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Chitosan microspheres prepared by spray drying

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Abstract

Non-crosslinked and crosslinked chitosan microspheres were prepared by a spray drying method. The microspheres so prepared had a good sphericity and a smooth but distorted surface morphology. They were positively charged. The particle size ranged from 2 to 10 μm. The size and seta potential of the particles were influenced by the crosslinking level. With decreasing amount of crosslinking agent (either glutaraldehyde or formaldehyde), both particle size and zeta potential were increased. Preparation conditions also had some influence on the particle size. DSC studies revealed that the H2 antagonist drug cimetidine, as well as famotidine was molecularly dispersed inside the microspheres, in the form of a solid solution. The release of model drugs (cimetidine, famotidine and nizatidine) from these microspheres was fast, and accompanied by a burst effect.

Keywords: Chitosan microspheres; Spray drying; Controlled drug delivery; H2-antagonists

1. Introduction

Chitosan is a hydrophilic, biocompatible and biodegradable polysaccharide of low toxicity which in recent years has been used for development of drug delivery systems. For example, it has been exploited for the preparation of microspheres for controlled release systems. Large (> 100 μm) chitosan microspheres (or beads), which were prepared by an emulsion method were used to deliver via different routes the anti-inflammatory drug diclofenac sodium (Acikgoz et al., 1996), the antineoplastic agent mitoxantrone (Jameela and Jayakrishnan, 1995) and other active materials, such as furosemide (Akbuga and Durmaz, 1994) theophylline, griseofulvin and aspirin (Thanoo et al., 1992). For all drugs, a sustained release profile was found. Small chitosan microspheres (< 10 μm) were also developed for site specific delivery of anticancer agents, such as oxantrazole (Hassan et al., 1992) and 5-fluorouracil (Ohya et al., 1993). During recent years, there has been an increasing interest in the use of such chitosan microspheres as mucoadhesive drug delivery systems (Lehr et al., 1992; He et al., 1998), especially for nasal (Illum et al., 1994) and

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peroral delivery of peptide drugs, in order to improve the drug absorption (Lueßen et al., 1996).

Spray drying is a well-known process, which is used to produce dry powders, granules, or agglomerates (Chawla et al., 1994) from drug-excipient solutions and suspensions. Recently, a number of articles have been published describing the preparation of microspheres by such spray drying methods. For example, microspheres composed of the water insoluble polymer polylactic acid (PLA) or poly(lactide coglycolide) (PLGA) were prepared for the delivery of progestone and theophylline (Bodmeier and Chen, 1988), vitamin D3 (Pavenetto et al., 1993) diazepam (Conte et al., 1994a), piroxicam (Wagenaar and Müller, 1994), and for the encapsulation of the water soluble material albumin (Gander et al., 1995) or vaccine antigens (Lee et al., 1997). Water soluble polymers such as proteins have been formulated into microspheres and used as the carrier for the intra-articular delivery of dexamethason (Pavenetto et al., 1994) and nicardipine (Conte et al., 1994b) by this technique. A herbicide (dicamba) formulation of ethyl cellulose microspheres has also been prepared by the spray drying (Tefft and Friend, 1993). The particle size of the microspheres prepared by spray drying method ranged from a few microns to several tens of microns, and had a relatively narrow distribution.

In the present study, chitosan microspheres, with H_2-receptor antagonists, such as cimetidine, famotidine and nizatidine entrapped were prepared by a spray drying method. The microspheres were positively charged in order to enhance the mucoadhesive properties and make these suitable for delivery of drugs via the nasal or gastrointestinal routes of delivery. The physicochemical properties of the microspheres related to preparation parameters, and release characteristics in vitro were also studied.

2. Experimental

2.1. Materials

Chitosan hydrochloride salts (Seacure CL 210, molecular weight: 140–160 kD, Seacure CL 310, molecular weight: 240–270 kD and 280–320 kD, respectively) were obtained from Pronova A/S, Norway. Chitosan as a free base and ethyl cellulose (EC) were purchased from Sigma (Dorset, UK). The following chemicals were obtained from different companies: Glutaraldehyde (50% aqueous solution, Aldrich, Dorset, UK); Formaldehyde (37–41% aqueous solution, BDH); Cimetidine (Aldrich); Famotidine (Merck Sharp & Dohme Research Laboratories, UK); Nizatidine (a gift from Eli Lilly Company, Indianapolis, USA); Phosphate buffered saline tablets (pH 7.4, Sigma). All chemicals, reagents and solvents used were of the highest grade available and used as provided.

2.2. Preparation of chitosan microspheres

The required volume (usually 250 ml) of a 0.1–0.5% aqueous solution of chitosan hydrochloride salt or an aqueous solution containing 1% acetic acid of chitosan free base were prepared. Dissolving chitosan free base in acetic acid produced the chitosan acetate salt. Various amounts of an 1% aqueous solution of formaldehyde or glutaraldehyde (0.5–16 ml) were added as crosslinking agents. Spray drying was co-currently performed using a SD-04 spray drier (Lab Plant, UK), with a standard 0.5 mm nozzle. When the liquid was fed to the nozzle with a peristaltic pump, atomization occurred by the force of the compressed air, disrupting the liquid into small droplets. The droplets, together with hot air, were blown into a chamber where the solvent in the droplets was evaporated and discharged out through an exhaust tube. The dry product was then collected in a collection bottle. In the standard condition, the inlet temperature, spray flow and compressed spray air flow (represented as the volume of the air input) were set at 160°C, 6 ml/min, 10 l/min, respectively. The effects of manufacturing parameters on the characteristics of the resulting microspheres, were studied by setting the pump rate, compressed air flow and inlet temperature at 5, 10, 15 ml/min, 6, 8, 10 l/min, and 140, 160 and 180°C, respectively and choosing nozzles with different size openings (0.5 and 1.0 mm). The chitosan microspheres were crosslinked to the

same level. The drug loaded chitosan microspheres were prepared by dissolving the model drugs (9 or 16.6% w/w of cimetidine, famotidine or nizatidine) in the chitosan solution prior to spray drying. The yield was from 200 mg to 1 g dependent upon the chitosan concentration used.

