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ANTIBACTERIAL ACTIVITY OF DIFFERENT TOOTHPASTES AND CHEWING STICKS ON SELECTED BACTERIA ISOLATED FROM THE ORAL CAVITY

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ABSTRACT: *The present study assessed the antibacterial efficacy of different toothpastes and chewing sticks purchased from a local market in Yenagoa, on selected bacteria isolated from the oral cavity. 100 oral specimens were collected from the primary school pupils of Community Primary School Otuoke, Bayelsa State. The specimens were analyzed by culture, biochemical test and agar well diffusion. Phytochemical and antibacterial properties of ethanol, and aqueous extracts of the chewing sticks were investigated in this study. The bacterial isolates were Escherichia coli, Staphylococcus aureus, Streptococcus mutans and Serratia marcescens. The highest bacterial isolate seen was E. coli, 27 (45%) while the lowest was Serratia marcescens, 2 (3.3%). The highest occurring bacterial isolates based on sex was seen in females with 32 (53.5%) while the least in male with 28 (46.7%). The age group with the highest bacterial isolate was 3–6years with 36 (60%) while the least was 11–14yaers with 8 (13.3%). Preliminary phytochemical screening of the chewing stick extracts revealed the presence of saponins, alkaloids and terpenes in Salvadora persica, and saponins, terpenes and glycosides were present in Massularia acuminata. The toothpastes (Close up and Oral-B) showed antibacterial activity ($p<0.05$) against all the bacterial isolates; however, no significant activity was observed for Close up on E. coli. The ethanol extracts of both chewing sticks (M. acuminata and S. persica) showed antibacterial activity ($p<0.05$) against the bacterial isolates than the aqueous extracts. However, the aqueous extracts of S. persica were shown to be effective against E. coli and S. mutans, with inhibition zones of 13.5mm at 62.5mg/ml concentration and 4.5mm at 125mg/ml concentration respectively; this showed significant difference ($p<0.05$), whereas no significant zone of inhibition was observed for M. acuminata. In comparison, this study showed Oral-B proved more effective than Close up, while for the chewing sticks, S. persica exhibited the greater antibacterial activity. Also, the toothpastes showed more effective antibacterial properties than the chewing sticks.*

KEYWORDS: Oral cavity, bacteria, toothpastes, chewing sticks, antibacterial properties.

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INTRODUCTION

One of the common oral infections suffered by the human population today is dental caries. It is a very harmful disease of the teeth (Moses *et al*., 2011). It causes decay of the teeth by the degradation of the acids released by the fermentation of the food particles left on the teeth after eating (Silk, 2014). Oral bacteria such *Streptococcus mutans, Lactobacillus acidophilus, Staphylococcus aureus,* etc. accumulate to form a thick whitish layer on the tooth known as dental plaque, by combining with the food remains and saliva (Marsh *et al*., 2011). The organism produces acid from the fermented foods which causes destruction of the tooth surface and form holes and cavities in the teeth (Verkaik *et al*., 2011).

One of the major factors contributing to tooth decay is poor oral hygiene due to lack of proper understanding of dental caries, which has led to a rise in the prevalence of dental caries around the world. Its treatment is expensive and requires the competence of highly skilled professionals (Tonetti *et al.,* 2015). In developing countries where there is a lack of dental care facilities for most people, it is important to educate them on dental care to promote traditional means of teeth cleaning in addition to conventional toothpastes. Toothpaste, as an abrasive, is used to promote oral health, help in removing dental plaque and food debris from the teeth, suppresses halitosis, and the active ingredients (fluoride) helps to prevent tooth decay and gum disease known as gingivitis (American Dental Association, 2010).

Although, before the use of toothpaste, early man was known to use miswak as natural hygiene tools for oral health. However, various studies have been carried out to explain the general effect of miswak on oral health, and equally elucidate certain natural chemical compounds contained in miswak which play important roles in maintaining good hygiene (Al-Bayati *et al*., 2018). Currently, miswak extract is now incorporated in toothpaste as both an active ingredient and as flavoring (Sudhir *et al.,* 2018). Thus, this study is set to determine the efficacy of different toothpastes and chewing sticks, and to determine their antibacterial activities on selected bacteria isolated from the oral cavity as a way of preventing dental caries.

