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Study of Formulation Variables on Properties of Drug-Gellan Beads by Factorial Design

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ABSTRACT The aim of the present study was to obtain cross-linked calcium-gellan beads containing diclofenac sodium as model drug, using full 3^3 factorial design. Drug quantity, pH of cross-linking solution, and speed of agitation were selected as variables for factorial design. The resultant beads were evaluated by scanning electron microscopy (SEM), percent yield, entrapment efficiency, micromeritic properties, swelling and drug release studies. The drug-loaded beads were spherical with size range of 0.85–1.8 mm. Percent yield and entrapment efficiency of various batches were in the range of 86.48–98.28% w/w and 72.52–92.74% w/w, respectively. Calcium-gellan beads containing diclofenac sodium showed pH-dependent swelling and drug release properties. Swelling and drug release were significantly higher in pH 7.4 phosphate buffer than 0.1N HCl. The swelling ratio for beads was up to 22 and 3 for phosphate buffer and 0.1N HCl, respectively. Cumulative diclofenac sodium release from calcium-gellan beads was 12–35% in 0.1N HCl within 2 h, whereas complete drug release was observed within 3–4 h in pH 7.4 phosphate buffer.

KEYWORDS Low-acyl gellan, Calcium-gellan beads, Ionotropic gelation, Factorial design, pH of cross-linking solution, Speed of agitation

INTRODUCTION

Gellan gum is an extracellular anionic, heteropolysaccharide produced by *Sphingomonas elodea* during aerobic fermentation (Kang et al., 1982; Anon, 1992). Gellan consists of a linear structure of repeating tetrasaccharide units of glucose, glucuronic acid, and rhamnose (Izumi et al., 1996; O'Neill, 1983). The native polysaccharide, high-acyl gellan, contains approximately 6% O-acetyl groups, which lost during alkali treatment of commercial extraction yielding low-acyl gellan gum. This commercial gellan is a potassium salt; in addition, it contains low levels of sodium, calcium, and magnesium.

Low-acyl gellan cross-link more effectively with divalent cations than the monovalent cations (O'Neill et al., 1983; Alhaique & Santucci, 1996; Morris,

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et al., 1996). Divalent metal ions bind to pair of carboxylic group on nearby helices, whereas monovalent metal ions bind individual helices. (Tang et al., 1996). The strongest gellan gels were obtained when the [cation]/[carboxylate] ratio corresponds to 0.5 for divalent and 10–30 for monovalent. At optimum concentration of respective cations, gellan form stronger gel with Ca^{2+} than Mg^{2+}. Low-acyl gellan forms gel at temperatures between 30 and 50°C. The high-acyl gellan has poor aggregation properties; it gels at around 70°C. Low-acyl gellan forms hard and brittle gel, whereas high-acyl gellan forms soft and elastic gel. Below pH 3.6, gellan forms acid gel, which has lower modulus and hardness value. (Grasadalen & Simdrod, 1987; Mortika et al., 1991). The high-acyl gellan has poor cation-mediated aggregation. The acetyl groups present on the surface of the duplex block helix-helix aggregation, and the stearic effect of bulky glyceryl group repels the carboxylic groups, making it incapable of accommodating site-bounded cations. (Morris et al., 1996; Standerson & Clark 1983; Kedzierewic et al., 1999).

Gellan is safe for human use, and it is widely used in the food industry as bacterial growth media and in plant tissue culture. (Milas et al., 1990). It has been studied for pharmaceutical applications including oral gels, beads, and eye drops. (Milas et al., 1990; Quigley & Deasy, 1992). Gellan beads offer an oral multiparticulate delivery system that provides many relative advantages over single-unit dosage forms such as predictable gastrointestinal transit time, greater product safety, etc. (Ritschel, 1991). Preparation of gellan beads by ionotropic gelation method involve very mild processing conditions in an aqueous, organic solvent-free environment. The purpose of the present research was to formulate cross-linked calcium-gellan beads containing diclofenac sodium as model drug using 3^3 factorial design. Drug quantity, pH of calcium chloride solution, and speed of agitation were selected as variables, and the resultant beads were evaluated by SEM, micromeritic properties, swelling, and drug release studies.

MATERIALS AND METHODS

Materials

Deacetylated Gellan gum, Kelcogel®, was supplied by C.P. Kelco Pvt. Ltd. Mumbai (India). Diclofenac sodium was supplied by Emcure Pharmaceuticals Pvt. Ltd., Pune (India). Calcium chloride was purchased from Sisco Research Lab, Mumbai (India). All other chemicals were of analytical grade.

Preparation of Beads

The beads were prepared according to the technique of ionotropic gelation. Diclofenac sodium (1, 2, or 3 g) was dissolved in 150 mL of deionised water at 55°C using cryostatic bath (Haake Phoenix C25P, Germany). Gellan gum (3 g) was added to this stirred solution (300 rpm) at the same temperature until a uniform dispersion was obtained. The resultant homogeneous bubble-free slurry was drip, using a disposable glass syringe preheated in boiling water having a needle of bore size, 18G, into 300 mL of calcium chloride solution (2% w/v) and was agitated continuously for 15 min with use of a constant speed stirrer with propeller blade (Eurostar power control-visc, IKA Labortecnik, Staufen, Germany). The beads obtained were separated by filtration and washed with deionized water. Beads were dried at room temperature. All batches were prepared in triplicate.

Effect of Variables

To study the effect of variables, batches were prepared by using 3^3 factorial design. Drug quantity, pH of cross-linking solution (2% w/v calcium chloride solution), and speed of agitation were selected as three independent variables. The pH of cross-linking solution was adjusted by using concentrated hydrochloric acid. Various batches prepared by using all possible combinations of different levels of experimental variables are listed in Table 1.

