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Application of Modified Bead Cellulose as a Carrier of Active Ingredients

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Summary: A new kind of very fine disperse porous bead cellulose is modified by chemical treatments to 2,3-dialdehyde cellulose, carboxymethyl and dihydrogenphosphate cellulose. Model drugs like benzocaine and prazosin are coupled to these modified bead celluloses by covalent and ionic linkage, respectively and compressed to tablets. Compression mixtures with low loaded benzocaine conjugates release with medium rate. Linkage of prazosin cation to the anionic derivatives of bead cellulose leads to fast release of poor soluble ionic drug.

Keywords: bead cellulose; drug delivery systems

Introduction

Bead cellulose is a special kind of regenerated cellulose. The characteristics of bead cellulose are: spherical particles with defined particle size and particle size distribution, highly porous structure, large specific surface area, hydrophilic properties, chemical reactivity, and high mechanical strength.

Bead cellulose has many different uses, mainly in supports of solid phase synthesis, supports of liquid chromatography, ion exchange processes, removal of heavy metals, and immobilization of enzymes.[1–6] Bead cellulose prepared by the acetate method is used for blood purification after chemical modification and coupling with specific substances.[7]

In former times 2,3-dialdehyde bead cellulose, dihydrogenphosphate bead cellulose and carboxymethyl bead cellulose were prepared from particle sizes in the range of 100–400 μm (swollen state).[8,9]

In the present study the small sized spherical high porous bead cellulose (1–10 μm) is investigated as potential drug carrier and part of multi-particulate drug delivery systems with controlled release. Conjugates with covalent as well as ionic linked model drugs are prepared to receive multi-particulate systems with high release rate useful for poorly soluble drugs. These are basic examinations for the use of the small beads as a drug delivery system.

Experimental Part

Materials

2.5-Cellulose acetate (FM3, Courtaulds), methyl cellulose (Fluka), sodium acetate trihydrate (Fluka), surfactant Triton X-100 (Fluka), sodium hydroxide (Riedel), phosphorous(V)oxychloride (Riedel), monochloroacetic acid (Riedel), perchloric acid (Fluka), sodium periodate (Fluka), ethylene glycol (Fluka), potassium phosphate dibasic (Merck), disodium hydrogen phosphate (Roth), Cellactose™80 (Meggle), Emcompress™ (J. Rettenmaier & Söhne), Primojel™ (Campina), Aerosil™100 (Merck), zinc stearate (Merck), benzocaine (Merck), prazosin hydrochloride (Ratiopharm), ethyl acetate (Riedel), diethyl ether (Fluka), methanol (Fluka), deionized water.

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Bead Cellulose

Cellulose beads are prepared from cellulose acetate by a well known method.[10,11]

2,3-Dialdehyde Bead Cellulose

10 g water wet never dried bead cellulose (about 3.3 g dry substance) are treated with a) 10 ml or b) 50 ml of 0.5 mol/l $NaIO_4$ solution. The flask is covered with an aluminum foil to exclude light. The mixture is shaken for 2 h at 40 °C. Thereafter 10 ml ethylene glycol are added and shaken for another 1 h. The oxidized bead cellulose is washed with water five times in a centrifuge. The content of dialdehyde groups is determined to a) 1315 μmol/g and b) 2069 μmol/g according to a described method.[12]

Benzocaine 2,3-Dialdehyde Bead Cellulose Conjugates

100 g water wet never dried 2,3-dialdehyde bead cellulose (about 3.3 g dry substance) are treated with 36 g benzocaine in 1.8 l methanol. The suspension is shaken 4 h and stood 20 h in the dark. Finally the product is washed with methanol five times in a centrifuge. The content of loaded benzocaine is determined and calculated from elemental analysis.

Carboxymethyl Bead Cellulose

According to the literature[9] 20 g water wet never dried bead cellulose (about 6.6 g dry substance) are suspended and stirred in 10 ml NaOH solution (4.6 g NaOH) for 30 min in an ice bath. 7.0 ml monochloroacetic acid solution (4.2 g MCA) are added under agitation and cooling. The suspension is heated to 70 °C, stirred over 30 min, cooled to room temperature and neutralized with acetic acid (pH 7). The suspension is transferred into a G3 frit, filtered off and washed with water. pH value of the last rinse should be equal to the pH of the water and absorption at 330 nm should not exceed 0.02 to be sure about removal of impurities. The degree of substitution is determined by titration according to an American Standard Test Method.[13]

Dihydrogenphosphate Bead Cellulose

As described in literature[9] 20 g water wet bead cellulose (about 6.6 g dry substance) are suspended in 10 ml NaOH solution (4.6 g NaOH) and agitated for 30 min in an ice bath. Ice-cooled mixtures of $POCl_3$/diethyl ether (1:1) of 5.5 and 22 ml, respectively are added under agitation. The reaction is continued for 30 min. The suspension is transferred onto a G3 frit, further treatment see above. Phosphorus content is investigated by elemental analysis.

