Extarction and charcterization of mucilage from Irvingia gabonensis seeds

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Abstract

This study focuses on the extraction and characterization of mucilage from Irvingia gabonensis seeds and its potential uses. The extraction process was carried out using water and the mucilage was characterized various physicochemical using and rheological parameters. All confirmatory tests for mucilage carried out (Molisch, iodine and ruthenium red test) were positive. The outcomes demonstrated that the mucilage had a yield of 52.6 %, a swelling capacity of 88.65 %, hydration capacity of 1.78 and a viscosity of 1.1805 mPa S. This study highlights the importance of utilizing natural for sustainable resources development and provides insights for

further research on the potential uses of mucilage from other plant sources.

Keywords: Ivringa gabonensis, extraction, characterization, mucilage.

Introduction

A molecule (macromolecule) made up of repetitive structural units is referred to as a polymer. Covalent chemical bonds are often used to join these subunits (Kulkarni et al., 2017) and they are used as excipients in pharmaceutical formulations to regulate the release of drugs from dosage forms. The globe is now becoming more and more interested in natural medications and excipients. Man has successfully used natural resources for ages in the medical and pharmaceutical industries (Choudhary & Pawar, 2014). They can also be altered to provide custom materials for drug delivery systems, which enables them to compete with synthetic items that are currently on the market (Bahadur et al., 2017). These natural materials are superior to synthetic ones because they are readily available, inexpensive, nontoxic, biodegradable and chemically inert (Alam et al., 2014) They can also be altered in a variety of ways to provide materials that are specifically designed for drug delivery systems (Jani et al., 2009). Several natural polymers have been characterized and evaluated as useful pharmaceutical excipients in various drug delivery systems (Akin-ajani et al., 2019). Notable them among are alginates, carrageenan, xanthan gum, guar gum, karaya gum, albizia gum, khaya gum and terminalia gum, which have been found useful as disintegrants, binders, emulsifiers and matrix systems (Bamiro et al., 2012).

Irvingia gabonensis (O'Rorke) Bail (family *Irvingiaceae*) is a fruit commonly referred to as ogbono in most oarts of Nigeria. Though it is also referred to as African bush mango, wild mango, dika nut and rainy season bush mango. *Irvingia gabonensis* bears edible fruits that resemble mango and is valued for their fat and protein-rich nuts. The nuts are used widely in Nigeria for thickening soups because they form a thick viscous sauce base when heated.

Different parts of the Irvingia plant have been utilized in herbal medicine for curing a variety of ailments including hernias, yellow fever, dysentery and poisons (Ewere et al., 2021). The antimicrobial, analgesic, antidiarrheagenic and anti-ulcer properties of the plant parts have been demonstrated (Mateus-Reguengo et al., 2019). The kernel or nuts and bark extract have been observed to reduce fasting blood glucose levels in both test animals and humans thus its possible usefulness in the management of diabetes, bodyweight reduction and blood cholesterol level control (Ngondi et al., 2005). The mucilage of its kernel has been investigated tablet binder. as a an emulsifying agent and a suspending agent in pharmaceuticals (Eraga et al., 2014). This

work aimed to extract the mucilage from *Irvingia gabonensis*, characterize it and evaluate its potential as a natural excipient.

Materials and Methods

Materials

Distilled water, N-hexane (Merck, USA), Ruthenium red solution (Otto chemie pvt Ltd, India), Molisch's reagent (Nice Chemicals Ltd, India), all other reagents used were of analytical grade.

Irivingia gabonensis fruits were obtained from Kaduna North Local Government Area of Kaduna State and it was identified at the Department of Biological Sciences Herbarium of Kaduna State University, Kaduna

Methods

Extraction of I. gabonensis mucilage

A brief modification of method described in (Coulibaly & Daouda, 2017) was used to carry out mucilage extraction of *I. gabonensis*. Five hundred grams (500g) kernels of *I. gabonensis* were grounded to powder using a mechanized grinding machine. The powder was delipidated by maceration in n-hexane for 24hours then the mixture was filtered using suction pump. The residue was collected, dried in hot air oven and was macerated for 24 hours in distilled water in a ratio of 1:50 (*I. gabonensis*: water), thereafter the mixture was filtered on a muslin cloth. The mucilage was collected in petri dishes, dried in hot air oven then the dried mucilage pulverized, transferred into an airtight container and stored in a desiccator for further use.

