



Ethnopharmacological Survey and Physiological Evaluation of Nutritional and Phytochemical Contents of Indigenous Plants Used for Treatment of Toothache and Mouth Odour in Ijebu Ode Local Government Area, Ogun State, Nigeria

Page | 3559

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ABSTRACT

The use of herbs for improvement of oral hygiene is increasing in many communities in Nigeria despite the usage of other dental care products. On the basis of this the present study was conducted to assess indigenous plants used to manage dental condition and mouth odour and to evaluate nutritional, phytochemicals and anti-nutrient content of the plants. A survey was conducted to document plants used for treatment of toothache and mouth odour in Ijebu-Ode Local Government Area. Using random sampling technique, a total of one hundred structured questionnaire was administered to Traditional Health Practitioners in three major markets of the study area. Nutritional, phytochemical and anti-nutrient contents of most used plants were evaluated. Data were analyzed using Statistical Analysis System. Means were separated using Duncan's Multiple Range Test at 5% level of significance ($p < 0.05$). A total of twenty-three (23) plants belonging to sixteen families were enumerated out of which *Capsicum frutescens*, *Piper guineense* fruits and *Zanthoxylum zanthoxloides* roots were the most exploited for management of the conditions. Crude fibre (8.86 %), fat (1.12%) and ash contents (4.73%) were significantly ($p < 0.05$) higher in *Z. zanthoxloides* roots while moisture (25.75 %) was significantly higher ($p < 0.05$) in *C. frutescens* fruits. Calcium (192.10 mg/100g), phosphorus (108.50 mg/100g), sodium (51.33 mg/100g), iron (4.85 mg/100g), Zinc (3.94 mg/100g), manganese (1.15 mg/100g) and copper (2.12 mg/100g) were significantly ($p < 0.05$) higher in *Z. zanthoxloides* roots compared *C. frutescens* and *P. guineense* fruits. Vitamin A (600.00 μ /100g), vitamin B (0.07.00 mg/100g), vitamin C (94.54 mg/100g) and vitamin E (720.00) were significantly ($p < 0.05$) higher in *C. frutescens* than *Z. zanthoxloides* and *P. guineense*. Results also showed that tannin (6.40 %), oxalate (30.00%), phytate (0.40%) and trypsin inhibitor (20.00 %) were significantly higher in *frutescens* than *Z. zanthoxloides* roots. Similar significant ($p < 0.05$) increase were observed in the quantity of flavonoid (3.25%), saponnis (1.30%), phenol (0.60%) and anthocyaninns (0.23%) in *Z. zanthoxloides* compared with *C. frutescens* and *P. guineense*.

Keywords: Ethnopharmacological, Mouth odour, toothache, phytochemical, anti-nutrient.

Introduction

Several efficacies of plants on personal hygiene such as oral hygiene had been reported by several researchers (Kalemba and Kunicka, 2003; Odugbemi, 2006; Maji *et al.*, 2011; Nasreen and

Radha, 2011; Ashidi *et al.*, 2013; Ojewumi and Kadiri, 2014). According to Ranjan *et al.* (2012), oral infections among others are common health challenges affecting people both in local

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communities and cities. Although the ailment affects adults but its prevalence among schooling children is higher (90%) (Peterson *et al.*, 2005). This record serves as high level of concern in rural areas. In an attempt to control high prevalence of this health challenge, various concerted methods had been adopted to keep teeth clean and make general mouth conditions hygienic (Kalemba and Kunicka 2003). Based on the attempts, the use of plants to manage teeth health care needs of people in many communities is very rampant mostly among the rural dwellers. This is due to, population increase, inadequate supply of drugs to some community centres, high cost of treatments, high level of poverty, side effects of several allopathic drugs, poor personal hygiene practice and availability of herbal products at the disposal of the people (Petersen, 2003; Pradeep, 2014; Michael and Sudeshni 2015). Motta *et al.* (2011); Bollen and Beikler. (2012); Madhushankari *et al.* (2015); Sara *et al.* (2016) and Aliyu (2018) revealed that mouth odour (halitosis) occurs due to decomposition of organic matter which develops from flakes of epithelial cells retained on the posterior portion of the dorsum of tongue. According to Nao *et al.* (2016) and Fernanda *et al.* (2019), the decomposition is facilitated by mucin precipitation, a reduction in salivary flow and/or water imbalance, microbial attack and alkalization of the oral environments, all of which enhance growth of proteolytic bacteria and consequently result in the production of volatile sulfur compounds. Also, Aylikc and Colak, (2013) and Kotti and Subramanyam (2015) opined that intensity of bad breath is associated with level of volatile sulfur compounds in the oral cavity of an individual.

According to reports of Aworinde *et al.* (2016), oral/dental conditions ranging from toothache /decay to black tongue are treatable using herbs. For example plants such as *Agremone maxicana*, *Azadirachta indica*, *Ocimum basilicum*,

Hedychium spicatum and *Zanthoxylum aromaticum* have been reported to be useful in dental health care (Sing and Dhakre 1989).

Also, Orange tree (*Citrus aurantium*) and lime tree (*Citrus aurantiifolia*) have been widely adopted as chewing sticks for maintenance of oral hygiene (Kalemba and Kunicka 2003), yet nutritional and therapeutic relevance of most of the plants need comprehensively elucidation. Also, in recent years many, people have become increasingly conscious of halitosis possibly as a result of poor hygienic status of the individuals. The most familiar smell is our personal odour, the odour generated by our own bodies. The odour is associated with negative impression thereby affecting social life and habits of most people. Humans are typically keen to eliminate or reduce it with affordable but effective approach. However, this study was conducted to assess indigenous plants used to manage dental condition and mouth odour and to elucidate nutritional, phytochemicals and anti-nutrient content of the plants.

