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Extraction and Physicochemical Properties of Barijeh (*Ferula galbaniflua*) Gum

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ABSTRACT

Ferula galbaniflua Boiss is one of the natural plants of Iran. Its exudates, Barijeh gum, can be used in food industry. The present study explored the properties of a new gum extracted from tubers of *Ferula galbaniflua*. For this purpose, Barijeh gum was extracted from crude exudates by alcoholic extraction with 90% ethanol. The purity of gum was relatively high and it composed mostly of saccharides. The composition of gum was analyzed using HPLC. The effect of temperature (50, 70, 90°C) and pH (4, 7, 10) on the extraction yield, viscosity and on the purity of extracted gum was investigated. It was found that extraction yield in acidic pH was highest and rising temperature lead to more yield. By the other way, increasing temperature and reducing pH reduced the protein content in extracted gum. Increasing temperature of extraction medium, decreased viscosity of gum suspension. Also, alkaline extraction yield caused less viscosity of gum suspension. Our analysis indicated presence of galactose and arabinose in 6:1 ratio.

Key Words: *Ferula galbaniflua*; Barijeh gum; Extraction; Galactose; HPLC

INTRODUCTION

Ferula galbaniflua Boiss is a perennial plant native to Central Asia, growing in the northern parts of Iran. The *Ferula galbaniflua* is a green annual herb of tribe Peucedaneae, sub-family Apioideae and family Umbelliferrae (Apiaceae) (Zargari, 1989). Among the *Ferula* species present in the area, *Ferula galbaniflua* has been used more than others and it has more ability to produce oleogum resin of higher quality in comparison with other species. One of the sources of pharmaceutical and industrial component is a plant by the name of *Ferula galbaniflua*. This plant has a tick sturdy stem with 0.8 - 3 m height. Stem is glabrous, only leaves are hairy and gray (Safaian & Shokri, 1993).

It is a resistant plant, native in humid mountain and semiarid regions in Iran. Its distribution could be observed mostly at an altitude of 2000 - 4000 m above sea level. It grows in sandy loam. The good quality habitats could be found mostly in north-facing slopes and on calcareous and well-dried soils. The best utilization time is July and August (Parsa, 1984; Ghahreman, 1986).

Barijeh is an air-dried oleogum resin exudation, which is obtained by incising the stems close to the ground. It is generally met with in lumps, consisting of large, irregular masses of brownish color composed of agglutinated tears that have a waxy density, but becomes soft and sticky at a temperature of 35 - 37.7°C. It has a pleasant odor and an acrid taste and diluted alcohol is its best solvent. Its specific gravity is 1/212 g/cm^3 and major compounds are komarins, terpens and glycosidic compounds. The gum exudates were used in medicine by both oral and topical administration. In ancient Iranian medicine the oleogum resin obtained from this plant has been used for stomach pain, chorea, epilepsy and as a wound-healing remedy (Agili-Khorasani, 1991). Nowadays, it is used as a food additive than as a drug. It is used as a flavoring additive in food stuffs such as non-alcoholic beverages and meat products.

Some scientists have taken interest in polysaccharides extracted from Barijeh by alcoholic solution. They identified gum composition by chemical methods. Their analysis indicated presence of galactose, arabinose, rhamnose and uronic acids that galactoronic acid was major component. Also, their study indicated presence of protein, Ca and Mg.

As well as Arabic gum Barijeh gum formed a low-viscosity solution (Jessennne *et al*., 1974). Howlett (1980) provides a clouding agent base comprising lanolin and a gum resin such as Barijeh gum for cloudy beverages (Howlett, 1980).

The present report is the first study on the extraction of gum from *F. galbaniflua*. The aim of the investigation was to isolate and characterize of Barijeh gum.

MATERIALS AND METHODS

Plant materials. The oleogum resin exudates of *Ferula galbaniflua* was collected in northern Iran by cutting up roots from June to July 2005 and kept in refrigerating room. All reagents were of analytical grade. Sulfuric acid, ethanol, glucose and galactoronic acid were purchased from Merck, Germany. Soybean oil was prepared from Sigma, USA. Deionized water was used for preparation of solutions and emulsions.

