective coating over the tooth. This consists mainly of calcium, phosphate and other ions in a structure known as "hydroxyapatite". Dental enamel is porous and is susceptible to acid dissolution during the process of demineralization.

The *Streptococcus mutans* has an ability of forming dental plaque by secretion of most sticky / mucous secretion known as dextran or glucan. This stick to the smooth surfaces of gums forming

plaque (1). The other bacteria which are also present in the mouth and are caries active are *Lactobacillus spp.*, *Veillonella spp.* and *Actinomyces spp* (3). All these and other bacteria get adhere to the plaque and secrete acid in microenvironment leading to dental caries.

Thus most important bacteria (*Streptococcus mutans*) which is responsible for plaque formation if eliminated from the mouth it reduces the chance of dental caries.

Streptococcus mutans is a gram positive, facultative anaerobic bacteria described by JK Clark(2) in 1924 after he isolated it from carious lesions. It metabolizes carbohydrates specially sucrose and produces extracellular polysaccharide “Glucan” or “Dextran” (4) by which the bacteria stick to smooth surfaces and forms plaque.

The effects of *Streptococcus mutans* can be counteracted through proper oral hygiene and use of fluoride. Fluoride has been found to be the most effective agent against caries because it acts through topical mechanisms inhibiting the demineralization of enamel and tooth structure, enhancement of remineralization and inhibition of bacterial enzymes such as enolases, phosphatases, pyrophosphatases etc.

Different antimicrobial agents have different actions in inhibiting the oral microflora. Fluoride directly inhibits enolase enzyme of cariogenic bacteria. Chlorhexidine reduces the population of cariogenic bacteria specially *Streptococcus mutans* by interfering with bacterial adherence. Green tree extract which is rich in catechin (antioxidant) inhibits *Streptococcus mutans* and kills other oral bacteria, combats oral plaque and inhibit collagenase activity.

Toothpastes has different ingredients (triclosan, bromochlorophene, sodium lauryl sulfate (SLS), sodium mono-fluorophosphate (MPF), and sodium fluoride (SF) can inhibit the activity of the cariogenic bacteria and hence the dental plaque.

The most predominant bacteria causing dental caries are *Streptococcus mutans* and *Streptococcus sanguis*. However the secondary invaders are *Actinomyces spp* and *Candida albicans*. Due to prolong infection or wound caused due to extraction of tooth these *viridians Streptococci* can cause endocarditis. Therefore it is of utmost important to prevent or treat the caries to stop the proliferation of infective agents. Thus new research in the field of tooth pastes and addition of different ingredients to enhance its antimicrobial activity is always welcome.

Review of Literature:

The caries can be prevented using tooth brush and pastes. Brushing on smooth surface helps dislodge the plaque, while the toothpaste inhibits the growth of caries causing bacteria.

Brushing of teeth is an age old phenomenon which is practiced since thousands of years. At the time of stone age there were no toothpastes or any chemicals available by which the bacteria causing dental caries can be eliminated. Also there was no sufficient knowledge of the causative agent of dental caries. People use to wash or rinse their mouth to avoid the bad breath. Later the plant twigs like miswak and neem were used to clean the smooth surface and keep away the bad breath.

It was in 1961 that Clark first demonstrated that *Streptococcus mutans* is predominant bacteria responsible for dental caries. It is gram positive cocci occurring in short chains and synthesizing polysaccharide "Dextran" forming a biofilm on smooth surface causing plaque. The bacteria secrete acid in microenvironment dissolving tooth enamel and causing caries.

Thus brushing of teeth without any lubricating agent could harm the smooth surface of gums leading to injury and to bleed. Thus it felt necessary to use any abrasives or chemicals which can remove the plaque, kill the bacteria and at the same time does not harm the gums.

The cleaning of tooth surface is an age old phenomenon. The Romans used the tooth pastes by adding abrasives such as crushed bones and oyster shells(12).

Ziryab developed a toothpaste which was most popular among Spain. The exact ingredients were unknown but were functional and pleasant to taste.

