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Isolation and Characterization of Aegle marmelos Gum– A Novel Controlled Release Polymer

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ABSTRACT

Wide ranges of drugs are formulated now in a variety of different forms for oral extended-release dosage forms. However, only those which result in a significant reduction in dose frequency and/or a reduction in toxicity resulting from high concentration in the blood or gastrointestinal tract are likely to improve therapeutic outcomes. To be a successful extended-release product, the drug must be released from the dosage form at a predetermined rate, dissolve in the gastrointestinal fluids, maintain sufficient gastrointestinal residence time, and may be absorbed at a rate and will replace the amount of drug being metabolized and excreted. In a nut shell, controlled-release formulations are a promising way to improve the patient's compliance by reducing dosing intervals and minimizing adverse effects. Out of many approaches to controlled drug release, matrix based approach is widely used due to its simplicity, scalability and from stability point of view. In order to control the release of drug from its dosage form, an effective controlled release polymer is essential. Though, there are several controlled release polymers available in the market, there is continuous need to develop controlled polymers which are safe and inexpensive. The aim of the work was to isolate and characterize the aegle marmelos gum as novel controlled relase polymer.

Keywords: Isolation, Controlled release, Aegle marmelos Gum.

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INTRODUCTION

Gum is obtained from fruits of *Aegle marmelos* belonging to family Rutaceae. The tree *Aegle marmelos* commonly known as Bael is indigenous to India and is generally found in the outer Himalayas, Shivaliks and South Indian plateau with altitudes ranging from 250 to 1200 m. It is a rich source of coumarins, vitamin C, and riboflavin. The ripe fruit pulp is red in color with mucilaginous and astringent taste. The pulp contains carbohydrates, proteins, vitamin-C, vitamin-A, angelenine, marmeline, dictamine, O-methyl fordinol and isopentyl halfordinol. The neutral oligosaccharides were characterized as 3-0-beta-D-galactopyranosyl-Larabinose, 5-0-beta-D galactopyranosyl-L-arabinose, and 3-0-beta-D galactopyranosyl-D-galactose, and the acidic oligosaccharides 3. The bark as well as fruit is reputed to be a valuable Ayurvedic medicine for dysentery and various intestinal complaints. The plant extracts were used against multi-drug resistant *Salmonella typhi* [1] [2]. The present investigation has been undertaken to find out the potential of gum from the fruits of *Aegle marmelos* to act as a release modifier in the formulation of controlled release matrix tablets.

MATERIALS AND METHODS

Isolation of Gum from *Aegle Marmelos* Fruits:

The edible pulp of *Aegle marmelos* fruit was collected and soaked in double distilled water. After soaking, it was boiled for 5 hours in a water bath until slurry was formed. Thus formed slurry was cooled and refrigerated over-night so that most of the undissolved portion was settled out. The upper clear solution was decanted off and centrifuged at 500 rpm for 20 minutes. The supernatant was allowed to concentrate at 60° C on a water bath until the volume reduced to one-third of its original volume. Solution was cooled to room temperature and was poured into acetone (three times the volume of slurry) with continuous stirring to form precipitate. Thus formed precipitate was separated, washed repeatedly with acetone and dried under vacuum at 50°C in an oven. The completely dried gum was powdered and was

passed through sieve #100, packed in a tightly closed container and stored in a dessicator for further usages.

Fig 1: (a) Photograph Showing Plant Part Selected For Study

Fig 1: (b) Photograph Showing Extracted *Aegle marmelos* Gum

Physicochemical Characterization of Gum:

Macroscopic properties of the gum were evaluated by observation of the color, taste and odor of the powdered gum. The gum was evaluated for solubility in water, ethanol, ether, methanol, acetone and chloroform in accordance with the standards. Other physicochemical properties of gum were also evaluated like loss on drying, total ash, P^H, angle of repose, bulk density, tapped density, hausner's ratio and Carr's index and the attained results are presented in Table 2 [2].

I. Angle of Repose:

The frictional force in a loose powder can be measured by 'Angle of Repose' (θ). It is defined as the maximum angle possible between the surface of the pile of the powder and the horizontal plane [2]. If more powder is added to the pile, it slides down the sides of the pile until the mutual friction of the particles producing a surface angle, is in equilibrium with the gravitational force.The angle of repose of gum was determined by the fixed funnel method. A funnel was secured with its tip at a given height (h), above a graph paper that is placed on a flat horizontal surface. The accurately weighed (10gms) gum was taken and was allowed to flow freely through the funnel on to the surface until the apex of the conical pile just touches the tip of the funnel. The radius of the formed cone was measured and angle of repose (θ) was calculated using the following equation:

$$\theta = \tan^{-1}\frac{h}{r} \quad \text{(or)} \quad \tan\theta = \frac{h}{r}$$

Where, θ = angle of repose; h = height of the cone; and r = radius of the cone base.

