

Phytochemical and Microbiological Studies on the Effect of Leaf Extract of *Mangifera indica* on Dental Caries Pathogens

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ABSTRACT

There is high need for medicinal plant to combat pathogen because of the adverse effect of synthetic drugs. Phytochemical screening and antimicrobial effects of *Mangifera indica* leaves were examined. The leaves were gotten from Agbani Enugu State, dried under room temperature for 48 days and ground with a milling machine in Chemistry Department, Enugu State University of Science and Technology. The solvent used for extraction were ethanol, methanol, N-hexane and water. The extraction was done via cold maceration. The phytochemicals screened for were; terpenoids, steroids, glycosides, phenols, saponins, tannins and alkaloids. The extracts were divided into dialyzed fraction and undialyzed fraction. The dialyzed was done for 48 hours in a refrigerator. In the antimicrobial screening, the isolated organisms were; *Streptococcus mutans*, *Escherichia coli*, *Staphylococcus aureus*, *Lactobacillus spp* and *Candida albicans*. The result of the phytochemical analysis showed that the methanolic extract yielded abundant quantities of flavonoids, steroids and glycosides, moderate quantities of terpenoids, saponins, tannins and phenols with absence of alkaloid. Ethanolic extract yielded abundant quantity of moderate quantities of flavonoids, terpenoids, saponins, tannins and phenols with absence of alkaloid. The N-Hexane extract yielded only moderate quantity of terpenoids and little quantities of steroids and tannins. Water extract yielded moderate quantities of steroid and absence of the rest. The antimicrobial screening test showed that the dialysed extracts had higher sensitivity against the isolated dental caries pathogens than undialyzed. The most inhibited organism was *Staphylococcus aureus* with IZD 9mm and MIC 6.25mg/ml. With the results obtained, it is clearly obvious that concentrated extracts from medicinal plants will help to activate more inhibition against dental caries pathogens and other microorganisms, and there would be little need to use N-Hexane in extraction of the leaf of *M. indica* since it has a little extraction power.

Keywords: Medicinal plant, *Mangifera indica*, Microorganism and antimicrobial

INTRODUCTION

The use of medicinal plants as herbal remedies to prevent and cure ailments differ from community to community [1], [2], [3], [4]. The advent of science into the search for antibiotics largely depends on some of these medicinal plants as raw materials [5]. For many years, medicine had depended exclusively on leaves, flowers and barks of plants; only recently have synthetic drugs come into use and in

many instances, these are carbon copies of chemicals identified in plants [6].

According to [7] medicinal plant is any plant in which one or more of its organ, contains substances that can be used for therapeutic purposes or which are precursors for the synthesis of useful drugs [8]. *Mangifera indica* is one of the most popular choices of fruit in India and it is highly prized as desert fruit.

Mango belongs to the genus *Mangifera* of the family *Anacardiaceae*. There are over 1000 named mango varieties throughout the world, which is a testament to their value to mankind. Mango is a common garden tree throughout the tropics. *Mangifera indica* have reddish or purplish leaves when tender and new and grow into dark green colour with a pale under side. They are fleshy and shiny with sharp tips. They provide shade for passerby in the hot season. They contain phenols, flavonoids, tannins, alkaloid and so many active ingredients that are known to be very effective in reducing the activities of micro-organisms [9], [10], [11]. The health benefits of these leaves are; in treating diabetes, controlling blood pressure and anxiety, Asthma, bronchitis and cough, Hiccups and sore throat, gum problems, kidney stone, ear aches and

digestion [12]. Apart from the leaves, the stem of *Mangifera* can also be used in combination with *Citrus sinensis* leaves to prepare decoction that can be taken against malaria parasite [13]. Dried mango flowers containing 15% tannin serve as astringents in cases of diarrhea and chronic dysentery [14], [15]. Photochemicals present in the leaf extracts of *Mangifera indica* have been noted to have high antimicrobial effect especially those that causes tooth decay; these micro-organisms are collectively called dental caries or cariogenic organisms [15].

Aim

The aim of this research is to determine the phytochemical properties of the leaf extracts of *Mangifera indica* and their effects in varying concentration against dental caries pathogen.

MATERIALS AND METHODS

Identification of sample

Mangifera indica .L. leaves were collected from Agbani Enugu West Local Government Area, Enugu State, Nigeria and official identification was done by Onyeukwu O of *herbarium* unit, University of Nigeria Nsukka. *Mangifera indica* .L. names (Igbo: Mangolo, Hausa: Mangworo, Yoruba: Mangoro, U.S.A: Mango). There are over 1000 named species (varieties), the trees are large trees, reaching ten to thirty (10-30) metres in height, with a broad round canopy, that may with age attain 30-38 metres in width, or with a more upright oval slender crown.

Preparation of Sample

The leaves of *Mangifera indica* .L. collected was carried out by plucking the leaves together with their stalk from the tree, this was followed by washing in distilled water to reduce the microbial load present on the leaves surfaces. The leaves were dried in a room, using the room temperature (37%), for 48 days. The dried leaves were grinded into fine powder using a milling machine in the chemistry department in Enugu State University of Science and Technology.

Extraction Procedure

The powdered leaves were extracted with four solvents; Ethanol, Methanol, N-hexane and Water (H₂O). This is done in

order to trap the bioactive ingredients present in the leaves. 100g of the powdered leaves was weighed for each of the solvents and poured into an air tight container, 300mls of the solvent each was measured and introduced into each container, then labeled for easy identification. The container was tightly sealed by wrapping with a masking tape. The set up was allowed for 48 hours, then filtered using Watman No.1 filter paper in order to concentrate the extract. The four extracts (ethanol, methanol, N-Hexane, and water (H₂O)) were divided into two portions each. The first portion is to be used directly for phytochemical screening and antimicrobial activities on dental caries pathogens, this portion was stored with a phosphate buffer solution of pH 6.8. While the second portions were dialyzed to further concentrate the extracts, then used also for antimicrobial screening against dental caries pathogen.

Dialysis Procedure

The dialysis bags were provided by Department of Applied Microbiology and Brewing, Enugu State University of Science and Technology. 100ml of each extracts (ethanol, methanol, N-Hexane and water (H₂O)) were poured into the dialysis bag, then sealed using a thread (Hair-thread). 5 molar concentration sucrose

solution was prepared, then the sealed bags were suspended inside the solution in such a way that they were suspended inside the solution in such a way that they were made to stand upright in the container by hanging them to a bridge (stick) made across the vessel containing the 5molar concentration sucrose solution. This was allowed to dialyze for three days, after which the concentrates were collected in an air tight container and equal volumes of phosphate buffer solution pH 6.8 was added and kept in a refrigerator.

Phytochemical Tests

Phytochemical screening of the undialyzed portions of the extracts (ethanol, methanol, N-Hexane and water (H₂O) were carried out to determine which solvent(s) extracted more of the bioactive ingredients in the leaves of *Mangifera indica* .L.

