# Microspheres of Carboxymethyl Guar Gum for *In Vitro* Release of Abacavir Sulfate: Preparation and Characterization

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Received 11 December 2010; accepted 13 January 2011
DOI 10.1002/app.34173
Published online 25 April 2011 in Wiley Online Library (wileyonlinelibrary.com).

**ABSTRACT:** Carboxymethyl guar gum (CMGG), an anionic semisynthetic GG derivative, was synthesized by incorporating CM group into GG chain. Microspheres were prepared by loading abacavir sulfate (AS) using water-in-oil (w/o) emulsion method. Formulations were characterized by Fourier transform infrared spectroscopy (FTIR), nuclear magnetic resonance (NMR), X-ray diffraction (XRD), thermogravimetry (TGA), differential scanning calorimetry (DSC) and scanning electron microscopy (SEM). *In vitro* release profiles of GG and CMGG loaded with AS performed in pH 1.2 as well as 7.4 buffer media at 37°C displayed the varying drug release profiles in stomach and intestinal conditions. Release of the drug was extended up to 28 h in both GG and CMGG matrices, but the burst release observed in case of GG matrix was reduced in the case of CMGG. The kinetics of *in vitro* release was analyzed using the empirical equations. © 2011 Wiley Periodicals, Inc. J Appl Polym Sci 122: 452–460, 2011

**Key words:** carboxymethyl guar gum; NMR; DSC; XRD; controlled release

## INTRODUCTION

Natural polysaccharides have been widely explored as drug delivery devices in view of their biocompatible and biodegradable nature compared to the synthetic polymers.[1–3] In addition, these polymers possess a large number of derivatizable groups, wide range of molecular weights, varying chemical compositions with low toxicity,[4,5] and are widely used in industrial and agricultural areas as viscosity controllers,[6] drag reducers,[7] flocculants,[8] adsorbents as well as controlled release (CR) devices.[9] However, the main problem associated with these polymers is the extensive swelling. To control these properties, chemical modifications of the structure are needed for their effective use as CR devices for drugs having short plasma half-life. Among the various chemical modifications, carboxymethylation is useful to impart better water solubility to polysaccharides.

Guar gum (GG), a polygalactomannan gum, is a natural polysaccharide, whose chain is made up of D-mannose units with 1–4 linkages, while the D-galactose unit is linked 1–6 on an average to every second D-mannose unit; it has a cyclic neutral structure containing numerous hydroxyl groups (an average of three per sugar unit), which are extracted from the seeds of *Cyamopsis tetragonoloba*.[10–12] Earlier, some reports have been published on drug delivery applications of GG[13–15] and on the structure modification of GG for CR applications.[16–18] For instance, Rubinstein[19] reported the CR systems of GG crosslinked with glutaraldehyde (GA) and phosphate for colon targeting. Soppimath and Aminabhavi[12] studied water transport and drug release characteristics of the crosslinked polyacrylamide-grafted GG hydrogel microspheres. Kumar et al.[20] reported the chitosan–carboxymethyl GG (CMGG)-based interpolymer complexes for colon delivery of fluticasone. Bajpai and Sharma[21] investigated the pH-sensitive swelling and vitamin $B_{12}$ release behavior of barium alginate/CMGG hydrogel beads. Thimma and Tammishetti[22] developed barium- and calcium-crosslinked CMGG beads for gastrointestinal drug delivery of protein to observe that $BaCl_2$-crosslinked beads protect the protein from low pH conditions to deliver in the simulated intestinal fluid.

In this work, we report the preparation of CMGG polymer by inserting carboxymethyl (CM) group into GG, whose structure was characterized by Fourier transform infrared (FTIR) and $^{13}$C-NMR methods.

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Contract grant sponsor: University Grants Commission [UGC; fellowship]; contract grant number: KU/SCH/UGC/RFSMS/2008-09.
Contract grant sponsor: CSIR [award of Emeritus Scientist]; contract grant number: 21(0760)/09/EMR-II.

