Phytochemical Screening and Mineral Analysis of Grewia mollis Stems Bark

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ABSTRACT

**Aims:** Aqueous extract of *Grewia mollis* stem bark was analyzed for its phytochemical constituents and mineral elements.

**Place and Duration of Study:** Department of Biochemistry, University of Jos, Jos, between June 2013 and July 2013.

**Methodology:** The phytochemical screening and mineral composition of the stem bark was carried out using methods by Sofowora, Trease and Evans, Harborne and Oshodi.

**Results:** The phytochemical screening of the stem bark showed the presence of Tannins, Saponins, Cardiac glycosides, Flavonoids, Steroids, Phenols and Resins. Alkaloids and Carbohydrates were not detected in the stem bark of the plant. Mineral analysis showed that the stem bark contains among others Manganese (1.899 g/100g), Zinc (0.375 g/100g), Magnesium (287.060 g/100g), Iron (47.941 g/100g), Copper (0.270 g/100g) and Lead (0.206 g/100g).

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1. INTRODUCTION

Plants and plant parts that possess medicinal values or exert important pharmacological effects in the animal body are generally known as medicinal plants. And it is now understood that the plants which naturally synthesize and accumulate some secondary metabolites and vitamins, possess medicinal properties [1]. Medicinal plants promote healthy life of a country. They play an important role by providing preliminary health care services to both urban and rural people. They also serve as an important therapeutic agents as well as important raw materials for manufacture of both traditional and modern medicine. This can serve as a source of foreign exchange for a country by exporting medicinal plants to other countries. Hence, indigenous medicinal plants play significant role in the economy of a country [1]. Medicinal plants contain both organic and inorganic constituents and many medicinal plants are found to be rich in one or more individual elements, thereby providing a possible link to the medicinal value of the medicines [2,3]. Minor elements play a vital role in the synthesis of bioactive chemical constituents in medicinal plants and are therefore responsible for both their medicinal and toxic properties [4]. Thus, a quantitative estimation of minor and trace mineral elements can be important in determining the effectiveness and toxicity of the medicinal plants in treating various diseases and understanding their pharmacological actions [5].

Grewia mollis (Tiliaceae) is a shrub or tree widely distributed in northern Nigeria and some African countries. It is known to be a strong fire-resistant plant and its parts are used in food and medicines [6]. In Nigeria, the stem bark powder or mucilage of G. mollis is used as a thickener in local cakes made from beans and corn flour commonly called “Kosai” and “Punkasau” respectively [6,7]. The dried stem bark is pulverized and the powder mixed with beans or corn flour enhances the texture of food product. Some findings have shown that mucilage collected from the stem bark of G. mollis can serve as a good binder in paracetamol formulations [8,9]. Furthermore, recent reports suggest that high concentration of this stem bark in diets may cause some adverse effects such as liver injury [6]. The phytochemical studies of G. mollis showed the presence of tannins, saponins, flavonoids, glycosides, phenols and steroids but absence of alkaloids in the leaves and stems bark [10] while the presence of alkaloids was revealed in the roots [11]. The infusion of stem bark obtained by cold or hot maceration in water is applied to give a smooth surface to mud walls and floors [12] while the wood ash is used as salt substitute including the ash of the leaves, roots and stems. Also, the flowers, buds and young shoots are added to soups and sauces as garnishing while in Sudan, the young leaves are cooked and eaten as vegetables. The fruit is eaten boiled or raw [13]. Studies have also demonstrated a wide spectrum of therapeutic effects such as hepatoprotective [14], antibacterial [15] and anti-inflammatory [15] activities. The present study was carried out to investigate some pharmacologically important phytochemicals and mineral compositions of Grewia mollis stem bark.

2. MATERIALS AND METHODS

2.1 Chemicals and Reagents

The sodium hydroxide, ferric chloride, copper acetate, petroleum ether (40–65°C), chloroform, naphthol, ammonium sulphate, Cupric sulphate, 5-hydrate, sulphuric acid, amyl alcohol, acetone, methanol, ethanol used were all products of BDH Chemicals Ltd., Poole, England. All other reagents used were of analytical grade and prepared in the Biochemistry laboratory, University of Jos, Nigeria, in glass apparatus using distilled water.

2.2 Plant Material

G. mollis stem bark was obtained from Akwanga-Nasarawa, Nasarawa State, Nigeria and was authenticated at Herbarium of the Department of Botany, University of Jos, Nigeria. A voucher specimen was placed at the Herbarium of this Institute.

2.3 Preparation of Extract

The stem barks of G. mollis were washed and oven dried at 37°C for 72 hours to a constant
The dried pieces were then pulverized using an electric blender. The powdered material was stocked in a glass container from which 150 g was extracted in 1500 ml of cold distilled water for 48 hours at 37°C. This was then filtered with Whatman No. 1 filter paper. The filtrate was concentrated on a steam bath to give the extract. The extract was then used for phytochemical screening and mineral analysis.

2.4 Phytochemical Screening of the Extract

The aqueous crude extract of *G. mollis* stem bark was subjected to various chemical tests in order to determine the phytochemicals present by using standard methods as described by Sofowora [16], Trease and Evans [17] and Harborne [18].

