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Amoxycillin Trihydrate Floating-Bioadhesive Drug Delivery System for Eradication of *Helicobacter pylori*: Preparation, *In Vitro* and *Ex Vivo* Evaluation

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Abstract

The aim of the present investigation was to formulate and evaluate floating-bioadhesive tablets of amoxicillin trihydrate (ATFBT), to provide local action in the treatment of *Helicobacter pylori* (*H. pylori*) by prolonging the gastric residence time. The FBT were prepared by direct compression method using hydroxy propyl methyl cellulose (HPMC K_4M) / chitosan (CH), carbopol (CP974P) / polymethacrylic acid (PMA) as release retarding agent / bioadhesive respectively, sodium bicarbonate ($NaHCO_3$) as a gas-former. The prepared tablets were evaluated for their physical characters such as drug content, *in vitro* buoyancy, swelling index, drug release and stability. Further, the bioadhesive strength (BS) was determined using porcine gastric mucosa. DSC studies indicate the compatibility of the drug and excipients used in the formulation. The floating lag time, total floating time, bioadhesive strength and swelling index of the optimized formulation (F7) was 32 ± 2.7 sec, more than 12 h, 1.86 ± 0.14 N and more than 3.5 respectively. Formulation (F7) was also found to be physically stable when stored at 40°C / 75% RH for 3 months. The drug release profiles of all the formulations were fitted into various kinetic models. The optimized formulation followed the Peppas model ($r^2 > 0.99$) with a non-Fickian diffusion mechanism (n = 0.625). From the results, a matrix floating-bioadhesive tablet incorporating an insoluble active substance such as amoxicillin trihydrate was developed for the successful eradication of *H. pylori*.

Keywords: Amoxycillin trihydrate; Floating-bioadhesive tablets; *Helicobacter pylori*; HPMC K_4M; Drug release; Bioadhesvie strength

Abbreviations: ATFBT: Amoxicillin Trihydrate Floating-Bioadhesive Tablets; BS: Bioadhesive Strength; HPMC: Hydroxy Propyl Methyl Cellulose; CH: Chitosan; CP974P: Carbopol; PMA: Polymethacrylic Acid

Introduction

Helicobacter pylorus (H. pylori) is a bacterium that causes chronic inflammation in the stomach and is a common cause of ulcers worldwide. It is also responsible for chronic gastritis, peptic ulcer disease and gastric malignancy in majority of healthy populations [1-3]. The treatment of *H. pylori* remains a challenging proposition; although it is highly sensitive to most antibiotics, difficult to eradicate from human body even with the current best therapies. Conventional formulations such as immediate release tablets are used for eradication therapy, but they do not remain in the stomach for longer time. Therefore, it is difficult to reach minimum inhibitory concentrations in the gastric mucus where *H. pylori* colonizes. The bioavailability and therapeutic efficiency of drugs was improved by many novel methods such as transdermal [4], iontophoretic, intranasal [5-7] and complexation technique [8] for poorly water soluble drugs. Bioadhesive floating drug delivery systems are also useful in this context. In order to extend the gastric residence period, gastro retentive drug delivery systems have been developed, which is an approach to prolong gastric residence time and also to provide site-specific drug release in the upper gastrointestinal tract (GIT) for local or systemic effects [9]. These include high density (sinking) systems, low density (floating) systems, mucoadhesive systems, swelling and expanding systems, modified shape systems, [10], floating-bioadhesive drug delivery systems [11] and other delayed gastric emptying devices which would improve the therapeutic effects of many drugs.

In case of floating drug delivery system (FDDS), drug remains buoyant in the stomach for a longer period of time without reducing the gastric emptying rate. This result in retardation of drug release at the desired rate from the system, an increased gastric retention time (GRT) and helps in better control of fluctuations in plasma drug levels [12]. But, the main drawback of FDDS is that it is effective only when the fluid level in the stomach is sufficiently high, which aids in the buoyancy of the dosage form [13]. This limitation can be overcome by using bioadhesive polymers which enable it to adhere to the mucous lining of the stomach wall [14]. Floating- bioadhesive drug delivery systems offer the advantages of increased contact time with stomach mucosa resulting in more effective absorption, improving the bioavailability of drugs with absorption window high in the stomach and proximal intestine and reduced dosing frequencies [13].

Previously, lectin conjugated multi particulate floating systems of clarithromycin for eradication of *H. pylori* was reported based on the mucoadhesive property of gastric mucosa [15]. Levofloxacin (single and mini floating tablets) was found to be a potential candidate for targeted drug delivery and are anticipated to be useful in the treatment of *H. pylori* [16].