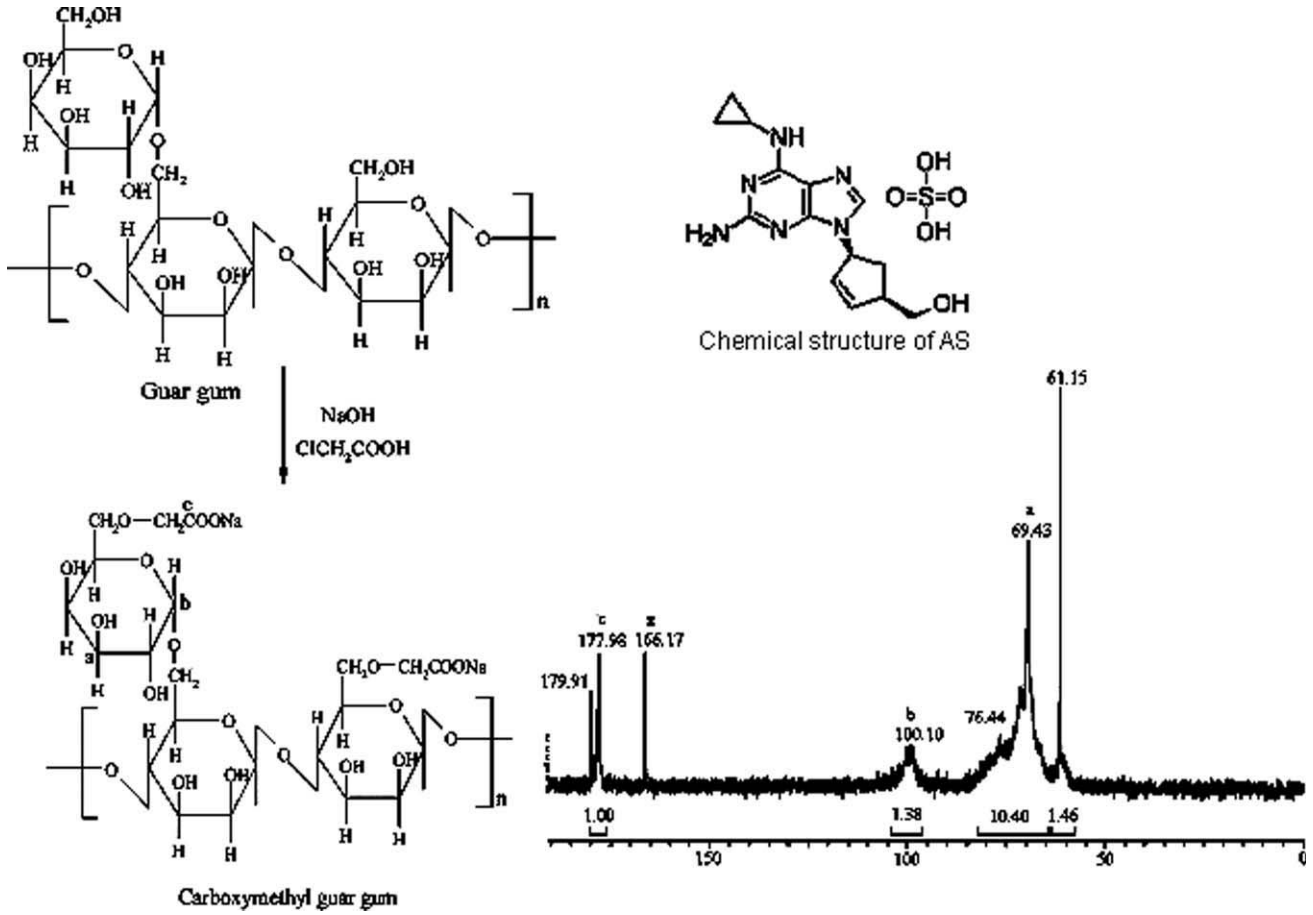


**Figure 1** Synthetic scheme, $^{13}$C-NMR spectrum of CMGG, and chemical structure of AS.

Microspheres were prepared using both GG and CMGG polymers to investigate the CR of abacavir sulfate (AS), an antidepressant drug of the antiviral/reverse transcriptase inhibitor class. Chemically, AS is (1*S*,4*R*)-4-[2-amino-6(cyclopropylamino)-9H-purin-9-yl]-2-cyclopentene-1-methanol sulfate (Fig. 1), which prevents HIV from reproducing in two ways. Metabolite of the drug inhibits the activity of an enzyme needed for the replication of DNA in viral cells. The metabolite is also incorporated into the viral DNA and terminates the formation of DNA. The drug has a plasma half-life of 1.45 h, and no reports on the CR devices of AS using natural polysaccharides are available.

In this research, we prepared the GG and CMGG matrices for extending the release time of short-lived AS in the stomach and intestine, by performing *in vitro* release experiments of the drug in acidic and alkaline pH conditions at 37°C. The kinetics of release data were analyzed using empirical equations.

## EXPERIMENTAL

### Materials and methods

AS was received as a gift sample from the local drug company (a proprietary drug). GG, sodium hydroxide, sulfuric acid, monochloroacetic acid (CAA), analytical reagent grade GA solution 25% (v/v), petroleum ether, and liquid paraffin oil were all purchased from s.d. fine Chemicals, Mumbai, India. The surfactant, Tween-80, was purchased from Loba Chemicals, Mumbai, India. Water used was of high purity grade after double distillation and deionization.

### Synthesis of CMGG

The synthesis of CMGG with various degree of substitution (DS) was reported before[23] and a slight modification was made in this work, wherein 3 g of GG was dispersed in 50 mL of 2-propanol (87 wt %) and stirred for 30 min under nitrogen atmosphere. The resulting suspension was mixed with 10 mL of NaOH solution (5 *N*), and the mixture was allowed to react for 1 h at ambient temperature (25°C). Then a solution of chloroacetic acid (CAA) in 2-propanol with two different concentrations (0.5 and 1 *M*) was added and kept for 1 h to react. Formulations were designated, respectively, as CMGG1 and CMGG2. Temperature of the reaction was raised to 70°C for 2 h; the mixture was cooled to ambient temperature, filtered and the solid was washed with 80% (v/v) methanol solution for 30 min to remove the

**TABLE I**
**Elemental Analysis, Degree of Substitution (DS), Equilibrium Swelling, % Encapsulation Efficiency (EE), and Diffusion Exponent (*n*) Values**

| Formulation codes | Elemental analysis (%) | | | DS | % Equilibrium swelling | % EE | *n* |
|---|---|---|---|---|---|---|---|
| | C | H | O | | | | |
| GG | 38.7 | 6.6 | 54.7 | – | 232 | 51 | 0.53 |
| CMGG1 | 29.1 | 5.6 | 65.4 | 0.11 | 178 | 89 | 0.34 |
| CMGG2 | 29.0 | 5.3 | 65.7 | 0.16 | 160 | 94 | 0.49 |

inorganic salts. The pale yellow colored product was isolated by filtration, washed with 80% methanol, and dried in an oven at 50°C for overnight. The synthetic scheme along with nuclear magnetic resonance (NMR) data is given in Figure 1. The DS of the derivatives was estimated from acidimetric titration.[24]