2.4.1 Test for alkaloids

1 ml of the extract was stirred with 5 ml of 1% aqueous HCl on a steam bath and filtered while hot. Distilled H$_2$O was added to the residue while 1 ml of the filtrate was treated with a few drops of Mayer's reagent (Potassium mercuric iodide solution). The formation of a cream color with Mayer's reagent indicates the presence of alkaloids.

2.4.2 Test for flavonoids

Few drops of 1% NH$_3$ solution was added to the extract of *G. mollis* stem bark in a test tube. Formation of yellow coloration was observed for the presence of flavonoids.

2.4.3 Test for steroids

0.5 ml of the aqueous stem bark extract was evaporated and dissolved in 2 ml chloroform. 2 ml of concentrated H$_2$SO$_4$ was carefully introduced into the test tube through the glass wall of the test tube. Formation of red colour ring confirmed the presence of steroid.

2.4.4 Test for phenols

5 ml of the aqueous extract was pipetted into a 30 ml test tube and then 10 ml of distilled H$_2$O was added. 2 ml of ammonium hydroxide solution and 5 ml of concentrated amyl alcohol were also added and left to react for 30 minutes. Development of bluish green colour was taken as a positive presence of phenol.

2.4.5 Test for resins

0.5 ml of the extract was evaporated and dissolved in 2 ml of petroleum ether, 2 ml of 2% copper acetate solution was then added and the mixture was shaken vigorously and allowed to separate, formation of green colour indicates the presence of resin.

2.4.6 Test for tannins

1 ml of the extract was boiled in 20 ml of H$_2$O in a test-tube and then filtered. Few drops of 0.1% ferric chloride was added and formation of green or a blue–black coloration which confirms the presence of tannin.

2.4.7 Test for saponins

2 ml of double distilled water was added to 1 ml of the extract. Few drops of olive oil were added and agitated. Formation of soluble emulsion indicated the presence of saponin.

2.4.8 Test for cardiac glycosides

0.5 ml of the extract was evaporated and dissolved in 1 ml glacial acetic acid. One drop of 10% ferric chloride was then added. 1 ml of concentrated H$_2$SO$_4$ was added by the side of the test tube. Appearance of brown colour ring at the interface indicated of presence of cardiac glycosides.

2.4.9 Test for carbohydrates

2 drops of 10% alcoholic solution of naphthol and 2 ml of concentrated H$_2$SO$_4$ were added to 2 ml of the extract, gently poured along the side of the test tube at an angle of 45°. Formation of a purple ring at the interface of the two liquid layers confirmed a positive result for the presence of carbohydrates.

2.5 Mineral Analysis

The atomic absorption spectrophotometer (AAS) was used for the analyses of magnesium, zinc, iron, manganese, copper and lead in the stem bark of *G. mollis*. Using AAS, a known amount of the sample was placed in a dish and heated with burnsen burner in a fume cupboard until there was no smoke emitted. This was transferred to the dessicator in order for it to cool after which 0.1 ml HCl solution was added to the ash. The resulting solution was filtered and diluted. Suitable salts of the metals in questions were...
used to make their standards, lamps were fixed and the analyses were done [19].

2.6 Statistical Analysis

Data were expressed as Mean ± Standard Error of Mean (SEM) values using statistical package for social sciences (SPSS) version 18.0.

3. RESULTS

Table 1 shows the phytochemicals present in G. mollis stem bark. Tannins, Saponins, Cardiac glycosides, Flavonoids, Steroids, Phenols, and Resins were detected in our study. However, Alkaloids and Carbohydrates were not detected which was in support with Onwuliri et al. [10]. The mineral analysis of G. mollis (Table 2) showed rich content of macro and micro elements like Manganese (1.899 g/100g), Zinc (0.375 g/100g), Magnesium (287.060 g/100g), Iron (47.941 g/100g) and Copper (0.270 g/100g). Stem bark of the plant also contains trace quantity of Lead (0.206 g/100g).

Table 1. Qualitative phytochemical screening of Grewia mollis stem bark

| Phytochemicals     | Grewia mollis stem bark |
|--------------------|-------------------------|
| Alkaloids          | -                       |
| Flavonoids         | +                       |
| Tannins            | +                       |
| Saponins           | ++                      |
| Steroids           | +                       |
| Cardiac glycosides | +                       |
| Phenols            | +                       |
| Resins             | +                       |
| Carbohydrates      | -                       |

Key = +: weak; ++: strong; -: absent

Table 2. Mineral composition of Grewia mollis stem bark

| Minerals       | Composition (g/100g) |
|----------------|----------------------|
| Manganese      | 1.899±0.003          |
| Zinc           | 0.375±0.001          |
| Magnesium      | 287.060±0.001        |
| Iron           | 47.941±0.002         |
| Lead           | 0.206±0.001          |
| Copper         | 0.270±0.001          |

Values are expressed as mean ± SEM (n = 3)

4. DISCUSSION

Medicinal plants are plants that have one or more of their organs containing substances used for the prevention and cure of diseases and they are used as starting materials for the synthesis of useful drugs. The crude extracts or purified form of plant has been used as medicines and cosmetics [1]. The medicinal value of these plants lies in bioactive phytochemical constituents that produce definite physiological action on the human body [20]. In this study, the phytochemical investigation of G. mollis stem bark indicates the presence of Tannins, Saponins, Cardiac glycosides, Flavonoids, Steroids, Phenols and Resins. This is in agreement with the work done by Onwuliri et al. [10]. These phytochemicals are well known for their pharmacological activities.