Table 4: Correlation between Angle of Repose and the Flow Property of Powders

Angle of Repose	Flow Property
< 25	Excellent
25-30	Good
30-40	Passable
> 40	Poor

II. Bulk Density:

Density is defined as weight per unit volume. Bulk density (D_b) was determined by measuring the volume (V_b) of known weighed quantity (W) of powdered gum using bulk density apparatus and can be calculated by using the formula:

$$\text{Bulk Density } (D_b) = \frac{\text{Mass of Powder (W)}}{\text{Bulk Volume of Powder } (V_b)}$$

The bulk density of a powder primarily depends on the particle size distribution, particle shape and the tendency of particles to adhere together [2].

III. Tapped Density:

Tapped density (D_t) was determined by measuring the volume (V_t) of known weighed quantity (W) of powdered gum after desired mechanical tapping using tapped density tester which provides a fixed drop of 14 ± 2 mm at a nominal rate of 300 drops per minute. The cylinder was tapped 500 times initially followed by an additional tap of 750 times until difference between succeeding measurement is less than 2% and then the Tapped Volume, V_t was measured, to the nearest graduated unit. The tapped density can be calculated by using the formula [2]:

$$\text{Tapped density } (D_t) = \frac{\text{Mass of powder (W)}}{\text{Tapped volume of powder } (V_t)}$$

IV. Hausner's Ratio:

The Hausner's ratio was obtained by dividing the tapped density by the bulk density of the gum powder. Lower the value of Hausner's ratio better is the flow property (Table 2).

$$\text{Hausner's ratio} = \frac{\text{Tapped density of powder } (D_t)}{\text{Bulk density of powder } (D_b)}$$

V. Carr's Index:

The Carr's index (% compressibility) of the gum powder was calculated from the difference between the tapped and bulk densities divided by the tapped density and the ratio is expressed as a percentage [2].

$$\text{Carr's index (\%)} = \frac{\text{Tapped density } (D_t) - \text{Poured density } (D_b)}{\text{Tapped density } (D_t)} \times 100$$

VI. Swelling Index (SI):

About 250 mg of gum powder was placed in 25 ml stoppered measuring cylinder and the initial volume occupied by the gum was noted. The volume was made up to 25 ml mark with distilled water. The contents were mixed gently for 2 minutes and set aside for 24 hours. Then the supernatant was carefully decanted and the volume of sediment was measured (Table 2).The Swelling Index was compute using the equation:

$$S = \frac{V_2}{V_1}$$

Where, S = Swelling Index

V_1 = Volume occupied by the gum prior to hydration

V_2 = Volume occupied by the gum after hydration [2].

VII. Water Retention Capacity:

Natural gum obtained from the fruits of *A. marmelos* is nontoxic. 250 mg of *A. marmelos* gum was allowed to hydrate in 25ml of distilled water at 25^{0}C in a 25 ml graduated cylinder and volume of water was measured at every 5 min. intervals until there was no further hydration observed [2]. The water absorption capacity was determined at different time intervals and the results are tabulated in Table 2.

VIII. Loss on Drying:

1 gm of the gum was transferred into a petri dish and then dried in an oven at 105 ± 5 ºC until a constant weight of gum was obtained[2]. The moisture content was then determined as the ratio of moisture loss to weight of sample expressed as a percentage. The result was presented in Table 2.

IX. Ash value:

1gm of gum was accurately weighed and evenly distributed it in the crucible. It was dried at 105ºC for one hour and ignited in muffle furnace at 600 ± 25 ºC. Ash content was estimated by the measurement of the residue left after the combustion in the furnace [2]. The result was presented in Table 2.

X. Acid Insoluble Ash:

The ash, obtained during the determination of ash value, was boiled with 25ml of 2M hydrochloric acid solution for 5 minutes and the insoluble matter was filtered and washed with hot water and ignited. Then subsequent weight was determined. The percent acid insoluble ash was calculated and the result was tabulated in Table 2.

