Compositional analysis and physicochemical evaluation of date palm (Phoenix dactylifera L.) mucilage for medicinal purposes

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Objectives: Date palm (Phoenix dactylifera) mucilage obtained from its dried fruits was evaluated to check the proximate composition and physicochemical properties.

Methods: Commercially available date palm mucilage was precipitated using ethanol. Both (crude and purified) mucilage samples were subjected for proximate, physicochemical, biochemical and antioxidant activity using standard experimental protocols. Elemental analysis of crude date palm mucilage was also performed using LIBS.

Results: Ethanol was used to purify the mucilage (58.4% yield). Proximate analysis was carried out on crude and purified mucilages showing crude fat, crude protein, crude fiber, total carbohydrates, nitrogen free extract and total energy in purified mucilage were more than the crude mucilage. Moisture and ash contents were found more in crude mucilage than the purified mucilage. Laser introduced breakdown spectroscopy (LIBS) detected Zn, Mg, Mn, K, Na, Cu, Fe and Ca metals as components of mucilage.

Biochemical profiling indicated that crude and purified mucilages have proteins, protease, superoxide dismutase, catalase, peroxidase, amylase, ascorbate peroxidase, free amino acids, total soluble sugars, reducing sugars, non-reducing sugars, total anthocyanin, free anthocyanin, total flavonoid contents and total phenolic contents.

Conclusion: The study shows that date palm mucilage could be potentially used as pharmaceutical and medicinal ingredient due to presence of bioactive compounds and its physicochemical properties.

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1. Introduction

Mucilage is a morpho-physiological product of plants having carbohydrates, proteins and small amounts of organic acids (Andrade et al., 2020). Mucilages have nutraceutical and pharmaceutical importance as an additive and binder (Shahid et al., 2020). Mucilage and gums are biodegradable natural materials and this property makes them important for the use in drug delivery system (Prajapati et al., 2013). The extraction and characterization of polysaccharide gums is an essential step in establishing their suitability as pharmaceutical excipients (Nep and Conway, 2010). Mucilage has structurally diverse biological molecules with wide range of physicochemical properties that makes the mucilage an important constituent in the pharmaceutical industry (Tietgen and Walden, 2013), food industry and medicines (Prajapati et al., 2013). However, uncontrolled rate of hydration, viscosity changes upon storage, uneven thickening ability and microbial contamination are some of the common drawbacks of natural materials (Jani et al., 2009) which highlight the need for modifications (Tekade and Chaudhari, 2013) either by physical methods (e.g. exposure to heat via microwave, steam, UV light and gamma rays) or chemical methods (e.g. including grafting and hydrolysis) to form these materials tailor-made to give specific properties and to compete the synthetic pharmaceutical excipients (Malviya et al., 2020). Ora-pro-nobis (Pereskia aculeata), fenugreek (Trigonella foenum-
graeum), okra (Abelmoschus esculentus), yam (Dioscorea alata) and many other mucilages from plant origin have been studied with least research for commercial scale (Faccio et al., 2015; Andrade et al., 2017).

Different plant organs have high concentrations of mucilages (Galloway et al., 2020). Date palm (Phoenix dactylifera L.) belongs to Arecaceae family (Mallhi et al., 2014). Arecaceae family has almost 200 genera and 2500 species (Jain 2012). Along with the nutraceutical importance, date palm has antimicrobial, antioxidant, anti-inflammatory, anti-mutagenic, anti-cancer, gastro- and hepato-protective activities (Biglari et al., 2009; Khalid et al., 2017). Fruit is the rich source of mucilage. High concentration of hydroxyl groups in mucilage results in the water binding and retention capacities (Hosseini and Nabid, 2020). Odor, color, taste, solubility, pH, swelling index and viscosity are important criteria for its commercial uses (Haruna et al., 2016).