Saponins are steroid or triterpenoid glycosides characterized by their bitter or astringent taste, foaming properties [21]. Studies have shown that saponin as a phytochemical found in plants have antitumor and antimutagenic properties and can also lower the risk of cancer in humans by reducing the growth of cancer cells. They reduce the growth and viability of the cancer cells by reacting with the cholesterol rich membranes of these cancer cells. Rao and Sung [22] showed in their studies that saponins have the ability to prevent colon cancer. Another properties exhibited by saponin include anti-hyperglycemic property as reported by Malinow et al., Olaleye [23,24]. Thus, stem bark of G. mollis may be used as an anti-diabetic agent due to the presence of saponins in it. Furthermore, studies have shown that plants containing saponins have hemolytic activities on red blood cell membrane [25]. This may also be applicable to G. mollis stem bark. Studies have shown that tannins and flavonoids have anti-diarrheal activity [26] by increasing the colonic water and electrolyte reabsorption while other phytochemicals involved act by preventing intestinal mobility [26]. The stem bark of G. mollis may be used in the treatment of diarrhea due to the presence of tannins and flavonoids. Also, tannins have astringent properties, the ability to hasten the healing of wounds and inflamed mucus membranes. Studies have shown that plants contain tannins are used in ethnomedicine for wound healing, haemorrhoids, varicose ulcers, burns and frost-bite [27,28]. Flavonoid as a very important phytochemical has varied biological activities which include inflammation, prevention against allergies, platelets aggregation and tumor[29]. They also exhibit free radical scavenging, anti-cancer and antioxidant activities [30]. Hence, the presence of flavonoid in the stem bark of G. mollis may make it suitable for
the treatment of inflammatory diseases and wound healing. *G. mollis* stem bark contains Cardiac glycoside, a phytochemical that has been used as an aid in the treatment of congestive heart failure and cardiac arrhythmia. The phytochemical inhibits the Na⁺/K⁺ pump therefore causing an increase in the level of sodium ion in the myocytes which brings about increase in the level of calcium ions. Hence, the availability of Ca²⁺ for the contraction of the heart is increase causing reduction in distention of the heart and improved cardiac output [31]. Phenols and phenolic compounds show antimicrobial activities, used as disinfectants and as a standard for other bactericides [32]. Thus, the presence of phenolic compounds in *G. mollis* stem bark might be responsible for its antimicrobial activities. Other phytochemicals such as Steroids and Resins might be responsible for the some pharmacological activities of the plant.

Minerals which are either macrominerals or microminerals are essential for the normal growth and maintenance of the body. *G. mollis* stem bark mineral analysis showed the presence of manganese, zinc, magnesium, iron, copper and minute amount of lead (Table 2). Magnesium is used as a cofactor in over 300 enzymatic reactions taking place in the human body [33,34] including Krebs cycle, nucleic acid synthesis, glycolysis, amino acid activation, creatine phosphate formation, cyclic AMP formation, cardiac and smooth muscle contractibility and protein synthesis etc. Also, it plays a vital role in the utilization of water soluble vitamin B complex and fat soluble vitamin E and helps to maintain the fluid and electrolyte balance of the body by the help of other elements such as sodium, calcium and potassium [35]. Copper is an essential trace element that plays a vital role in redox reactions and the scavenging of free radicals in the body and this is due to the ability of copper to accept and donate electrons [36]. Iron is very important in the formation of haemoglobin in red blood cells and deficiency of iron leads to anaemia. *G. mollis* stem bark could be used as a source of iron in the treatment of iron-deficiency anaemia. Manganese an important trace element plays an important role in a number of biological activities, as a constituent of multiple enzymes and an activator of other enzymes [37]. Manganese-activated enzymes found in the body play important roles in the metabolism of biomolecules such as carbohydrates, amino acids, and cholesterol. It is also thought to aid in the maintenance of epithelia tissue [38]. Zinc is essential in the activation of certain enzymes. These include dehydrogenase, alkaline phosphatase and carboxypeptidase. Zinc containing organic compounds is employed as astringent and antifungal agents. It aids wound healing and metabolism of nucleic acid and insulin. Excess zinc causes anaemia and deficiency of zinc in the body can cause dermatitis [39]. Effect of lead deficiency in the body results in anemia while excess lead in the body can cause brain damage [40].

5. CONCLUSION

This study shows that *G. mollis* stem bark is a rich source of significant phytochemicals and minerals which may be responsible for the various therapeutic uses attributed to it in ethnomedicine. However, the presence of minute quantity lead (Pb) in the stem bark of the plant may indicate some level of toxicity.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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