Evaluation of Beads

Yield and Entrapment Efficiency

Beads prepared were weighed after drying, and percent yield was calculated. The entrapment efficiency within the beads was determined indirectly by determining the amount of nonencapsulated drug, by measuring concentration of diclofenac sodium in the preparation medium and in the washing solutions. The entrapment efficiency in preparation medium of pH 1.0, pH 3.8, and pH 6.6 were, respectively, analyzed spectrophotometrically (JASCO-V500, Kyoto, Japan) at 273 nm and 277 nm

TABLE 1 Different Batches With Their Experimental Coded Level of Variables for 3^3 Factorial Design

Batch code	X_1 = drug quantity[a]	X_2 = pH of calcium chloride solution[b]	X_3 = speed of agitation[c]
S1	−1	−1	−1
S2	−1	−1	0
S3	−1	−1	1
S4	−1	0	−1
S5	−1	0	0
S6	−1	0	1
S7	−1	1	−1
S8	−1	1	0
S9	−1	1	1
S10	0	−1	−1
S11	0	−1	0
S12	0	−1	1
S13	0	0	−1
S14	0	0	0
S15	0	0	1
S16	0	1	−1
S17	0	1	0
S18	0	1	1
S19	1	−1	−1
S20	1	−1	0
S21	1	−1	1
S22	1	0	−1
S23	1	0	0
S24	1	0	1
S25	1	1	−1
S26	1	1	0
S27	1	1	1

[a]X_1 levels [1 g (−1), 2 g (0), 3 g (+1)].
[b]X_2 levels [1.0 (−1), 3.8 (0), 6.6 (+1)].
[c]X_3 levels [100 (−1), 300 (0), 500 (+1)].

after suitable dilution of samples with respective bulk preparation medium. The entrapment efficiency (EE) was calculated according to the formula given below.

$$EE = ((\text{Amount of added drug} - \text{amount of nonencapsulated drug})/(\text{Amount of added drug})) \times 100 \quad (1)$$

Scanning Electron Microscopy (SEM)

Beads were coated with a thin gold-palladium layer by sputter coater unit (VG- Microtech, United Kingdom), and the surface topography was analyzed with a Cambridge Stereoscan S120 scanning electron microscope (SEM; Cambridge, United Kingdom) operated at an acceleration voltage of 10 kV.

Surface Area

Surface area of the drug and the beads was determined by the gas adsorption method using Smart sorb 90/919, surface area analyzer (Smart Instruments, Mumbai, India). Beads of batches S7, S16, and S25 containing varying drug quantity and S7, S8, and S9 produced at varying speed were studied for surface area.

Particle Size Distribution

Randomly selected 50 beads were studied by using a stereomicroscope (Carl Zeiss, Germany) attached with a digital camera (Watec, WAT-202, Japan). Biovis image plus software (Expert Tech Vision, India) was used to analyze the images of beads. The shape and size of the beads are expressed by using different parameters such as diameter, roundness, and circulatory factor. Angle of repose (θ) of beads was determined by fixed funnel free-standing cone method.

Size and Weight Ratio

At the end of the process of the preparation of beads, the collected beads were measured for diameter and weight. Same beads were dried at 30°C for 24 h, and diameter and weight of each bead was again determined. Size and weight ratio were calculated by using the following formulas:

$$\text{Size Ratio} = \text{Diameter of dried beads}/\text{Diameter of wet beads} \quad (2)$$

$$\text{Weight ratio} = \text{Weight of dried beads}/\text{Weight of wet beads} \quad (3)$$

Swelling Study

Randomly selected 10 beads of each batch were studied for swelling characteristics. Beads were put in 20 mL of 0.1N HCl and phosphate buffer pH 7.4 separately, stirred slowly with a magnetic stirrer, and allowed to swell at room temperature. The beads were periodically removed at an interval of 15 min and weighed until three successive constant weights. The

swelling ratio was calculated as per the following formula (Shu & Zhu, 2002):

$$\text{Swelling Ratio} = \text{Weight of wet beads} / \text{Weight of dried beads} \quad (4)$$

Dissolution Studies

The dissolution studies were performed by using USP 26 type II dissolution test apparatus (Electrolab TDT-06P, Mumbai, India). Dissolution medium used were 0.1N HCl (pH 1.2) and phosphate buffer (pH 7.4), each 900 mL, temperature was maintained at 37 ± 2°C and 100 rpm stirring was provided for each dissolution study. Drug-loaded gellan gum beads equivalent to 100 mg of pure drug were used for each dissolution study. Samples were collected periodically and replaced with a fresh dissolution medium. After filtration through Whatman filter paper 41, concentration of diclofenac sodium was determined spectrophotometrically at 273 and 277 nm, respectively, for 0.1N HCl and phosphate buffer. Analysis of data was performed by using PCP Disso v2.08 software (Poona College of Pharmacy, Pune, India).

RESULTS AND DISCUSSION

Preparation and Characterization of Beads

Gellan beads have been produced by ionotropic gelation through interaction between negatively charged gellan and positively charged calcium ion. Preliminary study to obtain cross-linked gellan beads was carried out by using 0.1–2% w/v gellan solution and 0.1–2% w/v calcium chloride solution as cross-linking medium. It was observed that 2% w/v gellan gum dispersion in 2% w/v calcium chloride solution produced satisfactory beads at agitation speed of 100–500 rpm. Therefore, gellan concentration was kept constant, 3 g in 150 mL of deionized water. Because physicochemical properties of gellan vary according to pH, diclofenac sodium-loaded Ca-gellan beads were prepared by using calcium chloride solution of pH 1.0, 3.8, and 6.6 as cross-linking solution. Accordingly, drug quantity, pH of $CaCl_2$ solution, and speed of agitation were selected as factors for factorial design.

Surface Topography and Micromeritic Properties

Surface topography of beads is shown in Figs 1 and 2. The plain gellan beads (without drug), produced at 300 rpm with 2% w/v calcium chloride solution (pH 6.6), were spherical with minor ridges of shrinkage. The drug-loaded beads were spherical and white in appearance, and the whiteness gradually fades with increase in pH of cross-linking solution. The effect of each variable on the morphology of beads was studied by keeping other variables constant. The effect of drug quantity was studied by keeping constant pH 6.6 and speed 100 rpm. The loosely packed drug crystals were observed on the surface of high drug-containing beads, and the surface of beads became more open with increase in drug concentration. Packing of the beads is also revealed in the surface area of various batches, which were 0.334 ± 0.013, 0.392 ± 0.008, and 0.457 ± 0.017 m^2/g, respectively, for batches S7, S16, and S25.

The effect of pH of cross-linking solution was studied by preparing beads at constant drug quantity (1 g) and speed 100 rpm. As shown in Fig. 2.1, the surface deposits of densely packed finer drug crystals were observed when beads were prepared by using cross-linking solution of lowest pH. At pH 3.8, the surface was covered with thick nonuniform gel (Fig. 2.2), which was more uniform at pH 6.6 (Fig. 1.2). It reveals the possibility of preferential formation of acid gel at low pH. The acid gels are insoluble and less viscous than the gels formed at higher pH conditions. (Deasy & Quigley, 1991). Thus, the increased uniformity of the gel coat at higher pH may be attributed to viscosity and cohesivity of gel. Acid gel formed was embedded with fine drug crystals in beads that may have resulted because of less water-holding capacity of acid gel, preferential crystallization of drug at acidic condition, and leaching of drug toward bead surface under the force of agitation.

