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Research Article

Sustained Release of Amoxicillin from Ethyl Cellulose-Coated Amoxicillin/Chitosan–Cyclodextrin-Based Tablets

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Abstract. Sustained release mucoadhesive amoxicillin tablets with tolerance to acid degradation in the stomach were studied. The sustained-release tablets of amoxicillin were prepared from amoxicillin coated with ethyl cellulose (EC) and then formulated into tablets using chitosan (CS) or a mixture of CS and beta-cyclodextrin (CD) as the retard polymer. The effects of various (*w*/*w*) ratios of EC/amoxicillin, the particle sized of EC coated amoxicillin and the different (*w*/*w*) ratios of CS/CD for the retard polymer, on the amoxicillin release profile were investigated. The physicochemical properties of the EC coated amoxicillin particles and tablets were determined by scanning electron microscopy, Fourier-transform infrared spectroscopy, X-ray diffraction, and differential scanning calorimetry. The result showed that the release profiles of amoxicillin were greatly improved upon coating with EC, while the inclusion of CD to the CS retardant additionally prolonged the release of the drug slightly. Overall, a sustained release of amoxicillin was achieved using amoxicillin coated with EC at a (*w*/*w*) ratio of 1:1 and a particle size of 75–100 μm. Therefore, the tablet formulation of amoxicillin may be an advantageous alternative as an orally administered sustained-release formulation for the treatment of peptic ulcers.

KEY WORDS: amoxicillin; beta-cyclodextrin; chitosan; controlled release tablet; ethyl cellulose.

INTRODUCTION

Helicobacter pylori, a common human-specific pathogen, is the causative agent in chronic gastritis (1), gastric and duodenal ulcers, and gastric adenocarcinoma, a common form of cancer in humans (2). *H. pylori* are often observed to adhere to the epithelial cell surfaces of the human stomach and gastric metaplasi in the duodenum where they damage the stomach and duodenal tissue, causing inflammation and peptic ulcers (1,3). Therefore, access of antimicrobial drugs to the site is restricted from both the lumen of the stomach and the gastric blood supply.

Amoxicillin (α-amino-hydroxybenzylpenicillin) is a semi-synthetic, orally absorbed, broad-spectrum antibiotic that is especially effective against *H. pylori* infections (4), where it is widely used in the form of orally administered capsules. These conventional preparations have only a short active residency time in stomach and, furthermore, they may be degraded in gastric acid (pH 1.2) be1cause the β-lactam ring is more susceptible to hydrolytic degradation when the pH is significantly lower than the isoelectric point (pH 4.8; 5). Thus, traditional amoxicillin capsules may be unable to deliver the antibiotics to the site of infection in effective concentrations and in fully active forms (2). Consequently, to overcome these problems, amoxicillin is typically administered at high doses and frequencies, which are considered to bring about systemic toxicities and adverse effect (6), as well as raising the treatment cost. Therefore, delivery vehicles or other means of achieving an improved efficacy and extended residence period of amoxicillin in the gastric epithelial cell surfaces (metaplasi) are highly desirable traits. For example, a sustained-release dosage form that maintains a therapeutic concentration in the blood for a longer period of time is desired and would increase the efficiency of the drug (4).

Attempts have been made to develop a sustained-release dosage form for amoxicillin and to localize the antibiotic delivery in the acidic environment of the stomach. For example, the release of amoxicillin from a gastric retentive system based on alginates has been evaluated (7). In this present investigation, we used biodegradable polymers in controlling the release of amoxicillin as a model drug. Increasing interest and research effort has recently centered around the use of such biodegradable polymers in the formulation of pharmaceuticals, including the three polymers of this study, chitosan (CS), ethyl cellulose (EC), and beta-cyclodextrins (CD), providing the additional advantage of access to this wealth of other existent but required information without the need to self-obtain it each time (including FDA-approval tests).

CS is a natural polysaccharide derived from chitin by alkaline deacetylation. It consists mainly of the repeating unit of 2-amino- and 2- acetamido-2-deoxy-β-D-glucopyranose and is soluble in dilute aqueous acidic solutions (pH <6.5) (8). It has gained increasing attention in the pharmaceutical field due to its favorable biological properties, such as non-toxic, biocompatible, biodegradable, and mucoadhesive prop-

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erties, among others (9). As such, it is widely recognized as the preferred choice as a drug delivery carrier.

CDs are a family of cyclic oligosaccharides with a hydrophilic outer surface and a lipophilic central cavity (10). They are widely used as "molecular cages" in the pharmaceutical, agrochemical, food, and cosmetic industries (11). In the pharmaceutical field, they are used as complexing agents to increase the aqueous solubility of poorly soluble drugs and to increase their bioavailability and stability (12). CDs can also be used to reduce gastrointestinal drug irritation and prevent drug–drug and drug–excipient interactions (9). Moreover, the formation of inclusion complexes between amoxicillin and CDs retards the degradation of amoxicillin in strong acidic solutions (5), such as the gastric fluid in the stomach (pH ~1.2).

EC is a non-toxic, inert, hydrophobic polymer that has been widely used to prepare pharmaceutical dosage forms (13). It is used extensively as a coating material for tablets and granules, as a tablet binder, in microcapsules and microspheres and film- or matrix-forming material for sustained-release dosage forms (14).

The purpose of this work was to evaluate new sustained-release tablets of amoxicillin. To achieve that, amoxicillin was coated with EC and then either CS or a mixture of CS and CD was used as a retard polymer. The physicochemical characteristics were evaluated by scanning electron microscopy (SEM), Fourier-transform infrared spectroscopy (FT-IR), X-ray diffraction (XRD), differential scanning calorimetry (DSC), and hardness. Moreover, the *in vitro* amoxicillin release profiles were monitored in a stimulated gastric fluid (SGF; 0.1 N HCl, pH 1.2) at 37°C, and then compared with the release profile of a commercial drug.

MATERIALS AND METHODS

Materials

Amoxicillin trihydrate was purchased from The Union Chemical 1986, Ltd. (Thailand). Commercial drug tablets of 250/125 amoxicillin/clavulanate were purchased from GlaxoSmithKline, Thailand. A commercial drug capsule of amoxicillin was purchased from Siam Pharmaceutical, Ltd. CS, as food grade (CS BFM), with a degree of deacetylation of 85% and a $\overline{M}_w$ 50–300 kDa, was purchased from Bonafides Marketing Co., Ltd. CDs and EC with $\overline{M}_w$ of 30–60 kDa were purchased from The Union Chemical 1986, Ltd. (Thailand). The other reagents, such as lactose (food grade), magnesium stearate, 95% (*v*/*v*) ethyl alcohol and methanol, were purchased from The Union Chemical 1986, Ltd. (Thailand), except acetronitrile (HPLC grade), which was purchased from The Bonafides Marketing, Ltd.