### Preparation of AS-loaded microspheres

AS-loaded GG, CMGG1, and CMGG2 microspheres were prepared by water-in-oil emulsion method.[12] Briefly, 2% GG solution was prepared in water to which AS was added, stirred until the formation of a homogeneous solution, and acidified with 5 mL of dilute sulfuric acid followed by the addition of 5 mL of GA to crosslink the polymer(s). This solution was then emulsified into 100 mL of light paraffin liquid with 2% (w/v) Tween 80 using a Eurostar high-speed stirrer (IKA Labortechnik, Germany) at 600 rpm speed for 4 h at ambient temperature. The hardened microspheres were filtered and washed repeatedly with *n*-hexane and water to remove the liquid paraffin, unreacted GA, and any adhered Tween 80. The microspheres were dried under vacuum at 40°C for overnight and kept in a desiccator until further use. The same protocol was used to prepare the AS-loaded CMGG1 as well as CMGG2 microspheres.

### $^{13}$C-NMR spectroscopy

$^{13}$C-NMR spectrum of CMGG was recorded on Bruker's 400 MHz NMR spectrophotometer (courtesy of Dr. M. V. Badiger, Scientist, National Chemical Laboratory, Pune, India). The NMR spectrum is shown in Figure 1.

### Fourier transform infrared spectral analysis

Formation of CMGG was confirmed by FTIR taken on GG and CMGG on a spectrophotometer (Nicolet, Model Impact 410). Polymer samples were crushed to make KBr powder into pellets under a hydraulic pressure of 600 kg/cm$^2$ to record the spectra in the wavelength region of 500–4000 cm$^{-1}$.

### Elemental analysis

Elemental analysis for carbon and hydrogen was done using an elemental analyzer Leco (CHN), and the results are included in Table I.

### Thermal analysis

Thermal analysis of GG and CMGG was carried out on a differential scanning calorimetry (DSC)-Q20 (TA Instruments, Waters). Thermogravimetric analysis (TGA) was performed in the temperature range of 25–600°C in an inert atmosphere of nitrogen gas by maintaining a uniform heating rate of 10°C/min.

DSC experiments were performed on placebo GG, AS-loaded GG, placebo CMGG, AS-loaded CMGG, and pristine AS. Samples were heated from 25 to 400°C at the heating rate of 10°C/min in a nitrogen atmosphere.

### X-ray diffraction

Crystallinity of AS after encapsulation was evaluated by X-ray diffraction (XRD) recorded for placebo and AS-loaded CMGG microspheres as well as pristine AS using X-Pert, Philips, UK. Scanning was done at ambient temperature by varying the angle, 2θ up to 50°.

### Scanning electron microscopy

Scanning electron microscopy (SEM) images were taken using JEOL model JSM-840A, Japan instrument available at Indian Institute of Science, Bangalore, India. Particles were sputtered to form a thin gold coating of 10 nm thick to make them conducting. Before the actual measurements, samples were placed on a copper stub and SEM images were taken at different magnifications by applying a voltage energy of 20 kV.

### Swelling experiments

Equilibrium swelling of microspheres at 37°C was measured by determining the weights of samples to measure the extent of swelling in pH 7.4 buffer.[25] To ensure complete equilibrium, samples were allowed to swell for about 24 h and any excess surface-

adhered liquid drops were removed by blotting off with soft tissue papers. The swollen microspheres were weighed to an accuracy of ±0.01 mg on an electronic microbalance (Mettler, AT120, Greifensee, Switzerland), and particles were dried in vacuum oven maintained at 60°C for about 5 h until no further weight gain of the samples was observed. The % equilibrium swelling was calculated as discussed earlier.[25] Experiments were performed in triplicate, but average data are considered in data analysis and these results are included in Table I.

### Encapsulation efficiency

Estimation of drug concentration was done as per the protocol adopted elsewhere.[26] Particles of known weight (~10 mg) were grounded to get the powder using an agate-mortar, extracted with 50 mL of 7.4 pH buffer solution and sonicated for 30 min (UP 400s, Dr.Hielscher, GmBH, Germany). The solution was centrifuged (Jouan, MR23i, France) to remove polymeric debris and washed twice to extract the drug. The solution was centrifuged to remove the suspended polymer particles, and the clear supernatent liquid was diluted with buffer solution. Drug was assayed using UV–VIS spectrophotometer (Model Anthelie, Secomam, France) at the fixed $\lambda_{max}$ of 288 nm. The % encapsulation efficiency (EE) was calculated as described before.[26]

### *In vitro* release experiments

*In vitro* drug release was investigated in 0.1 *N* HCl aqueous solution initially for 2 h, followed by phosphate buffer of pH 7.4 until completion of the dissolution process. Experiments were performed in a tablet dissolution tester (LabIndia, Mumbai, India) equipped with eight baskets (glass jars) at the stirring speed of 100 rpm. Weighed quantity of each sample was placed in 500 mL of dissolution media maintained at the body temperature of 37°C. At regular intervals of time, sample aliquots were withdrawn and analyzed by UV spectrophotometer (Secomam, Anthelie, France) at the fixed $\lambda_{max}$ of 288 nm for the AS. The already withdrawn sample solution was replenished by adding 5 mL of fresh solvent to maintain the sink conditions. Triplicate data were collected and *in vitro* release curves were drawn through the average points, indicating standard deviations of ±3% in all the formulations.

