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DESIGN AND CHARACTERIZATION OF DICLOFENAC SODIUM MICROBEADS BY IONOTROPIC GELATION TECHNIQUE

P. S.GOUDANAVAR*, R.S.BAGALI, CHANDRASHEKHARA.S AND S. M.PATIL

K.L.E's College of Pharmacy, Nipani, Karnataka, India

*Corresponding Author p.goudanavar@rediffmail.com

ABSTRACT

Sustained release oral product namely microbeads for Diclofenac sodium prepared by ionotropic gelation technique using Sodium alginate alone and combination with Hydroxypropyl methyl cellulose, Chitosan, Pectin as release rate modifiers, and investigated for flow behavior, particle size, swelling properties, surface study by SEM, and in vitro drug release potential. While increase in the concentration of sodium alginate and other polymer dispersions increased sphericity, size distribution, mean particle size. Drug entrapment efficiency approached nearly 95%. Increasing calcium chloride concentration decreases the mean diameter of the microbeads, no appreciable change in morphology, and drug release behaviors. In vitro drug release was dependent on the pH of the medium and concentration of polymer dispersions. Among the nine formulation batches F5, F7 and F9 were found to show optimum sustained effect. The mechanism of drug release from the microbeads was found to be followed super case-II transport.

KEY WORDS

Sustained release, microbeads, Diclofenac sodium, ionotropic, Hydroxypropyl methyl cellulose

INTRODUCTION

In recent years a wide variety of newer drug delivery systems like oral sustained/ controlled release dosage forms are designed and evaluated in order to overcome the limitations of conventional therapy. The non-steroidal anti-inflammatory drug Diclofenac sodium is a good candidate for the development of oral controlled release formulation. It is used for long treatment of inflammation and pain, with dose range 50-75 mg as conventional tab/ capsules. Adverse gastrointestinal reactions have been observed, and its short biological half life (1-2 h) requires administration 2-3 times a day[1]. Microencapsulation has been employed to sustain

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the drug release, reduce or eliminate drug related adverse effects, dose-intake and improve the bioavailability in spite drug undergo extensive first-pass metabolism ultimately improve the compliance in pharmacotherapy of inflammation and pain[2]. Microencapsulation by ionotropic

gelation is one of the widely used method for preparation of calcium-alginate microspheres/beads which has ability to form gels reaction with calcium salts. Recently the use of calcium-alginate gel beads as a vehicle for controlled drug delivery system has attracted considerable attention because of their property of reswelling which is susceptible to environment pH. Consequently, acid sensitive drugs incorporated into beads would be protected from gastric juice. However, major disadvantages of alginate beads are their fast disintegration in simulated intestinal fluid and high porosity, which result in rapid drug release[3].

These considerations led to the objective of this study, which was to prepare and evaluate oral sustained release product namely microbeads for diclofenac sodium by ionotropic gelation method using sodium alginate alone and HPMC, chitosan, pectin as release rate modifiers to overcome the fast disintegration of alginate beads in simulated intestinal fluid. In the proposed method we dropped the mixture of drug and polymer dispersion into an aqueous calcium chloride solution gelation occurs instantaneously resulting the formation of spherical beads, with narrow particle size, high yield and optimum sustained release in various physiological GI conditions.

MATERIALS AND METHODS

Diclofenac sodium was obtained as a gift sample from Glenmark Laboratories Ltd., Aurangabad. Hydroxy propyl methylcellulose and Chitosan were from Loba Chemicals India. Sodium alginate, pectin and calcium chloride were purchased from SD Fine Chemicals Ltd., Mumbai. All other reagents used were of analytical grade.

Preparation of Microbeads:

The microbeads of Diclofenac sodium were prepared by using ionotropic gelation technique. In the present work four sets of microbeads were formulated by using sodium alginate alone and combination with HPMC, chitosan, pectin in different proportions and calcium chloride as cross-linking agent. The detailed composition of the various formulation batches (F1-F9) were mentioned in Table 1.

