



## From Plant To Pill: Investigating *Prosopis Juliflora* Fruit Mucilage As A Pharmaceutical Excipient Through Isolation And Physicochemical Analysis

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Article History	Abstract
Received: 10 January 2024 Revised: 25 January 2024 Accepted: 15 February 2024	This study investigates the extraction and characterization of mucilage from <i>Prosopis juliflora</i> fruits as a potential pharmaceutical excipient. Employing water as the extraction medium, two precipitation approaches (Acetone and 95% ethanol) were compared. Physical, chemical, and micrometric analyses, along with FTIR spectroscopy, were conducted on the mucilages, demonstrating their compliance with official pharmaceutical parameters. Notably, mucilage precipitated with acetone exhibited superior characteristics compared to ethanol. The research concludes that <i>Prosopis juliflora</i> fruit mucilage holds promise as a tablet binder and viscosity-modulating agent in pharmaceutical formulations, offering a cost-effective and sustainable alternative for pharmaceutical applications. Further studies, including in vivo testing and stability assessments, are warranted for comprehensive validation of its pharmaceutical utility.
<b>CC License</b> CC-BY-NC-SA 4.0	<b>Keywords:</b> <i>Prosopis juliflora</i> , fruits, Isolation, Characterization

### INTRODUCTION

*Prosopis juliflora*, commonly referred to as mesquite, is a resilient and adaptive tree that thrives in arid and semi-arid regions, particularly originating from south India. Its botanical classification places it within the Fabaceae family. Mesquite stands out for its ability to endure challenging environmental conditions, showcasing features such as distinctive elongated pods, feathery foliage, and an extensive root system that enables it to withstand periods of limited water availability.

One of the remarkable ecological attributes of *P. juliflora* is its capacity for nitrogen fixation. Through a symbiotic relationship with nitrogen-fixing bacteria in its root nodules, mesquite contributes to soil enrichment. This nitrogen-fixing ability renders the tree crucial for soil conservation efforts and ecological restoration initiatives in regions facing aridity and soil degradation.

*P. juliflora* has long been an integral part of the cultural and culinary traditions of the communities inhabiting its native regions. The edible pods of the tree have been utilized as a traditional food source, often ground into flour for baking or consumed directly. Moreover, the wood of mesquite is highly valued for its durability and resilience, finding applications in construction, furniture crafting, and the production of high-quality charcoal.

In recent years, the scientific community has turned its attention to the potential pharmaceutical and biotechnological applications of *P. juliflora*. The study described earlier highlights the extraction and characterization of mucilage from its fruits, showcasing promising properties for pharmaceutical use (Ahad *et al.*). The mucilage, when precipitated with acetone or ethanol, demonstrates qualities suitable for tablet binding and viscosity modulation in pharmaceutical formulations.

However, it is crucial to acknowledge that the introduction of *P. juliflora* to new regions has raised concerns about its invasive nature. Uncontrolled growth of mesquite can outcompete native vegetation, impacting biodiversity and upsetting ecosystem dynamics. Therefore, the sustainable management and controlled utilization of *P. juliflora* are essential, emphasizing a holistic approach that leverages its benefits while addressing potential ecological challenges in introduced environments. The taxonomy of the plant is described in Fig. 1.

<b>Kingdom</b>	• <b>Plantae</b>
<b>Sub kingdom</b>	• <b>Tracheobionta</b>
<b>Division</b>	• <b>Magnoliophyta</b>
<b>Class</b>	• <b>Magnoliopsida</b>
<b>Sub class</b>	• <b>Rosidae</b>
<b>Order</b>	• <b>Fabales</b>
<b>Family</b>	• <b>Fabaceae</b>
<b>Genus</b>	• <b>Prosopis</b>
<b>species</b>	• <b>juliflora</b>

**Fig. 1: Taxonomical classification of *P. juliflora***

## MATERIALS AND METHODS

### Materials

Fresh fruits of *P. juliflora* were gathered from plants in the vicinity of Bangalore, India, and subsequently authenticated (IIS/BD/22/C-37) by the Biology department of the Indian Institute of Science, Bangalore. Ethanol (95%), acetone, diethyl ether, and methanol were procured from Fisher Chemicals, while double distilled water was employed as needed in the research.

