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Copyright AJCEM 2024: <https://dx.doi.org/10.4314/ajcem.v25i4.10>**Short Communication****Open Access****Evaluation of the antimicrobial activity of chemical and herbal toothpastes on selected clinical microbial oral flora**\*<sup>1,2</sup>Buru, A. S., <sup>1</sup>Tola, A. P., <sup>1</sup>Akinseye, J. F., <sup>1</sup>Oluboyo, B. O., <sup>1</sup>Egbebi, A. H., and <sup>2</sup>Zagi, T. H.<sup>1</sup>Medical Microbiology and Parasitology Unit, Department of Medical Laboratory Science, Faculty of Allied Health, College of Medicine and Health sciences, Afe Babalola University, Ado-Ekiti, Ekiti State, Nigeria<sup>2</sup>Department of Medical Laboratory Science, Faculty of Allied Health Sciences, College of Health Sciences, Kaduna State University, Kaduna, Nigeria\*Correspondence to: [sunday.buru@kasu.edu.ng](mailto:sunday.buru@kasu.edu.ng)**Abstract:****Background:** Microbes are most commonly found associated with oral diseases. Globally, researchers along with the world Health Organization (WHO) suggested that this could be due to poor oral health and encourage all individuals to practice good oral hygiene using daily oral health products. The study aimed to determine the antimicrobial effects of toothpastes, as acclaimed by the manufacturers, to selected microbial flora involved in oral infections.**Methodology:** The antimicrobial activity of 6 toothpastes (3 herbal and 3 chemical) was tested against 3 clinical microbial isolates; *Staphylococcus aureus*, *Escherichia coli* and *Candida albicans*, with minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of each toothpaste determined for each microbial isolate using the broth dilution method.**Results:** Colgate exhibited the highest inhibitory activity against *S. aureus* with respective MIC and MBC of 125mg/ml and 125mg/ml, followed by Oral-B with MIC and MBC of 125mg/ml and 500mg/ml, Dabur herbal with MIC and MBC of 250mg/ml and 500mg/ml, and Longrich with MIC of 500mg/ml and MBC of 1000mg/ml. Colgate and CloseUp exhibited highest inhibitory activity against *E. coli* with respective MIC and MBC of 125mg/ml and 500mg/ml, followed by Dabur herbal with MIC of 500mg/ml but no MBC. Colgate exhibited the highest inhibitory activity against *C. albicans* with respective MIC and MBC of 125mg/ml and 500mg/ml, followed by CloseUp with MIC of 500mg/ml, Longrich with MIC of 500mg/ml and MBC of 1000mg/ml, and oral-B with MIC of 500mg/ml but no MBC. Longrich and Oral-B exhibited no MIC/MBC against *E. coli* while Gavia charcoal exhibited no MIC/MBC against all the 3 microbial isolates at the concentrations used in the assay.**Conclusion:** The results obtained showed that chemically formulated toothpastes (Colgate, CloseUp and Oral-B) had higher inhibitory activity on microbial isolates than herbal toothpastes (Longrich, Dabur herbal and Gavia charcoal). This supports the manufacturers claim for the chemical toothpastes used, however, the herbal toothpastes showed little or no inhibitory effects on the microbial flora.**Keywords:** Dental caries; Dabur; CloseUp; Oral-B; Microbial isolates

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Copyright 2024 AJCEM Open Access. This article is licensed and distributed under the terms of the Creative Commons Attribution 4.0 International License <http://creativecommons.org/licenses/by/4.0/>, which permits unrestricted use, distribution and reproduction in any medium, provided credit is given to the original author(s) and the source. Editor-in-Chief: Prof. S. S. Taiwo**Evaluation de l'activité antimicrobienne des dentifrices chimiques et à base de plantes sur une flore buccale microbienne clinique sélectionnée**\*<sup>1,2</sup>Buru, A. S., <sup>1</sup>Tola, A. P., <sup>1</sup>Akinseye, J. F., <sup>1</sup>Oluboyo, B. O., <sup>1</sup>Egbebi, A. H., et <sup>2</sup>Zagi, T. H.<sup>1</sup>Unité de Microbiologie Médicale et de Parasitologie, Département des Sciences de Laboratoire Médical, Faculté de Santé Paramédicale, Collège de Médecine et des Sciences de la Santé, Université Afe Babalola, Ado-Ekiti, État d'Ekiti, Nigéria<sup>2</sup>Département des Sciences de Laboratoire Médical, Faculté des Sciences de la Santé Paramédicale, Collège des Sciences de la Santé, Université d'État de Kaduna, Kaduna, Nigéria\*Correspondance à: [sunday.buru@kasu.edu.ng](mailto:sunday.buru@kasu.edu.ng)**Résumé:****Contexte:** Les microbes sont le plus souvent associés aux maladies bucco-dentaires. À l'échelle mondiale, des chercheurs et l'Organisation mondiale de la santé (OMS) ont suggéré que cela pourrait être dû à une mauvaise santé bucco-dentaire et ont encouragé tous les individus à pratiquer une bonne hygiène bucco-dentaire en utilisant quotidiennement des produits de santé bucco-dentaire. L'étude visait à déterminer les effets