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Amoxycillin trihydrate (AT) is a moderate-spectrum, bactericidal, β-lactam antibiotic used to treat bacterial infections caused by susceptible microorganisms. It is better absorbed from the upper part of GIT, following oral administration, with a half-life of 1 h – 2 h. AT is prescribed for the treatment of *H. pylori* induced peptic ulcers alone or in combination with other drugs for the eradiation from the human body [17]. Conventional oral dosage form of AT cannot bring out complete eradication *of H. pylori* [18]. The reason for the incomplete eradication of H. pylori is probably due to the short residence time of antimicrobial agents in the stomach so that effective antimicrobial concentration cannot be achieved in the gastric mucousal layer or epithelial cell surfaces where *H. pylori* exist. Hence, an attempt was made to formulate floating-bioadhesvie tablets of AT.

The present investigation was focused on the development of AT floating-bioadhesive tablets (ATFBT) by direct compression method and evaluation for the physical characters such as drug release, floating properties, swelling index and *ex vivo* bioadhesion study. Physical stability of developed optimized formulation at 40°C ± 2°C / 75% ± 5% RH for 3 months was to be studied.

Materials

Amoxicillin trihydrate (AT) was received as generous gift sample from M/s Aurobindo Pharma Ltd, Hyderabad, India. Hydroxypropyl methyl cellulose (HPMC K_4M), chitosan (CH) was received as gift sample from Dr. Reddy's Labs, Hyderabad, India. Carbopol974P (CP974P) and Polymethacrylic acid were received as gift sample from M/s Aurobindo Pharma. Ltd., Hyderabad, India. Sodium bicarbonate, citric acid, microcrystalline cellulose (Avicel PH102), magnesium stearate (MS) and talc were purchased from S.D. Fine-Chem. Ltd., Mumbai, India. All other reagents used were of analytical grade.

Methods

Preparation of AT floating-bioadhesive tablets

The floating-bioadhesive tablets of AT (ATFBT) were developed by using HPMC K_4M/CH or CP974P/PMA as release retarding agents / bioadhesive polymers. Direct compression method was used for the preparation of tablets. Sodium bicarbonate was used as effervescent base/gas generating agent to generate carbon dioxide [12]. All components of the formulation were sieved individually through sieve (mesh size 40). The components of the formulation were mixed thoroughly for 20 min. Magnesium stearate (sieved through 60-mesh) was added into powder blend as a lubricant and the resultant mass was mixed for an additional 3 min. Finally, the resultant powder mass obtained was compressed into tablets with 13 mm round punches at a hardness of 6 kg/cm^2 on 16 station punching machine (Cadmach, Ahmedabad, India). The composition of tablet formulations is shown in Table 1.

Evaluation of final blend

The flow properties of granules (before compression) were characterized in terms of angle of repose, tapped density, bulk density, Carr's index and Hausner ratio [19].

Evaluation of physicochemical properties

The formulated tablets were evaluated for weight variation, thickness, crushing strength, friability, weight variation, buoyancy, dissolution studies and stability studies. Further, *ex vivo* bioadhesion studies were conducted using porcine gastric mucosa.

The prepared floating-bioadhesive tablets were evaluated for weight variation using 20 tablets (IP, 1996), hardness (Monsanto tester) using 6 tablets, thickness (Vernier calipers) using 6 tablets, friability (Roche friabilator) using 10 tablets, drug content using 10 tablets, buoyancy using 6 tablets and dissolution studies using 6 tablets. The results were expressed as mean ± S.D (Table 2).

Drug content uniformity

Prepared tablets were accurately weighed and finely powdered by pestle in a mortar. A weighed portion of each powder equivalent to dose (500 mg) of prepared tablet, was transferred in to a volumetric flask and the drug was extracted with methanol as the solvent. The contents of the flask were sonicated for 10 min, filtered and diluted suitably with 0.1 N HCl as the solvent. The samples were analyzed spectrophotometrically at 272 nm [20].

Determination of *in vitro* buoyancy

The buoyancy of RSFBT was determined in six replicates using United States Pharmacopoeia (USP) dissolution apparatus II (Electrolab, TDT-06T, Mumbai, India) in 900 ml of 0.1 N HCl, maintained at 37°C ± 0.5°C with paddle rotation of 50 rpm [21]. The FLT as well as TFT were determined visually. The time taken by the tablet to emerge onto surface of dissolution medium and the total time, the tablet remained buoyant on fluid surface were noted as FLT and TFT, respectively for all the formulations.

