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Ethnobotanical survey and phytochemical analysis of selected medicinal plants used in treating digestive disorder

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Abstract
Digestive system disorders have a substantial effect on worldwide morbidity and mortality rates. The other remedy is to solve digestive disorder drug chemical drugs that are; Mixmag, Boscopand, Doxycycline, flangy, Hyoscine, Vitamin C, some of which have their side effect. Medicinal plants including *Vernonia amygdalina, Terminalia chebula, Ocimum gratissimum, Cinnamomum zeylanicum* are the main herbal agents traditionally used by most Nigeria people to treat digestive disorder particularly, in the rural areas. To value the traditional use of these plants, Ethno-medicinal survey of herbal plants used for digestive disorder was conducted in Ibarapa Area of Oyo state, Nigeria. Primary data was collected from 30 concoction and herbal sellers who were randomly selected through a cluster sampling procedure. The result revealed a modern age of 20-29 years sell the herbs and concoction of wh66.7% of which were females while 33.2% were males. According to the herbal sellers, *Ocimum gratissimum, Vernonia amgdalina* and *Cinnamomum zeylanicum* were the most used herbs for the treatment of digestive disorder. Phytochemical screening of the plants indicated that, plants metabolites like Alkaloids, flavonoids, tannin, steroids, terpenoids, saponin, phenol and coumarin were present. However, quantitative study of the plants revealed that *Ocimum gratissimum* had the highest concentration of flavonoids (12.33mg/g), followed by *Vernonia amygdalina* (6.222mg/g) while the least was *Cinnamomum zeylanicum* (3.213mg/g). All the three plants were relatively high in tannin and total phenolic constituents. Therefore, presence of these phyto-chemical components could permit to justify their usage against digestive disorder.

Keywords: Ethno-medicinal, survey, phyto-chemical, digestive disorder

Introduction
Digestive system disorders have a substantial effect on worldwide morbidity and mortality rates. There are a wide number of digestive system disorders, which impose a substantial influence on morbidity and mortality rates, worldwide. In 2012, world health statistics reported that, digestive system disorders, particularly diarrhoea, was the fifth leading cause of global mortality, as approximately 100 million people died worldwide. A global progression has been observed in the reformation of interest in traditional system of treatments. According to [3], 'Ethno-medicinal plant studies have become of particular interest and have become increasingly more valuable in the development of health care and conservation programs in different parts of the world'. In an under developed countries, medicinal plants continue to be a main source of medication and estimation revealed that approximately 88% of the people rely mainly on traditional medicine of their primary form of medicinal health care [4]. Digestive system disorders re one of the most common types of ailment affecting human identified as the third highest cause of morbidity among the people in 2010 [5]. Several ethno-medicinal plants by traditional people against digestive system disorder are a common practice throughout the world.

Plants have attracted the attention of human since the advent of man on the earth and also found many uses in human health care. Many traditional systems have evolved in the world, which use plants to cater to the needs of health care and they are still in practice around the world. The use of plants and natural products received a fillip when World Health Organization recognized plant and natural products based medicinal systems as alternative and complimentary therapy in the year 2002 [14]. The use of medicinal plants for human health care is well documented in India, China, Egypt and Arab world [15]. In the traditional system of medicine, drugs are prescribed either as single plant products or a mixture of several plants

depending on the disease diagnosed for treatment, and are mainly administered orally [9]. Based on trial and error experiment of diverse plants on human, traditional system of medicine has been developed and is in vogue in the modern world [11].

The healing properties attributed to plant have a long time been linked to the phytochemicals. Phytochemicals are plant chemicals which have no nutritional values but occur naturally in plants that have protective or disease preventive properties [10]. They are non-essential nutrients which are not required by the human body for life sustainability [4]. It is well known that plant produces these chemicals for protection but recent research demonstrates that, they can also protect human against diseases [15]. Over thousand known phytochemicals are available and of the well-known ones are; Lycopene in tomatoes, Lavone sin in soy and flavonoids in fruits [9]. Ethnobotanical studies had revealed that out of the total flowering reported for medicinal purposes in Nigeria, about 800 plant species are used in traditional health care to treat close to 300 physical and mental disorders. Therefore, this study focused on the investigation of some medicinal plant used for the treatment of digestive disorders in rural area of Nigeria and to know their plant chemicals.