Percentage yield of extracted mucilage

The percentage yield of the extracted mucilage was determined using Equation 1 (Dagogot et al., 2020)

%	Yield	of	mucilage
_		Weight of ext	tracted mucilage
_		Weight of I	gabonesis seed

X 100..... 1

Preliminary test for mucilage

The following preliminary tests for mucilage were carried out using the methods described by Coulibaly & Daouda (2017)

Red ruthenium test

Exactly 0.1g of dried mucilage powder was mounted on a slide with a solution of ruthenium red and observed under a microscope.

Molisch test

In a clean test tube, 0.1 g of dried mucilage powder was placed in a clean test tube, two drops of the freshly prepared Molisch's reagent were added, and then concentrated sulfuric acid was added gradually to the side of the tube to form a layer above the aqueous solution.

Iodine test

In a test tube, 0.1 g of dried mucilage powder was added to 1 M iodine solution with 0.2% dye, and the combination was examined.

Physicochemical characterization of extracted mucilage

pH determination

This was done by first calibrating the pH meter (Super –scientific, Arizona, United State of America) then , shaking 1 % w/v dispersion of the sample in water for 5 min

and the pH of the solution was determined. The process was done in triplicates to ensure reliability.

Viscosity

The viscosity of 1.0 % aqueous dispersion of the extracted mucilage was determined at room temperature (27.0 \pm 2°C) at 100 rpm using a digital rotary viscometer (NDJ-5S, China). Spindle number one was used and measurements were carried out in triplicates then the average was taken.

Hydration capacity

One gram (1 g) of the mucilage was placed in a centrifuge tube and covered with 10 ml of distilled water. The tube was shaken for 2 h. and left to stand for 30 min before centrifuging at 3000 rpm for 10 minutes. The supernatant was drained, and the weight of the mucilage after water uptake and centrifugation was determined (Oh and Kim, 2022). Hydration capacity was calculated with Equation 2 (Oh and Kim, 2022).

Hydration	capacity	=
<u> </u>	– weight of dry sample	
weight of	dry sample	
	2	

Swelling index of mucilage

One gram (1 g) of *I. gabonenesis* mucilage was accurately weighed and transferred to a 100 ml stoppered measuring cylinder. The initial volume of the powder in the measuring cylinder was noted. The volume was made up to 100 ml with distilled water. The cylinder was stoppered, shaken gently, and set aside for 24 h. The volume occupied by the mucilage was noted after 24 h (Sunitha *et al.*, 2020). The swelling index (SI) is expressed as a percentage and calculated according to Equation 3

Mucilage flow properties

Angle of repose determination

The angle of repose was determined using the method described by Ohwoavworhua & Adelakun (2005).

The heights (h), of the powder cones and the mean diameters (D), of the base of the

powder cones, were determined and the tangent of the angle of repose calculated using Equation 4.

Tan	θ =	=
$\frac{2 h}{D}$	4	

Bulk and tapped densities determination

The bulk and tapped density of the extracted mucilage was determined using the method described by Dagogot et al., (2020). Two (2.0) grams of the extracted mucilage of *I. gabonensis* was placed in a 10 ml measuring cylinder and the volume, V_0 , occupied by each of the samples without tapping was noted. After 100 taps on the table, the occupied volume V_{100} was read. The bulk and tap densities were calculated as the ratio of weight to volume (V₀ and V₁₀₀ respectively).

Hausner's ratio

The Hausner ratio is a measure of the flowability of a powder. It is calculated by dividing the bulk density by the tapped density of the mucilage.

This was calculated using the Equation 5 (ASTM International)

Hausner ratio = $\frac{Tapped \ density}{Bulk \ density} \dots 5$

Carr's index

Carr's compressibility index is a measure of the compressibility of a powder. This was calculated using Equation 6 (B.P.C, 2021) Carr's index = $\frac{Tapped \ density - Bulk \ density}{X}$

Flow rate determination

Flow rate determination involves measuring the rate at which a substance flows through a specific apparatus. Thirty grams (30 g) (w) of the samples were placed in the Erweka flow apparatus (Erwekaapparatebau-G.m.b.H, Germany) and allowed to flow through the funnel orifice. The time (t) taken for the powder to flow through the orifice was noted and the flow rate was computed using Equation 7 (USP 44)

Flow	rate
$=\frac{W}{t}$.7

Surface Morphology

The surface morphology of the extracted material was examined using a scanning electron microscope. The sample was mounted on aluminum stubs and coated with a platinum coating of electrically conducting material, deposited on the sample using a vacuum coater.