As a comparison, a water insoluble polymer, ethyl cellulose (EC) was used to prepare microspheres from a 2–4% polymer solution in dichoromethane. The spray drying conditions, inlet temperature and spray flow rate was set at 35–50°C and 6 ml/min, respectively.

2.3. Characterization of chitosan microspheres

The microspheres were sized using a Malvern MasterSizer (model MS 1002), determining the volume mean diameter (VMD) and polydispersity. The zeta potential of the microspheres was measured by laser doppler anemometry (Malvern Zetasizer 4) employing 0.005, 0.0005 and 0.0001 M phosphate buffers at pH 7.0 and 0.001 M acetate buffer at pH 4.0. The morphology of the microspheres was evaluated by a Jeol JSM-35 scanning electron microscope (Jeol, Tokyo, Japan). DSC was performed using a Perkin–Elmer DSC 2 instrument with intracooler. The samples were purged with the atmosphere of nitrogen. The heat flow rate was recorded from 280 to 520 K, at a rate of 10 K/min. Indium (m.p. = 429.8 K) was used as the standard reference material to calibrate the temperature and energy scales of the DSC instrument.

2.4. Determination of drug contents

Spectrophotometric methods were used for determination of drug contents. For the determination of cimetidine, ultraviolet absorption spectra of the sample solutions (2–20 μg/ml) were recorded using a spectrophotometer (UVIKON 860 spectrophotometer, Kontron Instruments, Switzerland) with 1 cm cells from wavelength 300–200 nm. The absorbance differences (ΔA) at 217 nm (maximum) and at 260 nm were calculated. The drug content was found from the absorbance differences read from the standard solutions of the drug (Bavin et al., 1984). For the determination of famotidine and nizatidine contents, the absorbance (A) of sample solutions (4–20 μg/ml) was recorded at 284 nm (famotidine, USP XXII, p559) or at 313 nm (nizatidine) (Wozniak, 1990). The same solvent was used as a reference. The drug contents were calculated form the absorbances read from the standard solutions of the drugs.

2.5. In vitro drug release

The rate of release of the model drugs from the microspheres in phosphate buffered saline (PBS) was determined in a dissolution apparatus (COPLEY ERWEKA DT-6) with the dissolution paddle assembly (USP Apparatus 2 or BP Apparatus II). 30–50 mg of microspheres were suspended in 300 ml of PBS, pH = 7.4 at 37°C, and at 50 rpm agitation rate. Triplicate samples were run.

3. Results and discussion

3.1. Characteristics of chitosan microspheres

Chitosan microspheres without the addition of any crosslinking agent (non-crosslinked) were firstly prepared by a spray drying method. The characteristics of the microspheres are shown in Table 1. The particle size of the chitosan microspheres ranged from 4 to 5 μm. The positive zeta potential of the chitosan microspheres, prepared from chitosan hydrochloride salts was higher than that produced from the free base of chitosan (Chitosan acetate). The characteristics of microspheres prepared form EC are also listed in the same table, as a comparison. EC microspheres were negatively charged. The particle size was about 5 μm.

Non-crosslinked chitosan microspheres can not be kept suspended in water because of swelling and dissolution. In order to prepare stabilized

chitosan microspheres, crosslinking agents (glutaraldehyde and formaldehyde) were generally used to solidify the microspheres (Akbuga and Durmaz, 1994; Jameela and Jayakrishnan, 1995). Stabilized drug free chitosan microspheres, crosslinked with formaldehyde or glutaraldehyde were thus prepared by a spray drying method and the characteristics listed in Table 2.

The chitosan microspheres were spherical, the size (VMD, swelled) ranged from 1.75 to 3.17 μm, and increased with decreasing amount of crosslinking agent. The size distribution of the microspheres was very narrow. The positive zeta potential of the chitosan decreased with an increase in the amount of the crosslinking agent. It should be noted that the zeta potential was strongly dependent on the media in which it was measured. In an aqueous environment, the electrolyte will dissociate into ions. The ions, especially the counterions, which have a charge opposite to that of the particles, will be attracted to the charged particles by an electrostatic (Martin, 1993). According to the DLVO theory and Schultz–Hardy rule, this effect is inversely proportional to the sixth power of the valence of the counterions. The influence of the tri-valent phosphate anions on the zeta potential of the positively charged chitosan microspheres is much greater than that of mono-valent anions, such as acetate anions. Thus, the zeta potential for chitosan microspheres decreased rapidly with increasing phosphate concentration. Therefore, in subsequent studies, dilute phosphate buffer (0.0001 M) was used as a neutral medium (pH 7) to measure zeta potentials, unless specified. In this medium, the positive zeta potential of chitosan microspheres was close to that in pH 4 acetate buffer. The chitosan microspheres, crosslinked by glutaraldehyde showed the same physicochemical characteristics as those crosslinked by formaldehyde. The particle size ranged from 1.5 to 9 μm (Fig. 1).

