Determination of Nutritive Value and Mineral Elements of Five-Leaf Chaste Tree (*Vitex negundo* L.) And Malabar Nut (*Adhatoda vasica* Nees)

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Abstract: This study was undertaken to assess the nutritive value and mineral contents from *Vitex negundo* and *Adhatoda vasica*. These two plant species are fairly used as medicine throughout the greater part of India. *Adhatoda vasica* is used to control pain, inflammation and other related diseases. Leaves of *Adhatoda vasica* are used for treatment of cold, cough, chronic bronchitis and asthma. It was also used by traditional midwives at the time of delivery. The leaves of *Adhatoda vasica* are extensively used in indigenous medicines remedies. Both the plants contained important macro and micro elements: K, Ca, Fe, Cu, Zn and Cr. These elements were found in more quantity in *Vitex negundo* than in *Adhatoda vasica*. The leaves of both the plants were analyzed for ash content, moisture, crude fat, crude fibre, crude carbohydrate and crude protein content. The results for percentage of ash content, moisture content, crude fat, crude fiber, carbohydrate and protein were 5.4 ± 0.35, 16.50 ± 1.2, 7 ± 0.7, $28.02 +1.03$, 8.5 ± 0.45, 13.7 ± 1.04 % respectively for *Vitex negundo*; and 5.2 ± 1.23, 15.3 ± 0.5, 1.6 ± 0.3, 6.4 ± 0.45, 16.4 ± 0.8, 6.5 ± 0.3 respectively for *Adhatoda vasica*. The leaves were also assessed for nutritional value. Nutritional value of *Vitex negundo* was 151.80 Cal/100g and that of *Adhatoda vasica* was 106.00 Cal/100.g.

Key words: *Vitex negundo* · *Adhatoda vasica* · Nutritive value · Mineral contents

INTRODUCTION

Plants have great importance due to their nutritive value and continue to be a major source of medicines as they have been found throughout human history [1]. About 30-40% of today's conventional drugs used in the medicinal and curative properties of various plants are employed in herbal supplements, botanicals, nutraceuticals and drugs [2]. Human beings require number of complex organic compounds as added caloric requirements to meet the need for their muscular activities, carbohydrates, fats and proteins, while minerals and vitamins form comparatively a smaller part, plant materials form a major portion of diet; their nutritive value is important [3, 4]. Human body comprises chemical compounds such as water, proteins, fatty acids, nucleic acids and carbohydrates, these in turn consists of elements such as carbon, hydrogen, oxygen, nitrogen and phosphorus and may or may not contain minerals such as zinc, calcium, iron, magnesium etc [5].

The minerals are obtained from the earth's crust. Through the effects of the weather, rocks that contain minerals are ground into smaller particles, which then become part of the soil. The mineral content in the soil is absorbed by growing plants. The plants are consumed by both animals and human beings as food. This mineral becomes part of the food chain. The plants absorb much of the essential elements from the soil in which they grow and serve as indicators of the materialization and are in fact used for this purpose [6]. Heavy metals are the matter of concern in the herbal drugs as certain plants have the tendency to store them from the soil, polluted water and atmosphere [7, 8].

Dental caries is one of the most important problems in public health because of its ubiquitousness in civilized populations. The prevalence of dental caries in industrialized countries like India is on the rise because the treatment is very costly and requires a lot of manpower thus the prevention at the primary level is the solution of choice [9]. Nano gram quantities of chromium

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are required in every insulin dependent system. It has been reported that by acting on ribosome, chromium facilitates the incorporation of insulin stimulated amino acid protein [10]. Insulin dependent diabetics excrete more chromium than the control subjects [11]. Chromium deficiency has also been held responsible for vascular complications associated with diabetes mellitus [11, 12]. Zinc plays a role in the synthesis, storage and secretion of insulin [13, 14]. The high concentration of potassium in the plants could be related to the diuretic action of the drug prepared from the plant materials. The high concentration of Ca and Mg can explain the absence of side effects as regard to stomach lesions [15, 16]. A number of metal complexes and ligands have been shown to be chemically useful as anti-tumour and antiviral agents. In the present study, the inorganic elements (Fe, K, Mg, Ca, Cu, Zn and Cr) of two medicinal plants, *Vitex negundo* and *Adhatoda vasica* were detected. These plants are used for the treatment of various diseases and are useful to the users of herbal medicine.

