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Phytochemical Screening and Elemental Analysis of Crude Extract of Mentha Piperita (Peppermint) Leaves

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Abstract Phytochemicals are biologically active, naturally occurring chemical compounds found in plants, which provide health benefits for humans as medicinal ingredients and nutrients. In this research work, extracts of Mentha Piperita were prepared in n-hexane, acetone, dimethyl ether, petroleum ether, chloroform, ethanol and water using cold maceration technique and the extracts obtained were screened qualitatively for some secondary metabolites. Similarly, some essential elements that are vital for human life were also determined quantitatively. Alkaloids, flavonoids, tannins, steroids, glycosides, terpenoids, Saponins and phenols together with some vital elements; Ca, Na, K, Mg, Fe, Zn, Cd, Cr and Mn were found to be present in the leaf extract of Mentha Piperita depending on the solvent used for extraction and in various concentrations. This research concludes that; Mentha Piperita leaves contain some vital phytochemicals and essential elements that are vital for human health.

Keywords Phytochemicals, Medicinal Plant, essential elements, Extract, Mentha Piperita

Introduction

Plants are vital source of drugs; especially in traditional medicine. It is a common tradition to use plant as crude extracts, decoction, infusion, to treat common infection and chronic conditions practice in Nigeria and other parts of the world [1]. According to WHO, a significant number; up to 70% of the world population rely on medicinal plants for primary health care [2,3] and there are reports from various studies on natural substances of plant origin which are biologically active with desirable activity [4]. It is believed that isolated compounds due to their synergistic effects are less biologically active than crude extract from medicinal plants [3]. Natural products, such as plant's extract, either as pure compounds or as standardized extracts, provide indefinite prospects for new drug discoveries because of the unsurpassed availability of chemical multiplicity [5].

Medicinal plants are of great importance to the health of individuals and communities. The medicinal worth of these plants lies in some chemical substances that produce a certain physiological action on the human body [6].

Approximately 20% of known plants have been used in pharmaceutical researches, impacting the healthcare system in affirmative ways such as treating cancer and harmful diseases and a large number of diverse bioactive compounds were derived from plants [7].

Phytochemicals are naturally occurring chemical compounds that are biologically active found in plants, which provide health benefits for humans as medicinal constituents and nutrients [8]. Phytochemical analysis of plants has

shown the presence of several chemicals including alkaloids, tannins, flavonoids, steroids, glycosides and saponins among others [3].

Peppermint or mint (Mentha piperita L.) is a perennial aromatic herb that belongs to the Lamiaceae (Labiatae) family and is a natural hybrid between spearmint (Mentha spicata L.) and water mint (Mentha aquatic L.) [9]. It is 30–90cm high. Its stems are square erect or ascending, branched, the upper portion always quadrangular. Its leaves are opposite; dark green on the upper surface with purplish Flowers [10].

The members of Lamiaceae family are wide spread around the world with high concentration in the Mediterranean region [9, 11]. Several studies have established that the active compounds present in the lamiaceae family have natural antioxidant [12], antibacterial [13], antifungal [14] and anti-tumor activities [15]. The plant is widely used for treatment of digestive disorders and nervous system actions because of its antitumor and antimicrobial properties, chemo- preventive potential, its renal actions, antiallergenic activities, and also for lessening cramping, digestive complaints, anorexia, nausea and diarrhea in folk remedies and traditional medicine [9].

It has a long history of safe use in medicinal preparations and that makes it one of the most widely used herbs worldwide. Common cold, inflammation of the mouth, pharynx, liver, as well as disorders in the gastrointestinal tract such as nausea, vomiting, diarrhea, cramps, flatulence and dyspepsia are some health defects that this leaf is used to provide remedy for. It is also used as antioxidant, antimicrobial, antiviral, anti- inflammatory, and anti-carcinogenic [16]. It is called Nana in Arabic, Po Ho in Chinese, Peppermint in English, Menthe in French and Pudina in Hindi [17]

Materials and Methods

Collection and Identification of Plant Material

The fresh plant material was collected from Kabuga, gwale local Government, kano state Nigeria, identified by the herbarium officer of Botany Department, Faculty of science Usmanu Danfodio University Sokoto, Sokoto state Nigeria, as plant *Mentha piperita*, family: *Lamiaceae.* Voucher specimen (UDUH/ANS/0217) and was deposited at university herbarium for reference.