Test for Alkaloids

Three reagents were used to determine the presence of alkaloids, hence each extract was divided into the three test tubes. For the first reagent; Dragendroff reagent, Add 1ml of the extracts and evaporate to dryness followed by addition of aqueous HCl and shaken. It was filtered, then few drops of the reagent was added to the filtrate. Turbidity or precipitate indicates the presence.

For the second reagent; Wagner's reagent. Add 1ml of the extract into test tubes, then few drops of the reagent will also be added to each test tubes and stirred. Precipitation indicates possible presence of alkaloids. For the third reagent; Mayer's reagent, Add 1ml of the extract to each tube. Turbidity indicates the presence of alkaloid [14].

Test for saponins

Add 1ml of each of the extracts to test tubes, 5ml of distilled water was also added and stirred vigorously. A steady foam formation indicates the presence of saponin [14].

Test for tannins

Add 1ml of each sample into test tubes, then few drops of FeCl₃ was added. Presence of blue or greenish black [14].

Test for phenols

Add 1ml of the extracts each to test tubes, then one drop of FeCl₃ (50%) was also added and stirred, an intense blue colour indicates the presence of phenol [14].

Test of steroids

Add 1ml of each extracts, plus 2-3 drops of acetic anhydride plus H₂SO₄ (conc). Bubbles formation indicates the presence of terpenoid [14].

Test for glycosides

Add 1ml of extracts, plus few drops of FeCl₃, plus 1ml of H₂SO₄ (15%). Reddish brown colour indicates its presence or bluish-green colour [14].

Test for terpenoids

Add 1ml of extracts to test tubes, then acetic anhydride, followed by chloroform and stirred. After that H₂SO₄ was added. Observe red or violet colour for the presence of terpenoids [14].

Test for flavonoids

Add 1ml of extract to test tubes, add 1.5ml of C₂H₅OH (150%), heat for 5 min, then add Mg (salt) + HCl (conc.). Observe red or orange colour for presence of flavonoid or flavin [14].

Determination for antimicrobial activity

This test was carried out to determine the effects of the extracts (dialyzed and undialyzed portions) on dental caries pathogens.

Collection of organisms

The test organisms were collected from a culture centre in University Teaching Hospital (UNTH), Ituku-Ozalla, Enugu West Local Government Area, Enugu State, Nigeria. The organisms collected were; *Escherichia coli*, *Staphylococcus aureus*, *Candida albicans*, *Lactobacillus*, and *Streptococcus mutans*.

Identification of organisms

The organisms collected were cultured to confirm their authenticity and viability. Specific selective media were used for identification of the different dental caries pathogens.

For *Escherichia coli*

Media used for identification of *Escherichia coli* was MacConkey agar for lactose and non-lactose fermenting bacteria.

Table 1: Media compositions

Composition	gm/litre
Peptic digest	20.00
Animal tissue	12.00
Agar	12.00
Lactose	10.00
Bile salts	5.00
pH	7.4±0.2 at 25°C
Distilled water	1 litre

Preparation

Dissolve 47gm in 1000ml of distilled water. Gently heat to dissolve the medium completely. Sterilize by autoclaving at 15

Psi (121°C) for 15 minutes to sterilize the media

For *S. aureus* The media used for identification of *S.aureus* was nutrient agar.

Table 2:**Media composition**

Agar	15.00
Peptone	5.00
Sodium chloride	5.00
Beef extract	1.50
Yeast extract	1.50
pH	7.4±0.2 at 25°C
Distilled water	1 litre

Preparation

Dissolve 28.0gm in 1000ml of distilled water. Gently heat to homogenize the

mixture. Sterilize by autoclaving at 15psi (121°C) for 15 minutes. Dispense the medium as desired.

Table 3: For lactobacillus

The media used for identification of lactobacillus was MRS agar.

Composition	gm/litre
Dextrose	20.00
Proteose peptone	10.00
Beef extract	10.00
Yeast extract	5.00
Sodium acetate	5.00
Ammonium citrate	2.00
Disodium phosphate	2.00
Tween 80	1.00
Magnesium sulphate	0.10
pH	6.5±0.2 at 25°C
Distilled water	1 litre

Preparation

Dissolve 67.15gm in 1000ml of distilled water and gently heat to homogenize the mixture. Sterilize by autoclaving 15psi

(121°C) for 15 minutes, allow to cool to about 35°C before pouring into plates with desired quantity.

For *Candida albicans*Table 4: The media used for identification of *C.albicans* was sabour and Dextrose Agar for fungi.

Composition	mg/litre
Peptone	5.0
Tryptone	5.0
Dextrose	40.0
Agar	15.0
Final pH (at 25°C)	5.6 ± 0.2
Distilled water	1 litre

Preparation

Suspend 65.0g of the powder in 100ml of distilled water mix thoroughly. Boil with frequent agitation to dissolve the powder completely. Avoid over heating the agar as it could cause a softer medium.

Sterilize by autoclaving at 121°C (15 psi) for 15 minutes.

Preparation of Mueller Hinton Agar

This media was used for the antimicrobial screening for all the isolates because it is a general purpose medium

Table 5:

Preparation	gm/litre
Beef infusion from casein	30.00
Acid hydrolysate	17.50
Agar	17.00
Starch	1.50
Distilled water	1 litre

Preparation

Dissolve 38gm in 1000ml of distilled water. Gently heat to dissolve the medium completely. Sterilize by autoclaving at 15 psi (121°C) for 15 mins.

Determination of minimum inhibition concentration (MIC)

A general purpose media, Mueller Hinton agar was used for this test. 20ml of the media was carefully poured into 10 plates, 5 plates for dialyzed portion and 5 plates for the undialyzed portion with the different extracts and the 5 microorganisms for each plates. After drying the plates, by passing it through flame, Agar well was made using a cork borer of diameter 8cm. The extracts were diluted to different concentrations 100, 50, 25, 12.5, 6.25 percent, using serial

dilution method. 1ml of each concentrations were carefully introduced in the well labeled agar wells made on every plates, while observing all the aseptic techniques. After that, the plates were incubated for 24 hours for bacteria and 48 hours for the fungi specie (*Candida albicans*). After which readings were taken using venier caliper and meter rule for measuring the zones of inhibition.

Preparation of control

A positive control was set up. Five separate plates were also provided and agar wells were made on each plate, the organisms were inoculated. The solvents used for extraction was used as control 0.5ml of each was added to the agar well and incubated for 24 hours.

RESULTS

The effects of different extracts on the organisms were determined and the inhibition zone diameter (IZD) of the isolates measured in millimeter as illustrated in tables 2-13.

