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Spray-dried carbamazepine-loaded chitosan and HPMC microspheres: preparation and characterisation

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Abstract

In this study, the potential of the spray-drying technique for preparing microspheres able to modify the release profile of carbamazepine was investigated. Low-, medium- and high-molecular-weight chitosan and hydroxypropyl methylcellulose (HPMC) in different drug–polymer ratios were used for the preparation of microspheres. The microspheres, characterized by X-ray powder diffractometry (XRD) and differential scanning calorimetry (DSC), were also studied with respect to particle size distribution, drug content and drug release. The results indicated that the entrapment efficiency (EE), as well as carbamazepine release profile, depended on polymeric composition and drug–polymer ratios of the microspheres prepared. The best entrapment efficiencies were obtained when chitosan of low-molecular-weight (CL) or HPMC were used for the microencapsulation. For all types of polymer used, the microspheres with low carbamazepine loading (6.3% w/w) showed better control of drug release than the microspheres with higher drug loadings. The HPMC microspheres showed the slowest carbamazepine release profile with no initial burst effect. Carbamazepine release profiles from ternary systems, carbamazepine–CL–HPMC microspheres, depended mostly on HPMC content and showed similar carbamazepine release profile as CL microspheres when HPMC content was low (9:1 CL–HPMC ratio, w/w). Otherwise, the carbamazepine release from CL–HPMC microspheres was remarkably faster than from either chitosan or HPMC microspheres. The release profile of carbamazepine from the microspheres was highly correlated with the crystalline changes occurring in the matrix.

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Introduction

Carbamazepine is a widely used anti-epileptic drug. It is used to control grand-mal seizure as well as in the treatment of trigeminal neuralgia. The drug is characterised by slow and irregular gastrointestinal absorption due to its low water solubility. Carbamazepine is also characterized by a short half-life on chronic dosing because of the auto-induction of hepatic metabolism. The initial half-life is about 24 h falling to approximately 12 h with chronic monotherapy and 8 h in those patients receiving other enzyme-inducing drugs (Larkin et al 1989). It is therefore both important and advisable to have a drug formulation characterized by prolonged carbamazepine release (Giunchedi et al 1991; Arnaud et al 1996; Katzhendler et al 1998, 2000).

The objective of this investigation was to study chitosan and HPMC microspheres as swellable and mucoadhesive carriers for carbamazepine. In such a hydrophilic system the water–polymer interaction would be modulated, giving a sustained release of a water-insoluble drug like carbamazepine. Microspheres differing in polymeric composition and drug–polymer ratio were prepared by spray-drying, which is a rapid high-yield technique applicable at industrial scale. Also, considering that carbamazepine can exist in at least four polymorphic forms and a dihydrate, spray-drying could lead to a modification of its solid state (Lowes et al 1987; Dugué et al 1991; Rustichelli et al 2000).

Chitosan is cationic polysaccharide derived by deacetylation of chitin that is, after cellulose, the most abundant polymer found in nature. Chitosan is a hydrophilic, biocompatible and biodegradable polymer of low toxicity and it has been extensively

investigated for pharmaceutical and medical purposes. In the context of drug delivery, chitosan has been used for preparation of microcapsules and microspheres with encapsulated proteins, enzymes, DNA and cells, as a nasal delivery system for insulin, as a system for oral vaccination and as a stabilizing constituent of liposomes. Several studies have highlighted the potential use of chitosan as an absorption-enhancing agent. Because of its bioadhesive properties, chitosan has also received considerable attention in novel bioadhesive drug delivery systems, which are aimed at improving the bioavailability of drugs by prolonging the residence time at the site of absorption (Paul & Sharma 2000).

Among the various hydrophilic polymers employed for drug release control, hydroxypropyl methylcellulose (HPMC) is the most commonly used polymer in matrices for extended release of drugs, due to its versatility, safety and compatibility with many drugs (Chien 1992). HPMC can take up and retain large amounts of water, which influences the physical and chemical properties of polymer and drug-release profile (Nokhodchi & Rubinstein 2001).

The microspheres were characterized by differential scanning calorimetry (DSC) and X-ray diffractometry (XRD) to elucidate the structure of the microspheres and to detect possible drug–carrier interactions. The drug encapsulation efficiency, the size and size distribution and the morphological properties of the microspheres were studied as a function of type and percentage of carrier. The release profiles were investigated for all preparations to evaluate the influence of polymeric composition and drug loading of microspheres on carbamazepine release rate.

Materials and Methods

Materials

Chitosans with different molecular weight (CH, CM and CL) were purchased from Fluka (Buchs, Switzerland): CH (M_r 600 000; deacetylation degree 83%), CM (M_r 400 000; deacetylation degree 83.5%), and CL (M_r 150 000; deacetylation degree 87.4%). HPMC was kindly donated by Synthpharm (Mülheim, Germany) and carbamazepine, reagent grade, by Zanchetta (Lucca, Italy). All other chemicals used were of analytical grade and purchased from Kemika (Zagreb, Croatia).

Microsphere preparation

All batches of microspheres were prepared by spray-drying using a Büchi 190 mini spray drier (Flawil, Switzerland) with a standard 0.5-mm nozzle. The liquid was fed to the nozzle with a peristaltic pump, atomised by the force of the compressed air and blown together with a hot air to the chamber where the solvent in the droplets was evaporated. The dry product was then collected in a collection bottle. The drying conditions were as follows: spray flow rate of 0.25 L h^{-1}, compressed air flow rate of 700 N L h^{-1}, inlet air temperature of 120 °C and outlet air temperature of 75 °C.

