Research Article

Isolation of polysaccharides from *Plantago ovata* and their evaluation as potential matrices for designing the controlled release floats of Piroxicam

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Citation

Abdul Karim, Tahir Mehmood and Muhammad Ashraf Shaheen. Isolation of polysaccharides from *Plantago Ovata* and their evaluation as potential matrices for designing the controlled release floats of Piroxicam. Pure and Applied Biology. Vol. 5, Issue 4, pp1026-1043. <http://dx.doi.org/10.19045/bspab.2016.50128>

Received: 24/08/2016

Revised: 07/09/2016

Accepted: 12/09/2016

Online First: 17/09/2016

Abstract

The present study deals with isolation of different polysaccharides from *Plantago Ovata* husk and their use in designing the controlled release Gastro-Retentive (GR) floats of piroxicam (PRC). Gastroretentive floats are actually drug release (DR) moieties of low density used to retain the drug in stomach for maximum time to increase its bioavailability. The polysaccharides extracted from *P. Ovata* were selected due to its therapeutic impacts and ease in accessibility although synthetic polymers have already been utilized for designing such types of floats. The floats of that fraction were selected for designing further formulations having least lag time with maximum drug release retarding ability. For modifying the drug release (DR) rate; flax seed gel, gum tragacanth and Stearic acid were used as variables. The direct compression technique was used for compression and floats were characterized for uniformity in weight, drug content, friability and *in vitro* buoyancy. All fractions were characterized by FTIR, TGA, GPC and SEM study to find the nature of polysaccharide, predicting its stability and compatibility with human body. Some mathematical tools i.e. kinetic study and regression analysis (RA) were applied to find the effect of different variables on DR rate and closeness among the theoretical and experimental DR profile. The ethanol separated part (ENa) was seen to be most suitable in view of its better swelling ability and minimum lag time. The formulation S₄F₉ was seen to be best fitted due to its minimum lag time, best drug entrapping ability and best suitability to Higuchi's model ($R^2 = 0.957$) and Power law ($R^2 = 0.963$). The DR mechanism was found to be non-Fickian diffusion ($n > 0.45$).

Keywords: Polysaccharide Gels; GR floats; GPC; FTIR; TGA; 3² Factorial design; Lag time

Introduction

The regular polysaccharides and their derivatives are richly utilized as a part of pharmaceutical formulations of various types where they assume an imperative part

as binder, film formers, DR modifiers, viscosity enhancers, stabilizers and suspending agents [1]. In a few cases, proteins, starches and plants hydro-gels are being used as a medium in CDDS (control

drug delivery system) but most of polymers used have no therapeutic importance. Presently an exceptional consideration is being paid on selection of polymers having double characters i.e. restorative significance alongside DR retardant. The *Plantago Ovata* is an herbal plant and generally used as a drug for treating unending ailments i.e. coronary heart diseases, reducing the blood glucose level in blood.

In this study, focus is towards the segregation of new swellable biopolymer from *P. Ovata* husk and their utilization as matricing agent for designing the gastroretentive floats. The GR floats are actually floating drug tablets used for increasing the gastric residence time to increase the drug bioavailability. The swelling property of husk gel is reported in literature but its fractionation and then their use in CDDS is under progress [2]. It is most suitable than other synthetics gels due to being non-toxic, minimal effort of extraction, simplicity of accessibility, high partiality for water. It is water insoluble and mechanical activity of nutritious trench can't absorb it. There are so many mechanisms for controlling the DR and one such mechanism is swelling through hydration which is apparently present in *P. Ovata* seed husk. Swelling increases the pore volume of hydrogel and permits the dispersion of gastric medium for discharging the absorbed medicine in gel and thermal study reveals that it can face the environmental changes due to its stability at room temperature [3-7].

The PRC is a viable NSAID drug utilized for the treatment of joint inflammation [8]. Its typical dose is 20 mg yet some time two fold doses are required to keep up the drug plasma level. The half-life of drug is around 40 h in man and is metabolized into dynamic metabolites and peak plasma level is achieved after 2 h subsequent to taking

single dosage and it stayed dynamic over next 48 h because of expanded plasma half-life. The drug has exceptionally protein binding capacity and in this way it replaces numerous different types of drugs that are only limited to proteins. The present study was directed to prepare PRC floats utilizing *P. Ovata* seed husk and some other characteristic polysaccharides as DR modifier and was compared to sustained release marketed tablets [9, 10]. This drug is utilized to assuage the joint inflammation. It works by preventing the production of some chemicals (prostaglandins) which are involved in mediation of pains in stiffness, swelling, tenderness and provocative conditions in joint inflammation [11-13].

The goal was to prepare GR floats of PRC. These extracted fractions meet all the physiological prerequisites required for matrix utilized for preparing the gastroretentive controlled release tablets i.e. being insoluble in water, having low density, high swelling/ re-swelling ability. By adjusting the matricing variables, the DR mechanism can be adjusted. The data acquired for *in vitro* DR were fitted to first-order, zero-order, Higuchi model and Power law [14]. The Statistical method [MANOVA: multivariate analysis of variance] was used to analysis the DR data at different time intervals [15, 16]. The highest value of regression coefficient (R^2) for Higuchi's model indicates that diffusion is predominant mechanism for controlling the release of drug. The main target was to increase the bioavailability by increasing its gastric residence time. All floats were tested for lag time, DR percentage, similarity and difference factors to find the optimum formulation [17, 18].