### Organoleptic Characters

The organoleptic strictures were depicted in fig.2.

<b>Colour</b>	• <b>Brownish-yellow</b>
<b>Odour</b>	• <b>Odourless</b>
<b>Taste</b>	• <b>Slightly sweetish</b>
<b>Fruit size</b>	• <b>10-15 cm</b>
<b>Shape</b>	• <b>Oblong</b>

**Fig. 2: Organoleptic rebukes of *P. juliflora* fruits**

### Chemical constituents

*P. juliflora*, commonly known as mesquite, is characterized by a diverse array of chemical constituents. Among these are polysaccharides, particularly abundant in the mucilage extracted from *P. juliflora* fruit. These polysaccharides contribute to the thickening and gelling properties of the mucilage. Additionally, mesquite pods contain proteins, making them a noteworthy nutritional component. The dietary fiber found in these pods serves as a valuable contribution to digestive health (SivaKumar *et al.*, 2009).

Furthermore, *P. juliflora* is known to contain flavonoids, which are plant compounds recognized for their antioxidant properties. These flavonoids play a role in the plant's defense against oxidative stress. Tannins, another group of chemical compounds, may be present in mesquite pods, imparting astringent properties and potential health benefits (Saravanakumar *et al.*, 2014).

Alkaloids, nitrogen-containing compounds, are reported in some species within the *Prosopis* genus, contributing to the plant's physiological effects. Saponins, glycosides with foaming properties, are also identified in various parts of *P. juliflora*, with potential biological activities (Sharifi-Rad *et al.*, 2019).

It is important to consider that the chemical composition can vary among different parts of the plant, such as pods or leaves, and among different varieties or species within the *Prosopis* genus. Additionally, factors like extraction methods and geographic location can influence the chemical profile. For a more specific focus on the chemical constituents of *P. juliflora* fruit mucilage, additional research or analysis of the particular sample in question may be necessary (Damasceno *et al.*, 2017; Elbehairi *et al.*, 2020).

This diverse array of chemical constituents underscores the multifaceted nature of *P. juliflora* and hints at its possible utilization in various sectors, including pharmaceuticals, agriculture, and industry, depending on the specific properties and applications of its components.

### Expected uses of *P. juliflora*

*P. juliflora* (mesquite), has several uses across various domains due to its hardy nature and versatile characteristics. Here are some of the notable uses (Damasceno *et al.*, 2017; de Lemos *et al.*, 2023; Patnaik *et al.*, 2017; Wakie *et al.*, 2016):

**Fuel and Energy:** Mesquite wood is renowned for its high energy content, making it a valuable source of fuel and firewood. The wood is used for cooking, heating, and as a source of renewable energy.

**Timber and Construction:** The durable and sturdy wood of *P. juliflora* is utilized in construction and carpentry. It is particularly valued for making furniture, fence posts, and building materials.

**Food and Forage:** The pods of mesquite are edible and have been traditionally used as a food source. They can be ground into flour, offering a nutritional ingredient for baking and cooking. Additionally, the pods serve as forage for livestock.

**Soil Conservation:** Mesquite has a deep and extensive root system that aids in soil conservation. The plant is often used in erosion control and for stabilizing soil in arid and semi-arid regions.

**Pharmaceutical Potential:** As highlighted in the research you mentioned earlier, *P. juliflora* has pharmaceutical potential, particularly the mucilage extracted from its fruits. It may be used as a tablet binder and viscosity-modulating agent in pharmaceutical preparations.

**Biomass and Biofuel Production:** Mesquite can be cultivated for biomass production, contributing to sustainable biofuel initiatives. The plant's rapid growth and adaptability make it a potential feedstock for bioenergy production.