H_2-antagonists (cimetidine, famotidine and nizatidine) were incorporated into the chitosan microspheres. Such incorporation of a drug may alter the composition of the microspheres, and therefore, could influence their characteristics. The characteristics of chitosan microspheres loaded with cimetidine and crosslinked by glutaraldehyde and formaldehyde are listed in Table 3. It can be seen that, although the cimetidine loaded chitosan microspheres showed the same trend as that of drug free microspheres in that particle size was reduced with an increase in the amount of crosslinking agent, the particle size of the loaded microspheres was larger than that of the drug free microspheres at the same crosslinking level. The zeta potentials of the microspheres were similar or slightly higher than those of drug free microspheres. This could be due to a reaction of the aldehyde group of the crosslinking agent with the amine group of the model drug. If part of the crosslinking agent had reacted with the drug, the amount of the crosslinking agent that reacted with chitosan would be reduced. The lower extent of crosslinking reaction resulted in a larger size of particle and a higher zeta potential.

Table 1
The physiochemical characteristics of non-crosslinked chitosan and EC microspheres

Microsphere type	Size (μm)	Zeta potential (mV)	
		0.001 M pH 4 acetate buffer	0.0001 M pH 7 phosphate buffer
Chitosan hydrochloride	5.58[a]	27.2	24.9
Salt			
Chitosan free base	4.19[a]	14.8	9.7
EC	5.1	−15.5	−5.2

[a] In isopropanol.

Table 2
The physicochemical characteristics of chitosan microspheres crosslinked by different amounts of formaldehyde, prepared by a spray drying method (250 ml of 0.1% chitosan solution)

Amount of formaldehyde (1%) ml	Size (μm)	Zeta potential (mV)			
		0.0001 M pH 4 acetate buffer	pH 7 phosphate buffer		
			0.0001 M	0.0005 M	0.005 M
0.5	3.17	19.6	19.6	12.8	8.9
1	2.32	19.0	17.9	12.0	8.7
2	1.80	17.2	17.1	11.7	8.6
4	1.75	16.8	17.0	10.1	8.1
8	2.02	15.8	15.6	10.0	8.0

3.2. Effects of manufacturing parameters on the characteristics of the microspheres

The results of the influence of preparation parameters on the characteristics of the chitosan microspheres are shown in Table 4. The size of the chitosan microspheres prepared using the standard nozzle (0.5 mm) was 3.63 μm, and became 4.83 μm when a larger nozzle (1.0 mm) was used. The size of the microspheres prepared under the condition of a faster pump rate was large, due to the fact that large droplets were formed during the process. There was an apparent increase in particle size from 3.32 to 3.81 μm when the air flow rate was reduced from 10 to 6 l/min. Since, the production was only performed twice, no statistical evaluation could be done. Liquid was disrupted into smaller droplets at a greater air flow rate. Therefore, microspheres with a smaller particle size were produced at greater air flow rate. The inlet temperature had little influence on particle size. When the temperature changed from 140 to 180°C, particle characteristics were almost the same, and the particle size was only slightly reduced.

Spray drying is a solvent evaporation process. The solvent in the droplets is removed very quickly due to heat energy provided in the spray drier. The thermal efficiency of the spray drying is related to the heat energy input (controlled by inlet temperature and blower) and the amount of heat used in the evaporation process. The optimum spray drying efficiency can be achieved from a balance of the amount of the energy input and the amount of the energy needed, which is related to the amount of the sample input. Since the boiling point of water is 100°C at standard condition, the inlet temperature used should be higher than this. The optimum inlet temperature for the preparation of chitosan microspheres from aqueous solutions of chitosan was found to be 160°C. Once the inlet temperature was set below 140°C, or the pump rate was chosen to be faster than 10 ml/min, the solvent in the droplets could not be fully evaporated. It was observed that some of the liquid droplets were attached inside the wall of the main chamber. The studies of Chawla et al. (1994) are relevant. They prepared spray dried salbutamol sulphate for use in dry powder aerosol formulations from aqueous drug solution. The inlet temperature and pump rate was controlled at 150–180°C and 5–7 ml/min, respectively.

As expected, the manufacturing parameters (nozzle used, pump rate and compressed spray air flow) affected the particle size of the resultant microspheres but did not affect the zeta potential, due to the fact that the microspheres were produced at the same level of crosslinking. Microspheres with different zeta potential could be prepared by changing the crosslinking level, through the quantity of added crosslinking agent. Thus, chitosan microspheres of different size and different zeta potential could be produced by the spray drying method under different manufacturing conditions.

3.3. Effects of the molecular weight and concentration of chitosan on the characteristics of chitosan microspheres

Three chitosan hydrochloride materials with different molecular weights (140–160, 240–270 and 280–320 kD) were used to prepare chitosan microspheres. The viscosities of these materials (1% of chitosan in 1% acetic acid aqueous solution) were 122, 300 and 480 mPa, respectively. The sizes of the drug free and drug loaded microspheres formed were increased with increasing chitosan molecular weight, at the same polymer concentration (Tables 5 and 6). Under the same preparation conditions, the droplets formed from the higher viscosity chitosan solution will be larger in size and result in a larger microspheres being formed. Pavenetto et al. (1993) prepared PLA microspheres by a spray drying method. At the same polymer concentration (1.25%), the particle size was similarly increased with polymer molecular weight.