LITERATURE REVIEW

Toothpaste is a gel dentifrice for cleaning and maintaining the beauty and health of teeth, and to promote oral hygiene. Toothpaste as an abrasive aid in removing dental plaque and food from the teeth, helps in suppressing foul-smelling breath to help prevent tooth decay and gum disease (American Dental Association, 2010). However large amounts of swallowed toothpaste while bushing can be toxic to humans (National Library of Medicine "Toothpaste overdose", 2014). Toothpaste contains 20%–42% water, and is derived from various components which include mainly abrasives, fluoride and detergents (Hujoel *et al*., 2019). These soluble particles are designed to help remove plaque and calculus from the teeth to prevent the accumulation of tartar and reduce cavities and periodontal disease (Hujeol, 2019). Furthermore, this gel dentifrice also contains antimicrobial agents like triclosan (Riley and Lamont, 2013), although clinical trials have shown the use of high fluoride dentifrices (Walsh *et al*., 2019) to reduce the amount of plaque accumulated and decrease the number of *Streptococcus mutans* and Lactobacilli (Ekstrand, 2016).

Chewing sticks are twigs or roots of certain plants that are chewed till one end becomes frayed. The end can be used to brush against the teeth (Panati, 2013), while the other end can be used

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as a toothpick (Zheng *et al*., 2013). Most common plants used as chewing sticks have high contents of tannins (astringent and antibacterial) or other compounds which benefit healthy gums and teeth (Shetty *et al*., 2010). The purpose of oral hygiene—using toothpaste and chewing sticks—is to efficiently reduce oral bacterial flora and promote dental health (America Dental Association, 2010).

MATERIALS AND METHODS

Two brands of toothpastes were purchased from a local market (Swali market) in Yenagoa, Bayelsa State, South-South Nigeria. Their batch numbers, registration numbers, manufacture and expiry dates as well as the presence of the manufacturer's seal were taken note off. Two different chewing sticks (*Massularia acuminata* and *Salvadora persica*) were also purchased from the same market.

Specimen Collection, Processing and Identification

A hundred oral specimens were collected from primary school pupils of Community Primary School, Otuoke, Bayelsa state. Consent was given by the school management. The specimens were cultured on blood agar, chocolate agar, MacConkey agar, CLED agar and nutrient agar. After 24 hours of incubation at 37^{o}C, the isolates were checked for growth. Pure isolates obtained were characterized using gram staining technique and biochemical tests which include catalase, oxiadase and coagulase. Haemolysis test was also carried out.

Preparation of Chewing Stick Extracts and Toothpaste Solution

500g of *Salvadora persica* and *Massularia acuminata* chewing sticks were cut into pieces and ground into powder with a commercially available food blender. 100ml of 95% ethanol and sterile water was added to 10g of the powder in a sterile well capped flask, left for 7 days at room temperature and then filtered using number 1 Whatman filter paper. The extract then evaporated in a mantle heater at 40^{O}C until ethanol was removed while the aqueous extract was centrifuged at 2000pm for 10 minutes (Al-lafi and Abadueh, 1995). The supernatant was passed through a 0.45mm membrane filter. The extracts were preserved in sterile screw-capped vials in the refrigerator for further use (Al-koubaisi, 2001).

The different toothpaste solutions were made by mixing the calculated amount of toothpaste (2g) in measured volume (2ml) of sterile pyrogen-free distilled water to give 1:1 dilution; they were further diluted in sterile water and three different dilutions of 1:2, 1:4 and 1:8 were made.

Phytochemical Analysis

Phytochemical screening were carried out on the two different chewing sticks *Salvadora persica* (Miswak) and *Massularia acuminata* (Pako Ijebu) to determine the presence of the following constituents: flavonoids, tannins, saponins, terpenes, basic alkaloids and glycosides, using the method described by Sony *et al.* (2011) and Thilagavati *et al.* (2015).

In-vitro Antibacterial Sensitivity Testing

The toothpastes and chewing sticks were tested on the isolates to determine their efficacy at various concentrations.

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Preparation of Concentration for the Chewing Stick Extract

2ml of the plant extract was measured using a 2ml syringe and was transferred into a test tube containing 2ml of 70% dimethyl sulfoxide (DMSO) to give a concentration of 500µg/ml; they were further diluted in 70% DMSO to give three different concentrations of 250, 125 and 62.5mg/ml respectively.

Preparation of McFarland Standard

A 0.5 McFarland standard was prepared by mixing 0.05ml of barium chloride dihydrate, with 9.95ml of 1% sulfuric acid (Cockerill *et al*., 2012). The standard was compared visually to a suspension of bacteria in sterile saline.