Methods

Preparation of EC-Coated Amoxicillin

To investigate the effect of EC content on the release of amoxicillin, three different (*w*/*w*) ratios of drug/polymer, namely 1:1, 1:2, and 2:1, were prepared. Solid dispersions of amoxicillin in EC solution were prepared by solvent evaporation as follows. Amoxicillin and EC were dry mixed to obtain a homogeneous mixture. Absolute ethyl alcohol was preheated to about 60°C and then gradually added to the amoxicillin–EC mixture to dissolve the blend while continuously heating the mixture on a hot plate and slowly evaporating the solvent. The mixture was poured onto glass plates and dried. The dried material was then ground and screened through sieves to separate it into three different sizes: size 1 (>100 μm), size 2 (75–100 μm), and size 3 (<75 μm). Each of these three size selected particle fractions was used for the separate preparation of tablets and physicochemical analysis.

Preparation of Tablets Using CS or CS–CD Mixtures as Retard Polymers

The evaluated tablets consisted of amoxicillin as the model drug and either CS or CS–CD as a release rate-controlling polymer. Lactose was used as a compression aid, and magnesium stearate was employed as a lubricant. The detailed compositions of each formulation (formulations A to E) are given in Table I.

Briefly, for the tablet preparation, the ingredients of each batch, separated into the respective size class as outlined

Table I. Composition of the Sustained-Release Amoxicillin Matrix Tablet Formulations, and their Respective Hardness

Formulation	Amoxicillin/EC (*w*/*w*) ratio	Size[a]	Amoxicillin (g)	EC (g)	CS (g)	CD (g)	Hardness (N)*
A	1:0	3	7.0	–	8.0	–	38.3±1.1**
B1	1:1	2	3.5	3.5	8.0	–	14.4±3.9***
B2	1:2	2	2.3	4.7	8.0	–	6.7±2.9***
B3	2:1	2	4.7	2.3	8.0	–	14.9±4.0***
B4	1:1	3	3.5	3.5	8.0	–	21.6±1.3***
B5	1:2	3	2.3	4.7	8.0	–	9.3±7.2***
B6	2:1	3	4.7	2.3	8.0	–	14.1±6.6***
C	1:1	2	3.5	3.5	4.0	4.0	27.9±3.1***
D	1:2	2	3.5	3.5	2.7	5.3	34.3±1.5**
E	2:1	2	3.5	3.5	5.3	2.7	23.1±1.5***

EC ethyl cellulose, *CS* chitosan, *CD* beta-Cyclodextrin
[a] Size 1 (>100 μm), size 2 (75–100 μm) and size 3 (<75 μm)
**P*<0.05 for all effects (one-way ANOVA)
***P*<0.05 (pairwise testing between the groups using Duncan's test)
****P*>0.05 (pairwise testing between the groups using Duncan's test)
For all formulations, 2.5 g of lactose and 1.5 g of magnesium stearate was also included

above (size 1, 2, and 3) were thoroughly blended. The resulting powder mixtures were directly compressed into tablets (~130 mg each) using a single-punched tablet machine.

Drug Content Assay

To obtain the amoxicillin standard curve, 10 mg of amoxicillin was dissolved in 100 ml of DI water as stock solution. Serial dilutions with distilled water were made to cover the working linear ranges of 1, 5, 10, 20, 50, and 100 μg/ml using HPLC (see below for HPLC condition). The linear regression analysis of the calibration curve in yielded equation of $y = 13,008x + 2,447 (R^2 = 0.9998)$.

For the amoxicillin content in the granules, 0.5 g of granules was ground to a powder sample. The powder containing a drug equivalent to 10 mg were accurately weighted and dissolved in 100 mL of water and shaken by a mechanical stirrer for 24 h. The solutions were filtered, suitably diluted and then analyzed by HPLC (see below) to determine the drug content.

The amoxicillin content in the tablets were performed as the same method described above except that 10 tablets of each formulation were ground instead of granules.

Characterization

Scanning Electron Microscopy

The surfaces morphology of the granules was observed using a SEM (Phillips XL30CP). The samples for SEM analysis were prepared by mounting the sample on one side of a double-adhesive stub. The stub was then coated by gold under vacuum.

Fourier Transform Infrared Spectroscopy

The functional groups of samples were studied using FT-IR (Perkin Elmer Spectrum RX-1 FT-IR system). Each sample (10 mg) for evaluation was dispersed in dry KBr, ground well in a mortar and pestle, and the drug–KBr disk was prepared. The disk was placed in FT-IR sample holder and purged with nitrogen gas for 5 min and reading taken. FTIR spectra were recorded in the wavelength region of 400–4,000 cm^{-1} at ambient temperature.

X-Ray Diffraction

The crystallinity of samples was evaluated by XRD using a PAN analytical X-Pert Pro ver. 1.6 with Cu as the anode material and a BB004 flat stage was used. One tablet of the sample was finely powdered and placed in a plastic sample holder of 1 in. square. Data were collected at 45 kV and 40 mA. Samples were scanned from 10 to 50°C at a scan rate of 0.02°/s.

Differential Scanning Calorimetry

Approximately 10 mg of each sample was weighed in an aluminum pan, crimped with a lid, and placed in the DSC unit along with an empty pan as a reference. DSC was performed with a Perkin-Elmer DSC 7 instrument under a nitrogen atmosphere. The sample was heated at a rate of 10°C/min from 25 to 350°C.

Hardness

Ten tablets from each formulation of the compressed tablets were tested for the diametrical crushing strength using the hardness tester (Pharmatest, USA). The crushing strengths (hardness values) were determined and reported as the mean±1 standard deviation (SD), derived from ten replications.

Encapsulation Efficiency

The drug content in the amoxicillin-loaded EC capsules, with either CS or a mixture of CS and CD copolymers as amoxicillin release retardants, was quantitatively determined as follows: 10 tablets of each sample were weighted and ground into fine powder. An amount of powder equivalent to 130 mg was weight accurately into a 100-ml calibrated flask, 100 ml of DI water was added and sonicated for 2 h. The solution was filtered and the drug content was determined by HPLC (see below). The encapsulation efficiency (%EE) was calculated according to the following equation (where AMOX is amoxicillin);

$$\text{AMOX encapsulation efficiency} = \frac{\text{weight of the total AMOX} - \text{weight of free AMOX}}{\text{weight of the total AMOX}} \times 100\%.$$

All experiments were performed in triplicate and the data are shown as means ± SD.