Prozosin Bead Cellulose Conjugates

Prazosin is coupled to carboxymethyl bead cellulose and dihydrogenphosphate bead cellulose under modification of an earlier described batch method.[14] 25.0 g water wet ionic bead cellulose derivative (about 8 g dry substance) are dispersed in 200 ml aqueous prazosin hydrochloride solution (0.1% w/w). The suspension is agitated over 4 h at room temperature under light protection, stored over 16 h and filtered off (G3 frit) into a flask to remove excess drug solution. Prazosin content of the filtrate is analyzed. For water exchange versus ethanol and careful drying the filter cake consisting of prazosin bead cellulose derivative conjugate is two times re-suspended in ethanol, filtered off and dried in a rotary evaporator. Finally, the dry product of drug loaded bead cellulose derivatives is weighed for yield and linked prazosin amount calculation.

Preparation of Tablets

For compression mixture preparation of tablets with benzocaine model drugs the components Cellactose™, Emcompress™, Primojel™, and Aerosil™100 are weighed and mixed by hand in a mortar (20 g lots), sieved (355 μm), zinc stearate is added, mixed and finally sieved (180 μm). Tablets with punch diameter 13 mm, thickness 2.8-2.9 mm, weight 500 mg, both side plane and facet; one side notch, compression force 11 kN, low rotation speed are compressed with a rotary tablet press.

In the case of prazosin hydrochloride powder as a model drug 500 g compression

mixture is mixed in a cube mixer for 10 min, 200 rpm, completely sieved (355 μm), mixed with zinc stearate in the cube mixer for 5 min and sieved (355 μm) again. Tablets with punch diameter 7 mm, value of height 2 scales, thickness 3.7–4.0 mm, weight 200 mg, both side plane and facet; one side notch, compression force 11 kN, low rotation speed are compressed with a rotary tablet press.

Methods and Instruments

Investigation of Loading Degree

The examination of the loading degree of benzocaine is carried out by elemental analysis. The amount of bounded benzocaine is calculated from the nitrogen content.

The concentration of prazosin in the filtrate of the bead cellulose suspension in prazosin hydrochloride solution is analyzed by UV spectroscopy at wave length 330 nm. The difference of prazosin amount in the original solution (200 mg) and the filtrate gives the amount linked to bead cellulose derivative.

In vitro Benzocaine and Prazosine Release

In vitro drug release is performed with a dissolution tester with 6 flasks, 1.0 l phosphate buffer solution (potassium hydrogen phosphate and disodium hydrogen phosphate, both 1/15 M) pH 6.8 at 37 °C and paddle speed 50 r.p.m. 10.0 ml aliquots are withdrawn after defined time intervals, the volume is even compensated by 10.0 ml buffer solution. Specimens are filtered by a membrane filter (pore diameter 0.2 μm). Drug analysis is performed with a spectrophotometer, 1 cm quartz cell and phosphate buffer solution as blind.

The calibration equation is determined with dilutions of 0.5 up to 10.0 mg/l benzocaine and 0.5 up to 20 mg/l prazosin hydrochloride. Conjugate amounts containing 5,0 mg benzocaine and 10,0 mg prazosin hydrochloride are weighed for drug release investigation, the tablet lots also contained doses of 5,0 mg and 10.0 mg, respectively. Wavelengths are 284 nm (benzocaine) and 330 nm (prazosin hydrochloride).[15]

Characterization of Products

Particle size distribution of bead cellulose lots is investigated by Laser particle counter, Beckman Coulter®. For scanning electron microscopy the dried bead cellulose specimen are coated with gold layer in the plasma phase, K550, Emitech Ltd. and investigated under 300-10000 fold magnification with a scanning electron microscope XL 30, Philips and CDU LEAP® detector.

