QUALITATIVE AND QUANTITATIVE ANALYSIS OF POLYMER LOCUST BEAN GUM WITH SALBUTAMOL BILAYERED TABLET

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Abstract

Background: The term "chronotherapeutics" describes a type of medicine where the availability of drugs in vivo is synchronized with the cycles of the disease to maximize therapeutic effectiveness and minimize negative effects.

Aim: The study was aimed to extract the gum mucilage from locust beans and evaluate the powdered drug as polymer when mixed with salbutamol for development of a tri-layered pulsatile tablet of salbutamol sulphate for Chronotherapeutic delivery in bronchial asthma.

Methodology: Locust bean gum was extracted by using water as solvent which was followed by purification of extract done by isopropanol resulting in overall 18% yield. The obtained purified powder of locust bean gum was evaluated for micromeritic/ flow properties. The powdered locust bean gum solely and along with mixture of salbutamol were also subjected to FTIR, and DSC.

Result and conclusion: The powdered mixture of locust bean gum and salbutamol was evaluated for flow properties including bulk density (0.56± 0.001 g/cm^3), tapped density (0.63± 0.001 g/cm^3), angle of repose (24.68º), Carr's compressibility index (11.1%) and Hausner ratio (1.12), Loss on drying (6.2 %), pH (5.5). The IR spectra of LBG revealed a large peak at 3389 cm-1 owing to O-H stretching of the hydroxyl group, and a peak at 2928 cm-1 due to stretching vibrations of-CH2, while, The spectrum of salbutamol-loaded LBG-, on the other hand, exhibits a strong peak at 3438 cm-1, which is due to O-H stretching of free hydroxyl. The DSC graph of LBG shows that the wide endotherm is at 92.23 °C, with a start at 71.08 °C and an end at 112.89 °C, with a heat flow of 55.73J/g. This might be attributed to evaporating structural water found in natural polymers. The thermogram of the solution of salbutamol sulphate shows an endotherm at 202.94 °C with a commencement of 195.07 °C and an end at 211.92 °C with a heat flow of 105.891J/g, indicating that an alteration has occurred with a change in thermal activity.

Keywords: chronotherapeutics, locust bean, Hausner ratio, micromeritic, salbutamol.

Introduction

Chronotherapeutics is a type of treatment wherein the availability of drugs in vivo is synchronized with the circadian rhythms of the disease to maximize therapeutic effectiveness and minimize side effects. (1;2). Numerous prevalent disorders, including rheumatoid arthritis (1), hypertension, asthma (2), angina pectoris (Portaluppi and Lemmer, 2007), and asthma are all impacted by chronobiology. The creation of chronotherapeutic drug delivery systems (CDDSs), which can release the necessary amount of medication at the precise location and time based on chronobiology and innate mechanisms, has been the subject of numerous recent studies. These mechanisms release drugs regardless of the environment or circadian cycles (3). The development of formulations to address therapeutic requirements associated with certain clinical disorders is the aim of drug delivery research. A new method for drug delivery systems has been introduced by chronopharmacological research, which has shown the significance of biological rhythm in medication therapy. In healthy organisms, the majority of physiological, biochemical, and molecular processes exhibit regular 24-hour cycles. Drug distribution can be synchronized with circadian rhythms with the help of chronotherapeutic agents, which can maximize effectiveness and/or reduce negative effects. Heart disorders, including cardiovascular conditions like hypertension and angina pectoris, can also be problematic. The purpose of creating chronopharmaceutic products is to maximize the desirable effects of a treatment and reduce its unwanted ones by utilizing recognized biological patterns in illness manifestation. **(4.)**

Other names for a chronomodulated system are sigmoidal release systems and pulsatile systems. The term "chronomodulated" originates from the chronobiologic system and is associated with chronopharmaceutics. The study of biological rhythms and their mechanisms is known as chronobiology. Our bodies include three different kinds of mechanical rhythms.

a) **Circadian rhythm:** Our bodies' oscillations that take place within a day are referred to as such.

b) **Ultradian rhythm:** Our bodies' oscillations that take place within a day or less are referred to as such.

c) **Infradian Rhythm:** - Oscillations lasting more than a day are referred to as Infradian rhythms.