The influence of the concentration of chitosan in aqueous solution on the particle size and zeta potential were tested with five different concentrations (0.1–0.5%) with the same ratio of chitosan and glutaraldehyde. As expected, with increasing concentration of chitosan, the size of the microspheres so prepared was increased (Table 5). This is due to the greater amount of chitosan contained in the same volume of a liquid droplet as the concentration of chitosan is increased. These results are agreement with those of Pavenetto et al. (1994), Wagenaar and Müller (1994). They prepared PLA microspheres or albumin microspheres by a spray drying method. Increasing the concentration of the polymer resulted in an increase in particle size.

3.4. Morphology

The morphology of the chitosan microspheres prepared by the spray drying method was examined by light microscopy as well as by scanning

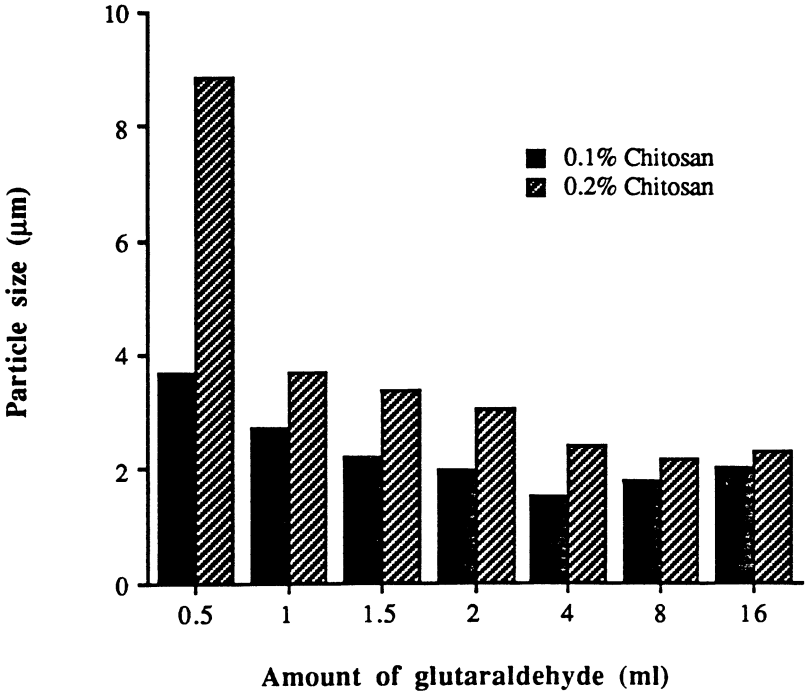


Fig. 1. The influence of the amount of glutaraldehyde on the particle size of the chitosan microspheres prepared from 0.1 and 0.2% chitosan aqueous solution by a spray drying method.

Table 3
Characteristics of cimetidine loaded chitosan microspheres prepared by the spray drying method

Concentration of chitosan (%)	Crosslinking agent (ml)	Drug contents (%)			Size (μm)	Zeta potential (mV)	
		Added	Found	Efficiency		pH4	pH7
Glutaraldehyde (4%)							
0.1	1	16.6	15.7	94.7	7.85	17.4	15.0
0.1	2	16.6	13.0	78.3	4.03	15.8	13.6
0.1	4	16.6	11.8	71.0	1.67	15.5	9.1
0.2	1	16.6	16.2	98	4.89	17.3	15.2
0.2	2	16.6	15.6	94	4.62	16.2	14.5
0.2	4	16.6	14.2	85.7	2.93	14.6	13.4
0.2	1	9	8.04	88.5	5.19	17.4	14.4
0.2	2	9	7.11	78.3	4.58	15.5	14.5
0.2	4	9	6.65	73.2	2.22	14.9	13.1
Formaldehyde (1%)							
0.2	2	16.6	16.2	97.6	7.91	22.5	17.4
0.2	4	16.6	15.3	92.4	3.89	18.8	15.3

electron microscopy (SEM). The sphericity of the microspheres was good, even for the non-crosslinked microspheres. SEM measurements of chitosan microspheres (non-crosslinked and crosslinked with glutaraldehyde and formaldehyde) are shown in Figs. 2 and 3. The non-crosslinked microspheres prepared from a low viscosity grade of chitosan had a depressed surface morphology (slightly wrinkled) but those prepared from a high viscosity grade of chitosan had a smooth surface. For the stabilized microspheres, a smooth but distorted surface of the microspheres was observed, especially for the microspheres crosslinked with a lower quantity of the crosslinking agent. A similar morphology was observed for the microspheres crosslinked with formaldehyde.

The drug loaded microspheres were spheroid-shaped, with a smooth surface, especially for famotidine loaded chitosan microspheres (Fig. 4).