The beads were also prepared at 1g drug quantity and pH 6.6 at various speeds. The formation of thick and uniform gel coat at 100 rpm is already explained (Fig. 1.2). The increased swirling action with increase in speed disturbed the formation of uniform surface gel coat, yielding rough and open structured beads (Fig. 2.3 and 2.4). Micromeritic properties such as size, angle of repose, and circulatory factor were mainly governed by drug amount. Drug-loaded gellan beads

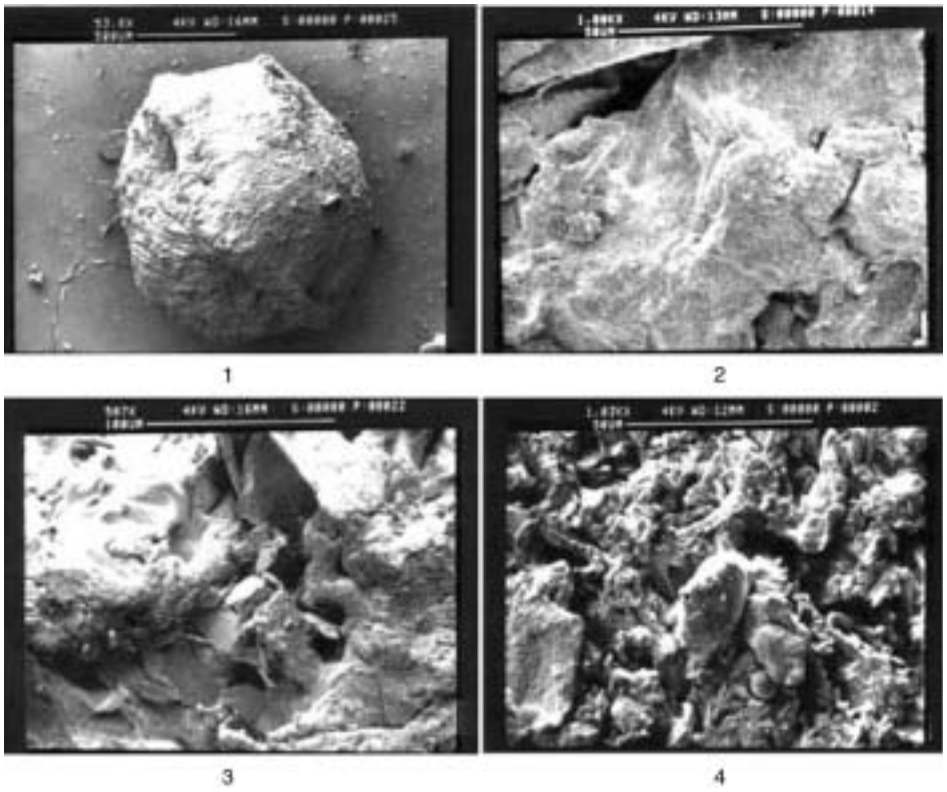

FIGURE 1 SEM microphotographs of plain gellan bead at 50× and diclofenac sodium-gellan beads at 1000×. (1) Plain gellan bead. (2) Batch S7. (3) Batch S16. (4) Batch S25.

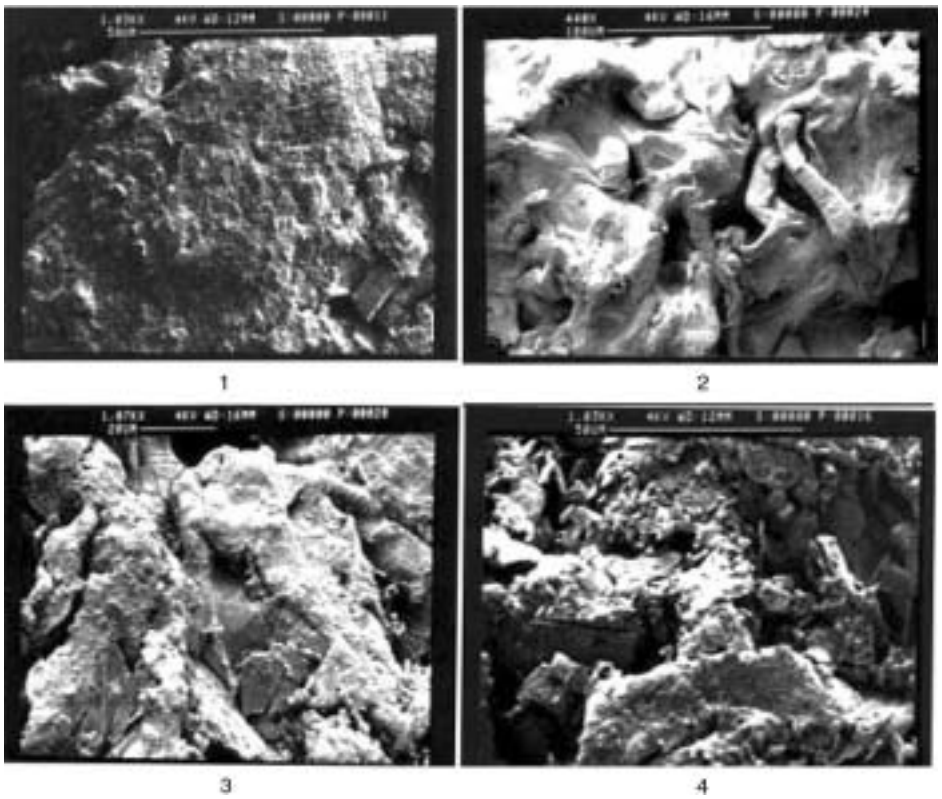

FIGURE 2 SEM microphotographs of diclofenac sodium-gellan beads. (1) Batch S1 at 1000×. (2) Batch S4 at 500×. (3) Batch S8 at 1000×. (4) Batch S9 at 1000×.

ranged in size from 0.85 to 1.8 mm. The bead size increased with increase in drug loading. Roundness of beads of different batches ranged from 0.5423 to 0.8225. The beads containing low diclofenac concentration were comparatively round and circular. The beads containing lesser drug showed greater reduction in both size and weight during drying of beads (Figs. 3 and 4).