Gum extraction and purification. In order to extraction of gum from gum-resin, crude sap was stirred with ethanol 90% v/v for 2 h. The ethanolic slurry was filtered through Whatman No. 1 filter paper. The residue was washed with ethanol 90% twice and then it was dried at 40°C for 12 h. After drying, the extracted powder was solubilized in distilled water at three different temperature (50, 70, 90°C) and pH (4, 7, 10) and then filtered through Whatman No. 1 filter paper. The filtrate was concentrated at 40°C in vacuum and dialyzed at cut-off 3500 Da. After filtering, solution was concentrated and dialyzed again in the same condition. Finally, the extracted gum was lyophilized. The effect of temperature and pH on yield and viscosity was studied. Fig. 1 shows analytically the stages of the isolation and purification of Barijeh gum.

Gum characterization. Moisture, crude protein and ash were measured by A.O.A.C. methods (1984). Briefly, moisture was measured in the form of weight decrease after heating the sample at 105°C for 4 h. Crude protein was measured by the method of Kjeldal using 6.25 as the conversion rate of nitrogen to crude protein (Anderson, 1986). Ash was measured as the residue remaining after heating at 600°C for 4 h. Total sugars was measured by the phenol-sulfuric method (Dubois *et al*., 1956). Using glucose (Merck, Darmstadt, Germany) and uronic acid was measured by the method of Blumenkrantz and Hansen (1973) using galactoronic acid (Merck, Darmstadt, Germany) as a standard.

Sodium, potassium, phosphorus, calcium and magnesium contents were analyzed using atomic absorption spectrum method (A O.A.C., 1984).

Sugar analysis by HPLC. The HPLC system used had a K-1001 Pump (Knauer, Germany) equipped with a 20 µL sample loop linked with a EUROKAT H column (8 × 300 mm) (Knauer, Germany) and a refractive index detector. Acetonitril/water (80/20, v/v) was used as the mobile phase. Data evaluation was performed using the peak area and an external standard. The respective sample of 0.05 g was weighed out accurately into stoppered Pyrex tubes and 5 mL of cold sulfuric acid (4% w/w) added to each. The tubes were heated for 4 h in a water bath at 100°C, reweighed and made up to the original weight by adding distilled water. The solutions were neutralized by adding 1 g barium carbonate and left to mix overnight at room temperature. The hydrolysates were filtered (0.45 µm) and analyzed by HPLC.

RESULTS AND DISCUSSION

Effect of extraction method on viscosity. The effect of pH and temperature on the viscosity was significant. There was significant difference ($p < 5\%$) between the viscosities of gum dispersions at the different values of pH studied. The same tendency was observed at each temperature. Decreasing pH increased viscosity of gum dispersions, no significantly difference was observed between acidic and neutral pH but it was more than alkaline extraction significantly (Fig. 2). Increasing temperature of extraction decreased viscosity of gum dispersions and there was significant difference between the viscosities of gum dispersions at the different values of pH studied.

Goycoolea *et al*. (1995) indicated that, in galactomannans, alkaline conditions caused a reduction in viscosity. Propably this is due to a reduction in the weight of the molecules and to the suppression of intermolecular association. Upon carrying out the extraction with acidic water, the formation of hydrogen bonds occurred, increasing the dispersion viscosity. Similarly as occurs in many other gums, the viscosity of the gum extracted by both methods diminished as the temperature increased. Casas *et al.* (2000) reported that the apparent viscosity of guar gum dispersions decreased upon heating, this behaviour being completely reversible.

