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Scanning electron microscopic analysis and swelling behaviour of ionotropically crosslinked carboxymethylcellulose and carboxymethylcellulose–gelatin matrices

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Water insoluble matrices of carboxymethylcellulose were made by the ionotropic crosslinking of sodium carboxymethylcellulose with cupric or ferric ions. Matrices were also made containing an interactive polymer, gelatin in combination with sodium carboxymethylcellulose. The surface texture of the matrices was analysed by scanning electron microscopy and the dynamic and equilibrium swelling properties of the insolubilised matrices in water were monitored. Micrographs of the controlled release matrices obtained by the incorporation of a larvicide, fenthion in these polymers were also analysed. The crosslinked matrices were highly porous and amorphous and fenthion would act like a plasticiser. Crosslinked matrices of carboxymethylcellulose and carboxymethyl-cellulose–gelatin (10:1 and 4:1) were prepared with three extents of crosslinker densities. The dynamic swelling was noted for about 24 h before attaining equilibrium. Among the cupric ion crosslinked samples, the values of swelling ratios ranged between 1·85 and 2·76 depending on the crosslinker density as well as the gelatin content. The values were in the range of 1·18 and 4·22 for ferric ion crosslinked matrices. The equilibrium swelling ratio was found to have an inverse relationship to crosslinker density and a direct relationship to gelatin content. Ferric carboxymethylcellulose matrix may satisfy the ideal conditions for erosion controlled release systems whereas other matrices may be classified under swellable and erodible matrices. The initial burst effect of fenthion noted from the controlled release matrices may be due to the porous nature and initial influx of water followed by the countercurrent diffusion of the active agent.

INTRODUCTION

Controlled release formulations of pesticides based on the entrapment of the active agent in biodegradable polymer matrices hold great promise for improving the efficacy of some of the low persistent pesticides with reduced environmental impacts associated with their application (Cardarelli, 1976; Kydonieus, 1980; Cross & Scher, 1988). Optimum design or selection of polymers for controlled release systems requires the understanding of the factors related to the structure and morphology of the polymer (Korsmeyer & Peppas, 1981; Langer & Peppas, 1983) and properties related to its interaction with the environment (Kydonieus, 1980). Optimum release conditions can be achieved by controlling the crystalline phase, porous nature, mesh size of the crosslinked macromolecular chain, thermodynamic transitions associated with its interaction with the environment and chemical erosion of the polymer (Langer & Peppas, 1983).

Sodium carboxymethylcellulose (NaCMC), a derivative of cellulose, is an anionic polyelectrolyte. Ionotrpically crosslinked and water insoluble derivatives, cupric carboxymethylcellulose and ferric carboxymethylcellulose were found to be useful as biodegradable controlled release matrices for mosquito larval control agents (Prasad & Kalyanasundaram, 1991, 1992). Controlled release matrices were also prepared with an interactive polymer, gelatin in combination with NaCMC followed by ionotropic crosslinking (Prasad & Kalyanasundaram, 1993*a*).

The analysis of the release data of the larvicide,

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fenthion, from these controlled release matrices indicated an initial 'burst effect' followed by a stabilised release pattern thereafter. The rate of release of an active agent from polymer substrates is influenced by the texture of the controlled release product (Shasha *et al.*, 1976; Korsmeyer & Peppas, 1981; Jeyanthi & Rao, 1990). The surface texture of the products is reported to have more importance on the release profile than their internal structures (Jeyanthi & Rao, 1990). Controlled release of active agents from polymeric matrices which swell continuously in the presence of a penetrant exhibit a constant rate of release as long as the swollen front advances to the glassy region at a constant velocity. The rate of permeation of the solvent into the polymer matrix (dynamic swelling) influences the release rates of the incorporated active agents (Hopfenberg & Hsu, 1978; Peppas & Franson, 1983). The equilibrium swelling ratio of polymers in water was reported to have an influence on the chemical erosion of the polymer and thereby an erosion controlled release mechanism (Heller, 1987).