The effect of undialyzed concentration of methanoic extract on different isolates was evaluated in table 6. The undialyzed methanol extract showed a moderate level of sensitivity, having the highest

sensitivity against *S. aureus* with inhibition zone diameter (IZD) 10mm at the concentration of 6.25mg/ml. Other isolates, *S. mutans*, *E. coli* and *C. albicans*

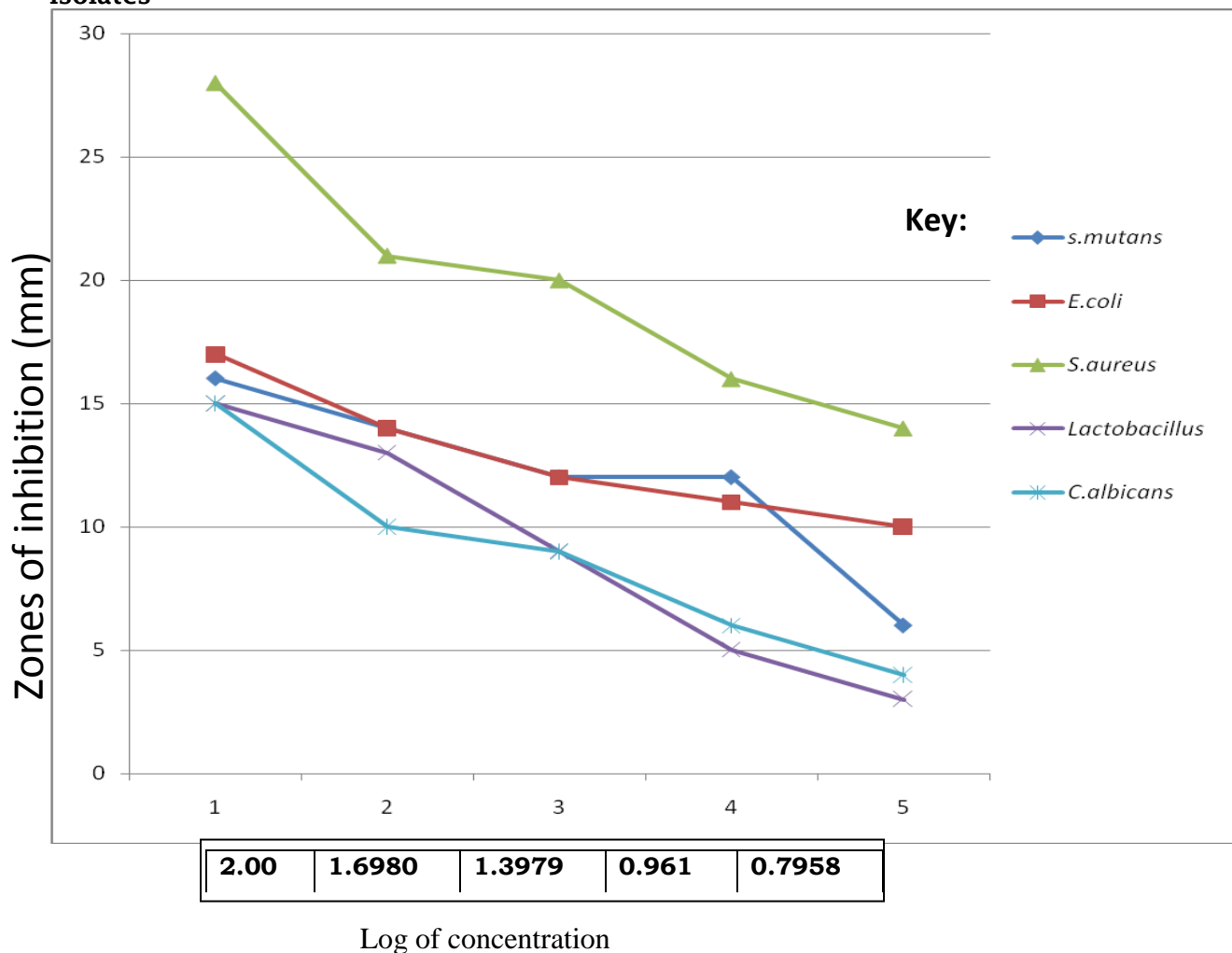
had IZDs of 5.9 and 3 respectively with MIC of 6.25mg/ml. It had no effect on *Lactobacillus*.

Table 6: The effect of undialyzed concentration of methanolic extract on different isolates.

Conc. (mg/ml)	100	50	25	12.5	6.25	+ control (100mg)
Log conc.	2.00	1.6980	1.3979	0.961	0.7958	
	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)	
<i>S.mutans</i>	20	14	14	12	5	10
<i>E.coli</i>	26	16	14	12	9	5
<i>S.aureus</i>	29	28	24	20	10	3
<i>Lactobacillus</i>	-	-	-	-	-	10
<i>C.albicans</i>	17	14	10	4	3	6

Key: IZD: Inhibition zone diameter (millimeter).

Figure1: The effect of undialyzed concentration of methanolic extract on different isolates



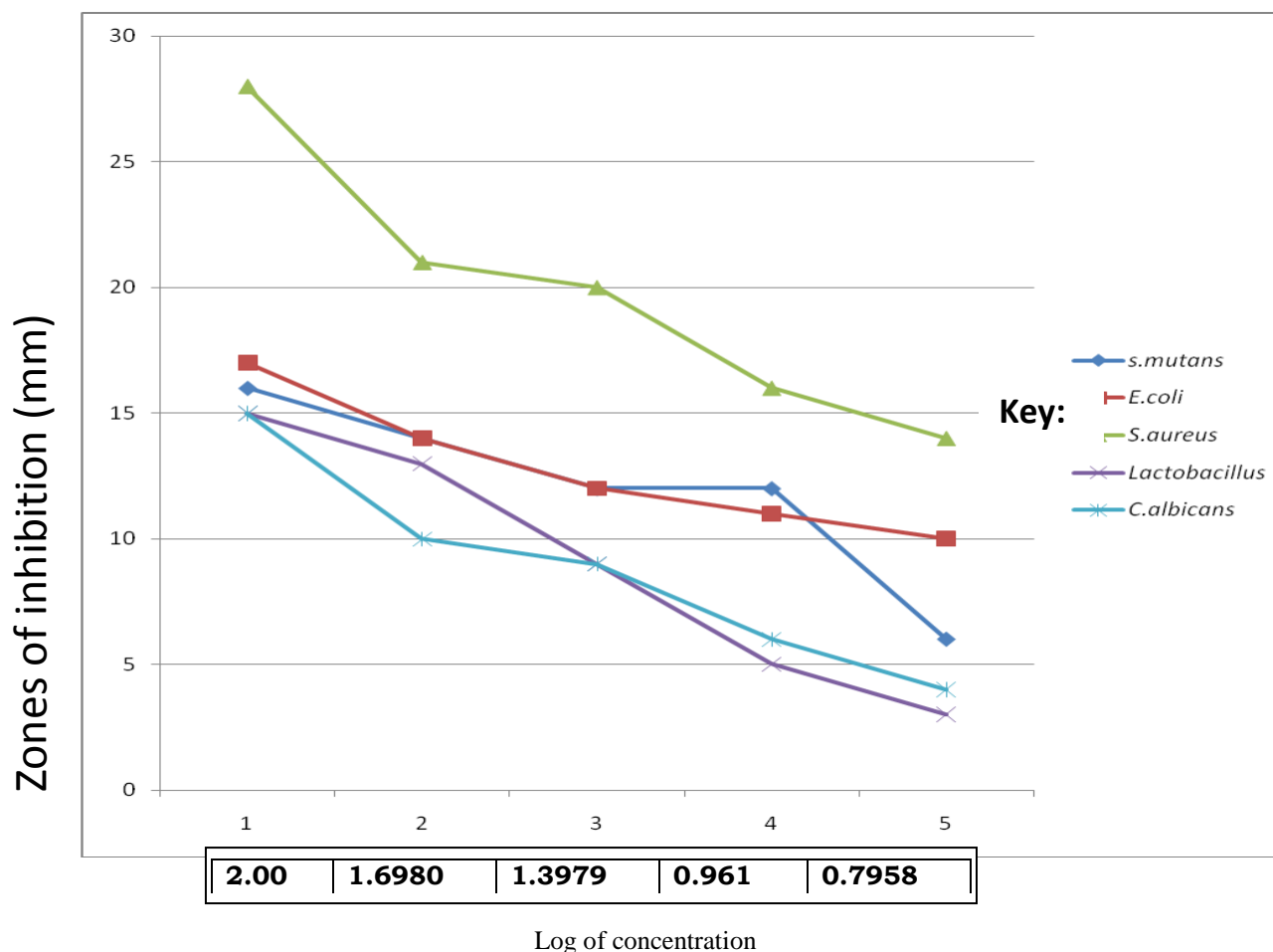
The effect of dialyzed concentrations methanol extract on different isolates was evaluated in table 7, dialyzed methanol extracts showed a high level of sensitivity against the isolates with the highest effect on *S.aureus* with inhibition zone of 12mm and MIC of 6.25 mg/ml.