In vitro Amoxicillin Release

The *in vitro* amoxicillin release behavior from tablets was investigated by the method reported previously (14). Adding a tablet of amoxicillin into 250 mL of stimulated gastric fluid (SGF; 0.1 N HCl, pH 1.2) in a flask, and then placed in a shaking water bath at 50 strokes per min at 37±1°C. Starting from time=0 h, and at selected intervals, 3 mL of sample was withdrawn and neutralized with 1 mL of 0.3 M NaOH to prevent any further degradation. The samples were then filtered through a 0.45-μm nylon membrane filter and the amoxicillin concentration was determined by HPLC with UV detection at 254 nm. For each formulation, the samples were analyzed in triplicate.

Degradation of Amoxicillin in pH 1.2 HCl Medium

Amoxicillin was observed to degrade rapidly under acidic conditions (14), resulting in decreasing the therapeutic dose of amoxicillin in the stomach; hence, the treatment of *H. pylori* may be failed. In order to determine the actual amount of amoxicillin; therefore, in this study it was important to correct for this degradation, and therefore evaluate the value of the degradation rate constant (k_2). To

this, the degradation behavior was fitted to the exponential decay equation as in:

$$C_t = C_o e^{-k_2 t} \quad (1)$$

where C_t is the amount of amoxicillin remaining in the solution at time *t, Co* is the initial amount of amoxicillin, and k_2 is the degradation rate constant.

From Eq. 1, it can be shown as the first order kinetics by the equation:

$$\ln C_t = \ln C_o - k_2 t \quad (2)$$

The degradation of amoxicillin was carried on the same method as previously reported by Sahasathian *et al.* (14). Briefly, 50 mg of amoxicillin (powder) were dissolved in 250 mL of pH 1.2 HCl in a flask. The flask was then placed in a shaken water bath at a speed of 50 strokes per min with the temperature maintained at 37°C. Starting from time=0 h and at appropriate intervals, 3 mL of samples were collected and neutralized with 1 mL 0.3 M NaOH to prevent further degradation reaction. The samples were then filtered through a 0.45 μm nylon membrane filter and determined by a HPLC method with UV detection at 254 nm (see below).

HPLC Assay of Amoxicillin

The HPLC analysis of amoxicillin was performed on a Spectra SYSTEM with a pump (P 4000), a UV detector (UV 6000 LP) and an automatic injector. Chromatographic separations were performed on a reverse-phase Pinnacle II C_{18} column (5.0 μm particle size, 250×46 mm ID) including a guard column (C_{18}, 5.0 μm particle size, 20×4.0 mm ID). The mobile phase used was a 9:1 (*v*/*v*) ratio of sodium phosphate buffer (0.01 M, pH 6.0)/acetonitrile at a flow rate of 1 ml/min. The detector wavelength was set at 254 nm and the amoxicillin retention time under these conditions was 3.327 min.

Statistical Analysis

Statistical analysis for hardness was performed by Duncan's test for pairwise testing and the statistical evaluation for errors by one-way ANOVA. All analyses were performed using SPSS version 11.5; with $P<0.05$ being accepted as statistically significant.

RESULTS AND DISCUSSION

Morphology of Amoxicillin with Different Levels of EC Coating

Representative scanning electron micrographs of amoxicillin, granules of amoxicillin coated with EC at three different (*w*/*w*) ratios of amoxicillin to EC (1:1, 1:2, and 2:1), and selected for three different sizes (size 1, 2, and 3) are shown in Fig. 1. Pure amoxicillin is seen to be a highly crystalline material with a flat rod shape and this crystalline shape is still maintained when the granules of amoxicillin were coated with EC at the three different (*w*/*w*) ratios of amoxicillin to EC. Thus, the amount of coating EC polymer had no effect on the morphology of amoxicillin.

After separating the EC-coated amoxicillin particles into three size groups by sieving the crystalline, nature of amoxicillin was still observed in all cases supporting that there was no change in the crystal form of the drug in the presence of EC in the granules. Moreover, the larger crystalline sizes of amoxicillin were only found in the larger size of granules, supporting that the range of particle sizes observed is determined by that of the amoxicillin crystals, that is the amoxicillin crystal size largely determines the amoxicillin–EC particle size, and is not due to heterogeneity in the amount of EC coatings.

FT-IR Studies

Effect of the Amount of EC Coated on Amoxicillin

The FT-IR spectrum of the pure amoxicillin, EC and the granules of amoxicillin/EC at the three different (*w*/*w*) ratios are presented in Fig. 2 for 75–100 μm sized particles.

The FT-IR spectrum of amoxicillin alone showed a band at around 3,470 cm^{-1} (O-H, N-H stretching vibration) and characteristic peaks at 1,769 cm^{-1} (C=O stretching of β-lactamic), 1,688 cm^{-1}(C=O stretching of amide) and 1,587 cm^{-1} (asymmetric stretching of carboxylate). Furthermore, the other bands at 1,008 cm^{-1} were attributed to the stretching vibration of C-O bending.

The FT-IR spectrum of EC displayed distinct peaks at 3,486 cm^{-1} (O-H stretching), 2,972 cm^{-1} (C-H stretching), 1,391 cm^{-1} (-CH_3 bending), and 1,120 cm^{-1} (C-O stretching in the cyclic ether).