The primary rhythm in the body that sustains all physiological, chemical, biological, and behavioral processes is the circadian rhythm. Therefore, circadian rhythms alter the pathophysiology of several illness states, potentially exacerbating the condition. Treatments for this kind of illness necessitate precisely timed, preprogrammed medicine delivery that synchronizes with the body's circadian rhythms. Thus, a novel method for treating this kind of illness is the chronomodulated or pulsed drug delivery system.(5). However, one of such illness can be considered as nocturnal asthma, a disorder that is common in 2/3rd of individuals with asthma, or those with the condition. This illness is signified by frequent nocturnal asthma flare-ups that worsen symptoms and necessitate medicines. Usually, warning signs appear between 8 a.m. and midnight. Taking medicine at midnight is convenient. (6)

Mostly found in the Mediterranean region, locust bean gum (LBG) is a high molecular weight non-ionic galactomannan polysaccharide that is derived from the seeds of the Ceratonia Siliqua, also known as the carob tree (7). Grafting or synergy with other appropriate polymers has increased its viscoelastic, swellability, and release-retarding capacities due to its flexible physicochemical properties, without compromising its biodegradable, biocompatible, nontoxic, or non-mutagenic qualities. LBG has been a part of several different medication delivery systems. (8).

MATERIALS AND METHODS

Extraction Procedure: LBGw (locust bean gum derived through water extraction) was obtained under the following conditions: For 1hour, whole carob seeds (100 g seeds) were immersed in 800 ml of boiling water at 100 °C. The seeds enlarge without tegument rupture during this processing. The seeds were removed from the water, rinsed, and the tegument was manually fractured and separated from the endosperm (Max Petitjean,2022). The germ was then separated from the endosperms, which were dried in an oven at 100 °C for 1-2 hours until constant weight was achieved. To obtain LBGw flour, the endosperms were milled and sifted through a 0.125 mm screen. (9)

Purification of Locust Bean Gum: After being distributed in aqueous solution, Locust Bean Gum was dissolved using heat. The resultant solution was filtered to remove non soluble particles. From this transparent solution, the locust bean gum was separated with isopropanol and filtered off. The white fibrous precipitate was recovered by vacuum filtering and washed twice with isopropanol, followed by acetone and diethyl ether. It is then kept for drying whole night in a hot air oven at 30 ºC (10), thereby producing 18% yield of locust bean gum after drying.

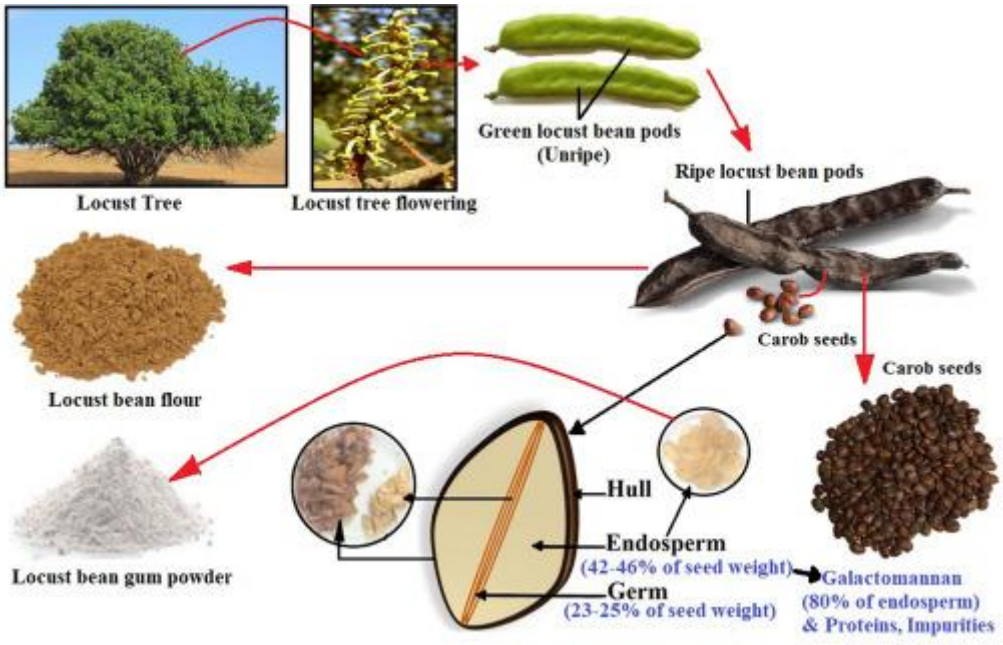

Figure 1: Diagrammatic representation showing locust bean gum powder after purification