3.5. DSC analysis

The physical state of the drug inside the chitosan microspheres was assessed by thermal analysis. DSC thermograms of cimetidine and chitosan materials, chitosan–cimetidine physical mixture with the same ratio (10:2) as the drug loaded microspheres (16.6% w/w of cimetidine), drug free chitosan microspheres and cimetidine loaded chitosan microspheres are shown in Fig. 5(A). Under the experimental conditions, no DSC peak was observed for the chitosan material and drug free chitosan microspheres. For the cime-

Table 4
The influence of preparation parameters on the particle size and zeta potential of chitosan microspheres prepared by a spray drying method

Preparation parameters	Level	Size (μm)	Zeta potential (mV)
Nozzle (mm)	0.5	3.63	18.8
	1.0	4.83	18.9
Pump rate (ml/min)	5	3.00	18.8
	10	3.56	17.5
	15	4.10	17.0
Compressed air flow rate (l/min)	6	3.81	17.5
	8	3.75	16.6
	10	3.32	17.0
Inlet temperature (°C)	140	3.59	18.0
	160	3.00	18.3
	180	3.27	17.5

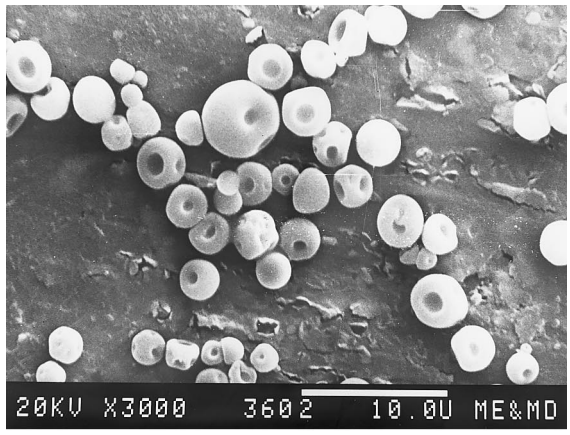
Table 5
The influence of chitosan molecular weight and the concentration of chitosan on the particle size and zeta potential of chitosan microspheres prepared by a spray drying method

Level	Size (μm)	Zeta Potential (mV)
Molecular weight of chitosan (kDa)		
140–160	3.04	18.5
240–170	4.16	16.3
280–320	5.03	16.3
Concentration of chitosan (140–160 kDa)		
0.1	2.69	17.1
0.2	3.00	16.6
0.3	3.27	16.5
0.4	3.95	17.7
0.5	4.22	16.0

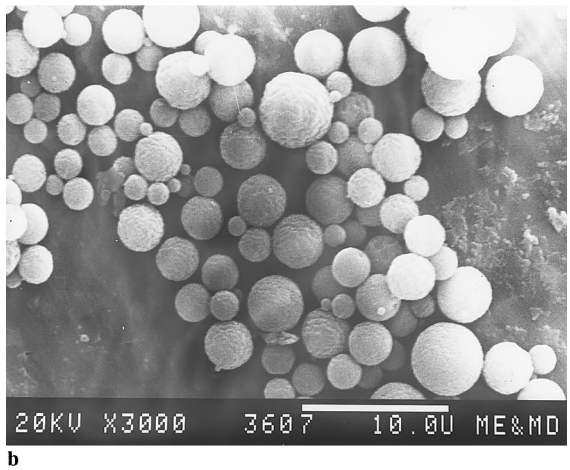
tidine–chitosan physical mixture, an endothermic peak of melting of cimetidine at 417 K (a) was broadened and shifted to about 380 K. This suggests that a crystalline form of cimetidine existed in the physical mixture of cimetidine–chitosan. The peak disappeared for the drug loaded chitosan microspheres, which indicated that the drug was molecularly dispersed inside of the matrix of chitosan as a solid solution. DSC thermograms of famotidine loaded chitosan microspheres are

Table 6
Drug loaded chitosan microspheres composed of different molecular weight chitosans prepared by a spray drying method

Chitosan	Drug loaded	Size (μm)	Zeta potential (mV)
CL 210, 122 mPa	Cimetidine	4.04	16.5
	Famotidine	4.18	16.2
	Nizatidine	4.19	16.4
CL 210, 300 mPa	Cimetidine	5.16	18.7
	Famotidine	5.00	18.4
	Nizatidine	5.30	16.8
CL210, 480 mPa	Cimetidine	5.90	12.9
	Famotidine	5.10	14.3
	Nizatidine	5.16	15.2



a



b

Fig. 2. Scanning electron microscopy of non-crosslinked chitosan microspheres: (a) chitosan hydrochloride salt; (b) chitosan free base, prepared by a spray drying method.

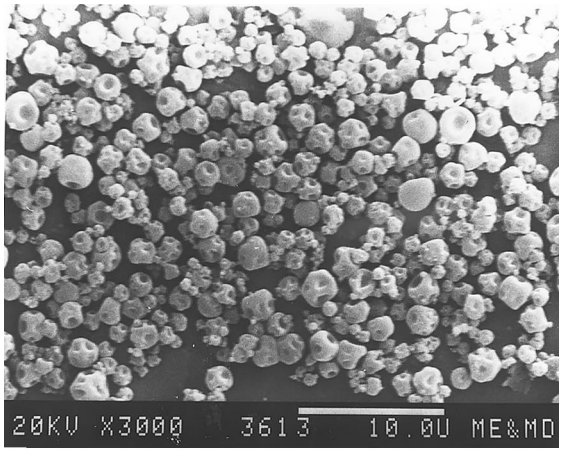
shown in Fig. 5(B). Similar to cimetidine, famotidine when dispersed inside the chitosan matrix, existed molecularly, as a solid solution.

Fig. 6 shows the DSC curves for the cimetidine material, physical mixtures of chitosan–cimetidine with different ratio (10:2; 50:50 and 30:70) and the cimetidine loaded (50 and 70% w/w) chitosan microspheres. For the physical mixtures, with increasing cimetidine content, the broadened endothermic peaks of the drug were closer to that of pure drug. This result indicates that the amount of crystalline cimetidine was increased when the ratio of the drug increased for the drug–polymer physical mixture. For the drug

loaded chitosan microspheres, however, no peak was observed, even when cimetidine content reached 70% w/w. The result indicated that the drug was dispersed inside the matrix of chitosan as a solid solution.