Dissolution studies

The dissolution of prepared ATFBT was studied in six replicates using USP dissolution apparatus II (Electrolab, TDT-06T, Mumbai, India). The dissolution medium was 900 mL of 0.1 N HCl (pH 1.2); temperature was maintained at 37°C ± 0.5°C with a paddle rotation at 50 rpm. 5 mL aliquots were withdrawn at predetermined time intervals

Ingredient	Formulation (mg)									
	F1	F2	F3	F4	F5	F6	F7	F8	F9	F10
AT	500	500	500	500	500	500	500	500	500	500
HPMC K_4M	50	75	100	125	-	-	-	-	-	-
Chitosan	25	25	25	25	-	-	-	-	-	-
Carbopol 974P	-	-	-	-	25	50	75	100	75	75
Polymethacrylic acid	-	-	-	-	25	25	25	25	50	75
Sodium bicarbonate	85	85	85	85	85	85	85	85	85	85
Avicel pH 102	123	98	73	48	148	123	98	73	48	23
Talc	8.5	8.5	8.5	8.5	8.5	8.5	8.5	8.5	8.5	8.5
Magnesium Stearate	8.5	8.5	8.5	8.5	8.5	8.5	8.5	8.5	8.5	8.5

Total weight of tablet equivalent to 800 mg

Table 1: Composition of AT floating-bioadhesive tablets.

Formulation	Carr's Index (%)	Angle of repose	Hanseur' s ratio
F1	21.4	25.9°	1.12
F2	23.5	24.3°	1.13
F3	21.6	25.8°	1.16
F4	22.3	26.6°	1.13
F5	21.1	24.6°	1.16
F6	21.8	24.6°	1.17
F7	22.3	26.9°	1.15
F8	23.1	27.3°	1.13
F9	21.6	27.8°	1.16
F10	24.5	25.4°	1.14

Table 2: Pre-compression properties of ATFBT.

and replaced with 5ml of fresh dissolution medium each time [22]. Samples were filtered using membrane filter (0.45 µm) and suitably diluted with dissolution medium wherever necessary and absorbance of the samples was measured at λ_{max} 272 nm by using double beam UV-Visible spectrophotometer (ELICO, SL 210, India).

Drug release kinetics

The drug release profiles were subjected to different kinetic models to explain the release kinetics for ATFBT. In this study, the drug release profiles were subjected to zero-order, first-order [23], Higuchi [24] and Korsmeyer-Peppas kinetic models [25,26]. The goodness of fit was evaluated using the correlation coefficient values (R^2).

Zero-order: $F = K_0 t$; where F is the fraction of drug released at time t, and K_0 is the zero-order release constant.

First-order: $\ln (1 - F) = -K_1 t$; where F represents the fraction of drug released at time t, and K_1 is the first-order release constant.

Higuchi model: $F = K_H t^{1/2}$; where F represents the fraction of drug released at time t, and K_H is the Higuchi constant.

Korsmeyer–Peppas model: $F = K_p t^n$; where F represents the fraction of drug released at time t, K_p is the rate constant and n is the release exponent, indicative of the drug release mechanism. A value of $n \leq 0.5$, indicates the Fickian release mechanism. The value of n between 0.5 and 1 is an indication of non-Fickian release mechanism (both diffusion controlled and swelling controlled). When, $n \geq 1$, it is case-II transport and this involves polymer dissolution and polymeric chain enlargement or relaxation.

Swelling studies

The swelling behavior of the tablets was determined in triplicate. Initial weight (W_0) of the tablets was noted individually and placed separately in a glass beaker containing 200 ml of 0.1 N HCl, maintained at 37°C ± 0.5°C. At regular time intervals, the tablets were removed and the excess surface liquid was carefully removed by means of a filter paper. The swollen matrix was then reweighed (W_1). The percentage swelling was calculated by using following equation.

$$\text{Percentage swelling} = \frac{(W1 - W0)}{W0} \times 100$$

Measurement of bioadhesive strength of the tablets

Bioadhesive strength of the prepared tablets was measured on a modified physical balance [27]. The porcine gastric mucosa was obtained from slaughter house and was stored in phosphate buffer solution prior to the bioadhesion study. The mucosal membrane was excised by removing the underlying connective tissue and isotonic phosphate buffer (IPB) pH 6.6 was used as the moistening fluid. It was washed thoroughly with IPB pH 6.6 and then tied over to the protrusion in the teflon block using a thread. The block was lowered into the glass container filled with IPB pH 6.6 at 37°C ± 2°C such that the buffer just touched the sides of the stomach membrane. The two sides of the balance were made equal, before the study, by keeping 5.0 g weight on the right pan. The glass container was kept below the left hand side of the balance. The tablet was stuck onto the lower side of the hanging Teflon cylinder using either a little moisture or a double sided tape. The surface of the stomach membrane was blotted with a whattman filter paper and 25 µL of IPB pH 6.6 was added to the stomach surface. This was done in order to obtain reproducible results. The 5.0 g weight from the right pan was removed. This lowered the Teflon cylinder along the patch over the stomach membrane with a weight of 5.0 g. This was kept undisturbed for 2.0 min. Then the weights on the right hand side were slowly added with an increment of 0.5 g till the tablet just separated from the stomach membrane surface. The excess weight on the right pan, that is, total weight minus 5.0 g was taken as a measure of the bioadhesive strength. The equipment was located in an air-conditioned room at 22°C and 60% relative humidity.