Materials and methods

The psyllium seed husk (H), flax seed gel (FSG), gum tragacanth (GT) from local market and PRC was gifted from a pharmaceutical industry. Different

chemicals: C_2H_5OH , CH_3OH , CH_3COOH , Magnesium stearate from Merck and HCl , $NaHCO_3$, $NaOH$, $CHCl_3$, and $(CH_3)_2CO$ from ICI Ltd and Sigma-Aldrich. The double distilled water from distillation unit. The apparatus: Dissolution apparatus, UV/Vis Spectrophotometer (Shimadzu 2100, Tokyo, Japan), CHNS analyzer, FTIR spectrophotometer (IR Prestige-21 Shimadzu, Japan) from High Tech Lab University of Sargodha. All chemicals were used without further purification.

Isolation of gel

The *P. Ovata* husk gel (H) was separated according to a published technique with some modifications [19, 20]. The *P. Ovata* husk around 100 g was mixed in 1 L distilled water and was kept for 24 h for

swelling. After complete swelling, whole mass was dissolved in 0.4 M $NaOH$ solution by stirring with glass rod. To remove any insoluble material, the solution was filtered and to the filtrate; different organic solvents (Figure 1) were added separately in different portion of filtrate to coagulate the different insoluble fractions from filtrate. The isolated gels were dialyzed, dried and finely ground to fine powder. The fine powder was used as matricing agent for drug to design GR floats. The citrus acid and $NaHCO_3$ was mixed in matrix to reduce the LT (The time after which tablets start floating on the dissolution medium) and increase the gastric retention time for increasing the bioavailability of drug.

Figure 1. Extraction of different fractions from raw gel of *P. Ovata*

Characterization of gels (Instrumentations)

Elemental analysis

The CHNS analyzer was used for elemental analysis of all samples and their results were reported.

FT-IR

The spectrometer (IR Prestige-21 Shimadzu, Japan) was used to get the spectra of all hydrogels sample using pellet KBr technique. After drying the samples under

vacuum for 20 h were heated at 60 °C and then these samples were ground to fine mass. Pellets were compressed by applying pressure 60 kN.

Thermal analysis (TA)

The thermal analysis was conducted by thermal analyzer (SDT, Q-600, TA USA) under N_2 atmosphere keeping heating rate 10°C, 15°C and 20°C min^{-1} ranging from room temp to 600°C in Pt crucible to find

the thermal stability, degradation pattern, heat flow, activation temp and glass transition temp. The major degradation pattern was determined by pre-exponential factor (A) using Broido's method (Broido, 1969) and for calculating activation energy (Ea) Eq. 1 was used.

$$\ln[\ln(1/y)] = -E_a/RT + \ln A R T_m^2 / E_a \quad (\text{Eq. 1})$$

It can be rearranged as,

$$\ln \ln(1/y) = -(E_a / R)(1/T) + \text{constant}$$

y is equal to $(w_t - w_f)/(w_0 - w_f)$, w_t was the mass of sample at time t, w_0 is initial mass and w_f is final mass, where Θ is heating rate and R is gas constant while T_m is the maximum reaction rate temp.

By plotting a graph between $1/T$ against $\ln \ln(1/y)$, a straight line is obtained and its slope and intercept gives E_a and A respectively. By single heating rate method, the kinetic parameters have been calculated but multiple effects heating rate method is most suitable [17]. The Doyle's method was used to find comprehensive index of intrinsic thermal stability (ITS), integral procedural decomposition temperature (IPDT) and thermal stability of samples [21- 23]. To find the energy of activation (ΔG^*), enthalpy of activation (ΔH^*) and entropy of activation (ΔS^*), the following standard equation was used

$$E_a = -(\text{slope} \times 8.314)$$

For data analysis, Universal Analysis 2000 software, version 4.2E, TA Instruments, USA and MS Excel® 2010 was used.

Gel permeation chromatography (GPC)

GPC technique was used to determine the GPC parameters i.e. molar masses of all fractions. The M_n = average molar mass based on number of molecules N_i in a particular weight class M_i .

$$M_n = \sum N_i M_i / \sum N_i \quad (\text{Eq. 1})$$

M_w = average molar mass based on weight fraction of molecules W_i in a particular weight class (M_i);

$$M_w = \sum N_i M_i^2 / \sum N_i M_i \quad (\text{Eq. 2})$$

Where $M_z = \sum N_i M_i^3 / \sum N_i M_i^2 \quad (\text{Eq. 3})$

From ratio of different average molar masses, the polydispersity index (PDI) was calculated (Eq. 4)

$$PDI = M_w / M_n \quad (\text{Eq. 4})$$

For GPC analysis, pullulan and dextran were used as standard (Polymer Standards Service, Germany). Dimethyl sulphoxide was used as eluent in a system that is fitted with two columns functioning at run rate 0.5 mL min⁻¹ at 70°C using volume 50 µL as injection.