3.6. In vitro drug release

Non-crosslinked chitosan microspheres did not maintain the form of spheres in water, especially in an acidic environment. The microspheres swelled and dissolved. Non-crosslinked chitosan



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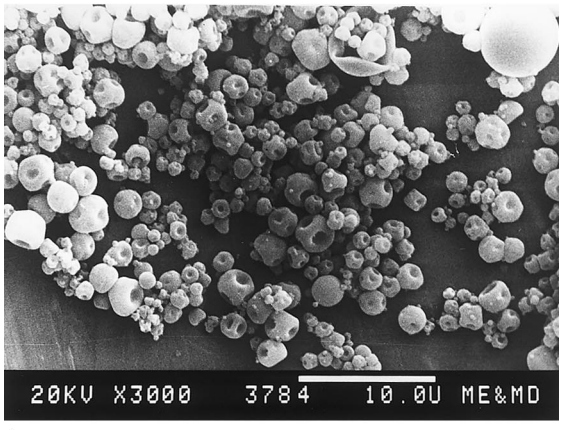
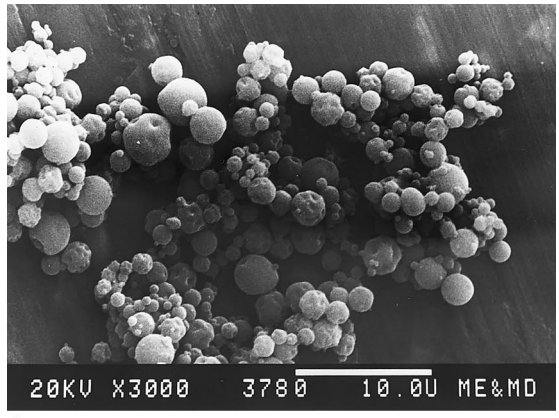
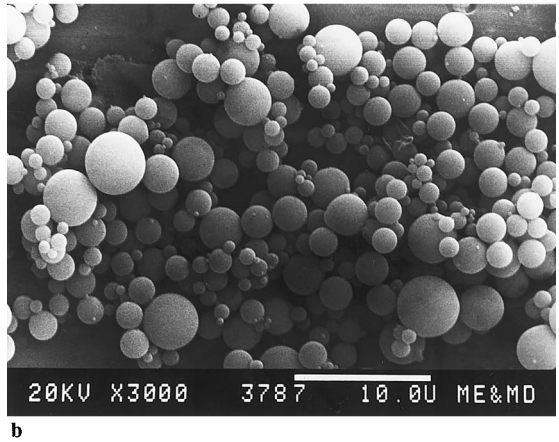


b

Fig. 3. Scanning electron microscopy of drug free chitosan microspheres prepared from 0.2% aqueous solutions for chitosan hydrochloride salt (M_w 140–160 kDa) by a spray drying method, crosslinked by glutaraldehyde (a), and formaldehyde (b).



a



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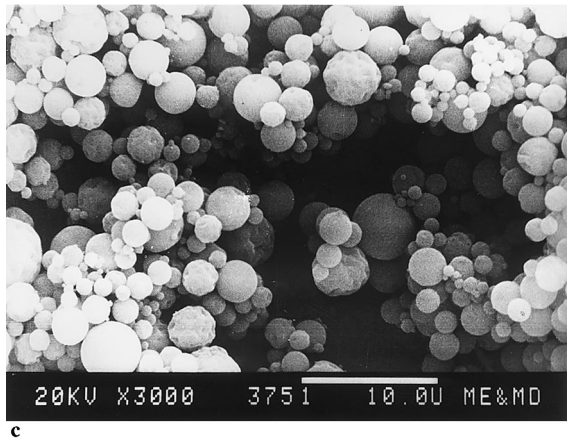

c

Fig. 4. Scanning electron microscopy of drug loaded (a) cimetidine; (b) famotidine; (c) nizatidine, chitosan microspheres prepared form 0.2% aqueous solutions of chitosan hydrochloride salt (M_w 140–160 kDa) by a spray drying method.