Antibacterial Assay

The antibacterial assay was determined using modified agar well diffusion. This method was carried out by streaking a loopful of each of the test isolates onto a prepared nutrient agar plate. A sterile 8mm cork-borer was used to cut one central and four wells at equidistance in each of the plates. 0.2ml of the toothpastes dilutions was introduced into each of the four wells while the same amount of sterile pyrogen-free water was introduced into the first well (central) as control. This method was repeated for the chewing extracts with the same volume of 70% DMSO serving as control in this case, and the plates were allowed to stay on a horizontal surface for one hour to enable the substances to diffuse before incubating at 37ºC for 24 hours. Zones of inhibition were measured in mm after incubation to determine the antibacterial efficacy. All experiments were performed in duplicate (Sarmad, 2013).

Statistical Analysis

The data were analysed using Anova IBM® SPSS® statistics version 21. One-way ANOVA was used to compare the mean value of the outcome variable followed by post hoc test. The significance level was set at $p<0.05$.

RESULTS

Table 1 shows the distribution of bacterial isolates based on sex. Out of the one hundred (100) specimens examined, a total of 60 isolates were obtained—28 from males and 32 from females. The frequency of bacteria isolated were *Escherichia coli*, 27 (45%); *Staphylococcus aureus*, 21 (35%); *Streptococcus mutans*, 10 (16.7%); and *Serratia marsescens*, 2 (3.3%).

Table 2 shows the frequency distribution of bacterial isolates based on age. Of the sixty bacterial isolates, 36 were obtained from pupils between the ages 3–6 years, 16 from 7–10 years and eight from 11–14 years. The highest number of isolates was *E. coli* with 20 (33.3%) as seen between the age 3–6 years while the least was 1 (1.7%) for *S. mutans*, as seen between the 3–6 years and *S. marsescens* between the ages 7–10 years and 11–14 years respectively.

The phytochemical constituents of *Massularia acuminate* and *Salvadora persica*, which showed the presence of saponins and terpenes in both plant extracts, was seen in relative abundance. However, the concentration of saponin was higher in *M. acuminata* while *S. persica* had higher concentration of terpenes. Alkaloids were present in *S. persica* only, while

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glycosides were seen in *M. acuminata* only. Flavonoids were absent in both extracts as shown in **Table 3**.

Table 4 shows the mean zone of inhibition (mm) of toothpastes on bacterial isolates at various concentrations. The result showed that Oral-B was more effective with a zone of inhibition of 19.5mm for *E. coli* while there was no significant zone of inhibition (0mm) for Close up.

Oral-B exhibited greater efficacy against *S. aureus* with a zone of inhibition of 15.5mm than Close up with 5.5mm. However, both dentifrices showed a level of significance ($p<0.05$) at all concentrations.

Close up showed greater effectiveness against *S. mutans* with a zone of inhibition at 18.5mm while Oral-B at 18mm. There was significant difference at $p< 0.05$ in all the concentrations.

The mean zone of inhibition of toothpastes on *S. marcescens* showed that Close up exhibited greater efficacy at 19.5mm than Oral-B at 14.5mm. At a significance level of $p<0.05$, all concentrations of both toothpastes were significant.

The mean zone of inhibition of the various chewing stick extracts on *E. coli* at various concentrations is shown in **Table 5**. The ethanol extract of both chewing sticks showed antibacterial properties with a mean zone ranging from 18.5mm–15.5mm and 10.5mm–4.0mm for *S. persica* and *M. acuminata* respectively. The largest zone of inhibition was observed at a concentration of 125mg/ml for *S. persica,* while observed at 500mg/ml for *M. acuminata* in which efficacy decreased as concentration decreased. However, the aqueous extract exhibited antibacterial properties at the concentration of 62.5mg/ml for *S. persica* while there was no significant zone of inhibition by *M. acuminata* at $p<0.05$. The aqueous extract of *S. persica* exhibited more antibacterial properties as the concentration decreased.

The result of the mean zone of inhibition of various chewing stick extracts on *S. aureus* at various concentrations is shown in **Table 6**. For *S. persica,* the ethanol extract produced a mean zone range of 13.5mm–11.0mm, and 6.5mm–4.0mm for *M. acuminata.* Whereas, the aqueous extracts of both chewing sticks exhibited no antibacterial properties against *S. aureus.* The largest zone of inhibition was seen with both ethanol extracts at a concentration of 125mg/ml for *S. persica* and 500mg/ml for *M. acuminata*.