Neem tree twigs in Indian sub continent and Miswak in Middle east are most popular and are used as tooth brushes removing the plaque and calculus and also are now known to have antibacterial property.

Tooth paste and powder are come into use in 19th century.

One of the most important ingredients in all the tooth pastes is fluoride. It is present in small amounts in natural water, plants and animals. It helps in formation of dental enamel and to

great extent prevents dental caries. However fluoride in greater concentration in tooth paste can be acutely toxic if swallowed in large amounts.

Another important ingredient found in tooth paste is Abrasives which include aluminium hydroxide, calcium carbonate, various silicas and hydroxyapatite. These insoluble particles due their abrasive action remove plaque and calculus, thus preventing dental caries.

Some of the tooth paste has surfactant which is mainly used as foaming agent and helps in cleansing tooth surface.

Antibacterial agent like triclosan or zinc chloride is added in the tooth paste in United kingdom (11). It helps in preventing gingivitis and reducing tartar and bad breath. However studies report from United states that triclosan can combine with chlorine in tap water to form chloroform and is classified by Environmental Protection Agency as probable carcinogen(10).

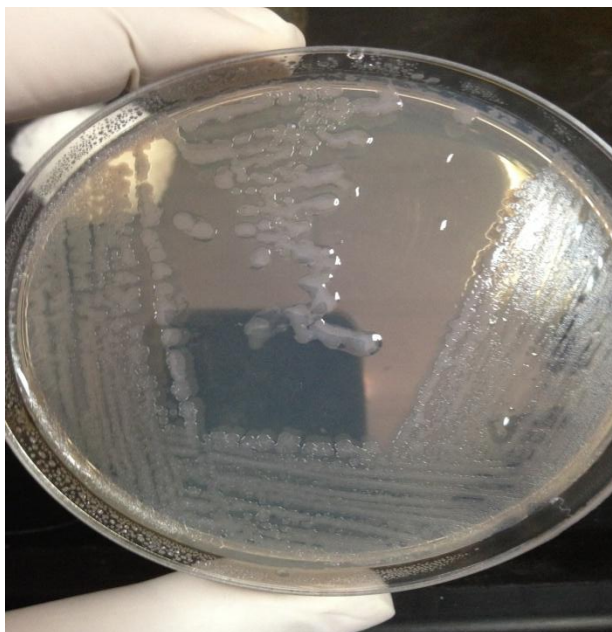
To avoid the side effects various chemicals used in the tooth paste the companies have developed herbal tooth paste. These tooth paste do not contain fluoride or sodium lauryl sulphate instead they contain baking soda, aloe, eucalyptus oil, plant extract and essential oils.

The objective of our research is to test the susceptibility of *Streptococcus mutans* the main caries causing bacteria against various tooth pastes used in Kingdom of Saudi Arabia.

Material and Method:

The *Streptococcus mutans* already isolated from our previous research study was used as test organism after confirming it by vitek. (Photo 1)

Photo1: *Streptococcus mutans* on Mitis Salivaris Agar



The *Streptococcus mutans* was further tested for its susceptibility against various toothpastes used in Kingdom of Saudi Arabia. The susceptibility was tested using Mitis Salivaris agar by Disc diffusion method of Kirby-Bauer technique (5). The toothpastes used for the study are Colgate, Close-Up, Crest, Siwak-F, Dabur-Herbal, Sensodyne, Mint Fresh, Aloe Dent and Signal.

Preparation of Inoculum:

The selected colonies of *Streptococcus mutans* are inoculated in 5ml sterile nutrient broth and incubated at 37⁰c for 2-8 hours under anaerobic condition till moderate turbidity is developed. The inoculum turbidity was compared by mixing 0.5ml of 1.75% barium chloride and 99.5ml of 0.36N sulphuric acid, as recommended by W.H.O. Wherever necessary the inoculum was diluted or incubated further to attain comparative turbidity. This culture is further used for the preparation of lawn culture on Mitis Salivaris agar.