Nutraceutical and pharmaceutical effects of date palm fruit are well known (Maqsood et al., 2020) but exposure of biochemical and physicochemical analyses to the date palm mucilage are on the way to research. It was hypothesized that date palm mucilage may be a good source of medicinal and pharmaceutical ingredient so the present study was designed to check the biochemical and physicochemical profile of the date palm mucilage. Objective of this study was to determine morpho-physiological attributes, phytochemicals and properties of date palm mucilage for its uses as pharmaceutical and medicinal ingredient.

2. Materials and methods
2.1. Raw material and chemicals

Date palm mucilage was purchased from herbal medicine seller (Bara Dawa Khana) located in Karkhana Bazar, Faisalabad, and identified from the Department of Botany, GCUF and kept in air-tight jar for further analysis. High quality analytical grade chemicals were purchased from Sigma-Aldrich (USA), Fluka (USA), BB1 (UK), Oxoid (UK), Merck (Germany), Pharmacia and ICN.

2.2. Purification and extraction of mucilage

Total, 500 g date palm mucilage was cut into pieces and soaked in four liters of distilled water for 36 h for dissolution. Superfluous materials were filtered using muslin cloth and filtrate was treated with 90% v/v ethanol to precipitate purified mucilage. For preservation, the purified mucilage was washed with diethyl ether and let it dry in hot air oven at 40 °C for 18 h (Akin-Ajani et al., 2018). Dried and purified date palm mucilage was minced mechanically to make it fine powder which was also kept in air-tight jar for further use. The percentage yield was calculated by the following formula (Ameh, 2013):

\[
\text{Percentage yield} = \frac{\text{weight of purified mucilage}}{\text{weight of soaked mucilage}} \times 100
\]

Organoleptic properties like smell, color, taste (Morris, 1994), solubility index and viscosity were also observed (Chandrappa et al., 2020).

2.3. Proximate analysis

Crude and purified mucilages were used for the proximate analysis using AOAC, 1990 protocols with certain modifications. Moisture, crude fat, crude protein, crude fiber and ash contents were estimated. Total carbohydrates (AOAC 2002), nitrogen free extract (NFE) and total energy were calculated by the following equations:

Total carbohydrates = 100 – (g moisture + g protein + g fat + g ash)

\[
\%\text{NFE} = 100 - (\% \text{ crude proteins} + \% \text{ crude fiber} + \%\text{ crude fat} + \%\text{ crude ash})
\]

Energy (kcal) = 4 x (g protein + g carbohydrate + 9 x (g lipid)

(Akl et al., 2020; Antigo et al., 2020; Gandji et al., 2019; Rostami and Esfahani 2019; Haiiu, 2018; Olkeh et al., 2013; Galla and Dubasi, 2010; Barros et al., 2007; Satchithanandam et al., 2001).

2.4. Elemental analysis of date palm mucilage

The crude date palm mucilage was analyzed by elemental analyzer. Laser introduced breakdown spectroscopy (LIBS) was performed for micro and macro elemental analysis of date palm mucilage (Nasr et al., 2018).

2.5. Biochemical profiling

Biochemical profiling of crude and purified date palm mucilage was carried out to find out protein contents and enzymes including proteases, superoxide dismutase (SOD), catalase (CAT) and peroxidase (POD), amylase and ascorbate peroxidase (APX). Total soluble sugars, reducing and non-reducing sugars were determined using standardized protocols (Kielkopf et al., 2020; Mæ hre et al., 2018; Naqvi et al., 2011; Varavinit et al., 2002; Sadasivam and Manickam 1992; Asada, 1987; Giannopolitis and Ries, 1977; Chance and Maehly, 1955).

2.6. Anti-oxidant activity

Antioxidants of crude and purified date palm mucilage were estimated by analyzing the anthocyanin, total flavonoid contents (TFC), and total phenolic contents (TPC) (Solomon et al., 2006; Karadeniz et al., 2005; Julkunen-Tiitto 1985).

2.7. Statistical analysis

The results of experimental work have been expressed in terms of mean ± SEM.