Physical stability studies

Physical stability studies were conducted according to International Conference on Harmonization (ICH) guidelines. The optimized formulation was enclosed in polyethylene bottle and placed in a desiccator containing saturated sodium chloride solution, which provided 75% ± 5% RH. The desiccator was stored at 40°C ± 2°C for 3 months [28]. At predetermined time intervals, the tablets were examined for hardness, drug content, buoyancy and drug release. Finally, the tablets were tested for any statistical difference using the Students unpaired t-test at $p < 0.05$.

Drug-excipient compatibility study

Differential scanning calorimetry (DSC): DSC analysis of pure drug, pure polymers, physical mixtures (1:1 ratio of drug: polymer) and optimized formulation were performed using Universal DSC instrument ((model DSC-60, Shimadzu, USA). Approximately 8 mg - 10 mg of sample was taken in aluminum crucible and heated within a heating range of 40°C - 300°C, at a rate of 10°C/min using dry nitrogen as the effluent gas, empty aluminum crucible was used as reference.

Results and Discussion

The floating bioadhesive tablets of AT were prepared by effervescent technique. Initially, concentration of gas-generating agent was optimized. Tablets with 9, 10 and 12.5% of sodium bicarbonate were prepared and evaluated for floating lag time (FLT) and total floating time (TFT). Tablets with 9% showed 108 sec ± 6.4 sec of FLT with > 12 h of TFT, whereas with 10% showed 66 sec ± 2.4 sec of FLT with 12 h TFT. But, in case of 12.5%, FLT was less than 40.3 sec ± 2.6 sec but TFT was less than 12 h. Hence, sodium bicarbonate at 10% concentration was selected for preparation of floating-bioadhesive tablets of AT.

Screening for polymer concentration

Initially, formulation of ATFBT was developed with HPMC K_4M (50, 100 and 125 mg) and carbopol (50, 75 and 100 mg) individually and subjected to buoyancy studies. All the formulations floated within 80 sec - 90 sec / 70 sec - 80 sec in HPMC K_4M / carbopol formulations respectively with TFT of 8 h – 10 h. Combination of HPMC K_4M with 25 mg of Chitosan resulted in floating lag time of 60 sec - 70 sec and combination of carbopol with 25 mg of PMA resulted in 40 sec - 60 sec

of FLT with TFT of more than 12 h. As the combinations showed better buoyancy properties, they were formulated and further studied.

Flow properties of pre-compressed powder

The powder blends prepared for compression of tablets were evaluated for their flow properties (Table 2). Angle of repose was in the range of 23.6° to 28.2°. Carr's index was found to be in the range of 12.55 to 15.49 and Hausner's ratio ranged from 1.143 to 1.181. These values indicate that the prepared powder blends exhibited good flow properties.

Characterization of floating tablets

The floating-bioadhesive tablets of AT were prepared by direct compression method using combination of HPMC K_4M / Chitosan (F1-F4), carbopol 947 / PMA (F5-F10) and lactose as a channelling agent. The physico-chemical characteristics are shown in Table 3. The weight variation was within the range of Indian pharmacopoeial specifications (IP, 1996). The hardness of different formulations was found to be between 6.19 ± 0.41 and 6.33 kg/cm^2 ± 0.43 kg/cm^2, indicating satisfactory mechanical strength. The thickness for different formulations was found to be in between 7.5 mm to 7.6 mm. The friability was less than 1% for all the formulations, which is an indication of good mechanical resistance of the tablets. The drug content of all the batches varied in between 98.88% ± 2.92% and 99.94% ± 1.84% indicating content uniformity in the prepared batches.