In the first set three batches of drug-loaded microbeads were prepared (F1, F2 & F3). In 50ml of sodium alginate solution, weighed quantity (100mg) of Diclofenac sodium was dispersed uniformly using mechanical stirrer at 500 RPM. Bubble free dispersion was dropped through a syringe with a needle of size no.18 into 100ml aqueous calcium chloride solution and stirred at 100 RPM. After stirred for 15 min the formed beads were separated by filtration, washed with distilled water, dried at 60^0 for 6 h in an oven[4].

In the second set two batches of drug-loaded microbeads were prepared (F4, F5) using fixed concentration of sodium alginate and HPMC in different proportions. The procedure was similar to the described above method. In the third set two batches of drug-loaded microbeads were prepared (F6, F7) using fixed concentration of sodium alginate and chitosan in different concentrations. In this, the mixture of drug and sodium alginate dispersion was dropped through a syringe with a needle of size no.18 into 100ml of chitosan solution containing 5% calcium chloride (Chitosan dissolved in 10ml of 5% (w/v) acetic acid) stirred at 100 RPM. After stirring for 30 min, the coated beads were separated by

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filtration, washed with water, dried at 60^0 for 6h in an oven[5].

In fourth set also two batches of drug-loaded microbeads were prepared (F8, F9) using sodium alginate and pectin in different concentration, as same as above described procedure.

Size Distribution of Microbeads:

Size distribution of the microbeads was determined using standard test sieves (Filterwel, Mumbai, India). Particles that passed through one sieve but were retained on the other were collected and weighed. Distribution of particles was analyzed based on weight fraction on each sieve[4].

Flow properties:

The flow properties of drug-loaded microbeads were investigated by measuring the angle of repose using fixed base cone method. The bulk and tapped densities were also measured in a 10ml graduated measuring cylinder as a measure of packability of the microbeads. The angle of repose (θ) was determined by the formula, $\theta = \tan^{-1}(h/r)$; h=cone height of microbeads; r = radius of the circular base[6]. Each experiment was carried out in triplicate.

Particle Size Analysis:

The mean diameter of drug loaded microbeads was performed by optical microscopy (Olympus Model 52x-12). Mount the sample containing microbeads on a clean glass-slide and observed in the microscope. The eye piece of the microscope fitted with a micrometer of calibrated for 1 unit was equal to 1/30mm (33.33 μm). All the three dimensions (lxbxh) of the microbeads were measured[6].

Drug Entrapment Efficiency:

Accurately weighed 50mg of drug-loaded microbeads were suspended in 100ml of simulated intestinal fluid of pH 7.2±0.4. The resulting solution was kept for 24 h. Next day it was stirred for 5 min and filtered. After suitable dilution, Diclofenac sodium content in the filtrate was analyzed spectrophotometrically at 276nm using a Shimadzu 1201 UV-Vis spectrophotometer[3]. The drug entrapment efficiency was determined using following formula:

$$\% \text{ DEE} = \frac{\text{Actual drug content}}{\text{Theoretical drug content}} \times 100$$

Swelling Properties:

Thirty beads were placed in a beaker containing 100ml of acidic buffer solution and then stirred with a magnetic stirrer at a speed 50 RPM. After 1h interval the equilibrium swollen beads were observed and measured under optical microscope. The magnitude of swelling was presented by ratio of the mean diameter of swollen beads to the mean diameter of the dried beads[7].

Scanning Electron Microscopy:

The drug-loaded microbeads observed under SEM (model JSM 35CF Jeol, Japan) at 15 Kv by mounting sample on the aluminium stubs, using double sided adhesive tape and vacuum coated with gold film using sputter coater (Bio Rad SC 502). The coated microbeads were observed under suitable magnification for surface characteristics[10].

***In vitro* Dissolution Study:**

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Dissolution studies were carried out in a USP XIII rotating basket apparatus containing 900ml of simulated gastrointestinal fluids of increasing pH namely pH 1.2 (2 h), pH 6.0 (1 h) and pH 7.2 (up to 10 h) maintained temperature $37^0 \pm 0.5^0$. Hard gelatin capsule filled with microbeads equivalent to 100mg of Diclofenac sodium were placed in basket rotated at a constant speed of 75 RPM. Aliquots of sample were withdrawn after predetermined periods and were replenished immediately with the same volume of fresh medium. Aliquots, following suitable dilution, were analyzed spectrophotometrically at 276nm[8].