## RESULTS AND DISCUSSION

### Synthesis of CMGG

The carboxymethylation reaction was designed to obtain the GG derivatives for substitution at hydroxyl groups of —$CH_2OH$, as the primary alcohol is more reactive than the secondary alcohol of the anhydroglucose rings. The reaction as confirmed by NMR is shown in Figure 1. To diminish the oxidative degradation of polysaccharide chains, the reaction was carried out in an oxygen-free nitrogen atmosphere. Carboxymethylation reaction was performed in the presence of slight excess of 5 *N* NaOH solution to act as a reactant as well as a catalyst. Both NaOH and GG interacted to form an alkoxide derivative, which then reacted with carboxymethylation agent through substitution reaction, leading to CM substitution along the polysaccharide backbone. The carboxymethylation reaction was carried out at normal pressure and at 70°C. By varying the concentration of carboxymethylation agent (CAA), two grades of CMGG with different DS were prepared, and the results are given in Table I.

### $^{13}$C-NMR spectroscopy

NMR tracings (Fig. 1) confirmed that carbonyl peak of CMGG appears at 177.9 ppm, while the carbon peaks of mannose and galactose appeared in the range of 98–100 ppm. The rest of the side chain carbons of galactose and mannose appeared in the range of 60–80 ppm, confirming the chemical structure of CMGG as given in Figure 1.

### Fourier transform infrared spectral analysis

In case of GG (Fig. 2), a characteristic peak appearing at 3423 $cm^{-1}$ is due to O—H stretching vibrations. The C—H stretching vibrations are observed at 2925 $cm^{-1}$, and additional characteristic bands of GG appearing at 1422 and 1023 $cm^{-1}$ are attributed to C—H and O—H bending vibrations, respectively. Majority of the peaks of derivatized GG almost resemble that of GG. The peak at 2925 $cm^{-1}$ due to —C—H absorption, resulting from the vibrational mode is quite weak in GG, whereas the peak at 2924 $cm^{-1}$ is due to the asymmetric stretching of $CH_2$, and symmetrical stretching of $CH_2$ in CMGG is intense. In case of CMGG, additional band observed at 1094 $cm^{-1}$ is due to C—O—C ether linkage, while sharp bands at 1601 and 1418 $cm^{-1}$ are originated from the resonance of carboxylic groups of CM substituents.[27]

### Elemental analysis

The results of elemental analysis for GG, CMGG1, and CMGG2 given in Table I suggest higher oxygen content in CMGGs than GG, confirming the insertion of CM group into the GG backbone.