antimicrobiens des dentifrices, tels que reconnus par les fabricants, sur une flore microbienne sélectionnée impliquée dans les infections buccales.

**Méthodologie:** L'activité antimicrobienne de 6 dentifrices (3 à base de plantes et 3 chimiques) a été testée contre 3 isolats microbiens cliniques; *Staphylococcus aureus*, *Escherichia coli* et *Candida albicans*, avec une concentration minimale inhibitrice (CMI) et une concentration bactéricide minimale (CMB) de chaque dentifrice déterminées pour chaque isolat microbien à l'aide de la méthode de dilution en bouillon.

**Résultats:** Colgate a montré l'activité inhibitrice la plus élevée contre *S. aureus* avec des CMI et CMB respectives de 125 mg/ml et 125 mg/ml, suivi par Oral-B avec des CMI et CMB de 125 mg/ml et 500 mg/ml, Dabur herbal avec des CMI et CMB de 250 mg/ml et 500 mg/ml, et Longrich avec des CMI de 500 mg/ml et CMB de 1000 mg/ml. Colgate et CloseUp ont montré l'activité inhibitrice la plus élevée contre *E. coli* avec des CMI et CMB respectives de 125 mg/ml et 500 mg/ml, suivi par Dabur herbal avec des CMI de 500 mg/ml mais pas de CMB. Colgate a montré l'activité inhibitrice la plus élevée contre *C. albicans* avec des CMI et CMB respectives de 125 mg/ml et 500 mg/ml, suivi de CloseUp avec une CMI de 500 mg/ml, Longrich avec une CMI de 500 mg/ml et une CMB de 1000 mg/ml, et Oral-B avec une CMI de 500 mg/ml mais pas de CMB. Longrich et Oral-B n'ont montré aucune CMI/CMB contre *E. coli* tandis que le charbon Gavia n'a montré aucune CMI/CMB contre les 3 isolats microbiens aux concentrations utilisées dans l'essai.

**Conclusion:** Les résultats obtenus ont montré que les dentifrices formulés chimiquement (Colgate, CloseUp et Oral-B) avaient une activité inhibitrice plus élevée sur les isolats microbiens que les dentifrices à base de plantes (Longrich, Dabur à base de plantes et charbon Gavia). Cela confirme les allégations des fabricants concernant les dentifrices chimiques utilisés, cependant, les dentifrices à base de plantes ont montré peu ou pas d'effets inhibiteurs sur la flore microbienne.

**Mots clés:** Carie dentaire; Dabur; Gros plan; Oral-B; Isolats microbiens

## Introduction:

Bacteria associated with oral diseases are one of the most common important worldwide oral health problems in the world today. It is considered one of the most common worldwide diseases related to the oral cavity affecting both children and adults (1). The oral cavity consists of microflora also that are of specific merits to the host by offering protection to the epithelial cells of the mouth from damage and promoting the digestion of some substrates. The oral cavity contains over 700 different species of bacteria and is regarded as the most complex ecosystem in the human body. Although, majority of the bacteria found in the oral cavity are considered as commensals, some of them are responsible for oral infections ranging from cavities to periodontal diseases and gum related infections and dental diseases and they include *Staphylococcus aureus*, *Escherichia coli*, *Streptococcus*, *Actinomyces*, *Lactobacillus*, *Candida albicans*, *Neisseria*, *Haemophilus* and many others (2).