Materials and Methods

The study area

The present study was carried out in major markets (Oke Aje, itale and New markets) located in Ijebu- Ode Local Government Area (LGA). It is one of the LGAs in Ogun State. It is located 110 km by road north-east of Lagos State. It is within 100 km of the Atlantic Ocean in the eastern part of Ogun State, possesses a warm tropical climate and land area of 192 km² (Ogundiran, 2013). The area has an estimated population of 222,653 (Adedeji *et al.*, 2019). It is largest city inhabited by the Ijebus, a sub-group of the Yoruba ethnic group who speak Ijebu dialect. It is the trade center of farming where crops and other economic plants are grown.

Population of the study: The population of the study consisted of herb practitioners and nursing mothers in major markets of Ijebu -Ode LGA.

Plant collection and identification

Samples of *C. frutescens*, *P. guineense* and *Z. zanthoxloides* were collected from Traditional Health Practitioners and identified at Lagos State University Herbarium. The voucher number of the plants are; *C. frutescens*(LUH 8556) *P. guineense* (LUH 8557) and *Z. zanthoxloides*(LUH 8558)

Validity and reliability of instrument: These were carried out using Pilot test according to Ojewumi *et al.* (2016b). Cronbach alpha values; 0.82 (82%) was obtained.

Administration of questionnaire: A total of 100 questionnaire was administered on the respondents to source for ethnobotanical information using random sampling technique.

Proximate analysis of *C. frutescens*, *P. guineense* fruits and *Z. zanthoxloides* roots

Crude fibre: One gram each of defatted samples of the three plants was boiled in 20ml of 1.25 % H₂SO₄ (sulphuric acid) for 30 minutes. After this, the content was filtered, washed with hot distilled water and boiled in 200 ml of 1.25% sodium hydroxide for about 30 minutes. Spotless beaker was dried at 100±5°C overnight, cooled in a desiccator and weighed to a constant weight. Then, the spoutless beakers with its content was put in a muffle furnace at 932°F-1112°F for 2-3-hour, cooled in a desiccator and weighed. Crude fibre was determined using formula (AOAC, 2000).

$$\text{Crude fibre (\%)} = \frac{\text{weight of spoutless beaker containing crude fibre} - \text{weight of spoutless beaker and crude fibre}}{\text{Weight of sample}} \times 100$$

Crude protein: Total nitrogen (N) was determined using Micro-Kjeldahl method in (2009)

Protein (%) was determined using the mathematical relationship below.

$$\text{Protein (\%)} = \frac{V \times 1.4 \times 6.25 \times 0.1N \text{ Hcl} \times \text{Vol (used)}}{W \times A \times 1000} \times 100$$

where;

V = Titter value. 1.4 -Weight of nitrogen expressed in gram in the formula.

6.25 = Protein factor.

W = Weight of sample. A -Aliquot digested sample used for distillation

Crude fat: One gram of crushed dried sample was taken in the paper thimble kept in a pre-weighed flask of fat extractor. Eighty (80ml) of petroleum ether was added and refluxed for 8hours. The flask was cooled and weighed and crude fat was determined using formula.

$$\text{Crude fat (\%)} = \frac{\text{Weight of flask with fat} - \text{weight of empty flask}}{\text{Weight of original sample}} \times 100$$

Moisture: Moisture was determined using hot air oven method as shown below

$$\text{Moisture} = \frac{\text{Weight of sample before drying} - \text{weight of sample after drying}}{\text{Weight of sample before drying}} \times 100$$

Ash content: Ten grams (10.g) of each samples was added to a reweighed crucible, weighed, placed in a muffle furnace at 932°F for 4hours, cooled in desiccator and reweighed. Ash content was determined using mathematical relationship;

$$\text{Ash (\%)} = \frac{\text{Weight of ash}}{\text{Weight of sample}} \times 100$$

Carbohydrate: Available carbohydrate was calculated using formula below;

Carbohydrate (%) = 100 - (moisture + crude fat + ash + crude protein) % (AOAC, 2000).

Mineral Analysis of *C. frutescens*, *P. guineense* fruits and *Z. xanthoxloides* roots

Minerals in each sample of the three plants were determined after sample wet digestion of 3.0 g of each plant with a mixture of $\text{HNO}_3/\text{HClO}_4/\text{H}_2\text{SO}_4$ in the ratio 9:2:1 v/v, respectively. Mineral such Mg, Ca, P, Fe, Cu, Zn and Mn, were determined using atomic absorption spectrophotometer. The K and Na of the sample were determined using atomic emission spectrometer and phosphorus by colorimetric method of AOAC, (1990).

Determination of Vitamins in *C. frutescens*, *P. guineense* fruits and *Z. xanthoxloides* roots

Vitamin A: Vitamins A was determined according to method of AOAC, (2000). Two (2 g) of sample of each plant was weighed into a flat bottom reflux flask, 10ml of distilled water was added and shaken to form a paste after which 25ml of alcoholic KOH solution was added and a reflux condenser attached. The mixture was heated using boiling water bath for one hour, shaken, cooled rapidly and about 30 ml of water was added after which hydrolysate obtained was transferred into a separatory funnel. The solution was extracted thrice with 250ml quantities of chloroform. In addition, 2g anhydrous Sodium sulphate was added to the extract to remove traces of water. The mixture was then filtered into 100ml volumetric flask and made up to mark with chloroform. Standard solution of B-carotene Vitamin A of ranged from 0 – 50 $\mu\text{g}/\text{ml}$ with chloroform by dissolving 0.003g of standard L-carotene in 100ml of chloroform. The above gradients of different standard solutions prepared were determined with reference to their absorbance from which average gradient was taken to calculate Vitamin A (B-carotene in $\mu\text{g}/100\text{g}$) using Spectrophotometer (Metrohm Spectronic 21D Model) at a wavelength of 328nm.