SEM studies

The pellets of all fractions were compressed at hydraulic press at pressure 3000 Psi mounted with Ag-painted aluminum stubs coated with Au working at accelerating voltage 10 kV. Different micrographs were taken at different magnifications to study the surface morphology. Greater the roughness on surface, greater is the adsorption power of gel to adsorb and uneven surface has greater absorption tendency. The drug release behavior of ENa was matching with the set criteria (minimum lag time), therefore other fractions were ignored. Moreover it was found that different fractions have different drug release behavior with different drugs. The ENa fraction was best fitted therefore was selected as a standard for designing the desired drug release floats and therefore its GPC chromatogram was inserted.

Calibration curve

A stock solution of PRC (10 mg/100 mL) was prepared in simulated gastric fluid (SGF) [pH 1.2 HCl (5 mL HCl concentrated in 991.5 mL distilled water) with purified pepsin]. The prepared samples were diluted 10 times and diluted medium was further diluted to 50% to obtain a solution of known concentration solutions ranging from 0.5 – 2.5 µg/mL. The absorbance was measured spectro-photometrically at 234 nm with the mean data (n = 5). The calibration graph for PRC (Figure 2a) was plotted against its concentration and was regressed into linear

line. The concentration of DR was calculated using equation

$$y = 0.070 X + 0.004$$

The calibration graph (Figure 2b) and DR profile of all fractions (Figure 3, Table 1) are shown. In equation, y is DR%, X is the

amount of drug released. By putting the value of other variables, we can calculate the value of X during that interval. This plot is drawn using DR data taken from pure drug only without any matrix.

Figure 2. a, Structure of PRC (L) b, Calibration curve for PRC (R)

Figure 3. (R) A plot of % release of drug from all hydrogels (L) The % release of drug from ENa matrix float, H_{matrix} and C_{std}. vs time for 10 h (Table 2)

Table 1. Comparative DR % from isolated fractions VS lag time

PRC	MNa	ENa	AANa	ClfNa	ANa	H	Cstd
Rel%	9.0	21.1	16.9	16.9	24.1	43.9	35.2
LT (min)	42	15	40	35	42	48	32

*Fractions extracted from hydrogel solution of NaOH using solvents, MNa = Methanol, ENa = Ethanol, AANa = acetic acid, Clf Na=chloroform, ANa= acetone, H= whole gel and Cstd= standard tablet

The floats prepared using ENa showed minimum LT, therefore ENa was selected as matricing agent for further study. Although

DR from all fractions was generally less than required for achieving effective drug blood plasma level. Further formulations

were designed to find optimized formulation. Different batches were prepared with different amounts of excipients using ENa as major matricing agent to explore the most potential formulation.

Preparation of matrix tablets

The wet granulation technique was used to prepare the ENa floats. Excipients including drug were blended geometrically and formulations were designed as per factorial model (Table 2). For sorption of drug, ENa was put in clear drug solution for maximum

swelling. After maximum swelling, the swelled lump was kept in long container having non-sticky surface fitted with pump. The whole mass was splashed through little opening on inner side of container fitted with heating heated at 45 °C. The dried granules were collected and compacted in to floats under pressure of 10 kN on single punch machine fitted with 9.5 mm flat punch. The iPrOH was added in blended material to homogenize the swelling rate and magnesium stearate was added as glident.

Table 2. Floats formulations of PRC (40 mg)

Batch	Variables	Resources	Low (-1)	Medium (0)	High (+1)	Constant*(mg)
1 st	X ₁	ENa	80	160	220	NaHCO ₃ = 10
	X ₂	SA	10	20	30	CA = 5
2 nd	X ₁	ENa	80	160	220	NaHCO ₃ = 10
	X ₂	SA	5	10	15	CA = 5
3 rd	X ₁	ENa	80	160	220	NaHCO ₃ = 5
	X ₂	FSG	5	10	15	CA = 2.5
4 th	X ₁	ENa	80	160	220	NaHCO ₃ = 5
	X ₂	GT	5	10	15	CA = 2.5

*CHCl₃ = 5 mL, iPrOH (isopropyl alcohol) = 2 mL, Magnesium stearate = 5 mg in each floats

Analysis of friability, drug content and stability study

Ten floats weigh equal to 100 mg were transferred to 100 mL volumetric flask and subsequent to blending in solution of pH 1.2. The blend was filtered and 1 mL of filtrate was further diluted to form a solution (100 µg mL⁻¹). The analysis was conducted at λ_{max} 234 nm using UV -Visible spectrophotometer. To test the friability, floats were kept in a drum rotating at a speed of 30 rpm min⁻¹ and sample was rotated at a speed 100 revolutions min⁻¹ for 5 min to find the disintegration of float. The floats were dusted off and were weighed again and % loss in weight was calculated from equation.

Friability = (initial mass - final mass) / initial mass × 100

Dissolution study

The USP paddle dissolution apparatus- II was used to study the DR at 37°C ± 0.1°C

rotating at 50 rpm. The buffer pH 1.2 was taken in each container (900 mL). After fixed time interval, 5 mL sample was taken out and was analyzed at double beam spectrophotometer (Schimadzu UV-1700) at λ_{max} 234 nm. After withdrawal of test sample, buffer medium kept at same temperature was added to have the volume 900 mL. The DR % was calculated from withdrawn sample.