The filter disc of standard size are cut and sterilized in autoclave. Approximately 10gm of each toothpaste was taken in a sterilized test tube containing 10ml of sterilized water. Each filter disc

was than soaked in the solution of tooth paste in test tube for about 30-40 seconds and is placed on the lawn culture of *Streptococcus mutans* on Mitis salivaris agar. The filter disc soaked in sterilized distilled water alone without tooth paste) was set as control. The plates are incubated in anaerobic jar at 37^oc for 24 hours. The experiment is done in triplicate for each toothpaste.

Result and Discussion:

The susceptibility of *Streptococcus mutans* against the above toothpastes was determined by measuring the zone of inhibition. The zone of inhibition was measured by zone reader and average of triplicate is recorded (Table 1). The highest zone of inhibition was of close-up measuring 45mm, followed by Dabur Herbal 42mm, colgate 37mm, Mint Fresh 31mm, signal 30mm, siwak-F 29mm, aloedent 25mm, crest 25mm and lowest was sensodyne 11mm (Photos 2-11).

Table 1: Efficacy of toothpastes against *Streptococcus mutans*.

S.No	Toothpastes	Zone of inhibition in mm (Average of triplicate)
1	Close-up	45mm
2	Dabur- Herbal	42mm
3	Colgate	37mm
4	Mint Fresh	31mm
5	Signal	30mm
6	Siwak-F	29mm
7	Aloedent	25mm
8	Crest	25mm
9	Sensodyne	11mm
10	Sterilized distilled water	nil

Chart 1: Pie chart showing Zone of inhibition in millimeters (Average of triplicate) of toothpastes against *Streptococcus mutans*.

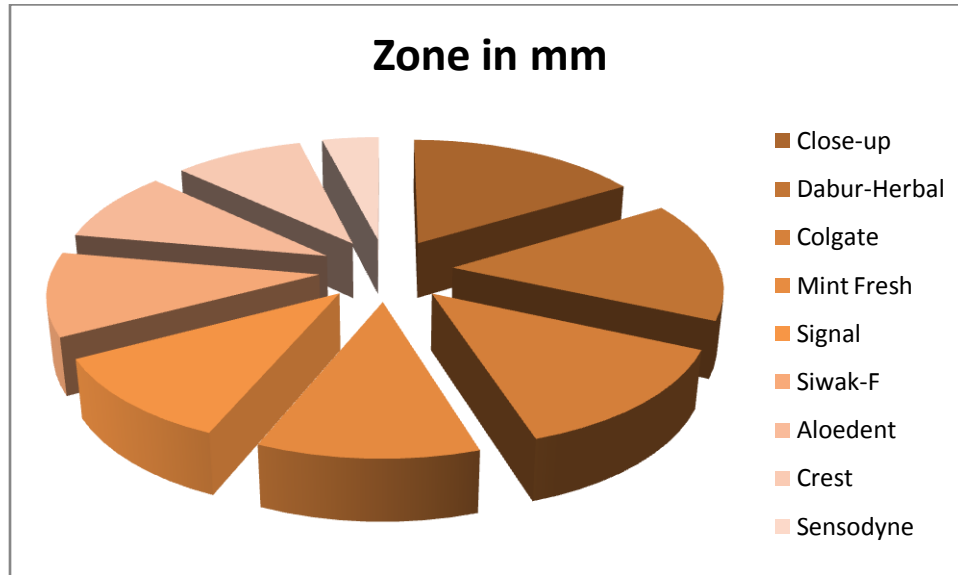


Photo 2: Effect of Closeup on *Streptococcus mutans*.