Fourier transform infra-red spectroscopy (FT-IR)

The FTIR spectra of the extracted mucilage were obtained using an FT-IR spectrophotometer. The sample was blended with solid KBr (100 mg), and about 40 mg of the blend was used to prepare a pellet. The spectra were scanned from 4000 to 400 cm-1 in a FT-IR spectrometer (Agilent Technologies Cary 630) under dry air at room temperature.

Results

The extracted mucilage had a light brown color, a characteristic smell, and was tasteless. The percentage yield of the mucilage after extraction was 52.6%. The preliminary mucilage confirmation tests with the extracted mucilage were all positive, as shown in Table 1. The physicochemical parameters of the extracted and swelling index) are presented in Table 2. The flow properties of the extracted mucilage are presented in Table 3.

mucilage (pH, viscosity, moisture sorption, **Table 1: Preliminary confirmatory tests for mucilage**

S/N	Test	Results
1	Molish's test(carbohydrates)	Positive
2	Iodine test	Positive
3	Ruthenium red test (mucilage)	Positive

Table 2: Physicochemical properties of the extracted mucilage

S/ Parameters		Results
Ν		
1	pH	5.4 ± 0.073
2	Viscosity	$1.1805 \pm 0.025 \text{ mpa.s}$
3	Hydration capacity	1.78 ± 0.33
4	Swelling index (swellability)	88.65 ± 1.16 %

Table 3: Flow properties of extracted mucilage

S /	Properties	Results
Ν		
1	Angle of repose (degree)	32.13 ± 0.526
2	Bulk density (g/cm ³)	0.75 ± 0.012
3	Tapped density (g/cm ³)	0.86 ± 0.005
4	Hausner's ratio	1.10 ± 0.002
5	Carr's index (%)	12.79 ± 0.013
6	Flow rate (g/sec)	2.30 ± 0.015

Flow characteristics	Carr's index (%)	Hausner's ratio	Angle of repose
Excellent	≤10	1.00-1.11	25 - 30
Good	11-15	1.12-1.18	31 - 35
Fair	16-20	1.19-1.25	36 - 40
Passable	21-25	1.26-1.34	41 - 45
Poor	26-31	1.35-1.45	46 - 55
Very poor	32-27	1.46-1.59	56 - 65

Table 4: Showing	Standard Ranges	for flow properties
Tuole II Showing	Standard Itanges	

The FT-IR spectrum for mucilage extracted from Ivringa gabonensis is shown in Fig

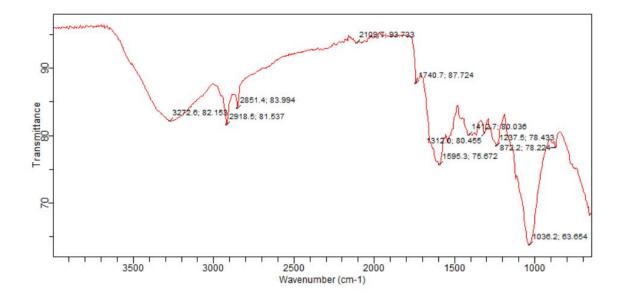


Fig 1: Showing the FTIR of the extracted Irivinga gabonesis mucilage

Table 4: FTIR peaks, intensity and assigned functional groups of *Irvingia gabonensis* mucilage

S/N	Peak	Intensity	Functional group
	(cm-1)		
1.	3272.6	82.153	O-H stretching (alcohol)
2.	2918.5	81.537	C-H stretching (alkane)
3.	2851.4	83.994	C-H stretching (alkane)
4.	2109.7	93.733	CEN stretching (nitrile)
5.	1704.7	87.724	C=O stretching (aldehyde)

6.	1595.3	75.672	C=C stretching (alkene)
7.	1302.0	80.455	C-O stretching (carbonyl)
8.	1412.7	80.036	C-H stretching (alkane)
9.	1237.5	78.433	C-O stretching (alkyl aryl
			ether)
10.	872.2	78.244	C-H bending (alkane)
11.	1036.2	63.654	C-O stretching (ester)

The result for the surface morphology as obtained from a scanning electron microscope is shown on Plate 1. It shows the morphology and surface characteristics of the particles. The particles appear flat, have different shapes, and look like flake.

