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Extraction and Characterization of Almond (*Prunus sulcis*) Gum as Pharmaceutical Excipient

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Abstract: This study involved extraction and characterization of almond (*Prunus dulcis)* gum as pharmaceutical excipient. The gum obtained from almond (*Prunus dulcis)* was extracted by using distilled water and precipitated with ethyl alcohol. Yield was calculated to evaluate the efficacy of process. The product was screened for presence of carbohydrate, protein, fat, starch, reducing and non-reducing sugars, alkaloids, glycosides (Cardiac glycosides, anthraquinone, saponins), steroids, tannins, phenolic compounds micromeritic properties, swelling index, flow behaviour, surface tension and viscosity were also studied.Result revealed that using water based extraction method, the percentage yield was found to be 8.89%. The result showed that extracted almond gum exhibited excellent flow properties. It had good swelling index (87.44 ± 0.310%). The pH was found to be 5.25 ± 0.813 and surface tension of the 0.01% gum solution was found to be 73.50 ± 0.078 dynes/cm. The ash values such as total ash, acid insoluble ash, water soluble ash was found to be 15.9%, 0.57% and 3% respectively, loss on drying was 4.85%. Extracted gum was soluble in warm water and insoluble in organic solvents. The scanning electron micrograph (SEM) revealed rough and irregular particles of isolated polymer. In conclusion, results of evaluated properties showed that almond derived gum can be used as pharmaceutical excipient to formulate solid oral dosage form. It has acceptable pH and organoleptic properties, so can be easily used to formulate various dosage form.

Key words: Almond · Gum · Pharmaceutical Excipient · Natural Polymer

INTRODUCTION

Polymer can be defined as a large molecule that built up from repeated small chemical units which linked together. The conversion of monomers to polymer leads to rearrangement of electrons. Polymers also have various arrangements of molecules to form linear, branched or cross-linked structures. Polymer can be natural polymer or synthetic polymer which is synthesized from various chemicals. Since polymer can be easily degraded and lost their effectiveness in turbulent flow in short duration even it is high molecular weight (>105) [1]. Natural polymers are generally obtained from plants. They are high molecular weight; water soluble polymers made up of monosaccharide unit and joined by glucosidic bond [1, 2]. Gummy exudate of natural polymers such as protein, enzyme, muscle, fibre and polysaccharide which have been used to formulate the various pharmaceutical products [2, 3]. Gum has obtained from hydrocolloids of plant and can be classified into two groups' i.e. anionic and non ionic polysaccharides. Hence by modification, gum can alter their physicochemical properties [4, 5]. It is a metabolised product which is intracellularly formed without injury to the plant [5]. Gums are readily soluble in water while mucilage forms slimy mass in the presence of water. Gum and mucilage are translucent, amorphous substances which are produced by plants as a protective during injury. Gum, cellulose, mucilage and resins are distinguished by the condensation of hexane and pentose [6-8]. Both can be obtained from middle lamella as in algae, cell wall as in the seed epidermis as well as endodermis, some secretary cells as in squill and also present in the

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schizogenous sacs [9,10]. Gum has various pharmaceutical applications such as suspending agent for insoluble solid component in mixture, emulsifying agent for resin oil and adhesive in troche masses and pill. They can be used as a thickener, emulsifier, sweetener, viscosity enhancer in pharmaceutical preparations [11-13]. The natural polymers have been used in the households, agriculture, food industries and in packaging and it help in decreasing the environmental pollution and resulting in disposal in landfills [14,15]. Almond gum is obtained from the tree *Prunusdulcis* which is a water soluble gum, extrudes from the wounds on almond trees. The constitution of almond gum includes aldobionic acid, L-arabinose, L-galactose, D-mannose etc. It contains different components which act as emulsifier, thickener, suspending agent, adhesive, glazing agent and stabilizer [16, 17].

MATERIALS AND METHODS

Extraction Procedure: Almond (*Prunus dulcis)* was obtained from local market of Greater Noida, India. Collected gum was carefully washed and dried under shade for 24 h, further dried at 30–40°C until constant weight was obtained. Size was reduced through grinder. Powdered fruit passed through sieve no. #22 and was stored in air tight container for further use. Extraction of gum included three steps[18].

Step 1: Extraction of Gum: Powdered fruit was put in 1000mL beaker containing 500ml of distilled water, then heated and stirred continuously at 60°C for approximately 4h. Concentrated solution was filtered through muslin cloth and cooled at 4°C-6°C [19, 20].