Table 7 shows the mean zone of inhibition of various chewing sticks on *S. mutans* at various concentrations. The result showed the ethanol extract of *M. acuminata* exhibited greater efficacy with a mean zone range of 10.0mm–4.0mm than *S. persica* with a mean zone range of 9.0mm–7.5mm. For the aqueous extract, the efficacy was from 4.5mm–2.5mm for *S. persica.* The largest zone of inhibition for *S. persica* and *M. acuminata* was seen at 125mg/ml and 500mg/ml respectively, with ethanol extracts, while for aqueous extract of *S. persica* at 125mg/ml, *M. acuminata* exhibited no significant zone of inhibition at $p<0.05$ significance level.

Table 8 shows the mean zone of inhibition of various chewing sticks on *S. marcescens.* The ethanol extracts of *S. persica* and *M. acuminata* exhibited antibacterial properties with the largest zone at a concentrations of 62.5mg/ml and 500mg/ml respectively. However, the aqueous extracts of both chewing sticks exhibited no significant zone of inhibition at $p<0.05$.

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Table 1: Distribution of bacterial isolates based on sex

Isolates	Number of isolates	Male	Female	Total (%)
Escherichia coli	27	13 (21.7%)	14 (23.3%)	27 (45.0%)
Staphylococcus aureus	21	11 (18.3%)	10 (16.7%)	21 (35.0%)
Streptococcus mutans	10	3 (5.0%)	7 (11.7%)	10 (16.7%)
Serratia marsescens	2	1 (1.7%)	1 (1.7%)	2 (3.3%)
Total	60	28 (46.7)	32 (53.4)	60 (100%)

Experiments were done in duplicates ± S.D of two replicates

Table 2: Frequency distribution of bacterial isolates based on age

Age Interval	*Escherichia coli*	*S. aureus*	*S. mutans*	*S. marsescens*	Total (Years)
3–6	20 (33.3%)	15 (25.0%)	1 (1.7%)	0 (0%)	36 (60%)
7–10	5 (8.3%)	3 (5.0%)	7 (11.7%)	1 (1.7%)	16 (26.7%)
11–14	2 (3.3%)	3 (5.0%)	2 (3.3%)	1 (1.7%)	8 (13.3%)
Total	27 (44.9%)	21 (35.0%)	10 (16.7%)	2 (3.4%)	60 (100%)

Experiments were done in duplicates ± S.D of two replicates

Key:

S. aureus = Staphylococcus aureus
S. mutans = Streptococcus mutans
S. marcescens = Serratia marcescens

Table 3: Phytochemical analysis

Phytochemicals	*Salvadora persica*	*Massularia acuminata*
Flavonoids	-	-
Saponins	++	+++
Tannins	-	-
Alkaloids	+	-
Reducing sugars	-	+
Terpenes	++	+

Key:
+++ = Highly present
++ = Moderately present
+ = Slightly present
- = Absent

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Table 4: Mean zones of inhibition (mm) of toothpastes on bacterial isolates

Isolates	Toothpaste	Concentration 1:1	1:2	1:4	1:8
E. coli	Close up	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
	Oral B	19.5±1.41	14.0±2.83	8.5±0.71	0.00±0.00
S. aureus	Close up	5.5±0.71	0.00±0.00	0.00±0.00	0.00±0.00
Oral B	15.5±2.12	13.0±1.41	5.0±0.00	0.00±0.00	
Streptococcus	Close up	18.5±0.71	11.5±0.71	8.5±0.71	5.0±0.00
mutans	Oral B	18.0±1.4	10.0±0.00	7.0±0.00	0.00±0.00
Serratia	Close up	19.5±0.71	12.5±0.71	0.00±0.00	0.00±0.00
marcescens	Oral B	14.5±0.71	10.5±0.71	9.5±0.71	0.00±0.00
Control A		0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
Control B		0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00

Values are mean inhibition zone (mm) ± S.D of two replicates

Key:
Control A = Control for Close up
Control B = Control for Oral-B

Table 5: Mean zone of inhibition (mm) of chewing stick extracts on *Escherichia coli*

