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QUANTTITATIVE ESTIMATION OF PRIMARY AND SECONDARY METABOLITES ON FLOWERS OF CARYOTA URENS.L.

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ABSTRACT: This paper describes the quantitative determination of primary and secondary metabolites present in the flowers of Caryota urens. The primary metabolites are the trace elements such as zinc, copper, boron and molybdenum quantified by AAS (Atomic Absorption Spectrometer) method and minerals such as sodium, potassium and calcium is quantified by flame photometer. Magnesium is quantified by the titrimetric method and nitrogen is quantified by kjeldhal's method. Iron, Phosphorous, Boron, Molybdenum and Sulphur are quantified by Spectrocolorimetric method. The secondary metabolites are quantified by HPLC method (Alkaloids, terpenoids, liginins, tannins, glycosides, serpentines and saponins). The phenols are quantified by Folin's Ciocalteau method and flavonoids by spectrophotometric method. This study indicates that the analysed species is a rich source of flavonoids, alkaloids and tannins etc..,
Key Words: Caryota urens. L. ; Primary and Secondary metabolites

INTRODUCTION

The Caryota urens is a member of the family Arecaceae (Palmae) (C. urens - Toddy fishtail, Jaggery palm, Koonthal panai in Tamil). It is Asian species that grows from India to Burma and on the Island country of Sri Lanka. Caryota urens flowers are long plait bunches hanging down in the tree (Figure 1). In Ayurveda recommends the use of C. urens for seminal weakness and urinary disorders, the juice is applied on the forehead for hemicrania. In traditional medicine porridge prepared from C. urens flower is used to treat gastric ulcer, migraine headaches, snake bite poisoning, as well as rheumatic swellings.Tender flowers are used as a home remedy to promote hair growth. Similarly, the root is used for tooth ailments. Sweet sap from inflorescence can be drunk fresh (toddy) or boiled to produce sugar (jaggery).Toddy can be fermented and distilled to alcohol (arrack) or to vinegar. Palm heart also used locally as flour (destructive), especially for control of diabetes and in auruvedic medicines. Fruit contains calcium oxalate and is not edible. In the present study, we have concentrated on the quantitative determination of primary and secondary metabolites from the flowers of C.urens.

MATERIALS AND METHODS

Plant Collection and Identification

The flowers of C.Urens were collected from the Thanjvur district Tamilnadu in April 2009.The plant was identified and it was authenticated with vouch specimen by Rapinant Herbarium, St.Joseph'College, Trichy,Tamilnadu India.

Quantitative Determination of Moisture and Ash Content

About 5g of fresh flowers were taken in a pre-weighed silica crucible. It was kept in air oven for one hour at 110°C. Then the weight of the dry flowers was found out. From the difference in weight, the amount of water was determined. The ash content was determined by incineration of the dry plant sample in muffle furnace at 400°C. The results were given in Table 1.

Figure.1. Caryota urens flowers

Table-1.Quantitative Estimation of Primary and Secondary Metabolites in flowers of Caryota Urens.

S.No	Name of the parameter	Sample (Caryota urens).
1	Ash (%)	0.78
2	Organic Carbon (%)	2.87
3	Nitrogen (%)	1.20
4	Phosphorous (%)	0.51
5	Potassium (%)	4.56
6	Sodium (%)	0.25
7	Calcium (%)	4.21
8	Magnesium (%)	3.05
9	Sulphur (%)	0.52
10	Zinc(ppm)	2.15
11	Copper(ppm)	1.05
12	Iron(ppm)	45.69
13	Manganese(ppm)	10.23
14	Boron(ppm)	0.06
15	Molybdenum(ppm)	0.09
16	Alkaloids(mg/kg)	0.42
17	Flavonoids(mg/kg)	0.62
18	Tannin(mg/kg)	0.13
19	Lignin(mg/kg)	0.24
20	Glycosides(mg/kg)	0.12
21	Serpentines(mg/kg)	0.06
22	Terpenoids(mg/kg)	0.05
23	Saponins(mg/kg)	0.06
24	Phenols(mg/kg)	0.17
Vitamins(mg/kg)		
25	A	143
26	B	365
27	C	66.6

Quantitative Determination of Primary Metabolites

Preparation of Sample solution

About 0.5g of ash was digested with con. HCl dissolved in water and filtered. The filtrate was made up to 100ml in a standard flask. This made up solution was used for further analysis.

Standard Graph Method

The standard Sodium ion solution was prepared (0.586g NaC1 in 100mL). From this NaCl solution, about nine differently concentrated solutions (1.0, 1.5, 2.0,…, 5.0 ml) were prepared. These solutions were taken for flame photometric studies (Systronics mediflame 127). A standard graph was plotted by taking concentration of sodium on the X-axis and flame photometric study and its reading is fitted with the standard graph. The percentage of sodium in plant sample was determined.