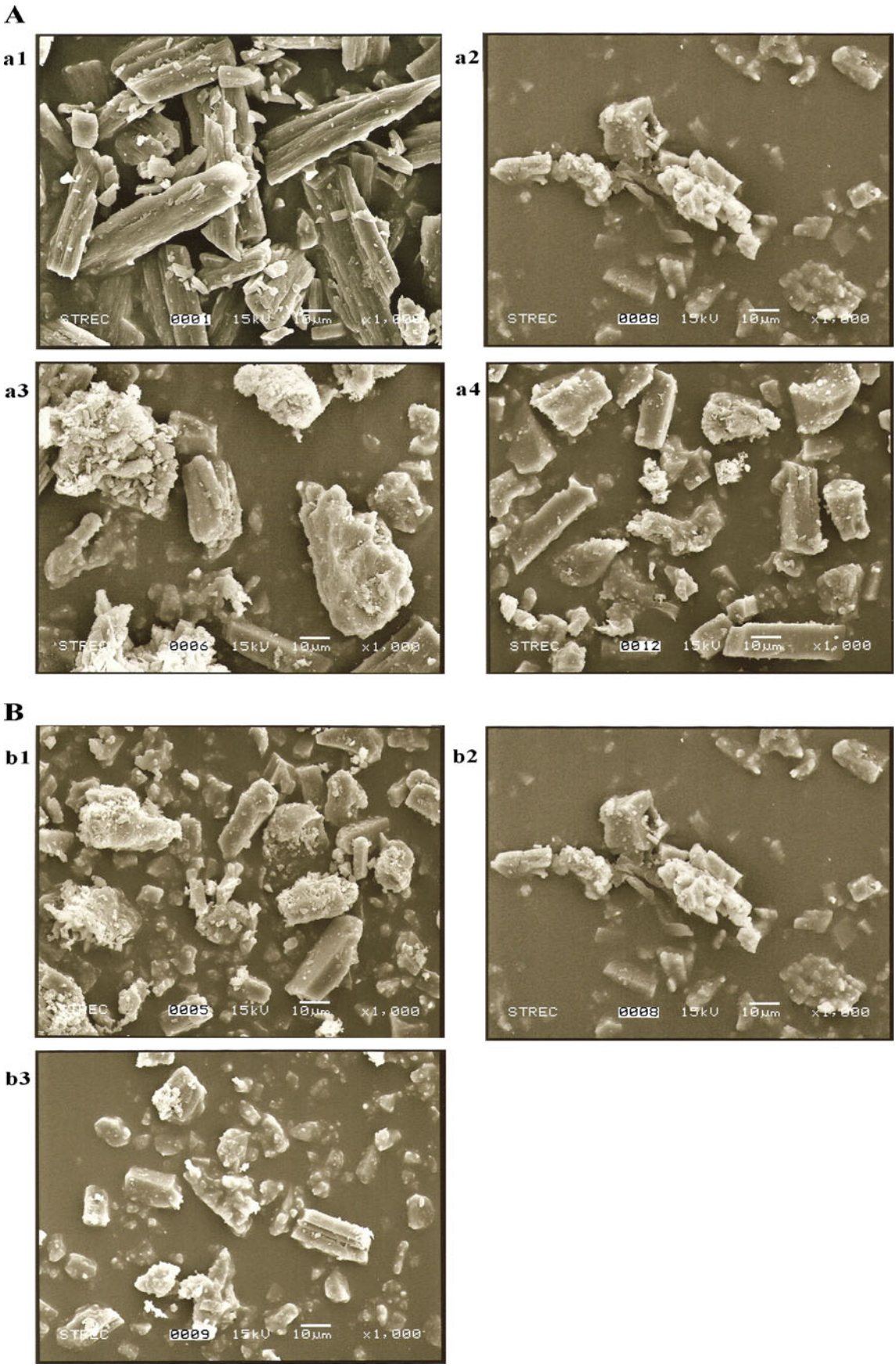
The FT-IR spectra obtained from the various mass ratios of EC-coated amoxicillin all showed the characteristic peaks of the pure amoxicillin and EC which were not shifted from the original peaks. If the drug and the polymer had chemically interacted, then the functional groups in the FT-IR spectra would have been expected to show band shifts and broadening compared to the spectra of the pure drug and polymer (15). Thus, the results suggested that there were no chemical interactions between amoxicillin and EC and an increase in the EC content did not initiate any amoxicillin–EC interactions either. Furthermore, the data suggest that amoxicillin is not decomposed when coated with EC using this solid dispersion technique, although the actual bioactivity evaluation is still required. This result was also consistent with the XRD patterns of the EC-coated amoxicillin.

Effect of Presenting CS or Various CS–CD Mixtures with Amoxicillin as Retard Polymers

The FT-IR spectrum for the amoxicillin, EC, CS, CD, and the granules of amoxicillin/EC at a 1:1 (*w*/*w*) ratio with various CS/CD mixtures are presented in Fig. 3.

The FT-IR spectrum of CD showed the characteristic peaks at 3,400 cm^{-1} and distinct single peaks at 2,926 cm^{-1} (O-H stretching), 1,630 cm^{-1} (N-H stretching), 1,429 cm^{-1} (O-

Fig. 1. a Representative SEM photographs (1,000×) of (a_1) pure amoxicillin, and granules of amoxicillin/EC with a (*w*/*w*) ratio of; (a_2) 1:1, (a_3) 1:2, and (a_4) 2:1. **b** Representative SEM photographs (1,000×) of granules of 1:1 (*w*/*w*) ratio of amoxicillin/EC 1:1 particles separated to size for, (b_1) size 1 (>100 μm), (b_2) size 2 (75–100 μm), and (b_3) size 3 (<75 μm)


A
a1
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a3
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B
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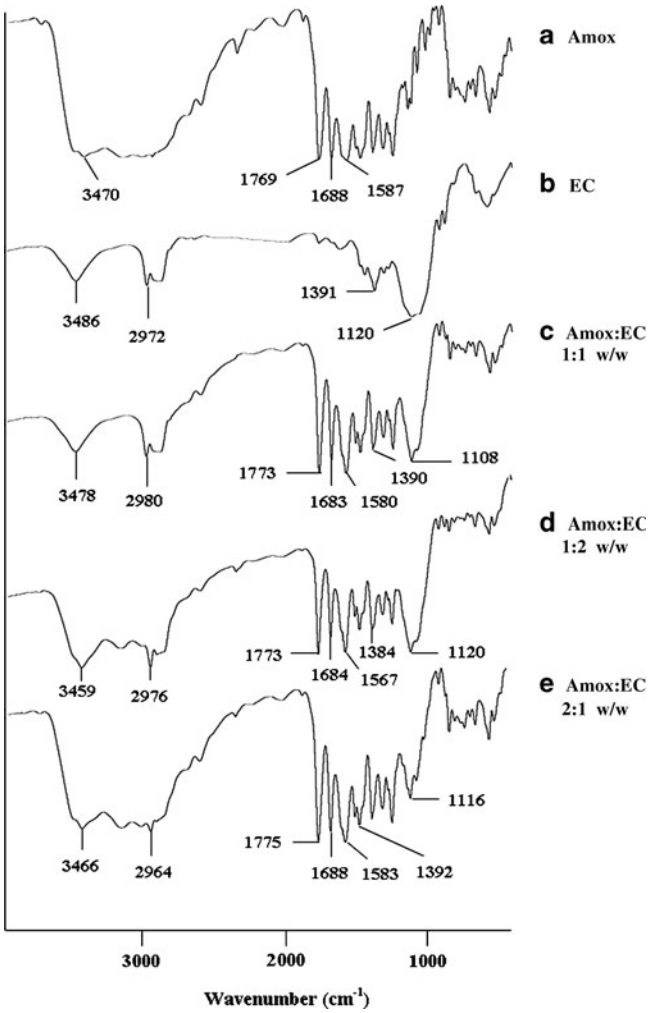

