Antimicrobial Activity of Different Colgate Toothpastes on Oral Flora of *Bacillus subtilis*

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Abstract:

The objective of this in vitro study is clinical and laboratory evaluation of different Colgate toothpaste on antimicrobial activity against Bacillus subtilis. The initially antimicrobial activity evaluation was performed by using disc diffusion method. Water was used as a control. Disc impregnated with the Colgate toothpaste were placed in Petri dishes containing culture media inoculated suspension of Bacillus subtilis by the pour plate method. The diameter of the zone of inhibition were measured in centimetre and recorded after 24 hours incubation at 37°C for each Colgate toothpaste and result were obtained. Antimicrobial activity of all Colgate toothpaste on bacterial types were approach one another. The antimicrobial activity of Colgate maximum cavity protection, Colgate cavity protection, Colgate total advanced health and Colgate maximum white were better result.

Keywords: *Oral, Bacillus subtilis, Colgate toothpaste, Antimicrobial activity*

Introduction:

Mouth represents a dynamic ecological niche. The composition of normal microbiota varies with age microbial Flora of oral cavity is highly Complex and various surface of normal mouth are inhibited by abundant microbial community. Oral cavity is an example of a complex ecosystem which is inhibited by more than 700 bacterial species present on the tongue, teeth, inner cheeks, platelet and tonsils. *Bacillus subtilis* is an example of bacteria present in the oral microbiota. Everyday in our life day starts with the use of cleaning of teeth's by Colgate toothpaste for maintenance of excellent dental hygiene.Generally people are unknown about the potential efficiency of Colgate toothpaste.They are under the influence of the various advertisement of Colgate toothpaste. Some of these Colgate toothpaste however, have undergone sophisticated and rigorous research concerning the effectivity of their products.The bacterial species of *Bacillus* especially *Bacillus subtilis* causes dental diseases by dental caries, dental plaque. The Awareness of using Colgate toothpaste is enhance day-by-day in urban as well as rural area also. The Colgate toothpaste contains the antimicrobial substance that inhibits and kills the microorganisms that are responsible for dental diseases (Jaganet. al., 2012; Pannutiet.al. 2003 Janab, ZainabDakhil Degiam, 2010.

The success of Colgate toothpaste depends on its ability to remove oral microflora which causes the dental diseases. The Colgate toothpastes containing sodium fluoride have been widely used in all over the world. So many researchers are working on the efficiency of different Colgate toothpastes containing chemicals that are working as antimicrobial agents functions as inhibitory effect against Plaque formations (Itthagarum and wei, 1996; fine et.al. 2006). The recent investigation tries to fulfil gap on study of efficiency of various Colgate toothpastes against oral Flora using standard disc diffusion method.

Table 1: Colgate toothpastes evaluated in the study, their respective compositions

Sr. No.	Toothpaste	Composition listed on Packages
1	Colgate MaxFresh	Sodium chloride 0.24 %, Sorbitol, Water, Hydrated silica, Polyethylene glycol, Sodium Lauryl sulphate, Flavour, Cellulose gum, Tetrasodium pyrophosphate, Cocamidopropyl Betaine, Sodium saccharin, Methyl cellulose
2	Colgate cibaca	Sodium Monofluorophosphate, Sodium carboxymethyl cellulose, White film, Sorbitol, Silica, Sodium Beurylsulphate, Flavour

3	Colgate triple action	Sorbitol, Water, Hydrated silica, Sodium lauryl sulphate, Flavour, Polyethylene glycol, Tetrasodium pyrophosphate, Cocamidopropyl Betaine, Cellulose gum, Sodium saccharin, Sodium fluoride, Xanthan gum, Titanium dioxide, Pigment green, Pigment blue
4	Colgate total whitening	Sodium fluoride 0.24%, Water, Hydrated silica, Glycerine, Sorbitol, Sodium lauryl sulphate, Flavour, Cellulose gum, Sodium hydroxide, Propylene glycol, Carrageenan, Sodium saccharin, Titanium dioxide, PVM/ MA copolymer
5	Colgate Visible white toothpaste	Silica, Sorbitol, Glycerine, Polyethylene glycol, Sodium triphosphate, Tetra potassium, Pyrophosphate, Sodium lauryl sulphate, Flavour, Cocamido propyl betadine, Sodium carboxymethyl cellulose, Sodium saccharin, Sodium fluoride, Xanthan, Sodium hydroxide, Blue poly 50, Titanium dioxide in aqueous base
6	Colgate maximum cavity protection	Sodium fluoride 0.24%, Sorbitol, Water, hydrated silica, Polyethylene glycol, Cellulose gum, Sodium lauryl sulphate, Flavour, Sodium saccharin, Mica, Titanium dioxide
7	Colgate optic white	Calcium pyrophosphate, Propylene glycol, Polyethylene glycol, Copolymer, Polyethylene glycol, Glycerine, Flavour, PVR, Sodium lauryl sulphate, Tetrasodium pyrophosphate, Silica, Hydrogen peroxide, Sodium saccharin, Phosphoric acid, Sucralose, Butylated, Hydroxytoluene, Water
8	Colgate Max white	Sorbitol, Hydrated silica, Sodium lauryl sulphate, Aroma, Polyethylene glycol, Tetrasodium pyrophosphate, Cellulose gum, Cocamidopropyl Betaine, Sodium saccharin, Sodium fluoride, Hydroxypropyl methylcellulose, Limonene
9	Colgate cavity protection	Sodium monofluorophosphate 0.76%, Calcium phosphate, Dehydrate, Water, Glycerine, Sodium lauryl sulphate, Cellulose gum, Favour, Tetrasodium, Pyrophosphate, Sodium saccharin
10	Colgate total advanced health	Water, Hydrated silica, Glycerine, Sorbitol, PVM/MA copolymer, Sodium lauryl sulphate, Flavour, Cellulose gum, Sodium hydroxide, Propylene glycol, Carrageenan, Sodium saccharin, Titanium