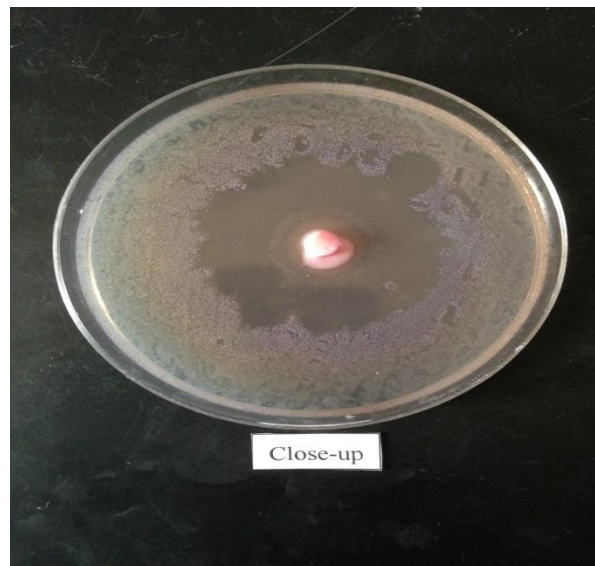


Photo 3: Effect of Dabur-Herbal on *Streptococcus mutans*.

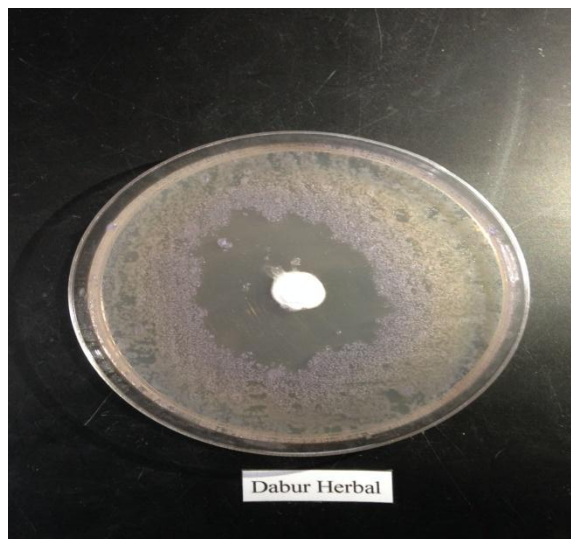


Photo 4: Effect of Colgate on *Streptococcus mutans*.

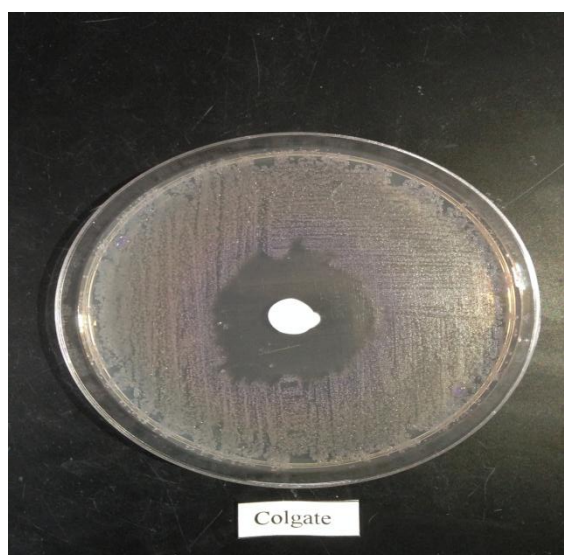


Photo 5: Effect of Mint Fresh on *Streptococcus mutans*.



Photo 6: Effect of Signal on *Streptococcus mutans*.

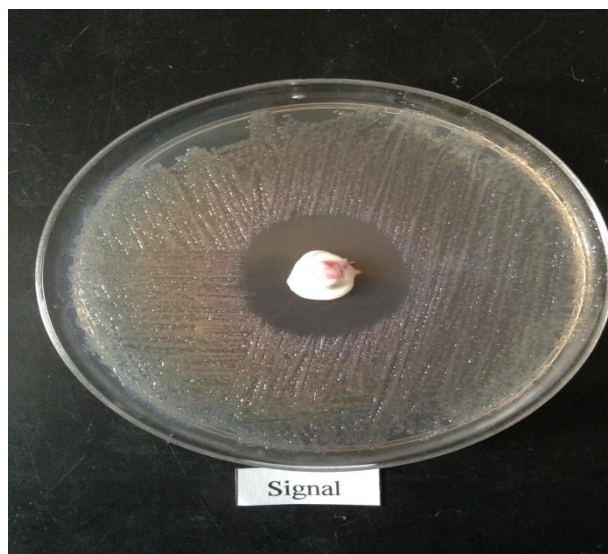


Photo 7: Effect of Sewak on *Streptococcus mutans*.