Dilution	500mg/ml	250mg/ml	125 mg/ml	62.5mg/ml
ASP	0.00±0.00	0.00±0.00	10.0±1.4	13.5±0.71
ESP	0.00±0.00	16.0±1.41	18.5±0.71	15.5±0.71
AMA	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
EMA	10.5±0.71	8.0±0.00	4.0±0.00	0.00±0.00
cASP	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
cESP	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
cAMA	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
cEMA	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00

Values are mean inhibition zone (mm) ± S.D of two replicates
Key:
ASP = Aqueous extract of *Salvadora persica*
ESP = Ethanol extract of *salvadora persica*
cASP = Control for aqueous extract of *salvadora persica*
cESP = Control for ethanol extract of *salvadora persica*
AMA = Aqueous extract of *Massularia acuminata*
EMA = Ethanol extract of *Massularia acuminata*
cAMA = Control for aqueous extract of *Massularia acuminata*
cEMA = Control for ethanol extract of *Massularia acuminata*

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Table 6: Mean zones of inhibition (mm) of chewing stick extracts on *Staphylococcus aureus*

Dilution	500mg/ml	250mg/ml	125 mg/ml	62.5mg/ml
ASP	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
ESP	0.00±0.00	8.5±2.12	13.5±2.12	11.0±1.41
AMA	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
EMA	6.5±0.71	5.5±0.71	4.0±0.00	0.00±0.00
cASP	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
cESP	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
cAMA	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
cEMA	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00

Values are mean inhibition zones (mm) ± S.D of two replicates
Key:
ASP = Aqueous extract of *Salvadora persica*
ESP = Ethanol extract of *salvadora persica*
cASP = Control for aqueous extract of *salvadora persica*
cESP = Control for ethanol extract of *salvadora persica*
AMA = Aqueous extract of *Massularia acuminata*
EMA = Ethanol extract of *Massularia acuminata*
cAMA = Control for aqueous extract of *Massularia acuminata*
cEMA = Control for ethanol extract of *Massularia acuminata*

Table 7: Mean zones of inhibition (mm) of chewing stick extracts on *Streptococcus mutans*

Dilution	500mg/ml	250mg/ml	125 mg/ml	62.5mg/ml
ASP	3.00±0.00	3.5±0.00	4.5±0.71	2.5±0.71
ESP	0.00±0.00	7.5±0.71	9.0±0.00	7.5.0±0.71
AMA	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
EMA	10.0±0.00	8.5±0.71	7.0±0.00	4.00±0.00
cASP	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
cESP	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
cAMA	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
cEMA	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00

Values are mean inhibition zones (mm) ± S.D of two replicates
Key:
ASP = Aqueous extract of *Salvadora persica*
ESP = Ethanol extract of *salvadora persica*
cASP = Control for aqueous extract of *salvadora persica*
cESP = Control for ethanol extract of *salvadora persica*
AMA = Aqueous extract of *Massularia acuminata*
EMA = Ethanol extract of *Massularia acuminata*
cAMA = Control for aqueous extract of *Massularia acuminata*
cEMA = Control for ethanol extract of *Massularia acuminata*

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Table 8: Mean zones of inhibition (mm) of chewing stick extracts on *Serratia marcescens*

Dilution	500mg/ml	250mg/ml	125 mg/ml	62.5mg/ml
ASP	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
ESP	0.00±0.00	3.5±0.71		10.0±1.41
11.0±1.41				
AMA	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
EMA	5.50.±0.71	0.00±0.00	0.00±0.00	0.00±0.00
cASP	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
cESP	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
cAMA	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
cEMA	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00

Values are mean inhibition zones (mm) ± S.D of two replicates
Key:
ASP = Aqueous extract of *Salvadora persica*
ESP = Ethanol extract of *salvadora persica*
cASP = Control for aqueous extract of *salvadora persica*
cESP = Control for ethanol extract of *salvadora persica*
AMA = Aqueous extract of *Massularia acuminata*
EMA = Ethanol extract of *Massularia acuminata*
cAMA = Control for aqueous extract of *Massularia acuminata*
cEMA = Control for ethanol extract of *Massularia acuminate*

DISCUSSION

In this study, both gram negative and gram-positive bacteria were subjected to different toothpastes (Close up and Oral-B) and chewing sticks (*Salvadora persica* and *Massularia acumunata*) to test for their efficacy. The result of the current research clearly showed that the chewing sticks and the toothpastes inhibited the growth of bacteria causing tooth decay. However, the range of effectiveness is concentration-dependent and varied against the different tested bacteria (Sudhir *et al*., 2018). The bacterial isolates seen were *Escherichia coli*, S. *aureus*, *S. mutans* and *S. marcescens.* The presence of these bacteria shows they are pathogenic and could be responsible for oral diseases (Anyiam *et al*., 2016). The highest occurring bacterial isolate was *E. coli* with 27 (45.0%) while the lowest was *S. marcescens* with 2 (3.3%). The age group seen with the highest bacterial isolates was 3–6 years with 36 (60%), while the least was ages 11–14 years with 8 (13.3%). This could be as a result of exposure to different diets and lack of good oral hygiene practices.