Among all these batches, the beads prepared by using 1-g drug quantity, agitated at 100 rpm and cross-linked with calcium chloride solution of pH 6.6 were comparatively spherical, smooth and dense. This may be explained on the basis that, in the beads that were obtained with use of pH 6.6 cross-linking solution, the crystallization of drug from the entrapped liquid occurs during drying stage, which is blanketed by thick uniform gel coat, whereas in the beads that were obtained with use of cross-linking solution of pH 1.0, the crystallization of drug occurs at interface during curing of beads, causing surface deposition of drug crystals. The smoothness and sphericity of beads is reflected by angle of repose. Angle of repose of drug–loaded beads of all batches was in range of 25–31°. The beads containing less drug quantity were readily able to flow.

Factorial Design Studies

The data obtained from the experiments were subjected to multiple-regression analysis using UNISTAT® statistic version 3, Meglon, USA. The data were fitted in the equation (Bolton, 1997):

$$Y = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_3 + \beta_{12} X_1 X_2 + \beta_{13} X_1 X_3 + \beta_{23} X_2 X_3 + \beta_{11} X_1 X_1 + \beta_{22} X_2 X_2 + \beta_{33} X_3 X_3 + \beta_{123} X_1 X_2 X_3 \quad (5)$$

Multiple-regression analysis and F statistics were used to identify statistically significant term. β_0 is the arithmetic mean response, and β_1 is the coefficient of factor X_1. The results of multiple-regression analysis are summarized in Table 2. Influence of formulation

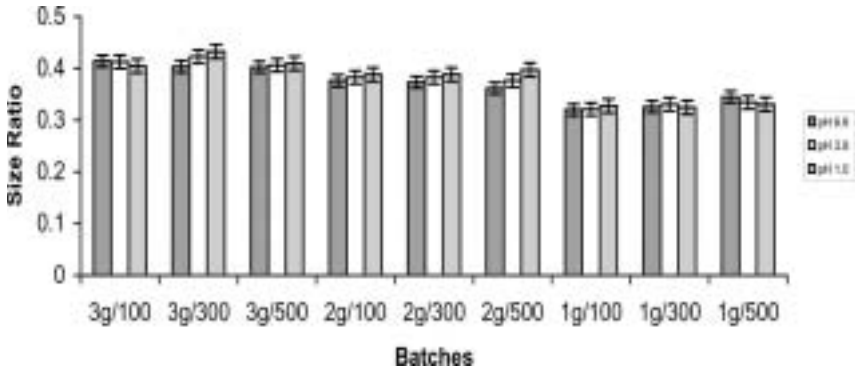

FIGURE 3 Size ratio of different batches of diclofenac sodium-gellan beads (n = 3).

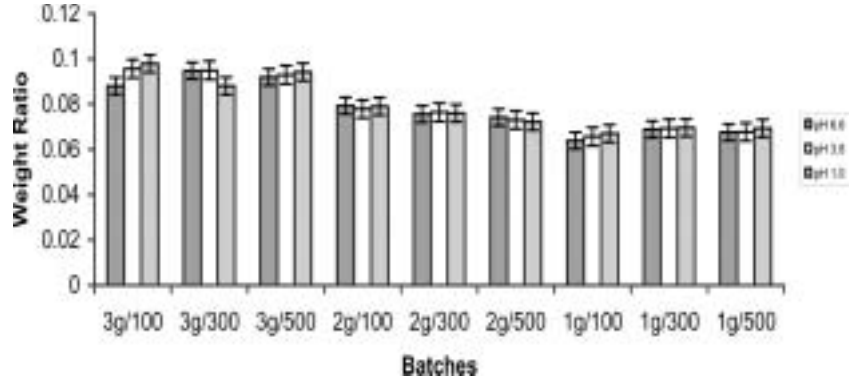

FIGURE 4 Weight ratio of different batches of diclofenac sodium-gellan beads (n = 3).

TABLE 2 Regression Analysis of Different Evaluation Parameters for Calcium-Gellan Beads (n = 3)

Coefficient	Percentage yield	Entrapment efficiency	Swelling ratio in 0.1N HCl at times in min.		Swelling ratio in pH 7.4 phosphate buffer at times in min.		Time for 50% drug release in pH 7.4 phosphate buffer($t_{50\%}$)
			30	60	30	90	
β_0	92.1481*	82.826*	1.3515*	1.6048*	2.703*	6.263*	43.852*
β_1	–1.5556*	–2.215*	–0.1039	0.4528*	–2.004*	2.877*	–17.133*
β_2	–4.0556*	–5.1044*	0.235*	–0.1756*	1.16*	–4.051*	–9.333*
β_3	–1.2778*	–2.7294*	0.0267	0.1294*	0.506	0.7761	–9.45*
$\beta_1\beta_2$	–.0.25	–0.1667	–0.0325	–0.1383*	–0.607	–1.361*	5.392
$\beta_1\beta_3$	–0.3333	–0.0942	0.0208	0.1125	–0.091	0.1475	3.608
$\beta_2\beta_3$	0	0.25	–0.0267	–0.0642	–0.087	–0.3758	–0.908
$\beta_1\beta_1$	0.2222	0.1194	–0.0494	0.3639	2.2817*	0.0422	18.478*
$\beta_2\beta_2$	–0.9444	1.1611	0.1372	–0.0845	–0.2433	3.3556*	–2.856
$\beta_3\beta_3$	0.7222	–0.7572	0.0422	–0.0394	0.0367	0.2006	3.494
$\beta_1\beta_2\beta_3$	0.625	0.125	0.0762	–0.0675	–0.171	–0.2737	2.35
R^2	0.984	0.917	0.819	0.968	0.948	0.969	0.939
F	95.43	17.75	7.24	48.53	29.01	50.57	24.55
P	0.0000	0.0000	0.000	0.000	0.000	0.000	0.0000

*Significant terms at $P < 0.005$.
n is the number of samples tested for each experiment.

variables on evaluation parameters is discussed under the following three subheadings;

Percentage Yield and Entrapment Efficiency

Percentage yield and entrapment efficiency of various batches was in the range of 86.48–98.28% w/w and 72.52–92.74% w/w, respectively. As shown in Table 2 and Figs. 5 and 6, yield of beads and entrapment efficiency of drug were governed by all three variables. The effect of pH may be attributed to the decrease in drug solubility with decrease in pH and formation of acid gel of gellan. The beads prepared at higher speed, higher pH, and high drug quantity of the cross-linking solution have lower yield and entrapment efficiency than those prepared at lower pH, lower speed, and at low drug quantity. At higher drug levels, the amount of gum may be insufficient to hold the increased drug amount. The aqueous solution of diclofenac sodium was used in the preparation of beads, and gellan was dissolved in drug solution. The resultant solution having pH 5.6 was dropped in the cross-linking solution of different pH and stirred at different speeds. Thus, the higher drug loss at higher pH and speed of agitation may be due to the squeezing of entrapped drug solution during rigidization, which was further enhanced by nonuniform gel coat produced at higher speed of agitation. (Park et al., 2001; Segi et al., 1989; Tomindo, et al., 1993). Crystallization of drug and difficulty in filtration of drug crystals through the polymer gel may be the governing factor for improved drug entrapment at lower pH and speed of agitation. Speed of agitation governs the force that solid drug particles exert on rigidifying gel and, hence, affects the structure of gel coat.