Vitamin B (Niacin): About 5 g of the sample was treated with 50 ml of 1 N H_2SO_4 and shaken for 30 minutes. Thereafter, 3 drops of the ammonia

solution were added to the sample and filtered. Afterwards, 10 ml of the filtrate was added into a 50 ml volumetric flask and 5 ml of 0.02 N H_2SO_4 470 nm (AOAC, 2000, Hussian *et al.*, 2006, Iqbal *et al.*, 2011).

Vitamin C: One gram of each sample was weighed in a 25 ml conical flask. Then 10 ml of oxalic acid (0.05 M)-EDTA (0.02 M) solution was added and placed in the sample for 24 hours to provide the required reaction time. After 24 hours, the samples were filtered through using 0.45 μm filter paper. Then 2.5 ml of each sample was transferred to a separate 25 ml volumetric brown flask, after which 2.5 ml of the oxalic acid (0.05 M)-EDTA (0.02 M) solution was added.

Subsequently, metal phosphoric acid was added separately with acetic acid (0.5 ml), H_2SO_4 (5 % v/v) solution (1 ml) and ammonium molybdate solution (2 ml) each volumetric brown flask and the volume made up to 25 ml with distilled water. The absorbance was measured at 760 nm in a UV/visible spectrophotometer.

Vitamin E: One (1g) of the sample was weighed into a 250ml conical flask and filtered with a reflux condenser after which 10ml of absolute alcohol and 20ml of 1M alcoholic H_2SO_4 were added. The condenser and flask were wrapped in Aluminum foil and refluxed for 45 minutes and cooled for another 15 minutes. Fifty (50 ml) of distilled water was added to the mixture and transferred to a 250ml separating funnel covered with Aluminum foil. The unsaponifiable matters in the mixture were extracted with 5 x 30ml dimethyl ether. The combined extracts were washed free of acid and dry evaporated at a low temperature and the residues obtained were dissolved in 10ml absolute alcohol. Aliquots of solutions of the samples and standards (0.3-3.0mg vitamin E) were transferred to a 20ml volumetric flask after which 5ml absolute alcohol was added, followed by addition of 1ml concentrated Nitric acid. The flasks were placed on a water bath at 90°C for exactly 3 minutes from the time the alcohol begins to boil,

cooled under running water and adjusted to volume with absolute alcohol and absorbance was taken at 470nm against a blank containing 5ml absolute alcohol and 1 ml conc. Nitric acid (HNO₃) was treated in a similar manner (AOAC, 2000).

$$\text{Vitamin E}(\mu\text{g}/100\text{g}) = \frac{\text{Absorbances of sample} \times \text{Gradient factor} \times \text{Dil. Factor}}{\text{Weight of sample}}$$

Determination of anti-nutrients in *C. frutescens*, *P. guineense* fruits and *Z. zanthoxoides* roots

Phytic acid: Phytic acid was determined according to method of Sofowora, (1993). Two (2g) of each sample was weighed into 250ml conical flask. 100mls of 2% Hydrochloric Acid was added to soak each sample in the conical flask for 3 hours and filtered through a double layer of hardened filter paper. Then, 50ml of each filtrate was placed in 0.50ml conical flask and 107mls distilled water was added in each case to give proper acidity. Thereafter, 10mls of 0.3% Ammonium Thiocyanate (NH₄SCN) solution was added into each solution as indicated. This was titrated with standard iron (III) chloride solution which contained 0.00195g Iron per ml. The end point was slightly brownish-yellow which persisted for 5 minutes. The % phytic acid was calculated using the formula:

$$\% \text{ Phytic Acid} = \frac{\text{Titre value} \times 0.00195 \times 1.19 \times 100 \times 3.55}{\text{Wt. of sample}}$$

Tannin: Approximately, 0.50 g of sample was measured into a 50ml beaker and 20ml of 50% methanol was added and covered with paraffin and placed in a water bath at 77-80°C for 1 hour and shaken to ensure a uniform mixing. The extract was quantitatively filtered using a double layered Whatman No. 41 filter paper into a 100ml volumetric flask, 20ml water added, 2.5m; folin-Denis reagent and 10ml of 1% Sodium carbonate were added and. mixed properly. The mixture was made up to mark with water mixed well and allowed to stand for 20min. The bluish-green color was developed at the end of range 0-10ppm and treated similarly as 1ml sample above. The

absorbance of the Tannic acid standard solutions as well as samples was read after color development on a spectronic 21D spectrophotometer at a wavelength of 760nm. % Tannin was calculated using the formula.

$$\% \text{ Tannin} = \frac{\text{Absorbance of sample} \times \text{average gradient factor} \times \text{dilution factor}}{\text{Wt. of sample} \times 10,000}$$

