World Journal of Pharmaceutical Sciences
ISSN (Print): 2321-3310; ISSN (Online): 2321-3086
Available online at: http://www.wjpsonline.org/
Original Article

Some physicochemical characteristics of *heinsia crinita* mucilage

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Received: 10-05-2019 / Revised Accepted: 29-06-2019 / Published: 01-07-2019

ABSTRACT

This study was undertaken to characterize mucilage from the leaves of Heinsia crinita. The dried mucilage was studied for its organoleptic properties, some physical properties such as swelling index, hydration capacity, true density, oil- holding capacity, loss on drying and particle size. The phytochemical and micromeritic properties were also investigated. The mucilage obtained was greyish in color without odor and a bland taste. It had a yield of 15 %, a pH of 5.36 and a true density of 1.65 g/cm^3. Its swelling index in water, phosphate buffer (pH 6.5) and 0.1 N HCl were found to be 30.6, 14.6 and 12 % respectively. The mucilage showed a very high hydration capacity of over 2000 % and an equally high oil- holding capacity of 582 %. The mean particle diameter was 330 µm and its loss on drying 0.9 %. The phytochemical analysis showed that it contained carbohydrates and mucilage. The micromeritic properties indicated cohesiveness of powder characterized by poor flow. The results obtained from the study show that the mucilage has great capacity to be utilized as a pharmaceutical excipient in both solid and liquid dosage forms.

Keywords: Mucilage, Heinsia crinita, Physicochemical, Excipient

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How to Cite this Article: Anukam NC, Izang EA, Poroye ED, Chris-Otubor GO, Damun PA. Some physicochemical characteristics of *heinsia crinita* mucilage. World J Pharm Sci 2019; 7(7): 31-37.

INTRODUCTION

In recent years, plant based polymers have awakened great interest due to the variety of ways in which they can be applied pharmaceutically such as binders [1], for controlled release of drugs[2], disintegrant in tablets[3] viscosifiers in oral liquids, protective colloids in suspensions[4] gelling agents in gels, film coating agents [5] and bases in suppositories [6]. While there is an abundance of synthetic polymers, the preference for natural gums and mucilages over the synthetic ones is justifiable due to their biocompatibility, lack of toxicity, low cost, non-irritant properties, environmental friendliness, and relative availability when compared to their synthetic counterparts [7].

Lipids, carbohydrates and proteins are natural polymeric materials which along with their derivatives represent a group of materials that are widely used in pharmaceutical formulations. These naturally occurring polymers have been employed as excipients in the pharmaceutical industry in the formulation of various dosage forms in which they play different roles[6].

Heinsia crinita (Afz) G. Taylor (Rubiaceae) is known as “Bush Apple” and it is called “Tonoposho” in Yoruba and “Atama” in Ibibio and “Etaabasi” among the Ogoni indigenes of southern Nigeria, in the Niger delta area of Nigeria[8]. It is a scrambling shrub or a small tree growing 8 - 13 metres tall in the under storey of high evergreen forests with persistent and very conspicuous leafy calyx-lobes [9], produces edible yellow or reddish fruits, sweet when ripe.

Heinsia crinita may be harvested from the wild as a local source of food and materials or is sometimes cultivated as a vegetable for its leaves. It may be found in Tropical Africa - Guinea and Sierra Leone to Somalia, Angola, Zimbabwe and Mozambique[10].

The Ibibio’s and Efiks in Southern Nigeria use the scented leaves in vegetable soup and also for the treatment of *craw craw* and head lice in children [11]. Other reported uses of the leaf include the treatment of umbilical hernia, hypertension, abscess and various ailments such as cough, catarrh, and sore throat among others[12].The antidiabetic and antiplasmodial activities of its ethanolic leaf extract in alloxan induced diabetes animals have been reported [13], The hypoglycaemic activity of the aqueous leaf extract in non-diabetic rats[14] and the hypoglycemic, hepatoprotective and nephroprotective effects of methanolic leaf extract in Alloxan-induced Diabetic Albino Wistar Rats, have also been reported [15].

Preliminary observation revealed that the plant contains a mucilage within its leaves. Presently, there are no reports of the characterization of any mucilage obtained from *Heinsia crinita* additionally, the mucilage is yet to be applied as a pharmaceutical excipient.

Before a natural product can be used as a pharmaceutical excipient, it must be characterized and appropriately investigated for safety, lack of toxicity, lack of activity, inertness and compatibility with the Active Pharmaceutical Ingredient (API) and other excipients. This study aims to evaluate some physicochemical and micromeritic properties of the mucilage.

