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Evaluation of acrylamide grafted moth bean starch as controlled release excipient

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ABSTRACT

The aim of this study was to investigate the applicability of acrylamide grafted moth bean starch as controlled release matrix former. Lamivudine was used as model drug and its controlled release tablets were formulated using various concentration of grafted copolymer. The grafted copolymer was tested for acute toxicity and drug–excipient compatibility study. The formulations were evaluated for physical characteristics like hardness, friability, % drug content and weight variations. The *in vitro* release study showed that the optimized formulation exhibited highest correlation (R) value in case of Higuchi model and the release mechanism study proved that the formulation showed a combination of diffusion and erosion based release process. There was a significant difference in the pharmacokinetic parameters (T_{max}, C_{max}, AUC, V_d, $T_{1/2}$ and MDT) of the optimized formulation as compared to the marketed conventional tablet Lamivir®, which confirmed controlled release potential of acrylamide grafted copolymer.

1. Introduction

In the last few years, novel synthetic techniques have been used to impart desirable chemical, physical, and biological properties to biomaterials. Materials have either been synthesized directly, so that desirable chain segments or functional groups are built into the material, or indirectly, by chemical modification of existing structures to add desirable segments or functional groups. Polymeric biomaterials can be produced by copolymerization of monomers to achieve nearly monodispersed polymers. It is possible to produce polymers containing specific hydrophilic or hydrophilic entities, biodegradable repeating units, or multifunctional structures that can render them into three-dimensional expansion of networks (Qiu & Park, 2001). Reservoir and matrix type tablets are the most commonly used orally administrated sustained release preparations. Especially matrix tablets, which are produced by direct compression using either hydrophilic polymer such as natural gums, HPMC, CMC, Carbopol or hydrophobic polymer like ethyl cellulose and amylodextrin, which are relatively easy to manufacture. For many reasons, oral drug delivery continues to be the preferred route of administration of drug substances. During the last two decades, polymers which swell in aqueous media have been used for preparation of oral sustained release dosage forms. Among the drug delivery systems, specifically among the diffusion-controlled and the dissolution-controlled delivery system, some of the most interesting are hydrophilic matrix systems, because they are easy to formulate, inexpensive and easy to produce, and have a good *in vitro–in vivo* correlation (Beneke, Vilijoen, & Hamman, 2009). Lamivudine (LAM) is the first nucleoside analogue approved to treat chronic HBV infection and AIDS. Conventional oral formulations of LAM are administered two times a day 150 mg each time because of its moderate half-life ($t_{1/2}$ = 5–7 h). Treatment of AIDS using conventional formulations of LAM is found to have many drawbacks, such as adverse side effects resulting from accumulation of drug in multi-dose therapy, poor patient compliance, and high cost (Perry & Faulds, 1997). The aim of the present study was to establish the grafted moth bean starch (GMBS) as controlled release excipient in the controlled release tablets of Lamivudine. In the present study lamivudine controlled release tablets were formulated using direct compression technique and evaluated as hydrophilic inert matrix former. The controlled release tablets were evaluated for various physical characteristics, *in vitro* dissolution profile and *in vivo* pharmacokinetic parameters in rabbit model.

2. Materials and methods

2.1. Materials

Lamivudine (LAM) was kindly gifted by Ranbaxy Limited, Paonta Sahib, Himachal Pradesh, India. Acrylamide, ceric ammonium nitrate (CAN) and hydroxyquinone were purchased from SD Fine Chem, India. Spray dried lactose (SDL) was kindly donated by DMV Fonterra excipients, The Netherland. Polyvinylpyrrolidine K-30 (PVP K-30), magnesium stearate and talc were purchased from Loba Chemie Limited, Mumbai, India. All other chemicals used were

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of analytical grade and purchased from SD Fine chemical limited, Mumbai, India.