Trypsin inhibitor: One gram of each sample was dispersed in 50 ml of 0.5 M Sodium Chloride solution. The mixture was stirred for 30 minutes at room temperature and centrifuged at 1500 rpm for 5 min. The supernatant was filtered and the filtrate was used for the assay. Two millilitres of the standard trypsin solution were added to 10 ml of the substrate of each sample. The absorbance of the mixture was taken at 410 nm using 10 ml of the same substrate as blank

Oxalates: Approximately, 2 g of the sample was boiled in 40 ml of water for 30 minutes in a reflux condenser and 10ml of 20% Sodium carbonate was added, boiled for another 30 minutes. The mixture was filtered and liquid extract washed with hot water until the wash water does not show any alkaline reaction. The combined wash water was filtered to a small volume and cool. With constant stirring, add Hydrochloric acid (HCL) (1:1) dropwise until the final acid concentration after neutralization was about 4% at which stage a heavy precipitate appeared and the extract was filtered into a 250ml flask to make up to mark and kept overnight. Aliquot of this filtrate was taken in a 400ml beaker, diluted with water to 200ml and make just ammoniacal, and reacidified with Lactic Acid. In the cold, medium, 10ml of a 10 % calcium chloride solution was added and stirred well to include calcium oxalate precipitate to appear and allowed to settle overnight. Clean supernatant liquid was decanted off through Whatman No. 42 filter paper, without disturbing the precipitate. The precipitate was dissolved in HCL (1:1). Oxalic acid was re-precipitated by adjusting the pH with ammonium hydroxide solution. The contents were boiled, allowed to

settle overnight and oxalic acid was determined by titrating against 0.05N Potassium permanganate solution.

Calculation

$$\begin{aligned}
 &1\text{ml of } 0.05\text{N KMnO}_4 \\
 &= 0.00225 \text{ anhydrous Oxalic Acid} \\
 &= \% \text{ Oxalic Acid} \\
 &= \frac{\text{Titre value} \times 0.00225}{2} \times \frac{100}{1} \\
 &= T.V \times 0.1125.
 \end{aligned}$$

Determination of phytochemicals in *C. frutescens*, *P. guineense* fruits and *Z. zanthoxloides*

Phytochemical contents of the samples were determined according to the methods of Harborne, (1973) in Awoyinka *et al.* (2016)

Alkaloids: Using distillation and titrimetric method described by (Harborne 1973), 2g of finely ground sample was weighed into a 100ml beaker and 20mls of 80% absolute alcohol added to give a smooth paste. The mixture was transferred to a 250ml flask and more alcohol was added to make up to 100ml after which 1g magnesium oxide added. The mixture was digested in a boiling water bath for 1.5hrs under a reflux air condenser with occasional shaking. The mixture was filtered while hot through a small bucher funnel. The residue was returned to the flask and re-digested for 30min with 50ml alcohol was evaporated after which, 3 drops of 10% HCl was added. The whole solution was later transferred into a 250ml volumetric flask 5ml of zinc acetate solution and 5ml of potassium ferrocyanide solution was added, thoroughly mixed to give a homogenous solution. The flask was allowed to stand for a few minutes and 10ml of the filtrate was transferred into a separatory funnel. The alkaloids present were extracted vigorously by shaking with five successive portions of chloroform. The residue obtained “as dissolved in 10ml hot distilled water

and transferred into a kjeldahl tube with addition of 0.20 g sucrose and 10ml Conc. H₂SO₄ and 0.02g selenium for digestion to a colorless solution to determine % N by Kjeldahl distillation method. %Nitrogen got was converted to % total alkaloid by multiplying by a factor of 3.26 i.e % Total alkaloid = %N X 3.26

$$\% \text{ alkaloids} = \% \text{N} \times 3.26$$

Flavonoids: About 0.50g of finely ground sample was weighed into a 100ml beaker and 80ml of 95% ethanol was added and stirred with a glass rod to prevent lumping and filtered into a 100ml volumetric flask and made up to mark with Ethanol. Also, 1ml of the extract was pipetted into 50 ml volumetric flask, four drops of Conc. Hydrochloric acid was added via a dropping pipette after which 0.5g of magnesium turnings added to develop a magenta red coloration. Standard flavonoid solution of range 0-5ppm were prepared from 100ppm stock solution and treated in a similar way with HCl and magnesium turnings like sample. The absorbance of magenta red coloration of sample and standard solutions were read on a digital Jenway V6300 Spectrophotometer at a wavelength of 520nm. The percentage flavonoid was calculated using the formula.

$$\begin{aligned}
 &\text{Flavonoids} \\
 &= \frac{\text{Absorbance of sample} \times \text{average gradient factor} \times \text{dilution factor}}{\text{Wt. of sample} \times 10000}
 \end{aligned}$$

Saponins: One (1g) of finely ground sample was weighed into a 250ml beaker and 100ml of isobutyl alcohol was added. The mixture was shaken on a UDY shaker for 5 hours to ensure uniform mixing. Thereafter the mixture was filtered through a Whatman No.1 filter paper into a 100ml beaker and 20ml of 40% saturated solution of magnesium carbonate was added. The mixture obtained with saturated Magnesium carbonate was again filtered to obtain a clear colorless solution. One (1ml) of the colorless

solution, was pipetted into 50 ml volumetric flask and 2ml of 5% Iron (III) chloride solution was added and made up to mark with distilled water. It was allowed to stand for 30min for blood red color to develop. 0-10ppm standard Saponin solutions were prepared from saponin stock solution. The standard solutions were treated similarly with 2ml of 5% Iron (III) chloride solution as done for 1ml sample above after which absorbance of the sample and standard saponin solutions were read after color development in a Jenway V6300 Spectrophotometer (380nm).