Statistical analysis

The 3³ full factorial designs (Table 1) were used for RA (regression analysis) to determine the effect of independent variables on DR%. The statistical equation was derived to find the effect of all variables.

$$S\% = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_{12} X_1 X_2 + \beta_1 X_1^2 + \beta_2 X_2^2$$

Where S% (DR %); β is coefficient of regression; X₁ and X₂ stands for effect of

both independent variables ; X_1X_2 is the interactions factor between X_1 and X_2 and X_1^2 and X_2^2 are quadratic terms.

Result and discussion

Description of ENa

Elemental analysis

Each sample was analyzed thrice. The percentage of Carbon was found to be lower (40.2%) than in other natural polysaccharides (45.0%) and Hydrogen content was greater (6.5%) than observed in other polysaccharides (6.0%) due to its better water upholding capability. The N was not detected in any sample showing that protein was absent in all sample.

FT IR spectroscopy

The FTIR spectrum of ENa (Figure 4) shows that following absorbance were indicated. The assignment at 3374-3443 cm^{-1} was due to -OH stretching broad band and a peak at 2882-2918 cm^{-1} was due to saturated C-H stretching and absorbance at 1563-1638 cm^{-1} was showing deformation by entrapped water, a neat band at 1014-1052 cm^{-1} was showing β - glycosides linkage and band at 643- 692 cm^{-1} and 529-550 cm^{-1} are indicating polymer backbone.

Figure 4. FTIR spectrum of ENa

GPC study (comparative study)

The molar mass distribution of all hydrogels was determined by GPC study (Table 2). The heterogeneity was determined from polydispersity index (PDI) that is the ratio of M_w and M_n . The molar mass averages for a monodisperse polymer are almost equal and their PDI values are close to 1. In a poly disperse system, $M_z > M_w > M_n$ as the result

the PDI value is greater than 1. For all fractions, the molar mass distribution is shown (Table 3) and their PDI are found to be; ANa = 2.87, AANa = 2.69, MNa = 3.16, ClfNa = 3.78 and ENa = 3.75. From these values it was found that their heterogeneity index is ranging from 2.682 to 3.781 indicating that these hydrogels are showing poly disparity.

Table 3. GPC parameters of hydrogel fractions

Parameters	AANa	ANa	ENa	MNa	ClfNa
Mn (g mol⁻¹)	4.991×10 ³	4.482×10 ³	3.326×10 ³	3.566×10 ³	2.158×10 ³
Mw (g mol⁻¹)	1.327×10 ⁴	1.265×10 ⁴	1.353×10 ⁴	1.124×10 ⁴	8.167×10 ³
Mz (g mol⁻¹)	2.641×10 ⁴	2.934×10 ⁴	2.955×10 ⁴	2.198×10 ⁴	2.185×10 ⁴
Mp (g mol⁻¹)	7.372×10 ³	8.473×10 ³	8.252×10 ³	1.597×10 ⁴	4.142×10 ³
Vp (cm³)	7.445	7.366	7.397	7.089	7.715
PDI	2.695	2.87	3.75	3.169	3.787

Scanning electron microscopy

Different photographs were taken at different magnifying power to study the

surface morphology. The SEM image of ENa is shown for representative purpose (Figure 5).

Figure 5. Scanning electron microscopy of ENa**Thermal Analysis**

The thermal analysis was conducted for isolated fractions ranging from room temperature to 600°C. An endothermic weight loss (10-12%) was seen at 190-200°C, might be due to loss of absorbed water [24]. The weight loss about ~46% occurred in range of 226-328°C due to main decomposition of structure (Figure 5a, b). This step is an exothermic in nature. Another degradation with weight loss ~25% was observed at 450-570°C might be due to complete decomposition of husk hydrogels to gaseous products (CO₂, CO, and H₂O) leaving behind carbon content ash residue. No significant difference among thermal

behavior of these fractions was seen. The average E_a values which were found to be 134 kJ mol⁻¹ at multiple heating rates. Using the parameters like $IPDT$ and ITS values (Doyle's method) the thermal stability of all fractions was assessed. The $IPDT$ and ITS values were ranging from 275-305 °C and 0.43-0.490 respectively. These values indicate that all fractions are fairly stable. FWO method was used to find the E_a values for major stage of decomposition (Table 4). The FWO and α - T plots for ENa are shown in Figure 6. The multistep degradation pattern was apparent from variability in E_a with α .

Figure 5 a, b. Thermal analysis of ENa and AANa (L→R)

Figure 6. Representative α -T curve and $\log \beta$ for selected fraction

Table 4. Thermal study of *Plantago Ovata* husk fractions

Sample code	Temp. range (°C)	weight loss (%)	E_a kJ mol ⁻¹	ΔH^* kJ mol ⁻¹	ΔS^* JK ⁻¹ mol ⁻¹	ΔG^* kJ mol ⁻¹	lnA	IPDT (°C)	ITS	weight at 600°C (%)
AANa	250-32	56.79	139.61	134.74	-74.06	179.07	21.22	275.42	0.44	0.401
MNa	230-31	42.46	141.45	138.71	-54.79	169.87	23.51	318.44	0.48	11.221
ENa	230-33	49.10	143.58	138.84	-52.34	168.56	23.81	289.22	0.49	0.981
ANa	255-32	46.60	136.54	131.67	-78.36	176.5	20.71	359.28	0.48	0.535
ClfNa	245-32	48.18	148.78	143.9	-56.79	178.18	23.3	315.99	0.51	14.598

Isolation of gel fractions

The relative amounts of hydrogels isolated from *P. Ovata* husk is, AANa (60 percentage) > ENa (44%)> ClfNa (30%)> MNa (50 %) > ANa (20%) (Figure 1).