MATERIALS AND METHODS

Materials: Matured leaves of *Heinsia crinita* were harvested from a local farm in Uyo, Akwa-Ibom State, Nigeria and submitted for taxonomy at the College of Forestry, Jos for authentication. Other chemicals and solvents used are of analytical grade.

Methods

Extraction of Mucilage: The leaves collected were dried under shade and milled. A 1.5kg quantity of the leaves was soaked in 30 L of boiling distilled water for 5 hours to allow for complete release of the polymer into the water.

An eight-fold muslin cloth was used to separate the marc and centrifuged for 30 mins at 3500 rpm and the supernatant collected. Concentrated ethanol was added to the filtrate in the ratio 3:1 and the polymer was precipitated out of solution, and rinsed severally with ethanol. The mucilage was subsequently placed in a wet mill and blended with acetone after which it was spread to dry at 60ºC for 1 hr. it was then milled and stored in dry powder jars.

Characterization of Extracted Mucilage

Physical characterization: The dried mucilage was studied for the percentage yield, organoleptic properties, solubility, pH, swelling index, hydration capacity, true density, oil-holding capacity, loss on drying, particle size distribution

*Solubility Behavior***:** The mucilage was tested for solubility using various solvents [16]

pH: pH was determined in 1%$^w/_v$ aqueous dispersion of the mucilage after it had been in water for 5 min, using a suitable pH meter at room temperature.

Swelling index: The swelling index is the volume in milliliter occupied by 1 g of a material, including any adhering mucilage, after it has swollen in aqueous liquid for 4 h. The method described in British Pharmacopoeia [17] was utilized, with slight modification. 1 g of mucilage was transferred into a 25 mL measuring cylinder, moistened with 1ml of ethanol 96% and 25 mL of distilled water. The cylinder was firmly closed and shaken vigorously every 10 min for 1 h then allowed to stand undisturbed for 3 h. The volume occupied by the material under test after the entire 4 h was measured and recorded as the swelling index. The test was repeated using Phosphate buffer (pH= 7.4) and 0.1 N HCl.

*Determination of hydration capacity***:** The method used by Musa, Muazu and Bhatia [18] was adopted. A 0.5g quantity of the mucilage was placed in a stoppered centrifuge tube. 10 ml of distilled water was added and shaken vigorously for 2 minutes, allowed to stand for 10 minutes during which it was mixed by inverting the tube three times at the end of 5 and 10 minutes. The sample was centrifuged at 1000rpm for 10 minutes, the aqueous supernatant carefully removed and the tube with the sediment reweighed. The hydration capacity was calculated as the ratio of the weight of sediment to initial weight of dry powder.

Determination of True density: according to Mbah et al., [19], a clean, dry density bottle was weighed, filled with xylene as the displacement fluid and reweighed. A 1.0 g quantity of the gum was weighed and poured into the density bottle containing xylene and the bottle and contents weighed.

Oil-holding capacity: A 0.2 g quantity of the gum was dispersed in 10ml arachis oil in a centrifuge tube shaken for 1 min and centrifuged at 2200 rpm for 30 min. The supernatant was removed and weighed. The oil-holding capacity was expressed as the grams oil held per gram sample of mucilage. difference between the initial volume of oil used and the volume of clear oil standing above the sedimented gum divided by the weight of gum expressed as a percentage.

Loss on Drying: One gram of powder was weighed accurately and dried in a hot air oven at 105°C.The weight was checked at intervals of 10min, until a constant weight was obtained.

Particle size analysis: A sieve-shaker assembly was used for this determination. A set of testing sieves (International and British test sieve series-ISO 3310-1, Endecotts Limited, London, UK) of sizes 180, 250, 500, 710, and 850 µm were arranged in descending order on a sieve shaker. A 20.0 g quantity of the sample was poured on the top sieve (850 µm) and a collecting plate placed at the bottom and the shaker was run for 20 min. At the end of the run, the amount of material retained on each sieve was weighed.

Chemical Characterization

Preliminary Phytochemical Studies: The mucilage was tested for preliminary phytochemicals such as carbohydrates, alkaloids, tannins, glycosides, flavonoids, reducing sugars, mucilage and proteins. [22].

Micromeritic Evaluation

Bulk density, Tapped density and Bulkiness: Using a 100ml capacity measuring cylinder and 10 g of mucilage, the bulk and tapped volume of polymer were determined.