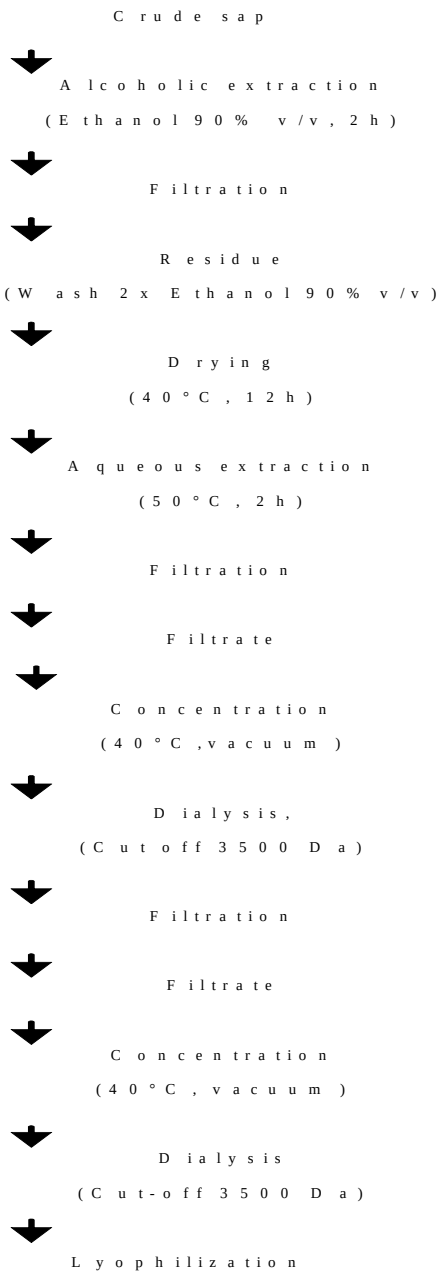
Effect of extraction method on yield. pH and temperature factors had significant effects on extraction yield. Decreasing pH increased Barijeh gum extraction yield. The gum yield obtained by acidic extraction was higher than that from alkaline treatment (Fig. 3). This trend was similar to the results obtained by others. Armisen and Galatas (2000) used acidic solvent extraction for agar gum extraction.

Increasing temperature increased Barijeh gum extraction yield. The role of temperature in extraction of another gums have been revealed as the most of pectin extraction in world is performed using hot acid as extraction medium (May, 2000).

Effect of extraction method on purity. Acid extraction resulted in lower protein content than alkaline extraction owing to molecular hydrolysis caused by the acid. In alkaline extraction the value observed for protein content was more than acid extraction (Fig. 4). Hence most pure was obtained at 90°C and pH 4. The value observed for protein content was more than that reported by Jessenne *et al*. (1974).

Gum characterization and sugar analysis. As shown in Table I, there is a slight difference in Barijeh gum composition as evidenced by the results we obtained compared to those in the literature. Our results showed 4.3% protein content more than result obtained previously by Jessenne *et al*. (1974) who observed 3.7% protein content. The total sugar content was 83.9%. Therefore, the purity of the gum was relatively high, composing mostly of saccharides. Also gum consisted of uronic acids (galactoronic acid) with a weight ratio of 24.3%. The

Fig. 1. Extraction–purification scheme of Barijeh oleogum resin



content of uronic acids obtained in this work was in agreement with that (22.7%) obtained by others (Jessenne *et al.*, 1974).

Gum consists of galactose and arabinose and uronic acids (galactoronic acid) in weight ratio of 65.26, 10.4 and 24.3%, respectively. Thus, galactose is the predominant sugar. It seems to be a arabinogalactan (Fig. 5).

Arabinogalactans (AGPs) are a group of macromolecules characterised by a high proportion of carbohydrate in which D-galactose and L-arabinose are the predominant monosaccharides. There is also a low proportion of protein, typically containing high levels of hydroxyproline. AGPs and AGs (arabinogalactan without protein) are found in flowering plants from every taxonomic group tested. In higher plants AGPs occur in leaves, stems, rods, floral parts, seeds and in large quantities in the trunks of some angio- and gymno- sperms. For example, the Acacia gums belong to a catholic family of structurally related AGPs. The component sugars are D-galactose, L-arabinose, L-rhamnose, D-glucuronic acid and 4 – 0 methyl glucuronic acid and the proportions vary in A. seyal and A. senegal. There are variations in protein content for various gums of the acacia species ranging from 0.13 to 10.4% (Phillips & Williams, 2006).