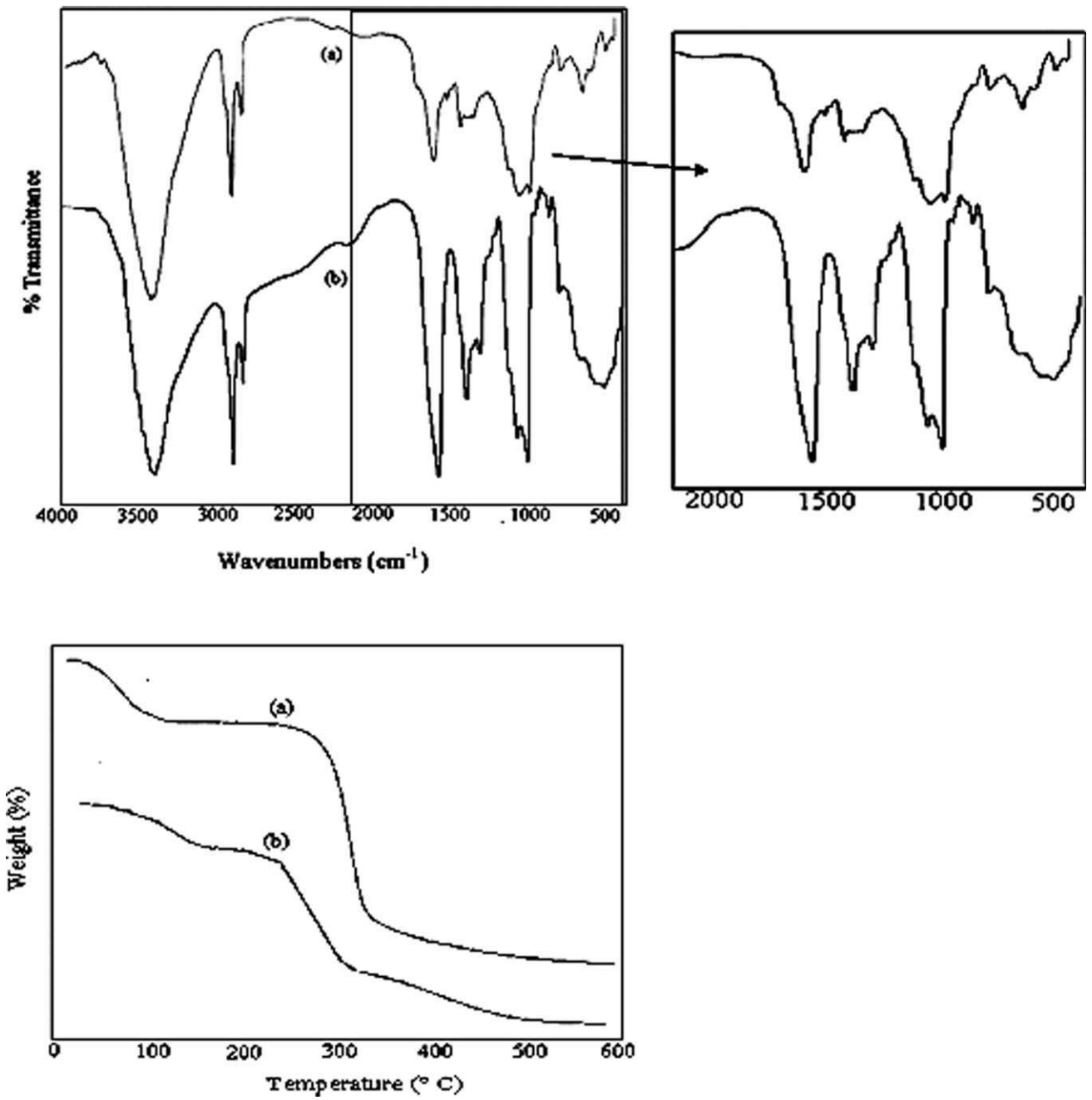


**Figure 2** FTIR and TGA thermograms of (a) GG and (b) CMGG.

### Thermal analysis

TGA curves of GG (Fig. 2) shows two distinct zones of weight loss. The initial weight loss (18%) occurs at 25–115°C, which is due to the trace amount of moisture in the sample. The second zone of weight loss (66%) is observed at 225–350°C due to polymer degradation. TGA curves of CMGG also shows the third zone of weight loss (13%) around 375–525°C, due to degradation of CM groups of the polymer. This third zone of weight loss is present only in CMGG, confirming the insertion of CM groups into GG matrix.

DSC curves of (a) placebo GG, (b) AS-loaded GG, (c) placebo CMGG, (d) AS-loaded CMGG, and (e) pristine AS are shown in Figure 3. Pristine AS shows an endothermic peak at 242°C, which has disappeared in both AS-loaded GG and AS-loaded CMGG formulations, indicating that drug is molecularly dispersed in both GG as well as CMGG polymer matrices.

### X-ray diffraction

XRD diffractograms of (a) placebo CMGG microspheres, (b) AS-loaded CMGG microspheres, and (c)

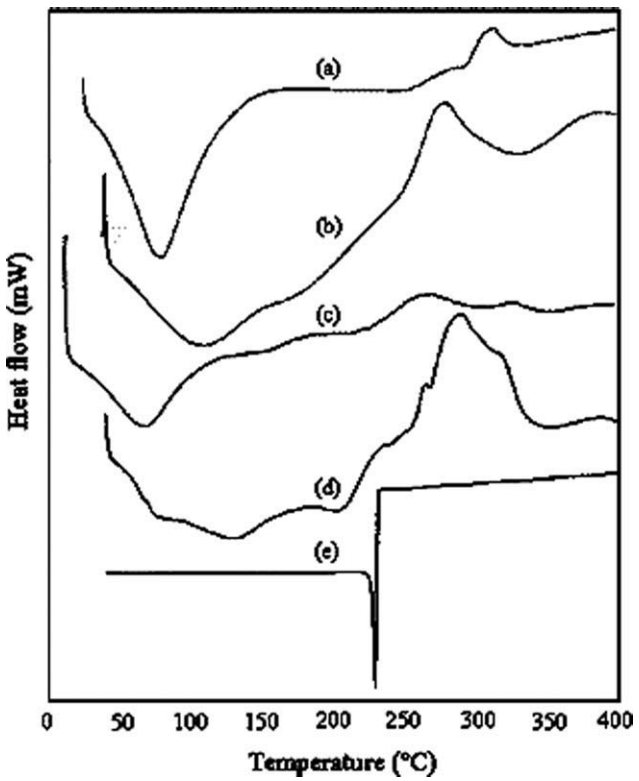


**Figure 3** DSC thermograms of (a) placebo GG, (b) AS-loaded GG, (c) placebo CMGG, (d) AS-loaded CMGG, and (e) pristine AS.

pristine AS presented in Figure 4 suggest the loss of crystallinity of drug after formulation. For instance, diffraction patterns of AS showed many peaks around 2θ of 9–30°, while a high intensity peak at $2\theta = 21°$ represents its crystallinity. However, these peaks have disappeared in the AS-loaded microspheres, but only peaks observed in the placebo polymer are seen, confirming that drug is molecularly dispersed in the polymer matrix and its crystals are not found in drug-loaded formulations.

### Scanning electron microscopy

Typical SEM images of group of AS-loaded CMGG microspheres and their surface morphology are shown in Figure 5(a and b), respectively, from which it is evident that average sizes of spherical microspheres are around 100 μm and also exhibit a rough surface as displayed in Figure 5(b).

### Equilibrium swelling

To investigate the effect of substituted CM group on swelling, we have studied % equilibrium swelling of the matrices in pH 7.4 buffer media at 37°C. As per data presented in Table I, equilibrium swelling is highest (232%) in case of GG, which is obvious due to its hydrophilic nature. However, in case of CMGG1, equilibrium swelling decreased considerably to 178% and a further decrease to 160% in case of CMGG2, suggesting that with increased DS, the matrix becomes less hydrophilic. Also, water-binding site of the CMGG matrix, a dominant factor in swelling, has decreased with increasing degree of CM substitution. This further suggests that higher CM substitution would favor the formation of stronger interaction, resulting in a structure with fewer water-binding sites. Similar observations were also made before by Sen and Pal.[28] This suggests that CMGG is a suitable matrix for encapsulating water-soluble drug such as AS compared to pristine GG.