Swelling of Beads

Swelling studies of the drug–loaded beads were carried out in 0.1N HCl, pH 1.2, and in phosphate buffer, pH 7.4. The swelling ratio was higher in phosphate buffer than in 0.1N HCl, and it was up to 22 and 3, respectively. The swelling t_{max} was around 45 min and 90 min, respectively, in 0.1N HCl and in phosphate buffer. In both mediums, swelling of gel increased with decrease in drug concentration, but the effect was more significant in phosphate buffer (Figs. 7 and 8). The effect of pH of cross-linking solution was similar in both these mediums. Beads that were prepared by using pH 6.6 of calcium chloride solution had higher swelling than beads that were prepared by using acidic calcium chloride solution. The beads obtained by using 1g drug, prepared at 500 rpm and in

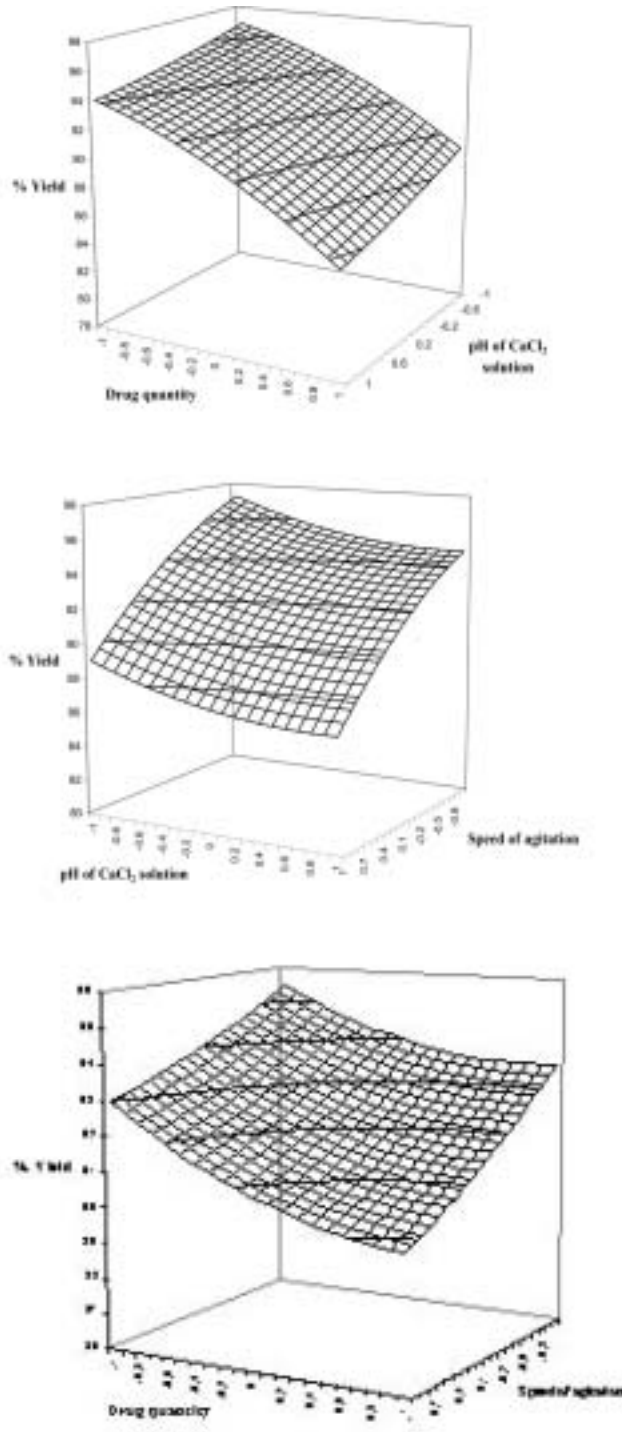

FIGURE 5 **Surface-graph showing effect on yield of the following: (1) drug quantity and pH of $CaCl_2$; (2) pH of $CaCl_2$ solution and speed of agitation; and (3) drug quantity and speed of agitation.**