Fig. 2. Viscosity of extracted gum at different conditions

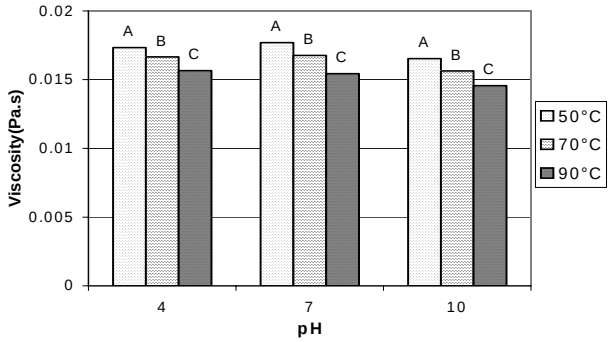


Fig. 3. Yield of Barijeh gum extraction at three temperature and pH values

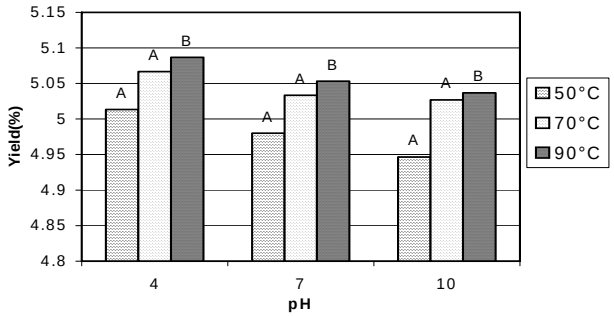


Fig. 4. The effect of various condition of extraction on protein content of gum

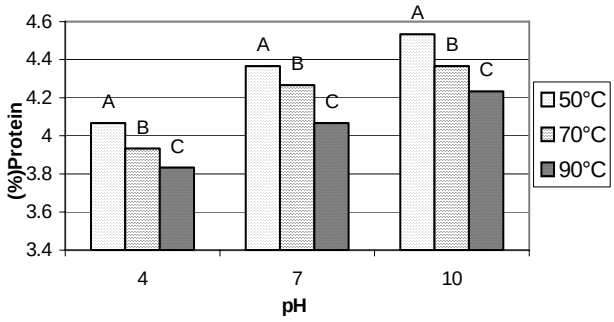
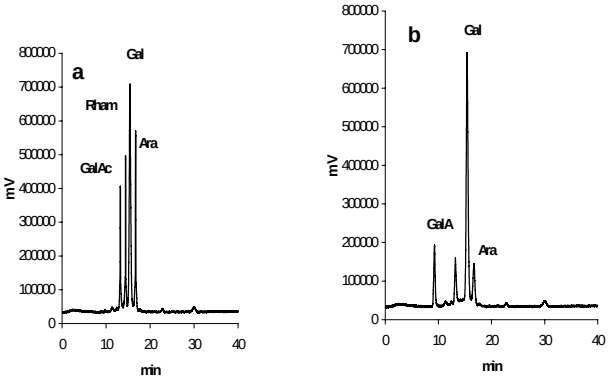


Table I. Composition of Barijeh gum

Barijeh gum	**%, w/w**
Moisture	5.1
Protein	4.3
Ash	6
From which;	
Na	0.05
Ca	3.02
K	0.62
Mg	0.17
P	0.05
Uronic acid	24.3
Total sugar	83.9
From which;	
Galactose	65.2
Arabinose	10.4

Fig. 5. HPLC chromatograms of standards sugars (a) and hydrolyzed gum (b)



The main components of gum arabic are arabinose and galactose in a 1:1 ratio (Defaye *et al*., 1986; Randall *et al*., 1989).

Larch gum is another arabinogalactan that is obtained from larch wood and its main components, galactose and arabinose are present in a variable ratio between 8:1 and 23:1. The gum is soluble in water up to 60% concentration and has a low viscosity with Newtonian behavior (Aspinall, 1962; Timell, 1965).

CONCLUSIONS

Results showed that Barijeh gum is an arabinogalactan-protein. Similar to another arabinogalactans it should have functional properties such as emulsifying properties. Though further studies are necessary to achieve a more comprehensive characterization of this gum; the properties measured in the present work reveal it as an interesting potential agent for food industry. Therefore, it has been concluded that although Barijeh is basically a polysaccharide.

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