Kinetics of Drug Release:

In order to understand the mechanism and various kinetics of drug release, the data obtained from the *in vitro* dissolution studies were analyzed with various kinetic equations like zero order (% release Vs. time), first order (log % retained Vs. time) and Korsmeyer Peppas equation ($m/_{m\infty}$ Vs. t_n). From these plots coefficient of correlation (r) values were calculated[9].

RESULTS AND DISCUSSION

Compatibility of drug with the various polymers was determined by IR spectrometer using a (Shimadzu FTIR-8000 model). I.R. spectrometer, suggest that the drug and polymer are compatible and free from chemical interactions. Increasing the concentration of sodium alginate and calcium chloride solution tended to make the particles more spherical and obtaining the uniform size beads (Fig. 2a). All the formulations showed an acceptable range of angle of repose. The formulation batches prepared with coating polymers such as F4, F5, F6, F7 and F8, F9 showed good flowability, as shown in Table 1.

Particle size of drug-loaded microbeads was performed by optical microscopy, obtained in the range 770-920 μm reported in Table 2. The mean diameter of the particles was found to decrease by increasing in the concentration of calcium chloride solution. It has been stated as Ca^{2+} ions penetrates into interior sodium alginate droplets, water is squeezed out, resulting in contraction of the beads, on other hand increases in the coating polymer concentration leading to increase in the diameter of formed beads. The drug entrapment efficiency of all the batches was in the range 70.4 percent to 95.2 percent, as reported in Table 1. Drug entrapment efficiency of microbeads increases with increasing in the percentage of sodium alginate as well as coating polymers concentration due to formation of dense matrix structure could decreases the loss of drug in the curing medium. But the amount of $CaCl_2$ has probably no significant effect on drug entrapment efficiency. The swelling behaviour of the drug-loaded microbeads was observed in pH 1.2 up to 2 h as shown in Table 2. Under acidic conditions swelling of calcium alginate beads scarcely occurs, because the osmotic pressure gradient that exist between the alginate gel and with acidic environment. However increasing concentration of calcium chloride solution produces the beads with higher level of Ca^{2+} ions could reduces the swelling of the beads. Scanning electron microscopy of the formulation batches F2, F5, F7 and F9 as shown in Fig 2(a), 2(b), 2(c) and 2(d). The SEM of the microbeads (F2) prepared with sodium alginate alone are spherical in shape exhibits rough surface has a sandy appearance Fig. 2(a), however the batches F5 (HPMC) Fig. 2(b), F7 (Chitosan) (Fig. 2(c) and F9 (Pectin) Fig. 2(d) appeared to be spherical, observed bridging and dense nature due to coalescence and fusion to the colloidal aqueous polymer dispersions in the alginate matrix leads to increase in diameter decreases porosity accounts for slow release of drug.

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In vitro dissolution studies were conducted by using three different dissolution mediums simultaneously in pH 1.2 for 2h. Then the beads shifted at pH 6.0 for 1h, and finally pH 7.2 up to 10 h. Cumulative % drug released was determined and reported in Table 2. The drug release behaviours in pH 1.2 was very slow due to slow solubility of drug in acidic medium, and moreover that calcium ions in alginate beads were totally discharged in an acidic environment and the carboxyl groups were shifted to an unionized form. In addition sodium alginates are the polyelectrolytes that exhibit the maximum swelling behaviour were observed at pH 6.0. Percentage cumulative drug release from the formulated microbeads showed increase drug release at pH 6.0 than in pH 1.2 (Table 2). Chitosan treated alginate beads (F5, F6) showed somewhat increase drug release in the acidic medium. Protonation of amine groups improves solubility and exists gel at low pH due interpolymeric complex between alginate and chitosan. The drug release behavior was increased in pH 7.2 were observed in almost all the batches due to increase swelling behavior as well as the drug entrapment efficiency. The cumulative percentage drug release at 10^{th} h for the batches F1 to F3 in the range 93% to 98% w/w (Fig. 1). The data shows increase in sodium alginate concentration however extent drug release to some extent. Cross-linking takes place only between carboxylate residue of GG-blocks and Ca^{2+} ions forms a tight gel network structure and subsequent disintegration and dissolution takes place through ion-exchange between the Ca^{2+} ions and Na+ ions present in the dissolution medium leading to drug release some extent, but more faster release compared with other batches because alginate beads having fast disintegration in simulated intestinal fluid and high porosity.

