

SPECTROPHOTOMETRIC ESTIMATION OF TOTAL POLYSACCHARIDES IN *KALANCHOE PINNATUM* AND *KALANCHOE CRENATA*

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ABSTRACT

Kalanchoe pinnatum and *Kalanchoe crenata* belong to the plant family Crassulaceae. They show close proximity in usage, habitat, preparation and identification. The present paper deals with the Spectrophotometric estimation of Total Polysaccharide Content of leaves & stems of *Kalanchoe pinnatum* & *Kalanchoe crenata* and evaluated for Total Polysaccharide Content using Phenol – Sulphuric Acid method. The Total Polysaccharide Content of *K. pinnatum* was found to be 2.21, 1.49 w/w% & of *K. Crenata* was found to be 2.04 & 1.35 w/w% in leaves & stems respectively.

Keywords: *K.pinnata*, *K.crenata*, Polysaccharide, Crassulaceae.

INTRODUCTION

The external morphological features of *K. crenata* resemble that of *K. pinnatum*. An inexperienced taxonomist and even unseasoned traditional practitioner can readily confuse the two plants. This is attested to by the same local name that is being used by Yorubas in Southwest Nigeria. Some People refer to both plants as "Odundun". Ethnobotanically, most often they are prepared and administered the same way. *K. pinnatum* is used in ethnomedicine generally for the treatment of ear-ache, cough, diarrhoea, dysentery, abscesses, ulcers, insect bites, heart-troubles, epilepsy, arthritis, dysmenorrhea and whitlow [1]. In southern Nigeria, it is used to facilitate the dropping and healing placenta wound of newly born babies. The plant leaf is mildly exposed to heat and the juice is squeezed out and applied as poultice to the baby's placenta on daily basis. Also, the crushed leaves as well as the extracted juice are mixed with shear butter or palm oil and rubbed on abscesses or other swellings. This is also applied on ulcers, burns and on the bodies of young children when they are ill. The leaves of this plant contain bryophyllin, potassium, malate, ascorbic, malic, and citric acids [2]. The plant is rich in both macro and micro elements, vitamins, calcium, phosphorus, ascorbic acid, inulin [3] and other compounds like saponin, flavonoids, anthraquinones, xanthenes, bryophyllin A and B [4]. Anti-inflammatory, hypoglycaemic, anti diabetic and anticancer properties have been reported [1].

The external applications of *K. crenata* are the same as those of *K. pinnatum*. The juice obtained by squeezing the leaves that have been passed over fire slightly, is most commonly used for the treatment of headache, general debility, dysentery, smallpox and convulsion. One or two drops of the leaf juice is dropped into the ear for earache. A poultice of the leaves is applied over wounds and sores. The leaves can be boiled in water and the extract is given as a sedative for asthma and palpitation. Also the leave juice mixed with salt and honey is a remedy for chronic cough. The extract of dried leaves is applied to septic wound [5].

In East Africa, the slightly heated leaves (heated over fire) are rubbed over the body as treatment for stiff joint and rheumatism [1]. Other parts of the plant especially the root is prescribed for gonorrhoea, vermifuge and abortion [5]. Alkaloids and saponins are present in the aqueous and alcoholic extracts of leaves and lectins in the juice from the fresh leaves [6]. The green callus of the plant contains malic acid, quinones and tocopherol [2,5]. Other works have also shown that this plant possesses analgesic, anticonvulsion, antiinflammatory, antiarthritic and antispasmodic properties [7]. The conventional method to extract plant materials is to use methanol, ethanol, acetone and so on as extracting solvents, but the ethnobotanical approach like the use of Palmwine, "Omidun/ekan-ogi" (the water derived from three days fermented milled maize), local gin as extracting solvent and ways in which they are prepared

locally, has received less attention. The type of solvents and methods of preparation affect antimicrobial activity of plants [8,9]. On the basis of this background, in-vitro antimicrobial activities of the extracts *K. pinnatum* and *K. crenata* from various solvents were tested against clinically important pathogens.

MATERIALS AND METHODS

Collection of plant material

The leaves of *K. pinnatum* & *K. crenata* were collected from Herbal Garden of Chandigarh College Of Pharmacy Landran, Mohali (Punjab) in July 2011, cleaned and dried at room temperature in shade and away from direct sunlight. The plants were authenticated by Dr. H. B. Singh Chief Scientist at NISCAIR (New Delhi) by comparing morphological features.

Instrumentation

Shimadzu UV –VIS Spectrophotometer was employed for all spectroscopic measurements using a pair of matched quartz cells.