The concentration of potassium and calcium were also calculated by the same procedure. The standard potassium (0.750g KCl in 100ml) and calcium solutions (0.55g$CaCl_2$ in 100ml) were prepared.

The determination of iron, boron, phosphorous and molybdenum was done spectrocolorimetrically by standard graph method.

The standard solutions of iron of different concentrations were prepared from the bulk solution (2.44g of FAS in 250ml). Each of the iron solution was treated with 4N HNO_3 and NH_4CNS. The percentage transmittance was measured at 470nm.

The standard solutions of boron of different concentrations were prepared from the bulk solution (0.286g of boric acid in 1000ml of water). Each of the boron solution was mixed with curcumin-oxalic acid solution and ethyl alcohol. It was taken for the measurement of percentage transmittance.

The nine different standard solutions of phosphorous were prepared from the bulk solution (0.1g of KH_2PO_4 in 250ml). Each of the phosphorus solution was treated with ammonium molybdate and ammonium vanadate. The percentage transmittance was measured.

The standard solutions of molybdenum of different concentrations were prepared from the bulk solution (01 50g of reagent grade molybdenum trioxide in 10ml of NaOH, made slightly acid with HCL and made up 1 liter with water) Each of the standard solution was treated with $FeCl_3$, $NaNO_3$, NH_4SCN and SnCl2.The percentage transmittance was measured spectrocolorimetrically.

Sulphur was also determined by Spectrocolorimeter method unlike other method here, the sulphur in plant sample was converted into sulphate using $BaCl_2$. The concentration of Copper, Manganese and Zinc in plants sample was determined by AAS (Atomic Absorption Spectrometer). A standard solution of Copper was prepared by dissolving 3.929g of $CuSo_4.5H_2O$ in 1000ml of water and 10ml of the solution was diluted to 100ml with water. The determination of Cu as also of Mn and Zn was done by using ASS with the following specifications for mono element hollow cathode lamp. The exact specifications should be as per the particular instrument used.

The standard solution of Mangesium was prepared by dissolving 3.076g of $MgSO_4$ in 1L of deionized water. 10ml of this solution was diluted to 100ml (100ppm of Mn). This solution was used as standard solution. The standard solution of Zn was also prepared by same method (4.398g of $ZnSO_47.H_2O$).

The magnesium was estimated by titrimetric method using standard EDTA with Erio T indicator at pH10.

Vitamin C was quantified by titrimetric method using idodine solution with Starch indicator. Vitamin B was determined spectocolorimetrically with the reagent ferric sulphate and KCNS. Vitamin A was estimated spectrocolorimetrically using acidic antimony chloride reagent by the standard graph method. Standard vitamin – A solutions were prepared in different concentration.

Quantitative Determination of Secondary Metabolites

The total flavonoid and phenolic contents were quantified by Spectrophotometeric method using Folin's Ciocalteaus reagent.

The other secondary metabolites such as alkaloids, tannins, lignin, glycosides, serpentines, terpenoids and sponins quantified by HPLC method and Cl8 general purpose column using the solvent the mobile phase consisted of solvent A (methanol) and solvent B (0.5% (v/v) ortho-phosphoric acid in water). The data were interpreted by the Millenium Chromatography Manager V4.0 Software.

RESULTS AND DISCUSSION

The results of quantitative estimation of primary and secondary metabolites are given in Table l. The highest percentage of iron and mangnesium was noticed in the flowers of C.Urens. The most abundant mineral in the sample was magnesium and then calcium, potassium, zinc and copper. The ash content of 0.78% was indicative of fact that the dried plant had been completely converted into ash. The organic carbon in plant sample was 2.87%. Lesser amounts of nitrogen, phosphorous, sodium, sulphur, zinc and copper were present in the sample. Boron and molybdenum were in trace amounts. The vitamin-C is found to be less in amount than that of vitamin-A.

The flavonoid and alkaloids contents were higher in amount than the other secondary metabolites in the flowers of C.Urens. This has given rise to its therapeutic properties. The significant amount of lignin has given toughness of the flower. An observable amount of phenolic compounds has given the plant greater resistivity against phytopathogenic micro organisms. The quantative estimation of phenols and flavanoids in the flowers of C.Urens gives an insight and understanding certain basis pattern of growth and metabolism. At the same time phenols and flavonoids can be used as chemical markers in taxonomic studies.

Conclusion

The results from this study indicate that analyzed species are a rich source of primary metabolites like iron and manganese and the secondary metabolites like flavonoids, alkaloids and lignins could be of equal value to those which have been characterized as medicinal properties.

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