Microspheres containing carbamazepine with different polymeric composition were prepared. Tables 1–3 report compositions and corresponding production yields (shown as a percent weight of microspheres obtained with respect to the initial amounts of drug and polymers).

Chitosan microspheres containing carbamazepine

Different types of chitosan (Table 1) at 1% (w/v) concentration were solubilized in 0.5% acetic acid solution. To obtain microspheres with different theoretical drug loadings, carbamazepine was dissolved at different concentrations (0.1, 0.2, 0.3 and 0.5% w/v) in 96% ethanol and mixed with chitosan solution in a 1:1.5 (v/v) ratio. The mixtures were subjected to spray-drying as described above. Table 1 reports the composition of carbamazepine-loaded chitosan microspheres produced.

HPMC microspheres containing carbamazepine

HPMC was dissolved in a mixture of 96% ethanol and water (2:3 v/v). The polymer concentration was 1% (w/v). To obtain microspheres with different theoretical drug loadings, carbamazepine was dissolved at different concentrations (0.1, 0.2, 0.3 and 0.5% w/v) in ethanol and added to HPMC solution in a 1:1 (v/v) ratio. The mixtures were spray-dried under the conditions described above. The characteristics of microspheres prepared are given in Table 2.

CL–HPMC microspheres containing carbamazepine

For the preparation of the CL–HPMC microspheres, CL chitosan and the HPMC solutions were prepared as described above at 1% (w/v) concentration. The CL–HPMC ratio varied between the preparations and was 1:1, 7:3 and 9:1 (w/w). The ethanolic solution of carbamazepine (0.1 or 0.3% w/v) was mixed with solution of polymers in 1:1.5 (v/v) ratio. Mixtures were spray-dried under the conditions described above. The microspheres obtained, and their characteristics, are given in Table 3.

Encapsulation efficiency determination

The drug content of the microspheres was determined spectrophotometrically ($\lambda = 285$ nm; Ultrospec Plus, Pharmacia LKB). The chitosan microspheres (50 mg) loaded with carbamazepine were dissolved in 15 mL of 0.1 M HCl under sonication. The microspheres consisting of chitosan and HPMC (50 mg), loaded with carbamazepine were dissolved in the mixture of 0.1 M HCl and 96% ethanol (1:1, v/v; 15 mL) under sonication. The solutions were filtered and the amount of carbamazepine was measured. Preliminary studies showed that the presence of dissolved polymers did not interfere with the carbamazepine absorbance at 285 nm.

Particle size analysis

A microscopical image analysis technique for determination of microsphere size distribution was applied. The

Table 1 Preparation and characteristics of chitosan microspheres with carbamazepine.

Sample	Theoretical drug loading (%)	Chitosan type								
		CL			CM			CH		
		Yield (%)	EE (%)	Diameter (μm)	Yield (%)	EE (%)	Diameter (μm)	Yield (%)	EE (%)	Diameter (μm)
1	6.3	37.9 ± 2.2	93.6 ± 4.2	2.77 ± 1.02	39.4 ± 5.9	41.6 ± 4.7*	2.63 ± 1.28	47.5 ± 2.9	52.8 ± 2.4*†	2.69 ± 1.12
2	11.8	42.9 ± 2.4	98.9 ± 1.6	2.74 ± 1.06	46.5 ± 2.7	55.3 ± 3.9*	2.68 ± 1.04	45.9 ± 1.1	68.0 ± 1.0*†	2.71 ± 0.98
3	16.7	42.7 ± 4.4	100 ± 0.2	2.69 ± 0.91	46.7 ± 4.3	76.8 ± 4.6*	2.61 ± 1.14	49.5 ± 1.9	76.2 ± 0.2*†	2.63 ± 0.83
4	25.0	44.4 ± 2.4	82.7 ± 2.6	2.43 ± 0.88	50.5 ± 1.1	84.0 ± 3.0	2.71 ± 0.96	39.4 ± 3.6	83.6 ± 3.3	2.68 ± 1.07

EE, Entrapment efficiency = drug loading/theoretical drug loading × 100. Values are mean ± s.d. (n = 3). *$P < 0.05$, compared with CL microspheres. $^{\dagger}P < 0.05$, compared with CM microspheres.

Table 2 Preparation and characteristics of HPMC microspheres with carbamazepine.

Sample	Theoretical drug loading (%)	Yield (%)	EE (%)	Diameter (μm)
H1	6.3	35.8 ± 2.0	89.1 ± 3.9	3.97 ± 1.69
H2	16.7	37.3 ± 2.1	88.2 ± 3.2	3.99 ± 1.44
H3	23.1	39.2 ± 3.2	99.7 ± 0.3	3.57 ± 1.35
H4	33.4	39.7 ± 1.6	96.0 ± 3.5	3.54 ± 1.39

EE, Entrapment efficiency = drug loading/theoretical drug loading × 100. Values are mean ± s.d. (n = 3).

morphology and particle size distributions (based on the numbers of particles) were determined using an Olympus BH-2 microscope equipped with a camera (CCD Camera ICD-42E; Ikegami Tsushinki Co., Japan) and computer-controlled image analysis system (Optomax V, Cambridge). The microspheres were dispersed on a microscope slide. The microscopical field was scanned by video camera. The images of the scanned fields were digitalised and analysed by the software (Optomax V Software, Cambridge). In all measurements at least 3000 particles were examined.