### Encapsulation efficiency

The EE values also depend on the nature of the matrix as well as the extent of CM substitution onto GG. For instance, EE is highest (94%) for CMGG2, which decreased to 89% for CMGG1, but the least EE value of 51% was observed for GG (see Table I). Thus, it appears that CM substitution onto GG increases the % EE of the matrices, suggesting their better suitability compared to neat GG.

### *In vitro* release

*In vitro* release of AS from GG and CMGG microspheres was investigated in gastric (pH 1.2) media for the initial 2 h, followed by intestinal (pH 7.2) conditions until the completion of dissolution process to understand its release pattern in the stomach as well as in the intestine. Each experiment was performed in triplicate, but results displayed are the average values of the corresponding determinations

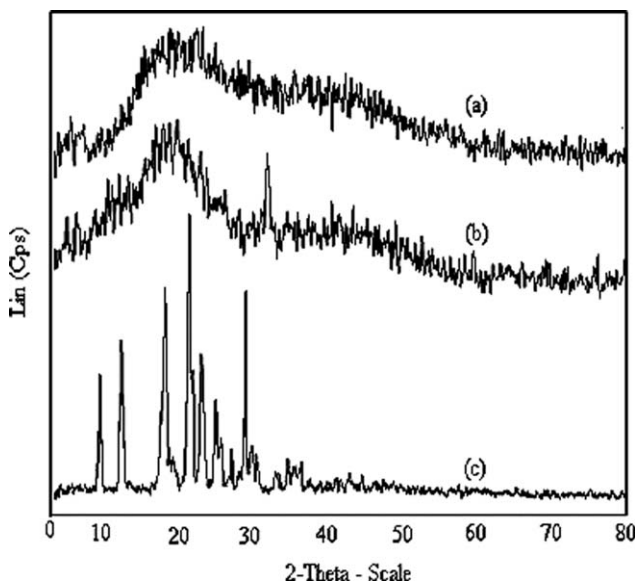


**Figure 4** XRD tracings for (a) placebo CMGG, (b) AS-loaded CMGG, and (c) pristine AS.

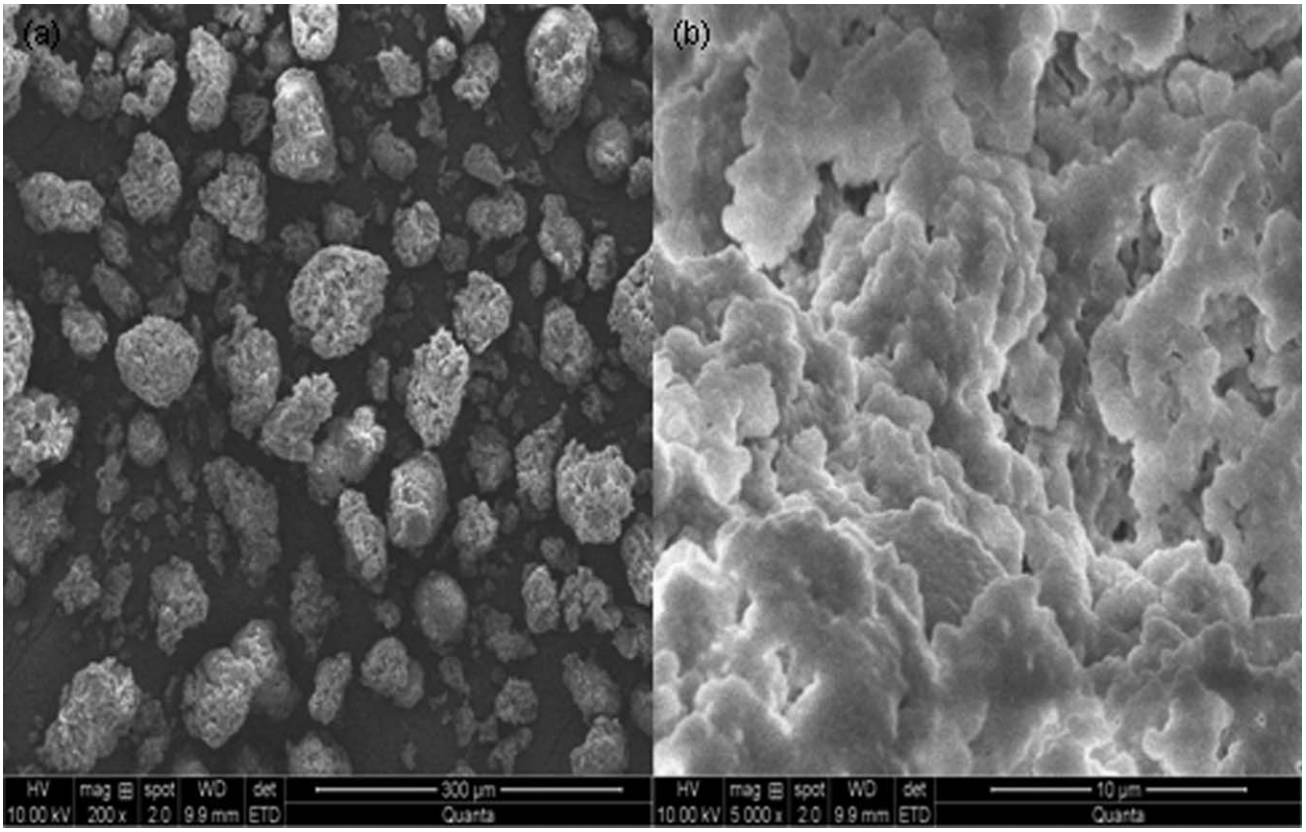


**Figure 5** SEM images of (a) AS-loaded CMGG microspheres and (b) surface morphology of AS-loaded CMGG microspheres.

at each time point. In each case, % cumulative drug release is plotted versus time in case of GG and CMGG matrices shown in Figure 6. It is evident that burst release of AS was observed in case of GG within the first 3 h, followed by a steady release up to 28 h, probably due to the direct exposure of the drug-loaded matrix to the dissolution media, giving a quick release of the drug. However, the *in vitro* release of AS from CMGG1 and CMGG2 did not show such a sudden burst release as that of GG matrix. The release was slowed up to 6 h, which later leveled off with a constant release of AS over an extended period of up to 28 h.