Carr's Index and Hausner Ratio Determination: Data values obtained from bulk density and tapped density above were used to calculate the Carr's compressibility index and Hausner ratio

Flow rate/Angle of repose: According to Nep and Conway [23], 10 g of the dry polymer was introduced into a funnel clamped to a stand with its tip 10 cm from a plane paper surface and allowed to flow freely unto the paper surface and the time taken noted.

Statistical analysis: The data were analyzed statistically using package SPSS 22.0(SPSS Inc. Chicago, USA). All the data are presented as the mean of three determinations.

RESULTS AND DISCUSSION

Characterization of the isolated mucilage: Physical characterization: The percentage yield of the mucilage was found to be 15% (Table 2). The mucilage appeared to be gray in color, (Fig. 1) was odorless, with a bland taste, soft to touch and a fluffy texture. The bland taste of an extract is a desirable quality as it removes the challenge of needing to mask taste and increases patient acceptability [19]

Solubility: The solubility test was performed using various solvents. The mucilage was readily soluble in cold and hot water forming viscous colloidal solutions. It was however insoluble in ethanol, pet. Ether, chloroform and acetone. This indicates that the mucilage is of hydrophilic colloid group. The high water solubility of the gum may be due to soluble cell wall materials of the plant and the absence of cross-linking between polymeric chains [24] The results are tabulated in Table 3.

pH of Mucilage: Generally, the pH of an excipient is an important parameter in determining its suitability in formulations since the stability and physiological activity of most preparations depends on pH. The pH of 1% w/v solution of the mucilage was found to be in the range of 5.4. This indicates a slightly acidic pH. The acidity of the mucilage is not unexpected since many of them are known to contain salts of acidic polysaccharides and contain uronic acids in their structures [25].Acidic excipients may however require a slight adjustment of pH in the formulation of oral and/or buccal drug delivery systems [26]. Additionally, the human stomach is capable of absorbing most acidic drugs and weakly acidic drugs are more readily absorbed in the stomach especially those in undissociated form [19]. By extension a weakly acidic mucilage is likely to also be readily absorbed in the stomach.

Swelling index: swelling index indicates the degree to which a material is capable of absorbing its own equivalent weight of water [27]. Isah et al., [28] described swelling capacity as a measure of increase in the volume of the substance sequel to hydration. The swelling index of the isolated mucilage was found to be 30.6, 14.6 and 12% in distilled water, phosphate buffer (6.5pH) and 0.1 N HCl respectively. As the time increased, the swelling in water was also observed to increase. This indicates that the mucilage has good water absorbing capacity. Swelling index plays a key role in the utilization of gum for various purposes and decides the quality of gum in industrial applications. The swelling index can also characterize the rate at which a tablet will dissolute and is also indicative of the mechanism [29]. This implies that the mucilage can be used as a disintegrant in tablet formulation. Disintegrants generally act by two mechanisms- swelling or capillary water absorption (wicking) [30].

Hydration capacity: the commonest feature of all theories of disintegration is that water penetration (hydration) must precede disintegration [31]. Water penetration into a material is assessed by determination of hydration capacity of the material [28]. Hydration capacity of a substance is defined as a measure of the relative amount of water that can be taken up by the substance[30]. A good hydration capacity value indicates that water is taken into the polymer by wicking. The mucilage under investigation showed an extremely high ability (>2000 %) to take up water (table 2).

Oil-holding capacity: The oil holding capacity (OHC) of a gum is their ability to absorb or hold oil [20]. The oil held may include both physically entrapped oil occupying the interstices of the powder mass and the oil which may be due to the presence of proteins and fat moieties of the gum. The mucilage under investigation gave very high OHC values. A high OHC indicates the hydrophobic character of proteins in the mucilage and is decided by the content and type of hydrophobic fraction present[31]. This is because the non-polar groups present in the gum/mucilage are lipophilic in nature thus binding more lipid molecules by capillary attraction [20].

Loss on drying: Loss on drying of the isolated mucilage was found to be 0.9%. This shows that the moisture content of the mucilage is low when compared to the normal limit of not more than 10.0 % as specified in the European Pharmacopoeia[32]. This suggests its suitability in formulating moisture sensitive drugs. High moisture content in pharmaceutical substances and preparations provides a favorable environment for bacterial growth and if given suitable temperature, high moisture content will lead to activation of enzymes thereby affecting the shelf life of the drug [23]. Thus moisture content plays an important role in physical and chemical stability of the active pharmaceutical ingredient and pharmaceutical preparations [33].