More sustained drug release was observed for batches F5 to F9 using polymer like HPMC, chitosan and pectin (Fig. 1). At neutral pH the viscous complex will swell and gel formed will slowly disintegrate, would be expected to resist erosion of alginate beads leads to prolong drug release. The diffusion co-efficient and correlation co-efficient were assessed and was found to be more linear for zero order release as compared to first order. The kinetic data was best fitted to Korsmeyer-Peppas model. The diffusion co-efficient (n) in the range 0.8806 to 1.4503, which indicates that the release of drug occurs by diffusion following super Case-II transport (Table 3). On the basis of estimated physicochemical properties and in vitro release data proved formulation F5 and F7 having 1% HPMC and 1% chitosan as coating polymers showed more optimum sustained release profile (Fig.1) with maximum entrapment efficiency followed by zero-order kinetics.

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Table 1

Composition, Drug entrapment efficiency and flow properties of the microbeads

Formu-lation Code	Sodium Alginate % (w/v)	Calcium Chloride % (w/v)	HPMC % (w/v)	Chitosan % (w/v)	Pectin % (w/v)	Drug Entrapment Efficiency (%)	Angle of Repose
F1	2	3				70.4	$32^0.25'$
F2	3	5				77.2	$30^0.15'$
F3	4	7				85.3	$28^0.20'$
F4	3	5	0.5			90.9	$26^0.20'$
F5	3	5	1.0			94.7	$25^0.30'$
F6	3	5		0.5		89.9	$25^0.40'$
F7	3	5		1.0		95.2	$24^0.60'$
F8	3	5			1	86.3	$27^0.15'$
F9	3	5			2	88.0	$26^0.80'$

Based on dry polymer weight, each formulation containing 100mg of Diclofenac sodium.
All values are mean of triplicates.
#means±SD of triplicates.

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Table 2
Physicochemical characteristics and in vitro release kinetic data of microbeads in various ph conditions

Formulation Code	Mean Diameter (µm)		Cum. Percent Drug Released (%)		
	Dried beads	Swelling beads in pH 1.2, 2 h	pH 1.2 for 2 h	pH 6.0 for 1 h	pH 7.2 up to 10 h
F1	822.14	862.30	5.20	18.20	98.40
F2	794.25	823.40	4.80	25.20	95.60
F3	778.50	803.20	4.20	28.40	93.80
F4	860.15	910.40	4.60	22.30	87.40
F5	890.55	931.30	4.80	20.40	81.40
F6	970.60	1029.10	6.10	34.30	88.40
F7	982.15	1026.15	5.80	32.40	86.10
F8	908.60	958.70	4.50	37.20	88.60
F9	920.15	962.70	4.10	35.40	86.60

Table 3
Model fitting data of the release profile for Diclofenac sodium using different models

Formulation Code	Mathematical Models (Kinetics)			
	Zero Order (r)	First Order (r)	Krosmeyer – Peppas (r)	'n' values
F1	0.9980	--	0.9984	0.8806
F2	0.9932	0.9076	0.9960	0.9160
F3	0.9981	0.9098	0.9955	0.9271

