Available online at http://www.journalcra.com

International Journal of Current Research
Vol.3, Issue, 4, pp.210-212, April, 2011

INTERNATIONAL JOURNAL OF CURRENT RESEARCH

ISSN: 0975-833X

RESEARCH ARTICLE

ISOLATION AND PHYSICOCHEMICAL CHARACTERIZATION OF *Hibiscus rosa-sinensis* LEAVES MUCILAGE

[1]Hindustan Abdul Ahad*., [2]Yesupadam, P., [1]Ramyasree, P., [1]Suma Padmaja, B., [1]Sravanthi, M and [1]Guru Prakash, P.

[1]Department of Pharmaceutics, College of Pharmacy, Sri Krishnadevaraya University, Anantapur- 515003, Andhra Pradesh, India, Tel.: +91 8554 255890, Fax.: +918554255244
[2]Department of Pharmacognosy, Balaji College of pharmacy, Anantapur- 515003, Andhra Pradesh, India

ARTICLE INFO

Article History:
Received 10th January, 2011
Received in revised form
14th February, 2011
Accepted 11th March, 2011
Published online 27th April 2011

Key Words:
Hibiscus rosa-sinensis;
Mucilage;
Isolation;
Characterization.

ABSTRACT

Hibiscus rosa-sinensis leaves are rich in mucilage. But there are no reports on isolation and characterization of *Hibiscus rosa-sinensis* leaves mucilage. Hence, the present study was designed to isolate, purify and characterization of *Hibiscus rosa-sinensis* leaves mucilage. *Hibiscus rosa-sinensis* leaves mucilage was extracted, purified and identified by official methods. The isolated mucilage was characterized for physical, chemical and flow properties. The mucilage was further characterized by Fourier Transform Infrared Spectroscopy. *Hibiscus rosa-sinensis* leaves gave sufficient quantity of mucilage and it has good physical, chemical and flow properties. The Fourier Transform Infrared Spectroscopic study revealed that the mucilage has characteristic peaks. It was concluded that the *Hibiscus rosa-sinensis* leaves mucilage can be used as matrix forming material in pharmaceutical dosage forms.

INTRODUCTION

Hibiscus rosa-sinensis, (Malvaceae family) commonly known as China rose is popular landscape shrub, creates a bold effect with its medium-textured, glossy dark green leaves with 4-6 inch wide and up to 8 inch long, showy flowers, produced throughout the year and grows upto 7-12 feet (Lee, 2007). These leaves were already proved its medicinal value as hypoglycemic (Fahmi, 2011) and other actions were under research. Presently the use of natural gums and mucilages is gaining importance in pharmaceutical formulations as an excipient in dosage forms. Synthetic polymers have certain drawbacks viz., high cost, non-renewable sources, side effects, toxicity, causing environmental pollution during their synthesis, non-biodegradable (where as biodegradable synthetic polymers are costlier) and less patient compliance (Donaruma, 1974; Breslow, 1976). While natural plant based materials are economical, devoid of side effects, biocompatible, biodegradable, renewable source, environmental-friendly processing and better patient compliance (Volfova, 1992). Mucilages are polysaccharide complexes formed from sugar and uronic acid units. Mucilages form slimy masses in water, are typically heterogeneous in composition. Upon hydrolysis mucilages gives Arabinose, Galactose, Glucose, Mannose, Xylose and various Uronic acids (Kulkarni, 2002). The prospects of natural polymers are brighter but even here extensive testing will be required. In present study the leaves of *Hibiscus rosa-sinensis* were selected for the isolation and purification of mucilage. However there are no reports on isolation, purification and characterization of *Hibiscus rosa-sinensis* leaves mucilage. Hence, the present study was planned to isolate and characterize mucilage of *Hibiscus rosa-sinensis* leaves. The data so obtained will be a standardizing parameter for future research work.

***Corresponding author:** abdulhindustan@rediffmail.com

MATERIALS AND METHODS

Materials

The fresh *Hibiscus rosa-sinensis* leaves were collected from plants growing in and around Anantapur, India. The plant and leaves were authenticated at the Botany Department, Sri Krishnadevaraya University, Anantapur, India. Ethanol (95%), Acetone, trichloro acetic acid, sodium hydroxide and diethyl ether were procured from SD Fine chemicals (Mumbai, India). All other chemicals used were of AR grade and double distilled water was used throughout the experiments.