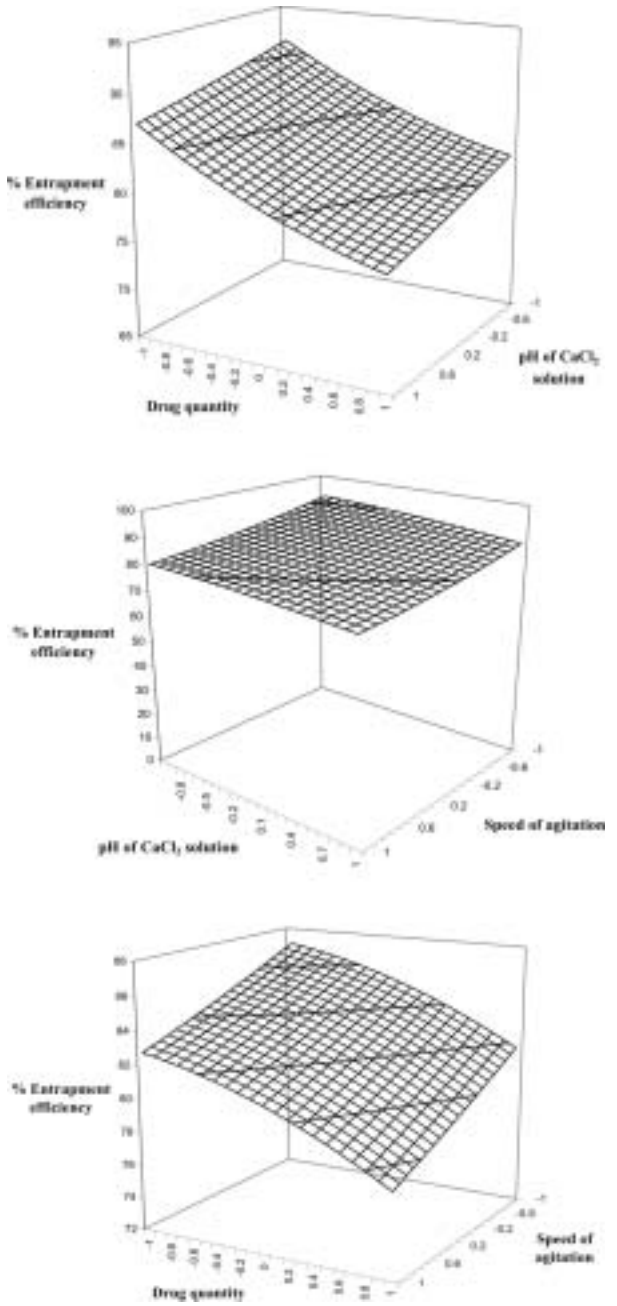

FIGURE 6 **Surface graph showing effect on entrapment efficiency of the following: (1) drug quantity and pH of $CaCl_2$; (2) pH of $CaCl_2$ solution and speed; (3) drug quantity and speed.**

pH 6.6 calcium chloride solution (batch S9) showed maximum swelling.

The regression data and surface responses shown in Table 2 and Fig. 7 reveal the effect of variables on swelling of beads in 0.1N HCl. The lower values of regression constants indicate insignificant effect of any of these individual factors on the swelling of beads in acidic medium. The curvilinear surface response curve shown in Fig. 7.1 and 7.3 is attributed to interaction between drug quantity with pH of cross-linking solution and speed of agitation, whereas the interaction between speed of agitation and pH of cross-linking solution is weak (Fig. 7.2).

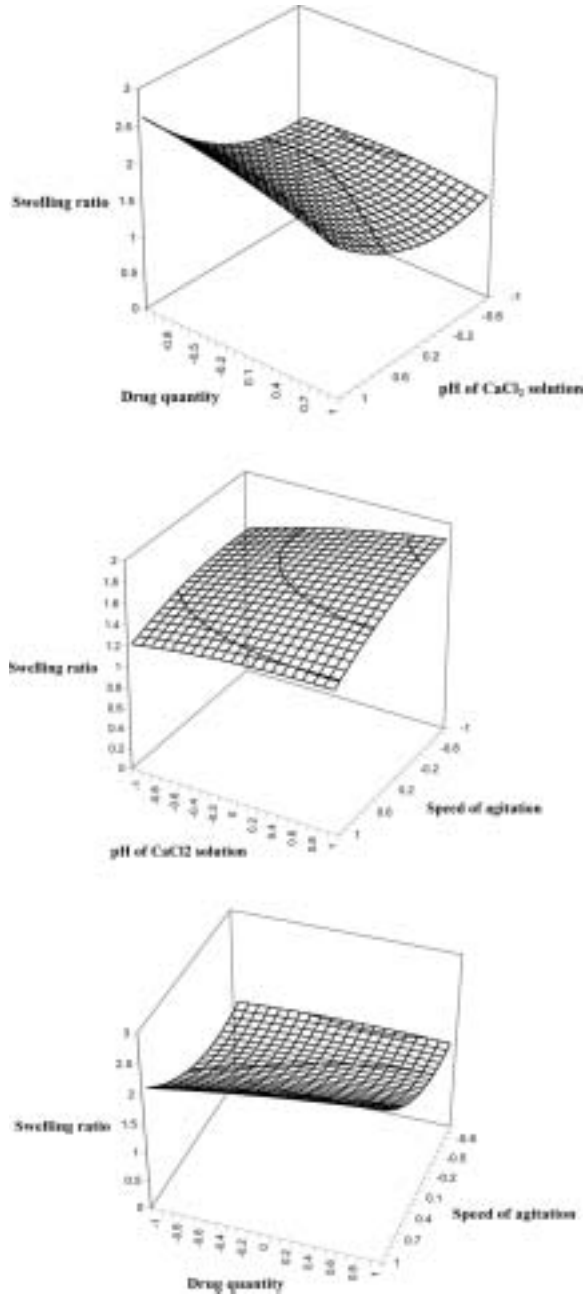

FIGURE 7 Surface graph showing effect on swelling ratio in 0.1N HCl of the following: (1) drug quantity and pH of $CaCl_2$ solution; (2) pH of $CaCl_2$ solution and speed of agitation; and (3) drug quantity and speed of agitation.

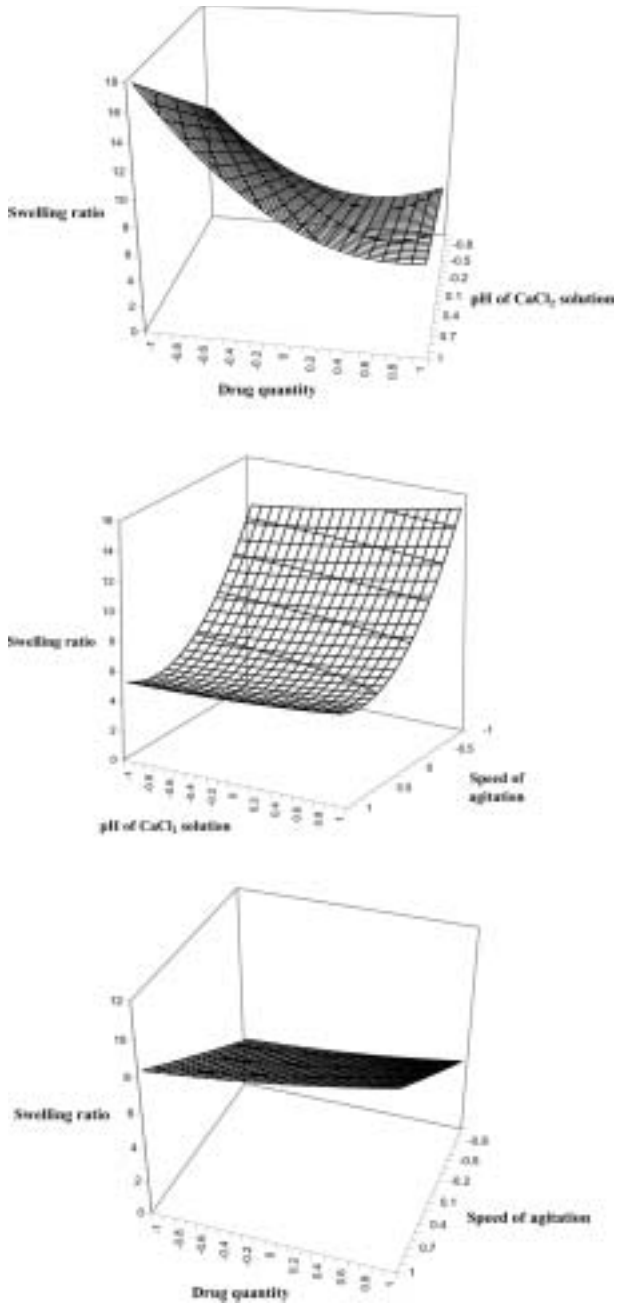

FIGURE 8 Surface graph showing effect on swelling ratio in 7.4 pH phosphate buffer of the following: (1) drug quantity and pH of $CaCl_2$ solution; (2) pH of $CaCl_2$ solution and speed of agitation; and (3) drug quantity and speed of agitation.