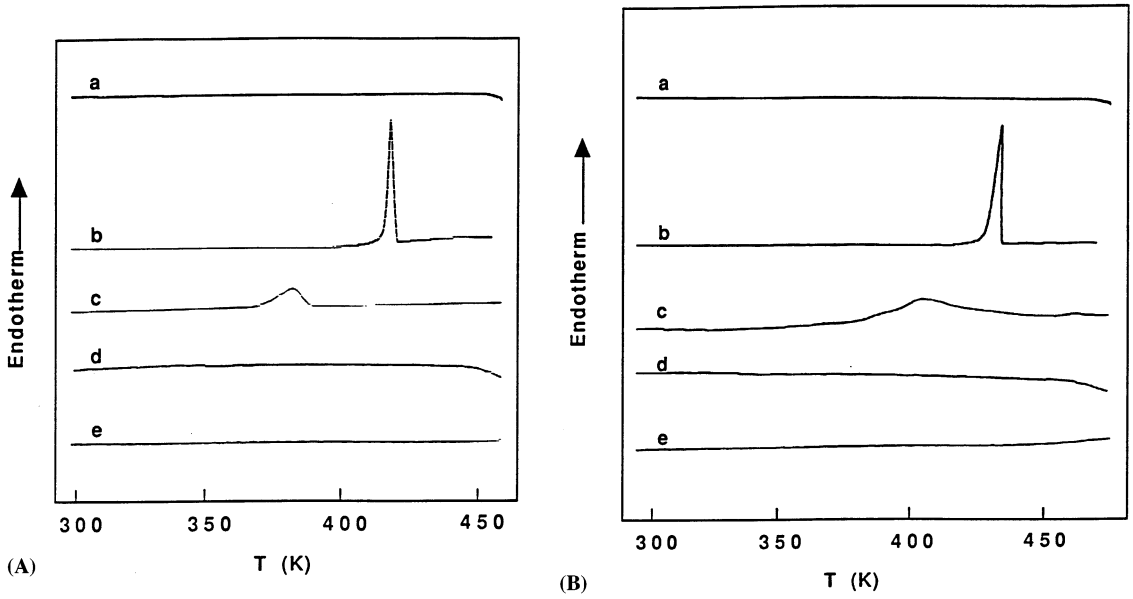

Fig. 5. DSC thermograms of chitosan (a) and model drug (cimetidine (A); famotidine (B)) materials (b), chitosan–drug physical mixture (c) 10:2; drug free chitosan microspheres (d); and the drug loaded chitosan microspheres, 10:2 (e).

microspheres were therefore unsuitable for the purpose of microsphere sustained delivery systems (Genta et al., 1995).

In order to evaluate the loading characteristics, chitosan microspheres, containing two different levels of cimetidine (9 and 16.6% w/w), crosslinked by glutaraldehyde or formaldehyde were prepared. The efficiency of the drug incorporation was high (Table 3). However, loading was decreased with an increasing amount of the crosslinking agent. This could be due to the increased binding of the amine groups of the drug with the added glutaraldehyde or formaldehyde, which complex would either be incorporated into the matrix of the microspheres during the spray drying process or be spray dried separately to the microspheres. The DSC results described above, points to the latter explanation.

The release of cimetidine from chitosan microspheres (theoretical drug content 16.6% w/w) prepared from a 0.2% w/w chitosan concentration is shown in Fig. 7. There was a 'burst effect' during the first stage of dissolution, and most of the drug was released in a few minutes. It was noticed that the amount of drug released was reduced when a greater amount of glutaraldehyde was used. Similar release profiles of the drug were observed for the chitosan microspheres loaded with a different

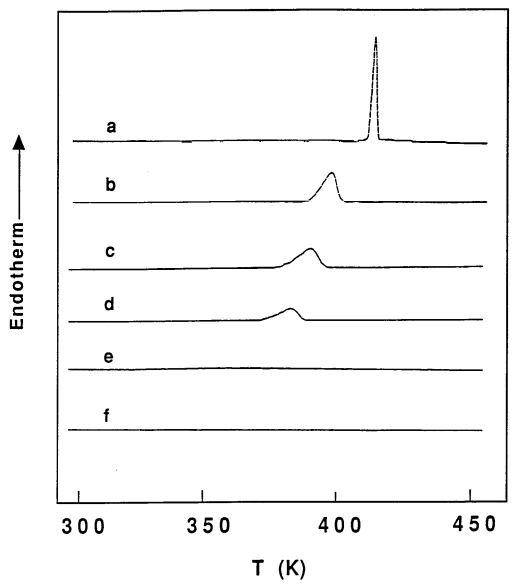

Fig. 6. DSC thermograms of cimetidine material (a) chitosan–cimetidine (b) 30:70; (c) 50:50; (d) 10:2, physical mixture and cimetidine loaded (e) 70%; (f) 50% chitosan microspheres.

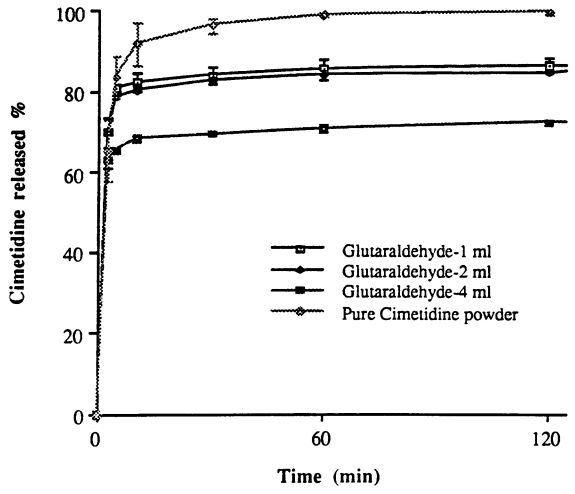

Fig. 7. In vitro release test for the cimetidine loaded chitosan microspheres prepared by a spray drying method from a 0.2% aqueous solution of chitosan hydrochloride salt (M_w 140–160 kDa).

amount (9% w/w) of the drug, or prepared form different concentration of chitosan (0.1% w/w). The rate of release of the drug from chitosan microspheres crosslinked by formaldehyde was as fast as that for those crosslinked by glutaraldehyde; especially for the microspheres crosslinked at a low level (Fig. 8). The theoretical amount of

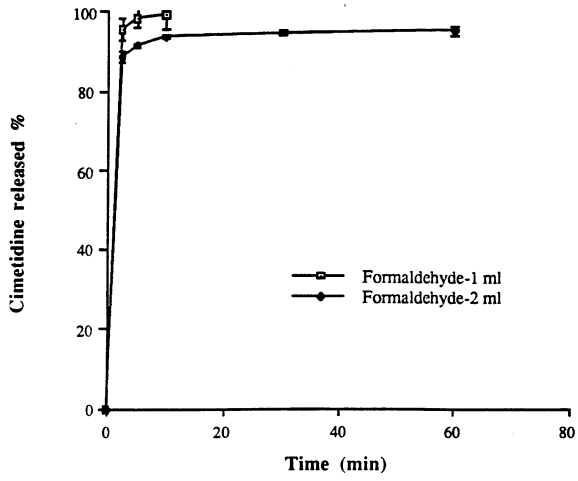

Fig. 8. In vitro release test for cimetidine loaded chitosan microspheres crosslinked by formaldehyde prepared by a spray drying method from a 0.2% aqueous solution of chitosan hydrochloride salt (M_w 140–160 kDa).