Digital weighing balance (Sartorius, Germany), Rotary ten station tablet punching machine (Shakti Engineering Limited, Ahmedabad, India). UV-Visible Spectrophotometer (UV-1700, Shimadzu, Japan), Differential scanning calorimetry (Perkin Elmer, JADE DSC, USA), HPLC (Waters®, USA), Eight basket digital *in vitro* USP dissolution apparatus (Electrolab, Mumbai, India), Monsano hardness tester, Roche friabilator (Campbell electronics, Mumbai, India) and Laboratory scale stability chamber (Model TH-90 S/G, Thermolab, Mumbai, India).

2.2. Microwave assisted acrylamide grafted starch

Moth bean starch (MBS) was pregelatinized by heating the aqueous dispersion of starch at 75 °C. The pregelatinized MBS was grinded and sieved through 100 mesh. Pregelatinized MBS (1.0 g) was dispersed in 50 mL of double distilled water. Various amounts of acrylamide (2.5–10.0 g) were dissolved in 15 mL of water and were added to the starch dispersion. They were mixed well using stirrer and the mixture was transferred to a conical flask (250 mL) and 0.25 g of CAN was added. The flask was subsequently placed on the turntable of a microwave oven (CE1111L, Samsung Electronics, India) and the flask was irradiated at various level of power (250–600 W). After formation of a gel mass the flask was placed in ice cooled water. The flask was kept undistributed for 12 h to complete the grafting reaction. After 12 h, 0.5 mL saturated hydroxyquinone solution was added to terminate the grafting reaction. The gel mass was poured into excess of acetone. The grafted copolymer was purified by solvent extraction method using a mixture of formamide and acetic acid (1:1). The resulting precipitate of graft copolymer was collected and was dried in hot air oven. The grafted material was grinded until a homogenous powder was obtained (Singh & Nath, 2011a, 2011b; Singh, Nath & Guha, 2011).

2.3. Acute toxicity study of grafted copolymer

Healthy male and female Swiss albino mice (8 weeks) used for the acute oral toxicity study were breed and reared at the Animal House of Gayatri College of Pharmacy, Orissa. The animals were housed in polypropylene cages and provided with bedding of clean paddy husk. The animals were acclimatized to laboratory conditions for one week prior to experiment. The temperature in the animal house was maintained at 25 ± 2 °C with a relative humidity of 30–70% and illumination cycle set to 12 h light and 12 h dark. The mice were fed with standard laboratory pelleted feed (M/s Gold Mohur Foods and Feeds Ltd., Bangalore, India). All the mice of both the sexes were fasted overnight before experimentation and were allowed to take food 1 h after the experiment. Modified starch was administered orally at a dose of 2000 mg/kg b.w. in distilled water. The animals were observed for any mortality and morbidity (convulsions, tremors, grip strength and pupil dilatation) at an interval of 12 h for 14 days. This study was approved by the Animal Ethical Committee of Gayatri College of Pharmacy (Reg. No. 1339/ac/10/cpcsea).

2.4. Preformulation study

2.4.1. Drug–excipient compatibility study by DSC

A differential scanning calorimetry (JADE DSC, Perkin Elmer, USA) was used to study the thermal analysis of drug–excipient compatibility. Firstly, binary mixtures of lamivudine and excipients (in 1:1 mass/mass ratio) were prepared by using physical mixture technique. The drug–excipient mixture was scanned in the temperature range of 50–220 °C under an atmosphere of nitrogen. The heating rate was 20 °C/min and the obtained thermograms were observed for any type of interaction.

Table 1
Corresponding peak temperatures and enthalpy values of lamivudine in various drug–excipient mixtures in DSC study.