Tablets are characterized according to Ph.Eur. 2005 by weight uniformity, hardness, disintegration time, friability with drum diameter 30 cm.

Analytical balance LA 230S, Sartorius GmbH, Goettingen; Diode-array Spectrophotometer Spekol 1200, Analytik Jena AG, Jena; Disintegration Tester PTZ, Pharmatest GmbH, Hainburg; Dissolution Tester PTW II, Pharmatest GmbH, Hainburg; Elemental Analyzer EA 1110, Carlo Erba, Milan; Friability tester TA 10, drum diameter 30 cm, Erweka GmbH, Heusenstamm; Laboratory cube mixer KB 15, Erweka GmbH, Heusenstamm; Membrane filter Chromafil PET-20/25, pore diameter 0.2 μm, Macherey-Nagel GmbH, Düren; Sieve shaker AS 200 basic, Retsch GmbH, Haan; Standard motor drive AR 402, Erweka GmbH, Heusenstamm; Tablet Hardness Tester PTB-411, Pharmatest GmbH, Hainburg; Tablet press Universal Perfecta II, FETTE GmbH, Schwarzenbek.

Results and Discussion

Preparation of Cellulose Beads

Cellulose beads are prepared from cellulose acetate by an emulsifying process. The particle size lies in the range of 1–20 μm with a narrow distribution (Figure 1). The surface structure is adjusted to be textured and porous (Figure 2).

Preparation of Covalently Linked Bead Cellulose Conjugates

For the preparation of covalent linked bead cellulose conjugates never dried water wet beads are oxidized with sodium periodate at first to obtain dialdehyde bead cellulose

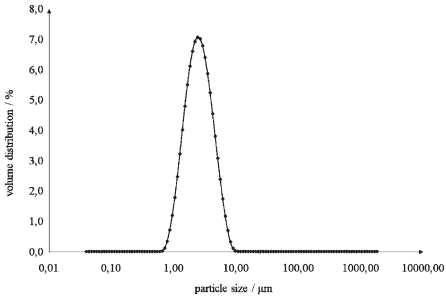


Figure 1.
Particle size distribution of bead cellulose.

(Scheme 1, equ. 1). The maximum reached dialdehyde group content is 2.17 mmol/g dried product. Benzocaine as a model drug is bound to the aldehyde groups in methanol solution under formation of an azomethine linkage (Scheme 1, equ. 2). Loading capacity is in the range of 0.16 to 1.67 mmol/g in dependence of the degree of oxidation and the concentration of benzocaine in the reaction.

Preparation of Ionic Linked Bead Cellulose Conjugates

Two different methods are used for the preparation of anionic charged bead cellulose. Carboxymethyl and dihydrogenphosphate cellulose are synthesized by the reaction of bead cellulose with monochloroacetic acid and phosphorous oxychloride respectively. For the carboxymethylation the bead cellulose is swollen in NaOH

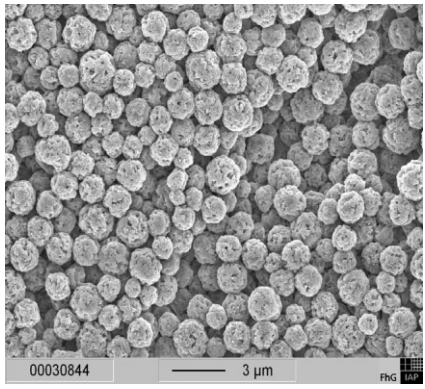
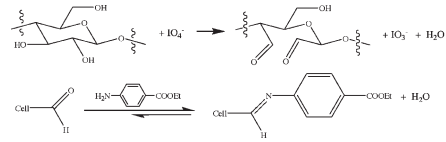


Figure 2.
Morphology of bead cellulose.



Scheme 1.
Reaction of cellulose with periodate to 2,3-dialdehyde cellulose and benzocaine coupling by azomethine linkage.

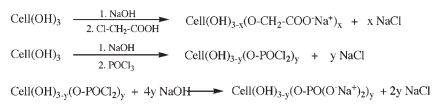
solution as the first step. The second step involves the reaction with monochloroacetic acid. At least, the product is neutralized with acetic acid and washed with water (Scheme 2, equ. 3). The obtained DS value for further coupling with prazosin is about 0.1. For the preparation of dihydrogenphosphate bead cellulose the first step is also the swelling and activation in aqueous NaOH solution. In nonprotic solvent (ether) sodium cellulose reacts with phosphorous oxychloride to cellulose dichlorophosphate which is hydrolyzed with NaOH to dihydrogenphosphate bead cellulose (Scheme 2, equ. 4 and 5). The obtained DS values are in the range of 0.05 and 0.07.