Fig. 2. FT-IR spectra of **a** amoxicillin (AMOX), **b** EC and **c**–**e** granules of amoxicillin/EC at a (*w*/*w*) ratio of **c** 1:1, **d** 1:2, and **e** 2:1

H stretching), and 1,326 cm^{-1} (C-O stretching), while that for CS revealed characteristic peaks at 1,663 cm^{-1} (amide of acetyl groups, O=C-NHR), 1,540 cm^{-1} (–NH_2 bending), 1,411 cm^{-1} (C=C stretching), and 890 cm^{-1} (C-H bending).

The FT-IR spectrum of a 1:1 (*w*/*w*) amoxicillin/EC with CS showed the characteristic peaks of amoxicillin, EC, and CS with no significant shifts from the original positions of each of these three constituents alone, again revealing no evidence for any chemical interactions between amoxicillin, EC, and CS.

Likewise, the FT-IR spectrum of a 1:1 (*w*/*w*) amoxicillin/EC with CS and CD still showed the characteristic peaks of all four constituents with no band shifts from their original positions, again supporting the absence of any chemical interactions and supporting the notion that all the ingredients can be mixed together without causing amoxicillin to degrade. However, again, actual amoxicillin bioactivity data is required for confirmation.

X-Ray Diffraction Studies

The X-ray diffraction patterns of pure amoxicillin and various amoxicillin/EC granules are presented in Fig. 4 for particles with a size of <75 μm as a representative formulation. Pure amoxicillin showed strong and characteristic sharp peaks at 2Θ (°) 12.06°, 15.02°, 16.12°, 17.08°, and 17.94°,

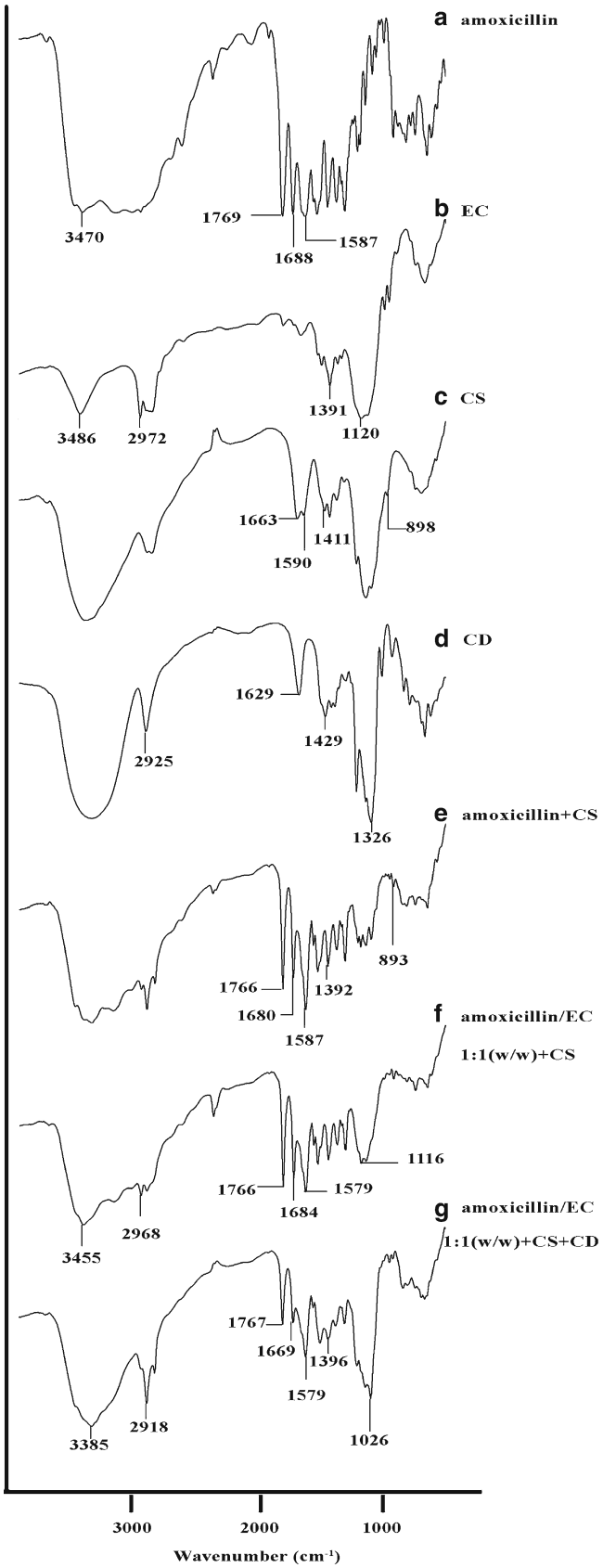

Fig. 3. FT-IR spectra of **a** amoxicillin, **b** EC, **c** CS, **d** CD, and **e**–**g** tablets of 1:1 (*w*/*w*) ratio of amoxicillin **e** alone, **f** plus 1.14:1 (*w*/*w*) of CS/amoxicillin and **g** plus 1.14 (*w*/*w*) CS–CD/amoxicillin with a 1:1 (*w*/*w*) ratio of CS/CD

