ABSTRACT

Pods of *Acacia nilotica* and leaves of *Tapinanthus dodoneifolius* were spectrophotometrically analysed for their mineral constituents (Ca, Cu, Fe, K, Mg, Mn, Na, P and Zn). Phytochemical analysis was conducted on successive Soxhlet extracts of ethanolic, n-hexane and ethyl acetate of the pods and leaves, respectively. Furthermore, the antibacterial activities of the extracts were investigated. Results showed high Ca and K with low Cu and Zn concentrations in both plants. The concentration of P in the pods of *A. nilotica* was highest. Phytochemical analysis revealed the presence of steroids, tannins and saponins in both ethanolic and ethyl acetate extracts respectively, for both *A. nilotica* and *T. dodoneifolius*. Flavonoids were present only in the ethanolic extract of *A. nilotica*. The ethanol extract generally had more phytochemical agents in both plants studied. All solvent extracts of *T. dodoneifolius* showed activity against *Escherichia coli*, *Staphylococcus aureus*, *Klebsiella aerogenes* and *Proteus mirabilis* showed varying activities only for *A. nilotica* solvent extracts, and only ethyl extract recorded activity against *S. aureus*, while the n-hexane extract showed no activity against any pathogen. The ethyl acetate extract of *A. nilotica* showed significant activity against *K. aerogenes* and *P. mirabilis*. The results in this work tend to agree with the ethno-medical claims.

Keywords: ethno-medical, mineral constituents, phytochemical analysis

INTRODUCTION

Human disease management in Nigerian history addresses in one accord evidence of the relationship of plants and medicine as elsewhere the world over (Bannerman et al. 1986; Hammer 1999; Raghabendra et al. 2006; Ayandele and Adebeyi 2007). In Kano, northern Nigeria, tuberculosis is a major health challenge, Family Health International (FHI) 2001; Nwankwo et al. 2005; Emokpae et al. 2006) and it is locally believed that the pods of *Acacia nilotica* and the leaves of *Tapinanthus dodoneifolius* possess medicinal properties that are effective in the management of tuberculosis as well as other ailments (stomach ache, diarrhea, dysentery, wound and cancer) (Deeni and Sadiq 2002). The pods of *A. nilotica* are strongly constricted, hairy, white-grey, thick and softly tomentose. The pods without seeds are known to have a high tannin content of about 50%. Generally, the *A. nilotica* tree is 5-20 m high with a dense spheric crown, stems and branches usually dark to black coloured, fissured bark, grey-pinkish slash, exuding a reddish low quality gum. In Ayurvedic medicine, *A. nilotica* is considered a remedy that is helpful for treating premature ejaculation. And in ancient Ethiopian traditional medicine, *A. nilotica* in certain mixtures have been effective in the treatment of rabies (Clement 1998; Murthy et al. 2003). *T. dodoneifolius* is a hemi-plant parasite usually growing on *Vitellaria paradoxa* trees with large galls, commonly known as African mistletoe and ‘Kauchi’ in Hausa (vernacular). In northern Nigeria, the Hausa and the Fulani tribes utilize it ethnomedicinally as a remedy for several human and animal ailments (Deeni and Sadiq 2002). According to Nacouma (1996), the aqueous extract is taken against gynecologic disturbances, digestive disorders and nervous confusions. More recently, the cardiovascular properties of *T. dodoneifolius* have also been described by Ouédraogo et al. (2005). Traoré (2000) analysed different extracts from *T. dodoneifolius* which showed the presence of triterpenes, sterols, carotenoids, saponoids, anthraconesides, anthocyanosides and tannins.

Both *A. nilotica* and *T. dodoneifolius* plants are found in abundance and used extensively in forms of decoction and concoctions for the management of tuberculosis in Niabawa, Kumbotos Local Government Area (LGA) of Kano State, Nigeria. Hence the need to investigate these plants originated from this locality. Though, Deeni and Sadiq (2002) have investigated the antimicrobial constituents and phytochemical constituents of *T. dodoneifolius*, but did not include samples from Kumbotos and other organic solvent extracts such as ethyl acetate. In this study, we provide basic information concerning the levels of some mineral constituents of both plants. These minerals are essential to human health. Further, phytochemical screening and analyses of extracts against important pathogen, will indicated concordance with previous works and show results of other solvent extracts not indicated earlier.