In vitro buoyancy studies

All the batches of tablets were prepared by effervescent technique. Sodium bicarbonate was used as a gas generating agent. Formulations F1-F4, prepared with HPMC K_4M / Chitosan floated with a lag time of 58 ± 1.8 sec to 66.4 ± 3.2 sec. Formulations F5-F10, prepared with carbopol 937 / PMA showed a floating lag of 32 ± 2.7 sec to 59.8 ± 1.4 sec. Tablets of all formulations showed Table 4 good buoyancy with maximum floating lag time of 66.4 sec ± 3.2 sec. This is mainly due to evolution of carbon dioxide entrapped inside the hydrated polymeric matrices, resulting from the interaction between gas generating agent and dissolution medium (0.1 N HCl), and this leads to lowering of density of matrices below 1 gm/ml enabling the matrices to float. At HPMC content of 20% or more, the particles of HPMC are close enough to permit a faster establishment of the gel layer, in a manner that the effect of different viscosities of the polymers and type of filler (soluble, insoluble or swellable) is minimized [29]. All formulations remained buoyant due to the presence of gas generating agent, for more than 12 h in dissolution medium (0.1 N HCl, pH 1.2) [30].

Dissolution studies

The dissolution studies revealed that formulations F1and F2 (containing 50 mg and 75 mg of HPMC K_4M) showed a release of 99.45% and 98.99%, respectively, in 6 h and 8 h (Figure 1). Formulation F3 (100 mg of HPMC K_4M) showed maximum drug release of 97.74% in 12 h. The variation in drug release was due to different polymer concentrations in all the four formulations. Formulation F1and F2 was unable to sustain the drug release for desired period of time but in case of formulation F3, 97.7% of drug was released in 12 h. All these four formulations floated for 12 h. Formulation F4 failed to show required drug release profile. Formulation F3 showed the desired drug release profile and floated with a lag time of 66.4 sec, for these reasons, it was considered as best formulation among all the four formulations.

The difference in drug release might be due to the amount of gel layer formed around the tablets. At higher concentrations of polymer, it resulted in a greater amount of gel being formed. This gel increased diffusion length so that drug release was decreased. As the concentration of polymers was increased, the tablets could retain their physical integrity and the drug release was significantly extended.

Formulations F5-F8, composed of carbopol 974P (25 mg - 100 mg) and PMA, showed a release of 98.75, 98.85, 99.83 and 90.8% respectively. These variations in drug release were due to changes in polymer concentrations of the tablets. The main reason for retarded drug release is due to the presence of Carbopol 974P, which readily absorbs water and swells. In addition to its hydrophilic nature, cross-

Formulation code	Weight variation (mg)[a]	Hardness (kg/cm^2)[b]	Thickness (mm)[b]	Friability (%)[d]	Drug Content (%)[c]
F1	800.2 ± 2.4	6.33 ± 0.43	7.76 ± 0.16	0.31	99.56 ± 2.33
F2	799.4 ± 3.1	6.15 ± 0.38	7.86 ± 0.23	0.27	99.74 ± 1.83
F3	798.2 ± 2.2	6.28 ± 0.65	7.76 ± 0.24	0.43	98.91 ± 1.34
F4	801.8 ± 2.8	6.21 ± 0.62	7.63 ± 0.16	0.37	98.85 ± 2.43
F5	802.2 ± 2.4	6.24 ± 0.32	7.68 ± 0.25	0.33	98.88 ± 2.928
F6	799.5 ± 2.7	6.19 ± 0.41	7.55 ± 0.15	0.26	99.69 ± 0.98
F7	799.2 ± 1.7	6.26 ± 0.52	7.65 ± 0.06	0.53	99.94 ± 1.84
F8	800.8 ± 1.6	6.31 ± 0.62	7.62 ± 0.09	0.18	99.24 ± 1.45
F9	801.9 ± 1.9	6.29 ± 0.63	7.56 ± 0.06	0.53	99.41 ± 2.36
F10	802.2 ± 2.4	6.23 ± 0.41	7.48 ± 0.04	0.19	99.77 ± 2.72

Mean ± SD: a-*n* = 20, b-*n* =6, c-*n* = 3, d-n=10

Table 3: Physical evaluation parameters.

Formulation	Floating lag time (sec)	Floating time (h)
F1	58 ± 1.8	> 12
F2	59.7 ± 2.3	> 12
F3	66.4 ± 3.2	> 12
F4	60.8 ± 1.9	> 12
F5	51.7 ± 2.6	> 12
F6	58.6 ± 2.5	> 12
F7	32 ± 2.7	> 12
F8	41.9 ± 0.9	> 12
F9	58.9 ± 2.4	> 12
F10	59.8 ± 1.4	>12

Table 4: Floating properties of prepared tablets.