DR and *in vitro* buoyancy study

All physical parameters i.e. content uniformity, thickness, weight variation (less than $\pm 5\%$), floats size were found in acceptable limits (Table 5).

Table 5. Physiochemical properties and floating lag time of floats of different fractions

Tablet matrix	Crushing strength (kN)	Content uniformity (%)	Friability (%)	LT (min)
MNa	9.8± 0.5	97.5± 0.5	0.46	42
ENa	9.7± 0.5	96.7± 0.5	0.35	15
AANa	9.7± 0.5	98.9± 0.5	0.46	40
ANa	9.4± 0.5	98.4± 0.5	0.48	42
ClfNa	9.2± 0.5	96.4± 0.5	0.67	35
H	9.4± 0.5	97.6± 1.5	0.68	48
Cstd	9.3± 0.5	97.4± 1.5	0.49	32

Due to best retarding ability of ENa for PRC, it was selected for designing the other formulations. The drug remained within the matrix was relatively greater, therefore new Formulations were designed by changing the amount of variables to increase the availability of drug. The design criteria was as; after 2 h; $25 \pm 5\%$, after 5 h; $45 \pm 5\%$ and after 10 h; $65 \pm 5\%$. 1st batch could not meet the set criteria except S₁F₉, therefore a 2nd batch was designed where DR was also

less than 25% in 2 h but after 5 h it was very close to stated criteria. After 10 h, release rate was less than 65%. Therefore this batch was also rejected. In 3rd batch; the DR% in 2 h and 5 h were very close to set target ($> 25\%$, $> 45\%$) but after 10 h, it was again less than stated criteria. Therefore 4th batch was designed by replacing TG by LG where DR% was $> 25\%$, $> 45\%$ and $\approx 65\%$ for 2 h, 5h and 10h respectively (Table 6).

Table 6. DR of PRC at various interval

Ist batch	S1 _{2h}	S1 _{5h}	S1 _{10h}	2 nd batch	S2 _{2h}	S2 _{5h}	S2 _{10h}
F1	8.8	10.60	27.3	F1	7.6	25.2	39.0
F2	12.5	16.2	30.0	F2	8.5	27.0	42.9
F3	15.4	22.9	39.6	F3	10.4	28.7	45.5
F4	7.2	8.51	26.1	F4	8.5	25.0	36.2
F5	12.1	7.50	35.7	F5	9.3	26.5	38.1
F6	15.9	11.10	42.0	F6	10.0	27.7	41.2
F7	5.3	15.01	19.5	F7	8.4	23.9	34.7
F8	10.7	18.10	35.1	F8	10.9	17.7	37.3
F9	13.8	22.13	42.2	F9	11.8	19.6	41.1
3 rd batch	S3 _{2h}	S3 _{5h}	S3 _{10h}	4 th batch	S4 _{2h}	S4 _{5h}	S4 _{10h}
F1	27.6	43.5	24.2	F1	28.9	46.2	51.6
F2	29.2	47.3	26.8	F2	30.5	48.1	57.1
F3	30.9	48.7	28.1	F3	31.4	42.5	63.0
F4	26.4	42.7	22.5	F4	21.4	44.8	50.2
F5	27.5	44.8	24.7	F5	30.3	48.3	62.5
F6	28.5	77.1	25.6	F6	41.9	50.0	70.6
F7	22.1	41.8	22.6	F7	24.5	34.1	48.1
F8	28.5	43.9	23.8	F8	29.3	46.2	52.0
F9	33.7	46.5	24.2	F9	32.1	52.1	67.9

So this batch was found to be favorable for designing the desired formulations. In 4th batch, the formulation S₄F₉ was found to be best fitted due to minimum LT (Table 7) and maximum DR % sufficient for achieving the drug plasma level (Figure 7).

Kinetic study

DR data with all kinetic parameters are given in Table (8-11).

Table 7. Lag time study of all batches

Fractions	MNa	ENa	AANa	ClfNa	ANa	H	Cstd		
Time (min)	42	15	40	35	40	38	25		
1st batch	F1	F2	F3	F4	F5	F6	F7	F8	F9
Time (S)	140	85	65	150	120	90	175	155	100
2nd batch	F1	F2	F3	F4	F5	F6	F7	F8	F9
Time (S)	140	125	100	160	145	120	180	150	100
3rd batch	F1	F2	F3	F4	F5	F6	F7	F8	F9
Time (S)	105	85	65	85	70	60	150	125	90
4th batch	F1	F2	F3	F4	F5	F6	F7	F8	F9
Time (S)	200	150	135	225	185	160	275	205	160