Materials and Methods

Ethno-medicinal plant survey

This survey was conducted in Ibarapa Local Government Zones, Ibarapa Central Local Government, Ibarapa North and Ibarapa East Local Government area of Oyo State Nigeria on latitude 7.53^0 and longitude 3.08^0 with a population of 102, 979 according to the 2006 population census. The major occupations of the people in the survey area are; farming, trading, hunting and teaching. Few are herbalists, artisans and traditionalists. A semi-structured questionnaire was distributed among thirty respondents in the surveyed area. Selection of the respondents were based on social status, occupation, name, age, gender, marital status and those approved by the community to have the knowledge of ethno-botanical use of plants. Questions emphasized on types and parts of plants often used for the treatment of digestive disorders cum mode of preparation and administration. Information on the medicinal plants was put together according to Generic name, family names, common names and parts of plants used. Fresh samples of the plant were obtained from the respondents and were dried using absorbent paper moistened with methylated spirit and mounted in accordance with conventional herbarium practice. These samples were identified by Professor A. E. Ayodele of the department of Botany, University of Ibadan, while authenticity was done by University of Ibadan Herbarium (UIH).

Plant preparation for therapeutic purpose as described by the respondents

Fresh and clean leaves of *Ocimum gratissimum* are crushed to squeeze the juice out. Salt is added and poured into a clean bottle. Half of a glass cup to be taken anytime and once daily for 5 day.

Clean leaves of *Vernonia amygdalina* are cut into smaller pieces in a bottle and some quantity of clean water is added, to soak for some minutes. Once the plant chemicals have been liberated into the water, a glass cup is taken two times daily for 5 days.

Clean inner bark of *Cinnamomum zeylanicum* plant is cut into smaller pieces, and soaked with clean water in a clean bottle for some minutes, after which half a glass cup could be taken two times daily for 5 days.

The seed of the unripe pawpaw are removed and discarded. The unripe pawpaw is cut into smaller pieces in a bottle with portable water added and shaken thoroughly. It is left for about 1-2 weeks and after which a glass cup can be taken once daily for five days.

The shrubs of *Terminalia chebula* are cut into pieces and squeezed. Then, it is poured into a clean bottle and water or alcohol is added and left for 15 mins. Half a glass cup is taken once a day for 5 days.

Phytochemical analysis of used for the treatment of digestive disorder

Determination of saponin

Twenty grammes of each sample were put into a conical flask and 100 cm^3 of 20% aqueous ethanol were added. The samples were heated over a hot water bath for 4 hours with continuous stirring at about 550 ^{0}C. The mixture was filtered and the residue re-extracted with another 200ml 20% aqueous ethanol. The combined extracts were reduced to 40ml over water bath at about 900 ^{0}C. The concentrate was transferred into a 250ml separating funnel and 20ml of di-ethyl ether was added and shaken vigorously. The aqueous layer was recovered while the ether layer was discarded, the purification process was repeated. 60ml of n-butanol was added and the combined n-butanol extract was washed twice with 10ml of 5% aqueous sodium chloride. The remaining solution was heated in a water bath, after evaporation, the samples were dried in the oven to a constant weight. The saponin content was calculated as a percentage [13].

Determination of alkaloids

5g of the sample was weighed into a 250ml beaker and 200ml of 10%acetic acid in ethanol was added, covered and allowed to stand for 4 hours. This was filtered and allowed the extract was concentrated on a water bath to one- quarter of the original volume. Concentrated ammonium hydroxide was added drop wise to the extract until the precipitation was complete. The whole solution was allowed to settle and the precipitated was collected and washed with dilute ammonium hydroxide and then filtered. The residue is the alkaloid, which was dried and weighed. Harborne [13].

Determination of total phenolic

The concentration of phenolics in plant sample was determined using Spectrophotometric method. Folin-ciocalteu assay method was used for the determination of the total phenol content. The reaction mixture consists of 1ml of extract and 9ml of distilled water was taken in a volumetric flask (25ml). 1ml of Folin-clocalteu phenol reagent was added to the mixture and shaken well. After 5 minutes, 10ml of 70% Sodium carbonate (Na_2CO_3) solution was treated to the mixture. The volume was made up to 2ml. A solution of standard solution of Gallic acid (20, 40,60,80 and 100µg/ml) were prepared in the same manner as described earlier, incubated for 90 mins at room temperature and the absorbance for test and standard solutions were determined against the reagent blank at 550nm with an Ultraviolet (UV)/Visible Spectrophotometer. The total phenol content was expressed as mg of GAE/gm of extract [13].