Materials and methods

Collection of samples:

The samples were taken by swabbing the oral cavity by rotating the sterile swab and were it had limitations, dental probes; endodontic paper points and scalers were used.

Isolation and identification of bacterial species:

The samples that were collected were streak it on the surface of nutrient agar plate using four quadrant methods and incubated at 30^{o}C for 24 to 48 hours. Once samples were taken and Gram staining was done. These isolated samples were spread onto a number of freshly prepared agar plates and incubated at allow cells to form microbial colony.The media used nutrient agar. The above agar plate inoculated by streak method and incubated at 30^{o}C for 24 to 48 hours. After incubation period of 24 to 48 hours the colonies were identified by morphology, Gram staining and biochemical reactions.

Morphological identification:

Bacillus subtilis is Gram Positive bacteria, rod shaped, motile and is above 4 to 10 micrometre long and 0.25 to 1.0 micrometre in diameter. *Bacillus subtilis* is a facultative anaerobe and heavily flagellated.

Biochemical characteristics identification:

Main biochemical index of isolated *Bacillus subtilis* were determined by via the biochemical identification tubes. Strains cultured overnight at 30°C were inoculated into the biochemical identification tube via the sterile inoculating loop. Every strain inoculation was triplicated. Negative control tubes were void of bacteria results were read within 12 to 48 hours.

Table 2: Biochemical Characterization of the isolated organism

Sr. No.	Test	Result
1.	Indol production	-
2.	Methyl red	-
3.	Voges-Proskaeur	+
4.	Citrate	+
5.	Catalase	+
6.	Oxidase	+
7.	Nitrate Reduction	+
8.	Urea hydrolysis	-
9.	Hydrogen sulphide production	-

Result and discussion:

The antimicrobial activity of the Colgate toothpaste was determined by the disc diffusion method, using standard diffusion technique.The Results of the present investigation showed that the bio efficiency of Colgate maximum cavity protection, Colgate cavity protection, Colgate total advanced health, Colgate max white are highest among all the toothpaste against the test organism.The zone of inhibition were average range in Colgate triple action, Colgate total whitening, Colgate optic white.The zone of inhibition were less in Colgate MaxFresh, Colgate cibaca, Colgate Visible white toothpaste.

The results obtained in this study suggest differences among the tested dentifrices regarding antimicrobial properties. Each and every test comparing zone of inhibition amongst the oral bacteria called *Bacillus subtilis*. The reason for this could be attributed to the differences in interactions between the bacteria and different Colgate.

Table: 3. Zone of Inhibition in different Colgate toothpaste

Sr. No.	Name of Colgate toothpaste	Zone of Inhibition (in Cm)
1.	Colgate MaxFresh	2.3
2.	Colgate cibaca	2.4
3.	Colgate triple action	2.9
4.	Colgate total whitening	2.9
5.	Colgate Visible white toothpaste	2.6
6.	Colgate maximum cavity protection	3.4
7.	Colgate optic white	2.8
8.	Colgate Max white	3.0
9.	Colgate cavity protection	3.2
10.	Colgate total advanced health	3.1

When compared to water and conventional toothpaste, all Colgate toothpaste containing antimicrobial agents showed antimicrobial activity with significant differences. The addition of antimicrobial agents to conventional toothpaste to increase effectiveness in the control or elimination of microorganisms involved in a wide variety of microbial infections in the human mouth, such as *Bacillus subtilis* the primary etiological agents of dental caries. Thus, the objective of this study was evaluated commercially available Colgate

toothpastes that include the most widely used and most studied antimicrobial agents in the composition. Water was used as a control like in other studies to confirm the microbial growth around the disk.

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