DR profiles of all batches

Figure 7. DR profiles of designed batche

Table 8. Kinetic studies of 1st batch

		Z- Order		F-Order		H-Model		H-C Model		P- law		
PRC	S _{10h} (%)	k _o	R ²	k ₁	R ²	k _H	R ²	k _{HC}	R ²	k _P	R ²	n
F1	17.3	0.022	0.968	0.00	0.96	0.66	0.89	-0.007	0.968	0.0042	0.905	0.534
F2	19.9	0.029	0.8989	0.00	0.988	0.882	0.95	-0.009	0.989	0.002	0.954	0.666
F3	21.6	0.032	0.993	0.00	0.988	0.959	0.939	-0.01	0.993	0.003	0.947	0.628
F4	16.5	0.022	0.904	0.00	0.894	0.644	0.807	-0.007	0.904	0.003	0.858	0.549
F5	15.2	0.025	0.848	0.00	0.837	0.734	0.735	-0.008	0.848	0.002	0.789	0.586
F6	17.5	0.028	0.845	0.00	0.834	0.817	0.731	-0.009	0.845	0.002	0.782	0.619
F7	13.5	0.019	0.875	0.00	0.866	0.558	0.76	-0.006	0.875	0.005	0.759	0.465
F8	15.1	0.022	0.883	0.00	0.872	0.66	0.773	-0.007	0.883	0.003	0.805	0.538
F9	16.1	0.028	0.830	0.00	0.824	0.802	0.718	-0.009	0.835	0.002	0.756	0.611

*Z= zero,P=power, F=first, H-C= Hixon Crowell

Table 9. Kinetic studies of 2nd batch

		Z- Order		F-Order		H-Model		H-C Model		P- law		
PRC	S _{10h} (%)	k _o	R ²	k ₁	R ²	k _H	R ²	k _{HC}	R ²	k _P	R ²	n
F1	29.0	0.042	0.994	0.00	0.987	1.283	0.947	-0.014	0.994	0.003	0.971	0.666
F2	32.8	0.046	0.992	0.00	0.984	1.414	0.954	-0.015	0.992	0.004	0.977	0.657
F3	35.4	0.051	0.9965	0.00	0.99	1.542	0.963	-0.017	0.996	0.004	0.984	0.653
F4	26.2	0.038	0.994	0.00	0.996	1.181	0.981	-0.012	0.994	0.004	0.992	0.637
F5	28.0	0.038	0.995	0.00	0.996	1.184	0.979	-0.012	0.995	0.006	0.987	0.574
F6	31.2	0.043	0.998	0.00	0.996	1.316	0.965	-0.014	0.998	0.008	0.965	0.549
F7	24.7	0.035	0.988	0.00	0.987	1.077	0.954	-0.01	0.988	0.006	0.961	0.555
F8	27.2	0.035	0.993	0.00	0.997	1.101	0.989	-0.012	0.993	0.011	0.988	0.492
F9	31.1	0.04	0.995	0.00	0.996	1.224	0.982	-0.013	0.995	0.013	0.977	0.479

Table 10. Kinetic studies of 3rd batch

		Z- Order		F-Order		H-Model		H-C Model		P- law		
PRC	S _{10h} (%)	k _o	R ²	k ₁	R ²	PRC	S _{10h} (%)	k _o	R ²	k ₁	R ²	PRC
F1	25.1	0.038	0.988	0.001	0.999	1.155	0.946	-0.012	0.985	0.003	0.966	0.632
F2	26.7	0.041	0.985	0.001	0.985	1.236	0.985	-0.013	0.987	0.005	0.987	0.621
F3	27.9	0.043	0.986	0.001	0.983	1.244	0.996	-0.013	0.981	0.004	0.994	0.595
F4	21.8	0.038	0.946	0.001	0.964	1.186	0.977	-0.012	0.943	0.003	0.975	0.654
F5	24.6	0.038	0.984	0.001	0.997	1.213	0.972	-0.013	0.982	0.004	0.976	0.663
F6	26.0	0.036	0.957	0.001	0.974	1.174	0.985	-0.012	0.951	0.005	0.994	0.582
F7	23.5	0.037	0.933	0.001	0.956	1.119	0.986	-0.01	0.932	0.005	0.993	0.573
F8	24.0	0.039	0.978	0.001	0.994	1.198	0.985	-0.012	0.976	0.006	0.988	0.536
F9	25.6	0.037	0.994	0.001	0.997	1.113	0.987	-0.012	0.991	0.014	0.977	0.462

Table 11. Kinetic studies of 4th batch

		Z- Order		F-Order		H-Model		H-C Model		P- law		
PRC	S _{10h} (%)	k _o	R ²	k _i	R ²	k _H	R ²	k _{HC}	R ²	k _p	R ²	n
F1	31.5	0.046	0.994	0.001	0.985	1.422	0.951	-0.014	0.994	0.004	0.976	0.631
F2	37.2	0.052	0.993	0.001	0.988	1.564	0.957	-0.016	0.994	0.004	0.981	0.635
F3	43.3	0.063	0.994	0.001	0.971	1.866	0.933	-0.022	0.992	0.006	0.964	0.656
F4	30.4	0.038	0.992	0.001	0.994	1.214	0.973	-0.012	0.995	0.005	0.983	0.584
F5	35.6	0.045	0.999	0.001	0.993	1.332	0.962	-0.013	0.997	0.008	0.962	0.536
F6	37.5	0.044	0.997	0.001	0.992	1.406	0.962	-0.014	0.995	0.012	0.951	0.511
F7	28.2	0.039	0.995	0.001	0.997	1.165	0.986	-0.013	0.993	0.009	0.992	0.528
F8	31.9	0.043	0.993	0.001	0.991	1.227	0.972	-0.014	0.992	0.014	0.968	0.477
F9	34.8	0.044	0.996	0.001	0.988	1.357	0.957	-0.016	0.996	0.013	0.963	0.482