3. Results
3.1. Physical parameters of date palm mucilages

Percentage yield of date palm mucilage reviled that crude date palm has impurities. Precipitate of purified date palm is only obtained using 90 % v/v ethanol. The percentage yield was found to be 58.4%. The color, smell, taste, physical appearance, solubility, pH, swelling index and viscosity have been summarized in Table 1. The results of physical properties (presented in table 1) showed that dark brown color of crude date palm mucilage changes to light brown when it gets purified and taste also becomes sweeter. This change might be due to the removal of impurities. The physical appearance also changes from hard and spongy to crystalline and shiny (Fig. 1).

Mucilage (crude and purified) when dissolved in different solvents, solubility of purified mucilage was greater in both hot and cold water (7.12 ± 0.011% and 5.92 ± 0.024%, respectively) as compared to the crude mucilage (6.75 ± 0.012% and 5.2 ± 0.019%, respectively). In ethanol, crude mucilage is slightly soluble (0.25 ± 0.018%) and purified mucilage is insoluble. Acetone and chloroform were unable to dissolve both crude and purified mucilages indicating that date palm mucilage has polar (hydrophilic) groups that enable the mucilage to get dissolved in polar solvent (water) as
The increase in the viscosity of date palm mucilage (crude and purified) due to pH change tends to increase at random. Variations in mucilage (crude and purified) with pH change. The change in viscosity by the change in concentration of mucilage, temperature of the environment and pH of the medium also affected the viscosity of the solution. Fig. 2 discloses the change in viscosity by the change in concentration of mucilage in the solution. From the plot it is evident that viscosity of date palm mucilage (both crude and purified) tends to increase as there is increase in the concentration of mucilage. Fig. 3 shows the graphical representation of variations in viscosity of date palm mucilage (crude and purified) with pH change. The change in viscosity due to pH change tends to increase at random. Variations in the viscosity of date palm mucilage (crude and purified) due to temperature change has been expressed in Fig. 4. The increase in temperature leads to decrease in the viscosity of mucilage.

### Table 1

| Sr. No. | Property       | Crude                          | Purified                      |
|---------|----------------|-------------------------------|-------------------------------|
| 1       | Colour         | Dark brown                    | Light brown                  |
| 2       | Smell          | Aromatic                      | Aromatic                     |
| 3       | Taste          | Sweet Blandy taste            | Sweetish                     |
| 4       | Physical appearance | Hard and spongy          | Crystalline and shiny         |
| 5       | Solubility     |                               |                               |
| 6       | Cold water     | 5.2 ± 0.019% (w/v)            | 5.92 ± 0.024% (w/v)           |
| 7       | Hot water      | 6.75 ± 0.012% (w/v)           | 7.12 ± 0.011% (w/v)           |
| 8       | Ethanol        | 0.25 ± 0.018% (w/v)           | 0.00                          |
| 9       | pH             | 6.64                          | 6.72                          |
| 10      | Distilled water| 1.90 ± 0.022                  | 1.99 ± 0.037                  |
| 11      | Phosphate buffer| 2.1 ± 0.020                  | 2.25 ± 0.014                  |
| 12      | 0.1 N HCl      | 1.22 ± 0.017                  | 1.15 ± 0.013                  |
| 13      | Viscosity      | 8.3 ± 0.154                   | 9.1 ± 0.062                   |

### 3.2. Proximate analysis

The results of proximate analysis have been summarized in Table 2. Analysis indicated that crude fats in purified date palm mucilage were more (3.6 ± 0.058%) than crude date palm mucilage (1.6 ± 0.123%). The moisture contents in crude mucilage (13.3 ± 0.035 %) were more than the purified mucilage (8.3 ± 0.080%). Crude proteins were found in higher concentration in purified mucilage (1.95 ± 0.018%) than that of crude mucilage (1.55 ± 0.025%). Ash contents similar to moisture contents were more in crude mucilage (7.33 ± 0.015%) as compared to purified mucilage (2.66 ± 0.037%).