Table 7: The effect of dialyzed concentrations of methanoic extracts on different isolates

Conc. (mg/ml)	100	50	25	12.5	6.25	+ control (100mg)
Log conc.	2.00	1.6980	1.3979	0.961	0.7958	
	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)	
<i>S.mutans</i>	26	20	14	12	9	10
<i>E.coli</i>	15	14	10	4	3	5
<i>S.aureus</i>	25	23	17	14	5	3
<i>Lactobacillus</i>	20	20	15	5	5	10
<i>C.albicans</i>	24	16	13	11	10	6

Key: IZD: Inhibition zone diameter (millimeter).

Figure 2: The effect of dialyzed concentrations of methanol extracts on different isolates



The effect of undialyzed concentrations of undialyzed ethanolic extract on the different isolates was evaluated, in table 8, the undialyzed ethanolic extract also showed high level of sensitivity on *S.aureus* with IZD of 9mm with MIC of

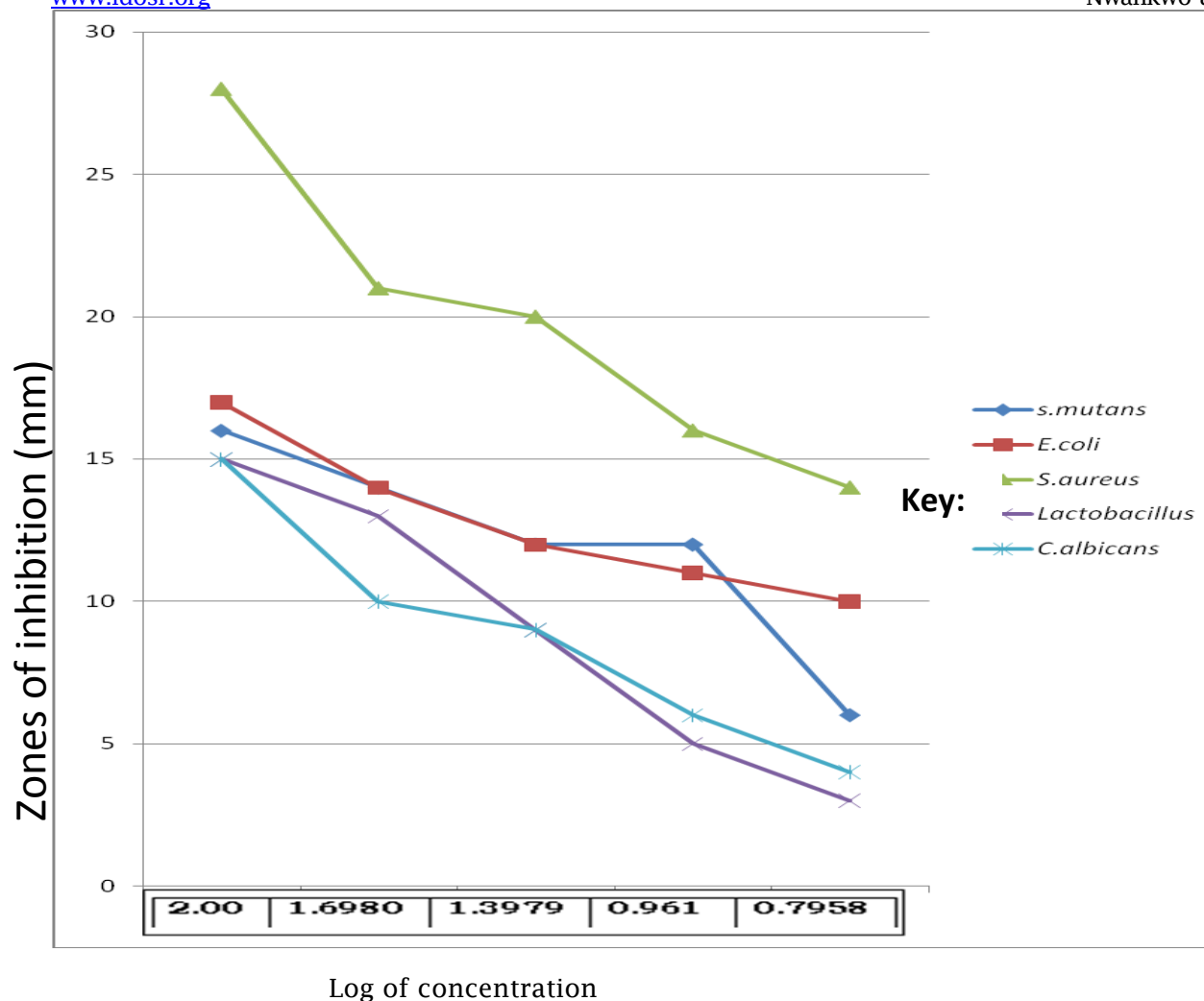
6.25mg/ml and moderated effect on *S. mutans*, *E. coli*, and *Lactobacillus* with IZDs of 5,4 and 1 respectively and MIC of 6.25mg/ml for all the isolates, then little effect on *C. albicans* with MIC 50mg/ml with IZD of 5mm.

Table 8: The effect of undialyzed concentrations of undialyzed ethanolic extract on the different isolates

Conc. (mg/ml)	100	50	25	12.5	6.25	+ control (100mg)
Log conc.	2.00	1.6980	1.3979	0.961	0.7958	
	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)	
<i>S.mutans</i>	26	21	20	18	9	8
<i>E.coli</i>	12	12	10	6	5	5
<i>S.aureus</i>	26	23	21	10	4	5
<i>Lactobacillus</i>	16	7	6	4	1	10
<i>C.albicans</i>	8	5	-	-	-	9

Key: IZD: Inhibition zone diameter (millimeter).