Similarity and difference factors

The DR profile of all formulations was compared with sustained release marketed tablet and their similarity factors were found to be >50. From these values (Table 12) it was found that their DR mechanism was

non-Fickian diffusion. The value of difference factor for no difference lie in-between 0-15 but most of the formulations deviated from this range showing that their release pattern is anomalous and need to explore further.

Table 12. Similarity and difference factors

PRC		F1	F2	F3	F4	F5	F6	F7	F8	F9
1 st batch	f1	66.58	69.67	72.36	70.25	73.24	75.95	73.44	74.96	76.61
	f2	67.17	73.59	77.04	65.14	65.15	65.26	57.90	60.41	62.57
Cstd	f1	61.23	64.80	67.95	65.49	68.97	72.10	69.19	70.95	72.85
	f2	50.31	55.45	58.82	49.21	49.30	49.56	45.11	46.67	47.99
2 nd batch	f1	39.98	48.74	56.64	65.14	67.39	70.04	68.75	72.34	74.63
	f2	59.64	53.91	49.45	61.62	58.74	53.28	64.17	57.32	51.70
Cstd	f1	30.36	40.52	49.69	59.55	62.17	65.24	63.74	67.91	70.56
	f2	76.55	85.88	78.49	73.12	79.71	85.61	70.27	79.40	78.99
3 rd batch	f1	14.58	20.85	26.48	26.45	29.60	41.31	23.73	32.11	36.30
	f2	71.69	63.46	60.40	77.58	72.34	67.50	76.11	71.82	66.83
Cstd	f1	10.49	12.51	15.06	14.83	18.32	31.91	11.87	21.23	26.09
	f2	67.56	76.32	78.50	61.63	68.58	73.09	63.34	68.81	70.87
4 th batch	f1	10.03	20.84	31.55	22.77	30.32	36.06	38.81	42.25	45.76
	f2	53.86	49.00	43.26	57.29	50.94	47.50	60.46	55.67	48.79
Cstd	f1	11.20	9.95	20.58	11.88	19.15	25.81	29.00	32.99	37.07
	f2	85.66	79.51	62.77	81.42	85.37	73.77	77.07	92.10	76.49

Regression analysis

The regression analysis was conducted to find the effect of variables on drug release rate. The negative sign of factors in equation indicates that its amount should be decreased while positive sign indicates that

its amount should be increased to get targeted release rate and mechanism. The effect of all variables in all batches is shown in table 13.

*X₁= A, X₂= B, X₁²= C, X₂²= D and X₁X₂= F

i) 1st Batch

2h study

$$S1_{2h} = 0.0137t + 0.013A - 0.043B - 7E^{-05}AB + 7.1E^{-05}A^2 + 0.0008B^2 + 2.82$$

(CRC = 0.913, $R^2 = 0.83$, Adj. $R^2 = 0.785$, n = 27)

5h study

$$S1_{5h} = 0.016t - 0.031A + 0.1005B - 0.0004AB + 6.07E^{-05}A^2 - 0.0003B^2 + 4.79$$

(CRC=0.922, $R^2 = 0.8511$, Adj. $R^2 = 0.8311$, n = 54)

10h study

$$S1_{10h} = 0.0231t - 0.0622A + 0.2501B - 0.0007AB + 0.0001A^2 - 0.0015B^2 + 5.085$$

(CRC = 0.9611, $R^2 = 0.9262$, Adj. $R^2 = 0.9213$, n = 99) (Figure 8). **Figure 8. The Scattered plot for 1st batch is shown in Figure 1(a, c and e).****ii) 2nd Batch**

$$S2_{2h} = 0.046t - 0.0391A + 0.0556B + 0.00034AB + 0.000167A^2 - 0.00056B^2 + 3.349$$

(CRC = 0.97231, $R^2 = 0.94611$ Adj. $R^2 = 0.93022$, n = 27)

5h study

$$S2_{5h} = 0.041t - 0.041A + 0.118B + 0.0008AB + 0.0001A^2 + 0.0096B^2 + 2.853$$

(CRC=0.987, $R^2 = 0.974$, Adj. $R^2 = 0.971$, n = 54)

10h study

$$S2_{10h} = 0.0411t - 0.0651A + 0.09B + 0.0001AB + 0.0002A^2 + 5.2E^{-05}B^2 + 5.039$$

(CRC = 0.995, $R^2 = 0.980$, Adj. $R^2 = 0.979$, n = 99) (Figure 9). **Figure 9. Scattered plot between experimental and predicted release figure 2(a, c and e).**

iii) 3rd batch

2h study

$$S3_{2h} = 0.0463t - 0.03A + 0.266B - 0.004AB + 0.0003A^2 - 0.002B^2 + 2.27$$

(CRC = 0.941, $R^2 = 0.908$, Adj. $R^2 = 0.877$, n = 27)

5h study

$$S3_{5h} = 0.0397t - 0.09A + 0.334B - 0.007AB + 0.0003A^2 - 0.002B^2 + 5.6724$$

(CRC=0.989, $R^2 = 0.9696$, Adj. $R^2 = 0.9578$, n = 54)

10h study

$$S3_{10h} = 0.0363t - 0.1360A + 0.3875B - 0.0005AB + 0.0004A^2 - 0.0037B^2 + 9.1875$$

(CRC = 0.9919, $R^2 = 0.9839$, Adj. $R^2 = 0.9827$, n = 99) (Figure 10).