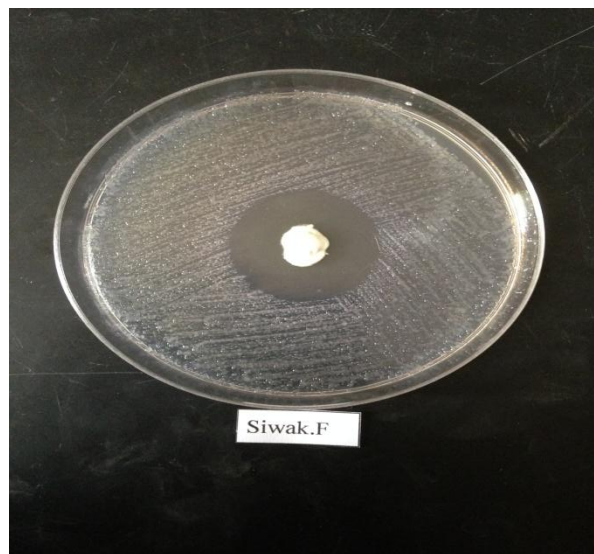


Photo 8: Effect of Aloedent on *Streptococcus mutans*.

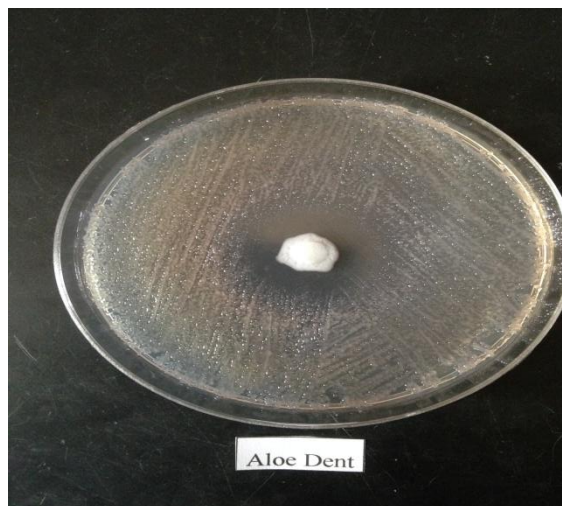


Photo 9: Effect of Crest on *Streptococcus mutans*.

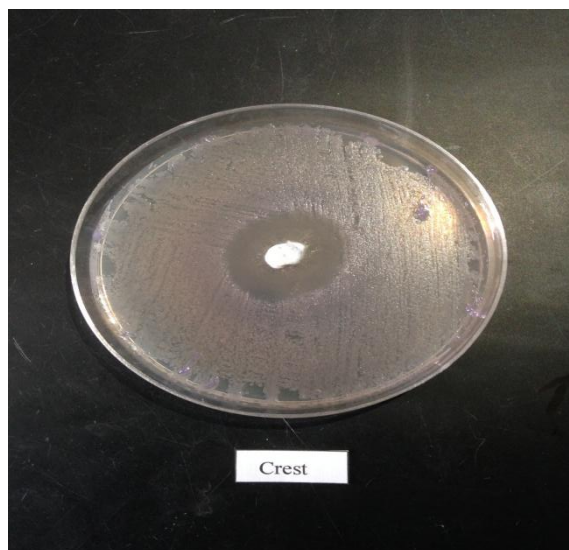


Photo 10: Effect of Sensodyne on *Streptococcus mutans*.