Abundant research work has been carried out on the organic constituents of the medicinal plants while little attention has been paid on the role of inorganic elements in the medicinal use of these plants [17-21]. A literature survey revealed that trace elements play a significant role in curing various diseases. It has been found that alteration of trace elemental homeostasis in an organism has direct correlation with different pathological conditions [22]. The present investigation is an attempt to gain an insight into the trace elemental composition of Vitex *negundo* and *Adhatoda vasica*.

MATERIALS AND METHODS

***Vitex Negundo*:** *Vitex negundo* Linn. Belongs to family Verbenaceae commonly known as Nirgundi. It is a large, aromatic shrub; with typical five foliate leave pattern found throughout the greater part of India at warmer zones and ascending to an altitude of 1500 m in outer, Western Himalays. The shrub is one of the common plants used in Indian medicines. It has been claimed to possess many medicinal properties. It contains various chemical compounds of various classes such as alkaloids, tannins, flavonoids, carbohydrates and tannins.

***Adhatoda Vasica*:** *Adhatoda vasica* Nees (Acantheceae) commonly known as vasaka distributed throughout India up to an attitude of 1300m. The leaves, flowers, fruit and roots are extensively used for treating cold cough, whooping cough, chronic bronchitis, asthma and as sedative, expectorant and antispasmodic. It was also used by traditional midwives at the time of delivery. The leaves, the roots and flowers of *Adhatoda vasica* are extensively used in indigenous medicine as remedy for cold, cough, bronchitis and asthma. It has been reported that methanolic extracts of plants generally possess terpines and phenolics.

Collection: The leaves of *Vitex negundo* and *Adhatoda vasica* were collected from Ranchi district of Jharkhand state during February, 2013.

Powder Preparation: The plant shoot were washed with deionised water and disinfected with 0.1% $HgCl_2$ solution for 5 min and dried in shade to prepare the sample for mineral analysis, the washed and dried materials were ground to fine powder with mortar and pestle and used for dried ashing [23].

Analysis for K and Ca: For analysis of K and Ca the powdered plant shoot was taken in pre cleaned and constantly weighed silica crucible and heated in muffle furnace at 400°C till there was no evolution of smoke. The crucible was cooled in desiccator at room temperature. The ash totally free from carbon moistened with Concentrated H_2SO_4 and heated on Hot plate till fumes of sulphuric acid get evolved the silica crucible with sulphated ash was again heated at 600°C in muffle furnace till weight of sample was constant (3-4 hrs) one gram sulphated ash were taken in beaker which dissolved in 100 ml 5% concentrated HCl to obtain solution for determination of K and Ca through flame photometry (FPM), standard solution of each mineral was prepared and calibration curve drawn for each element using FPM [4].

Determination of Protein and Nitrogen: Determination of protein and Nitrogen was done using Micro Kjeldahl method, 1 g of sample of each plant taken in a Pyrex digestion tube and 30 ml of concentrated H_2SO_4 carefully added, then 10 g potassium sulphate and 14 g copper sulphate. This mixture was placed on sand both on a low flame just to boil the solution, it was further heated till the solution becomes colorless and clear, allowed to cool, diluted with distilled water and transferred 800 ml Kjeldahl flask, washing the digestion flask. Three or four pieces of granulated zinc and 100 ml of 40% caustic soda were added and the flask was connected with the splash heads of the distillation apparatus. Next 25 ml of 0.1 N sulphuric acid was taken in the receiving flask and distilled; it was

tested for completion of reaction. The flask was then removed and titrated against 0.1 N caustic soda solution using Methyl Red indicator for determination of nitrogen, which in turn give the protein content [24].