Particle Size Analysis

An increase in particle size or a more uniform shape leads to a smaller angle of repose and smaller value of Carr's Index [19]. The mean diameter of particle was obtained to be 330 µm which represents a small particle sized distribution. This could be responsible for the high angle of repose and the equally high value for Carr's compressibility index data obtained in the present study

Chemical Characterization

Preliminary phytochemical studies

The mucilage was tested for chemical characteristics. Test for Alkaloids, Steroids, Flavonoids, Saponins, Tannins, Carbohydrate, Terpenoids, Glycosides, Amino acids and Mucilagewere conducted. The preliminary phytoconstituents studies showed the presence of carbohydrates and mucilage in the isolated mucilage which further confirmed that the sample is a mucilage. The results are presented in Table 4.

Micromeritic Evaluation of mucilage

The flow properties of powders are of significance in determining whether a material is suitable as a direct compression excipient. The angle of repose, Hausner index and Carr's percent compressibility are considered as indirect measurements of powder flow property, The Hausner index is indicative of interparticle friction, while the Carr's index shows the aptitude of a material to diminish in volume [34]. As the values of these indices increase, the flow of the powder decreases. In general, Hausner ratio greater than 1.25 indicates poor flow; Carr's index below 16 % is indicative of excellent flow ability while values above 21 % indicate cohesiveness [34}. Angle of repose is characteristic of the internal friction or cohesion of the particles. It is high if cohesive and other forces are high and vice versa. Generally, if the angle exceeds 50°, the powder will not flow satisfactorily while materials having values near 25°, flow easily and well [19]. This could be as a result of rougher and more irregular surface of particles or a high moisture content[35]. The Hausner's ratio previews the degree of densification which could occur during tableting. The higher the ratio, the greater the propensity of powder densification and this phenomenon may cause tablets which lack uniformity of weight and content to be produced. The result obtained (Table 5) for Hausner's ratio range signifies cohesiveness of the powder hence poor flow. However, for powders with a Hausner's quotient which falls between 1.25 and 1.5, addition of glidants will improve flow. This does not happen in powders with values greater than 1.5.It is known that porosity determines the swelling capacity of polymers, i.e. the higher the porosity the more the inter-particulate spaces where water could be absorbed[35] Generally the results (Table 5) shows a high porosity value for the mucilage under investigation. This result tallies with the correspondingly high swelling index of the mucilage (Table 2). Bulkiness (or specific bulk volume) is the reciprocal of bulk density. It is observed to increase with a decrease in particle size. It is an important consideration in the packaging of powder and also affects mixture of materials.

Table 1: Organoleptic Properties of the Mucilage

Parameters	Observations
Color	Gray
Odor	Odorless
Taste	Bland
Texture	Fluffy
Touch	Soft

Table 2: Physical properties of Mucilage

Parameters	Value
Percentage yield (%)	15
pH	5.36
Swelling Index (%)	
Water	30.6
Phosphate buffer (pH 6.5)	14.6
0.1N HCl	12
Hydration Capacity (%)	2019
True Density (g/cm^3)	1.65
Oil-Holding-Capacity (%)	582
Loss on Drying (%)	0.9
Mean Particle Diameter (µm)	330


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Figure 1: Isolated Mucilage

Table 3: Solubility of the mucilage with various solvents

Solvents	Observation
Ethanol	Insoluble
Chloroform	Insoluble
Water	Soluble
Pet ether	Insoluble
Acetone	Insoluble

Table 4: Preliminary Phytochemical Analysis

Test	Observation
Alkaloids	-
Steroids	-
Flavonoids	-
Saponins	-
Tannins	-
Carbohydrate	++
Terpenoids	-
Glycosides	-
Amino acids	-
Mucilage	++

Key: ++ = present, - = not present

Table 5: Micromeritic evaluation of mucilage

Property	Observation
Bulk Density (g/ml)	0.12
Tapped Density (g/ml)	0.17
Hausner's Quotient	1.38
Carr's Compressibility (%)	27.28
Bulkiness	8.3
Porosity (%)	93
Angle of Repose (◦)	50.09

CONCLUSION

Natural polymers play a vital role in the drug delivery. Thus prior to selection of a natural polymer, information regarding its physical and chemical properties should be carefully obtained. Natural gums and mucilagesare promising biodegradable polymeric materials and several polymers from plant origin have been successfully utilized as pharmaceutical excipient. This report deals with the isolation of mucilage from *Heinsia crinita* and its physicochemical and Micrometric characterization. The chemical characterization revealed the presence of carbohydrates and mucilage. The physical characterization of mucilage indicates its suitability for use as an excipient. The micrometric characterization indicated the high cohesiveness of the mucilage.

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