Sample Preparation and Extraction

The identified plant material was washed with distilled water and was air dried for 7 days in the laboratory at room temperature then was sorted to remove dead ones and debris, pounded to powder with a clean wooden mortar and pestle. The pulverized powder plant material was carefully weighed using an electric weighing balance. The weighed plant material was transferred in to a clean polythene bag and sealed perfectly. 10g of the powdered plant material were weighed and transferred in to seven (7) different glass vessels. Carefully, 100ml of different solvents; acetone, methanol, chloroform, diethyl ether, n-hexane, petroleum ether and water were into the vessels containing the plant material, stirred properly and allowed to stand for seven (7) days. The stirring was repeated periodically. It was filtered using Whatman no. 1 filter paper. The filtrate was then concentrated by evaporation on water bath at 65 ^{o}C and finally stored at 4 ^{o}C for use.

Phytochemical Analysis

Qualitative analysis of phytochemicals constituents such as Alkaloids, flavanoids, steroid, terpenoids, saponins, tannins, phenols and glycosides were carried out for all the extracts using procedures adopted from [18].

Akaloids

The extracts were dissolved individually in dilute HCl and filtered. The filtrates were treated with Wagner's reagent (iodine in potassium iodide). Formation of brown/reddish precipitate was observed which indicates the presence of alkaloids.

Flavonoids

2ml of the extracts were treated with conc. H_2SO_4. The solutions turns to orange colour, indicating the presence of flavonoids

Steriods and Terpenoids (Libermann – Burchard test)

4mg of extracts were treated with 0.5ml of acetic anhydride and 0.5ml of acetic acid. Then concentrated H_2SO_4 were added slowly. The reaction mixture change to blue green colour and reddish brown in separate layer, indicating the presence of terpenoids and steroids respectively

Saponins (foam test)

About 2g of the plant extracts were mixed with 10ml of distilled water in test tubes. The mixture was shaken thoroughly while corked for about 30 seconds, and was allowed to standard for half an hour. The presence of saponins was confirmed by the presence of a honey comb forth

Tannins

0.5g of dried powdered sample was boiled in 20ml of water in a boiling tube and then filtered. A few drops of 0.1% $FeCl_3$ were added to the filtrates. The reaction mixture turns to blue black coloration which is an indication of the presence of tannin in the sample

Phenols

About 2ml plant extracts was taken to water bath and warm at 45 ºC– 50 ºC. Then 2ml of 0.3% $FeCl_3$ were added. The mixture changed to blue, indicating the presence of phenols

Glycosides

1ml of Fehling's solution A and B were diluted with 8ml of distilled water and boiled for 1minute. To this clear blue solution (1ml) and 8 drops of plant extracts were added and boiled in a water bath for 5min. The presence of glycoside was indicated by a brick-red precipitate

Elemental Analysis

The concentration of mineral elements such as calcium, potassium, magnesium, sodium, manganese, copper, zinc, chromium, cadmium were determined by digestion of 2g powdered sample with 30ml of diacid mixture ($4HNO_3$: $1HClO_4$) in a conical flask heated on a hotplate in a fume hood till clear white precipitate settled down at the bottom of the conical flask. The precipitate was dissolved in 1% HCl prepared in double glass distilled water, filtered and volume of the filtrate was made to 100ml with deionised water [19]. The concentrations of mineral elements were determined by atomic absorption spectrophotometer according to Perkin –Elmer method [20]. Analysis of mineral element contents was carried out in triplicates; means were determined and reported in milligram per gram (mg/g)

Results and Discussion

Qualitative phytochemical analysis

The preliminary phytochemical analysis of leaf extract of Mentha Piperita is presented in Table 1. The results indicate that; flavonoids were present in the extracts of all solvents employed in the course of this research except in acetone. Similarly, terpenoids were detected in all solvents except in ethanol. Saponins on the other hand are found to be present in all solvents including water. Steroids and alkaloids are however found to be present in all solvents excluding water. Tannins and phenols are somewhat similar, been absent in petroleum ether and diethyl ether but present in n-hexane, acetone and ethanol. While tannins are present in chloroform and absent in water; phenols shows the opposite; present in water and absent in chloroform.