Sample	Ratio (drug:excipient)	T_{onset} (°C)	T_{peak} (°C)	ΔH (J g^{-1})
LAM	–	177.40	182.73	74.54
LAM + GMBS	(1:1)	167.40	178.03	34.27
LAM + PVP K-30	(1:1)	174.32	180.09	72.43
LAM + SDL	(1:1)	171.56	179.44	48.78
LAM + Mag. Stearate	(1:1)	177.63	182.91	80.66
LAM + Talc	(1:1)	178.64	183.32	98.38

2.4.2. Isothermal stress testing (IST) analysis

In isothermal stress testing (Singh & Nath, 2011a, 2011b) samples of drug and different excipients (Table 3) were weighed directly in 5 mL glass vials (n = 3). After mixing on a cyclomixer for 3 min, 10% (w/w) water was added in each of the vial. The glass vials, after teflon sealing, were stored at 50 °C in hot air oven. Drug–excipient blends without adding water and stored in refrigerator served as controls. The drug–excipient blends were periodically examined for any change in physical appearance. Samples were quantitatively analyzed using UV-Vis spectrophotometer (Pharmaspec 1700, Shimadzu, Japan) after 4 weeks of storage at above conditions.

2.5. Formulation of tablets

The lamivudine controlled release tablets were formulated by incorporating selected excipients using model drug lamivudine. Lamivudine, grafted moth bean starch (GMBS), spray dried lactose and PVP K-30 were dispensed accurately. Each ingredient was shifted through #80 sieves, transferred in to a polyethylene bag and mixed for 20 min. The dry mixture was evaluated for flow properties and subjected to direct compression. The slugs prepared were crushed into granules and the mass was shifted through #20 sieves, weighed and subjected to the evaluation of flow properties. The quantity of remaining ingredients of the Table 1 i.e. magnesium stearate and talc required for tablets was calculated as per the formula of the Table 1 to compensate any loss during the direct compression process. The lubricated dry powder was compressed into tablets at 8 kg/cm^2 pressure by using 10 mm standard concave and flat punch set in 10 station rotary tablet punching machine. The tablets were double wrapped in polyethylene bag till further study.

2.6. Evaluation of tablet

The tablets from each batch was picked randomly in order to evaluate the weight variation, the hardness, % drug content and the friability (Banker & Andsi, 1987). The hardness and friability of the tablet were measured using Monsanto hardness tester and Roche friabilator respectively.

2.7. In vitro dissolution rate study

In vitro dissolution rate study of the formulations (n = 6) was carried out in USP Type-II dissolution rate test apparatus. Dissolution rate study was performed in simulated gastric fluid (0.1 M HCl) and in simulated intestinal fluid (pH 6.8) for first 2 h and for successive 10 h respectively. The each dissolution medium (900 mL) was maintained at 37 ± 0.5 °C throughout the study. The samples (5 mL) were withdrawn at predetermined time (1–4, 6, 10, and 12 h) and replaced with an equivalent volume of fresh medium. The samples

were filtered through membrane filter (0.45 μm) and analyzed by UV-visible spectrophotometer at 270 nm (λ_{max}). The cumulative percent drug release was plotted against time to determine the release profile.

2.8. Kinetics of drug release

The kinetics of drug release (Costa & Loba, 2001; Hadjiioannou, Christian & Koupparis, 1993; Higuchi, 1963; Korsemeyer, Gunny, Doelker, Buri & Peppas, 1985) is important because it is an useful tool to correlate the *in vitro* drug release and *in vivo* drug responses or to compare the results of pharmacokinetics with dissolution profiles of the formulations. Different mathematical models i.e. zero order, first order, Higuchi and Korsemeyer–Peppas equations were applied for describing the kinetics of the drug release process from controlled released tablets of LAM, the most suited being the one which fitted best in the experimental results.

2.9. Pharmacokinetic study

The pharmacokinetic study of optimized controlled release tablet (F4) of lamivudine was carried out in two groups of six male white albino rabbits each weighing 1.5–2.5 kg. All animals were fasted overnight (12 h) before dosing and continued till 4.0 h after administration of tablets, thereafter rabbit chew diet was provided ad libitum. Drinking water was deprived of before dosing and continued till 2 h of post dose, thereafter it was provided ad libitum. The tablets ($n = 3$) of batch, F4 were administered orally to three rabbits of each group along with 10 mL of water by using feeding tube.