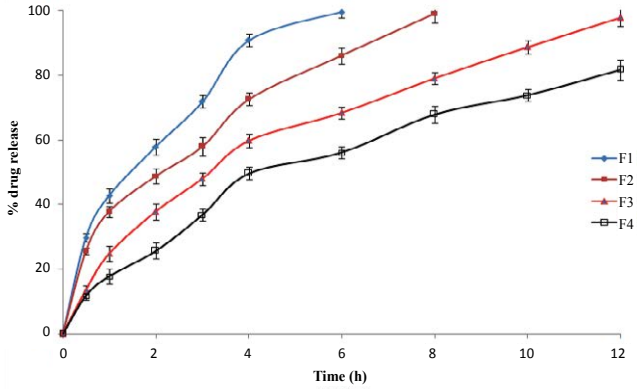

Figure 1: *In vitro* drug release of amoxicillin trihydrate from FBT (F1-F4) formulations.

linked structure and insolubility in water makes the Carbopol 974P as a potential candidate for use in controlled release drug delivery systems.

Formulations F9 and F10 prepared with increased concentrations of PMA (50 mg and 75 mg) resulted in the floating lag time of 58.9 ± 2.4 sec and 59.8 ± 1.4 sec, and the drug release was 97.87% and 89.95% in 12 h. The results are shown in Figure 2.

In all the formulations, formulation F7 (containing 75 mg of carbopol and 25 mg of PMA) met the desired drug release profile in 12 h and floated with a lag time of 32 ± 2.7 sec. It was, therefore, considered the best formulation among all the four formulations of this series.

Release kinetics

The data obtained from dissolution studies were fitted into different kinetic models viz., zero order, first order, Higuchi and Peppas model. The Higuchi plots were found to be linear as indicated by their high regression values (R^2 = 0.988 to 0.997). To confirm the exact mechanism of drug release from these tablets, the data were fitted to Peppas model. Regression analysis was performed and R^2 values were found in between 0.980 to 0.995 for different formulations. The values of the release exponent were in the range of 0.47 to 0.63; this suggested that the release of AT from FBT tablets followed non-Fickian transport mechanism. This means that water diffusion and also the polymer rearrangement played important role in the drug release. As can be seen in Table 5, release rate constants (k) of the developed tablets were significantly different.

Swelling studies

The ability of hydration of the formulation is important because it effects on: (a) tablet buoyancy, and adhesion ability of swellable polymers and drug release kinetics. The percentage swelling obtained from the water uptake studies of the formulations is shown in Figures 3A and 3B. Complete swelling was achieved at the end of 8 h, then diffusion and erosion takes place. The formulation F4 and F8 shows higher swelling index compared to that of the other formulations. The swelling index of the tablets increased with an increase in the polymer concentration.

Ex-vivo bioadhesion study

Apart from buoyancy of the tablet, bioadhesive property could be an important property for gastro retentive drug delivery systems. The developed formulations contained CP 974P, which has bioadhesive property. HPMC polymers are also reported to have the bioadhesive property. The bioadhesive strength of the F7 formulation was found to be 1.86 ± 0.14 N.

Physical stability studies

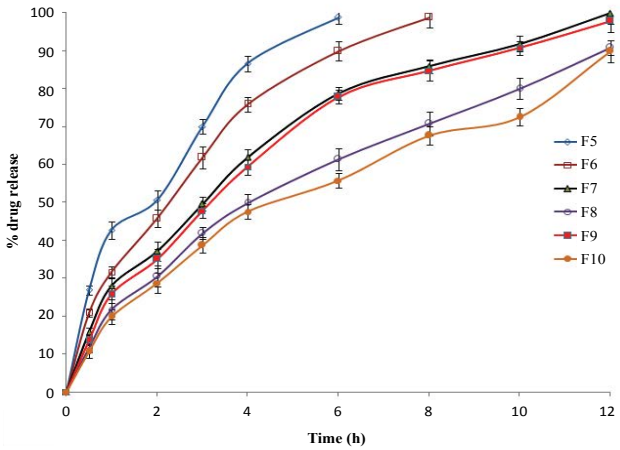

Figure 2: *In vitro* drug release of amoxicillin trihydrate from FBT (F5-F10) formulations.

Formulation	R^2			Peppas	
	Zero order	First Order	Higuchi	R^2	n
F1	0.892	0.909	0.992	0.992	0.498
F2	0.909	0.876	0.997	0.995	0.479
F3	0.922	0.916	0.995	0.987	0.596
F4	0.937	0.994	0.989	0.991	0.623
F5	0.911	0.916	0.989	0.980	0.521
F6	0.915	0.930	0.992	0.994	0.579
F7	0.907	0.800	0.990	0.988	0.576
F8	0.946	0.970	0.995	0.993	0.62
F9	0.912	0.957	0.988	0.985	0.614
F10	0.952	0.935	0.988	0.992	0.632

Table 5: Mathematical models and release kinetics of AT from FBT based on regression coefficient.