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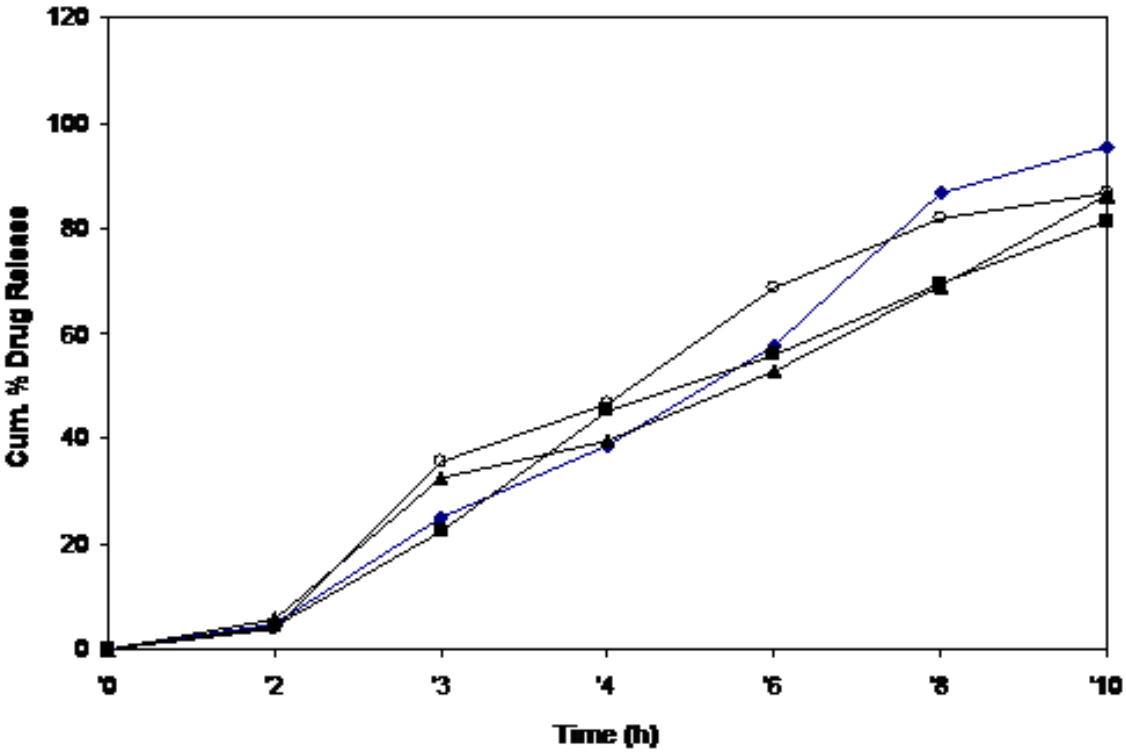
F4	0.9942	0.9854	0.9937	1.0682
F5	0.9988	0.9877	0.9969	1.1304
F6	0.9943	0.9026	0.9806	1.3520
F7	0.9964	0.9591	0.9968	1.2206
F8	0.9918	0.9818	0.9848	1.4506
F9	0.9890	0.9885	0.9747	1.1506

Figure 1

Graphs of Cumulative % Drug Release V/s. Time for Formulations F2, F5, F7 and F9.



In vitro drug release profile of drug loaded micro beads containing sodium alginate (F2) [♦], Sodium alginate with HPMC (F5) [■], Sodium alginate with Chitosan (F7) [▲] and Sodium alginate with pectin (F9) [○].

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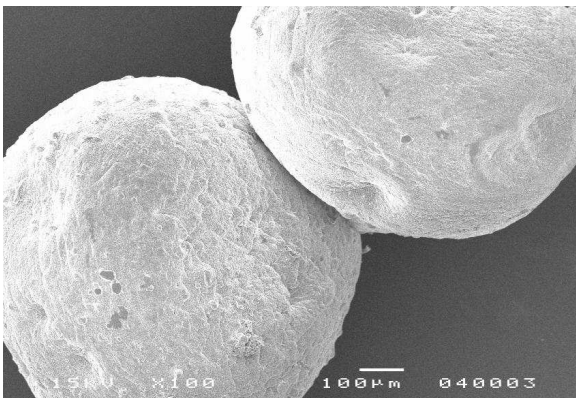
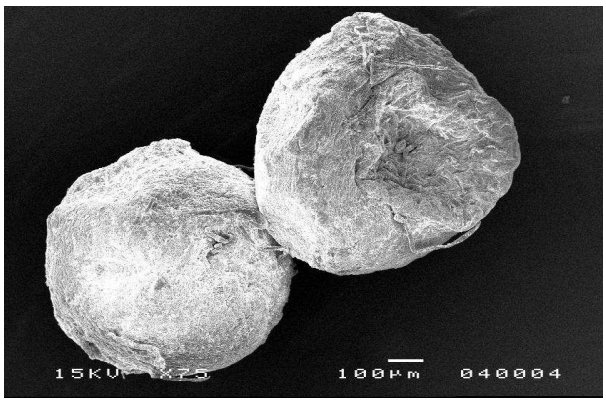
Figure 2