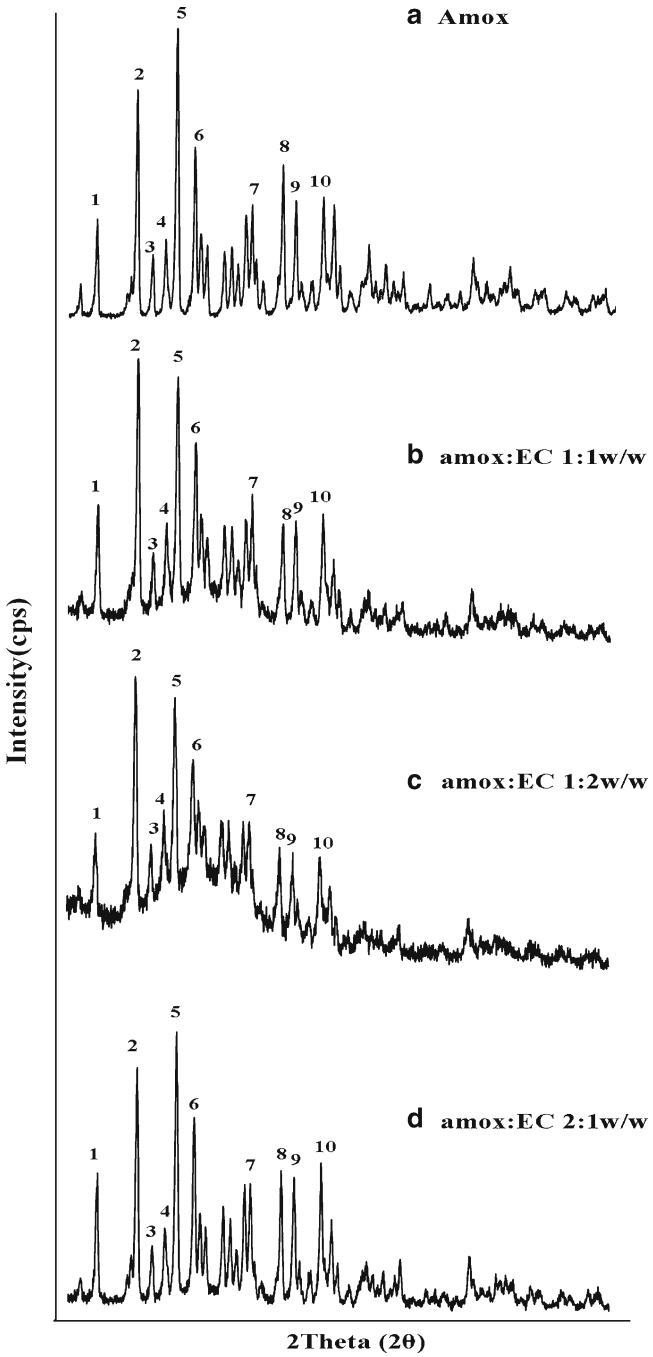

Fig. 4. XRD patterns of **a** amoxicillin and amoxicillin/EC with a (*w*/*w*) ratio of **b** 1:1, **c** 1:2, and **d** 2:1

demonstrating its crystalline nature. The three different weight ratios of amoxicillin/EC also showed the same characteristic peaks at 2Θ (°) as those seen with amoxicillin alone, and are listed in Table II. Thus, there were no changes in the crystalline form of amoxicillin in the presence of EC or EC plus CS–CD, in the physical mixture.

Differential Scanning Calorimetry

The DSC thermograms of amoxicillin, EC and the coated particles derived from the three different (*w*/*w*) ratios of amoxicillin/EC are shown in Fig. 5 for particles of 75–100 μm.

Table II. Intensity (cps) and Diffraction Angle 2θ (°) of Amoxicillin and of Particles of Amoxicillin Coated with Different (*w*/*w*) Ratios of EC

	2θ(°) of amoxicillin/EC (*w*/*w*) ratio			
Position	1:0	1:1	1:2	2:1
1	12.06	12.22	12.14	12.20
2	15.02	15.16	15.08	15.14
3	16.12	16.26	16.24	16.24
4	17.08	17.26	17.20	17.20
5	17.94	18.10	18.02	18.06
6	19.20	19.40	19.36	19.34
7	23.40	23.54	23.50	23.48
8	25.64	25.82	25.72	25.76
9	26.58	26.74	26.70	26.72
10	28.60	28.74	28.72	28.70

Amoxicillin and EC alone showed the melting endothermic peaks at 136°C and 184°C, respectively, while the amoxicillin/EC composites at (*w*/*w*) ratios of 1:1, 1:2, and 1:3, showed two melting endothermic peaks, that of EC in the range of 172–174°C and that of amoxicillin in the range between 108 and 118°C. The endothermic peaks for both amoxicillin and EC decreased because the effect of colligative property of the mixture on the melting point.

Hardness

The sustained-release amoxicillin tablets formed from all the evaluated formulations (Table I) were white and round. The diameter and thickness of tablets were approximately 7.45 and 3.30 mm, respectively.

Comparison of the hardness, summarized in Table I, between preparations showed that formula A (containing only CS, without EC) had the highest hardness values (38.29± 1.11 N) because the composition is more densely packed than in the other formulas. Increasing the proportion of EC in the

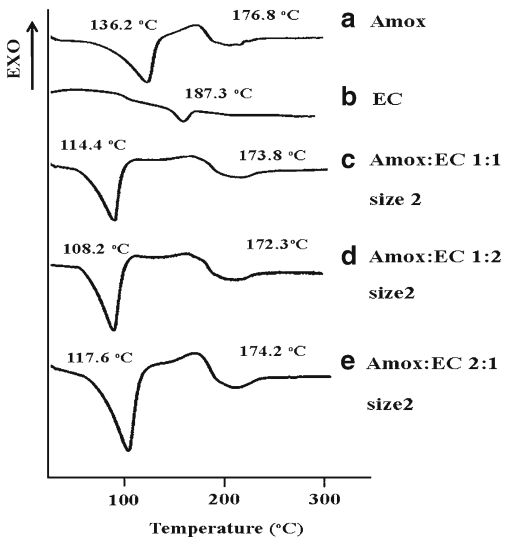

Fig. 5. DSC thermograms of **a** amoxicillin, **b** EC and granules made from 75 to 100 μm size-selected particles of amoxicillin/EC with a (*w*/*w*) ratio of **c** 1:1, **d** 1:2, and **e** 2:1