Determination of Fe: For determination of Iron 1 g of sample in 125 ml deionised water was taken in conical flask pH adjusted to 2-3 by using Congo red paper. About 5 drops of Variamine blue indicator was added then content was warmed at 400 °C on hot plate and titrated with standard 0.05 M EDTA the initial blue color changes to gray just before the end point and final drop of reagent changes to yellow. Concentration of Iron was calculated by using the formula 1 mol EDTA = 1 mol Iron [25].

Determination of Cu: For determination of copper the titration cell was charged with 1 g. of sample with 10 ml distilled water, 20 ml of acetate buffer (pH 2.2) followed by addition of 120 ml of distilled water. Spectrophotometer adjusted to zero, the solution was then filtered, stored and titrated with standard EDTA. The absorbance was then recorded at every 0.50 ml until the value is about 0.20 and subsequently every 0.20 ml titration continued up to 1.0 ml. The end point observed when readings become constant. The absorbance plotted against volume of titrant added; the inter section of the two straight lines gives concentration of copper in sample which compared with true value [25]

Determination of Cr: For determination of Chromium 0.50g of sample dissolved in 100 ml distilled water, 20 ml of 0.1 M Silver nitrate solution followed by 50 ml of 10% solution of ammonium persulphate were added. The mixture was then boiled for 10 min cooled and diluted to 250ml in graduated flask up to the mark then 50 ml of solution removed and 50 ml 0.1M ammonium iron sulphate solution, 200 ml 1M sulphuric acid and 0.5 ml of N-phenyl-anthranilic acid indicator is added and titrated with standard 0.02 M potassium dichromate solution until the color changes from green to violet red. Ammonium iron sulphate solution standardized against 0.02M potassium dichromate, using N-phenyl-anthranilic acid as indicator. The volume of iron in the solution calculated which was oxidized by the dichromate originating from the chromium salt and from this the percentage of Chromium was calculated [25].

Determination of Zinc: For determination of Zinc 1g of sample in 20 ml deionised water were taken and placed in titration flask and 1ml pure cyclohexylamine was added, -1.4 V Vs. SCE potential applied to deaerate the solution and titrated with standard EDTA using a semi micro burette. From volume of EDTA used concentration of zinc was determined using the relation 1 ml 0.01 M EDTA = 0.6538 mg Zn [25].

Determination of Crude Fat: Crude fat were determined by extracting 1g of moisture free plant material of each plant with petrol in a soxhlet extractor heating the flask on sand bath for about 1 hr. This petroleum extract that contained crude fat, was taken in a pre-weighed beaker (W_1) and petroleum was evaporated. The weight of beaker along with the residual extract (Crude fat, W_2) was taken and crude fat content of the sample was calculated using the formula [26].

$$\% \text{ crude fat} = (W_2 - W_1) \times 100/S$$
(where S is the weight of the sample)

Determination of Crude Fibre: For determination of crude fibre, 2g of moisture and fat free material were treated with 200ml of 1.25% H_2SO_4, after filtration and washing, the residue was treated with 1.25% NaOH, filtered, washed with hot distilled water and then 1% HNO_3 and again washed with hot distilled water. The residue was ignited and the ash weighed. Loss in the weight gives the weight of crude fiber [26].

Determination of Moisture Content: For determination of moisture content plant material is kept in pre weighed watch glass and dried at 150°C over night in an oven. The sample with watch glass is cooled at room temperature in a desiccator before weighing, the weight loss in sample regarded as moisture content.

Determination of Ash Content: For determination of ash content, 5 g of each plant sample weighed and taken in silica crucible and heated first over a low flame till all the material was completely charged, followed by heating in a muffle furnace for about 3-5 hrs at 600°C. Then the sample was cooled in a desiccator and weighed to ensure the completion of ashing; it was again heated in muffle furnace for 1 hour, cooled and weighed. This was repeated consequently till the weight of sample became constant (Ash became white or grayish white). The loss in weight of plant sample gives the ash content [27]. All above procedures were carried out for both plant materials.

Nutritional Value: The nutritional value of both test plants was calculated as per the formula used by Nile and Khobragade [28].

RESULTS AND DISCUSSION

The results of mineral elements of *Vitex negundo* and *Adhatoda vasica* are given in Table 1, while the result of various nutrients is summarized in Figure 1 and the nutritive value is summarized in Table 2.