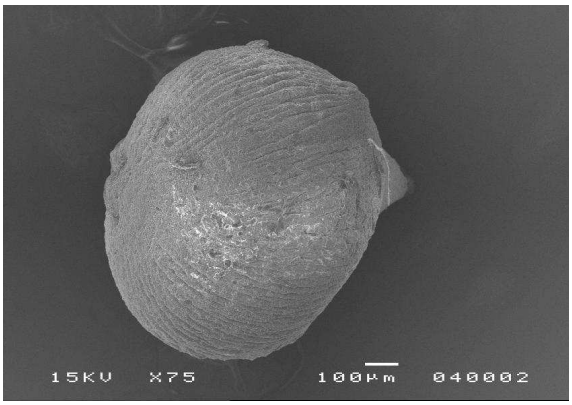
Scanning Electron Microscopy (SEM) Photographs of Drug-Loaded Microbeads



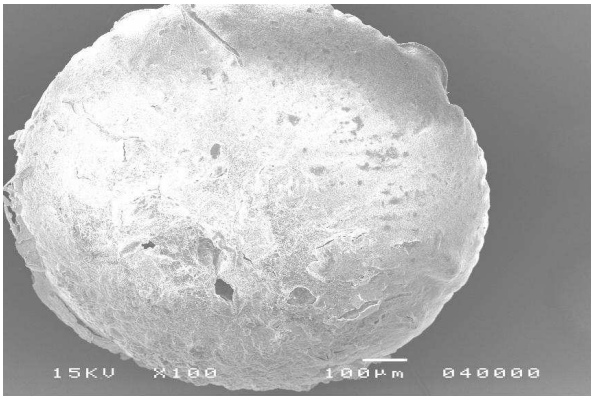
a



b



c



d

2(a) SEM of formulation F2 containing sodium alginate at 100X, using 15 K.V.
2(b) SEM of formulation F5 containing sodium alginate and HPMC at 75X using 15 KV.
2(c) SEM of formulation F7 containing sodium alginate with chitosan at 75X using 15KV
2(d) SEM of formulation F9 containing sodium alginate with pectin at 100X using 15 KV

CONCLUSION

The micro encapsulation ionotropic gelation technique is inexpensive, prevented drug related adverse effects and used to formulate oral controlled release formulations for Diclofenac sodium using HPMC and chitosan as drug release modifiers.

REFERENCES

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1. Robert Jackson, Land Marrow D. Jason in Goodman S and Gilman A. The Pharmacological Basis of Therapeutics, 10th edn. MacGraw Hill Publishers New York, 1445, 2001
2. Patric B Deasy, in 'Drug and Pharmaceutical Science', 2nd edn. Marcel Dekker Inc., New York, 22, 1984.
3. Rajesh KS, Khanrah A and Biswanath Sa. J. Sci. Ind. Res. 62,985, 2003.
4. Manna A, Gosh I, Goswami N, Gosh LK and Gupta BK. J. Sci. Ind. Res., 58, 717,1999.
5. Anil K Anal, Deepak Bhopatkar, Sellehi Tokuria and Hiroshi Tamura. Drug Develop. Ind. Pharm. 29, 713, 2003.
6. Martin, Alfred Eds., in 'Physical Pharmacy', 4th Edn., BI Waverly Pvt. Ltd., New Delhi; 340, 1991.
7. Pornsak Srimornasak, Jurairat Nunthanid. Int. J. Pharm., 106 ,207, 1998
8. Gang Cheng, Feng An, Mei-Juan Zou, Jin Sun, Xiu-Hua, Yun-Xia. He. Word J. Gastroenterol.; 10(12), 1769, 2004.
9. Paulo Costa, Jose Manual Sousa Lobo. Eur. J. Pharm. Sci.; 13, 123, 2001
10. Alf Lamprechet, Ulrich Schafer and Claus-Michael Lehr. AAPS Pharm. Sci. Tech., 1(3), 17, 2000.