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EXTRACTION AND CHARACTERIZATION OF MUCILAGE FROM CROTALARIA SENEGALENSIS LINN (FAMILY – FABACEAE)

P. O. Onah^{1*} and M. Shok²

¹Department of Pharmacognosy, University of Jos, Jos. Nigeria ²Department of Pharmacognosy and Drug Development;Ahmadu Bello University, Zaria. Nigeria.

Abstract

Mucilages from the leaves, stem and root of *Crotalaria senegalensis* Linn. (Fam. Fabaceae) has been extracted using both the cold and hot extraction methods. The results obtained showed that the hot extraction method (HEM) is a better extraction method than the cold extraction method (CEM) as the yields were:- 10.2%; 5.9% and 1.2% w/w (HEM) as opposed to: 8.0%; 3.7% and 1.0% w/w (CEM) for the leaves, stem and root respectively. Characterization of the mucilage showed that it is composed of galacturonic acid, fructose, arabinose, galactose and xylose. The elemental analysis indicated the presence of magnesium.

Keywords: Crotalaria senegalensis; Mucilage; Extraction; Characterization.

INTRODUCTION

The plant, *Crotalaria senegalensis* L., a herb belonging to the family Fabaceae, is widely distributed in tropical countries. In Nigeria, the plant is used as fodder for livestock. The present investigation was motivated by the observation that the plant contains a lot of mucilage

(Shok, 1999). Mucilage containing plants have been and still are the subject of vast investigations because of their importance in the pharmaceutical, textile and other industries (Ibrahim, 1990).

Chemically mucilage is a complex polysaccharide with colloidal properties, it forms a slimy mass in cold water but dissolves easily in hot water. It is considered as calcium, potassium and magnesium salts of polyuronides i.e. polysaccharides containing one or more uronic acids in its molecular structure. It absorbs water readily to form a viscous mass however, when dry, it is hard and horny (Wallis, 1967).

It usually occurs as an amorphous mass that is insoluble in alcohol and other organic solvents.

MATERIALS AND METHODS

Plant collection and identification:

The leaves, stem and roots of *C. senegalensis* were collected in July, 2000 from a farmland behind Government College, Samaru-Zaria, Kaduna state, Nigeria.

Identification and authentification were performed by Mr. A. Gallah, Department of Biological Sciences, Ahmadu Bello University, Zaria, Nigeria the plant

^{*} Corresponding author.

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materials were separately dried in the open for two weeks and subsequently reduced to coarse powder using a mortar and pestle. The powders were stored in airtight containers until required.

Extraction of Mucilage.

The mucilage from the three plant parts was each extracted using the hot and cold extraction methods respectively as outlined by Karawya *et al*, (1971).

i) Hot extraction method (HEM)

10g of each powdered plant part was extracted with 250ml of hot water for six hours with continuous agitation and filtered through a Muslin cloth. The marc from the leaf and stem powders was extracted with an additional 250ml of hot water $(100^{\circ}c)$ and again filtered. The filtrate was treated with four times it's volume of 95% ethanol. The colloidal mass was collected and washed with some acetone. The acetone was decanted and the mucilage obtained was labeled as M_R (mucilage from leaves), M_S (mucilage from the root).

ii) Cold Extraction Method (CEM):-

The above procedure was repeated using cold water (room temperature)

iii) Percentage yield :-

The percentage yield of the mucilage for each procedure was calculated with reference to the original weight of the sample used (10g) as follows:-

Percentage yield of mucilage

 $= \frac{\text{Amount of mucilage obtained}}{\text{weight of plant sample (10g).}} \times 100$

Characterization of the mucilage-

i) Acid hydrolysis of the extracted mucilage:-2ml of dilute sulphuric acid was added to 0.1g of the mucilage and boiled over a water- bath for about 20 minutes to achieve hydrolysis. The aqueous solution was allowed to cool, neutralized with 1ml of barium carbonate solution and filtered (Harborne, 1984). The filtrate was concentrated over a water-bath. The residue was made up to 1ml with 10% isopropanol.

ii) Analysis of sugar:-

Sugar analysis of the hydrolysate was done by means of paper chromatography on Whatman No. 1 paper using authentic samples of maltose, arabinose, galactose, xylose sucrose, glucose and galacturonic acid as reference compounds.

Two solvent systems were used:-

- I. Chloroform : Ethanol (70:30).
- II. Ethylacetate: Acetic acid: Methanol : Water (60:15:15:10).

Detection of spots was performed by means of anisaldehyde -sulphuric acid and Aniline hydrogen phthalate respectively.

III). Elemental analysis of the mucilage

a) Calibration curve:-

With the use of the Atomic Absorption spectrophotometer 969 Unicam AAS, the elemental analysis of 1g of the mucilage was done after it had been dissolved in 5ml of hot water $(100 \ ^{0}C)$ and 10ml of concentrated nitric acid.

Using the spectrophotometer and the characteristic wavelength for the elementspotassium, magnesium and calcium, various concentrations ranging from 0-80/100ml were used to obtain the calibration curve for each element. Thereafter, the mucilage sample solution was introduced into the spectrophotometer with the calibration curve, it became possible to extrapolate the concentration of the various elemental reference elements, present in the mucilage.

RESULTS AND DISCUSSION

Extraction Of Mucilage:

The percentage yield of mucilage from the three plant parts using either the CEM or HEM procedure is presented in Table 1. The mucilage yield from the powdered leaves was the greatest being 8.0% w/w (CEM), and 10.2% w/w (HEM). The percentage yield of the mucilage from the three plant parts using the two extraction methods was in this order:- % yield from leaves> % yield from stem> % yield from root.

The HEM yielded more mucilage as compared to what was obtained with the CEM. This is in agreement with the fact that mucilage is more soluble in hot water than cold water (Wallis, 1967).

Characterization of Mucilage

The chromatographic analysis of the hydrolyzed mucilage from *C. senegalensis* revealed the following:

Solvent system II gave better separation than solvent system I.

On drying the developed chromatogram at 105^{0} c for 2 minutes and observing under U.V light, the sugar spots were seen to fluoresce.

The R_F values for glucose and galactose were found to be the same for solvent II that is 0.12; 0.24 and 0.21 respectively for solvent system 1, spots from

the sample were found to correspond with these R_F values. In all the R_F values for the spots observed for the sample, they corresponded with those of arabinose galacturonic acid.

After spraying with the detection reagent the colours of the spots for glucose and galactose were brown; arabinose reddish –brown and galacturonic acid- orange – yellow.

Elemental analysis

The result of the quantitative analysis of the element in the mucilage from the leaves of *C. senegalensis* using the atomic absorption spectrophometer is given in Table 2;

Magnesium had the highest concentration in the mucilage. than the other two elements, potassium and calcium.

This finding is in agreement with the fact that mucilage is chemically defined as calcium, potassium and magnesium salts of polyuronides. (Balbaa, 1976).

Table 1: Percentage- Yield of Mucilage using CEM or HEM procedure

	Yield of Mucilage (%	
Samples	CEM	HEM
M_L	8.0	10.2
M _S	3.7	5.9
M _R	1.0	1.2
T T 1 .1	0.1	

Values are the average of three determinations.

Key: CEM = Cold Extraction Method HEM = Hot Extraction Method M_L , M_S M_R = Mucilage from the leaves, stem and root respectively

Table 2: Elemental Analysis Results		
Element	Concentration (mg/ L)	
Calcium	247.3	
Magnesium	298.5	
Potassium	198.4	

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