**Environmental Remediation:** Due to its nitrogen-fixing ability, mesquite can contribute to soil improvement and restoration. It is often used in reforestation and afforestation projects to rehabilitate degraded lands.

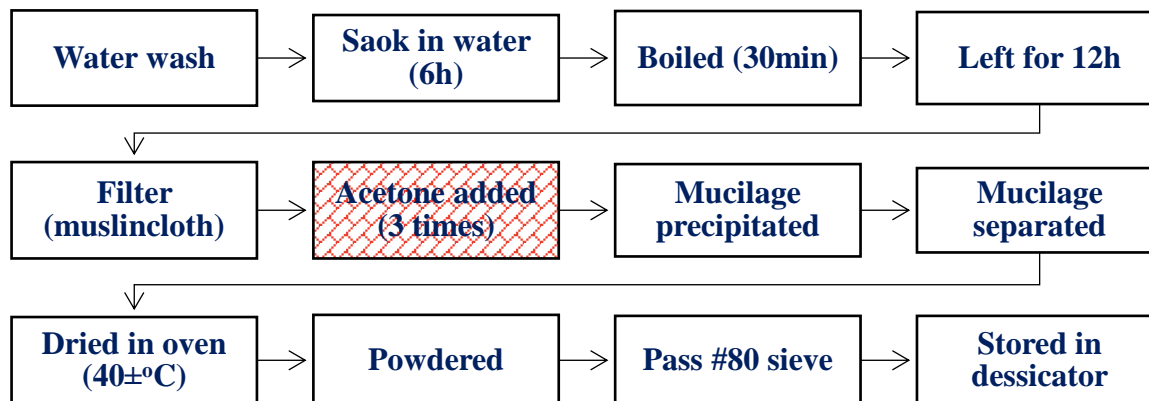
**Bee Forage:** Mesquite flowers produce nectar that attracts bees, making it a valuable forage source for honey production. In regions where it is abundant, mesquite can support local beekeeping and honey industries. While *P. juliflora* offers numerous benefits, it's important to note that in certain regions, it has become invasive and poses ecological challenges. Therefore, careful management and sustainable utilization practices are necessary to balance its advantages with potential environmental impacts.

### Extraction of Mucilage

PJFM was extracted by 2 methodologies.

#### Approach 1:

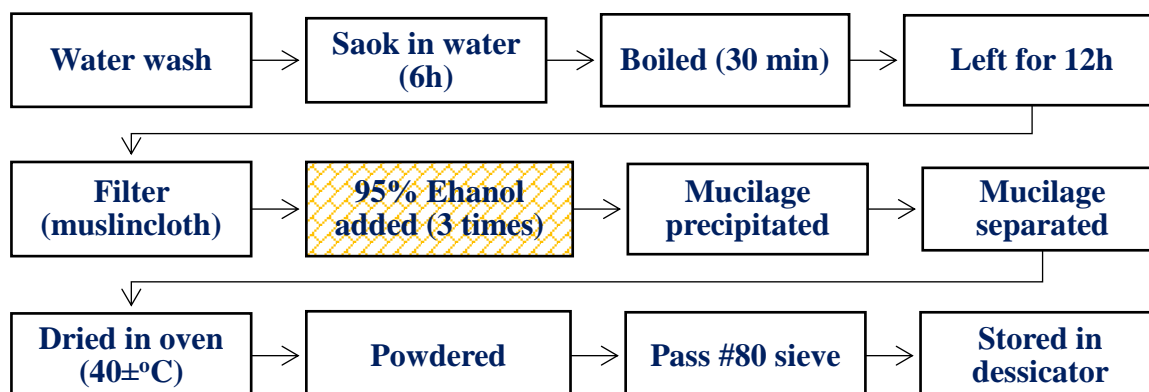
The mucilage from *P. juliflora* fruits was obtained through water extraction, and subsequently, the mucilage was precipitated (Krishna *et al.*, 2011) using acetone (Fig.3).