MATERIALS AND METHODS

Collection of plant material and preparation

Composite samples of healthy mature pods of *A. nilotica* (Family: Mimosaceae) from an about 8 years old tree, and leaves of *T. dodoneifolius* (Family: Loranthaceae), during the flowering stage respectively, were both collected at the peak of the dry season (April, 2007) at Naibawa in Kumbotos LGA of Kano State, Nigeria. The samples were identified by an expert botanist, Prof. S. Sanusi, Department of Biological sciences, Bayero University Kano. A voucher specimen of the samples with numbers GSU 042 for *A. nilotica* and GSU 103 for *T. dodoneifolius* has been deposited in the herbarium of the Department. Samples were thoroughly washed, shade dried away from sunlight at room temperature. Air-dried samples were ground into fine powder.
Mineral determination

Analyses of mineral constituents (Ca, Cu, Fe, K, Mg, Mn, Na, P and Zn) were carried out on about 2 g of each sample of the pods of *A. nilotica* and the leaves of *T. dodoneifoliu*s. This was ashed in the oven at about 400°C for 2 h. The elements in the ash were quantitatively taken up with 20 ml concentrated HNO₃, and filtered into 50 ml volumetric flasks. The volumes were made up to the mark with distilled water. Minerals were determined by atomic absorption spectrophotometry using Pye Unicam SP9 spectrometer (Pye Unicam Ltd., Cambridge, England). Phosphorus was determined by UV spectrophotometry using Spectronic 20 (Thermo Electron Scientific Instruments LLC, USA) (Ogugbua et al. 1997). The standard calibrations method was used as described by Vogel (2000).

Solvent extractions

The powdered samples (50 g each) of the pods of *A. nilotica* and the leaves of *T. dodoneifolius* were separately charged in thimbles and extracted successively with 400 ml each of 98% ethanol, *n*-hexane and ethyl acetate using a Soxhlet extractor for 48 h. All the extracts were concentrated using a rotary flash evaporator and preserved at 5°C in airtight, well labeled bottles until further use (Raghavendra et al. 2006). All the extracts were subjected to phytochemical analysis and antibacterial activity assay.

Phytochemical screening

Phytochemical analysis of all the evaporated solvent extracts was conducted in accordance with a standard procedure (Harborne 1998). Tests for alkaloids, steroidal, saponins, tannins, flavonoids and terpenoids were carried out in all the fractions.

Antibacterial activity assay

Five bacteria species: *Escherichia coli*, *Staphylococcus aureus*, *Klebsiella aerogenes*, *Psuedomonas aeroginosa* and *Proteus mirabilis* stock cultures were collected from Murtala Mohammed Specialist Hospital Kano, Nigeria. The antibacterial assay was by the cup diffusion method on nutrient agar medium according to the British Society for Antimicrobial Chemotherapy (BSAC 1997). The standard calibrations method was used as described by Vogel (2000).

![Fig. 1 Concentration of elements in *A. nilotica* pods and *T. dodoneifoliu*s leaves.](image)

Data obtained were subjected to statistical analysis using coupled MSExcel-Analyse-it® (2006). Independent Student’s *t*-test at *p*<0.05 was considered significant in results of both mineral contents and antibacterial assay.

RESULTS

Mineral constituents

Fig. 1 shows the results of mineral constituents in the pods of *A. nilotica* and the leaves of *T. dodoneifolius*. Cu and Zn were present in the least concentration in both plants. Ca followed by K showed highest concentrations in both plants. The concentration of P in the pods of *A. nilotica* was highest (12.3 × 10⁵ μg/g) and least (5.6 × 10³ μg/g) in the leaves of *T. dodoneifolius*. In general the concentrations of mineral constituents were higher in the pods of *A. nilotica* than in the leaves of *T. dodoneifolius*.

Phytochemical analysis

Results of phytochemical analysis (Table 1) revealed the presence of steroids, tannins and saponins in both ethanol and ethylacetate extracts respectively, for both *A. nilotica* and *T. dodoneifolius*. Flavonoids were present only in the ethanol extract of *A. nilotica*. The ethanol extract generally presented more phytochemical agents in both plants studied.