Dental plaque is a common term associated with the oral cavity which consists of the biofilms or a group of bacteria that invade the mouth and can result in a variety of diseases. These plaques can be found around the surfaces of the mouth and are described as sticky colorless deposits during onset of appearance but later take up a pale yellow to brown coloration after transformation into a deposit known as tartar sometimes referred to as calculus and can be found in-between the teeth, front and back of the teeth and all along the gum. At the stage where dental plaques become tartar or calculus, self-cleaning and use of toothbrushes cannot help in its eradication.

Dental plaque can also be referred to

as microbial plaque, oral or dental biofilm or bacterial plaque biofilm. Although plaques are usually associated with oral diseases, which include cavities and other gum diseases, their formation is normal and cannot be prevented, as they develop 4-12 hours after oral cleaning through brushing (2). If not properly treated however, dental plaques can result in tooth decay which as a consequence can lead to tissue damage. These dental biofilms can be quite serious as their acidic nature causes demineralization of the teeth caused by the formation of tartar. It is therefore very essential to eradicate these plaques to avoid formation of dental caries and related gum diseases (2). The World Health Organization (WHO) highlighted the fact that the challenge of oral disease still persists globally despite great improvements in oral health of the population in several countries. The poor oral health has been observed to have profound effect on the general health and wellbeing, and several oral diseases are linked to chronic diseases that can eventually be fatal (3).

Toothpastes are daily oral care products that aim to improve oral health. However, the chemical and herbal composition and combinations of these toothpastes differ due to competition by manufacturers. Toothpastes are recognized as the best products that allegedly effectively protects both milk and permanent teeth from dental caries. While chemical toothpastes use fluoride as their active ingredient, herbal toothpastes make use of various natural extracts. There are many claims by the manufacturers that these tooth pastes have bacteriostatic and/or bactericidal activities against microbial agents of dental caries, provided by the active ingredients they contain. There are also claims that the toothpastes have a number of ingredients

with specific purposes to solve specific oral health problems (4).

Despite the acclaimed antimicrobial properties of these toothpastes, dental caries is still commonly reported globally today (3), and this is what necessitated the conduct of this study. The objective of this study is to evaluate the antimicrobial activity of selected chemical and herbal toothpastes on three clinical microbial isolates (*S. aureus*, *E. coli* and *C. albicans*) by determining the minimum inhibitory and minimum bactericidal concentrations of each toothpaste against the selected microorganisms. This is with the aim of providing evidence or otherwise of the manufacturers' claim of antimicrobial activities of these toothpastes and to put into proper perspectives of the types of healthcare products for oral hygiene practice.

## Materials and method:

### Study design and setting:

This was a descriptive, analytical evaluation study conducted at the Afe Babalola University, Ado-Ekiti, the capital and head-quarter of Ekiti State, southwestern Nigeria (5).

### Ethical clearance:

Ethical approval for the study was obtained from the Research Committee of College of Medicine and Health Sciences, Afe Babalola University, Ado Ekiti.

### Samples of toothpastes & microbial isolates:

Six commonly used toothpastes were tested against three clinical microbial isolates. These include 3 herbal [Longrich (white-tea extract), Dabur herbal, Gavia charcoal] and 3 chemical [Oral-B (pro-health), CloseUp (deep action), and Colgate] toothpastes. The test microorganisms (*S. aureus*, *E. coli* and *C. albicans*) were oral microbial isolates obtained from Ekiti State University Teaching Hospital (EKSUTH). All pure cultures were stored and maintained in nutrient slants at 4°C until use.

### Determination of minimum inhibitory and bactericidal concentrations of the toothpastes:

The minimum inhibitory concentration (MIC) of the toothpastes against the microbial isolates was determined using the broth dilution method. A 5-part serial dilution of the toothpaste in Mueller-Hinton (MH) broth was made. About 2 ml of MH broth was then pipetted into the test tubes, and two-fold dilutions of the toothpastes were prepa-

red. Approximately 2ml of the dilution was pipetted into the first test tube and properly mixed, and 2ml from the first test tube was pipetted into the second test tube, and this was repeated until the last tube, which was discarded. Approximately 0.2ml of the pure cultures of each of the test organism was pipetted into the prepared test tubes and incubated at 37°C for 24 hours. The lowest concentration of the toothpaste that inhibited the growth of the test organism was recorded as the MIC (6).