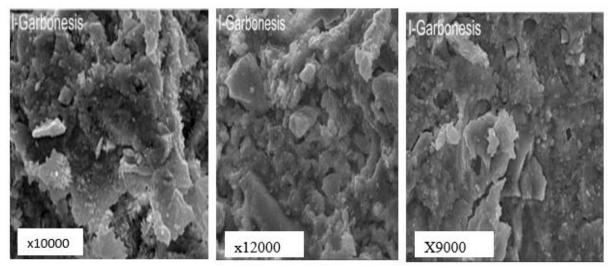


Plate 1: Showing the extracted mucilage surface morphology at x10000, x12000 and x9000 magnification.

Discussions

Yields around 100% are called *quantitative*, yields above about 90% are called *excellent*, yields above about 80% are *very good*, yields above about 70% are called *good*, yields below about 50% are called *fair*, and yields below about 40% are called *poor* (Vogel *et al.*, 1996). However, these values *JCBR Vol 3 Is 5 Sept-Oct 2023* are not universally accepted, and depending on the nature of the product in question, these expectations may be unrealistically high. Yields may appear to be 100% or above when products are impure, as the measured weight of the product will include the weight of any impurities (Petrucci *et al.*, 2002). *Irvingia gabonensis* extracted mucilage gave a yield of 52.6%, which is a good yield for natural products. It shows *Irvingia gabonensis* mucilage has a good likelihood for use in the pharmaceutical industry.

Red ruthenium test is used to confirm the presence of mucilage. The Molisch test is used to confirm the presence of carbohydrate in the mucilage. Iodine test is used to confirm the presence or absence of starch in the mucilage. The results were all positive, which signifies that the mucilage was extracted properly. The presence of carbohydrates in Irvingia gabonensis mucilage is an indication that it is a polysaccharide. This corroborates the findings of Assi et al. (2017), in which he carried out the same confirmatory tests on Irvingia gabonensis mucilage.

The pH of the extracted *Irvingia gabonensis* mucilage was 5.4. *Irvingia gabonensis* mucilage extract is thus acidic. The stability and physiological activity of most preparations are influenced by their pH; therefore, it is important to have knowledge of the pH of an excipient (Bamiro, 2011; Liang *et al.*, 2017). The pH of mucilages plays an important role in their functionality and has the tendency to form gels at pH values between 5.0-7.0 due to the presence of charged groups that can interact with the water molecules. A lower pH can result in a decreased stability of the mucilage gel and can affect its thickening and emulsifying properties in pharmaceutical products (Chime *et al.*, 2019).

The extracted mucilage had a viscosity of 1.180 ± 0.025 mPa. Mucilage typically displays non-Newtonian behavior, meaning that its viscosity fluctuates depending on the applied shear rate (Goswami & Saikia 2017). The range of mucilage viscosity is 100 to 5000 cP. High viscosity in mucilage is desirable for many industrial applications, including the food and pharmaceutical industries, where they are used as thickening agents. *Ivringa gabonensis* mucilage has been reported to have a high viscosity, which makes it suitable for its use in the

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food and pharmaceutical industries, and a low viscosity can lead to poor product quality and inferior performance (Onwuka *et al.*, 2014).

The hydration capacity refers to the ability of the mucilage to absorb and hold water molecules and was 1.78 ± 0.33 . The hydration capacity of mucilage depends on various factors such as the botanical source, time of harvest, and extraction method (Karki & Kim 2016). The hydration capacity affects how viscous liquids form, which might help industrial processes. When in contact with water, mucin creates a three-dimensional network that traps the liquid and produces highly viscous solutions. Similar to pectins, gums, and other algal polysaccharides, mucilage has a high capacity to bind or retain water since it is predominantly made up of galactose, mannose, xylose, and other sugars. Mucilage can dissolve, disperse, and create colloids in foods. cosmetics. and pharmaceuticals, where it may be used because of its high propensity to absorb

water (Monrroy et al., 2017). A high hydration capacity is desirable for many industrial applications, as it can improve texture, rheological properties, and stability of products, whereas a low hydration capacity can result in a lack of water binding capacity and can affect the stability and shelf-life of products (Okonkwo et al., 2017). So many scientific studies have shown that high water holding capacity is due to the presence of hydroxyl groups and protein substituents in the gum and mucilage structure, and a low amount of hydration capacity can be attributed to the high solubility of mucilage even at a high concentration, which leads to an inability to form a gel (Hadad and Goli 2018).

The swelling index of a mucilage which is the extent to which it swells in the presence of water was 88.65 ± 1.16 %. Swelling indices of mucilage can vary depending on the source used. Generally, the swelling index of mucilage lies in the range of 2-10 g/g (Yadav & Mishra 2013). A high swelling index can be desirable in many

industrial applications, as it can improve the water binding capacity of products and enhance their texture and our mucilage showed a high swelling index which makes it a useful ingredient in pharmaceutical preparations (Onwuka *et al.*, 2014). On the other hand, a low swelling index can result in poor water binding capacity and affect the texture and stability of products.