X-Ray powder diffraction (XRD) analysis

Samples of spray-dried systems and raw materials were studied by means of X-ray powder diffraction technique (XRD) using a diffractometer (STOE D500; Siemens, Munich, Germany) with CuK_α radiation ($\lambda = 1.5418$ Å), monochromatised by a secondary flat graphite crystal. The scanning angle ranged from 3 to 35° of 2θ, the steps were 0.1° of 2θ and the counting time was 1 s/step. The mains-power was 20 mA and the accelerative-voltage was 40 kV.

Differential scanning calorimetry (DSC) analysis

The physical state of the drug in the samples was assayed by differential scanning calorimetry (DSC). The calibration of the instrument was performed with indium and lead for the temperature, and with indium for measurement of the enthalpy. This analysis was carried out using a differential scanning calorimeter (Mod. TA 4000, equipped with a measuring cell DSC 20 Mettler). Samples containing about 0.06 mg of carbamazepine were placed in pierced aluminium pans and heated from 30 to 210 °C, under air atmosphere. For better distinguishing between the polymorphs of the drug, the scanning rate was 40 °C per min, as suggested for carbamazepine by several authors (Krahn & Mielck 1987; Lowes et al 1987; Rustichelli et al 2000; Bettini et al 2001; Moneghini et al 2001).

Release of carbamazepine from microspheres

The release profiles of carbamazepine from microspheres were examined in phosphate buffer (pH 6.8). The drug-loaded microspheres containing 10 mg of carbamazepine were put into a rotating basket (50 rev min^{-1}) and placed in 500 mL of the dissolution medium, thermostatted at 37 °C. At scheduled time intervals agitation was stopped, the samples (2 mL) were withdrawn and replaced with fresh medium. The samples were filtered and assayed spectrophotometrically at 285 nm.

Statistical analysis

All values are expressed as means ± s.d. obtained from n separate experiments. The differences between data for the effect of polymer composition and drug loading on production yield, entrapment efficiency, and microsphere size were analysed using a Kruskal–Wallis test. Statistical ignificance for the comparison of release profiles was tested by one-way analysis of variance. Statistically significant differences were defined as P values of less than 0.05. Calculations were performed with the GraphPad Prism program (GraphPad Software, Inc., San Diego, CA; www.graphpad.com).

Results and Discussion

Characterisation of chitosan microspheres with carbamazepine and drug release

Three types of chitosan with different molecular weight were used for the preparation of microspheres. The characteristics of microspheres are shown in Table 1. The amount of carbamazepine varied among the preparations while the amount of chitosan was kept constant.

The preparation method produced well-formed microspheres with good morphological characteristics for all batches. Scanning electron microscope studies confirmed that the molecular weight of chitosan had no influence on the size and appearance of microspheres (data not shown). Particle size analysis indicated narrow logarithmic-normal distribution for all samples with 60–70% of particles having spherical diameter ranging from 1 to 3 μm (Table 1). Neither chitosan molecular weight nor drug loading of the microspheres influenced particle size characteristics. This fact was already reported for chitosan microspheres prepared by other methods (Berthold et al 1996; Genta et al 1998), even though He et al (1999) found influence of chitosan molecular weight on microsphere sizes.

The entrapment efficiency ranged between 41.6 ± 4.7 and 100% (Table 1). Comparing the entrapment efficiency

Table 3 Preparation and characteristics of microspheres with carbamazepine made of mixture of HPMC and CL chitosan.

Theoretical drug loading (%)	CL chitosan–HPMC ratio (w/w)											
	1:1				7:3				9:1			
	Sample	Yield (%)	EE (%)	Diameter (μm)	Sample	Yield (%)	EE (%)	Diameter (μm)	Sample	Yield (%)	EE (%)	Diameter (μm)
6.3	CLH1	28.3 ± 2.5	76.2 ± 1.6	2.99 ± 1.12	CLH2	29.9 ± 2.1	100 ± 1.0*	2.87 ± 0.97	CLH3	33.2 ± 3.9	100 ± 0.7*	2.78 ± 1.12
16.7	CLH4	31.6 ± 2.3	92.9 ± 3.6	3.25 ± 1.23	CLH5	38.8 ± 0.6*	94.0 ± 1.4	3.03 ± 1.11	CLH6	35.5 ± 1.9*	69.6 ± 0.8*†	3.17 ± 1.16

EE, Entrapment efficiency = drug loading/theoretical drug loading × 100. Values are mean ± s.d. (n = 3). *$P < 0.05$, compared with 1:1 microspheres. $^{\dagger}P < 0.05$, compared with 7:3 microspheres.

obtained with different types of chitosan used (CL, CM and CH), the highest entrapment, 82.7 ± 2.6–100%, was obtained when CL chitosan was used for encapsulation, probably due to the highest degree of deacetylation of CL chitosan (87.4%). Similar findings have already been reported (Genta et al 1998; Martinac et al 2002). Further, increase of drug loading efficiency was obtained at higher concentrations of chitosan in the preparative mixture (Table 1).