$$\% \text{ Saponin} = \frac{\text{Absorbance of sample} \times \text{gradient factor} \times \text{dilution factor}}{\text{Wt. of sample} \times 10000}$$

Steroids: About 0.50g of sample was weighed into a 100ml beaker and 20ml of Chloroform-Methanol (2:1) mixture was added to dissolve the extract after which the mixture was filtered into another 100ml Conical Flask. The resultant residue was repeatedly treated with Chloroform-Methanol mixture until free of Steroids. One (1ml) of the filtrate was pipetted into a 30ml test tube and 5ml of alcoholic potassium hydroxide was added and shaken thoroughly to obtain a homogenous mixture. The mixture was later placed in a water bath set at 37°C-40°C for 90minutes, cooled to room temperature and 10 ml of petroleum ether added followed by the addition of 5ml distilled water and later evaporated to dryness on the water bath. Six (6ml) of Liebermann Burchard reagent was added to the residue in dry bottle and was absorbance taken at a wavelength of 620nm on a Spectronic 21D digital Spectrophotometer.

Standard Steroids of concentration of 0-4mg/ml was prepared from 100mg/ml stock steroid solution and treated similarly like sample as above.

$$\text{Steroids} = \frac{\text{Absorbance of Sample} \times \text{Gradient} \times \text{Dilution Factor}}{\text{Wt of sample} \times 10000}$$

Anthocyanins: Approximately, 1g of sample was blended in a blender with 75ml of (Methanol: Water: Acetic Acid) (25: 24: 1) mixture to extract the anthocyanin. The extract was then centrifuged at 12,000rpm for 20mins at 15°C. The residue remaining was mixed thoroughly with the 75ml of (methanol/water/ acetic acid) mixture. The extraction was repeated thrice. The three extracts were pulled together into a 250ml beaker to evaporate to dryness in a rotary evaporator. The residue obtained above was re dissolved in 10ml of 15ml of 15% methanol and 85% of 5%(w/v) formic acid solution. This extract was diluted to 250ml with 135ml of a mixture of methanol/0.1M HCl at ratio of 85:15. Working standard solutions of anthocyanin of range 0-10mg/ml were prepared from stock 50mg/ml anthocyanin solution and treated like sample above. Absorbances of sample extracts as well as anthocyanin working standard solutions were read at a wavelength of 535nm on a UV Spectronic 21D Spectrophotometer.

$$\% \text{ Total Anthocyanin} = \frac{\text{Absorbance of sample} \times \text{gradient} \times \text{Dilution Factor}}{10,000}$$

Phenol: Approximately, 0.20g of sample was weighed into a 50ml beaker, 20ml of acetone was added and homogenized properly for 1hr to prevent lumping. The mixture was filtered into a 100ml Volumetric flask using acetone to rinse and made up to mark with distilled water. One (1ml) of sample extract was pipetted into 50ml Volumetric flask, 20ml water added, 3ml of phosphomolybdic acid added followed by the addition of 5ml of 23% Sodium carbonate and mixed thoroughly, made up to mark with distilled water and allowed to stand for 10min to develop bluish-green color. Standard phenol of concentration range 0-10mg/ml were prepared from 100mg/L stock Phenol solution from Sigma-Aldrich chemicals, U.S.A. The absorbance of sample and standard concentrations of Phenol were read on a Digital Spectrophotometer at a

wavelength of 510nm. The percentage Phenol is calculated using the formula:

$$\% \text{Phenol} = \frac{\text{Absorbance of sample} \times \text{gradient factor} \times \text{dilution factor}}{\text{Wt. of sample} \times 10,000}$$

Page | 3566

Statistical Analysis

Data obtained were analysed using Statistical Analysis System. One-way Analysis of Variance (ANOVA) was conducted to determine significant difference between parameters. Means were separated using Duncan's Multiple Range Test at $p < 0.05$.

Results

Distribution of respondents based on their socio-economic characteristics showed that the respondents were predominantly females (75.0%), mainly between 50-59 years, married (42.0%) with primary school as their highest educational attainment. Also, majority (94.0 %) of them practised Islam and were predominantly traders (78.0%). More than half (55.0%) of the respondents claimed to have between 10 -20 years in the sales of herbs used to treat toothache (76.0%). (Table 1). A total of twenty-three (23) plants belonging to 16 families were recorded, out of which *C. frutescens*, *P. guineense* and *Z. zanthoxloides* were the most exploited for management of the ailments (Table 2).

Roots (42.0%), leaves (10.0%), fruits (36.0%), and stems (10.0%) are the distribution of the plant parts commonly used. The herbal products are sourced mainly by foresters/ farmers (59.0%) both in fresh and dry form (96.0%), prepared mainly by infusion and applied predominantly by mouth washing, followed by chewing (40.0%) majorly one week (91.0%). The herbal preparations used for the ailments are majorly single plants

preparation (56.0%). Also, the preparations are often used in combination with non-plant materials such as salt (84.0 %), hot water (7.0%) and alum (9.0) (Table 3)

Significant difference ($p < 0.05$) was observed in proximate contents of *Z. zanthoxloides* roots, *C. frutescens* and *P. guineense* fruits studied. Crude fibre (8.86%), fat (1.12%), and ash (contents (4.73%) were significantly higher in *Z. zanthoxloides* roots, carbohydrate (96.23%) and moisture (25.75% dry matter basis) in *C. frutescens* fruits while similar values of crude protein (5.88, 5.99% dry matter basis) and were recorded in *Z. zanthoxloides* roots and *P. guineense* fruits (Table 4).