**Fig. 3: Segregation of mucilage by Acetone precipitation**

#### Approach 2:

The extraction of mucilage from *P. juliflora* fruits involved the use of water, and the ensuing precipitation (de Brito Damasceno *et al.*, 2020) of the mucilage was carried out using 95% v/v ethanol (Fig.4).



**Fig. 4: Separation of mucilage using ethanol precipitation**

### Description of Mucilage

The mucilage from *P. juliflora* fruits underwent evaluation for the following parameters.

#### Organoleptic constraints

The evaluation of *P. juliflora* mucilage encompassed a thorough assessment of its color, odor, taste, and texture. The color analysis aimed to characterize the visual appearance of the mucilage, providing insights into its hue and overall pigmentation. Odor assessment sought to identify any distinctive or characteristic smells associated with the mucilage, contributing to a comprehensive sensory profile. The taste evaluation delved into the flavor attributes, examining any discernible tastes that could influence its palatability or potential applications. Finally, the texture analysis explored the consistency and feel of the mucilage, shedding light on its physical properties. Together, these assessments aimed to provide a comprehensive understanding of the sensory attributes of *P. juliflora* mucilage, essential for its potential utilization in various applications, particularly in the pharmaceutical and food industries (Ahad *et al.*, 2011).

#### Physical constraints

The mucilage from *P. juliflora* was subjected to a comprehensive analysis, investigating various aspects and constraints (Hindustan *et al.*, 2012).

**Solubility**

The solubility characteristics of *P. juliflora* were elucidated through a comprehensive examination involving different solvents, including water and various organic solvents. This investigation aimed to provide insights into the plant's dissolution behavior in diverse chemical environments. By assessing solubility across different solvents, ranging from polar to non-polar, a more nuanced understanding of the plant's chemical composition and potential applications could be gained. This information is crucial for determining the solubility profile of *P. juliflora* in different contexts, influencing its suitability for various industrial, pharmaceutical, or research applications (Syiemlieh *et al.*, 2023).

**Yield**

The quantification of extracted mucilage from *P. juliflora* was achieved by applying a specific formula. The formula, tailored to the nature of mucilage extraction, facilitated the precise determination of the amounts obtained. This quantitative approach played a pivotal role in assessing the yield and efficiency of the extraction process. The utilization of such a formula adds a quantitative dimension to the research, allowing for accurate and reproducible measurements, which are essential for drawing meaningful conclusions regarding the extraction efficiency of *P. juliflora* mucilage for potential applications in various industries (Ngamwonglumert *et al.*, 2017).

$$\frac{\text{weight of mucilage after drying}}{\text{weight of } P. \text{ovata mucilage taken}} \times 100$$

**Particle size**

The mucilage extracted from *P. juliflora* underwent a size assessment using a compound microscope, and the mean size was subsequently determined. This microscopic examination allowed for a detailed analysis of the particulate dimensions within the mucilage sample. The calculated mean size provides valuable information about the average particle size, contributing to a comprehensive understanding of the morphological characteristics of *P. juliflora* mucilage. Such size assessments are instrumental in elucidating the potential applications of the mucilage, particularly in pharmaceutical formulations or other industries where particle size plays a critical role.

**Loss on drying (LOD)**

The determination of LOD serves as a critical parameter for assessing the moisture content in *P. juliflora* mucilage. In this analysis, a representative sample weighing 1g was subjected to control heating at 105°C within an oven. The process continued until a constant or unceasing weight was achieved, signifying that the mucilage had reached a state of equilibrium, and no further loss in weight was observed. The recorded loss of moisture during this drying procedure was meticulously documented. This methodological approach enables precise quantification of the moisture content within the mucilage, offering valuable insights into its stability, storage requirements, and potential applications in various industries, particularly in pharmaceuticals or food formulations where moisture content can significantly impact product quality (Nagesh *et al.*, 2012).

$$\frac{\text{mass of water in mucilage}}{\text{total mass of mucilage}} \times 100$$