The release profiles of carbamazepine from the chitosan microspheres are shown in Figure 1. All batches of carbamazepine-loaded microspheres showed the most remarkable differences in drug release in the first 5 h.

A better control of drug release was obtained with the microspheres of the lowest carbamazepine loading (6.3% w/w) than with the microspheres of the highest drug loading (25% w/w) for all types of chitosan used (Figure 1). When microspheres of hydrophilic polymers are immersed in water, they swell and form a gel diffusion layer that hinders the outward transport of the drug within the matrix, hence producing a controlled-release effect (Lim et al 2000). As the amount of polymer increases, the thickness of the hydrogel layer increases as well, and the drug diffusion is more retarded. That can explain the slower release of lipophilic carbamazepine from the microspheres with low drug loading.

When comparing the carbamazepine release profiles from microspheres composed of different molecular weight chitosans, but with the same drug loading, it could be seen that the molecular weight of the chitosan has very scarce effect on carbamazepine dissolution rate, in agreement with previous findings (He et al 1999). Although there was a significant difference ($P < 0.05$) in carbamazepine release between CL and CM microspheres for all drug loadings, CL microspheres significantly differed ($P < 0.05$) in carbamazepine release from CH microspheres only for lowest drug loading (6.3%; Figure 1A) while release of carbamazepine from CM and CH microspheres was significantly different ($P < 0.05$) for higher drug loadings (16.7 and 25%; Figure 1C, D).

Figure 1 also shows an initial burst (10–50% in 10 min) of carbamazepine release from all batches of microspheres. This fact, already noticed by He et al (1999), is most likely due to the hydrophilic character of the chitosan and to the small dimensions of the microspheres. The initial rapid release has been reported not only to occur

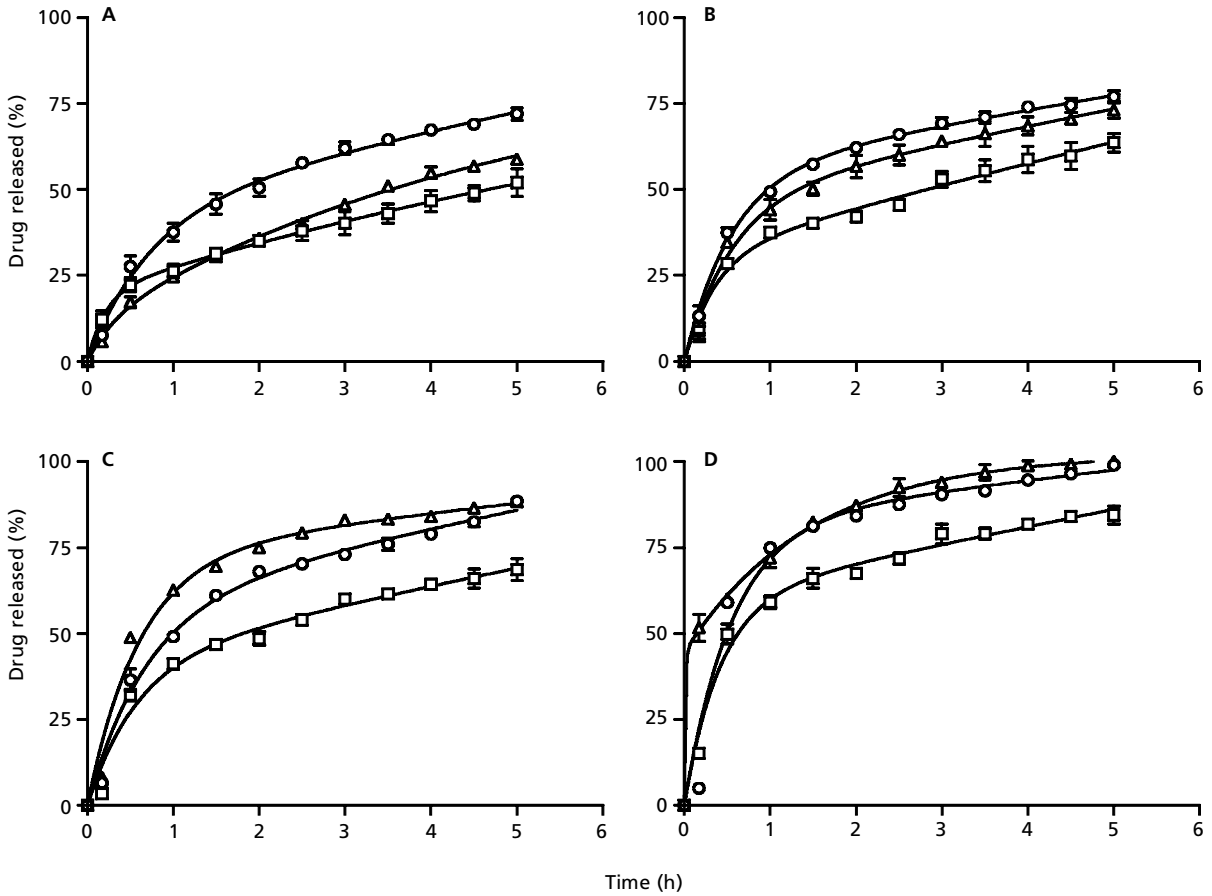

Figure 1 The release profiles of carbamazepine from chitosan microspheres made of theoretical drug loading (w/w) 6.3% (A), 11.8% (B), 16.7% (C) and 25% (D). Type of chitosan: CL (○); CM (□); CH (△). Data are mean ± s.d., n = 3. In some cases, the error bars are within the size of data points.

with water-soluble drugs but also with some less water-soluble drugs, such as dexamethasone (Pavanetto et al 1994) and nicardipine (Conte et al 1994). Further, in this case the carbamazepine reduction in crystallinity (see below), caused by spray-drying, enhanced its dissolution rate (Moyano et al 1995). It must be pointed out that the initial rapid drug release may have a functional use in providing an initial dose during the drug delivery, minimising any lag period. The microspheres with the lowest drug loading presented the lowest burst effect.