Fig 3: The effect of undialyzed concentrations of undialyzed ethanolic extract on the different isolates



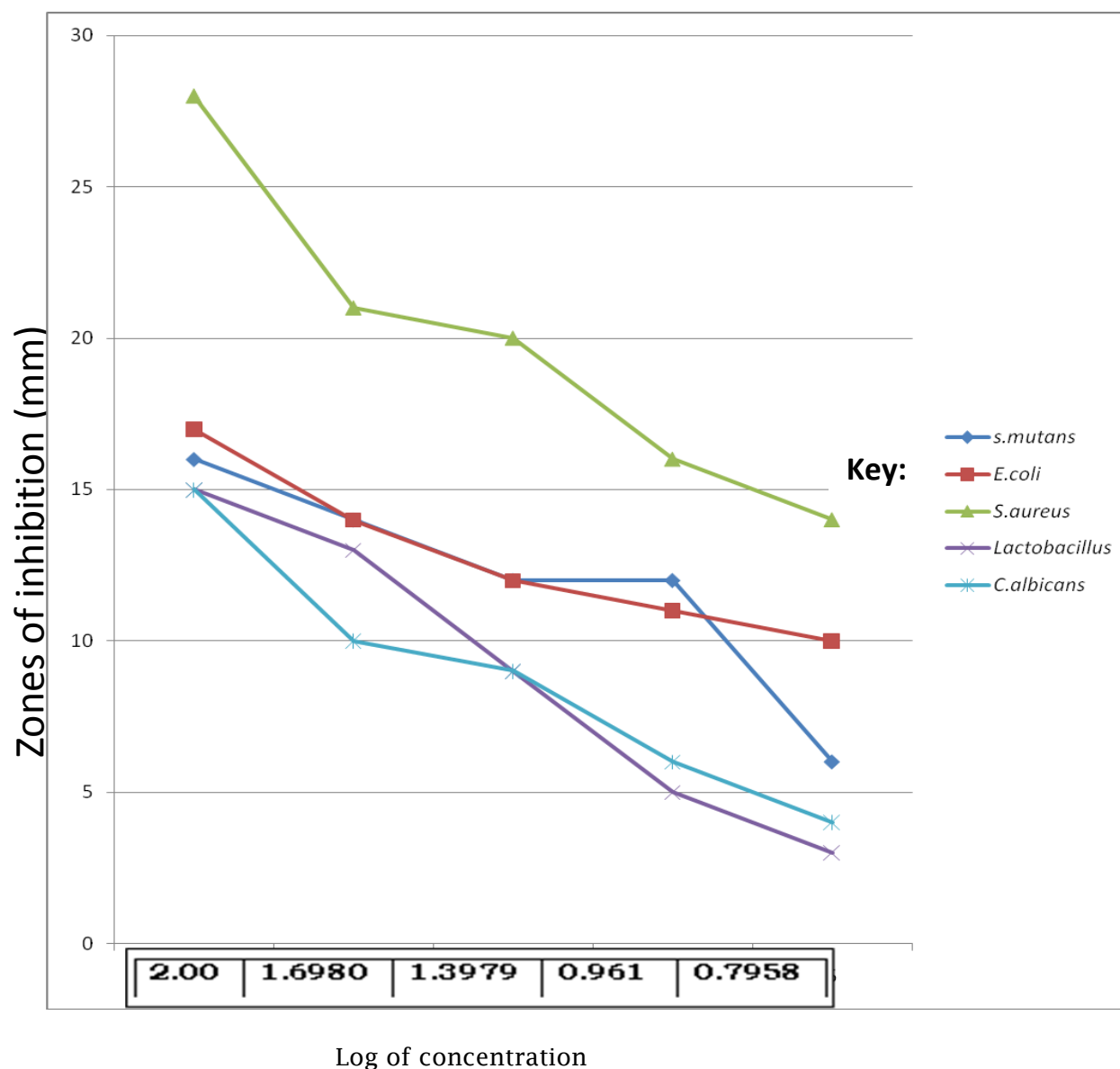
The effect of the dialyzed concentrations of ethanolic extract on different isolates was evaluated in table 9. The result showed that there was high level of sensitivity against *Lactobacillus* and *S. mutans* with the inhibition zones IZD of 12mm and 11mm respectively and MIC of 6.25mg/ml.

Table 9: The effects of dialyzed concentrations of ethanoic extract on different isolates

Conc. (mg/ml)	100	50	25	12.5	6.25	+ control (100mg)
Log conc.	2.00	1.6980	1.3979	0.961	0.7958	
	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)	
<i>S. mutans</i>	23	21	16	12	11	8
<i>E. coli</i>	14	10	6	5	2	5
<i>S. aureus</i>	26	17	13	9	4	5
<i>Lactobacillus</i>	21	18	14	13	12	10
<i>C. albicans</i>	16	15	12	12	4	9

Key: IZD: Inhibition zone diameter (millimeter).

Figure 4: The effects of dialyzed concentrations of ethanoic extract on different isolates