Table 1

	Extracting Solvents						
Phytochemical constituents	**Petroleum Ether**	**n-hexane**	**Diethyl ether**	**Acetone**	**Chloroform**	**Ethanol**	**Water**
Alkaloids	+	+	+	+	+	+	_
Flavonoids	+	+	+	_	+	+	+
Terpenoids	+	+	+	+	+	_	+
Saponins	+	+	+	+	+	+	+
Tannins	_	+	_	+	+	+	_
Phenols	_	+	_	+	_	+	+
Glycosides	+	+	_	_	+	_	_
Steroids	+	+	+	+	+	+	_

A number of biological activities that protect from most of chronic diseases are exhibited by different extracts of Phytochemicals such as alkaloids, flavonoids, steroids, cardiac glycosides, phenols, Saponins and tannins [2]. Plant alkaloids are one of the largest groups of natural products representing a highly diverse group of chemical entities; many of which possess potent pharmacological activities such as antibacterial, antifungal and antiviral properties [21].

In their review, Tungmunnithum *et al*., reported the anti-oxidant, antibacterial, anticancer, anti-inflammatory, immune system promoting and cardio-protective potentials of flavonoids [22]. Different studies have illustrated the health benefits of Saponins among which are their effect on blood cholesterol levels, cancer, bone health and stimulation of the immune system [11]. Similarly, they are found to be strong expectorant and aid the absorption of nutrients [18]. Dietary intake of flavonoids containing foods potentially lowers the risk of certain free radical pathophysiology. Central Nervous System, cardiotonic, lipid lowering, anti-ulcer, hepato-protective and hypoglycemic activities were also reported for flavonoids. Cardio glycosides are also of medicinal significance and used in the treatment of congestive heart failure and cardiac arrhythmia. Phenols and phenolic compounds have marvelous antimicrobial potential and have been extensively used in disinfections. Steroids are pharmacologically active compounds and show analgesic properties and central nervous system activities. Some tannins stimulate glucose uptake and exhibit insulin like activity acting as glucose transport activators of fat cells [2]. Terpenoids display momentous pharmacological activities which include but not limited to antiviral, antibacterial, antimalarial, anti-inflammatory, cholesterol synthesis inhibition and anti-cancer activities [18].

Mineral Elements Analysis

The results of mineral elemental analysis in the leaves of Mentha Piperita are presented in Table 2. Mineral elemental composition of the leaves of Mentha Piperita decreases in the following order: Ca (21.5223mg/g), K (3.3000mg/g). Mg (2.0077mg/g), Fe (0.4777mg/g), Na (0.1200mg/g) Mn (0.0119mg/g), Cu (0.0035mg/g), Zn (0.0034mg/g), Cr (0.0014mg/g) and Cd (0.000058mg/g). The results presented above indicate that leaves of Mentha Piperita contain adequate amount of essential elements that are beneficial to human health.

Table 2

Element	**Value (mg/g)**
Ca	21.5223
K	3.3000
Mg	2.0077
Fe	0.4777
Na	0.1200
Mn	0.0119
Cu	0.0035

Zn	0.0034
Cr	0.0014
Cd	0.000058

Conclusion and Recommendations

Conclusion

Phytochemicals and macro elemental composition of Mentha piperita leaves were investigated in this research work. Findings revealed the presence of some important secondary metabolites and essential elements beneficial to human and his health. A variation of phytochemical parameters encountered depends on the solvent used for extraction. It's therefore concluded that Mentha Piperita leaves possess some vital medicinal ingredients that can augment the wellbeing of the body and stimulate its functions. The study consequently provides further evidence on the traditional usage of this plant extract as beverage and in treating diseases.

Recommendations

On the basis of the findings obtained in this research work; the following recommendations are drowned:

- Further study is necessary to isolate and characterize the secondary metabolites detected in this work
- Biological activities of this plant needs to be evaluated and their actions against various groups of pathogens be ascertain
- Finally, the antinutrientional factors needs to be ascertain.

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