Micromeritic properties: The locust bean powder was evaluated for flow properties including bulk density, tapped density, angle of repose, Carr's compressibility index and Hausner ratio.(11)

Bulk density:

Apparent bulk density was determined by pouring the blend into a graduated cylinder.
The bulk volume and weight of the powder was determined.(12) It is determined by following equation,

$$\rho_b = W / V_b$$

Where, ρ_b = Bulk density, V_b = Bulk volume of blend (cm^3), M = Weight of power (gm).

Tapped density: The measuring cylinder containing a known mass of blend was tapped for a fixed time (100 tapping). The minimum volume occupied in the cylinder and weight of the blend was measured.(13)

It is determined by following equation,

$$\rho_t = W / V_t$$

Where, ρ_t = Tapped density, V_t = Final volume of blend after tapping (cm^3)

Angle of repose: Angle of repose was determined using funnel method. Funnel was set at a height of 2 cm from the surface. The blend was discharged from the funnel until the tip of pile of powder touches the lower end of funnel. Radius of the heap was measured and angle of repose was calculated. (14) It is determined by following equation,

$$\tan \theta = h / r$$

Therefore, $\theta = \tan^{-1} (h / r)$

Where, θ = Angle of repose, h = Height of pile of powder blend, r = Radius of heap

Carr's compressibility index: Carr's compressibility index is determined by following equation. (15)

$$C = (\rho_t - \rho_b) / \rho_t \times 100$$

Where, C = % compressibility, ρ_b = Bulk density, ρ_t = Tapped density

Hausner's Ratio: This is an indirect index of ease of powder flow.[9] It is calculated by following formula; (16)

$$H = \rho_t / \rho_b$$

CHARACTERIZATION of LOCUST BEAN GUM POWDER

FOURIER TRANSFORM-INFRARED SPECTROSCOPY (FTIR): FTIR is a versatile technique used for the analysis of polymers, drug and final formulation. (17) The spectrometer (IR, affinity, DRS-8000A, Shimadzu, Japan) was used for the analysis of functional group present in the samples using diffuse reflectance method. In this technique, before starting the process, samples were thoroughly dehydrated, in order to fully eliminate the moisture however the compressed disc preparation is not needed in the diffuse reflectance method. Therefore, dried samples were carefully mixed in very small amount of dried potassium bromide and then placed in the sample stub having shape of cup for detecting the structure and functional groups. Infrared region with a scale of 4000–400 cm−1 was analysedfurther using which spectra were obtained between the X-axis i.e.,wave number (cm−1) and Y-axis i.e., transmittance (%). (18)

DIFFERENTIAL SCANNING CALORIMETRY (DSC): Differential Scanning Calorimetry (DSC) is a thermal analytical technique that measures the heat against time and examines how the physical properties of a sample change with increasing temperature over time. (13) Differential scanning calorimetric analysis was done for studying thermal behaviour and performed using diffraction scanning calorimeter (Perkin-Elmer, USA). Sealing of samples was done by placing the samples in aluminium pan in addition to heating in the range of (30-300) °C with the rate of heating 10 °C/min in nitrogen atmosphere. (19)

SOLUBILITY MEASUREMENTS:

Four samples were made at 0.1% w/w concentration on a dry weight basis (i.e., solids content was constant) at room temperature (20-25 °C) for 0.5, 1, 2, and 3 hours with mechanical stirring.

Other preparations were made under the same conditions but at a temperature of 80 °C for 5, 10, 30, and 60 minutes. The equivalent solution was then taken and centrifuged (6000g for 30 minutes at 20 °C) to remove the insoluble particles. The supernatent was recovered, and the final polymer concentrations were determined (Gaisford, 1986).

$$\text{solubility (\%)} = \frac{\text{supernatent concentration (mg/ml)}}{\text{initial preparation concentration (mg/ml)}} \times 100.$$

RESULT

Evaluation of powder properties of locust bean gum: The locust bean powder was evaluated for flow properties including bulk density, tapped density, angle of repose, Carr's compressibility index and Hausner ratio. (Cheng, Malik)