Methodology

The plant material collected & dried under shade at room temperature. Then plant material i.e leaves & stems were subjected to grinding in mixer grinder. Then passed through 120 mesh size to remove coarse powder and fine powder was used for estimation of polysaccharide content in plant material.

Preparation of blank solution

To 1ml of distilled water added 1ml of 5% phenol followed by 5ml of concentrated H₂SO₄

Preparation of standard solution

A stock solution 100µg/ml of glucose was prepared in distilled water. Aliquots were taken from

this solution to obtain sugar concentrations 60-90µg/ml. 1ml of 5% phenol solution was added to 1 ml of sugar solution followed by 5ml of concentrated H₂SO₄. The absorbance was measured after 10 minutes at 488nm against blank.

Estimation of Total Polysaccharide Content in *K. pinnatum* & *K. crenata*

Dissolve about 10mg of powder drug in 100ml of distilled in water. From this use 1ml for sugar analysis to estimate the polysaccharide content in *K. pinnatum* & *K. crenata*, add 1ml of 5% phenol to the 1ml of sample solution, and then add 5ml of concentrated H₂SO₄ and measure the absorbance after 10 minutes at 488nm against blank. Then compare it with standard solution of glucose [10]. The experiment was carried out in triplicate (i.e. Test-1, Test-2 & Test-3) [11].

RESULT & DISCUSSION

The calibration curve for different concentrations of glucose is represented in Figure -1. Using the proposed method, the calibration curve was found to be linear in the range of 10-80µg/ml. A correlation coefficient of 0.996 indicates good linearity between the

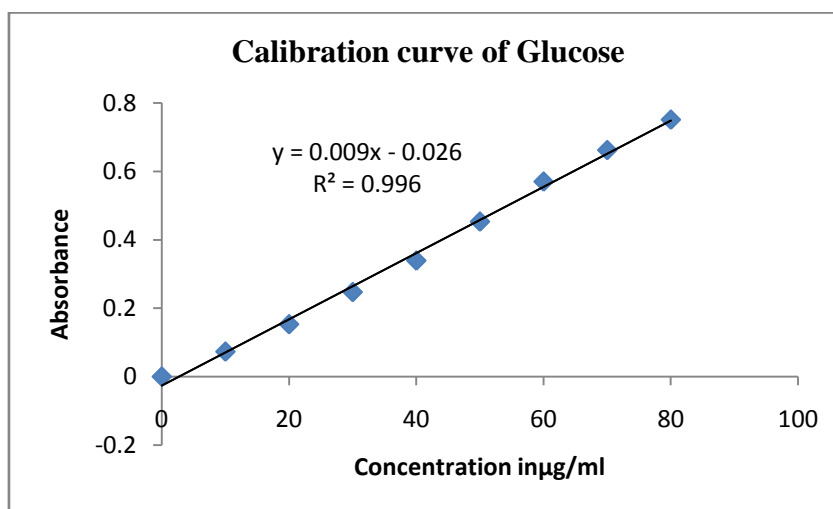
concentration and absorbance. The % Relative Standard Deviation (% RSD) indicates that the used method is precise & accurate. The total polysaccharide content of *K. pinnatum* & *K. crenata* was calculated using regression equation obtained from the calibration curve (Table 2).

Table 1: Absorbance for Total Polysaccharide Content

S. No.		Absorbance of Test			Mean	% RSD*
		Test-1	Test-2	Test-3		
1.	$K_1(L)$	0.262	0.265	0.264	0.264	0.59
2.	$K_1(S)$	0.156	0.154	0.157	0.155	0.48
3.	$K_2(L)$	0.236	0.238	0.241	0.238	0.63
4.	$K_2(S)$	0.138	0.135	0.137	0.137	0.42

*Relative Standard Deviation

K_1 -*Kalanchoe pinnatum*, K_2 -*Kalanchoe crenata* (L-Leaves, S-Stem)

**Fig. 1: Calibration curve of Glucose****Table 2: Total Polysaccharide Contents of *K. pinnatum* & *K. crenata* (Leaves & Stems)**

Powdered extract	Total polysaccharide content (%W/W)
<i>K. pinnatum</i> Leaves	2.21
<i>K. pinnatum</i> Stems	1.49
<i>K. crenata</i> Leaves	2.04
<i>K. crenata</i> Stems	1.35

CONCLUSION

The use of herbal plants is increased in development of various pharmaceutical dosage forms because they are economical, readily available, non-toxic, and capable of chemical modifications. Phenol-sulphuric acid technique is one of the simple, rapid, precise and accurate spectroscopic technique for the determination of total polysaccharides in *K. pinnatum* & *K. crenata*.

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