Crude fibers and total carbohydrates were also in extra amount in purified mucilage (82.6 ± 0.093% and 83.49 ± 0.014%, respectively) than the amount present in crude mucilage (78.3 ± 0.097% and 82.6 ± 0.093%, respectively) (Table 2). Nitrogen free extract as calculated on difference bases were found more in crude mucilage (88.78 ± 0.019%) as compared to purified mucilage (90.81 ± 0.014%). As purified date palm mucilage has more crude fats, crude proteins and total carbohydrates than the crude mucilage so total energy produced by purified mucilage (374.16 ± 0.037 kcal/g) was also more than the energy produced by crude mucilage (325.48 ± 0.051 kcal/g).

### 3.3. LIBS analysis

Date palm mucilage was subjected to laser introduced breakdown spectroscopy (LIBS) for the detection of elements. The spectrum obtained by the analysis of Phoenix mucilage is given in Fig. 5. The spectrum indicated that date palm mucilage had Zn (I), Mg (I), Mn (II), K (II), Na (II), Cu (II), Fe (II) and Ca (II).

### 3.4. Biochemical profiling

Table 3 summarizes the biochemical profiling of crude and purified date palm mucilage. The result indicated that enzymes were present in relatively small amount in date palm mucilage (crude and purified) as compared to the protein contents. The protein contents in purified mucilage (405.17 ± 0.037) were more than the crude mucilage (380.49 ± 0.044). Crude and purified mucilages had higher amounts of amylases (58 ± 0.033 and 68.8 ± 0.141, respectively) and proteases (60.9 ± 0.070 and 63.15 ± 0.061, respectively) as compared to other enzymes present in date palm mucilage. Peroxidases were present in the least amount among others but its concentration in crude date palm mucilage (28.71 ± 0.01) is relatively more than in purified date palm mucilage (13.2 ± 0.006). Superoxide dismutase (SOD) and catalase (CAT) were also found during biochemical profiling of crude (37.55 ± 0.032 and 43.55 ± 0.032, respectively) and purified mucilage (59.53 ± 0.014 and 44.40 ± 0.033, respectively). Ascorbate peroxidase was found to have same concentration in crude (34.94 ± 0.012) and purified (34.92 ± 0.021) mucilages. Free amino acids were also found in crude (1.9 ± 0.004) and purified (0.779 ± 0.001) mucilages.

### 3.5. Soluble sugars

Soluble sugars were also present in crude (206.78 ± 0.012) and purified (210.56 ± 0.037) date palm mucilage. Among soluble sugars, reducing sugars in crude date palm mucilage (47.54 ± 0.035) and purified date palm mucilage (46.79 ± 0.021) were present in relatively small amount than non-reducing sugars present in crude (159.24 ± 0.01) and purified (163.76 ± 0.024) date palm mucilage (Table 3).

### 3.6. Antioxidants

Antioxidants (i.e. anthocyanin, TFC and TPC) were also found during the biochemical profiling of crude and purified date palm mucilage (Table 3). TPC was found in greatest amount in both the crude (14.69 ± 0.018) and purified (18.04 ± 0.031) date palm mucilage. TFC was more in crude (12.23 ± 0.008) date palm mucilage than purified (9.32 ± 0.024) date palm mucilage. Anthocyanin (0.28 ± 0.002 and 0.33 ± 0.002) was present in small amount in

![Fig. 1. Selected crude (a) and purified (b) date palm mucilage.](image)
date palm mucilage (crude and purified, respectively). The results also indicated that most of the anthocyanins were present in free-state in crude (0.25 ± 0.003) and purified (0.28 ± 0.002) date palm mucilage.