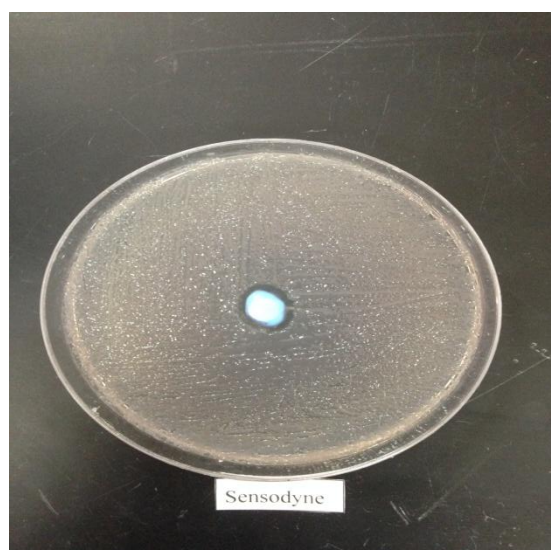


Photo 11: Control (With Sterilized Distilled Water)



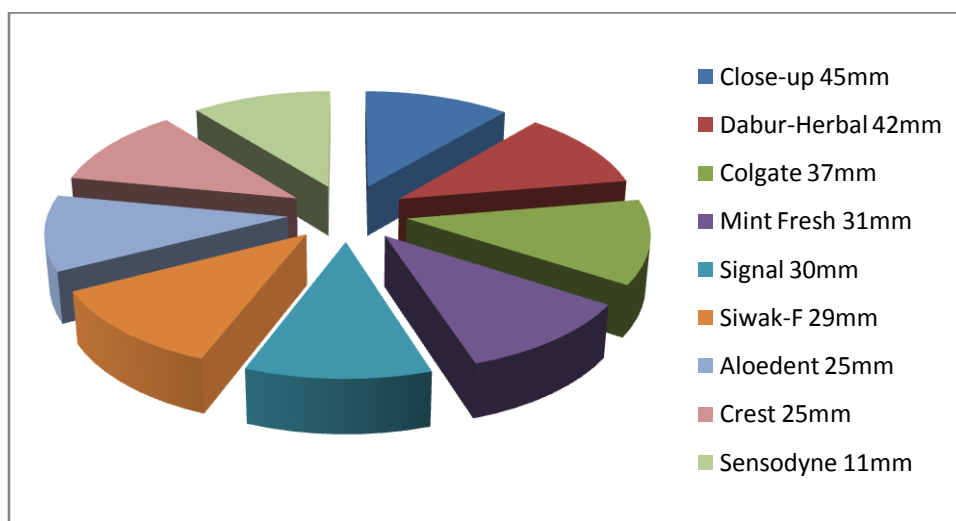
Total ten number of isolates of *Streptococcus mutans* were tested against nine different toothpastes used in Kingdom of Saudi Arabia. The average of triplicate of each of these toothpastes for the above nine isolates was determined . The consistency of results does vary among the toothpastes for its activity against the isolates. The highest consistency of results were found to be of Colgate with 95% isolates showing same results. It is followed by Siwak-F showing 93% , Close-up and Crest 90%, Dabur-Herbal 89%, Signal and Mint Fresh 87%, Sensodyne 86% and Alodent 85%.

Table 2: Percentage consistency of Isolates for its susceptibility against various toothpastes.

S.No	Toothpastes	Average of Triplicate	% consistency among Nine isolates
1	Colgate	37mm	95%
2	Siwak-F	29mm	93%
3	Close-up	45mm	90%

4	Crest	25mm	90%
5	Dabur-Herbal	42mm	89%
6	Signal	30mm	87%
7	Mint Fresh	31mm	87%
8	Sensodyne	11mm	86%
9	Aloedent	25mm	85%

Chart 1: Pie chart showing Consistency of Isolates for its susceptibility against various toothpastes.