Swelling in phosphate buffer was increased with decrease in drug concentration and increase in pH of cross-linking solution (Fig. 8.1). The swelling of cross-linked gellan beads was greater in phosphate buffer, probably because like other cross-linked polysaccharides, swelling of Ca-gellan beads in phosphate buffer involves exchange of the Ca^{++} with the monovalent cation and sequestering effect of phosphate on Ca^{++}. (Khihiko et al., 1997). The curvilinear surface responses (Fig. 8) were observed between pH of cross-linking solution with drug quantity or speed of agitation. The uneven gel coat hastens rate and extent of penetration of medium in the beads. The increase in medium–gel contact in phosphate buffer causes maximum swelling exchanging divalent metal ions by monovalent ions. The water-holding capacity of

monovalent cross-linked gels is more. (Alhaique & Suntucci 1996) Although the batch S9 has maximum swelling, its erosion rate was also faster than S8 and S7 batches that were prepared at lower speed. The longer plateau of constant swelling with delayed erosion can be attributed to water-holding capacity of undisturbed and more uniform gel coat.

Drug Release

Diclofenac sodium release from gellan beads was 12–35% in 0.1N HCl within 2 h (Fig. 9). The beads obtained from cross-linking solution of pH 1.0 released 11–21% drug within 2 h in acidic medium, whereas beads obtained from cross-linking solution of pH 3.8 and 6.6 released 18–35% drug. Although the drug

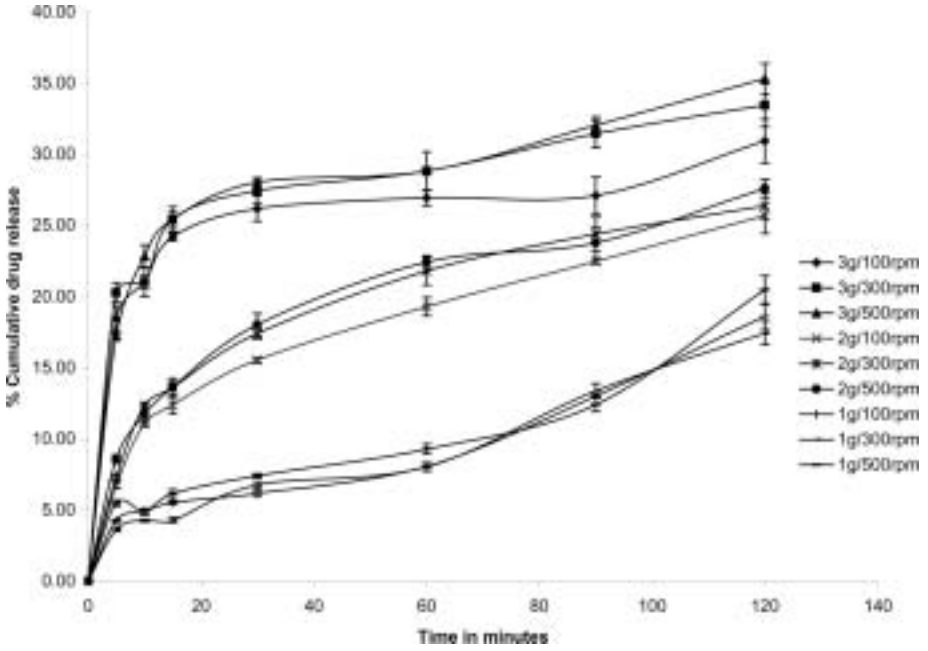

FIGURE 9 **Drug release profile of beads, prepared by using pH 6.6 $CaCl_2$ solution, in 0.1N HCl (n = 3).**

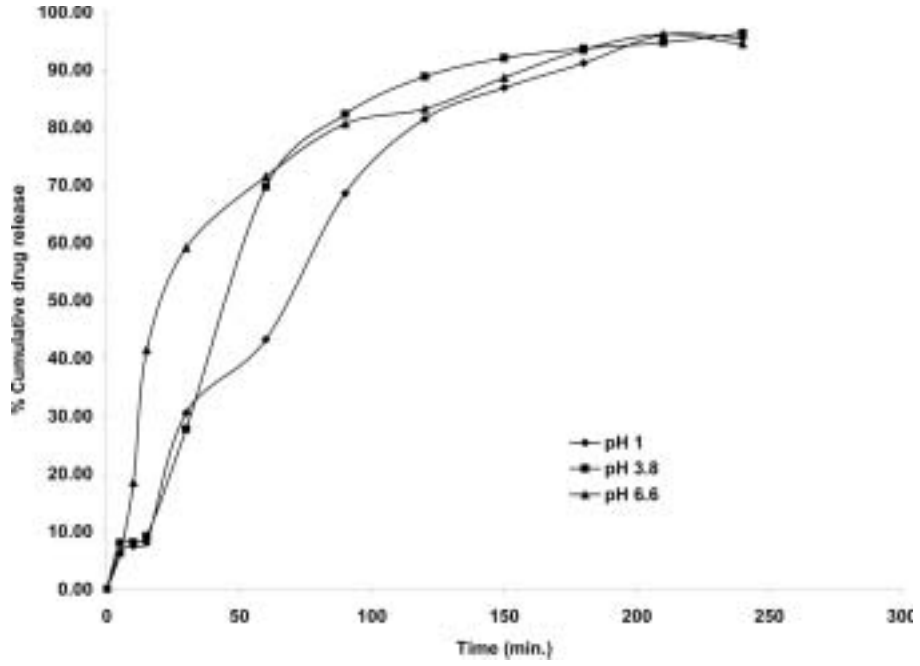

FIGURE 10 **Drug release profile of beads, prepared by using $CaCl_2$ solution of increasing pH keeping speed of agitation and drug quantity constant as 500 rpm and 3 g, respectively, (batches S3, S6 and S9), in pH 7.4 phosphate buffer (n = 3).**

crystals were deposited on the surface of beads when prepared with cross-linking solution of pH 1.0, they showed slower drug release than beads covered with thick polymeric coat. The speed of agitation has not significantly affected drug release in acidic medium.