**Swelling index (SI)**

A measured quantity of 1g of *P. juliflora* mucilage was carefully introduced into a 100 ml stopper measuring cylinder with a total volume capacity of 100 ml. Distilled water was then added to the cylinder containing the mucilage, and the mixture was thoroughly shaken to ensure uniform dispersion. Subsequently, the setup was allowed to stand undisturbed for a designated period of 24 h, during which the mucilage underwent hydration and swelling. Following this hydration period, the extent of swelling was meticulously measured. This experimental approach serves to assess the water absorption and swelling capacity of *P. juliflora* mucilage, providing valuable data on its physicochemical properties that are pertinent to various applications, particularly in pharmaceutical and industrial formulations where swelling behavior is a crucial parameter (Yahya *et al.*, 2018).

$$\frac{\text{final volume} - \text{initial volume}}{\text{initial volume}} \times 100$$

### **Melting point**

*P. juliflora* mucilage was introduced into an open capillary, and the capillary was carefully positioned within a melting point apparatus. The heating process was initiated, and the mucilage was subjected to gradually increasing temperatures until it reached its melting point. This meticulous procedure involved observing the physical transformation of the mucilage from a solid to a liquid state, with the melting point serving as a key parameter in characterizing the thermal properties of PJFM. The utilization of a controlled and standardized apparatus ensures precision in determining the melting point, contributing valuable data for understanding the thermal behavior of PJFM and informing potential applications in various industries, such as pharmaceuticals and material sciences (Hamuda *et al.*, 2016).

### **pH**

A measured quantity of 1g of PJFM was meticulously combined with 10 ml of distilled water. The resulting mixture was then subjected to pH assessment using a universal pH paper. This method allowed for the determination of the mucilage's acidity or alkalinity based on the color changes observed on the pH paper. By employing a universal pH indicator, the pH of the PJFM solution could be qualitatively assessed across the entire pH spectrum. This pH analysis is integral for understanding the mucilage's chemical nature, and it provides valuable information for potential applications, particularly in industries such as pharmaceuticals and cosmetics where pH can influence product stability and efficacy (Ingle *et al.*, 2017).

### **Chemical characterization**

The PJFM underwent a comprehensive analysis to identify specific chemical constituents, encompassing steroids, saponins, flavonoids, and tannins. The presence of these compounds is crucial as they contribute to the mucilage's potential applications, particularly in pharmaceutical and nutritional contexts where these chemical entities can impart beneficial properties.

Furthermore, the mucilage was subjected to tests aimed at detecting potentially undesirable substances. This included scrutiny for foreign matter to ensure the purity of the mucilage, heavy metal analysis to assess the presence of metallic contaminants, and testing for arsenic, a toxic element that could pose health risks. These analyses serve to confirm the safety and quality of the PJFM, making it imperative for applications where purity and safety are paramount considerations, such as in pharmaceutical or food industries. The dual approach of identifying beneficial compounds while rigorously assessing for potential contaminants ensures a comprehensive understanding of the chemical composition of PJFM for its safe and effective utilization in various applications (Sharifi-Rad *et al.*, 2019).

### **Flow properties**

The dried PJFM underwent a meticulous evaluation to assess its flow properties, encompassing several key parameters. The angle of repose, a fundamental measure of powder flowability, was determined by carefully allowing the mucilage to flow freely through a funnel onto a flat surface, and the resultant cone angle was measured.

Bulk densities, another critical parameter influencing material handling and compaction, were ascertained. This involved measuring both the loose and tapped densities of the mucilage, providing insights into its packing characteristics. The compressibility index, indicative of the powder's compressibility and flowability, was calculated based on the bulk and tapped density values.