Results of the study based on quantities of mineral elements in the three plants revealed that sodium (51.33%), potassium (211.90%), calcium (129.00%) phosphorus (108.50 %), iron (4.85 %), Zinc (3.94 % s) manganese (1.15%) and copper (2.12 %) were significantly higher ($p < 0.05$) in *Z. zanthoxloides* roots compared with *C. frutescens* and *P. guineense* fruits (Table 5 and 6). Across the three plants studied, vitamin A (600.00µ/100g), vitamin B (0.07 mg/100g), vitamin C (94.54 mg/100g) and vitamin E (720.00µg/100g) were significantly ($p < 0.05$) higher in *C. frutescens* fruits studied compared with *Z. zanthoxloides* and *P. guineense* (Table 7).

Higher values of amount anti-nutrient studied such as tannin (6.40%), oxalate (30.0%) phytate (0.40.00%) and trypsin inhibitor (20.00%) were significantly recorded in *Z. zanthoxloides* roots (Table 8). Similar significant ($p < 0.05$) increase were reported in the amount of phytochemical contents such as flavonoid (3.25%), saponins (1.30%), phenol (0.60%), anthocyanins (0.23%) reported in *Z. zanthoxloides* than *C. frutescens* and *P. guineense* (Table 9)

Table 1: Socio-economic characteristics of respondent covered by the study

Variable	Frequency	% frequency	Mode
Gender			
Male	25	25.0	
Female	75	75.0	75.0
Age (years)			
20-29	3	3.0	
30-39	5	5.0	
40-49	22	22.0	
50-59	42	42.0	42.0
60 years and Above	28	28.0	
Marital status			
Single	6	6.0	
Married	58	58.0	58.0
Divorced	6	6.0	
Widow	30	30.0	
Highest education attainment			
Primary school	46	46.0	46.0
Secondary school	37	37.0	
Tertiary	17	17.0	
Religion			
Christianity	6	6.0	
Islam	94	94.0	94.0
Occupation			
Civil servant	6	6.0	
Trading	87	87.0	87.0
Farming	7	7.0	
Years of traditional herbal practice			
Less than 10 years	12	12.0	
10-20 years	55	55.0	55.0
20-30 years	33	33.0	

Table 2: Ethnobotanical information of plants used in managing toothache and mouth odour in Ijebu Ode Local Government Areas

Common name	Botanical name	Family	Part used	Frequency
Red pepper	<i>Capsicum frutescence</i>	Solanaceae	Fruit	30
Uziza pepper	<i>Piper guineense</i>	Piperaceae	Fruit	25
Artar root	<i>Zanthoxylum zanthoxyloides</i>	Rutaceae	Root	15
Bitter leaf	<i>Vernonia amygdalina</i>	Asteraceae	Leaf	2
Cashew	<i>Anacardium occidentale</i>	Anacardiaceae	Bark	5
Manding dyula	<i>Olaxsubscorpioidea</i>	Olacaceae	Root	2
Satinwood	<i>Distemonathus benthamianus</i>	Fabaceae	Root	2
Ugwu	<i>Telfaria occidentale</i>	Cucurbitaceae	Leaf	1
Tobacco	<i>Nicotiana tabacum</i>	Solanaceae	Leaf	5
Ginger	<i>Zingiber officinale</i>	Zingiberaceae	Root	3
Alligator pepper	<i>Aframomum melegueta</i>	Zingiberaceae	Fruit	5
African Birch.	<i>Anogeissus leiocarpus</i>	Combretaceae	Root/stem	4
Barbados nut	<i>Jatropha curcas</i>	Euphorbiaceae	Stem	11
African mesquite	<i>Prosopis Africana</i>	Leguminosae	Stem	3
Dogoyaro	<i>Azadirachta indica</i>	Meliaceae	Twigs	12
coffee senna	<i>Cassia occidentalis</i>	Caesalpinaceae	Root	5
Cashew	<i>Mangifera indica</i>	Anacardiaceae	Twigs	5
Stool wood	<i>Alstonia boonei</i>	Apocynaceae	Stem	7
African basil	<i>Ocimum gratissimum</i>	Lamiaceae	Stem	6
Cassia tree	<i>Senna siamea</i>	Fabaceae	Stem	5
Bitter	<i>Garcinia kola</i>	Guttiferae	Stem	7
Lime	<i>Citrus aurantifolia</i>	Rutaceae	Stem	8
Tuit	<i>Terminalia schimperiana</i>	Combretaceae	Root	6

Table 3: Plant parts and mode of administration of herbal preparations used for management of toothache and mouth odour in Ijebu Ode Local Government Areas

Variable	Frequency	% Frequency	Mode
Disease cured			
Toothache	76	76.0	76.0
Mouth odour	24	24.0	
Plant parts used			
Roots	42	42.0	42.0
Leaves	10	10.0	
Fruit	36	36.0	
Stems	10	10.0	
Source of herbal materials used			
Immediate house environs	18	18.0	59.0
Markets	23	23.0	
Foresters/Farmers	59	59.0	
Method of preparation adopted			
Infusion	44	44.0	44.0
Decoction	14	14.0	
Tincture	32	32.0	
Powder	10	10.0	
Method of administration adopted			
Bathing	4	4.0	54.0
Mouth-washing	54	54.0	
Massaging	2	2.0	
Chewing	40	40.0	
Duration of usage of the preparations			
5days- 1 week	91	91.0	91.0
2 weeks – 1 month	8	8.0	
More than a month	1	1.0	
How plant material is used			
Single plant preparation	56	56.0	56.0
Combination with plant materials	33	33.0	
Combination with non-plant material	11	11.0	
Non- plant material used			
Salt	84	84.0	84.0
Palm oil	7	7.0	
Alum	9	9.0	
Solvent used			
Pap water	5	5.0	70.0
Local gin	70	70.0	
Hot water	25	25.0	