It's worth noting that mucilage composition and properties can vary widely depending on the specific plant source and processing method, so these ranges may not apply to all mucilage-based pharmaceutical preparations. The flow properties of a mucilage play a crucial role in pharmaceutical product formulation as they determine the ease of processing, handling, and dosing of the product therefore, good flow properties are desired as they ensure consistent and uniform distribution of active pharmaceutical ingredients (APIs) and excipients, enhance blend homogeneity, and enable efficient manufacturing processes and on the other hand, bad flow properties

can lead to issues such as segregation, powder caking, or uneven distribution of APIs, which can negatively impact the quality, efficacy, and safety of the final pharmaceutical product (Shesky et al., 2012) The bulk and tapped densities of the gum were 0.75 and 0.86 g/cm³, respectively. Bulk and tapped densities of material affect the handling and process method to be adopted when included in a formulation. A material with low tapped density would be difficult to compress while a highly compressible material should have a high tapped density (Ogaji et al., 2012). In the case of the extracted mucilage, the tapped density was more than that of the bulk suggesting it is highly compressible, a good quality attribute for direct compression operation. The bulk and tapped densities, provide an understanding of the particle packing and rearrangement when zero or low pressure has been applied (Bernard et al., 2022).

Hausner ratio measures interparticle friction and flowability, whereas Carr's index

measures a powder's ability to decrease in volume, which is a measure of powder compressibility and both the Hausner ratio and Carr's index are useful in predicting powder flowability therefore the lower a material's Carr index, the better its flowability but the worse its compressibility (Humbert-Droz *et al.*, 1983). Higher values indicate poor flow, non-uniformity and variations in drug release (Merchant *et al.*, 2015). A high Hausner ratio will imply poor flow characteristics and a low ratio is more desirable and indicates good flow properties (Zhmud *et al.*, 2018).

Carrs index of 12.79 and Hausners ratio of 1.10 were reported for the mucilage, indicating good flowability comparing to standard values (Carrs index 11 – 15 exhibit good flow and Hausners ratio 1.0 - 1.11exhibit excellent flow). This implies that that the obtained mucilage had a good flow. The angle of repose is a quantitative measure of the internal cohesion and coefficient of friction impact under relatively low loading. such as that

experienced during powder blending, tablet dies, or capsule shell filling (Odeku 2005). Angles between $25 - 35^\circ$ are typically indicative of free-flowing materials. whereas angles of $\geq 40^{\circ}$ indicate poor flow (Odeku and Picker-Freyer 2007). The extract mucilage gave an angle of repose of 32.13°, which indicates the free flowing nature of the mucilage. Powders with poor flowability may have difficulties in flowing uniformly during the manufacturing process and filling into capsules or moulds (Li et al., 2020)

The flow rate of the mucilage was 2.30g/sec. The flow rate of mucilage can vary based on several factors such as concentration of the mucilage solution, temperature and viscosity of the solution (Chai *et al.*, 2018). It reflects the ease of powder flow and is influenced by particle size, shape, and surface properties and good flow properties are associated with higher flow rates, as they ensure efficient and consistent powder flow during dosing, blending, and tableting (Kaya *et al.*, 2021).

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FT-IR spectroscopy was used to identify the functional groups present in mucilage and confirm the occurrence of peak characteristics of polysaccharides. The frequency of vibration of a part of molecule in the sample is the energy at which any peak in absorption spectrum appears (Stuart, 2004; Michael, 2017). The 3272.6 cm^-1 band corresponds to stretching vibrations of O-H bonds, indicating the presence of hydroxyl groups. Hydroxyl groups can act as hydrogen bond donors, contributing to the viscosity and gel-forming properties of mucilage. These properties make Irvingia gabonensis mucilage a potential candidate for sustained-release drug delivery systems (Adel et al., 2010). The 2918.5 cm⁻¹ and 2851.4 cm⁻¹ bands signify the stretching vibrations of C-H bonds, indicating the presence of aliphatic compounds. Aliphatic compounds can contribute to the stability emulsifying properties of and formulations. pharmaceutical These properties suggest that Irvingia gabonensis mucilage may find applications as а