Differential scanning calorimetry (DSC) and X-ray powder diffraction (XRD) were performed on the raw materials and on the microspheres to assess the physical structure of microspheres in the solid state.

The DSC trace of pure carbamazepine (Figure 2A) showed only a melting endotherm at 191.2 °C with a heat of fusion of 106.0 Jg^{-1}. This fact attested that the starting drug was a sample mainly composed of the trigonal polymorph.

In the thermogram of pure chitosans (regardless of the M_r) (Figure 2A), two endothermic events could be recognised: a broad endotherm ranging from about 50 to 110 °C corresponding to the hydration water and a second one with a maximum at about 167.2 °C, followed by an inflection on the baseline, due to the thermal degradation of the substance at temperatures superior to 175 °C.

The carbamazepine–chitosan microspheres prepared with low-molecular-weight chitosan (CL samples; Figure 2B) always showed the melting peak of the carbamazepine but shifted to a lower temperature than of the pure drug. The lowering of the peak and the loss of its sharp appearance became more evident as the carrier content increased. It seems like a significant reduction of degree of the drug crystallinity appeared due to both the carrier and the spray-drying process. Further, the shift of the melting peak to a lower temperature could be due to an interaction between drug and chitosan, or due to the formation of a crystalline complex between them. In the system with the lowest drug loading (CL1, 5.9%), only one endotherm of the drug was detected, while in the systems of increased carbamazepine loading (CL2–4), a further peak at 190 °C was also detected representing a residual amount of crystalline carbamazepine that melted. It could be assumed that at concentrations superior to 5.9% not the whole amount of drug is involved in the crystalline complex with the carrier.

Carbamazepine might interact with chitosan in the position of an amino group, as confirmed by DRIFT analysis conducted as previously reported by Passerini et al (2002). In fact, a dramatic shift in the carbonyl stretching peak was noticed (from 1691 in the original carbamazepine to 1658 cm^{-1} in the spray-dried systems), absent in the corresponding physical mixtures (data not shown).

The DSC thermograms of the chitosan microspheres with carbamazepine prepared with medium-weight chitosan (CM samples) are reported in Figure 2B. Similarly to CL systems, in both CM samples, the carbamazepine peak could be distinguished; however when chitosan content increased (sample CM1) a great shift of the endotherm

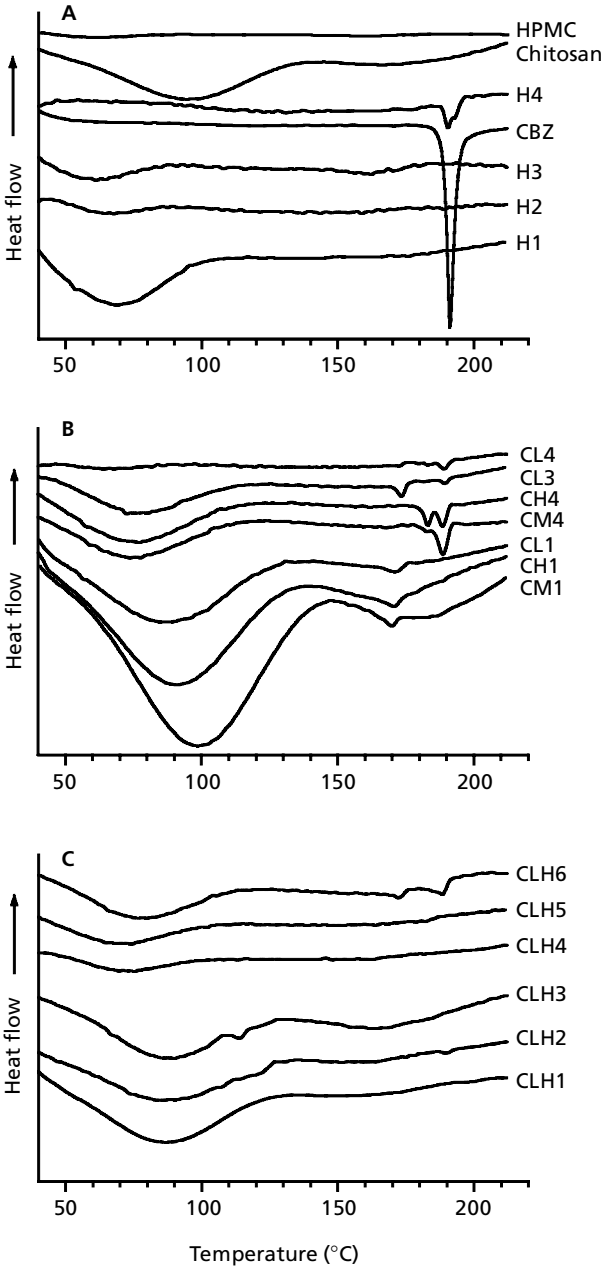

Figure 2 DSC curves of raw materials and HPMC microspheres (A), chitosan microspheres (B) and ternary systems CL–HPMC (C). CBZ, carbamazepine.

of fusion of the drug could be attested (from 191.2 of the starting carbamazepine to 188.2 of the sample CM4 and finally to 171 °C of the sample CM1).