The present study was, therefore, carried out to analyse the surface texture of the polymers and controlled release matrices with the help of a scanning electron microscope and to monitor the dynamic and equilibrium swelling behaviour of the ionotropically crosslinked carboxymethylcellulose and carboxymethylcellulose–gelatin (10:1 and 4:1) matrices with two gelling agents, cupric or ferric ions at three different extents of crosslinking.

EXPERIMENTAL

Materials

Sodium carboxymethylcellulose was a commercial grade material obtained from Gujchem Distillers India Ltd, Gujarat (India). The estimated molecular weight was 5.496×10^4 (viscosity method) and viscosity of 5% w/v neutral aqueous solution was 140 cps (Haake Viscometer, VT 02, Germany) at 25°C. Bacteriology grade gelatin was obtained from Loba-Chemie, Bombay, India. Inorganic chemicals used were of Analar grade. Technical grade fenthion (*O*, *O*-dimethyl-*O*-4-methylthio-m-tolylphosphorothioate) was obtained from Bayer AG 5090, Leverkusen, Germany.

Methods

Preparation of the samples for scanning electron microscopy

Neutral aqueous slurry of NaCMC (20%) was prepared, spread on a Petri dish and dried at room temperature to constant weight. Similarly, dried sheets of NaCMC containing 20% fenthion, NaCMC–gelatin (4:1) were also prepared. Water insoluble samples were obtained by crosslinking with cupric and ferric ions in the respective 1·0 M solutions of cupric sulphate and ferric chloride by immersing for 24 h at room temperature. The withdrawn samples were further dried at room temperature to constant weight to obtain samples of 2 mm thickness.

Scanning electron micrographs of polymer samples were studied under the electron microscope (Stereoscan-180, Scanning Electron Microscope, Cambridge Instruments, UK). Surface morphology of polymer samples of NaCMC, NaCMC–gelatin (4:1), samples of CuCMC and FeCMC, crosslinked samples containing fenthion (20%) were studied. The samples were mounted on an aluminium stub using quick drying silver paint (Agar Scientific Ltd., UK). The samples were then coated with gold by sputter coating to a thickness of about 10 m. The scanning was carried out at an accelerated voltage of 20–29 kV and 300× magnification.

Preparation of the samples for swelling studies

Dried NaCMC sheet was made from 20% neutral aqueous slurry of NaCMC by uniformly spreading over a Petri dish and allowed to dry in air. The dried sheets of 4 mm thickness thus obtained were cut into samples of 4 × 4 cm size. Polymer sheets were also prepared which contained NaCMC–gelatin in the ratios 10:1 and 4:1. The dried samples thus obtained were crosslinked in 1·0 M solution of the gelling agent, copper sulphate or ferric chloride by immersing each sample in 75 ml of the gelling medium for 12, 24 and 48 h. The withdrawn samples were washed in water and dried in an air oven at 50°C to constant weight.

The swelling behaviour of the polymer samples in water was monitored by volume balance method (Brannon-Peppas & Peppas, 1990). Weights of the samples in air and hexane were noted and the sample volume V was calculated as:

$$V = (W_a - W_h); d_h, \qquad (1)$$

where W_a and W_h are the weights of the polymer in air and in hexane, respectively, and d_h is the density of hexane. Initially the swelling behaviour was noted at hourly intervals and at daily intervals later, until there were no appreciable changes in the weight and volume of the samples.

The swelling ratio, r^* was calculated from the swollen polymer volume, V_s and the volume of the dry polymer, V_p as:

$$r^* = V_s / V_p. \qquad (2)$$

The dynamic swelling ratio characteristics of different samples were presented by plotting swelling ratios against time during the initial stages of swelling and the equilibrium swelling ratios of the samples were noted when there were no appreciable changes in the value of swelling ratio with respect to time.