Prazosin is coupled to these derivatives in a batch process with water wet anionic bead cellulose and aqueous prazosin hydrochloride solution (0.1% w/w) by an ionic linkage (Scheme 3).

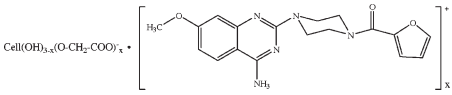
Model Drug Release

Powder like bead cellulose conjugates are mixed with suitable excipient and compressed to tablets. Tablet compositions and parameters are given in Table 1 comply with the demands of Ph.Eur. 2005.[16]

The release of the drugs in phosphate buffer solution is shown in Figure 3 and Figure 4. In the case of benzocaine compression mixtures (CM_{low}) and tablets



Scheme 2.
Reaction of cellulose with chloroacetic acid and with phosphorous oxychloride.



Scheme 3.
Prazosin cation linkage to carboxymethyl bead cellulose.

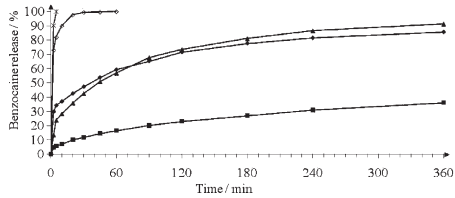


Figure 3.
Benzocaine release in phosphate buffer solution; -×- = benzocaine compression mixture (PM), -◇- = benzocaine tablet (TB3), -▲- = conjugate compression mixture low loaded (CM_{low}), -◆- = conjugate tablet low loaded (TB1), -■- = conjugate gh loaded (TB2).

with low loaded conjugates (TB1) release occurs with medium rate (release 58% after 1 hour) due to the processes of wetting and swelling of the conjugate, hydrolysis of azomethine linkage and diffusion of molecules. With a high loaded conjugate tablet (TB2) 100% release is not achieved during 24 h resulting from bead agglomeration (release 22% after 1 hour). Vice versa, dissolution is complete after 5 min and 30 min from a benzocaine powder compression mixture (PM) and the corresponding tablet (TB3), respectively. In the case of non-linked benzocaine, high dispersion, good wetting and solubilizing influence of the excipients lead to very fast dissolution rate of the amount of 5 mg substance in a volume of 1 l buffer solution (solubility of benzocaine 400 mg/l in water [Ph.Eur. 2005]). On the other hand, the covalent azomethine linkage between benzocaine and dialdehyde bead cellulose leads to retarded release of the drug in buffer solution pH 6.8, and with increased loading degree the release is more and more retarded.

Prazosin is released from dihydrogenphosphate bead cellulose in phosphate buffer solution pH 6.8 with very high rate (Figure 4) by ionic exchange reaction of prazosin cation versus sodium cation. The release can be decelerating by using ethanol-dried carboxymethyl bead cellulose (TP2). The influence of the drying method is shown in the case of water-dried carboxymethyl bead cellulose. The release is retarded and incomplete due to sintered product and slow particle disintegration and swelling.

In contrast to benzocaine, prazosin hydrochloride is dissolved with slow rate due to low solubility in buffer solution pH 6.8 (25 mg/l[16]), after 1 hour only 22% are dissolved. Contrary, release from tablets with non-linked prazosin hydrochloride

Table 1.
Composition and parameters of tablets.

	TB1	TB2	TB3	TP1	TP2	T1
model drug [weight %]	16.5[a)]	3.7[b)]	1[c)]	5[d)]	38.9[e)]	–[f)]
Cellactose [%]	40.9	47.2	48.5	46.5	30.6	49
Emcompress [%]	33.4	38.5	39.6	37.5	19.5	40
Primojel [%]	8.4	9.6	9.9	10	10	10
Aerosil 100 [%]	0.4	0.5	0.5	0.5	0.5	0.5
Zink stearate [%]	0.4	0.5	0.5	0.5	0.5	0.5
weight conformity [g]	485.9	495.2	484.0	197.3	192.3	198.6
hardness [N]	41.3	64.8	48.2	81.2	101.6	29.1
disintegration time [s]	73	27	34	25	80	20
friability [%]	0.10	0.60	0.26	0.69	0.46	0.29

[a)]Low loaded benzocaine bead cellulose;
[b)]High loaded benzocaine bead cellulose;
[c)]Benzocaine;
[d)]Prazosin powder;
[e)]Prazosin loaded carboxymethyl bead cellulose;
[f)]Placebo.