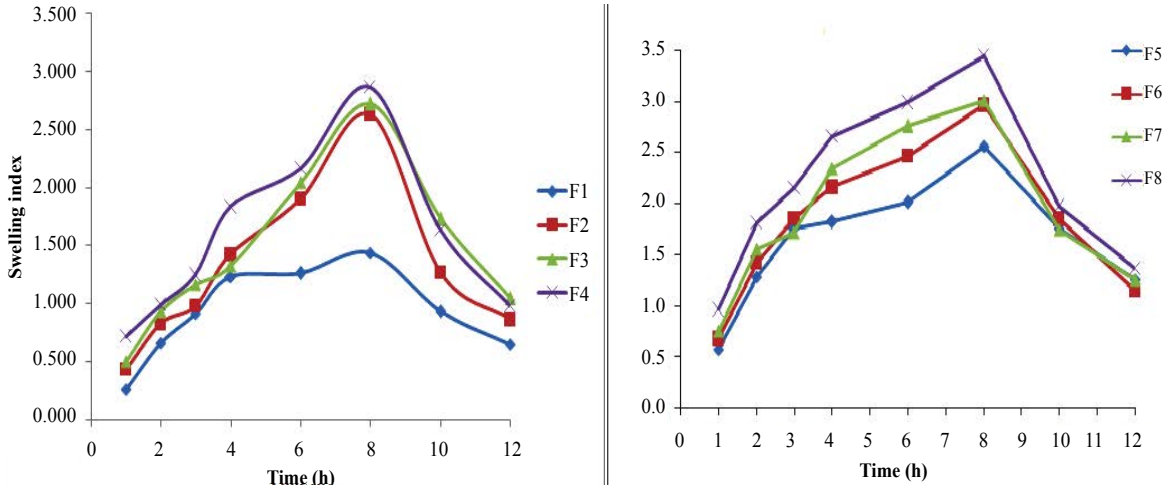

Figure 3: Swelling studies of amoxicillin FBT prepared with (A) HPMCK$_4$M and (B) Carbopol.

Characteristic	0 day*	15th day*	30th day*	60th day*	90th day*
Hardness (kg/cm^2)	6.31 ± 0.47	6.42 ± 0.71	6.42 ± 0.47	6.06 ± 0.70	6.01 ± 0.64
Drug content (%)	99.89 ± 1.82	99.93 ± 2.51	99.88 ± 2.74	99.68 ± 2.17	99.48 ± 12.02
Floating lag time (s)	32.11 ± 3.21	31.73 ± 3.81	30.89 ± 2.84	33.77 ± 3.62	33.89 ± 3.71
Duration of floating (h)	> 12	> 12	> 12	> 12	> 12
Drug released at 12 h (%)	99.89 ± 2.43	99.84 ± 3.72	99.73 ± 2.84	98.97 ± 3.47	99.10 ± 3.84

*The difference was not statistically significant ($p > 0.05$)

Table 6: Stability studies of ATFBT.