Determination of tannin

0.1ml of the sample solution was added to a volumetric flask (10 ml) containing 7.5ml of distilled water and 0.ml of Folin-

Ciocalteu phenol reagent, 1ml of 35% Na_2CO_3 solution and dilute to 10ml with distilled water. The mixture was shaken well and kept at room temperature for 30 mins. A set of reference standard solutions of gallic acid (20, 40, 60, 80 and 100µg/ml) were prepared in the same manner as described earlier. Absorbance for test and standard solutions were measured against the blank at 725nm with a spectrum lab 752s UV/Visible Spectrometric. The tannin content was expressed as mg of GAE/gm of extract [13].

Determination of flavonoid

Total Flavonoid content was measured by the Aluminum chloride Colorimetric assay. The reaction mixture consists of 1ml of extract and 4ml of distilled water was taken in a 10ml volumetric flask. To the flask, 0.30ml of 5% sodium nitrite was treated and after 5 minutes, 0.30 of 10% Aluminium chloride was mixed. After 5 minutes, 2ml of 1M sodium hydroxide was treated and diluted to 10ml with distilled water. A set of reference standard solutions of quercetin (20, 40, 60, 80 and 100µg/ml) were prepared in the same manner as described earlier. The absorbance for test and standard solutions were determined against the reagent blank at 510 nm with a UV/Visible Spectrophotometer. The total flavonoid content was expressed as mg of QE/g of extract [13].

Results and Discussion

Over the years with wide use of herbs, large number of drugs have been developed and introduced into international markets through exploration of ethno-pharmacology and traditional medicine [7], which have demonstrated different pharmacological potency [10]. In Ibarapa local government of Oyo state, Nigeria being rural, a large number of indigenes use herbal concoction purchased from herbal sellers and accessibility of the people to herbs is more than Western health care provider.

A total of 5 plant species belonging to five different families were found useful locally for the treatment of digestive disorders including diarrhoea, dysentery, abdominal pain and constipation as provided by the concoction sellers in the Ibarapa region of Oyo State, Nigeria. The local nomenclature of the plants and their specific use is presented in the table 1. *Ocimum gratissimum* belonging to the family of Lamiaceae and *Vernonia amygdalina* (Asteraceae) were observed to be used broadly for the treatment of digestive disorder such as diarrhoea, dysentery, constipation and abdominal pain as said by the respondents, affirms the research of [9] who reported that Lamiaceae had the highest medicinal usage for the treatment of digestive disorders in central Macedonia Greece followed by Asteraceae family. Asteraceae (*Vernonia amygdalina* leaves) was commonly used for the treatment of diarrhoea in this region (Ibarapa). This further confirmed the research of [18] on the treatment of diarrhoea with Asteraceae family (*Brachylaena ilicifolia* L leaves). Also observed and recorded from the herbal sellers was *V zeylanicum* belonging to the family of Lauraceae which is also frequently used for the treatment of diarrhoea.

Table 1: Ethnomedicinal plants in Ibarapa, Nigeria used for the treatment of Digestive disorder

Plant species	Local name	Family	Digestive disorder	Used parts
Ocimum gratissimum	Efirin	Lamiaceae	Abdominal pain, Diarrhoea, Dysentery, Constipation,	leaves
Unripe Carica papaya	Ibepe dudu	Caricaceae	Abdominal pain	Unripe fruit
Cinnamomum zeylanicum	Egbo cassia	Lauraceae	Diarrhoea	Inner bark
Terminalia chebula		Combretaceae	Diarrhoea	shrub
Vernonia amygdalina	Ewuro	Asteraceae	Diarrhoea, Dysentery	leaves

Table 2: Demographic profile of the respondents

Demographic characters	Frequency	Percentage (%)
Marital status		
singles	9	30
married	21	70
Age range		
20-29	13	26.6
30-39	14	20
40-49	18	20
50-59	11	10
60-69	13	10
70-79	10	6.7
	11	6.7
Occupation		
Concoction seller	11	36.7
Herbal seller	19	63.3
Local government		
Ibarapa central	13	43.3
Ibarapa East	10	33.4
Ibarapa North	7	23.3
Sex		
Female	20	66.7
Male	10	33.3