Step 2: Isolation of Gum: The extract was poured into ethyl alcohol, thus precipitation of the gum occurred. This allowed the filtration through muslin cloth. Washed with ethyl alcohol and precipitated gum has collected through muslin cloth by filtration. Gum was further dried to constant weight at 35–45°C in hot air oven. Hard gum cake was grinded and sieved through sieve # 22 [21].

Step 3: Removal of Oil from Almond Gum: Extracted almond gum was kept in petroleum ether for 4-5h. Filtration of above petroleum ether solution using muslin cloth or Whatman filter paper was done and filtered gum was dried in oven at 45°C. Dried polymer was powdered and stored in a dessicator for further study [22].

Physicochemical Characterization of Almond Gum: Aqueous extract was mixed with Molish's reagent followed by addition of sulphuric acid. The violet colour ring appeared at junction, showing presence of carbohydrates [20, 23]

Determination of Purity of Almond Gum: To measure the purity of extracted gum, tests for alkaloids, proteins, gum, fats, tannins and amino acids were performed [18, 20].

Organoleptic Evaluation of Isolated Gum: Isolated gum was characterized for organoleptic properties such as colour, odour, taste, fracture and texture [20].

Ash Values: Ash values such as total ash, acid insoluble ash and water- soluble ash were determined using Equations 1, 2, 3 respectively [23].

$$Total\ ash\ value = \frac{weight\ of\ ash}{weight\ of\ ploymer} \times 100 \quad (1)$$

$$Acidin\ solubleash = \frac{weight\ of\ acidib\ solubleash}{weight\ of\ dried\ powder} \times 100 \quad (2)$$

$$Water\ solubleash = \frac{weight\ of\ water\ solubleash}{weight\ of\ dried\ powder} \times 100 \quad (3)$$

Solubility Behaviour: Dry gum powder was shaken with different solvents and further solubility was determined [23]

pH of Gum: The gum was weighed and dissolved in water separately to get a 1%w/v solution. The pH of solution was determined using digital pH meter [24].

Swelling Index: Swelling index were calculated as per equation 4 [20].

$$Swelling\ index = \frac{final\ volume - initial\ volume}{final\ volume} \times 100 \quad (4)$$

Surface Tension: The surface tension of the selected gum was determined by drop weight method, using a stalagmometer [20, 23]. The surface tension of the polymer has been reported to influence the binding quality of the polymer. Surface tension was calculated as per equation 5.

$$\sigma_{solution} = \sigma_{water} = \frac{m(solution)}{m(water)} \quad (5)$$

where,

$\sigma_{solution}$ = Surface tension of solution
σ_{water} = Surface tension of water
M (Solution) = Weight of solution
M (Water) = Weight of water

Viscosity: Viscosity of almond gum was determined by Oswald viscometer as per equation 6.

$$\eta_s = \eta_w \times \frac{t_s \rho_s}{t_w \rho_s} \quad (6)$$

where
η_s = Viscosity of solution
η_w = Viscosity of water
t_s and t_w = Time of solution and water
ρ_s and ρ_w = Density of solution and water

Loss on Drying: One gram of powder was weighed accurately in a weighing bottle and was dried in a hot air oven at 105°C and the weight was checked at intervals of 10min, until a constant weight was obtained. The percentage of weight lost by the powder was calculated using equation 7 [20, 23].

$$Losson\ drying = \frac{initial\ weight - final\ weight}{initial\ wight} \times 100 \quad (7)$$

Bulk Density and Bulkiness: Bulkiness is the inverse of bulk density. Accurately weighed quantity of powder was introduced into a graduated measuring cylinder. The cylinder was fixed on the bulk density apparatus and the volume occupied by the powder was noted. Then, the powder was subjected to tapping in a bulk density apparatus until constant volume was obtained. The final volume (Bulk volume) was noted [22, 24]. Bulk density, tapped density and bulkiness were calculated using equation 8, 9 and 10 respectively.

$$Bulk\ density = \frac{weight\ of\ powder\ blend}{weigth\ of\ apparent\ volume} \quad (8)$$

$$Tapped\ density = \frac{weight\ of\ powder\ blend}{tapped\ volume} \quad (9)$$

$$Bulkiness = \frac{1}{bulk\ density} \quad (10)$$

True Density: Among various methods available for the determination of true density, liquid displacement method is the simplest method and was used in the present study. Acetone was selected as the liquid for displacement, because, gum is insoluble in acetone [23, 25].