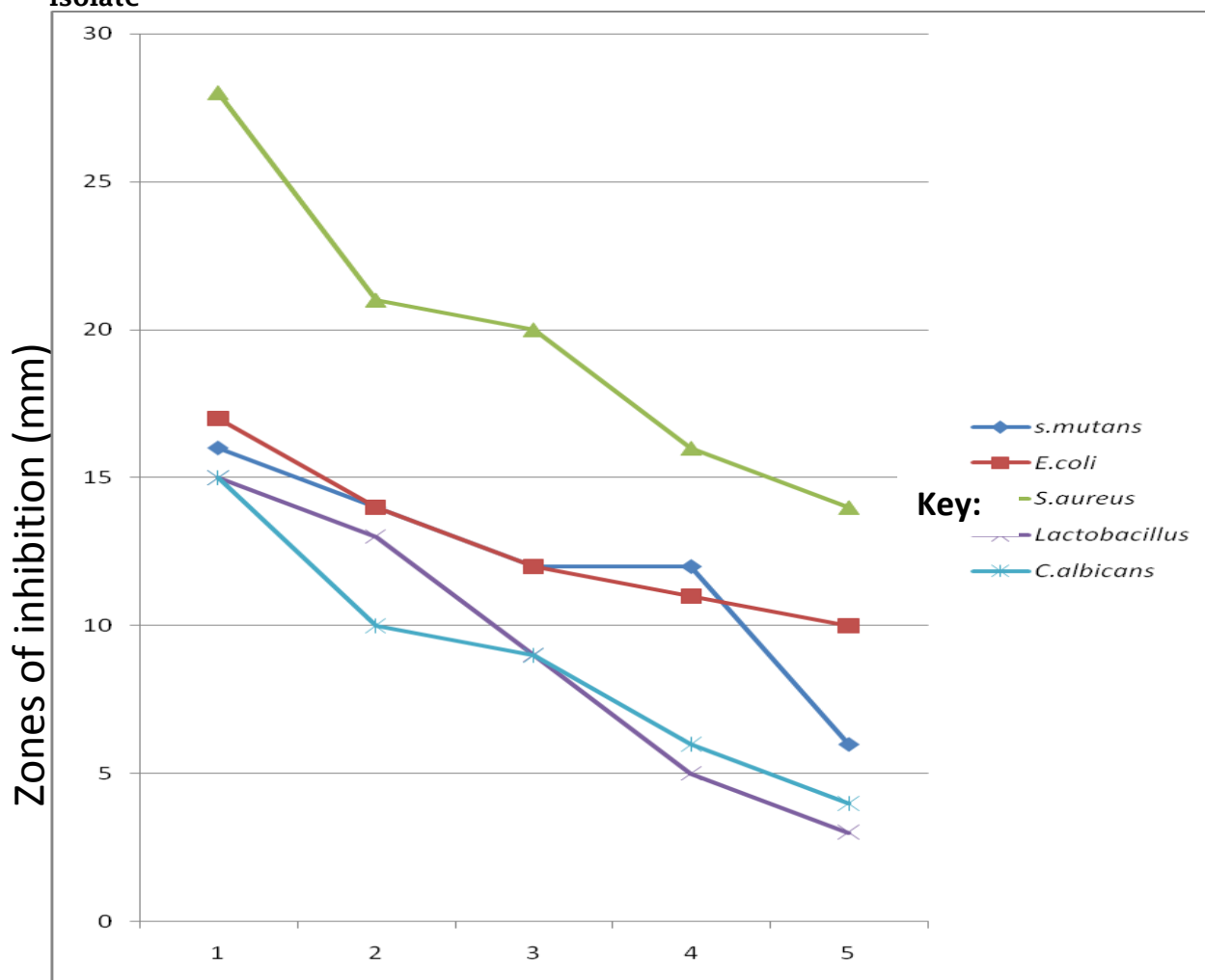
The effect of the undialyzed concentrations of N-Hexane extract on different isolates was evaluated in table 10. The undialyzed N.Hexane fraction showed little effect in all of the isolates, the highest effect was on *Lactobacillus*

with IZD of 5mm and MIC of 6.25mg/ml, the least was *S. mutans* with IZD of 1mm and MIC of 6.25, there was no inhibition in *E. coli* and *S. aureus*.

Table 10: The effect of the undialyzed concentrations of N-Hexane extract on different isolate

Conc. (mg/ml)	100	50	25	12.5	6.25	+ control (100mg)
Log conc.	2.00	1.6980	1.3979	0.961	0.7958	
	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)
<i>S.mutans</i>	10	6	4	4	1	2
<i>E.coli</i>	4	-	-	-	-	4
<i>S.aureus</i>	-	-	-	-	-	4
<i>Lactobacillus</i>	9	7	6	6	5	3
<i>C.albicans</i>	-	-	-	-	-	6

Key: IZD: Inhibition zone diameter (millimeter).

Figure 5: The effect of the undialyzed concentrations of N-Hexane extract on different isolate


2.00	1.6980	1.3979	0.961	0.7958
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Log of concentration

The effect of the dialyzed concentrations of N-Hexane extract on different isolates was evaluated in table 11. The dialyzed N-Hexane fraction showed high sensitivity on *S.aureus* with inhibition zone diameter

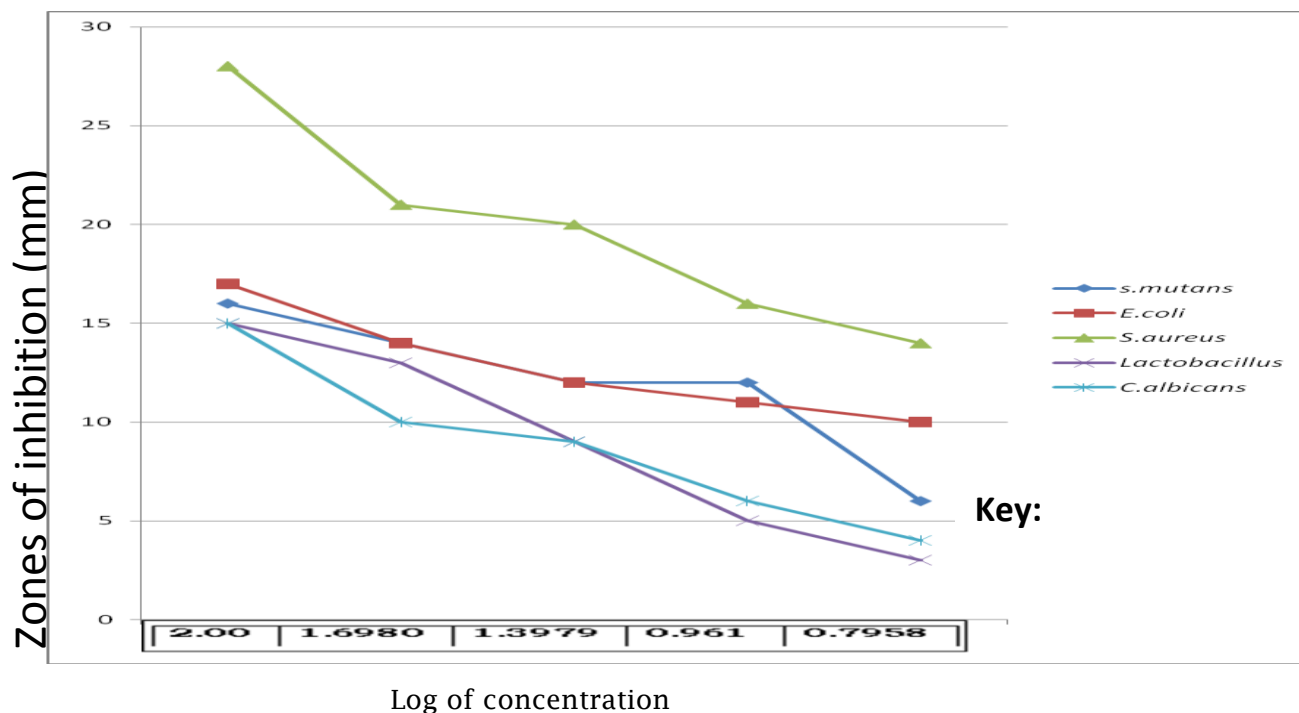
of 5mm at MIC of 6.25mg/ml., and little effect on *C.albicans*, IZD of 5mm at MIC of 100mg/ml while that of *Escherichia coli* was IZD 12mm and MIC of 12.5mg/ml.

Table 11: The effect of dialyzed concentrations of N-Hexane extract on different isolate

Conc. (mg/ml)	100	50	25	12.5	6.25	+ control (100mg)
Log conc.	2.00	1.6980	1.3979	0.961	0.7958	
	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)	
<i>S.mutans</i>	20	14	4	2	2	2
<i>E.coli</i>	27	16	-	12	-	4
<i>S.aureus</i>	28	18	16	13	5	4
<i>Lactobacillus</i>	8	7	4	4	-	3
<i>C.albicans</i>	5	-	-	-	-	6

Key: IZD: Inhibition zone diameter (millimeter).