The MBC was determined from the MIC test tubes. A loopful from the tubes in the MIC test showing no visible growth was inoculated onto plain MH agar (for bacteria) and Sabouraud Dextrose Agar (SDA) plate (for *Candida*), using the streak plate method. The plates were incubated for 24 hours. The MIC plate with no visible growth was recorded as the MBC (7).

### Statistical analysis:

Data were analysed with SPSS and results presented in frequency distribution tables.

## Results:

Table 1 shows the composition of the selected toothpastes and their active ingredients. Table 2 represents the MIC and MBC values of the different toothpastes against the microbial isolates. Colgate exhibited the highest inhibitory activity against *S. aureus* with respective MIC and MBC of 125mg/ml and 125mg/ml, followed by Oral-B with MIC and MBC of 125mg/ml and 500mg/ml, Dabur herbal with MIC and MBC of 250mg/ml and 500mg/ml, and the lowest was by Longrich with MIC of 500mg/ml and MBC of 1000 mg/ml.

Colgate and CloseUp exhibited the highest inhibitory activity against *E. coli* with respective MIC and MBC of 125mg/ml and 500mg/ml, followed by Dabur herbal with MIC of 500mg/ml but no MBC. Colgate exhibited the highest inhibitory activity against *C. albicans* with respective MIC and MBC of 125mg/ml and 500mg/ml, followed by CloseUp with MIC of 500mg/ml, Longrich with MIC of 500mg/ml and MBC of 1000mg/ml, and least is oral-B with MIC of 500mg/ml but no MBC. Longrich and Oral-B exhibited produced no MIC/MBC against *E. coli* while Gavia charcoal produced no MIC/MBC against all the 3 microbial isolates at the concentrations used in the assay.

Table 1: Toothpastes with their ingredients

Toothpastes	Ingredients	Active ingredients
Longrich	Water, sorbitol, Hydrated silica, Glycerin, Sodium Lauryl Sulfate, Xylitol, PEG-32, Parfum, Menthone Glycerin Acetal, Cellulose Gum, Xanthan Gum, Sodium Saccharin, Calcium Glycerophosphate, Strontium Chloride Hexahydrate, Trisodium Phosphate, Menthol, Maltodextrin, Aloe Barbadensis Leaf Juice, Camellia Sinensis Leaf Extract, Lactic Acid, Sodium Benzoate, Potassium Sorbate, CI 77891, CI 42090	Xylitol
Dabur herbal	Sorbitol, Silica, Treated water, Poly Ethylene Glycol 1500, Sodium Lauryl Sulphate, Sodium Caboxy Methyl Cellulose, Sodium Saccharin, Tri Sodium Ortho Phosphate, Citric Acid, Preservative, FD and C Blue #1, FD and C yellow #5	Natural lemon extract, Flavor containing Natural blend of Mint, Eucalyptus, Rosemary, Chamomile, Sage, Myrrh and other natural oils.
Gavia charcoal	Coconut Charcoal, Aniseed, Licorice, Clove, Peppermint, Aloe vera, Eucalyptus, Cinnamon, Glycerine, Basil, Camphor, Calcium Glycerophosphate.	
Colgate (Max fresh)	Sorbitol, Aqua, Hydrated silica, Sodium lauryl sulfate, Aroma, PEG-12, Cellulose gum, Cocamidopropyl betaine, Sodium saccharin, sodium fluoride, Hydroxypropyl Methylcellulose, Menthol, Limonene, CI 47005, CI 74260, CI 77891.	Sodium fluoride
CloseUp	Sorbitol, Water, Hydrated silica, Sodium lauryl Sulphate, Flavour, Cellulose Gum, Sodium Saccharin, Sodium fluoride, Zinc Sulphate, Sodium hydroxide, Synthetic Fluorophlogopite, Eugenol, Limonene, Linalool, Geraniol, CI 17200, CI 16035, CI 77891.	Sodium fluoride Zinc sulphate
Oral-B	Sorbitol, Aqua, Hydrated Silica, Sodium Lauryl Sulphate, Cellulose Gum. Sodium Phosphate, Trisodium Phosphate, Sodium Saccharin, Cocamidopropyl Betaine, Sodium Fluoride, Carbomer, Limonene, Mica, Sodium Benzoate, CI 77891, CI 74160, CI42090	Sodium fluoride

Table 2: Minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of the toothpastes against the oral microbial isolates.