Table 4: Variations of proximate content of plants used to treat toothache and mouth odour in Ijebu Ode Local Government Areas

Plants/parts	Proximate contents (%) dry matter basis					
	Crude protein	Crude fibre	Fat content	Ash content	Carbohydrate	Moisture
<i>Zanthoxylum zanthoxyloides</i> (root)	5.88±0.07 ^a	8.86±0.05 ^a	1.12±0.01 ^a	4.73±0.05 ^a	79.41±0.06 ^c	9.71±0.02 ^b
<i>Capsicum frutescense</i> (fruits)	0.94±0.02 ^b	2.38±0.04 ^b	0.12±0.01 ^c	0.32±0.01 ^c	96.23±0.04 ^a	25.75±0.02 ^a
<i>Piper guineense</i> (fruits)	5.99±0.03 ^a	3.31±0.04 ^b	0.22±0.02 ^b	1.33±0.02 ^b	89.15±0.05 ^b	8.75±0.08 ^c
P values (P<0.05)	0.00	0.03	0.01	0.00	0.02	0.04

Means ± standard error with different superscripts in columns are significantly different using Duncan Multiple Range Test P<0.05

Table 5: Variations of macro elements in plants used to treat toothache mouth odour in Ijebu Ode Local Government Areas

Plants/parts	Minerals (mg/100g)				
	Sodium	Magnesium	Potassium	Calcium	Phosphorus
<i>Zanthoxylum zanthoxyloides</i> (root)	51.33±0.47 ^a	5.24±0.01 ^b	211.90±0.80 ^a	192.00±1.33 ^a	108.50±0.19 ^a
<i>Capsicum frutescense</i> (fruit)	2.58±0.02 ^c	11.84±0.07 ^a	195.9±0.72 ^b	102.30±0.12 ^c	23.80±0.05 ^c
<i>Piper guineense</i> (fruits)	12.63±0.06 ^b	3.23±0.03 ^c	115.1±0.78 ^c	98.56±0.04 ^b	85.59±0.32 ^b
P values (P<0.05)	0.01	0.00	0.00	0.04	0.00

Means ± standard error with different superscripts in columns are significantly different using Duncan Multiple Range Test P<0.05

Table 6: Variations of macro elements in plants used to treat toothache mouth odour in Ijebu Ode Local Government Areas

Plants/parts	Mineral content (mg/100g)			
	Iron	Zinc	Manganese	Copper
<i>Zanthoxylum zanthoxyloides</i> (roots)	4.85±0.03 ^a	3.94±0.01 ^a	1.15±0.01 ^a	2.12±0.01 ^a
<i>Capsicum frutescense</i> (fruits)	0.55±0.02 ^c	0.16±0.01 ^c	0.07±0.01 ^b	0.07±0.01 ^b
<i>Piper guineense</i> (fruits)	1.96±0.02 ^b	2.13±0.02 ^b	0.11±0.02 ^b	0.09±0.01 ^b
P values (P<0.05)	0.03	0.00	0.04	0.04

Means ± standard error with different superscripts in columns are significantly different using Duncan Multiple Range Test P<0.05

Table 7: Variations of vitamin in plants used to treat toothache mouth odour in Ijebu Ode Local Government Areas

	Vitamin A (µg/100g)	Vitamin B (mg/100g)	Vitamin C (mg/100g)	Vitamin E (µg/100g)
<i>Zanthoxylum zanthoxyloides</i> (roots)	30.00±0.01 ^b	0.04±0.02 ^b	62.40±1.53 ^b	40.00±0.01 ^b
<i>Capsicum frutescense</i> (fruits)	600.00±0.03 ^a	0.07±0.01 ^a	94.54±0.20 ^a	720.00±0.03 ^a
<i>Piper guineense</i> (fruits)	20.00±0.01 ^c	0.02±0.01 ^c	54.60±1.10 ^c	10.0±0.00 ^b
P values (P<0.05)	0.00	0.02	0.00	0.01

Means ± standard error with different superscripts in columns are significantly different using Duncan Multiple Range Test P<0.05

Table 8: Variation of anti-nutrients in plants used to treat toothache mouth odour in Ijebu Ode Local Government Areas

Plants/parts	Anti-nutrient content (%)			
	Tannin	Oxalate	Phytic acid	Trypsin inhibitor
<i>Zanthoxylum zanthoxyloides</i> (roots)	60.00±0.05 ^a	30.04±0.01 ^a	0.40±0.02 ^a	2.00±0.03 ^a
<i>Capsicum frutescense</i> (fruits)	1.60±0.02 ^c	3.45±0.01 ^c	0.14±0.01 ^b	2.00±0.01 ^a
<i>Piper guineense</i> (fruits)	5.06±0.03 ^b	21.01±0.01 ^b	0.18±0.02 ^b	1.40±0.02 ^a
P values (P<0.05)	0.00	0.00	0.03	0.01

Means ± standard error with different superscripts in columns are significantly different using Duncan Multiple Range Test P<0.05