The equilibrium water intake per gram of the dry polymer samples, W, was calculated as:

$$W = (W_s - W_d)/W_d, \qquad (3)$$

where W_s and W_d are the equilibrium swollen and dry weights of the polymer samples (Walker & Peppas, 1990).

RESULTS AND DISCUSSION

The release mechanism of an incorporated active agent from a polymeric monolithic device is determined by the rate limiting step of the release process. The weekly release profile analysis of the larvicide, fenthion from these formulations showed a high initial release rate of the active agent at the initial periods of the release study (Prasad & Kalyanasundaram, 1991, 1992, 1993*a*). These matrices were also found to swell and undergo erosion when introduced in water.

SEM analysis

The results of the scanning electron microscopic studies carried out to analyse the surface texture of the samples, textural variation with respect to the crosslinking agents, the influence of interactive polymer and interaction of fenthion with the crosslinked matrices are discussed below.

Micrographs of NaCMC and NaCMC–gelatin (4:1)

The scanning electron micrographs of the aqueous solution casted NaCMC and NaCMC–gelatin (4:1) are presented in Fig. 1. The micrographs show that the surface texture of NaCMC (Fig. 1a) is crystalline and the crystalline nature is lost by the addition of the interactive polymer, as a result of the strong electrostatic interaction between the macromolecular chains leading to agglomeration (Fig. 1b).

Micrographs of crosslinked samples

The micrographs of CuCMC and FeCMC samples obtained by the crosslinking of NaCMC with cupric and ferric ions, respectively, are shown in Fig. 1c and d. The micrographs show that the surface texture of both CuCMC (Fig. 1c) and FeCMC (Fig. 1d) are amorphous and porous. Textural differences are noticed between