Powder Flow Property: Flow characteristics were measured by angle of repose. Same study was repeated here. Using the readings and the formula, the angle of repose was calculated using equation 11 [23, 25].

$$Tan\theta = \frac{h}{r} \quad (11)$$

where,
θ = Angle of repose
h = Height of pile
r = Radius of pile

Powder Compressibility (Carr's Consolidation Index): This property is also known as compressibility. Finely powdered gum (5 g) was transferred into a measuring cylinder and calculations were done using bulk density apparatus [21, 24].

$$Carr'sindex = \frac{Tapped\ density - Bulk\ density}{Tapped\ density} \times 100 \quad (12)$$

$$Hausner'sratio = \frac{Tapped\ density}{Bulk\ density} \quad (13)$$

Particle Size Analysis: The particle size was determined by using microscope, which was calculated as per equation 15, 16.

$$Size\ findividual\ particle = No.\ of\ individualineyepiece \times calibration\ factor \quad (15)$$

$$Calibration\ factor = \frac{stagereding \times 0.01}{ocularreading} \quad (16)$$

RESULTS AND DISCUSSION

After extraction and further precipitation by ethyl alcohol, the yield of gum was found to be 3.89%. The purified polysaccharide stained pink with ruthenium red solution which confirmed it as gum. For determination of purity of the isolated almond gum, different phytochemical tests were carried out including Molish test, Benedict's test, Barfoed's test and Selivenoff's test. This showed that almond gum has failed the given test. Proteins, starch, reducing and non-reducing sugars, alkaloids, glycosides (Cardiac glycosides, anthraquinone, saponins), steroids, tannins and phenolic compounds were found to be absent, which confirmed the purity of isolated gum. This can be considered as proof for purity of the isolated gum as depicted in Table 1.