Further, in the carbamazepine microspheres prepared with high-molecular-weight chitosan (CH samples; Figure 2B) a remarkable interaction between carrier and carbamazepine could be recognised, which is responsible for the shift of the melting peak towards temperatures lower than 190 °C. It must be pointed out that in sample CH4 an anomalous feature was found: the splitting in two peaks of the endotherm of fusion of the drug. This fact could be

attributed to a partial interaction of the drug with the polymer, which caused the shift of the peak to 183.3 °C. On the other hand, part of the carbamazepine crystals remained unaltered and melted at 188.6 °C, as usual. The sample CM4 showed a similar behaviour, even if with a lesser magnitude.

The X-ray diffraction patterns of raw materials and chitosan microspheres are presented in Figures 3A and 3B, respectively. The diffractogram of pure carbamazepine (Figure 3A) revealed the presence of some peaks at value of 2θ lower than 10°, having low intensities, in particular at 6.1 and 9.4° of 2θ, and some major signals at 12.2, 19.9 and 22.8° of 2θ, typical of the trigonal polymorph I (Lowes et al 1987; Rustichelli et al 2000), thus confirming the previous DSC results.

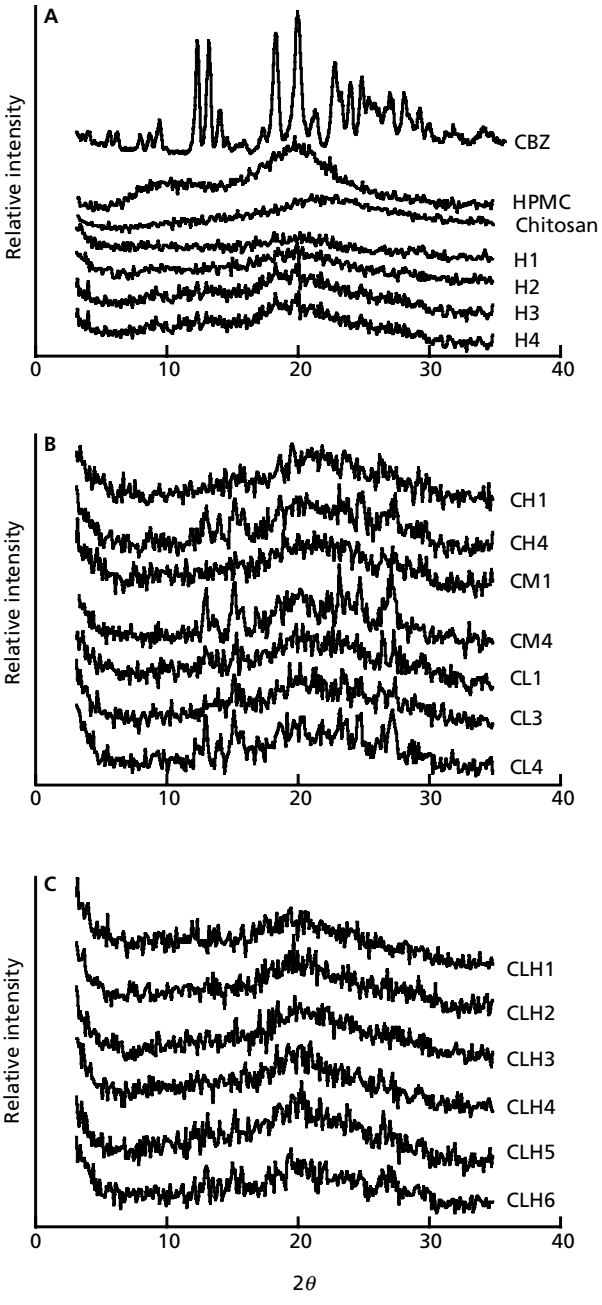

Figure 3 XRD patterns of raw materials and HPMC microspheres (A), chitosan microspheres (B) and ternary systems CL–HPMC (C). CBZ, carbamazepine.

The XRD patterns of pure chitosans (Figure 3A) were typical of amorphous substances, without any intense peak in their diffractograms being detectable and being characterised by an evident phenomenon of scattering.

The carbamazepine–CL systems always indicated the presence of crystalline carbamazepine but with a dramatic decrease of the intensity of the signal due to both a dilution effect and to a decrease in crystallinity of the drug in the microspheres. As expected, the XRD patterns of these systems (Figure 3B) revealed an increase of the intensity of the signal when increasing the amount of carbamazepine in the microspheres. Further, several signals not attributable to the drug could be detected in all systems, indicating the presence of interaction between drug and CL, in agreement with previous DSC findings.

The XRD patterns of the carbamazepine–CM systems (Figure 3B) still revealed the presence of carbamazepine in a crystalline state, even if a remarkable reduction of the intensity of its peaks could be noticed. Also, in this case it must be underlined that a precise correspondence with the signals of the pure drug could not be found. The presence of signals other than those of pure carbamazepine indicated the existence of some interactions between drug and chitosan CM, as already suspected by DSC analysis.