It can be observed from Figure 6 that 58% of AS was released from the GG matrix in 2 h, while for CMGG1 and CMGG2 matrices, only 25% and 18% of drug release occurred, respectively in 2 h. The reason for such a fast release of AS from GG is due to its fast hydrating nature compared to CMGG.[29] Thus, the chemical modification of GG by carboxymethylation has helped to release AS in a slow and steady manner. It can also be notified that % cumulative release of AS decreased at higher carboxymethylation of GG. In any case, the formulation based on GG exhibits higher release than either CMGG1 or CMGG2, and for all formulations, the release was extended up to 28 h. The higher release of drug from the GG matrix compared to CMGG matrices is due to the more hydrophilic nature of GG than CM-substituted GG. Notice that % cumulative release from the GG matrix was >80%, whereas with the

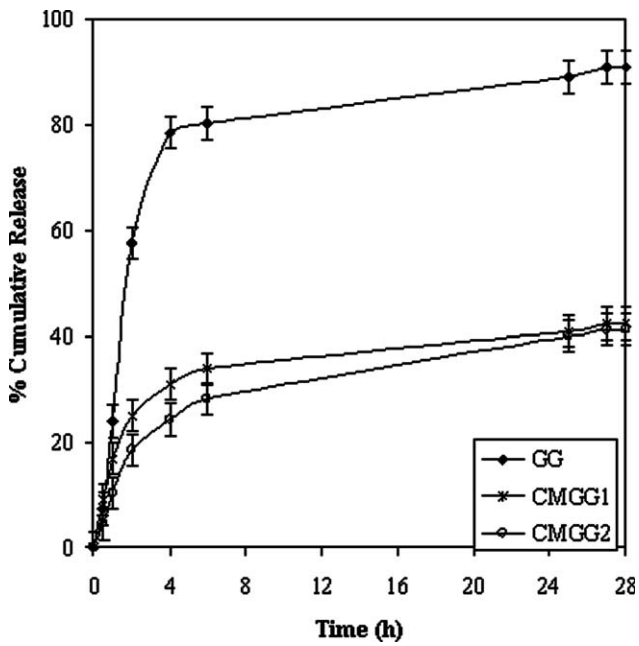


**Figure 6** *In vitro* release of AS-loaded formulations.

**TABLE II**
**Estimated Parameters of Empirical Equations**

| | Zero-order equation, $Q = Q_0 - K_0t$ | First-order equation, $\ln Q = \ln Q_0 - K_1t$ | Higuchi square root equation, $M_t = K_Ht^{1/2}$ | Hixson–Crowell equation, $Q^{1/3} = Q_0{}^{1/3} - K_ct$ |
|---|---|---|---|---|
| Formulation codes | | | | |
| GG | $K_0 = 0.121$ | $K_1 = 0.007$ | $K_H = 0.83$ | $K_c = 0.011$ |
| | $r^2 = 0.518$ | $r^2 = 0.567$ | $r^2 = 0.725$ | $r^2 = 0.550$ |
| CMGG1 | $K_0 = 0.268$ | $K_1 = 0.003$ | $K_H = 1.789$ | $K_c = 0.072$ |
| | $r^2 = 0.620$ | $r^2 = 0.65$ | $r^2 = 0.825$ | $r^2 = 0.64$ |
| CMGG2 | $K_0 = 0.152$ | $K_1 = 0.003$ | $K_H = 0.963$ | $K_c = 0.007$ |
| | $r^2 = 0.776$ | $r^2 = 0.802$ | $r^2 = 0.929$ | $r^2 = 0.793$ |

$M_t$ is the amount of drug released at time $t$; $M_\infty$ is the amount of drug released at infinite time; $Q$ is the amount of drug remaining at time $t$; $Q_0$ is the amount of drug remaining at $t = 0$; $k$, $K_0$, $K_1$, $K_H$, and $K_C$ are the release constants obtained from the linear curves of Korsmeyer–Peppas, zero-order, first-order, Higuchi, and Hixson–Crowell cube root equations, respectively.[34]

CMGG matrices, the extent of drug release was much lower, that is, it varied between 30% and 40%, but the advantage is that no burst release was observed with CMGG matrices.

### Drug release kinetics

To examine the relationship between drug release and molecular transport, cumulative release data were fitted to an empirical equation as analyzed before[30] to estimate the values of diffusion exponent, $n$ at 95% confidence level.[31,32] These data and correlation coefficients are included in Table I. The $n$ values varied from 0.34 to 0.53, suggesting a deviation of transport from the Fickian trend. Different empirical equations, viz., zero order, first order, Higuchi square root, and Hixson–Crowell were also used to analyze the kinetics of *in vitro* release data.[33] However, the criterion for selecting the most appropriate equation was based on the goodness-of-fit test. Evaluated parameters from different equations are presented in Table II. It is observed that the kinetics of release of AS fitted well the Higuchi equation, suggesting the diffusion-controlled process of *in vitro* release of AS from the matrices of this study.