XI. pH:

This was done by shaking 1% w/v dispersion of the gum in water for 5 min and then the P^H was determined using Digital P^H meter (Elico). The data was tabulated in Table 2.

Phytochemical Examination [3-10]:

For the detection of the presence of carbohydrates, reducing sugars, tannins, mucilage and peroxide enzymes, the standard tests, Molisch's test for carbohydrate, reduction of Fehling's solution for reducing sugars, ferric chloride test for tannins, ruthenium red test for *Aegle marmelos* gum were done.

1. Test For Carbohydrates (With aqueous test solution):

a. Molisch's Test:

To the aqueous solution of *Aegle marmelos* gum, few drops of alcoholic α-naphthol were added and to it few drops of concentrated sulphuric acid was added through sides of the test tube.

b. Barfoed's Test: To the aqueous solution of *Aegle marmelos* gum, Barfoed's reagent was added and then this solution was boiled.

c. Benedict's Test:

Procedure: To the aqueous solution of *Aegle marmelos* gum, Benedict's reagent was added and then this solution was boiled.

2. Test For Proteins:

a. Ninhydrine Test:

To the aqueous solution of *Aegle marmelos* gum, ninhydrine solution was added and then this solution was boiled.

b. Xanthoproteic Test:

To the aqueous solution of *Aegle marmelos* gum, concentrated nitric acid solution was added and then this solution was boiled.

3. Test For Alkaloids:

i. Wagner's Test:

To the aqueous solution of *Aegle marmelos* gum, Wagner's reagent was added.

4. Test For Tannins:

Ferric Chloride Test:

The extract was treated with ferric chloride solution.

5. Confirmatory Test for Chlorides: (Silver Nitrate Test):

Small amount of sodium extract was taken in a semi micro test tube and it was neutralized with dilute nitric acid and then silver nitrate was added.

Result: No white precipitate was formed indicating the absence of chlorides in *Aegle marmelos* gum.

6. Test for Sulphates:

Small amount of sodium carbonate extract was taken in a semi micro test tube and it was neutralized with dilute nitric acid. To this solution 5 drops of Barium chloride solution was added finally.

7. **Test for Flavonoids:**

To the alcoholic solution of *Aegle mar*melos gum, few fragments of magnesium ribbon and concentrated hydrochloric acid was added.

Characterization of *Aegle marmelos* gum:

A. X-ray Diffraction:

Diffraction pattern of *Aegle marmelos* fruit gum powder was recorded with an X-ray diffractometer (Panalytical species Pvt.Ltd, Singapore), X-ray diffraction was performed at room temperature (30^0 C) with a diffractometer; target, Cu (λ=1.54A^0), filter - Ni; Voltage - 40 KV; current - 30mA; time constant - 10mm/s; scanning rate - 2^6/min; measured from 10-350 at full scale of 200. The X-ray Diffraction of *Aegle marmelos* gum is shown in Fig 2.

B. Fourier Transform Infrared (FTIR) Spectroscopy:

FTIR spectra of *Aegle marmelos* fruit gum powder were recorded on samples prepared in potassium bromide (KBr) disks using Shimadzu Corporation (Tokyo, Japan) Model-1601 PC. Samples were prepared in KBr disks by means of a hydrostatic press at 6-8 tons pressure. The scanning range was 500 to 4000cm^{-1}. The FTIR spectra of *Aegle marmelos* gum is shown in Figure 3 and the peaks shown at different wave lengths are mentioned in Table 4.

In the spectroscopy, infrared radiation is absorbed by the sample and some of it is transmitted. The resulting spectrum represents the molecular absorption and transmission, creating a molecular fingerprint of the sample. Like a fingerprint, no two unique molecular structures produce the same infrared spectrum.

RESULTS AND DISCUSSIONS

Physicochemical characterization of gum:

Macroscopic properties showed that *Aegle marmelos* gum obtained after extraction from its fresh fruit pulp was an amorphous, free flowing powder, with yellow color, sweet in taste and with characteristic sweetish odor. The gum was found to be soluble in water and gave viscous solution on standing but practically insoluble in ethanol, acetone and chloroform. Its pH was found to be around 6.4 with acceptable limit of loss on drying (7.2%) and total ash (7%)

Flow properties of gum were determined in terms of angle of repose, bulk density, tapped density, hausner's index and Carr's index. All these physicochemical properties are tabulated in Table 2. The hausner's index and Carrs's index of the gum, indicate that it has good flow properties in the formulation of controlled release polymers. The swelling index of the gum indicates that the gum is suitable for the controlled release of the drug.