Demographic study as given by the respondents (table 2) revealed that 66.7% female and 33.3% males sell herbal concoction for the treatment of digestive disorder. That is, more female were herbal seller while those who had prepared the herbs into concoction before sales were also more of female (36.7%) than the male (11%) this conforms to the work of [1] who reported that females in Ekiti and Ondo state, Nigeria sell herbs than male. There are three local governments in the Ibarapa region and of these Ibarapa central had the highest population of herbal sellers (43.3%)

followed by Ibarapa East (33.4%) while the least was Ibarapa north (23.3%). Considering the age range of the herbal seller, 26.6% fall within the age range of 20-29 and as the ages increases population percentage of herbal seller reduces this might be due to agility and activeness derived from young ages which reduces as human being ages.The phytochemical investigation revealed the presence of saponins, flavonoids, coumarin and alkaloid in all the tested plants (table 3). In the other phyto-constituents results varied between plants. These commonly known phyto-compounds particularly flavonoids, saponin tannin and alkaloids have been reported to create barrier against sicknesses and diseases in man. These phyto-chemicals have been linked to different pharmacological activities which include antibacterial, antioxidants and analgesic properties, which could justify their traditional therapeutic use [2]. The quantitative phytochemical composition of the plants metabolites showed that tannin had the highest concentration in all the plant except *Terminalia chebula* which lacked tannin (table 4).

This might be one of the contributory factors that made *Ocimum gratissimum, Vernonia amygdalina* and *Cinnamomum zeylanicum* to be the most used plants for the treatment of digestive disorders. The high concentration of flavonoids (12.33mg/g) and tannin (7.05mg/g) in *Ocimum gratissimum* may contribute to its wide use as anti-diarrhoea because flavonoids and tannin are said to increase colonic water and electrolytes re-absorption [12]. The high total phenols in all the plants except *Terminalia chebula* is an indication that these plants may serve as good sources of antioxidants that scavenge any free radical in human body with digestive system disorder. The main threat on medicinal plants parts for traditional medicine preparation could also be used in the communities for maintaining their primary health.

Table 3: Qualitative phytochemical screening of the medicinal plant used in Ibarapa, Nigeria for the treatment of Digestive disorder

Phytochemicals	**Medicinal plant/inference**			
	Ocimum gratissimum	***Terminalia chebula***	***Vernonia amygdalina***	***Cinnamomum zeylanicum***
Saponins	+	+	+	+
Tannin	+	-	+	+
Flavonoids	+	+	+	+
Steroids	-	+	+	+
Coumarin	+	+	+	+
Emodin	-	+	-	-
Anthocyanin	-	-	-	-
Alkaloid	+	+	+	+
Cardiac glycoside	-	-	-	+
phenols	+	-	+	+
Chacones	+	-	+	-

Key: (Absent), + (Present)

Table 4: Quantitative screening of medicinal plants used in Ibarapa, Nigeria for the treatment of Digestive disorder

Phytochemicals (mg/g)	***Ocimum gratissimum***	***Terminalia chebula***	***Vernonia amygdalina***	***Cinnamomum zeylanicum***
Total phenols	$10.00^{\pm 0.051}$	-	$15.51^{\pm 0.001}$	$7.21^{\pm 0.003}$
Tannin	$7.05^{\pm 0.040}$	-	$9.90^{\pm 0.003}$	$6.31^{\pm 0.000}$
Alkaloid	$0.0074^{\pm 0.000}$	$0.0039^{\pm 0.000}$	$0.007^{\pm 0.000}$	$0.005^{\pm 0.001}$
Flavonoid	$12.33^{\pm 0.03}$	$4.32^{\pm 0.02}$	$6.22^{\pm 0.000}$	$2.20^{\pm 0.000}$
Saponin	$0.0067^{\pm 0.05}$	$0.0009^{\pm 0.04}$	$0.0072^{\pm 0.04}$	$0.0022^{\pm 0.000}$

Conclusion and recommendation

Medicinal plants still play an important role among people in Ibarapa Local Government. The present information on these medicinal plants, may serve as the baseline data to initiate further research for the discovery of new compounds and the biological activities of these potential plant remedies. Further research on these plants, may provide some important clues for the development of new drugs for the treatment

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