Toothpastes	Microbial isolates	MIC (mg/ml)	MBC (mg/ml)
Longrich	<i>Candida albicans</i>	500	1000
	<i>Staphylococcus aureus</i>	500	1000
	<i>Escherichia coli</i>	-	-
Dabur herbal	<i>Candida albicans</i>	500	1000
	<i>Staphylococcus aureus</i>	250	500
	<i>Escherichia coli</i>	500	-
Gavia charcoal	<i>Candida albicans</i>	-	-
	<i>Staphylococcus aureus</i>	-	-
	<i>Escherichia coli</i>	-	-
Colgate	<i>Candida albicans</i>	125	500
	<i>Staphylococcus aureus</i>	125	125
	<i>Escherichia coli</i>	125	500
CloseUp	<i>Candida albicans</i>	500	500
	<i>Staphylococcus aureus</i>	500	500
	<i>Escherichia coli</i>	125	500
Oral-B	<i>Candida albicans</i>	500	-
	<i>Staphylococcus aureus</i>	125	500
	<i>Escherichia coli</i>	-	-

## Discussion:

The toothpastes used in this study are amongst the most commonly used toothpastes across Nigeria as it cuts across the ones formulated using chemicals and natural products such as plant extracts and natural herbs. The manufacturers of both groups of toothpastes have claimed that their toothpastes achieve the goal of destroying bacteria that causes tooth decay and plaque formation. The MIC and MBC results from our study showed that among the chemical-formulated toothpastes, Colgate exhibited the highest inhibitory activity against most of the tested organisms, while among the herbal-formulated toothpastes, Dabur herbal exhibited high inhibitory activity against the oral microbial isolates tested, but Gavia charcoal toothpaste showed no inhibitory effect on all of the three isolates tested.

The inhibitory effects of the selected toothpastes against the isolates tested have been documented and the effectiveness of these herbal and chemical-formulated toothpastes have been reported showing without doubt that the chemical-formulated toothpastes in our study are more effective against some microbial isolates associated with oral infections than the herbal-formulated toothpastes. The effectiveness of chemical and herbal formulated toothpastes may be dependent on the concentration or presence of active antimicrobial ingredients, which may explain why the inhibitory activity of the chemical toothpastes was generally higher against the selected organisms compared to the herbal toothpastes. This agrees with the finding of the study by Oluboyo et al., (8).

The fact that our study was an *in vitro* one implies that it cannot be definitely established that our finding is directly proportional to *in vivo* activity of the toothpastes in the oral cavity. The oral cavity contains variety of commensal microbes that can cause opportunistic infections under certain conditions and considering the limited amount of time these toothpastes are used during tooth brushing, these microorganisms can still multiply tremendously, hence fluoride present in the chemical toothpastes may not be as effective as reported on certain dental diseases (9).

## Conclusion and recommendation:

The oral cavity consists of over 200 different bacteria species and fungi that are generally regarded as commensals, but they can cause disease when given the opportunity. Therefore, the use of oral health products is necessary to prevent these microbes from causing oral diseases, especially as poor oral health has been found to be responsible

for high prevalence of oral diseases.

Our study observed that Longrich, Dabur herbal and Gavia-charcoal herbal toothpastes do not possess appreciable antimicrobial properties and may perhaps need to be used more often to have any desirable antimicrobial effect. On the other hand, chemical toothpastes, especially Colgate and Oral-B, exhibited higher antimicrobial activity against the selected organisms compared to the herbal toothpastes and the results obtained supports the manufacturer's claim of their antimicrobial activity against plaque forming bacteria.

Based on our findings, we recommend the use of Colgate anti-caries and Oral-B pro-health toothpastes for tooth brushing two times daily as they both possess antimicrobial properties against the selected organisms but individuals who prefer herbal toothpastes may need to use them more frequently. It is also recommended that this study be carried out on a larger scale against other organisms capable of causing dental caries.

## Contributions of authors:

The study was conceptualised by ASB and APT. The laboratory aspects of the study and methodology application were carried out by APT, ASB, AJF, EAH, and BOO. The original draft preparation was done by ASB, APT, AJF, BOO, THZ, and EAH. Review and editing were done by ASB, APT, AJF, BOO, THZ, and EAH. All authors read and approved the final manuscript submitted for publication.

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## Conflict of interest:

No conflict of interest is declared.

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