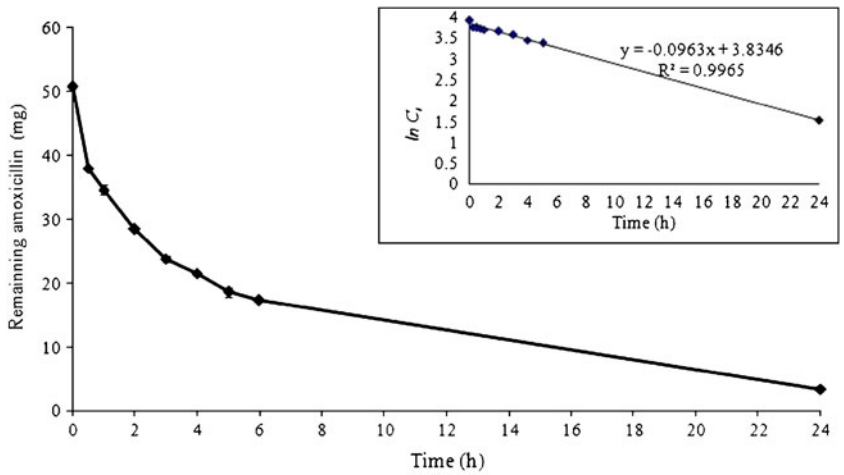

Fig. 6. Degradation of amoxicillin in SGF medium. Data are shown as the mean±1 SD and are derived from three repeats

composition resulted in a decrease in the hardness of the tablet (ANOVA, $P<0.00$). For example, formulation B2, which contained twice as much EC as that of formulation B1, had a nearly twofold decreased hardness (ANOVA, $P<0.03$). Considering that tablets from formulations C, D, to E (containing both CS and CD) displayed a stronger hardness than those from formulations A and B (without CD), it is also likely that CD can improve the binding properties and so hardness (ANOVA, $P<0.05$).

Degradation of Amoxicillin in SGF (0.1 N HCl, pH 1.2)

The typical degradation behavior of amoxicillin in SGF was fitted to the exponential decay equation $C_t=C_o e^{-k_2 t}$, which can be shown as the first-order kinetics by the equation of $\ln C_t = \ln C_o - k_2 t$. The plot in Fig. 6 showed the data fitted a straight line well (R^2=0.9965), supporting that it followed the first-order kinetics, and from this the value of the degradation rate constant (k_2) was evaluated as −0.0963.

In vitro Drug Release

Effect of Amoxicillin Coating with EC

Sahasathian *et al.* (4) reported that amoxicillin-containing CS as a retardant polymer showed a sustained-release profile relative to the tablets of amoxicillin without CS. Therefore, in this work, the tablets of amoxicillin-containing CS were used for comparison.

The release profiles of amoxicillin from the tablets with and without polymer coating were compared with a commercial amoxicillin capsule (Figs. 7 and 8). However, particles of >100 μm could not be compressed into a tablet, likely because

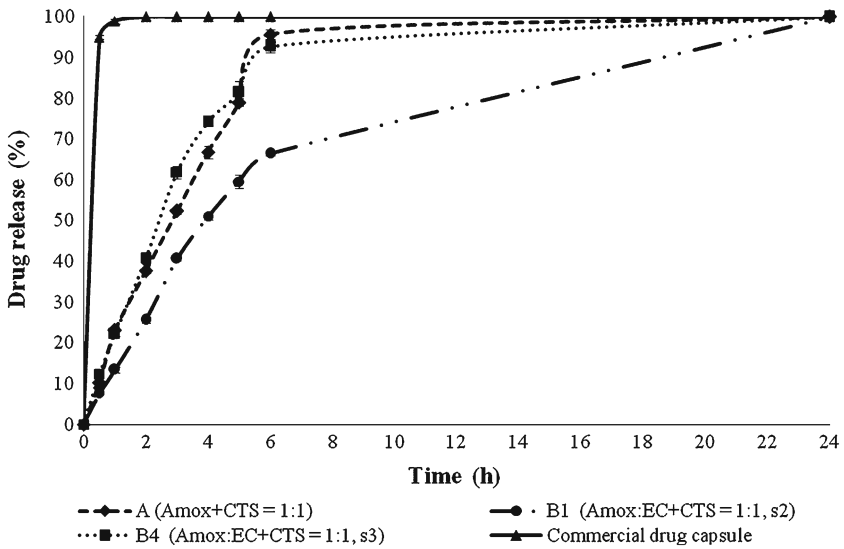

Fig. 7. Release profile of amoxicillin from tablets formulations A, B1 to B4 (see Table I for composition) and a commercial drug capsule, when immersed in SGF medium. Data are corrected for the amoxicillin degradation factor, and are shown as the mean±1 SD derived from three repeats

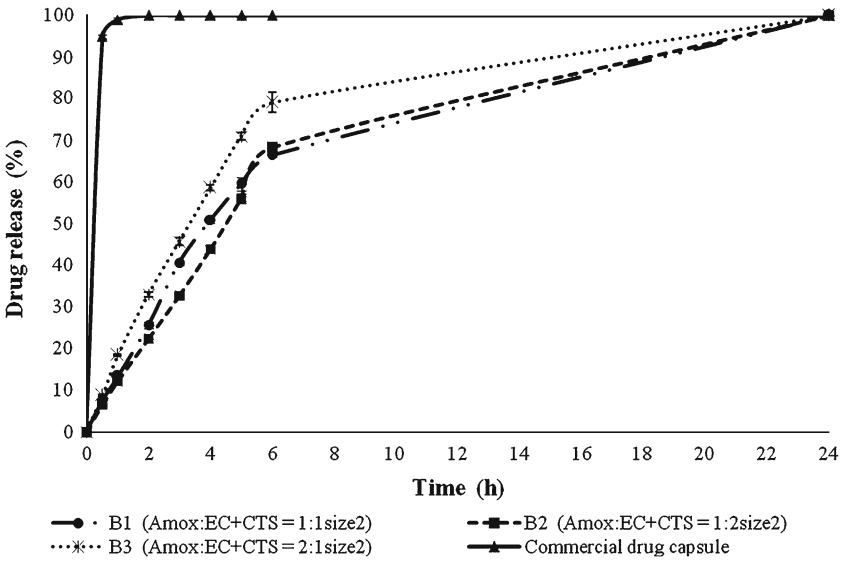

Fig. 8. Release profile of amoxicillin from tablets formulations B1, B2 to B3 (see Table I for composition) and a commercial drug capsule, when immersed in SGF medium. Data are corrected for the amoxicillin degradation factor, and are shown as the mean±1 SD derived from three repeats

the particle size of these granules is very large. This notion is consistent with the disintegration behavior observed in the release studies that showed an immediate amoxicillin release profile (data not shown).