Once again, among carbamazepine–CH samples (Figure 3B), the CH1 sample was found to be very scarcely crystalline, while the sample CH4 showed some new signals not attributable to the drug.

Characterisation of, and drug release from, HPMC microspheres with carbamazepine

The samples prepared, the carbamazepine content and entrapment efficiency of the HPMC microspheres are reported in Table 2. The averaged efficiency of drug encapsulation into microspheres improved with respect to previous CM and CH chitosan microspheres, while it was similar to the entrapment efficiency of CL chitosan microspheres. The highest encapsulation efficiency (99.7%) was obtained for H3 microspheres. It may be attributed to HPMC ability to promote the amorphisation of carbamazepine, and it could also be the reason why it was possible to prepare the HPMC microspheres with a drug loading of 33.4% by spray-draying procedure.

Figure 4 represents the effect of HPMC content on carbamazepine release from the microspheres. It was revealed that increasing the HPMC content in the matrix decreased the fraction of carbamazepine released. Only 25% of carbamazepine was released from the HPMC microspheres with a drug loading of 6.3% (w/w) within 5 h, while 75% was released in the same period with a drug loading of 33.4% (w/w). The microspheres with high drug content were expected to be more porous than those with low drug con-

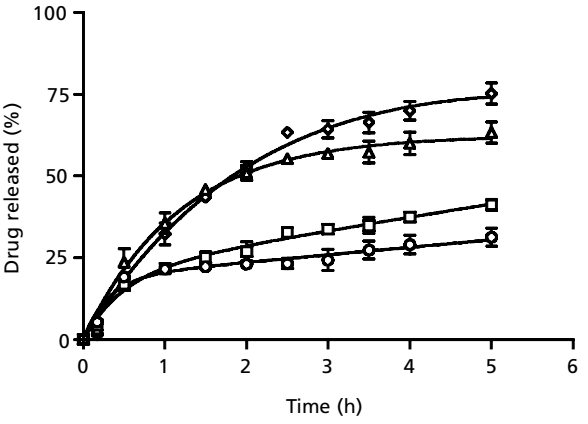

Figure 4 The release profiles of carbamazepine from HPMC microspheres made of theoretical drug loading (w/w) 6.3% ( ), 16.7% ( ), 23.1% ( ) and 33.4% ( ). Data are mean ± s.d., n = 3. In some cases, the error bars are within the size of data points.

tent, which might facilitate the release of residual drug from microspheres (Wan et al 1994). This could explain the significant difference ($P < 0.05$) in the carbamazepine release between the HPMC microspheres with the lowest (H1; 6.3%) and the highest drug loading (H4; 33.4%). However, there was no significant difference in the carbamazepine release ($P > 0.05$) between H1 and H2 (6.3% and 16.7% drug loading, respectively) or between H3 and H4 (23.1% and 33.4% drug loading, respectively) microspheres.

The DSC curve of the HPMC (Figure 2A) showed a broad endotherm of dehydration with a maximum at about 75 °C, and the polymer started slowly to degrade at 170 °C. According to these results, its diffractogram (Figure 3A) is a halo pattern, typical of an amorphous substance.

No endothermic peak corresponding to the fusion of carbamazepine was observed in the thermograms of the HPMC microspheres having drug concentrations lower than 32% (w/w) (Figure 2A). This can be explained by the complete dissolution of carbamazepine crystals in the spray-dried HPMC microspheres (i.e., the presence of carbamazepine in an amorphous state). This fact is not surprising since both the spray-drying process and HPMC have been reported to be able to promote the amorphisation of drugs (Moyano et al 1995; Katzhendler et al 1998). As for H4 microspheres, no shift of the drug melting peak was noticed.

In agreement with DSC data, the diffractograms of the carbamazepine–HPMC systems (Figure 3A) showed the capacity of the HPMC to change the crystallinity of carbamazepine with the formation of a less organised crystal structure and amorphous appearance in the samples H1, H2 and H3. On the other hand, the diffractogram of the system having the highest content of carbamazepine (sample H4) showed a few signals characteristic of the drug.

Characterisation of, and drug release from, CL–HPMC microspheres with carbamazepine

Finally, ternary systems based on mixtures of CL and HPMC in different ratios were prepared (carbamazepine–CL–HPMC). The characteristics of these microspheres are given in Table 3. No direct correlation could be drawn between the polymeric compositions and the entrapment efficiency.

Figure 5 shows release profiles of carbamazepine from the CL–HPMC microspheres in comparison with release profiles of carbamazepine from HPMC and CL microspheres with the same theoretical drug loading. The microspheres having low theoretical drug loading (6.3% w/w) showed no significant differences in drug release profiles. An exception were the microspheres with CL–HPMC weight ratio of 9:1 (sample CLH3) that exhibited the slowest carbamazepine release among the ternary systems ($P < 0.05$; Figure 5A). Further, despite the difference in polymeric composition, about 80% of carbamazepine