4. Discussion

The nutritional value of mucilage is exposed by the presence of crude proteins (Soukoulis et al., 2018), crude fats (Al-Samarai et al., 2018), ash contents (Ismail, 2017). In this study, the percentage yield was found to be 58.4% which was lesser than the yield of date palm mucilage (73.75%) determined by Shahid et al., 2020. Mucilage yield (3.6–9.4%) in linseed (Linum usitatissimum L.) was recorded for its composition and physicochemical properties (Fedeniuk and Biliaderis, 1994). Mucilage yield from different plants is dependent upon extraction time, process and solvent system used (Oikeh et al., 2013). It was revealed that decrease in yield might be due to the purification steps (Keshani-Dokht et al., 2018). Properties of mucilage(s) are reflective of their significance, functional attributes and constituents. For instance, our recorded pH of purified date palm mucilage (6.72) shows that it would be more suitable to use because pH nearer to neutral pH indicates that date palm mucilage would be less irritating to mucosal membrane and formulations formed by date palm mucilage at this pH would be comparatively more stable (Haile et al., 2020). Similarly, pH of 1% w/v aqueous solution of crude (6.64) and purified (6.72) mucilage is slightly acidic indicating the presence of cations (Murray et al., 2006).

### Table 2

| Sr. No | Phoenix dactylifera | Results          |
|--------|---------------------|-----------------|
|        |                     | Crude Gum       | Purified Gum   |
| 1.     | Crude fat (%)       | 1.6 ± 0.123     | 3.6 ± 0.058    |
| 2.     | Moisture content (%)| 13.3 ± 0.035    | 8.3 ± 0.080    |
| 3.     | Crude protein (%)   | 1.55 ± 0.025    | 1.95 ± 0.018   |
| 4.     | Ash content (%)     | 7.33 ± 0.015    | 2.66 ± 0.037   |
| 5.     | Crude fiber (%)     | 78.3 ± 0.097    | 82.6 ± 0.093   |
| 6.     | Total carbohydrates | 76.22 ± 0.010   | 83.49 ± 0.014  |
| 7.     | Nitrogen free extract (%) | 88.78 ± 0.019 | 90.81 ± 0.014 |
| 8.     | Total energy (kcal/g) (%) | 325.48 ± 0.051 | 374.16 ± 0.037 |

**Fig. 2.** Variation of viscosity with concentration.

**Fig. 3.** Variation of viscosity with pH.

**Fig. 4.** Variation of viscosity with change in temperature.

**Table 2**

| Sr. No | Phoenix dactylifera | Results | Crude Gum | Purified Gum |
|--------|---------------------|---------|-----------|--------------|
|        |                     |         | Crude Gum | Purified Gum |
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| 8.     | Total energy (kcal/g) (%) | 325.48 ± 0.051 | 374.16 ± 0.037 |         |
Different gums and mucilages have different concentrations of crude proteins (Capitani et al., 2015). Higher amount of protein in date palm mucilage indicates its possible use as protein supplement (Akin-Ajani, 2018). Purification process of date palm mucilage boosts the crude protein contents in purified date palm (1.95 ± 0.018%) from crude date palm (1.55 ± 0.025%). Presence of crude proteins in date palm mucilage (2.19 ± 0.011%) was encouraged by Shahid et al. (2020).

The residue, inorganic in nature, obtained after complete oxidation of organic matter is the ash contents and it discloses the nutritional values (Ismail, 2017), handling and adulteration of drugs. Ash contents in natural substances ranged 0.03–0.07% by weight, although the oily substances may have higher values (Sarkar, 2016). In our experimental work, the ash contents in crude date palm mucilage were 7.33 ± 0.015% while in purified date palm mucilage ash contents were reduced to 2.66 ± 0.037%. Our study on ash contents in date palm mucilage was supported by Shahid et al., 2020 where they found 2.95 ± 0.05% ash contents in date palm mucilage.

The insoluble residue obtained after acidic hydrolysis is the crude fiber that is rich in cellulose and insoluble lignin (Chien et al., 2017). While conducting the study, the crude fiber in crude date palm mucilage was found to be 78.3 ± 0.097% while in purified date palm mucilage it was 82.6 ± 0.093%. In another study by Shahid et al. (2020), the crude fibers were found to be 98.7 ± 0.060% in date palm mucilage. Nutritional value is determined by the presence of crude fiber. Guar gum has relatively low crude fibers (2–3%) (Chudzikowski, 1971). Ndouenkeu et al. (1997) found that mucilage obtained from different parts have different concentrations of crude fibers. The higher fiber contents in natural gums and mucilages will lead to the increase in colonic fermentability and holding capacity (for water and oil) as well as lower the phytic acid contents and caloric value (Figueroa et al., 2005).