Additionally, Hausner's ratio, a dimensionless parameter derived from bulk and tapped densities, was determined. It serves as an indicator of the powder's cohesiveness and is integral for understanding its flow properties. This comprehensive analysis of flow properties is essential for applications in pharmaceutical and powder processing industries, where the ease of handling, mixing, and compaction are critical factors in the formulation and manufacturing processes. The obtained data collectively offers valuable insights into the practical utility of dried *P. juliflora* mucilage in various industrial applications (Busch *et al.*, 2018).

### **Rheological studies**

A solution of 1% w/v PJFM underwent a thorough evaluation of its viscosity, employing an Oswald viscometer. This experimental setup involved carefully preparing a solution with a known concentration of 1% w/v, ensuring accurate measurements of both the mucilage and the solvent.

The Oswald viscometer, a specialized instrument for viscosity measurements was employed to gauge the flow properties of the PJFM solution. The procedure involved introducing the solution into the viscometer, and the time taken for a specific volume to flow through the capillary was recorded. This time measurement

allowed for the determination of the solution's viscosity, providing crucial information about its resistance to flow (Vadapalani Nallasivam *et al.*, 2022).

Viscosity testing is of particular significance in various industries, especially pharmaceuticals and food, where the flow behavior of solutions plays a vital role in product formulation and processing. The data derived from this assessment offers insights into the rheological properties of the PJFM solution, aiding in its potential applications as a viscosity-modulating agent in various formulations.

#### Fourier transform infrared (FTIR) studies

FTIR spectra of PJFM were meticulously recorded utilizing a high-precision FTIR spectrophotometer (Bruker). The analysis involved the preparation of samples incorporated into KBr disks, ensuring a uniform and representative distribution of the mucilage for spectral analysis. The FTIR spectrophotometer was set to scan within the wavenumber range of 500 to 4000  $\text{cm}^{-1}$ .

This spectral analysis in the infrared region is instrumental for identifying and characterizing the functional groups present in the PJFM. The wavenumber range selected is particularly significant as it covers a broad spectrum, allowing for the detection of various chemical bonds and molecular vibrations. The resulting FTIR spectra provide a fingerprint-like representation of the mucilage's chemical composition, revealing valuable information about the types of bonds, molecular structures, and functional groups present (Bishnoi *et al.*, 2019).

The utilization of sophisticated instrumentation such as the Bruker FTIR spectrophotometer enhances the accuracy and reliability of the spectral data, contributing to a comprehensive understanding of the chemical makeup of PJFM. This information is pivotal for elucidating the potential applications of PJFM in diverse industrial sectors, including pharmaceuticals, food, and materials science.

## RESULTS AND DISCUSSIONS

The confirmation of PJFM was established through the ruthenium red test, resulting in a distinct pink coloration. The visual and sensory analysis revealed that PJFM appeared as a light brown substance with no discernible odor or taste. In terms of solubility, PJFM exhibited insolubility in acetone, ethanol, methanol, dichloromethane, and diethyl ether, forming a gel-like solution only in water.

The acetone-precipitated mucilage demonstrated a higher yield, measuring  $42.25 \pm 0.09$  g per 100g of the sample. This mucilage exhibited fine particle size ( $151.76 \pm 3.2$   $\mu\text{m}$ ), low LOD at  $2.15 \pm 0.03$ , and substantial swelling capacity of  $74.84 \pm 2.81\%$ . Both isolated PJFM samples demonstrated similar melting points with a nearly neutral pH. The presence of foreign matter was negligible, and the heavy metal and arsenic content fell within acceptable limits.

PJFM exhibited negative reactions for steroids, saponins, flavonoids, and phenolic contents. The angle of repose for the dried PJFM was less than  $25^\circ$ , indicative of excellent flow properties following the Indian Pharmacopoeia. Bulk and tapped density values facilitated the calculation of the compressibility index and Hausner's ratio, both of which were found to be below 10% and 1.25, respectively, confirming the outstanding flow properties of PJFM.