Almost complete drug release from beads was observed within 3–4 h in phosphate buffer pH 7.4, with almost a similar release profile for all batches (Fig. 10). As shown in Table 2 and Fig. 11, the drug release was retarded with decrease in drug concentration in turn due to high gellan content. In phosphate buffer medium also the beads obtained at pH 1.0 showed slower drug release (Fig. 10). Batch S3 that is obtained at pH 1 and 500 rpm should have the faster dissolution in phosphate buffer. But beads obtained at same speed at pH 6.6 (batch S9) showed faster drug release than beads prepared at pH 3.8 (batch S6). As discussed above, crystallization of drug at interphase occurs during rigidization in acidic cross-linking solution, whereas crystallization of drug occurs during drying in beads that were prepared with use of pH 6.6 calcium chloride solution. The beads prepared at pH 3.8 and 6.6 showed surface coat of acid gel and Ca^{++} cross-linked gellan gel, respectively, which increases chances of crystallization of drug in viscous gel, yielding crystals of reduced crystallinity because of the inhibition of crystal growth. In addition, the drug release at medium rate in batch S6 and faster rate in batch S9 were attributed to swelling and erosion of porous gel in phosphate buffer.

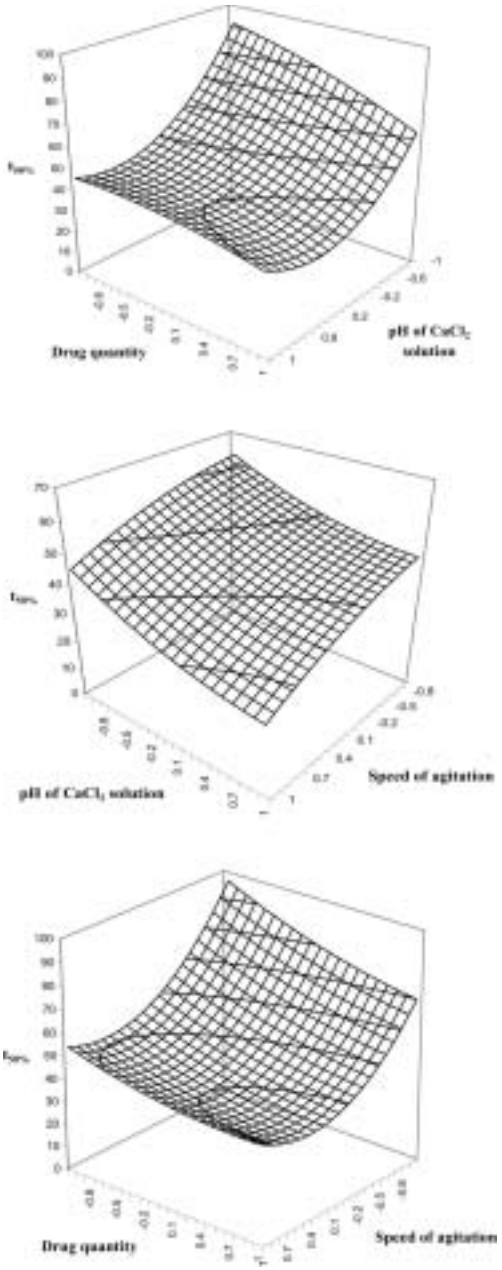

FIGURE 11 Surface graph showing effect on $t_{50\%}$ in 7.4 pH phosphate buffer of the following: (1) drug quantity and pH of $CaCl_2$ solution; (2) pH of $CaCl_2$ solution and speed of agitation, and (3) drug quantity and speed of agitation.

Because higher solubility of drug in phosphate buffer, the drug deposited on the surface of beads underwent fast dissolution. It was reflected in the SEM of beads after its dissolution in 0.1 HCl and phosphate buffer pH 7.4 (Fig. 12). Gonzalez et al. (2002) postulated the chances of formation of calcium–diclofenac precipitate, the side reaction during cross-linking of alginate. The higher dissolution values indicate occurrence of no such reaction.

CONCLUSION

Studies indicate that, in addition to drug:polymer ratio, pH of cross-linking solution and speed of agitation also influence the formation and properties of calcium-gellan beads. The pH of manufacturing liquid is more important for drugs having pH-dependent solubility and gums having pH-dependent gelling. The effect of speed of agitation is more influential for the polysaccharides showing slow rigidization during cross-linking process. Release of diclofenac sodium from calcium-gellan beads in phosphate buffer was not only dependent on drug solubility but also governed by formation of drug precipitate, which occured during processing.

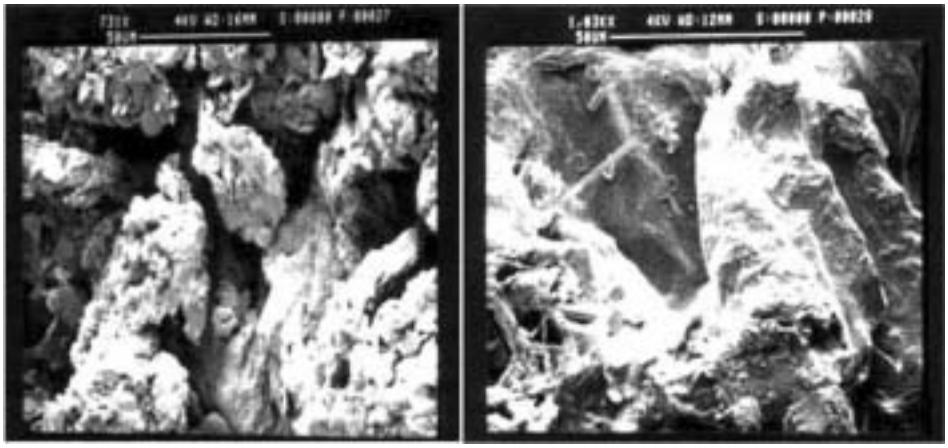

FIGURE 12 **SEM microphotographs of diclofenac sodium-gellan beads prepared by using 1 g of drug, 100 rpm speed of agitation, and pH 1.0 $CaCl_2$ solution (batch S1) after a 2-h dissolution in (1) 0.1N HCl and (2) phosphate buffer, pH 7.4.**

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