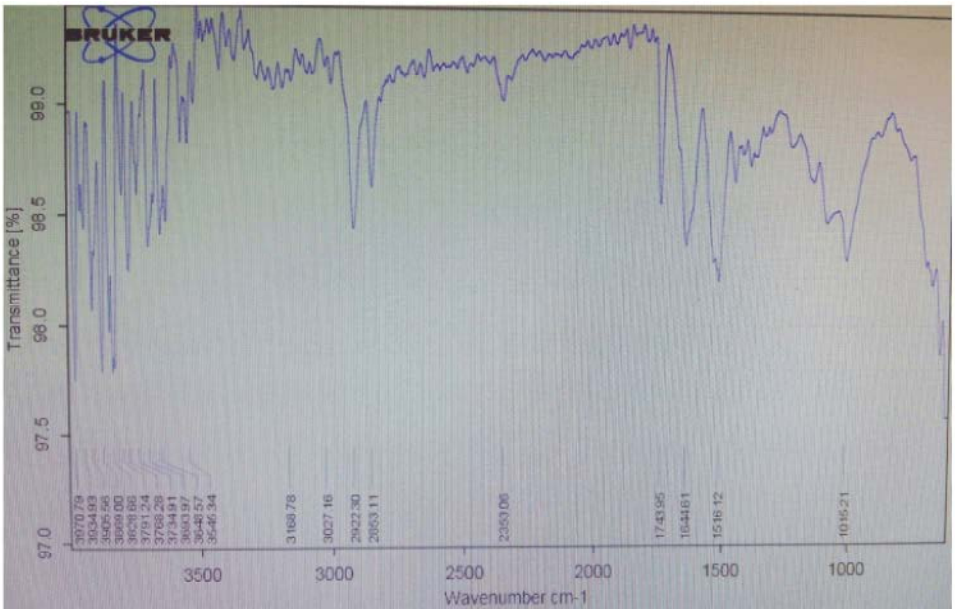

Fig. 1: IR spectra of almond gum

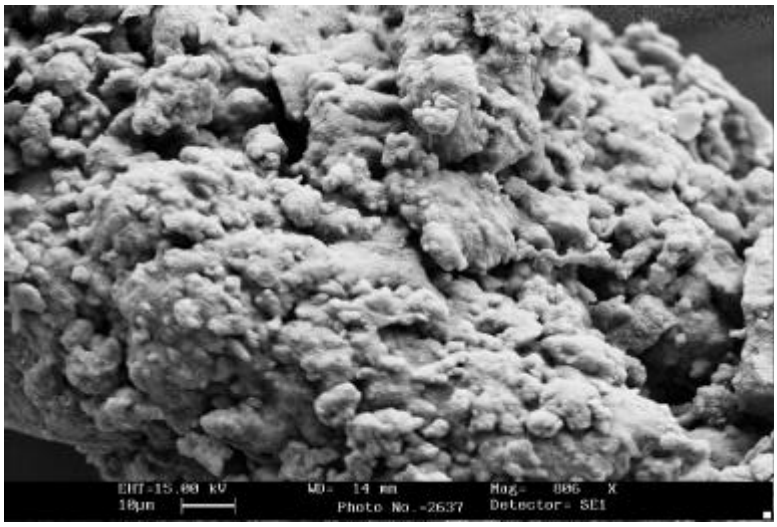

Fig. 2: Scanning electron micrograph of almond gum

Table 1: Determination of isolated gum

S. No.	Test	Present/absent
1.	Carbohydrates	-
2.	Hexose Sugar	-
3.	Monosaccharides	-
4.	Proteins	-
5.	Fats and oils	-
6.	Tannins and Phenolic Compounds	-
7.	Alkaloides	-
8.	Amino acids	-

+ Present, -absent

The pH of the 1% solution of gum was found to be 5.25 ± 0.813, which was slightly acidic. The pH of gum indicated that adjustment of pH might be required in the formulation of oral and buccal drug delivery systems. Surface tension of the 0.01% gum solution was found to be 73.50 ± 0.078 dynes/cm. The lower value of surface tension might allow better penetration and wetting ability of gum dispersion over the powder mass and hence, resulted in formation of better quality of granules.

The result for loss on drying was found to be 4.85%.This indicated that gum was hygroscopic in nature and need to be stored in air-tight containers. In solubility behaviour of almond gum was found to be soluble in warm water, slightly soluble in cold water and insoluble in benzene, ether, chloroform, n-butanol, ethanol, acetone, glycerine, paraffin.

Table 2: Micromeritic study data of gum

S No.	Parameters	Values
1.	Angle of repose (°)	22.08 ± 2.93°
2.	Carr's index (%)	11.6 ± 0.16
3.	Hausner's ratio (%)	1.93 ± 0.070
4.	Tapped density (g/ml)	0.517 ±0.03
5.	Bulk density (g/ml)	0.399±0.003
6.	Bulkiness (ml/g)	2.52±0.02
7.	Mean particle size (μ)	45±1.1

Values with "±"shows standard deviation and all measurement were done in triplicate studies.

Table 3: IR study data of almond gum

S. No.	Functional group	Peak
1.	Carboxylic acid	3168.78
2.	Alkenes	3027.16
3.	Aldehyde	2853.11
4.	Ester	1743.95
5.	Amines	1516.12

Irregular particles size was found to be 45±1μm. Result of almond gum showed that gum was off white colour, odourless, tasteless, rough and irregular in shape. Ash values were calculated to characterize gum; total ash, acid insoluble ash and water soluble ash were found to be 15.9%, 0.57% and 3% respectively. Swelling index of isolated mucilage was found to be 87.44 ± 0.310% indicating that almond gum might have good water uptake capacity. The viscosity of 2.5% solution of gum was found to be 650.66 ± 0.50cp and value of Brookfield rheometer was found to be 301.224pa/sec.

The Carr's consolidation index, Hausner's ratio and angle of repose were found to be 11.6 ± 0.16%, 1.93 ± 0.070 and 22.08 ± 2.93° respectively. The value of Carr's consolidation index indicated that almond gum has excellent flow property. The result of Hausner's ratio and angle of repose indicated excellent flow property of gum powder. Physical characterization of gum was carried out for bulk density and bulkiness, true density, total porosity, powder flow behaviour. The bulkiness value indicated that powder is 'light' in nature. Result obtained in micromeritic characterization of gum was shown in Table 2.

IR spectra showed that almond gum contain carboxylic acid, alkene, ester, aldehyde and amines as shown in Figure 1 and peaks of these groups have shown in Table 3.

The SEM photograph as shown in Figure 2 of the almond (*Prunus dulcis)* gum revealed that the surface of the particles was found to be rough and irregular. Earlier, it has been observed that rough surface of polymer can retard the drug release from the dosage form due to the entrapment of drug in the pores [25, 26].

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Conflict of Interest: Authors have no conflict of interest.

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