Table 9: Variation of phytochemical contents in plants used to treat toothache mouth odour in Ijebu Ode Local Government Areas

Plants/parts	Phytochemical Contents (%)					
	Alkaloids	Flavonoids	Saponins	Steroids	Phenol	Anthrocyanins
<i>Zanthoxylum zanthoxyloides</i> (roots)	3.23±0.13 ^a	3.25±0.01 ^a	1.30±0.03 ^a	0.20±0.04 ^b	0.60±0.05 ^a	0.23±0.05 ^a
<i>Capsicum frutescense</i> (fruits)	0.50±0.02 ^b	1.71±0.04 ^b	0.12±0.02 ^c	0.05±0.01 ^c	0.04±0.02 ^c	0.02±0.00 ^b
<i>Piper guineense</i> (fruits)	3.22±0.12 ^a	0.62±0.01 ^c	0.40±0.01 ^b	0.22±0.02 ^a	0.40±0.03 ^b	0.04±0.01 ^b
P values (P<0.05)	0.04	0.00	0.00	0.01	0.02	0.00

Means ± standard error with different superscripts in columns are significantly different using Duncan Multiple Range Test P<0.05

DISCUSSION

Hygienic oral health condition is one of the essential factors of quality life that determines

state of well-being of an individual because an unhealthy mouth and teeth condition affects all other parts of body (Idu *et al.*, 2009; Pradeep

2014; Richa *et al.*, 2014; Anyanwu and Nwosu, 2014; Awonrinde *et al.*, 2016). Based on the results of the present study, natural plant products are increasingly adopted as popular method of maintaining oral health care of people most especially among rural dwellers as indicated by 23 plants reported in this study.

C. frutescens, *P. guineense* and *Z. zanthoxyloides* prioritized by the Traditional Health Practitioners captured in the present study are adopted for treatment and general oral care cases as well as preventive measure of the challenge depending on the severity of the conditions.

This is probably due to the exorbitant financial health demand of dental therapy or on the other perspective; it may indicate that people are beginning to realize relevance of natural products including plants in the maintenance of personal hygiene and general wellbeing status (Nitika *et al.*, 2012). The significant number of plants recorded being prescribed by the Traditional Health Practitioners for treatment of oral ailments may suggest improvement in discovery on number of herbs used for treatment of oral health challenge of people in the study area. This is in agreement with submissions of Henley-Smith *et al.* (2013) and Pradeep (2014). Also, studies of Borokini *et al.* (2013) revealed *Z. zanthoxyloides* root, *Nicotiana tabacum* leaf powder, *Oxythenanthera abyssinica* leaves with little potash together and water of fermented corn extract as effective method of managing toothache. Also, results of several other studies also showcased roles of plants for management of diseases including unhealthy oral condition with or without non plants materials (Petersen 2003; Henley-Smith *et al.*, 2013; Pradeep 2014; Michael and Sudeshni 2015). In the same vain, Borokini *et al.* (2013) revealed that *Capsicum frutescens* fruit and *Piper guineense* fruit, together with a spoon of salt, small alum, little potash that are blended together as remedy to manage toothache.

According to Mayaud *et al.* (2008) and Bachir and Benali (2012) human oral cavity harbours diverse ranges of bacteria, fungi and protozoa as normal flora, but these micro-organisms when in excess may cause dental disease in poor oral hygiene and suppress immunological system of an individual which can lead to disintegration of organic substance of tooth, dental plaque, gingivitis, caries and periodontitis (Aworinde *et al.*, 2016). High adoption of the *Z. zanthoxyloides* root in the treatment of the ailment in the present study could be ascribed to the appreciable amount phytochemicals most especially saponins reported in the plant which has ability to act as cleansing agent. This may serve as basis for the use of the plants as chewing sticks most especially in the local communities.

Results of the present study are in line with findings of several researchers in other locations who posited that presence of phytochemicals such as saponins, flavonoids, tannins and phenolic present in *Z. zanthoxyloides* may be responsible for the use of the plants for health care service (Sofowora, 2008; Aworinde *et al.*, 2016; Elizabeth *et al.*, 2016). Also, findings of (Idu *et al.*, 2009) showed that root and stem of *P. guineense*, *Z. zanthoxyloides* and *Vernonia amygdalina* are used as remedy for treating toothache and other teeth related diseases.

Also, minerals have been reported to be playing lead role in oral health, for example, calcium strengthens teeth and health of the jaw bones. Magnesium for formation of teeth and in conjunction with calcium help to mineralize teeth while phosphorus collaborates with calcium on the formation of teeth (Ojewumi *et al.*, 2016a). However, appreciable quantities of these nutritional contents recorded in *C. frutescens*, *P. guineense* suggest the multipurpose usage of the plant as medicine as well as food (Eze and Obinwa, 2014; Ojewumi *et al.*, 2016a)

Conclusion

This study showed that several forest plants are used for management of toothache and mouth odour in Ijebu area but *Z. zanthoxloides* C. *frutescens* and *P. guineense* are the most adopted. Also, the plants contain nutritional and therapeutic index suitable for prevention of the ailments. This survey also clamors for concerted efforts towards research that can enhance better documentation of adequate records of indigenous method of treating toothache, body odour and mouth odour.

Recommendation

Results of this study recommend that the use of root, and fruits of *Z. zanthoxloides* C. *frutescens* and *P. guineense* should be considered as relevant method of treating toothache, mouth odour and body odour not only in Ijebu Ode but in any other communities where the plants can be found.

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Conflict of interest

The authors declare no conflicts of interest in this research.

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