Extraction of mucilage

The fresh *Hibiscus rosa-sinensis* leaves were collected and washed with water. The leaves were opened and seeds were removed. The leaves were crushed and soaked in water for 5–6 h, boiled for 30 min and left to stand for 1 h to allow complete release of the mucilage into the water. The mucilage was extracted using a multi-layer muslin cloth bag to remove the marc from the solution. Acetone (in the volumes of three

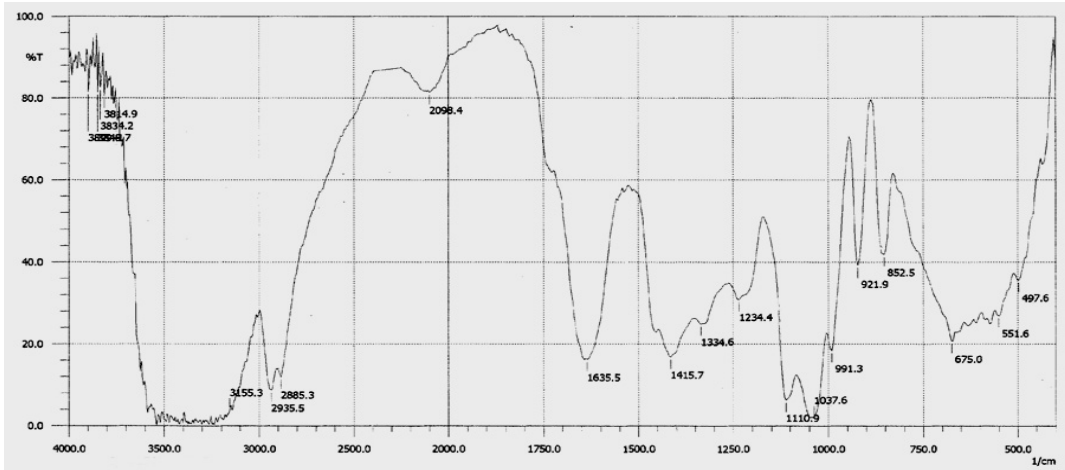

Fig. 1. FTIR spectrum of *Hibiscus rosa-sinensis leaves* mucilage

Table 1. Taxonomical Classification of *Hibiscus rosa-sinensis*

Kingdom	Plantae
Order	Malvales
Family	Malvaceae
Genus	Hibiscus

Table 2. Physical characterization of *Hibiscus rosa-sinensis* leaves mucilage

Physical Properties	Observation
Appearance	Greenish powder
Odour	Characteristic
Solubility	Slowly soluble in water produces hage viscous solution
*Percent yield (g /kg)	28 ±2.457
*Average particle size (μm)	159.32±9.543
*Weight loss on drying (mg)	2.58±0.159
*Swelling Index (%)	47.0±2.575
*Charring (^{0}C)	152±6.851
*Density of liquid (1.0% w/v)	1.058±0.018
*pH	7.1±0.111
*Microbial count (cfu/g)	Bacteria: 6 ; Fungi: 2

Cfu = Colony forming units; *= experiments were conducted in triplicates (n=3)

Table 3. Chemical characterization of *Hibiscus rosa-sinensis* leaves mucilage

Chemical properties	Observation
Mounted in 96% ethanol	Transparent angular masses
Mounted in ruthenium red	Particles stained red
Mounted in Iodine solution	Particles stained blue
Mollish test (for Carbohydrates)	+ve
Ferric chloride test (for Tannins)	-ve
Silver-nitrate test (for chlorides)	-ve
Barium chloride test (for Sulphates)	-ve
Test for Uronic acid	+ve
Test for foreign matter (%)	NMT 0.1
Test for heavy metal (lead)	20 ppm
Test for Arsenic	<1 ppm

NMT = Not more than

Table 4. Flow properties of *Hibiscus rosa-sinensis* leave mucilage

Flow properties	Observation
Angle of repose (θ°)	25.77±1.68
Loose Bulk density (g/cm^3)	0.53±0.05
Tapped bulk density(g/cm^3)	0.67±0.05
Carr's Index (%)	20.8±0.04
Hausner's ratio	1.26±0.03

Number of trials (n)=5

Table 5. FTIR spectral data of *Hibiscus rosa-sinensis leaves* mucilage

Absorption Peak value	Absorption range	Specific type of bond
3155.3	2400–3200	N—H bond (ammonium ions)
2935.5	2925	C—H bond (alkyl, methyl)
2885.3	2400–3200	N—H bond (ammonium ions)
2098.4	2140–1990	C—N bond (any)
1635.5	1615–1700	C═N bond (any)
1635.5	1560–1640	N—H (primary amine)
1234.4	1220–1260	C—O bond (ethers, aromatic)
1110.9	~1100	C—O bond (alcohols, secondary)
991.3	990	C—H bond (vinyl, mono substituted alkenes)
852.5	800–860	C—H bond (aromatic, para-di substituted Benzene)

times to the volume of filtrate) was added to precipitate the mucilage. The precipitated mucilage was separated, dried in an oven at 40°C, collected, ground, passed through a # 80 sieve and stored in desiccator at 30°C & 45% relative humidity till use (Ahad *et al.,* 2010).

Purification of the Mucilage

The crude mucilage (1%) was homogenized (Potter S homogenizer, Sartorius AG, Germany) with cold dilute trichloro acetic acid solution (5%). The solution was centrifuged (3500 rpm for 20 min), neutralized with sodium hydroxide by drop wise addition and then dialyzed for 30 h against distilled water. The mucilage was precipitated with ethanol (in the quantities of three times the volumes) and washed successively with ethanol, acetone and diethyl ether. The mucilage so obtained was dried under vacuum (less than 1 Torr at 25°C for 12 h). The so obtained mucilage was passed through a # 80 sieve and stored in desiccator at 30°C & 45% relative humidity till use (Mark, 1917).