suspending and emulsifying agent in oral liquid dosage forms (Faccio et al., 2015). The 2109.7 cm⁻¹ band corresponds to stretching vibrations of C≡N bonds, suggesting the presence of nitrile groups. Nitriles have potential applications in drug delivery systems due to their ability to form with metal ions. complexes Irvingia gabonensis mucilage could therefore be explored for its chelating properties in pharmaceutical formulations (Petera et al., 2015). The 1704.7 cm⁻¹ band corresponds to stretching vibrations of C=O bonds, indicating the presence of carbonyl groups. Carbonyl groups common are in polysaccharides and contribute to their filmforming and mucoadhesive properties. The mucoadhesive nature of Irvingia gabonensis mucilage may make it suitable for buccal drug delivery systems (Pachuau et al., 2012). The 1595.3 cm⁻¹ band corresponds to stretching vibrations of C=C bonds. suggesting the presence of unsaturated compounds. Unsaturated compounds can act as radical scavengers, protecting drugs from

oxidative degradation. Incorporating Irvingia gabonensis mucilage into pharmaceutical formulations may enhance their stability (Eddy et al., 2013; Udoh et al., 2017). The 1302.0 cm⁻¹ band corresponds to stretching vibrations of C-O bonds, indicating the presence of ether or ester groups. These functional groups could contribute to the solubility-enhancing of mucilage, properties making it a promising candidate for improving the dissolution rate of poorly soluble drugs in oral solid dosage forms (Njanbi et al., 2018). The 1412.7 cm⁻¹ band corresponds to bending vibrations of C-H bonds, suggesting the presence of methyl groups. Methyl groups can enhance the lipophilicity of drug molecules, potentially improving their permeability across biological membranes. This property could be leveraged in the development of transdermal drug delivery systems (Augustin et al., 2014). The 1237.5 cm⁻¹ band corresponds to stretching vibrations of C-O bonds, indicating the presence of alcoholic or

phenolic groups. These groups confer antioxidant properties, making Irvingia mucilage gabonensis suitable for formulations requiring oxidative stability (Ozougwu et al., 2020). The 872.2 cm^-1 band corresponds to bending vibrations of C-H bonds, indicating the presence of aromatic or substituted aromatic compounds. Aromatic compounds possess antioxidative, antimicrobial, and anti-inflammatory properties, potentially broadening the applications of Irvingia Gabonensis mucilage in pharmaceutical formulations (Augustin et al., 2014). The 1036.2 cm^-1 band corresponds to stretching vibrations of C-O bonds, suggestive of the presence of alcohols, ethers, or esters. These functional groups may contribute to the solubilityenhancing and surfactant properties of the mucilage, making it suitable for improving drug bioavailability (Ozougwu et al., 2020).

Scanning electron microscopy of the *Irvingia gabonensis* extracted mucilage indicates the morphology and surface characteristics of the particles. *Irvingia*

gabonensis extracted mucilage particles were mostly irregular in shape and appear flaky. Particle shape has been shown to affect the compaction characteristic of powders (Wray, 1992). The more irregular the particles of a material, the greater the likelihood of them fragmenting during compaction thus better compatibility (Odeku *et al.*, 2013). In addition, particle irregularity allows for closer packing during compression, hence higher compact strength.

Conclusion

In conclusion, the extraction and characterization of mucilage from Irvingia gabonensis seeds through preliminary confirmatory physicochemical tests, properties, and flow properties provide valuable insights into its potential uses. The characterizations involving preliminary confirmatory tests help in determining the presence of key components such as carbohydrates and proteins, while physicochemical properties shed light on its viscosity, solubility, and stability. Understanding and optimizing the flow

properties of mucilage used in pharmaceutical product formulation is essential for achieving high-quality, uniform, and consistent drug products. Various techniques, such as modifying particle size, particle shape, surface properties, or adding flow aids, can be employed to improve the properties of mucilage-based flow formulations, ensuring proper handling, processing, and administration of pharmaceutical products. The FTIR spectra analysis of Irvingia gabonensis mucilage revealed the presence of several functional groups that can influence its applications in pharmaceutical formulations. Further studies, such as in vitro and in vivo evaluations, are warranted to validate these implications and explore the feasibility of Irvingia gabonensis mucilage in diverse pharmaceutical formulations. Furthermore, its natural origin makes it an attractive alternative synthetic additives, to contributing to the increasing demand for sustainable and eco-friendly solutions. In summary, the extraction and

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characterization of mucilage from *Irvingia* gabonensis seeds pave the way for the development of new uses, further highlighting its potential as a valuable resource in various industries.

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