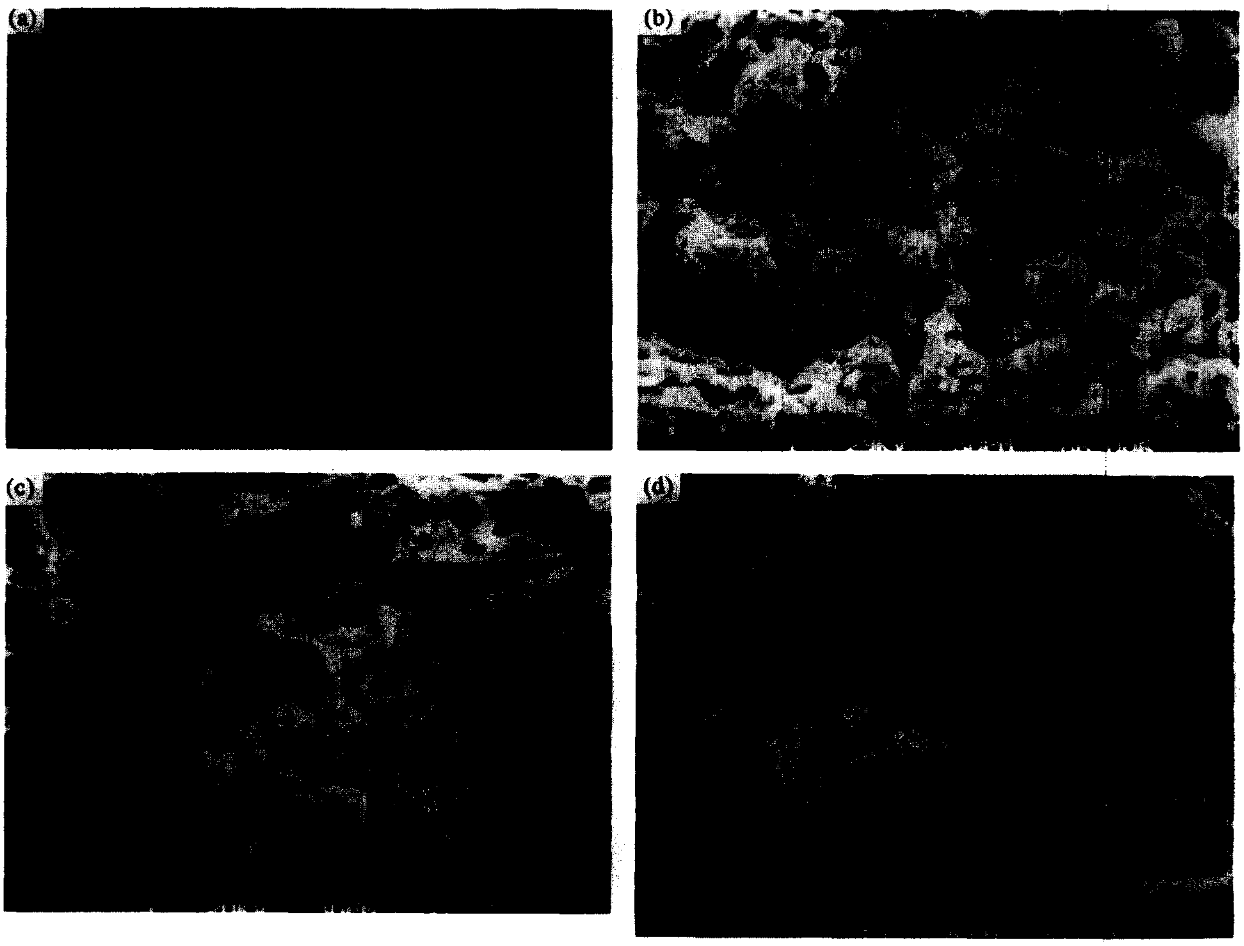

Fig. 1. (a) Scanning electron micrograph of : (a) sodium carboxymethylcellulose; (b) sodium carboxymethylcellulose–gelatin; (c) cupric carboxymethylcellulose; (d) ferric carboxymethylcellulose.

the two samples. The CuCMC matrix was more porous than FeCMC.

Micrographs of crosslinked samples containing fenthion

The crosslinked matrices containing 20% fenthion were also subjected to scanning electron microscopic analysis to determine the interaction between fenthion and polymer matrices. The micrographs (Fig. 2a, b) show that the active agent, fenthion, is uniformly distributed and interacts with the polymer matrices like a plasticiser. Differences are noticed between the matrices prepared with two crosslinking agents.

Micrographs of crosslinked samples containing gelatin and fenthion

The surface micrographs obtained with the samples containing the interactive polymer gelatin and the larvicide fenthion are presented in Fig. 2c and d. The micrographs show that these two matrices are amorphous and their surface textures are very different from the micrographs of those matrices without the interactive polymer, gelatin.

Analysis of the micrographs show that the surface texture of solution casted NaCMC matrix has been altered by the incorporation of the interactive polymer, gelatin. The ionotropic crosslinking used for insolubilising NaCMC has also resulted in textural changes. The micrographs obtained with the matrices containing fenthion show that fenthion may act as a plasticiser, thereby reducing macromolecular interactions.

Dynamic and equilibrium swelling behaviour

The dynamic swelling properties of the samples crosslinked for 12, 24 and 48 h in 1·0 M copper sulphate are presented in Figs 3, 4 and 5, respectively, for the matrices of CuCMC, CuCMC–gelatin (10:1) and CuCMC–gelatin (4:1). The swelling ratios of the samples increased rapidly during the initial 24 h and the increase was relatively slow thereafter till the equilibrium swelling ratio was reached. The values of the swelling ratios at specific time intervals were found to be influenced by the duration of crosslinking and the gelatin content in the matrices. The values of the dynamic swelling ratios were found to vary substantially when the ferric ion crosslinked matrices were studied (Figs 6, 7 and 8). The