The blood samples each of about 50 μL from each animal were collected from orbital sinus into the microcentrifuge tubes containing 50 μL of 10% w/w of disodium EDTA as anticoagulant according to the sampling schedule (pre dose, 0.25, 0.5, 1.0, 2.0, 3.0, 4.0, 6.0, 9.0, 12.0 and 24.0 h). The collected blood samples were centrifuged immediately at 1000 × *g* for 10 min. The supernatant plasma layer was separated and stored at −20 °C till analysis.

2.9.1. Plasma sample analysis

The 200 μL of each sample was taken into 2 mL centrifuge tube and 50 μL of nelfinavir solution (50 μg/mL) was added as an internal standard (IS). The mixtures were vortexed for 10 s. Acetonitrile (1.5 mL) was added into the mixture, vortexed for 3 min and centrifuged at 10,000 rpm for 10 min. The supernatant was transferred into a glass centrifuge tube and evaporated to dryness at 45 °C under a stream of nitrogen .The residue was reconstituted with 200 μL of reconstitution solvent (mobile phase). The samples were filtered through 0.45 μm membrane filter using syringe filter. An aliquot of 20 μL of the sample was injected into the injector of the HPLC system. The samples were analyzed by using the in house developed chromatographic condition. The area under the curve of peaks of LAM and IS was determined and the concentration of drug present in sample was estimated by using the linear regression equation of standard calibration curve (concentration of LAM vs ratio of LAM to IS. The amount of drug present in 200 μL of plasma [(quantity obtained from linear regression equation/20) × 1000)] was calculated.

2.9.2. Determination of pharmacokinetic parameters

The pharmacokinetic parameters were determined from the data of plasma drug concentration at different time points by using MS-Excel 2003 software according to the procedure described elsewhere (Bourne, 2002; Jambhekar & Breen, 2009; Singh, Nath & Pani, 2011).

2.10. Stability study

Stability study is used to assess expiration dating and storage conditions for pharmaceutical products. Stability study of lamivudine controlled release tablets were performed as per ICH guideline. The optimized controlled release tablets were kept in polypropylene bottle and stored in stability chamber maintained at 40 °C ± 2 °C and 75% ± 5% RH for six months. Samples were checked initially, after three months and further after six months.

3. Results and discussion

3.1. Synthesis of acrylamide grafted copolymer

The acrylamide grafted copolymer was successfully synthesized on starch backbone in the presence of catalyst (CAN) was occurred by free radical reaction mechanism. The grafted copolymer was found more hydrophilic as number of free hydrogen group was increased after acrylamide grafting.

3.2. Acute toxicity study

The purpose of this study was to evaluate toxicity profile of the modified starch. A 14 day acute oral toxicity study was performed in swiss albino mice. The LD_{50} of all the modified starches were not further studied as they were found to be safe up to 2000 mg/kg on 24 h study basis. It was observed that the animals fed with the modified starch were found healthy. No unusual changes in behavior, or in locomotor activity, ataxia, and signs of toxicity were observed during the 14 days period. No differences were found in growth behavior between the control and treatment group in 14 days study. The body weight of male and female swiss albino mice was found to be normal after treatment. There was no change observed in body weight of control and modified starch treated mice.

3.3. Preformulation study

3.3.1. Drug–excipient compatibility study by DSC

DSC thermograms of drug and drug–excipient mixtures with their corresponding peak temperatures and enthalpy values (ΔH) of lamivudine (LAM) with various excipient mixtures are summarized in Table 1 DSC thermogram of LAM showed a sharp endothermic peak at 182.73 °C corresponding to its melting point. The endothermic peak of the drug was well retained in majority of cases. However, in some combinations there were slight change in peak temperature and peak shape, which might be due to reduction of the purity level of component in mixtures, on mixing of excipients with the drug.

3.3.2. Isothermal stress testing study

In the isothermal stress testing, drug–excipient binary mixtures showed no change in physical appearance at ambient temperature. The blends remain physically stable and no discoloration, liquefaction or gas formation was observed during storage. There was no significant drug degradation observed in the excipients. Table 2 showed percent drug remaining at the end of the study at 50 °C.