Phytochemical characterization of gum:

In Molisch's test, the violet coloured ring appeared at the junction and this confirmed the presence of carbohydrates in the *Aegle marmelos* gum. In Barfoed's test, the test solution showed brick red precipitate up on boiling. In Benedict's Test, the test solution showed reddish brown precipitate up on boiling. Therefore, all the above tests, indicate the presence of carbohydrates in *Aegle marmelos* gum. The results are indicated in the Table 2.

Table 1: Swelling Property of *Aegle marmelos* Gum

Natural Gum	After 5 min (ml)	After 10 min (ml)	After 15 min (ml)	After 20 min (ml)	After 25 min (ml)	After 30 min (ml)	After 35 min (ml)	After 24h (ml)
Aegle marmelos	0.6	0.8	0.9	1.1	1.2	1.3	1.3	4.2

Table 2: Physicochemical Properties of *Aegle marmelos* Gum

S.No.	Parameters		Results
1.	Macroscopic Property	Color	Yellow
		Taste	Sweet
		Odor	Characteristic sweetish
2.	Solubility	Water	Soluble
		Ethanol	Insoluble
		Acetone	Insoluble
		Methanol	Insoluble
		Ether	Insoluble
		Chloroform	Insoluble
3.	Angle of Repose (^{o})		28.09 ± 0.70*
4.	Bulk Density (g/cc)		0.47 ± 0.01*
5.	Tapped Density (g/cc)		0.56 ± 0. 01*
6.	Hausner's ratio		1.19 ± 0. 006*
7.	Carr's Index (%)		16.23 ± 0. 44*
8.	P^{H}		6.4
9.	Loss on Drying (%)		7.2
10.	Ash value (%)		7
11.	Acid Insoluble Ash (%)		1.0
12.	Swelling Index		16.8

* indicating the data presented for triplicate determinations, i.e. n=3.

Table 3: Phytochemical Characterization of *Aegle marmelos* Gum

S.No	Tests	Observation
1.	Test for Carbohydrates (Molischs's test)	+
2.	Test for Carbohydrates (Barfoed's test)	+
3.	Test for Carbohydrates (Benedict's test)	+
4.	Test for Tannins (Ferric chloride test)	–
5.	Test for proteins (Ninhydrin test)	+
6.	Test for proteins (Xanthoproteic test)	+
7.	Test for alkaloids (Wagner's test)	–
8.	Test for glycosides (Keller-killaini test)	–
9.	Test for flavonoids (Shinoda test)	–
10.	Mounted in 95 % alcohol	Transparent angular masses under microscope.
11.	Mounting in the iodine	No blue coloured particles (starch absent)
12.	Test for chlorides (silver nitrate test)	-ve
13.	Test for sulphates (barium chloride test)	-ve

Characterization of *Aegle marmelos* gum:

A. X-ray Diffraction Analysis:

The X-ray diffraction pattern (Fig 2) of *Aegle marmelos* gum did not show any characteristic peak, which indicates that the structure is completely amorphous.

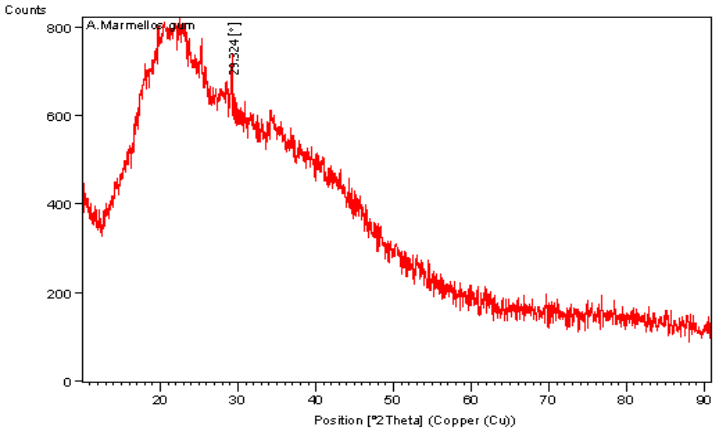

Figure 2: X-ray Diffraction Pattern of *Aegle marmelos* Fourier Transform Infrared (FTIR) Spectroscopy:

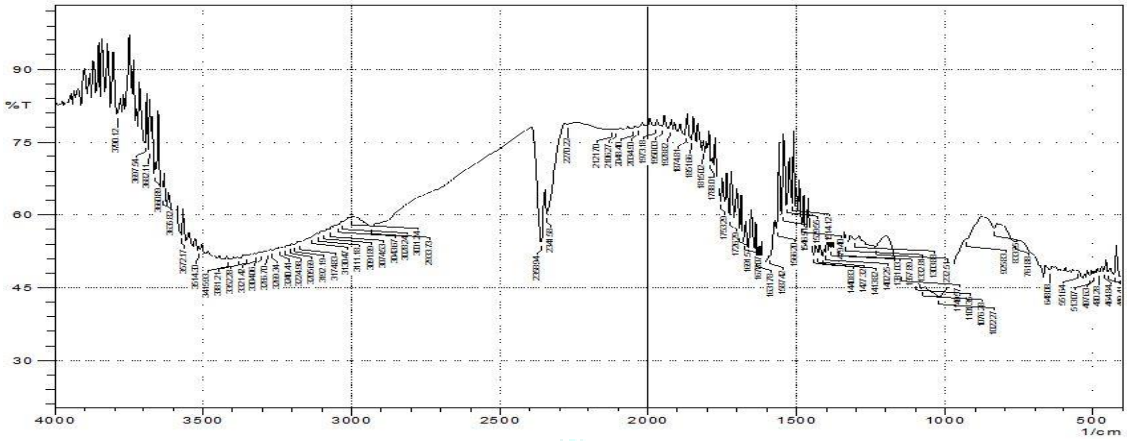

Fig 3: FTIR Spectra of *Aegle marmelos* gum

Table 4: FTIR Spectral Analysis of *Aegle marmelos* gum

Sample	Frequency of Peak	Functional Group
Aegle marmelos gum	3415.93	OH – Stretching
	2933	C-H Stretching
	1726.29	C=O Stretching
	1631.78	C-O Stretching
	1427.32	C-C Deformation
	1022.27	Secondary OH

CONCLUSION

The results of the present study demonstrated that the *Aegle marmelos* gum obtained from the fruits of tree *Aegle marmelos* is light yellow colored granular powder which is amorphous in nature. It is slightly soluble in water, practically insoluble in alcohol, chloroform and acetone and forms thick gel can control the drug release. Gum showed good flow property. Results suggest that the gum is suitable for use as a release retardant for the manufacture of controlled release tablets.

Since the primary ingredients are inexpensive, devoid of toxicity, biocompatible, biodegradable and easy to manufacture, they can be used in place of currently marketed controlled release polymers. Moreover, as this tree is widely distributed in nature, available chiefly in India and many other countries and easily available option without destroying the natural sources as compared to that of the other available natural option will be one of the suitable options to utilize as pharmaceutical controlled release polymer.

REFERENCES

1. Patil D.N, Kulkarni A.R, Hatapakki B.C And Patil B.S, Preparation And Evaluation Of *Aegle Marmelos* Gum As Tablet Binder, International Journal Of Pharma And Bio Sciences, 2010, Volume 1, Issue (1).
2. Rohini S. Kharwade, Neha S.Vyavhare, Sachin M.More, Formulation Of Mucoadhesive Tablet By Using *Aegle Marmelos* Gum, International Journal Of Applied Biology And Pharmaceutical Technology, Jan-Mar -2011, Volume:2, Issue(1).
3. C.K.Kokate, A.P.Purohit. S.B.Gokhale, Pharmacognosy volume–I, June 2010, 45th edition, Pg.No- A.2.
4. C.K.Kokate, A.P.Purohit. S.B.Gokhale, Pharmacognosy volume–I, June 2010, 45th edition, Pg.No-A.1.
5. C.K.Kokate, A.P.Purohit. S.B.Gokhale, Pharmacognosy volume–I, June 2010, 45th edition, Pg.No-7.9.
6. C.K.Kokate, A.P.Purohit. S.B.Gokhale, Pharmacognosy volume–I, June 2010, 45th edition, Pg.No- A.5.
7. Agarwal's-Text book of Biochemistry, 13th edition, 2005, Pg.No. 85.
8. BP -2008, Volume – IV, Appendix – IX – A 252.
9. G.R.Chatwal, Pharmaceutical Chemistry Inorganic (volume I), 2010, Pg.No-449.
10. C.K.Kokate, A.P.Purohit. S.B.Gokhale, Pharmacognosy volume –I, June 2010, 45th edition, Pg.No-7.9.