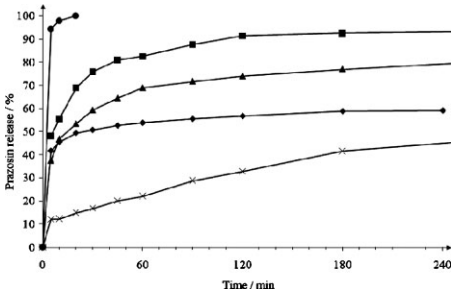


Figure 4.
Prazosin release in buffer solution; -●- = prazosin dihydrogenphosphate bead cellulose, -■- = prazosin powder tablet (TP1), -▲- = ethanol-dried prazosin carboxymethyl bead cellulose tablet (TP2), -◆- = water-dried prazosin carboxymethyl bead cellulose tablet, -×- = prazosin hydrochloride.

powder (TP1) occurs with high rate (more than 80% after 1 hour).

Conclusion

Bead cellulose is modified by chemical treatment to 2,3-dialdehyde cellulose, carboxymethyl and dihydrogenphosphate cellulose. Model drugs like benzocaine and prazosin are coupled to bead cellulose derivatives by covalent and ionic linkage, respectively and compressed to tablets. The release of these drugs is determined. Benzocaine 2,3-dialdehyde bead cellulose conjugates are received as spherical granulate-like products with sufficient flow properties and therefore excellent conditions for further manufacturing. Compression mixtures with low loaded benzocaine conjugates release with medium rate. Retarded benzocaine release of 75% after 2 h is achieved from conjugates and tablets, and in dependence of oxidation, drug loading degree and drying method the release rate is varied in a certain range. Linkage of prazosin cation to the anionic derivatives of bead cellulose leads to fast release of poor soluble ionic drug, Release rate of prazosin was high due to high dispersion degree in the conjugate and fast ion exchange reaction compared to very slow dissolution of pure substance. Therefore, ionic linkage to bead cellulose carriers may be used to increase release rate of poor soluble drugs. On the other hand, retardation occurs with sintered conjugates in dependence of drying regime (e.g. drying from water) so that release rate may be varied in a limited range for controlled release.

[1] B. Wolf, W. Horsch, *Pharmazie* **1991**, *46*, 392.
[2] F. Loth, C. Fanter, *Das Papier* **1993**, *12*, 703.
[3] W. de Oliveira, W. Glasser, *J. Appl. Polym. Sci.* **1996**, *60*, 63.
[4] Xuejun. Guoa, Yonghua. Duc, Fuhua. Chena, Hung.-Suck. Parkb, Yaning. Xie, *J. Colloid Interface Sci.* **2007**, *314*, 427.
[5] D. Zhoua, L. Zhanga, S. Guo, *Water Res.* **2005**, *39*, 3755.
[6] C. Fanter, *Das Papier* **2004**, 4 Zellcheming, Wiesbaden, 16.-19.06. 2003; CD-Beilage.
[7] V. Weber, I. Linsberger, M. Ettenauer, F. Loth, M. Höyhtyä, D. Falkenhagen, *Biomacromolecules* **2005**, *6*, 1864.
[8] B. Wolf, I. Finke, *Pharmazie* **1992**, *47*, 121.
[9] B. Wolf, I. Finke, *Pharmazie* **1991**, *46*, 861.
[10] W. Wagenknecht, C. Fanter, F. Loth, EP 0750007, (**2000**).
[11] S. Fischer, K. Thümmler, B. Volkert, K. Hettrich, I. Schmidt, K. Fischer, *Macromol. Symp.* **2008**, *262*, 89.
[12] K. Pommering, H. Rein, D. Bertram, R. Müller, *Carbohydr. Res.* **1992**, *233*, 219.
[13] American Standard Test Method (ASTM) D 1439-03.
[14] B. Wolf, I. Finke, *Pharmazie* **1992**, *47*, 35.
[15] B. Wolf, *Pharmazie* **1993**, *48*, 223.
[16] European Pharmacopoeia **2005**, 4. Ausgabe, Deutscher Apotheker Verlag.