3.4. Formulation of controlled release tablets

The formulae of the Lamivudine (LAM) controlled release tablets have been showed in Table 3. Spray dried lactose was selected as direct compression diluents by considering its advantages in terms of good compressibility, easy availability, cost effectiveness and low moisture sensitivity. PVP K-30 was used as dry binder considering its widespread applicability in industry and relative low moisture sensitivity. Magnesium stearate and talc was used as lubricant and

Table 2
Results of UV Analysis of the drug–excipient mixtures, under isothermal stress testing after 4 weeks of storage.

Sample	Ratio (drug:excipient)	% drug remaining[a]		Change in physical appearance
		Control[b] sample	Stressed[c] sample	
LAM	–	101.12 ± 3.2	99.97 ± 3.1	No
LAM + GMBS	1:2	100.13 ± 1.5	99.09 ± 2.4	No
LAM + SDL	1:2	102.51 ± 2.1	102.34 ± 0.7	No
LAM + PVP	1:1	103.67 ± 2.2	101.87 ± 1.1	No
LAM + Magnesium stearate	1:1	101.45 ± 1.5	100.12 ± 2.2	No
LAM + Talc	1:1	101.22 ± 4.1	100.02 ± 1.1	No

[a] Values expressed as average ± standard deviation ($n = 3$).
[b] Drug excipient blends without added water and stored in refrigerator.
[c] Drug excipient blends with 10% added water and stored at 50 °C for 4 weeks.

glidant respectively due to their widespread applicability in industry and relative low moisture sensitivity. Highly substituted grafted moth bean starch (GMBS) was used as hydrophilic polymeric carrier material for the production of oral controlled release tablets. The batches of tablets from F1 to F6 were prepared to select the suitable grades of GMBS. The tablets were formulated to attain 8 kg/cm^2 hardness with varying concentration of GMBS (25–150 mg/tablet). The tablets were formulated without any processing difficulties (stickiness, lamination, capping and picking).

3.5. Evaluation of physical parameters of tablets

3.5.1. General appearance and thickness

The tablets of all the batches were white in color, flat, round shaped and plane from both sides. The thickness (4.11 ± 2.2 mm to 4.16 ± 1.5 mm) of all the batches of tablets is shown in Table 4.

3.5.2. Weight variation

The average weight of 20 tablets along with standard deviation of entire formulations has been presented in Table 4. The percentage of weight variation of individual tablets from the average weight was found to be within ±5% w/w which proved that the entire tablets have passed the USP weight variation test.

3.5.3. Drug content

The drug content of all the tablets in each batch was found to be in the range of (101.98 ± 2.4% to 103.34 ± 1.2%) as shown in Table 4. The results indicated that tablets of entire batches have passed the USP criteria for the drug content of tablets.

3.5.4. Hardness

The hardness of tablets of entire batches was found to be in the range of 6.2 ± 1.1 kg/cm^2 to 6.4 ± 2.0 kg/cm^2 and the results are showed in Table 4.

3.5.5. Friability

The results of the friability test of entire formulations are showed in Table 4. It was observed that the tablets of entire batches had passed USP criteria of friability testing (<1.00% w/w). The results revealed that the tablets possessed good mechanical strength.

Table 3
Composition of lamivudine controlled release tablets using GMBS.

Ingredients (mg)	Formulation code					
	F1	F2	F3	F4	F5	F6
Lamivudine 100	100	100	100	100	100	
Grafted moth Bean starch	25	50	75	100	125	150
PVP K-30	15	15	15	15	15	15
Spray dried lactose	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.
Magnesium stearate	3	3	3	3	3	3
Talc	3	3	3	3	3	3
Total weight	300	300	300	300	300	300

qs = quantity sufficient.