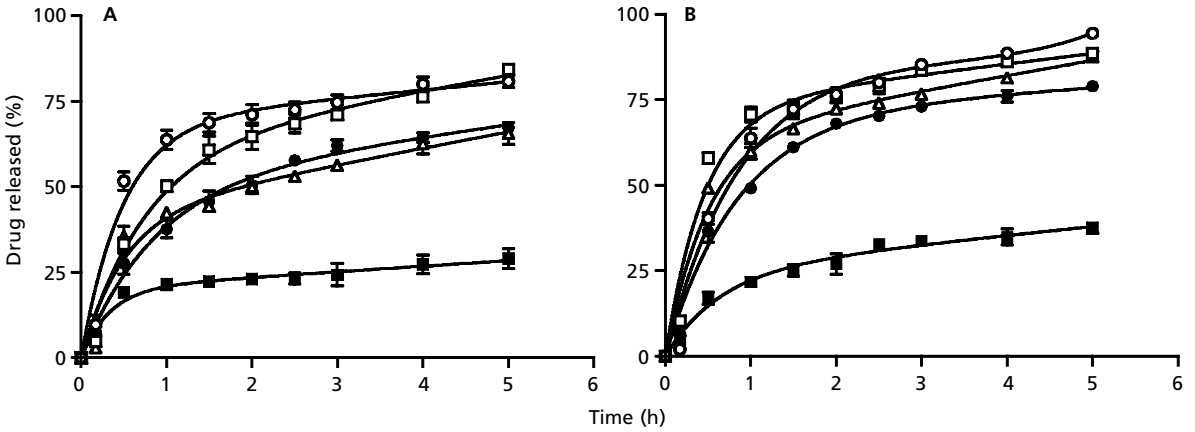

Figure 5 Carbamazepine release from CL–HPMC, HPMC and CL microspheres with theoretical drug loading (w/w) of 6.3% (A) and 16.7% (B): CL–HPMC 1:1 w/w (○); CL–HPMC 7:3 w/w (□); CL–HPMC 9:1 w/w (△); CL (●); HPMC (■). Data are mean ± s.d., n = 3. In some cases, the error bars are within the size of data points.

was released in 2 h from all CL–HPMC microspheres with theoretical drug loading of 16.7% (w/w) and they all showed similar release profiles (Figure 5B).

The release of carbamazepine was faster from all ternary systems (CL–HPMC microspheres) than from the HPMC microspheres, for both drug loadings. Comparing the CL–HPMC and CL microspheres of the same theoretical drug loading, it was revealed that the CL microspheres and CL–HPMC microspheres with high CL–HPMC weight ratio (9:1, w/w) showed similar dissolution profiles, while the carbamazepine release was somewhat faster from CL–HPMC microspheres with higher HPMC content. A possible explanation of these dissolution performances could be that an interaction between HPMC and chitosan occurred, since they are oppositely charged, and that it influenced the hydration and swelling of the ternary systems matrix, which consequently behaved more like pure chitosan matrix with more amorphous carbamazepine due to HPMC.

Figure 2C presents the DSC curves of the CL–HPMC systems. At drug proportions ranging from 4.8 to 6.3% (CLH1–3), the peak of the drug is not possible to distinguish, probably because of the total amorphisation of the drug. At a carbamazepine content superior to 11.6% (CLH4–6), the carbamazepine peak starts to be visible. However, the degree of crystallinity seems to be dependent on the content of HPMC. In fact, on increasing the amount of HPMC, the amorphisation of the drug was again evident. These data are in agreement with those regarding the binary mixture carbamazepine–HPMC. It must be pointed out that in the case of the sample CLH6, containing the highest amount of CL chitosan, the thermogram showed two carbamazepine peaks. It appears that the same phenomenon occurred as with the binary mixtures carbamazepine–CL.

The diffractograms of these systems are depicted in Figure 3C. The samples CLH1 to CLH3 revealed a solid state of the drug, completely amorphous. In their diffractograms, no carbamazepine peaks could be recognised, only a halo pattern similar to that of pure HPMC and chitosan. The ternary systems CLH4, CLH5 and CLH6 showed a progressively increasing degree of crystallinity.

Conclusion

In this study, well shaped spherical microspheres able to promote a sustained release of carbamazepine were produced by spray-drying technique, using chitosan or HPMC (or both) as swellable carriers. The drug entrapment efficiency, as well as the carbamazepine release profile, was influenced by the polymeric composition and drug–polymer ratios of the microspheres prepared. The best entrapment efficiency was obtained when chitosan of low molecular weight (CL) or HPMC was used for the microencapsulation. Independently of the type of polymer used, the microspheres with low carbamazepine loading (6.3% w/w) showed better control of drug release than the microspheres with higher drug loadings. The HPMC microspheres showed the slowest carbamazepine release profile with no initial burst effect, whereas an initial rapid carbamazepine release was found in the chitosan microspheres, regardless of the type of chitosan used. The sustained-release action of carbamazepine from the three types of chitosan appeared to be due to both the formation of a gel diffusion layer that controls the transport of the drug, and to the drug–polymer interaction, revealed by the physicochemical characterisation. Conversely, no drug–polymer interaction was observed in HPMC–carbamazepine microspheres, thus the carbamazepine release prolonging effect may be attributed to the ability of HPMC to rapidly form a gel layer at the microsphere periphery exposed to aqueous media. Finally, in the carbamazepine–CL–HPMC systems (representing a sum of the characteristics of the binary systems from both the physicochemical and dissolution point of view), a major effect of the CL–HPMC ratio on the carbamazepine release rate was noticed in the systems with low drug loading.

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