Amoxicillin from the commercial drug capsules was released rapidly, with 100% complete dissolution being attained within 1 h. Tablets from compositions A to B4 (Table I) showed much slower amoxicillin release rates with ~23%, 50%, and 93% being released by 1, 3, and 6 h, respectively, for both compositions and 100% release attained at 24 h. However, the amoxicillin release rate was delayed even more with tablets from preparation B1, with 13% released in the first hour, 50% at 4 h and <70% at 6 h, with 100% release being attained sometime before 24 h. The results indicated that the formulations containing amoxicillin coated with EC could sustain but not prevent the release of amoxicillin from the tablets better than that of the formulation containing only amoxicillin without an EC coating. Thus, the entire payload (amoxicillin) was made available, with none excluded by remaining trapped in the voided delivery particle, but just made available at a lower concentration over a longer time period. However, the sustained amoxicillin release rate observed also depended upon the particle size, where the smallest particles (<75 μm) gave an amoxicillin

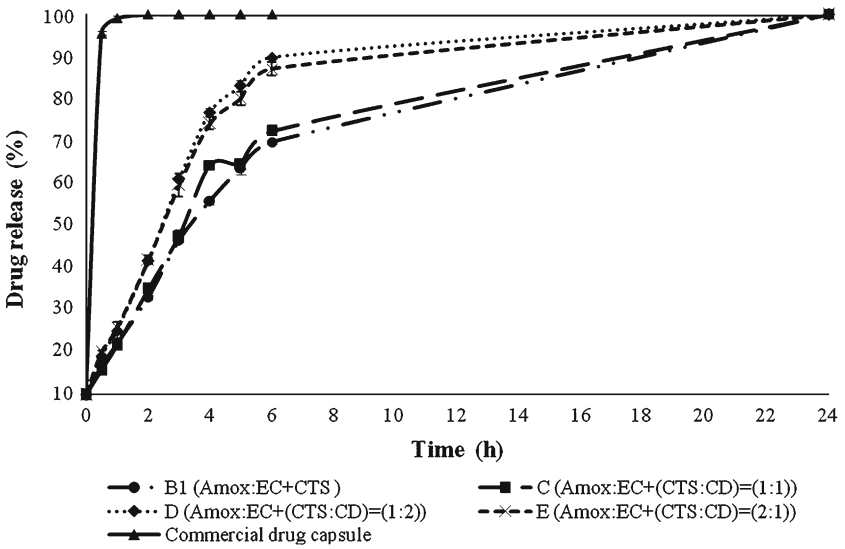

Fig. 9. Release profile of amoxicillin from tablets formulations B1, C, D, E (see Table I for composition) and a commercial drug capsule when immersed in SGF medium. Data are corrected for the amoxicillin degradation factor, and are shown as the mean±1 SD derived from three repeats

release profile that was similar to that of the uncoated formulation A. This is consistent with the SEM photographs of size three sorted particles (<75 μm) derived from a 1:1 (*w*/*w*) ratio of amoxicillin/EC that showed that there was a lower amount of EC coated onto the amoxicillin particles, and thus large areas of unprotected crystals that would be exposed to the water and so solvate rapidly. In contrast, the larger particles of tablet B1 (75–100 μm) showed the most delayed release profile of amoxicillin when compared to other formulations. Therefore, tablet B1 was chosen for further studies to try to improve the release profile.

Effect of the Amoxicillin/EC Ratio on the Drug Release Profile

The above results of release profiles revealed that the particle sizes in the range of 75–100 μm (size 2) produce the sustained-release profile of amoxicillin in the acidic conditions of SGF. Therefore, in the subsequent studies, amoxicillin–EC particles of (75–100 μm) size were used as a fixed parameter. Three different (*w*/*w*) ratios of amoxicillin/EC (1:1, 1:2, and 2:1) were next evaluated as a potential variable factor to retard the drug release.

The release rates of amoxicillin from the tablets derived from the three different (*w*/*w*) ratios of amoxicillin/EC were essentially the same as each other, and thus the different (*w*/*w*) ratios of amoxicillin/EC did not seem to affect the release rates of amoxicillin (Figs. 7 and 8). However, tablet B3 presented the faster release rate (80% released at 6 h) than the other two formulations. Thus, tablets B1 and B2 showed a very similar amoxicillin release profile with 80% of drug released at nearly 14 h. Therefore, tablet B1 was selected for further studies as it contained a lower amount of EC for coating the amoxicillin, and so would decrease the cost of production.

Effect of a CS–CD Mixed Polymer Matrix on the Amoxicillin Release Rates

The release profile of formulation B1 (Table I) tablets, comprised of size 2 (75–100 μm) particles, were selected for further studying the effect of the inclusion of CD to CS as the retardant, upon the amoxicillin release rate profiles by varying the (*w*/*w*) ratios of CS/CD (1:1, 1:2, and 2:1).

The release profiles of amoxicillin from polymer matrix tablets with different ratios of CS/CD (formulations C, D, and E) with a particle size of 75–100 μm compared with the commercial drug capsules are shown in Fig. 9. All three tablet formulations (C, D, and E) showed a more sustained-release profile than that of the commercial capsule. However, tablet C (with CD) exhibited a release profile more similar to that of Tablets B1 (without CD). Furthermore, the different weight ratios of CS/CD did not appear to significantly influence the release profile of amoxicillin. The results were consistent with the results obtained from FT-IR to XRD spectrum that amoxicillin were no chemical reaction with CD, therefore, the CD could not help to retard the amoxicillin.

CONCLUSION

Sustained release of amoxicillin under SGF conditions was achieved by coating amoxicillin with EC for protection and sustained release, and then further coating the preparation with the CS or CS–CD mixture as a retard polymer and selecting particles of 75–100 μm in size. Amoxicillin is not decomposed when coated with EC by solid dispersion, and increasing the EC contents did not initiate any detectable amoxicillin-EC interactions. The degradation behavior of amoxicillin in SGF revealed that the *in vitro* release of amoxicillin followed first-order kinetics ($k_2 = -0.0963$), and was greatly improved (sustained release) upon coating with EC and a CS or CD–CS composite. This is probably due to the fact that CD did not interact with CS as an interpolymer complex, consistent with the observed FT-IR spectra. Overall, it was clearly shown that a sustained release of amoxicillin under SGF conditions was achieved with amoxicillin coated with EC at a (*w*/*w*) ratio of 1:1 with subsequent coating with a (1:1) ratio of CS/CD and selecting for particles in the size range of 75–100 μm. As pointed out before, it is important to note that only the release kinetics was delayed and that all of the amoxicillin antibiotic was released within 24 h (desired effect), with none remaining trapped in the delivery vehicle to be voided (undesired effect). Thus, more exact dosages can be calculated and administered, reducing wastage, costs, and non-desired side effects from *off situ* drug delivery. The tablet formulations of amoxicillin may be an advantageous alternative for an orally administered sustained-release formulation, and be helpful for the treatment of peptic ulcers.

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