3.6. In vitro drug release study

The results ($n = 6$) of *in vitro* drug release study of all the batches of tablets are shown in Fig. 1. From the drug release profile of the batches from F1 to F3 it was seen that the total amount of drug was released within 6 h. Release profile of batch F4 showed a superior fit to the required drug release profile among all the batches. The F5 and F6 batch (with highest concentration of GMBS) also showed a controlled release pattern but significantly a less amount of drug was released in 12 h study so they were not considered for further study. The result of drug release profile of the batch F4 showed 25% w/w of drug released during the first 2 h, while 63% w/w drug was released within 6 h and the remaining 37% w/w of the drug was released in the next 6 h. Hence, a controlled release pattern of drug was observed from the batch F4 throughout the 12 h of dissolution study.

3.7. Kinetics and mechanism of drug release

The release rate constant was calculated from the slope of the appropriate equations and the correlation coefficient (R) was determined for all formulations (Table 5). The *in vitro* drug release of optimized batch (F4) was best fitted and explained by zero order ($R_O = 0.9785$), followed by Higuchi kinetics ($R_H = 0.9673$), and finally first order ($R_F = 0.871$) kinetics. This explains that the batch F4 found to be best fitted in zero order kinetics as compared to other kinetic study. All other batches showed neither good (high) correlation value (R) nor acceptable K value (low). This explains that the controlled release tablets prepared with GMBS with highest grafting value released drug due to swelling of the polymers followed by diffusion controlled mechanism. It is assumed that the hydrophilic matrices, when came in contact with the solvent they

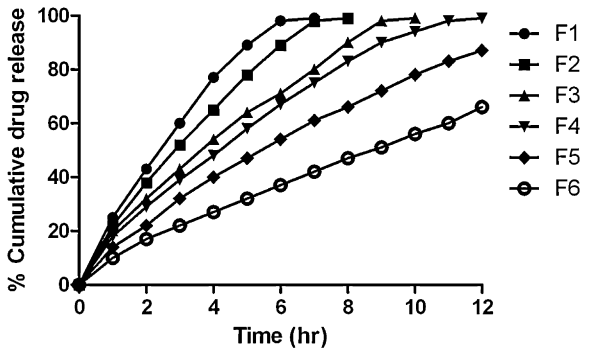

Fig. 1. *In vitro* dissolution rate profile of all the batches of controlled release tablet.

Table 4
Physical properties of lamivudine controlled release table using grafted moth bean starch as release retardant.

Batches	Drug content (%)	Weight deviation (%)	Hardness (kg/cm^2)	Friability	Thickness (mm)
F1	103.2 ± 1.4	303 ± 1.4	6.4 ± 1.4	0.43 ± 2.3	4.16 ± 1.5
F2	101.98 ± 2.4	302 ± 2.3	6.3 ± 1.7	0.34 ± 0.7	4.15 ± 1.2
F3	102.56 ± 1.3	305 ± 1.6	6.3 ± 2.1	0.32 ± 2.1	4.13 ± 1.4
F4	102.14 ± 1.4	309 ± 1.7	6.4 ± 2.0	0.41 ± 1.7	4.14 ± 3.1
F5	103.34 ± 1.2	304 ± 2.4	6.3 ± 1.3	0.35 ± 2.5	4.11 ± 2.2
F6	102.98 ± 2.4	299 ± 2.4	6.2 ± 1.1	0.30 ± 1.4	4.13 ± 2.1

All values represent mean ± standard deviation ($n = 3$).

Table 5
Comparative release kinetics parameter of all the batches of controlled release tablets.

Formulation code	Release kinetic Parameters							
	Zero order		First order		Higuchi		Korsemeyer and Peppas	
	R_O	K	R_F	K	R_H	K	R_p	n_p
F1	0.952	14.44	0.9091	−0.288	0.9722	41.08	0.9886	0.74
F2	0.967	16.53	0.8803	−0.243	0.9721	38.3	0.9951	0.749
F3	0.976	9.74	0.8419	−0.182	0.9752	33.85	0.9984	0.716
F4	0.9647	8.2	0.893	−0.153	0.978	31.93	0.9992	0.716
F5	0.9815	7.01	0.9774	−0.07	0.9745	27.01	0.9984	0.751
F6	0.9917	5.15	0.9898	−0.036	0.9622	19.63	0.9983	0.751