Characterization of Mucilage

Physical characterization: The collected mucilage was evaluated for physical characteristics (Mark, 1917) viz., Appearance, Odour, Solubility, percentage yield, average particle size, swelling ratio, weight loss on drying, pH, Charring, density and bio burden. All these values were tested in triplicate.

Chemical characterization: The extracted mucilage was tested for chemical characteristics for identification, test for Carbohydrate, test for Tannins, test for chloride, test for sulphate, and test for Uronic acid. The mucilage was also tested for unwanted chemicals (Mark, 1917) viz., foreign matter, heavy metal and Arsenic.

Flow properties: The dried *Hibiscus rosa-sinensis* leaves mucilage was tested for the flow properties viz., Angle of repose, Bulk densities, compressibility index and Hausner's ratio. All these evaluations were carried out as per procedures described in official books (Martin, 1991; Aulton, 1988). All these experiments were conducted for five times.

Fourier Transform Infrared (FTIR) Spectroscopy: FTIR spectrums of dried mucilage were recorded on samples prepared in potassium bromide disks using FTIR spectrophotometer (Shimadzu 1601 PC, Tokyo, Japan). Samples were prepared in KBr disks. The scanning range was 500 to 4000 cm^{-1}.

RESULTS AND DISCUSSION

Physical characterization: The extracted mucilage was greenish in colour with a characteristic odour and soluble in water produces huge viscous solution. The leaves gave 28 ±2.457 g of yield per kg. The Average particle size of dried mucilage was 159.32±9.543 µm. The weight loss on drying was 2.58±0.159 and the percentage of swelling was 47.0±2.575%. The dried mucilage was melted and charred at 152±6.851^{0}C. The density of 1.0% w/v solution was 1.058±0.018 and a pH of 7.1±0.111. The mucilage has very negligible bio burden. All these values were shown in Table 2.

Chemical characterization: The mucilage gave positive test for carbohydrates and uronic acid (common for all mucilages) and negative test for tannins, chlorides and sulphates. The amount of foreign matter was negligible. The heavy metal concentration was also found to be within the limits. All these values were shown in Table 3.

Flow properties: The dried *Hibiscus rosa-sinensis* leaves mucilage has excellent flow properties as the Angle of repose was 25.77±1.68^0. The Bulk density values were considered for calculating compressibility index and Hausner's ratio, which were 20.8±0.04% and 1.26±0.03 respectively. All these values were shown in Table 4.

FTIR spectrum: The FTIR spectrum of *Hibiscus rosa-sinensis* leaves mucilage showed sharp and characteristic peaks at 3155.3, 2935.5, 2885.3, 2098.4, 1635.5, 1234.4, 1110.9, 991.3 and 852.5cm^{-1}. The FTIR spectral values and probable bonds present were represented in Table 5 and shown in Fig. 1.

Conclusion

This study revealed that *Hibiscus rosa-sinensis* leaves mucilage has good physicochemical characteristics with good flow properties. The mucilage can be used as binder in tablet formulations and as matrix forming material in pharmaceutical dosage forms.

REFERENCES

Aulton M.E., 1988. Pharmaceutics -The Science of Dosage Forms Design, 2nd ed. London: Churchill Livingstone.pp.600.

Breslow, D. S, 1976, Biologically active synthetic polymers. *Pure Appl. Chem,* 46, 103–113.

Donaruma, L. G, 1974. Synthetic biologically active polymers. *Progr. Polym. Sci.,* 4, 1–25.

Fahmi SM, prakash RN, Najma HM, Selvaraj S, 2011. Anti-diabetic properties of *Hibiscus rosa sinensis* L. leaf extract fractions on non-obese diabetic mouse. *Indian Journal of experimental Biology,* 49 (1), 24-29

Hindustan Abdul Ahad, Chitta Suresh Kumar, Pilli Yesupadam, Sandhya Rani P, Chandra Sekhar A, Sivaramakrishna GV, 2010. Fabrication and Characterization of Diclofenac Sodium Hibiscus rosa-sinensis leaves Mucilage Sustained Release Matrix Tablets. *Der Pharmacia Lettre*, 2 (1), 452-456.

Kulkarni. G.T, Gowthamarajan K, Rao B.G and Suesh B., 2002. Evaluation of binding properties of Plantago ovata and Trigonella foenum graecum mucilages. *Indian drugs*, 39(8), 422-425.

Lee, David Webster, 2007. Nature's Palette: the Science of Plant Color. University of Chicago Press. pp. 183

Mark L. Woolfe,a Martin F. Chaplid and Gifty Otchere, 1917. Studies on the Mucilages Extracted from Okra Fruits and Baobab Leaves. *J. Sci. Fd Agric.,* 28, 519-529

Martin Alfred, 1991. Physical Pharmacy, 4th ed. Maryland, USA: Lippincott Williams & Wilkins, pp. 423.

Volfova I, 1992. Biocompatibility of biopolymers. J. Bioact. Biocompat. *Polymers*, 7, 175–190.