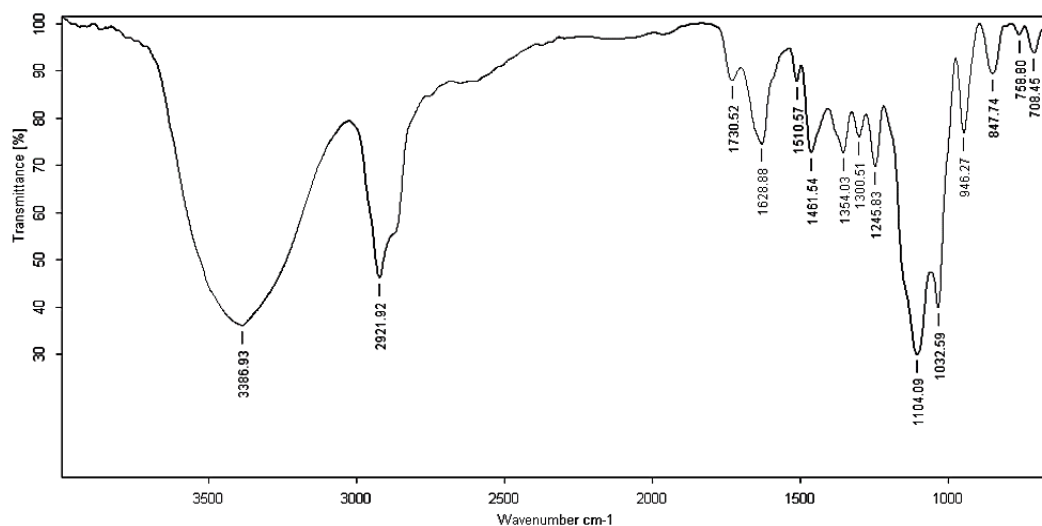
In viscosity testing, a 1% w/v PJFM solution with acetone precipitation demonstrated higher viscosity ( $12.33 \pm 0.17$  MPas) compared to the ethanol-precipitated counterpart. This multifaceted analysis underscores the diverse characteristics and potential applications of PJFM in pharmaceutical, food, and industrial settings. All these values were made in triplicates and depicted in Table 1.

**Table 1: Physical, chemical, and flow properties of *P. juliflora* fruit mucilage**

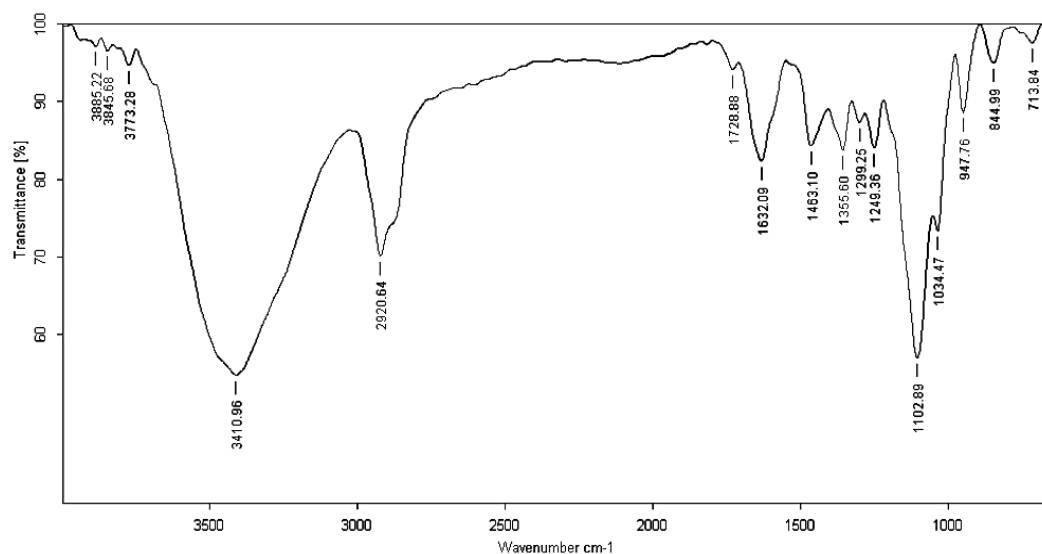
Parameter	<i>P. juliflora</i> mucilage	
	Acetone precipitated	95% Ethanol precipitated
Ruthenium red solution added	Pink coloration	Pink coloration
<b>Organoleptic assets</b>		
Colour	Brownish-yellow	Light brown
Odour	Odorless	Odorless
Taste	Tasteless	Tasteless
Texture	Rough	Rough
<b>Physical assets</b>		
<b>SOLUBILITY</b>		
Acetone, dichloromethane, diethyl ether, ethanol, and methanol	Insoluble	Insoluble
Water	Yellowish-brown sticky liquid	Yellowish- brown sticky liquid
Yield /100g (g)	$52.36 \pm 0.11$	$54.65 \pm 0.04$
MPS ( $\mu\text{m}$ )	$189.75 \pm 5.6$	$196.12 \pm 6.6$
LOD (%)	$2.11 \pm 0.02$	$2.09 \pm 0.01$