S.No	Parameter	Result
1	Bulk Density (g/cm3) (±SD) (n=3)	0.56± 0.001
2	Tapped density (g/cm3) (±SD) (n=3)	0.63± 0.001
3	Angle of repose (θ) in degree	24.68
4	Carr's compressibility index (%)	11.1
5	Hausner ratio	1.12
6	Swelling index	12
7	Loss on drying (%)	6.2
8	pH	5.5
9	Viscosity (cps) at 50 rpm (1%)	6.4

SD=standard deviation, n=3 the parameters shown are based on 3 replicate and expressed as mean

Table 1: Powder evaluation of locust bean

Evaluation of powder properties of locust bean gum and salbutamol (F1): The locust bean and salbutamol blend was evaluated for flow properties including bulk density, tapped density, angle of repose, Carr's compressibility index and Hausner ratio.

S.No	Parameter	Result
1	Bulk Density(g/cm3) (±SD) (n=3)	0.61± 0.002
2	Tapped density(g/cm3) (±SD) (n=3)	0.70± 0.004
3	Angle of repose(θ) in degree	30.64
4	Carr's compressibility index (%)	12.8
5	Hausner ratio	1.14

SOLUBILITY MEASUREMENTS: Carob gum is somewhat 50-55% soluble at 20 °C/1 hr in cold water and achieves a solubility of 80-85% at 80 °C/30 min). This difference in solubilization may be due to the fact that at high temperatures, some molecules could dissolve which was not possible at low temperatures.

BILAYER CORE TABLET FORMULATION: Blend of locust bean gum and salbutamol was prepared to develop different formulations (F1, F2, F3) for core tablet (bilayer) and the ingredients were mixed, passed through sieve number 60.

Outer layer- immediate release	**Present in one tablet in milligrams (approximate values)**		
	F1	F2	F3
Salbutamol sulphate	2	2	2
Mannitol	0.5	1	1.5
Lactose	12.5	12	11.5
Inner layer- sustained release			
Salbutamol sulphate	4	4	4
$PVPK_{30}$	0.25	0.5	0.75
Locust bean gum	40.5	40	40
Talc	0.25	0.5	0.25

Table 2: Salbutamol and Locust bean powder blend

FTIR and DSC analysis of Salbutamol and Locust bean gum:

FOURIER TRANSFORM-INFRARED SPECTROSCOPY (FT-IR)

The FTIR method is widely used in the analysis of functional groups. The FTIR spectra of LBG and salbutamol sulphate are shown in Fig. 1. The IR spectra of LBG revealed a large peak at 3389 cm-1 owing to O-H stretching of the hydroxyl group, and a peak at 2928 cm-1 due to stretching vibrations of-CH2. The emergence of the peak at 1657 cm-1 is because of aryl-substituted C=C. The larger peak at 1438 cm-1 is due to symmetrical stretching of LBG carboxylate. The IR-spectrum of salbutamol sulphate revealed strong peaks at 1103 cm-1 (C-O stretching) and 1499 cm-1 (O-H bending). The spectrum of salbutamol-loaded LBG-, on the other hand, exhibits a strong peak at 3438 cm-1, which is due to O-H stretching of free hydroxyl.

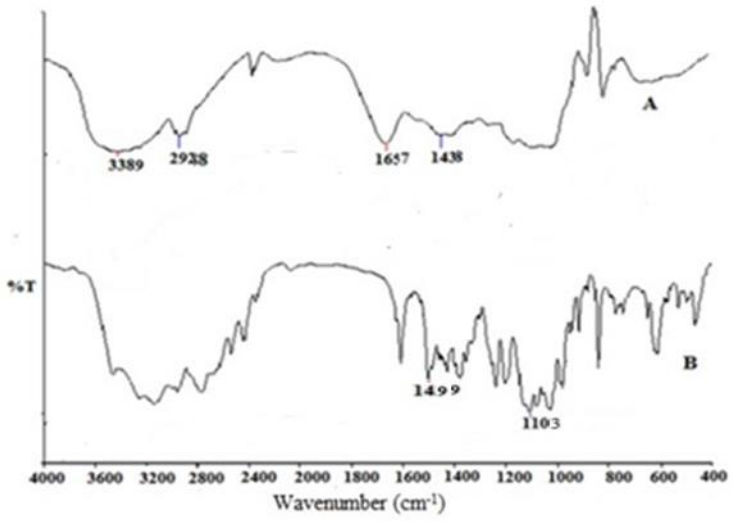

Fig.1: FT-IR spectra of LBG (a), salbutamol sulphate(b)