The above toothpastes were found to have two ingredients (8) in common i.e sodium fluoride and sodium lauryl sulfate. However other ingredients varied including hydrated silica, cellulose gum and glycerine in Close-up, sodium saccharin and limonene in Crest, Miswak powder and Calcium carbonate in Siwak-F, salvadora persica extract and lemon extract in Dabur-Herbal, Cellulose and Minthol in Mint Fresh, Aloe barbadensis and Xylitol in Aloedent, Sorbitol and Phineal carbinol in Signal, Potassium Nitrate in Sensodyne and Eucalyptus and chamomile in Colgate

Table 2: Ingredients in Toothpastes.

S.No	Toothpastes	Zone of inhibition in mm (Average of triplicate)
1	Close-up	Hydrated silica, cellulose gum and glycerine
2	Dabur-Herbal	Salvadora persica extract and lemon extract
3	Colgate	Eucalyptus and chamomile
4	Mint Fresh	Cellulose and minthol
5	Signal	Sorbitol and phineal carbinol
6	Siwak-F	Miswak powder and Calcium carbonate
7	Aloedent	Aloe barbadensis and xylitol
8	Crest	Sodium saccharin and Limonene
9	Sensodyne	Potassium Nitrate

The antibacterial property was seem to be due to sodium fluoride and sodium lauryl sulfate which is common in all the toothpastes (9), however different toothpastes having varied ingredients as shown in table 2, above has the influence on the antibacterial property. This was observed by measuring the zone of inhibition of various toothpastes against *Streptococcus mutans*.

Conclusion:

In the present study it has been observed that the most effective toothpaste among the above was Closeup against *Streptococcus mutans*. However the efficacy of Colgate even though less than Close-up was more consistent among all the isolates and therefore is also the choice of preference. Since this bacteria is highly cariogenic and is the main cause of dental caries, it can easily be prevented by brushing the mouth daily before the breakfast and after the dinner.

References:

1. Carlsson J. (1968), "Plaque formation and streptococcal colonization on teeth", *Odontol.Rev.*, Vol. 19: 1-5.
2. Clarke J K (1924) "On the Bacterial Factor in the Etiology of Dental Caries" *Br J Exp Pathol.* 5(3): 141-147.
3. Dzink JL, Socransky SS, Haffajee AD(1988): "The predominant cultivable microbiota of active and inactive lesions of destructive peridontal disease". *J Clin Periodontol* 15:316:323.
4. Farwa Sarwat, Shah Ali Ul Qader (2008) " Dextran" *Int J Biol Sci* 4(6):379-386. ()
5. Kirby W and Bauer A. (1966), "Antibiotic susceptibility of bacterial isolates". *Am. J. Clin. Pathol.*, Vol. 45: 493
6. Kalesinskas P, Kačergius T (2014)." Reducing dental plaque formation and caries development. A review of current methods and implications for novel pharmaceuticals". *Stomatologija.* Vol 16(2):44-52
7. Loesche WJ. (1975), "The association of streptococcus mutans with human dental caries". *Infect. Immun.*, Vol. 11:1252-1260.
8. Lippert F(2013) "An introduction to toothpaste - its purpose, history and ingredients". *Monogr Oral Sci"* Vol 23:1-14.
9. Marinho VCC, Higgins JPT, Logan S, Sheiham A (2003) " Fluoride toothpastes for preventing dental caries in children and adolescents. *Cochrane Database Syst Rev.*(1):CD002278.
10. Rule KL, Ebbett VR, Vikesland PJ (2005). "Formation of chloroform and chlorinated organics by free-chlorine-mediated oxidation of triclosan". *Environ. Sci. Technol.* Vol 39 (9): 3176-85.
11. Trombelli L, Farina R (2013) "Efficacy of triclosan-based toothpastes in the prevention and treatment of plaque-induced periodontal and peri-implant diseases" *Minerva Stomatol.* Vol 62(3):71-88.
12. *The History of Toothpaste and Toothbrushes.* *Bbc.co.uk.* Retrieved on April 4, 2013