became hydrated and form a gel and model drug LAM was diffused from the gel. The drug release mechanism from controlled release devices is very complex to explain and still not yet completely understood. Although some controlled release processes may be classified as either purely diffusion or purely erosion controlled and many others can only be interpreted as being governed by both the mechanism. To evaluate the *in vitro* drug release profile the data at various time points were fitted into the Korsemayer–Peppas equation to know the mechanism of the drug release. With an n_p value of 0.5, the equation becomes equal to the square root model described by Higuchi, which signifies that drug release from the matrix is governed by Fickian diffusion, for $n_p > 0.5$, anomalous non Fickian drug diffusion occurs i.e. combination of both diffusion or swelling and erosion mechanism takes place. For $n_p > 1$, non-Fickian case-II, erosion controlled or zero order release kinetics is followed. The R_p and n_p values of various trial batch formulations are showed in Table 4. The R_p values of 0.9886, 0.9951, 0.9984, 0.9992, 0.9984, and 0.9983 for the tablets of batches F1, F2, F3, F4, F5 and F6 respectively showed good linearity between log cumulative amount drug releases versus log time. The R_p value of all the tablets was found to be linear and the highest linearity was observed with the batch F4, where the concentration of GMBS as release retardant polymer was quite high. The n_p value was found to be 0.740, 0.749, 0.716, 0.716, 0.751, and 0.751 for the tablets of batches F1, F2, F3, F4, F5 and F6 respectively. Hence, the mechanism of drug release from the tablets was predicted from Korsemayer–Peppas equations and from the obtained n_p values of all the batches of tablets ($n_p > 0.5$) it is revealed that the mechanism of drug release was a coupling of both the process of diffusion and erosion.

3.8. In vivo pharmacokinetics

The results of the plasma drug concentration at different time intervals, after administration of controlled release tablets containing 100 mg of lamivudine to three rabbits, are presented in Table 5. The drug was remained in the body of the animals up to 24 h of administration of the tablet. The data of plasma drug concentration at various time intervals was analyzed by one way analysis of variance (ANOVA). A value of $p < 0.05$ was considered statistically significant (Fig. 2). The pharmacokinetic parameters were derived from plasma drug concentrations versus time profile of all the subjects and the results are shown in Table 6. The T_{max} of all the subjects in case of controlled release formulation was found to be 4.0 h, as compared to 1.0 h for the marketed conventional tablet (Lamivir®), which indicated the slow absorption rate from the controlled release tablets due to extended release effect of hydrophilic polymer present in controlled release tablets. The average C_{max} value of the controlled release tablets was decreased as compared to conventional tablets (from 12784.913 ± 2.1 ng/mL to 8123.31 ± 4.1 ng/mL). The $AUC_{0-\infty}$ of controlled release tablets exhibited high value (57926.78 ± 2.8 ng h/mL) as compared to conventional tablets (54206.28 ± 1.9 ng h/mL). The mean $AUMC_{0-\infty}$ of controlled release tablets was found to be 298573.80 ± 4.0 ng h/mL 227067 ± 0.6 ng h/mL, which is higher than that of the conventional tablets. The plasma half-life has been increased from 3.594 ± 0.2 h (Lamivir®) to 7.0 ± 2.2 h (F4), which confirmed higher duration of action of the drug in systemic circulation from the controlled release tablets (F4). The mean V_d and clearance values for the tablets were 15.98 ± 4.1 L and 1.721 ± 1.1 L/h respectively. The *Ka* and *Ke* value for controlled release tablets were found to be 0.698 ± 2.6 h^{-1} and 0.099 ± 3.1 h^{-1} which are lower than Lamivir® (2.142 ± 1.8 and 0.193 ± 1.5). The mean residence time (MRT) of controlled release tablets was found to be higher (5.170 ± 2.5 h) than that of the conventional tablets (4.189 ± 2.1 h) confirming the controlled release property of the GMBS. The results revealed that the drug was made available to the body in a controlled release manner and the controlled release effect was due to the presence of higher proportion of GMBS in the tablets.