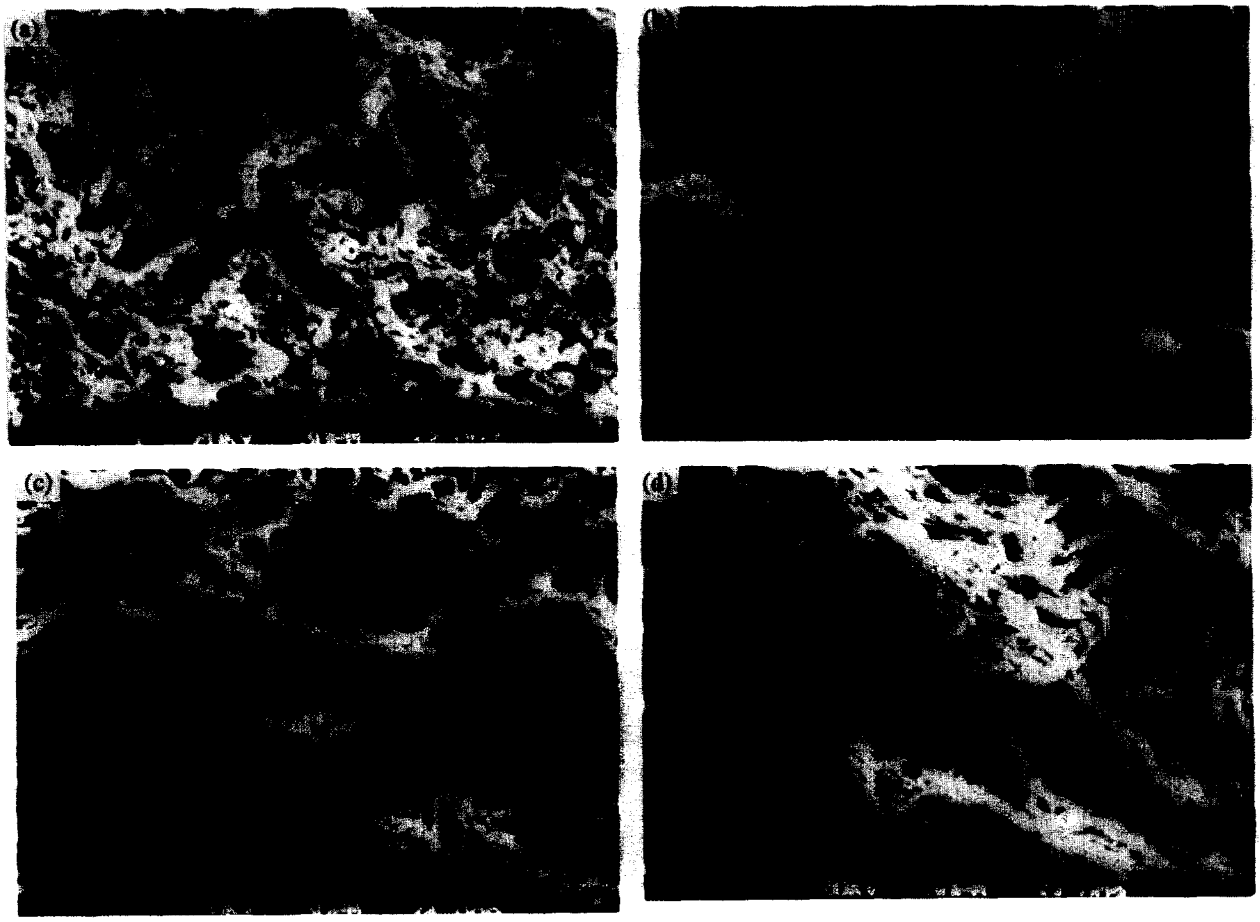

Fig. 2. Scanning electron micrograph of: (a) cupric carboxymethylcellulose–fenthion; (b) ferric carboxymethylcellulose–fenthion; (c) cupric carboxymethylcellulose–gelatin–fenthion; (d) ferric carboxymethylcellulose–gelatin–fenthion.