SI (%)	49.04±2.51	52.28±0.95
MP (° C)	135±3.94 and charred	139.6±4.55 and charred
pH	7.2±0.15	7.6±0.37
<b>CHEMICAL CHARACTERISTICS</b>		
Steroids (Liebermann – burchard test)	Absent	Absent
Saponins (foam test)	Absent	Absent
Flavonoids (Shinoda test, Zinc/HCl reduction test)	Absent	Absent
Tannins/ Phenols (Ferric chloride test, Gelatin test)	Absent	Absent
Foreign matter (%)	NMT 0.1	MT 0.1
Heavy metals-Lead (PPM)	<15PPM	<15PPM
Arsenic (PPM)	<1	<1
<b>Flow assets</b>		
AR (°)	29.85±0.08	31.54±0.14
BD	0.856±0.07	0.682±0.05
TD	0.894±0.05	0.7.4±0.07
CI	4.251±0.06	3.125±0.04
HR	1.044±0.01	1.0322±0.02
<b>Rheological assets</b>		
Viscosity (mPas)	9.77±0.35	8.72±0.42
Values in mean ±S.D; n=3; NMT-Not More Than; PPM: parts Per Million; MPS: mean Particle Size; LOD: Loss on Drying; SI: Swelling Index; MP: Melting point; AR: Angle of repose; BD: Bulk density; TD: Tapped density; CI: Carr's index; HR: Hausner ratio		

The FTIR spectrum of PJFM exhibited distinct and well-defined peaks, indicative of the mucilage's high purity. The specific values and potential bonds identified in the FTIR spectrum were depicted in Figures 5 and 6.



**Fig. 5: FTIR spectrum of PJFM (Acetone precipitated)**



**Fig. 6: FTIR spectrum of PJFM (Ethanol precipitated)**



## CONCLUSIONS

In conclusion, the findings of this study highlight the favorable physicochemical attributes of *Prosopis juliflora* fruit mucilage (PJFM), affirming its potential utility across diverse pharmaceutical applications. The mucilage's exceptional flow properties, as evidenced by a low angle of repose and favorable bulk and tapped densities, position it as a promising viscosity-modifying agent for liquid oral formulations. Moreover, its suitability as an excipient in tablet formulations underscores its versatility in solid dosage forms, offering potential benefits in terms of binding and disintegration properties. The study also indicates that PJFM can serve as a matrix-forming material in the development of various pharmaceutical dosage forms. The distinct characteristics of the mucilage, particularly when precipitated with acetone, contribute to its superior excipient properties compared to the ethanol-precipitated variant. This difference in excipient quality suggests that the choice of precipitation method significantly influences the mucilage's performance as a pharmaceutical aid. These revelations not only underscore the potential applications of PJFM in pharmaceutical formulations but also emphasize the importance of selecting appropriate extraction and precipitation techniques to optimize its excipient properties. The study paves the way for further exploration and development of PJFM as a valuable and versatile component in the pharmaceutical industry.

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