DIFFERENTIAL SCANNING CALORIMETRY

Differential scanning calorimetry is a useful thermal analytical method for determining the alteration in thermal conductivity with temperature. Thermal evaluation may be used to identify the characteristics of the matrix material

in composite materials. In comparison to the separate polymers, the mix typically exhibits an increase in glass transition temperature. LBG's thermogram is shown in Fig. 2(a). The DSC graph of LBG shows that the wide endotherm is at 92.23 °C, with a start at 71.08 °C and an end at 112.89 °C, with a heat flow of 55.73J/g. This might be attributed to evaporating structural water found in natural polymers. The thermogram of the solution of salbutamol sulphate in fig 2 (b) shows an endotherm at 202.94 °C with a commencement of 195.07 °C and an end at 211.92 °C with a heat flow of 105.891J/g, indicating that an alteration has occurred with a change in thermal activity.

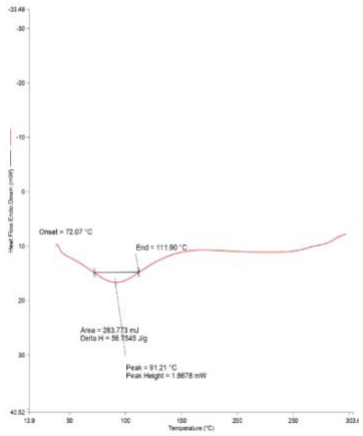

Fig 2(a) DSC thermogram of LBG

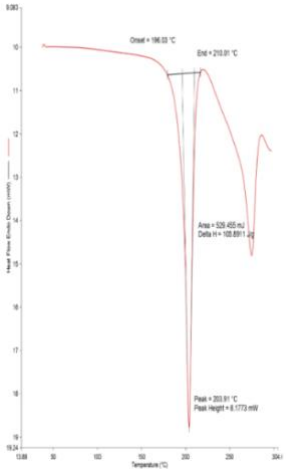

Fig 2(b) DSC thermogram of Salbutamol sulphate

DISCUSSION

Although sustained and controlled drug delivery systems have gained a lot of success and application in field of medication, these systems fail to deliver drug according to circadian behavior of diseases for which pulsatile systems are beneficial. For successful development of chronotherapeutic dosage form, knowledge of circadian time structure, rhythm in disease pathophysiology or 24 hour pattern in symptom intensity of chronic medical conditions and chronopharmacology of medication is needed. Significant progress has been made towards achieving pulsatile drug delivery system that can effectively treat diseases with non-constant dosing therapy.(20)

The concept of formulating tablets for chronotherapeutic drug delivery system containing salbutamol offers a suitable, practical approach to achieve sustained release of the drug. In present work, locust bean gum and salbutamol powdered drug was prepared in different concentrations. Other excipients used for preparation of powdered drug included Mannitol as binder and diluent, talc as glidant, lactose, salbutamol suphate, $PVPK_{30}$ & Aspartame as sweetener. All the micromeritic parameters like angle of repose, bulk density, Carr's index were studied.

The drug and excipients compatibility were studied by FTIR which revealed that no chemical or physical interaction took place. Differential scanning calorimetry is a useful thermal analytical method for determining the alteration in thermal conductivity with temperature. In comparison to the separate polymers, the mix typically exhibits an

increase in glass transition temperature. Micromeritic parameters of locust bean gum and salbutamol powder having better flow properties were studied such as- bulk density, tapped density, angle of repose, Carr's compressibility index and Hausner ratio.

CONCLUSION

We can conclude that chronotherapeutic dosage form can be prepared by using natural polymer i.e., locust bean gum which showed the drug can be used effectively for sustained released as part of chronotherapeutic drug delivery system. Also, there are several advantages of natural polymer over that of synthetic polymers. Hence, from the results of FTIR, DSC study profile of locust bean gum and salbutamol powdered drug it was concluded that it can be used for the compression of triple layered sandwitched chronotherapeuic dosage form which can effectively suppress the symptoms of asthma in the early morning.

AUTHORS CONTRIBUTIONS

All the authors have contributed equally.

CONFLICT OF INTERESTS

There is no conflict of interest with reference to the publication of this paper.

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