Figure 6: The effect of dialyzed concentrations of N-Hexane extract on different isolate



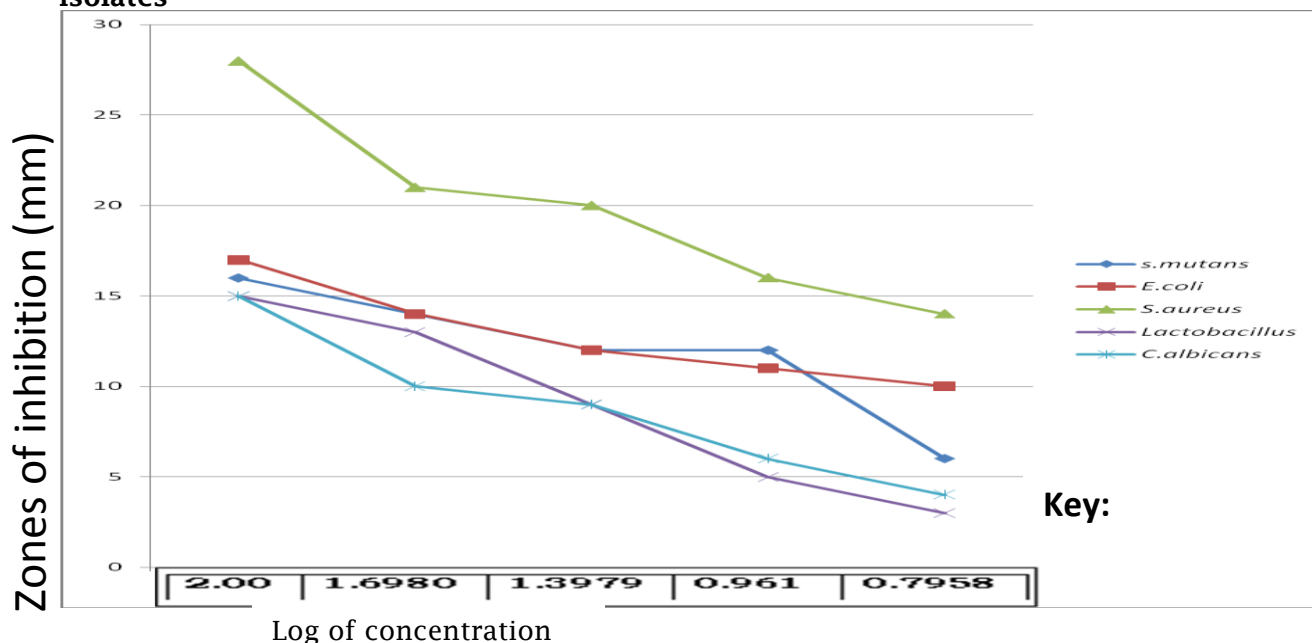
The effect of undialyzed concentrations of crude water extract on different isolates was evaluated in table 12. The undialysed water extract showed high sensitivity on *E.coli* with IZD of 9mm at MIC of 6.25mg/ml and there was no inhibition against *S.aureus*.

Table 12: The effect of undialyzed concentrations of crude water extract on different isolates

Conc. (mg/ml)	100	50	25	12.5	6.25	+ control (100mg)
Log conc.	2.00	1.6980	1.3979	0.961	0.7958	
	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)	
<i>S.mutans</i>	18	14	10	8	4	-
<i>E.coli</i>	16	14	13	10	9	-
<i>S.aureus</i>	-	-	-	-	-	-
<i>Lactobacillus</i>	10	6	4	2	2	-
<i>C.albicans</i>	15	10	9	6	4	-

Key: IZD: Inhibition zone diameter (millimeter).

Figure 7: The effect of undialyzed concentrations of crude water extract on different isolates



The effect of dialysed concentration of water extract on different isolates was evaluated in table 13. The dialyzed fraction of water had high sensitivity on all the isolates, having highest IZD

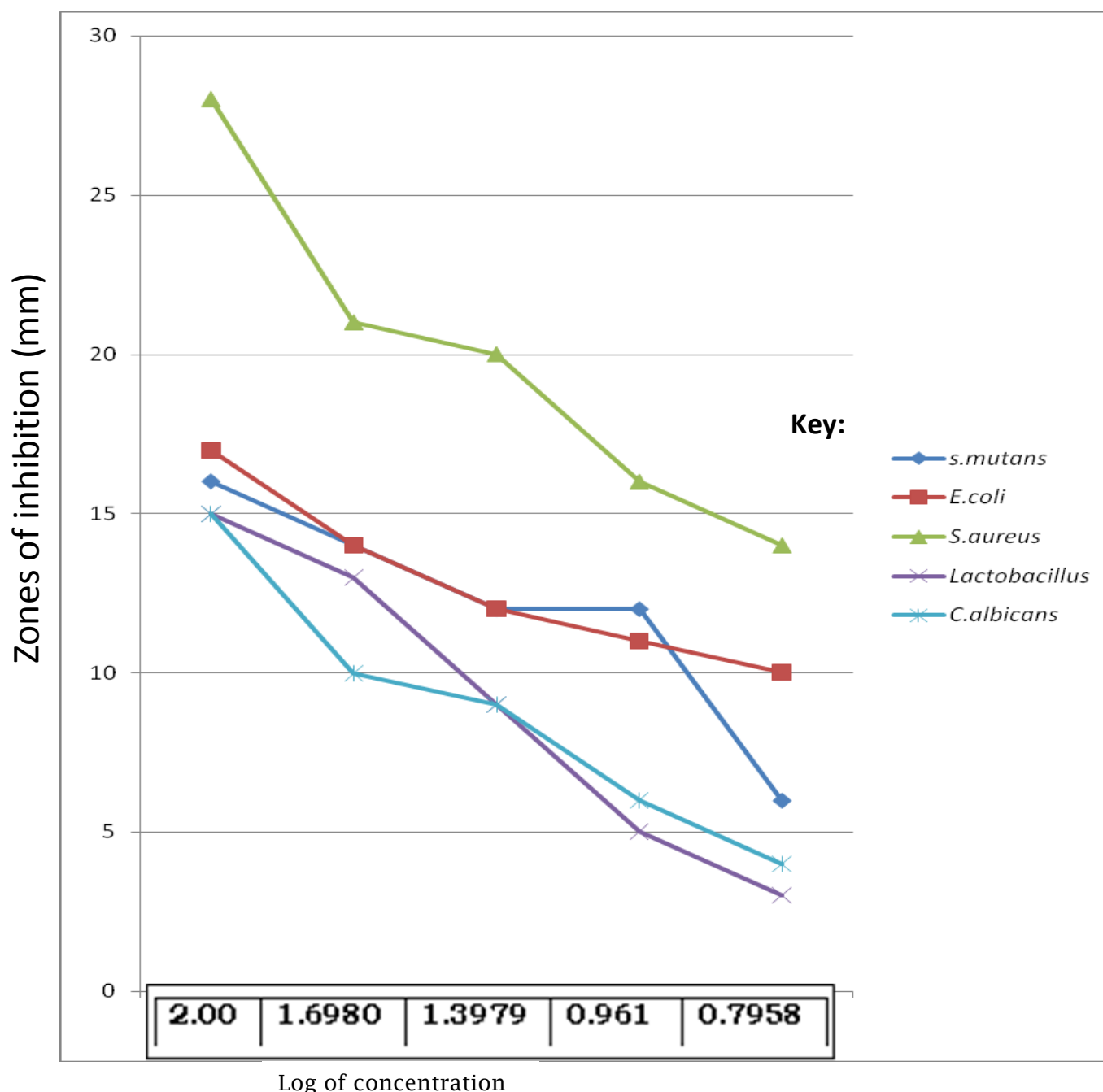
starting from *S.aureus*, *E.coli*, *S.mutans* and *C.albicans*, their IZDs are 14, 10, 6, 4 and 3 mm, respectively with MIC of 6.25 mg/ml.