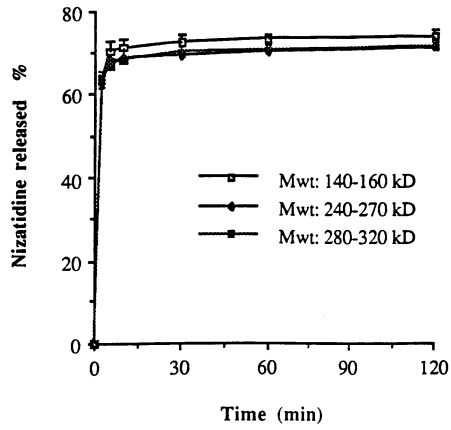

Fig. 9. Influence of the chitosan molecular weight on the release rate of nizatidine from the drug loaded chitosan microspheres prepared by a spray drying method.

the drug was released within a few minutes.

The effect (if any) of chitosan molecular weight on the release rate was small (Fig. 9). The release of nizatidine from the drug loaded chitosan microspheres prepared with different molecular weights of chitosan was almost the same.

The particle size, the properties of the matrix and solubility of drug in the dissolution medium would be expected to influence the rate of release of the drug from the microspheres. The particle sizes of various polymer microspheres, prepared by similar spray drying methods were found to be relatively small, usually less than 10 μm in size and had a porous appearance and a convoluted surface (Tefft and Friend, 1993). When a water soluble (or even slightly water soluble) drug was incorporated into such microspheres, the release rate of the drug would be expected to be rapid, and accompanied by a burst effect (Bodmeier and Chen, 1988; Giunchedi et al., 1994). For example, the drug theophylline was released immediately from Eudragit RS microparticles prepared by a spray drying method. However, the release rate could be retarded when the microspheres were compressed into tablets (Palmieri et al., 1994).

Drug release form microspheres composed of a hydrophilic polymer, such as albumin is still fast. Usually there is an initial fast release (Burst effect). This characteristic of fast release not only occurs with water soluble drugs (Conte et al.,

1994a), but also for some less water-soluble drugs, such as dexamethasone (Pavenetto et al., 1994).

It is believed that only if both the matrix material and drug are lipophilic in nature, would it be possible to prepare drug loaded microspheres with a slow release character, using the spray drying method. The release of vitamin D3 from PLA microspheres prepared by a spray drying method lasted several hours (Pavenetto et al., 1993).

The fast release of cimetidine and other H_2-antagonists from chitosan microspheres prepared by the spray drying method could be due to several causes. Firstly, both the matrix (chitosan) and drug (cimetidine) are hydrophilic in nature, and both, therefore have a high affinity for water. The drugs have been shown to be molecularly dispersed in the polymer matrix (DSC results). This could cause a fast release of the drug. Lastly, the microspheres formed were small in size and porous in nature.

4. Conclusions

Non-crosslinked chitosan microspheres with a size of 4–5 μm, crosslinked chitosan microspheres with a size of 2–10 μm and a positive zeta potential could be prepared by a conventional spray drying method. The microspheres had a good sphericity, a uniform distribution of particle size and were positively charged. A smooth, but distorted surface was observed for drug free microspheres, especially for the microspheres crosslinked with a lower quantity of crosslinking agent. The drug loaded microspheres crosslinked with a lower quantity of crosslinking agent. The drug loaded microspheres were still spheroid in shape, and had a smooth surface. The particle size and the positive zeta potential were influenced by the quantity of crosslinking agent employed. With a decrease in the amount of crosslinking agent (either glutaraldehyde or formaldehyde), both particle size and zeta potential increased.

The preparation parameters (the size of nozzle, feeding pump rate, inlet temperature, and compressed air flow rate) influenced the particle size of the microspheres. Large particles were formed at a faster feeding pump rate and at a larger size of nozzle. Smaller particles were formed at a greater volume of air input. Comparatively, there was a relatively little influence of inlet temperature on the particle size for the range 140–180°C.

The particle size of chitosan microspheres, prepared from a higher concentrations of chitosan and from higher molecular weights of chitosan was large, compared with microspheres prepared from lower chitosan concentrations and molecular weights.

DSC studies revealed that cimetidine, as well as famotidine, were molecularly dispersed inside of the microspheres, in the form of a solid solution. Even when the percentage of drug (cimetidine) in the microspheres reached 70% w/w, no crystalline drug was detected in the microspheres.

The release of model drugs (cimetidine, famotidine or nizatidine) from these microspheres was fast, and accompanied by a large burst effect. The amount of the drug released was slightly influenced by the amount of crosslinking agent, and the solubility of the drug. When the crosslinking density increased or the solubility of the drug decreased, the amount of the drug released was slightly reduced.

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