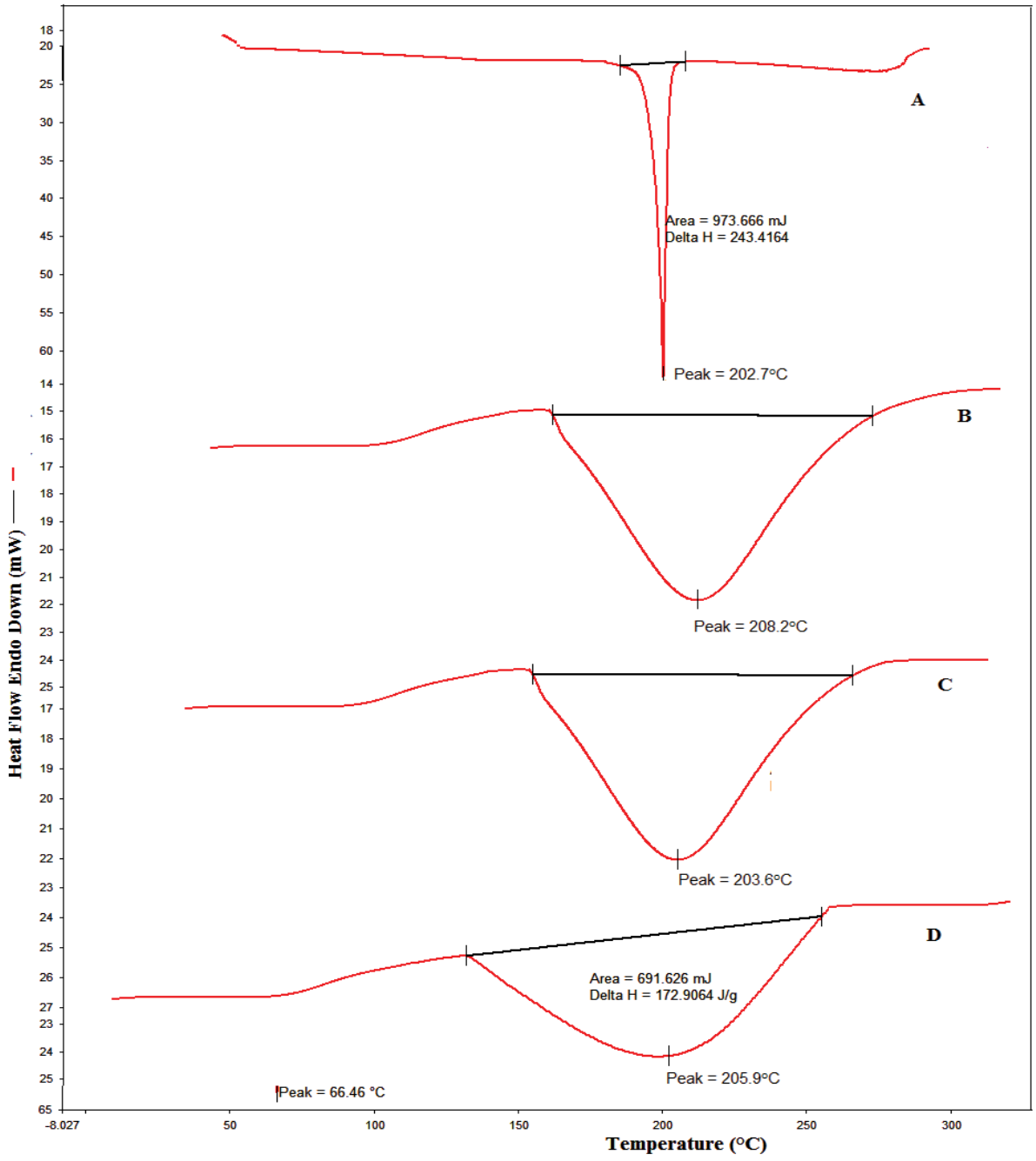

Figure 4: DSC thermograms of (A) pure drug, (B) physical mixture (1:1 ratio) of HPMC K_4M and drug, (C) physical mixture (1:1 ratio) of Carbopol and drug and (D) optimized formulation (F7).

The optimized ATFBT was selected for stability studies. Before and after conducting the stability studies for 3 months, the results were analyzed by using Student's unpaired t-test. No significant difference ($p > 0.05$) was observed in the tablet hardness, drug content, buoyancy or dissolution (Table 6). Therefore, the ATFBT were found to be stable for at least 3 months under these storage conditions.

Drug-excipient compatibility studies by differential scanning calorimetry

The thermal properties of the drug and the mixture of drug and excipients are of important interest since this can help to assess the interaction among different components of the formulations. The DSC thermogram of pure drug (Figure 4) showed an endothermic peak (broader) at 202.7°C in a melting range of 208 to 215°C. The 1:1 mixture of drug and HPMC K_4M is having endothermic peak (broader) of drug at 208.2°C. Similarly, 1:1 mixture of drug and carbopol showed an endothermic peak (broader) of drug at 203.6°C, which was well preserved with slight changes in terms of broadening or a shifting of peak towards the lower temperature. This minor change in the melting endotherm of drug could be due to the mixing of drug and polymer, which lowered the purity of each component in the mixture and may not necessarily indicate potential incompatibility. The DSC thermogram of optimized formulation F7 showed an endothermic peak of drug at 205.9°C which was well preserved with slight change in terms of broadening of peak towards the higher temperature. From the results it was concluded that the drug had compatibility with polymers and other excipients used in the formulation.

Conclusion

Floating-bioadhesive tablets of AT were prepared using combination of HPMC K_4M / Chitosan, and carbopol 947P / Polymethacrylic acid, a promising approach for treatment of *H. pylori*. Gas generating agent (sodium bicarbonate) is essential to achieve optimum buoyancy. The optimized formulation (F7) floated with a lag time of 32 ± 2.7 sec and floated for 12 h. The non-Fickian diffusion was the release mechanism from these tablets. The formulation was stable up to 90 days and exhibited sufficient bioadhesive strength which was proven from *ex vivo* studies.

Declaration of Interest

Authors declare that there is no conflict of interest in this study.

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