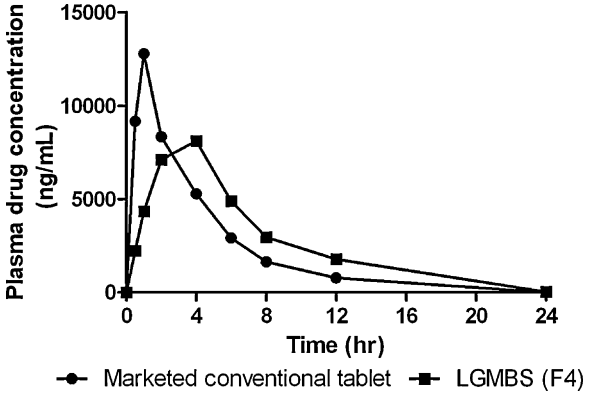

Fig. 2. Comparative *In vivo* pharmacokinetic study of marketed conventional tablet (Lamivir®) and optimized formulation (F4).

Table 6
Pharmacokinetic parameters of marketed and optimized lamivudine controlled release tablet (F4) after a single oral dose to rabbits ($n = 3$).

Pharmacokinetic parameters	Observed value (Lamivir®)	Observed value (F4)
Maximum plasma concentration, C_{max} (ng/mL)	12784.91 ± 2.1	8123.31 ± 4.1
Time required to reach maximum plasma concentration, T_{max} (h)	1.00 ± 1.6	4.00 ± 1.7
Area under curve at 24 h, AUC $(_{0\rightarrow\infty})$ (ng h/mL)	54206.284 ± 3.7	57926.78 ± 2.8
Area under momentum curve At 24 h, AUMC $(_{0\rightarrow\infty})$ (ng h^2/mL)	227067.968 ± 0.6	298573.80 ± 4.0
Volume of distribution, V_d (L)	9.574 ± 2.8	15.98 ± 4.1
Plasma half life ($T_{1/2}$) (h)	3.594 ± 0.2	7.00 ± 1.8
Absorption rate constant, *Ka* (h^{-1})	2.142 ± 1.8	0.698 ± 2.6
Elimination rate constant, *Ke* (h^{-1})	0.193 ± 1.	0.099 ± 3.1
Mean residence time, MRT (h)	4.189 ± 2.1	5.170 ± 2.5
Clearance, Cl (L/h)	1.846 ± 3.0	1.721 ± 1.1

3.9. Stability study

The selected optimized formulation (F4) was evaluated for various parameters (drug content and dissolution study) after 3 and 6 months of storage at accelerated stability conditions (40 ± 2 °C and 75% ± 5% RH). There was no significant amount of change observed in the drug content of tablets after 6 months of storage at accelerated stability conditions. The dissolution profile of formulation at initial stage was considered as reference for dissolution study. The results obtained revealed that the dissolution profile of the formulation after 6 months of storage at accelerated condition was found to be similar to that of the reference one. Based on the results it was opined that the tablets of batch F4 is stable after 6 months of storage at accelerate stability condition.

4. Conclusion

The present study focussed on the formulation of 12 h controlled release tablet of lamivudine using GMBS as matrix forming hydrophilic polymer. It was summarized from the dissolution study of the entire batches that the tablets contain less concentration of GMBS were disintegrated with in 1.0 h and unable to control the release of drug. At higher concentration the tablets released the drug in controlled manner over 12 h study. The study of release mechanism exhibited anomalous non-Fickian diffusion which involved both diffusion and erosion processes. The pharmacokinetic study of the optimized batch (F4) in rabbits was also carried out in triplicate. The plasma drug concentration versus time interval was estimated by using in house developed RP-HPLC method. The optimized batch F4 exhibited first order rate kinetics in absorption of drug from the tablets. The increase in T_{max} value and decrease in C_{max} value in case of the optimized formulation F4, from the values of the conventional tablets of lamivudine (Lamivir®) revealed that the GMBS in the controlled release tablets rendered the drug to be released in a controlled manner.

Conflict of interest

The authors declared no conflict of interest.

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