Figure 10. Scattered plot between experimental release and predicted release figure 3(a, c and e)

iv) 4th batch

2h study

$$S4_{2h} = 0.043t - 0.047A + 0.333B - 0.0001AB + 0.0001A^2 - 0.004B^2 + 3.375$$

(CRC = 0.8811, $R^2 = 0.7832$, Adj. $R^2 = 0.7182$, n = 27)

5h study

$$S4_{5h} = 0.0421t - 0.096A + 0.423B + 6.6E-05AB + 0.000277A^2 - 0.006B^2 + 5.738$$

(CRC=0.96, $R^2 = 0.9194$, Adj. $R^2 = 0.984$, n = 54)

10h study

$$S4_{10h} = -0.0068A + 1.598B - 0.001AB - 9E-06A^2 - 0.013B^2 + 15.063$$

(CRC = 0.77621, $R^2 = 0.592$, Adj. $R^2 = -0.0833$, n = 9) (Figure 11).

Figure 11. Scattered plot between experimental and predicted release for 4th batch

The all given plots indicate best correlation between theoretical and experimental drug

release profiles.

Table 13. Effect of all variables on DR profiles of formulations of all batches at different time intervals

1st batch	
2 h study	B(2.205)>A ² (0.024)>AB(0.023)>A(0.021)>B ² (0.006)
5 h study	A(0.358)>AB(0.238)>B(0.230)>A ² (0.208)>B ² (0.033)
10h study	A(0.327)>B(0.263)>A ² (0.233)>AB(0.203)>B ² (0.064)
2nd batch	
2 h study	B(3.727)>AB(0.096)>A(0.054)>A ² (0.046)>B ² (0.008)
5 h study	A(0.261)>A ² (0.259)>B(0.224)>AB(0.1749)>B ² (0.13)
10h study	A(0.215)>A ² (0.193)>B(0.093)>AB(0.037)>B ² (0.002)
3rd batch	
2 h study	B(13.67)>AB(0.157)>A(0.057)>A ² (0.0463)>B ² (0.02)
5 h study	A ² (0.619)>A(0.59)>B(0.437)>AB(0.232)>B ² (0.1154)
10h study	A(0.552)>A ² (0.535)>B(0.314)>B ² (0.120)>AB(0.111)
4th batch	
2 h study	B(25.997)>A(0.076)>B ² (0.076)>A ² (0.050)>AB(0.04)
5 h study	B(0.799)>B ² (0.687)>A(0.601)>A ² (0.524)>AB(0.031)
10h study	B(3.20)>B ² (2.336)>AB(0.776)>A(0.045)>A ² (0.018)

Conclusions

All fractions are polysaccharides in nature and due to having swelling, reswelling properties these can be used for designing the sustained release formulations and due to low density they can float over the surface of dissolution medium and hence are suitable for gastro retentive drug release tablets/floats. The *Plantago Ovata* husk gel has many pharmaceutical properties and is successfully used in diseases like constipation, hyperglycemia and coronary heart diseases. Due to being herbal medicine, they can be successfully applied for designing the controlled release floats. The full factorial design 3² reveals that all independent variables shows significant effect on DR rate, lag time and floating time of floats. The SRA (standard regression analysis) shows that the A and B are basic parameters and their alteration can be helpful to achieve the desired DR profile. The positive and negative signs in regression

analysis indicates that the value of that variable must be decreased or increased to get required DR profile. Similarity and difference factor is helpful in finding the resemblance or difference with standard DR profile. The thermal study indicates that ENa is thermally stable can withstand environmental changes. The GPC study of these fractions revealed that their PDI values are changing from 2 to 3.74 showing that polymers are poly disperse in nature. All excipients used were herbal in nature and biocompatible to body and are included in GRAS list. Due to these exceptional properties, we can conclude that these fractions are totally safe to use as matrix formation in CDDS, and also have a potential as food preservative films [25]. The values of R² from kinetic study and RA are helpful in interpreting the nature of matrix. In the last formulation, the value of R² for power law 0.965 with n value $\approx$ 0.482 indicates best fit to required release

mechanism. So this formulation was considered to be optimized formulation. Same values or even better values for R^2 are also seen in some other formulations but their DR rates or mechanism are not best fitted to targeted criteria (swelling/diffusion).

Authors' contributions

Conceived and designed the experiments: A Karim, Performed the experiments: A Karim, Analysis of data: A Karim, Contributed reagents /materials/ analysis tools: T Mehmood, Wrote the paper: A Karim & MA Shaheen.

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