<table>
<thead>
<tr>
<th>Table 1 Results of phytochemical analysis of extracts from pods of <em>A. nilotica</em> and leaves of <em>T. dodoneifolius</em>.</th>
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<td>Test for:</td>
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</tr>
<tr>
<td>Alkaloids</td>
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<td>Steroids</td>
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<td>Tannins</td>
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<td>Saponins</td>
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<td>Flavonoids</td>
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<td>Terpenoids</td>
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<td><strong>Key:</strong> Absent (-). Present (+)</td>
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<th>Table 2 Results of antibacterial activity assay of the extracts from pods of <em>A. nilotica</em> and leaves of <em>T. dodoneifolius</em>. (Inhibition zones measured in mm)</th>
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<tbody>
<tr>
<td>Plant</td>
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<tr>
<td><em>A. nilotica</em></td>
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<td><em>T. dodoneifolius</em></td>
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<td>Control</td>
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*Data represented as mean of three trials ± standard error*
Antibacterial activity assay

Table 2 shows the results of the antibacterial activity assay; activity was not recorded for P. aeroginosa for all fractions from both plants. All solvent extracts of T. dodoneifolius showed activity against E. coli and showed the only significant variations for the ethyl acetate extract when compared to the standard Gentamicin control. S. aureus, K. aerogenes and P. mirabilis showed varying activities only for A. nilotica solvent extracts, and only the ethanol extract showed activity against S. aureas, while the n-hexane extract showed no activity against any pathogen. Thus the ethyl acetate extract of A. nilotica showed significant activity against K. aerogenes and P. mirabilis.

DISCUSSION

The mineral composition of the pods of A. nilotica and the leaves of T. dodoneifolius indicated a higher likelihood of mineral enrichment by the use of these plants. The structural and functional roles of mineral composition of some medicinal plants to biosystems have been well established (O’Dell and Campbell 1971; Ogugbua et al. 1997). However the presence of P has been exploited for synthesizing numerous phytochemical compounds in plants (Habibovic et al. 2002). However the results of this work are in accordance with those presented earlier for some medicinal plants of northern Nigerian origin with regards to the accumulation of certain minerals (Fe, K and Mn) in fruity parts of plants than in the stems and leaves (Ogugbua et al. 1997).

Generally, the result of phytochemical analysis revealed a greater likelihood of tannins being the active phytochemical agents in both plants. This may likely be due to the presence of phenolic compounds (Harborne 1998). Terpenoids were only detected in the ethanol and hexane fractions of A. nilotica; this portrays a general trend of the components in medicinal plants, but T. dodoneifolius showed a wide spectrum of antimicrobial activities and shows great similarity with the results of Deeni and Sadiq (2002), in terms of phytochemical and the antibacterial activity, especially against E. coli. These give credence to the ethnomedicinal usage of the plant.

Pseudomonas species, showed resistance to all extracts, but the ethanol and the ethyl acetate fractions of A. nilotica were moderately promising on K. aerogenes and P. mirabilis. The n-hexane fraction of A. nilotica showed no inhibition against all the bacteria investigated in this study. This may be due to very low concentration or absence of active agents in this fraction, which indicates ineffectiveness of n-hexane in the extraction of the requisite inhibitory agents against the bacteria tested. Worthy to note is also the fact that biological actions are primarily due to these components in a very complicated concert of synergistic or antagonistic actions. Mixtures of such chemicals show a broad spectrum of biological effects and pharmacological properties (Albert 1952; Mayo 1994). To a large extent, the phonomological age of the plant, the percentage humidity of the harvested material, place and time of harvest, and the method of extraction are possible sources of variation in the chemical composition, toxicity and bioactivity of the extracts (Felix 1982). It is very difficult though, to make connections between the present findings and the acclaimed efficacy of these plants on the management of tuberculosis. However studies have indicated that certain biflavonoids have inhibitory activity against Mycobacterium tuberculosis (Lin et al. 2001) and other components that act in similar way as the Rifampin and Pyrazinamide, anti- tuberculosis drugs (Sadoff 2006) towards tuberculosis patients utilizing these plants studied in this work.

CONCLUDING REMARKS

Pods of A. nilotica and the leaves of T. dodoneifolius contain sufficient mineral constituents and their extracts indicate a relatively moderate number of phytochemical agents that showed significant activity on some bacteria. Hence their ethno-medical claims in the management of some human diseases are justified. It is suggested that more research be conducted that will further elucidate the effective components and possible mode of actions involved in the use of these plants in ethno-medical practices.

REFERENCES

Ogugbua VO, Akinniyi JA, Ogawuru VC, Abdulrahman F (1997) Element- als Contents of Medicinal Plants: A Monograph, Faculty of Science Mono- graph Series, University of Maiduguri, 1, pp 3-23

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