Table 13: The effect of dialyzed concentrations of water extract on different isolates

Conc. (mg/ml)	100	50	25	12.5	6.25	+ control (100mg)
Log conc.	2.00	1.6980	1.3979	0.961	0.7958	
	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)	IZD (mm)	
<i>S.mutans</i>	16	14	12	12	6	-
<i>E.coli</i>	17	14	12	11	10	-
<i>S.aureus</i>	28	21	20	16	14	-
<i>Lactobacillus</i>	15	13	9	5	3	-
<i>C.albicans</i>	15	10	9	6	4	-

Key: IZD: Inhibition zone diameter (millimeter).

Figure 8: The effect of dialyzed concentrations of water extract on different isolates



Results of the phytochemical analysis showed abundant quantities of flavonoids, steroids, glycosides, under the methanolic extract in Table 14. It also contained moderate quantities of Terpenoids, tannins, phenols and No presence of alkaloids in the three reagents used (Dragendroff, Mayer's and Wagner's reagents), Table 14. The result

also showed that there was moderate quantity of Terpenoids, small quantities of steroids and tannins and absence of result showed that there was abundant quantity of Saponins, moderate quantities of flavonoids, terpenoids, glycosides, tannins, small quantity of steroid, then absence of alkaloids.

Table 14: Phytochemical constituents of *Mangifera indica* leaf extracts.

Extract	Methanol	Ethanol	N-Hexane	Water
Constituents				
Flavonoids	+++	++	-	++
Terpenoids	++	++	++	++
Steroids	+++	+++	+	+
Glycosides	+++	+++	-	++
Phenols	++	++	-	++
Saponins	++	+++	+	+++
Tanins	++	+++	+	++

DISCUSSION

It is thought that the anti-bacterial activities of the extracts of *Mangifera indica* against some dental caries pathogens specified was due to their phytochemical constituents such as Flavonoids, Terpenoids, Steroids, Glycosides, tannins and phenols [1]. Tannins have been reported to have anti-microbial properties [2]. Also [3] had reported their inhibitory effects on the growth of intestinal bacteria such as *Bacteroid fragilis*, *Clostridium perfringens* and some dental caries pathogen such as *Staphylococcus aureus* and *Escherischia coli* among others.

From table 12, the abundance of saponins, might also be partly related to the antibacterial activity, as it was reported that Saponins increase antibiotic activity, especially against Gram-positive bacteria [4]. This probably explains the inhibition of *S. aureus* with MIC of 6.25mg/ml and IZD of 14mm in table 13. Reports by [8] showed that flavonoids, steroids and tannins which were mostly extracted by methanol and ethanol in abundance have wide range of activities. Flavonoids are hydrooxylated polyphenolic compounds known to be produced by plants in response to microbial infections; they have an array of antimicrobial activates in the body [9]. Steroids have some health benefits like absorption of Na⁺ and water [13]. Tannins are known to have great effect on fungi [12], this may partly be the reason for the high sensitivity against *C. albicans* with MIC of 6.25mg/ml and inhibition zone diameter of 10mm.

From the result shown in table 10, it was observed that among all of the solvents used in extracting; methanol and ethanol extracted most of the bioactive ingredients, followed by the water and lastly N-Hexane in *Mangifera indica* leaves. This may be as a result of the high extracting capacity in methanol and ethanol [5].

Several studies had revealed the antimicrobial efficacy of *Mangifera indica* leaves water extract, hexane, ethanol and methanolic extracts against some dental caries pathogen, as well as Gram-positive and Gram negative bacteria responsible for majority of multidrug resistant infections in Nigeria [7], and Salmonella [8], as well as Streptococcus and Bacillus species [13].

Extrapolations from the results of susceptibility of the concentrations of extracts and fractions on the tested isolates showed their minimum inhibitory concentration (MIC) values in Tables 2-13. From the result of the MIC, it was observed that the greater the inhibition zone diameter (IZD) produced, the greater the MIC and less potent of the agent. However, the extracts and their concentrations had varying MICs on individuals organisms.

Generally, the dialyzed fractions had higher sensitivity on the dental caries pathogens specified, than the undialyzed fractions. In the methanolic extracts, there was high sensitivity against *S. aureus*, but the highest inhibition was noted in the dialyzed fraction, this was with an inhibition of concentration of 6.25mg/ml and diameter of 12mm; while

in the undialyzed methanol fraction, the inhibition of *S. aureus* was a MIC of 6.25mg/ml and IZD of 10mm. Ethanolic extract also had a moderate effect on the different isolates with MIC of 6.25 for *S.mutans*, *E. coli* and *S. aureus* and MICs of 6.25 and 1(mm) respectively, then for *C. albicans* MIC was 50mg/ml and IZD OF 4mm because Ethanol and methanol has high extracting power.

N-Hexane fraction had and insignificant effect on the isolates, and it was assumed

CONCLUSION

The phytochemical analysis has shown that the leaves of *Mangifera indica* contains a lot of bioactive ingredients and these ingredients are high in antimicrobial effect as well as in nutritional content. The N-Hexane used in extraction yielded insignificant quantity

to be as a result of its inability to extract most of the bioactive ingredients present in the leaves of *Mangifera indica*. The highest sensitivity was on *S. aureus* and *S. mutans* in the dialyzed fraction with IZD of 5 and 2mm respectively. The water extract had a moderate effect on both the dialyzed and undilayzed fractions, but it was observed that there was no action on *S. aureus* in the dialyzed fraction, and in the dialyzed fraction an IZD of 14mm with MIC of 6.25 was observed.

of the ingredients, therefore should not be used in extraction of mango leaves because of its low extracting power. In the antimicrobial screening it can be noted that the extract especially the dialyzed fractions were more effective.

RECOMMENDATION

It is recommended that extracts of medicinal plants should be dialyzed in order to obtain more effective result on pathogenic microorganisms and that

instead of using N-Hexane during extraction of *Mangifera indica* leaves for extraction, Ethanol, methanol and water would be more preferable.

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