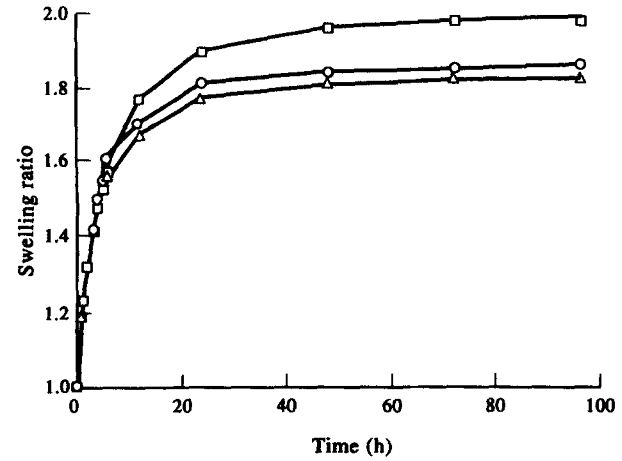

Fig. 3. Dynamic swelling behaviour of NaCMC samples crosslinked in 1.0 M copper sulphate for 12 (□), 24 (○) and 48h (△).

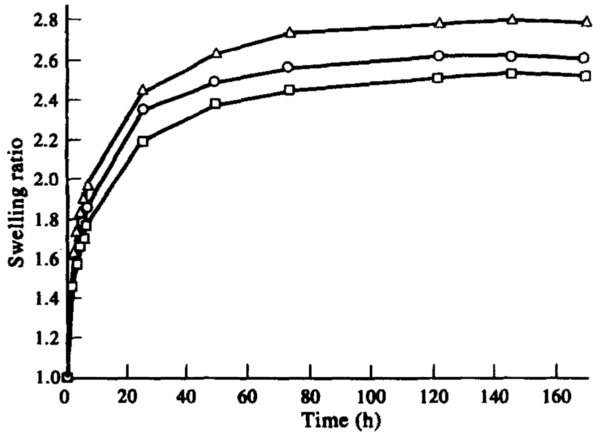

Fig. 5. Dynamic swelling behaviour of NaCMC–gelatin (4:1) crosslinked in 1.0 M copper sulphate for 12 (□), 24 (○) and 48h (△).

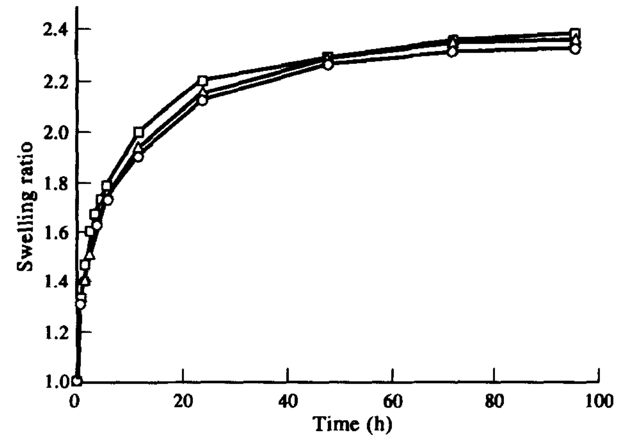

Fig. 4. Dynamic swelling behaviour of NaCMC–gelatin (10:1) samples crosslinked in 1.0 M copper sulphate for 12 (□), 24 (○) and 48h (△).

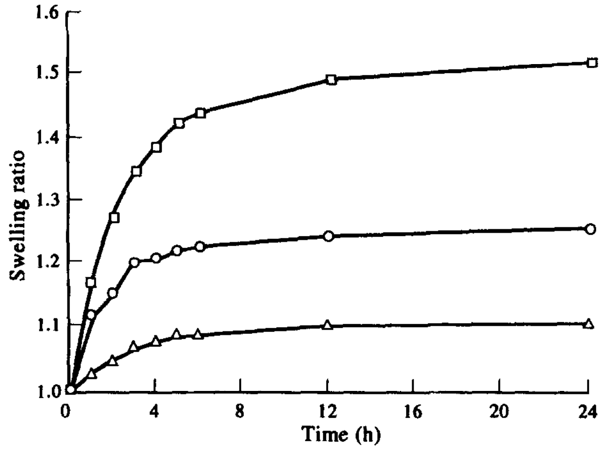

Fig. 6. Dynamic swelling behaviour of NaCMC samples crosslinked in 1.0 M ferric chloride for 12 (□), 24 (○) and 48h (△).

dynamic swelling ratios were inversely proportional to the crosslinking duration of the matrices and directly proportional to the gelatin content.