Results showed that chromium was very low in comparison with other mineral elements in both plants; comparatively higher in *Vitex negundo*. Cr is vital element as it works with insulin to stabilize blood sugar level, help to absorb energy from blood and increase muscle mass by reducing fat mass in human body [29]. Deficiency of Cr results in growth failure, cataract, hyperglycaemia, neuropathy, atherosclerosis and leads to diabetes in humans [30].

Potassium was higher in both plants but contained less Na. Sodium and Potassium take part in ionic balance of the human body and maintains tissue excitability, carry normal muscle contraction and help in formation of gastric juice in stomach [31]. K helps in release of chemicals which acts as nerve impulses, regulate heart rhythms, deficiency causes nervous irritability, mental disorientation, low blood sugar, insomnia and coma [32].

Iron sufficient in both the plants, is involved in making of body tendons and ligaments, certain chemicals of brain are controlled by presence or absence of iron. It is essential for formation of hemoglobin, which carry oxygen around the body [33]. Iron deficiency causes anemia, weakness, depression, poor resistance to infections [34].

Ca is higher in *Vitex negundo* but also sufficient in *Adhatoda vasica*. Calcium play important role in building and maintaining strong bones and teeth and also large part of human blood and extra cellular fluids. It is also necessary for normal functioning of cardiac muscle, blood coagulation, milk clotting and regulation of cell permeability [35]. Calcium deficiency causes rickets, back pain, osteoporosis, indigestion, irritability, premenstrual tension and cramping of uterus [36].

Zinc is again higher in *Vitex negundo* than *Adhatoda vasica*, Cu is an important component of many enzyme systems such as cytochrome oxidase, lysyl oxidase and ceruloplasmin, an iron oxidizing enzyme in blood [37].
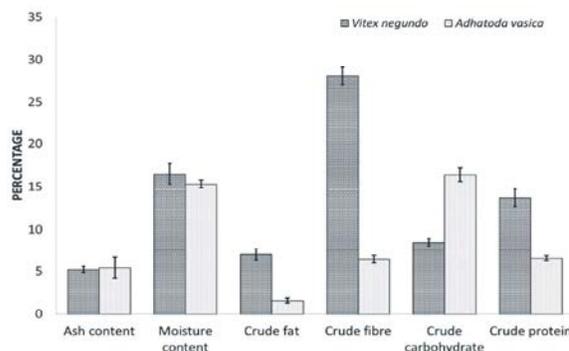

Fig. 1: Percentage of Ash, Moisture Contents, Fibre, Carbohydrate and Protein (N = 3; M ± Sd)

Table 1: Various elemental concentration (ppm) of *Vitex negundo* and *Adhatoda vasica*. (n=3; M ± SD)

Plant	K	Ca	Fe	Cu	Zn	Cr
Vitex negundo	161800 ± 36	143400 ± 23	5330 ± 16	200 ± 9	280 ± 10	120 ± 5
Adhatoda vasica	31190 ± 20.3	68070 ± 35	705 ± 8	64 ± 5	67 ± 6	41 ± 3

Table 2: Nutritive value of *Vitex negundo* and *Adhatoda vasica* (n = 3; M ± SD)

Sl. No.	Plants	Nutritive value (Cal/100g)
1	*Vitex negundo*	151.80 ± 3.33
2	*Adhatoda vasica*	106.00 ± 4.74

Cu deficiency has been associated with cardiac abnormalities in human and animal; causes anaemia and neutropenia [37]. Zn maintain various reaction of body, which help to construct and maintain DNA, required for growth of body tissues, important element of ligaments and tendons [38]. Zn deficiency causes clinical consequences, including growth delay, diarrhoea, pneumonia, disturbed neurophyshological performance and abnormalities of foetal development [38].

Nutritive Value: The nutritive value of both the plants is low, although the nutritive value of *Vitex negundo* is higher than that of *Adhatoda vasica*. The low nutritive value shows that the plants cannot be used as food or fodder, although both the plants have good mineral content, which shows that the plants have good medicinal value and extracts can be used for medicinal formulations.

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