The findings from this study revealed the presence of alkaloids, saponins, terpenes, and the absence of flavonoids and tannins in the extracts. The efficacy of the plant materials could be as a result of the presence of some basic phytochemicals such as saponins and terpenes in both extracts, and the presence of glycosides and alkaloids in *M. acumunata* and *S. persica* respectively. In comparison, the extracts of *S. persica* exhibited greater antibacterial properties than *M. acuminata*. This could be due to the absence of alkaloids in *M. acuminata* which are known to exhibit antibacterial properties (Rao *et al*., 2000).

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The evaluation of the antibacterial properties of extracts on bacterial isolates showed they all possess antibacterial properties. The antibacterial activity was seen at varying concentrations indicating that the chewing stick extract had a broad antibacterial spectrum.

The aqueous extract of both chewing sticks exhibited no antibacterial properties against *S. aureus.* This result agrees with a similar work done by Naziru *et al.,* (2015) who compared the antibacterial activity of chewing sticks and toothpastes commonly used in Kano (Nigeria) on *Staphylococcus* and *Streptococcus* spp. However, the aqueous extract of *S. persica* exhibited significant antibacterial properties at $p<0.05$ on *E. coli* and *S. mutans* (only at a concentration of 125mg/ml and 62.5mg/l respectively), which seems to agree with the findings of Sarmad, (2013) who revealed that the aqueous extract of *S. persica* showed antibacterial properties against many cariogenic bacteria including *E. coli* and *S. mutans*. There was no significant zone of inhibition for *M. acuminata* on all the bacterial isolates.

The ethanol extracts of the chewing sticks showed greater inhibitory effect than aqueous extracts; this is in accordance with the research carried out by Odeleye *et al*., (2016). The aqueous extracts of the chewing stick showed a very poor inhibitory activity against the bacterial isolates. This may be due to the poor solubility nature of the active principles of the plant material.

Of the toothpastes used, Oral-B proved to be more efficacious (based on mean zone of inhibition) than Close up; this is in agreement with the research carried out by Josephs *et al*., (2016) on the evaluation of antibacterial activity of toothpastes on some selected microorganisms.

Comparably, the toothpastes were more effective than the aqueous and ethanol extracts of both chewing sticks on all bacterial isolates (based on mean zone of inhibition), except for Close up which had no inhibitory effect against *E. coli*. This agrees with the previous study by Sarmad, (2013) who concluded that toothpastes were more effective in inhibiting cariogenic and pathogenic bacteria than *S. persica.* However, this is contrary to the result obtained by Odeleye *et al*., (2016) who concluded that toothpastes had weaker antimicrobial properties than chewing sticks, on his work on the antibacterial activity of Macleans and Close up in comparison to some chewing sticks, including *M. acuminata*.

CONCLUSION

The finding suggests that the ethanol extracts of both chewing sticks (*S. persica* and *M. acuminata*) as well as the two different toothpastes (Close up and Oral-B) have an inhibitory effect on all tested isolates, with the exception of Close up which had no inhibitory effect against *E. coli.* Advocacy should be planned to increase the use of chewing sticks based on the evidence of the current trial, especially in the developing countries with lack of funds and confined oral health care services for the general population. Also, measures should be put in place to thoroughly check for the efficacy of all the toothpastes sold within the country to ensure they are all effective against pathogenic oral bacteria. However, this research was an in-vitro test; therefore, further research needs to be conducted on the in-vivo efficacy of these toothpastes and chewing sticks in managing bacterial load in the oral cavity, to determine the detrimental effects of toothpastes and chewing sticks on beneficial oral bacteria. In addition, further studies should be conducted to determine the time interval of the inhibitory effect of

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the toothpastes and chewing sticks on oral pathogens, since not all bacteria found in the oral cavity are pathogenic.

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