The values of the equilibrium swelling ratios of the insolubilised samples are presented in Fig. 9. Among the cupric ion crosslinked samples, samples obtained by crosslinking NaCMC matrices exhibited lower values compared with those contained the interactive polymer, gelatin. Ferric ion crosslinked matrices of CMC exhibited minimum values of the equilibrium swelling ratios than the rest of the matrices and those containing gelatin exhibited the highest values compared with the respective matrices obtained under identical conditions of crosslinking.

The equilibrium water uptake per gram of the dry polymer samples are presented in Fig. 10. Cupric ion crosslinked samples exhibited values which ranged between 0·74 and 3·19 whereas with ferric ion crosslinked matrices the values were between 0·29 and 4·71.

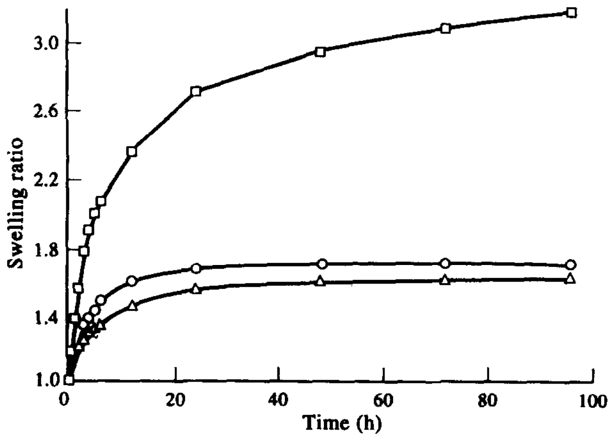

Fig. 7. Dynamic swelling behaviour of NaCMC–gelatin (10:1) samples crosslinked in 1.0 M ferric chloride for 12 (□), 24 (○) and 48h (△).

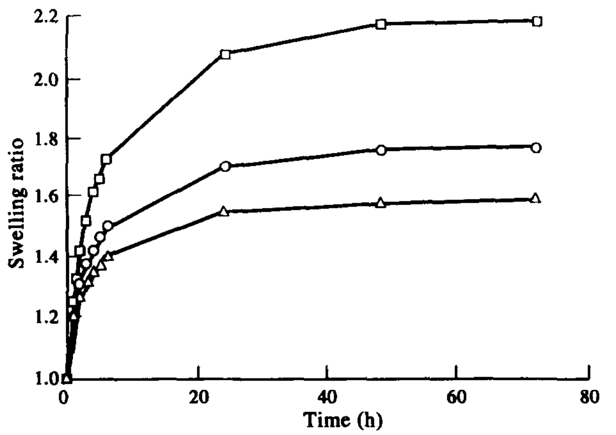

Fig. 8. Dynamic swelling behaviour of NaCMC–gelatin (4:1) samples crosslinked in 1.0 M ferric chloride for 12 (□), 24 (○) and 48h (△).