## CONCLUSIONS

CMGG, an anionic semisynthetic GG derivative, was synthesized and its potential was compared with neat GG as a oral drug-delivery device to deliver AS. The polymers were characterized by a variety of techniques, which indicated successful carboxymethylation reaction. Equilibrium swelling and extent of EE strongly depend on the extent of CM substitution. *In vitro* release of CMGG matrices performed in pH 1.2 and 7.4 media demonstrated the CR profiles of AS that avoided the burst release of the matrix, which was observed in case of GG matrix. Analysis of release kinetics revealed that the release of AS from the microspheres followed the Higuchi kinetics, indicating a diffusion CR. *In vitro* release studies indicated that CMGG microspheres effectively extended the release of AS up to 28 h. The *in vitro* release kinetics confirmed the non-Fickian transport trends for the systems of this study.

Miss. A.G. Sullad and Prof. L.S. Manjeshwar thank the University Grants Commission (UGC), New Delhi, India (KU/SCH/UGC/RFSMS/2008-09) for a fellowship to A.G. Sullad. Prof. T.M. Aminabhavi thank CSIR, New Delhi for award of Emeritus Scientist [21(0760)/09/EMR-II].

### References

1. Wu, W.; Aiello, M.; Zhou, T.; Berliner, A.; Banerjee, P.; Zhou. S. Biomaterials 2010, 31, 3023.
2. Shi, J.; Liu, X.; Shang, Y.; Cao, S. J Membr Sci 2010, 352, 262.
3. Coviello, T.; Matricardi, P.; Marianecci, C.; Alhaique, F. J Control Release 2007, 119, 5.
4. Chourasia, M. K.; Jain, S. K. Drug Deliv 2004, 11, 129.
5. Cheung, R. Y.; Ying, Y.; Rauth, A. M.; Marcon, N.; Wu, X. Y. Biomaterials 2005, 26, 5375.
6. Wunderlich, T.; Stelter, M.; Tripathy, T.; Nayak, B. R.; Brenn, G.; Yarin, A. L.; Singh, R. P.; Brunn, P. O.; Durst, F. J Appl Polym Sci 2000, 77, 3200.
7. Bolto, B. A. Progr Polym Sci 1995, 20, 987.
8. Singh, R. P. Encyclopedia of Fluid Mechanics; Houston: Gulf Publishing Inc., 1990, p 425.
9. Sumathi, S.; J Pharm Pharm Sci 2002, 5, 12.
10. Chan, A. W.; Neufeld, R. J. Biomaterials 2009, 30, 6119.
11. Mundargi, R. C.; Agnihotri, S. A.; Patil, S. A.; Aminabhavi, T. M. J Appl Polym Sci 2006 101, 618.
12. Soppimath, K. S.; Aminabhavi, T. M. Eur J Pharm Biopharm 2002, 53, 87.
13. Toti, U. S.; Aminabhavi, T. M. J Control Release 2004, 95, 567.
14. George, M.; Abraham, T. E. Int J Pharm 2007, 335, 123.
15. Li, X.; Wu, W.; Wang, J.; Duan, Y. Carbohydr Polym 2006, 66, 473.
16. Sen, G.; Mishra, S.; Jha, U.; Pal, S. Int J Biol Macromol 2010, 47, 164.
17. Huang, Y.; Yu, H.; Xiao, C. Carbohydr Polym 2007, 69, 774.
18. Thakur, S.; Chauhan, G. S.; Ahn, J. H. Carbohydr Polym 2009, 76, 513.
19. Rubinstein, A. Biopharm Drug Dispos 1990, 11, 465.
20. Kumar, V.; Tiwary, A. K.; Kaur, G. Int J Drug Deliv 2010, 2, 242.

21. Bajpai, S. K.; Sharma, S. J. Macromol Sci Part A 2006, 43, 1513.
22. Thimma, R. T.; Tammishetti, S. J Appl Polym Sci 2001, 82, 3084.
23. Pal, S. J Appl Polym Sci 2009, 111, 2630.
24. Wang, Y.; Yu, Y.; Mao, J. J Agric Food Chem 2009, 57, 10913.
25. Sullad, A. G.; Manjeshwar, L. S.; Aminabhavi, T. M. J Appl Polym Sci 2010, 117, 1361.
26. Sullad, A. G.; Manjeshwar, L. S.; Aminabhavi, T. M. J Appl Polym Sci 2010, 116, 1226.
27. Shin, Ji-Y.; Lee, S.; Bae, I. Y.; Yoo, S. H.; Lee, H. G. J Agric Food Chem 2007, 55, 3368.
28. Sen, G.; Pal. S. J Appl Polym Sci 2009, 114, 2798.
29. Khan, A. B.; Nanjundaswamy, N. G. Arch Pharm Sci Res 2009, 1, 203.
30. Mundargi, R. C.; Shelke, N. B.; Rokhade, A. P.; Patil, S. A.; Aminabhavi, T. M. Carbohydr Polym 2008, 71, 42.
31. Gudasi, K. B.; Vadavi, R. S.; Shelke, N. B.; Sairam, M.; Aminabhavi, T. M. React Funct Polym 2006, 66, 1149.
32. Basu, S. K.; Rajendran, A. Chem Pharm Bull 2008, 56, 1077.
33. Maswadeh, H. M.; Semreen, M. H.; Abdulhalim, A. A. Acta Polon Pharm—Drug Res 2006, 63, 63.
34. Sullad, A. G.; Manjeshwar, L. S.; Aminabhavi, T. M. Ind Eng Chem Res 2010, 49, 7323.