Food material contain multiple nutrients, including sugars, dietary fibers and starch (Berrios et al., 2010). These all are the types of carbohydrates and have their significant roles in the energy provided by the food material (Tondt et al., 2020). During the research it has found that total carbohydrate in crude date palm mucilage was 76.22 ± 0.011% while amount of carbohydrates increased to 83.49 ± 0.014% when date palm mucilage was purified. In another study by Shahid et al., 2020, total carbohydrates in date palm mucilage were found to be 85.16 ± 0.036%.

The extract of gum and mucilage that depicts total sugars and starch in it could be the nitrogen free extract of mucilage (Fraps, 1931). It is the only part of proximate analysis that is not determined analytically (Detmann and Valadares Filho, 2010). During the study on date palm mucilage, it was found that crude date palm had 88.78 ± 0.0119% nitrogen free extracts while purified date palm mucilage had 90.81 ± 0.0114% nitrogen free extract. During research by Munir et al., 2016 on A. modesta and D. sissoo gums nitrogen free extracts were found to be 94.48% and 92.81%, respectively.

The presence of useful metals in date palm mucilage. Zinc, magnesium, potassium, sodium, copper and calcium were found in measurable quantities while some other metals may also be present in traces as explored by Akin-Ajani et al. (2019).

Rheological measurements gave useful, predictive and behavioral information about the materials which helped to understand the aging phenomenon and formulation changes (Ebewele, 2000). Materials always give their responses to the mechanical forces and rheology is the study that deals with such properties of the materials. In solids this involves in the study of plasticity and elasticity while in liquids and fluids it involves in the measurement of internal friction. In natural and synthetic polymers, among the other commercial and analytical parameters, viscosity is an impor-
tant parameter as size and shape may affect the viscosity (Anderson and Dea, 1967). Concentration changes, pH of solvent and temperature of environment affect the viscosity of the system. Viscosity of date palm mucilage (both crude and purified) tends to increase as there is increase in the concentration of mucilage. The similar results were found by Jefferies et al. (1977) and Ashton et al. (1975) when they studied the Ghatti and Albizia gums. Ameh (2013) also declared similar type of results when he discussed the rheological properties of F. glumosa gum. The possible increase in the viscosity could be due to the increase in the strength of molecule–molecule interaction and reduction in molecule-solvent interaction. Viscosity of date palm mucilage increases at random with the increase in pH and this affects the emulsifying property of gums and mucilages (Calvo et al., 1998). The increase in temperature leads to decrease the viscosity of mucilage. In order to verify the onset of degradation or conformational transitions during heating, the viscosity of the gums was again measured as cooling proceeded. The tests did not indicate any difference between the two sets of data suggesting that there was no degradation or conformational transition when the gums were heated (De Paula et al., 2011).

Biochemical profiling of date palm mucilage (crude and purified) indicated the presence of primary and secondary metabolites. The medicinal and pharmaceutical effects of plant residue are due to these metabolites (Kayani et al., 2015). Total soluble sugars (crude and purified). These antioxidants may involve in protection of cells from free radical damage (Omoregie and Osagie, 2012) or may involve in reducing thrombosis, cholesterol level (Talapata and Talapata, 2015) or may help to inhibit tumor production during cancer (Devasagayam et al., 2004) or may stimulate hormones and antibacterial activity (Mathew et al., 2012). Presence of enzyme shows the biocatalytic activity (Arsalan and Younus, 2018) of date palm mucilage.

5. Conclusion

The physiochemical and biochemical profiling of date palm mucilage (crude and purified) showed that this mucilage is biologically important and have its medicinal and pharmaceutical effects. Minor changes in the biochemical profile are produced during purification of crude date palm mucilage but the purified mucilage has better proximate composition. The authors, therefore, recommended further studies for the quantification, isolation and characterization of pure compounds from the Phoenix dactylifera mucilage, as well as, want to check its use as pharmaceutical excipient.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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