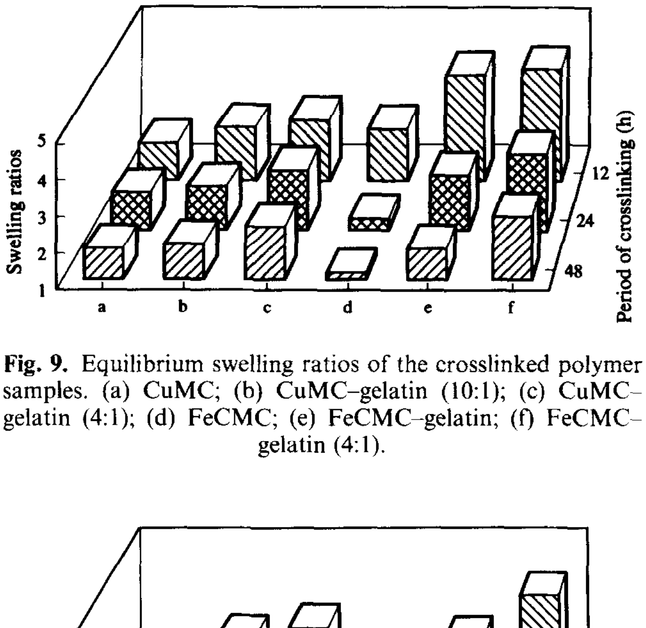

Fig. 9. Equilibrium swelling ratios of the crosslinked polymer samples. (a) CuMC; (b) CuMC–gelatin (10:1); (c) CuMC–gelatin (4:1); (d) FeCMC; (e) FeCMC–gelatin; (f) FeCMC–gelatin (4:1).

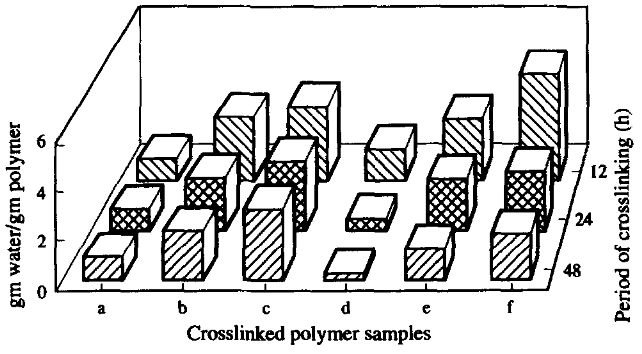

Fig. 10. Equilibrium water uptake per gram of the crosslinked polymer samples. Samples as in Fig. 9.

The equilibrium water intake was found to decrease with increase in crosslinking duration and values were directly proportional to gelatin content.

When a polymer comes into contact with any medium which is thermodynamically compatible, the solvent penetrates into free space between the macromolecular chains resulting in an increase in radius of gyration and end to end distance of the polymer molecules and is observed macroscopically as swelling (Rao & Padmalatha Devi, 1988). The dynamic and equilibrium swelling behaviour of hydrogels is determined by the hydrophilicity, nature and extent of the crosslinking agent. The interaction of the matrices with water suggests that initially water molecules form hydrogen bonding with the hydroxyl groups of cellulose. As the water content increases, some of the hydrogen bonds weaken and the hydration of the cation is made possible and in the final stage water is absorbed to reach the equilibrium swelling. The hydrophilic nature of gelatin may be responsible for the higher values of the swelling ratios observed with the matrices containing gelatin.

In conclusion, the high initial release rate of fenthion observed with these matrices may be responsible for the relatively high porous nature coupled with the initial influx of water followed by the countercurrent diffusion of the active agent. The matrices obtained by ionotropic crosslinking of NaCMC with cupric or ferric ions were relatively hydrophobic and studies on the swelling behaviour in water indicated that FeCMC matrix was very hydrophobic and the presence of gelatin enhanced its hydrophilicity. The ionotropic crosslinking was found to be reversible resulting in erosion of the crosslinks followed by matrix erosion at later periods (Prasad & Kalyanasundaram, 1993*b*). FeCMC matrix may satisfy the ideal conditions for the erosion controlled release systems whereas other matrices may be classified under swellable and erodible matrices (Langer & Peppas, 1983). The controlled release of various low molecular weight and water soluble biologically active agents from these matrices may be effected by permeation of water followed by countercurrent diffusion of the active agent during the initial periods and later the release is initiated by crosslink erosion followed by erosion of the matrices. These matrices can be utilised for the controlled release of various pesticides of agricultural and public health importance with minimum impact on the environment as the matrix material is biodegradable.

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