CHAPTER 5

Drug encapsulation into polymeric nanoparticles.

Contents

Drug encapsulation into polymeric nanoparticles synthesised through emulsion polymerisation of MMA – HEMA.

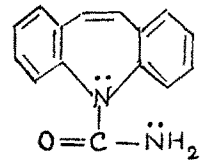
[5.1] Introduction

One of the primary objectives in the design of novel drug delivery systems is controlled delivery of the pharmacological agent to its site of action at a therapeutically optimal rate and dose regimen [1, 2]. This site – specific or targeted delivery combined with delivery at an optimal rate will not only improve the efficacy of the drug but would also reduce the possibility of unwanted toxic side effects of the drug thus improving the therapeutic index [3]. Among the most promising systems to achieve this goal are the colloidal drug delivery systems. Colloidal drug delivery systems include the drug carrier systems liposomes, niosomes, nanoparticles and microemulsions. Liposomes, niosomes, nanoparticles and microemulsions are very similar in their size, shape and mode of administration, and for this reason may be used alternatively. Nevertheless, these systems have a number of different advantages and disadvantages. The great advantage of liposomes, for instance, is that their main components are lecithins and in most cases, cholesterol. Since they exist in the body in large amounts good bio-acceptability may be expected. Nanoparticles, on the other hand, possess a better stability due to their rigid structure. One of the methods to prepare nanoparticles is emulsion polymerisation of a suitable system. It offers the advantage of reduction in particle size and thus enables an use in intravenous injection. Little and Parkhouse [4] have demonstrated that a reduction in particle size of the polymeric particles can minimise the possible irritant action at the injection site. Moreover, Stinson [5] showed that the cancerogenic effects are not related to the chemical nature of materials but the particle size. One of the essential prerequisite of a successful nanoparticle system is to achieve

higher percentage entrapment of the drug molecule into the nanoparticle. Since the present work is directed to oil – in – water kind of system oil soluble model drug **carbamazepine** was selected to attain high % entrapment with SDS as surfactant. The oil MMA – HEMA system was selected since it provides a broad range of microstructure after polymerisation and biocompatibility of HEMA in addition to the solubility of the drug in the co monomer mixture.

[5. 2] Structure of the Drug and Use

Carbamazepine [5H – dibenzazepine – 5 – carboxamide] is an iminostilbene derivative with a tricyclic structure having very little solubility in water but freely soluble in ethanol , dichloromethane and acetone. It is an antiepileptic drug widely used for treatment of simple and complex partial seizures and trigeminal neuralgia. Carbamazepine is slowly absorbed in the liver and is distributed erratically following oral administration. It enters the brain rapidly due to its higher lipid solubility. It reduces the propagation of abnormal impulses in the brain by blocking sodium channels, thereby inhibiting the generation of repetitive action potentials at the epileptic focus.



[5. 3] Nanolatex Synthesis through Emulsion Polymerisation

The emulsion system comprising of MMA – HEMA as oil , SDS as surfactant and water was initiated with redox initiator H_2O_2 – Ascorbic acid [1: 1mole] at 40 ^{0}C. The oil

soluble drug carbamazepine was dissolved in the monomer prior to emulsification. The amount of drug dissolved was 5% based on monomer. The polymerised latex was made free from the surfactant by cooling below the Kraft temperature, which is reported to be 16 ^{0}C for SDS. This procedure removes bulk of the surfactant. Remaining adsorbed surfactant was removed by repetitive washing with hot water. The resulting polymer was dried under vacuum at 50 ^{0}C for two days. [2] The alternative procedure for removal of surfactant involves precipitation of the latex in methanol. Thereafter the surfactant was removed by repetitive washing of the polymer.

[5 . 4] Drug Estimation

In order to estimate the amount of drug entrapped, the calibration plot in ethanol was constructed. The drug showed maximum absorbance at 276 nm and was free from any interference due to drug polymer interaction. In order to increase the % entrapment the reaction conditions were optimised with respect to variation in method of purification of the polymer particles, cross linker concentration and comonomer ratio.

[5 . 4. 1] Method of purification

Precipitation of the polymer in methanol resulted in lower % entrapment of the drug in polymer matrix and is given in column A of Table 5.1. Whereas cooling below Kraft temperature followed by repeated washing resulted into better entrapment and is given in column B of Table 5.1. This can be explained on the basis of the solubility of the drug in methanol.

Table 5.1 : Effect of comonomer ratio, cross linker concentration and method of purification on the % entrapment of the drug in the nanoparticle.

Co monomer ratio [MMA/HEMA]	% Entrapment [A]	% Entrapment [B]	Effect of cross linker concentration		
			1%	2%	3%
90 / 10	61.2	69	72	73	73
80 / 20	63.2	74	76.4	78	79
70 / 30	65	78	80	84.4	85

[5. 4. 2] Effect of comonomer ratio

HEMA is completely soluble in water whereas MMA has only partial solubility in water. This offers a wide range of microstructure after polymerisation. Moreover increase in HEMA in the comonomer can be advantageous due to its biodegradability. Therefore the co monomer ratio was varied from 90 / 10, 80 / 20 to 70 / 30. Increase in HEMA % above 30 leads to phase separation. SEM shows more or less discrete spherical polymer particles at 90 / 10 [MMA / HEMA] [Fig 5.1]. This changes to a more or less porous structure at 70 / 30 ratio. Increase in HEMA content leads to a greater degree of homogeneous nucleation in aqueous phase resulting into some coagulation and greater connectivity in the polymeric matrix. This results into a more closed structure and hence higher entrapment was observed at 70 / 30 compared to 80 / 20 and 90 / 10.

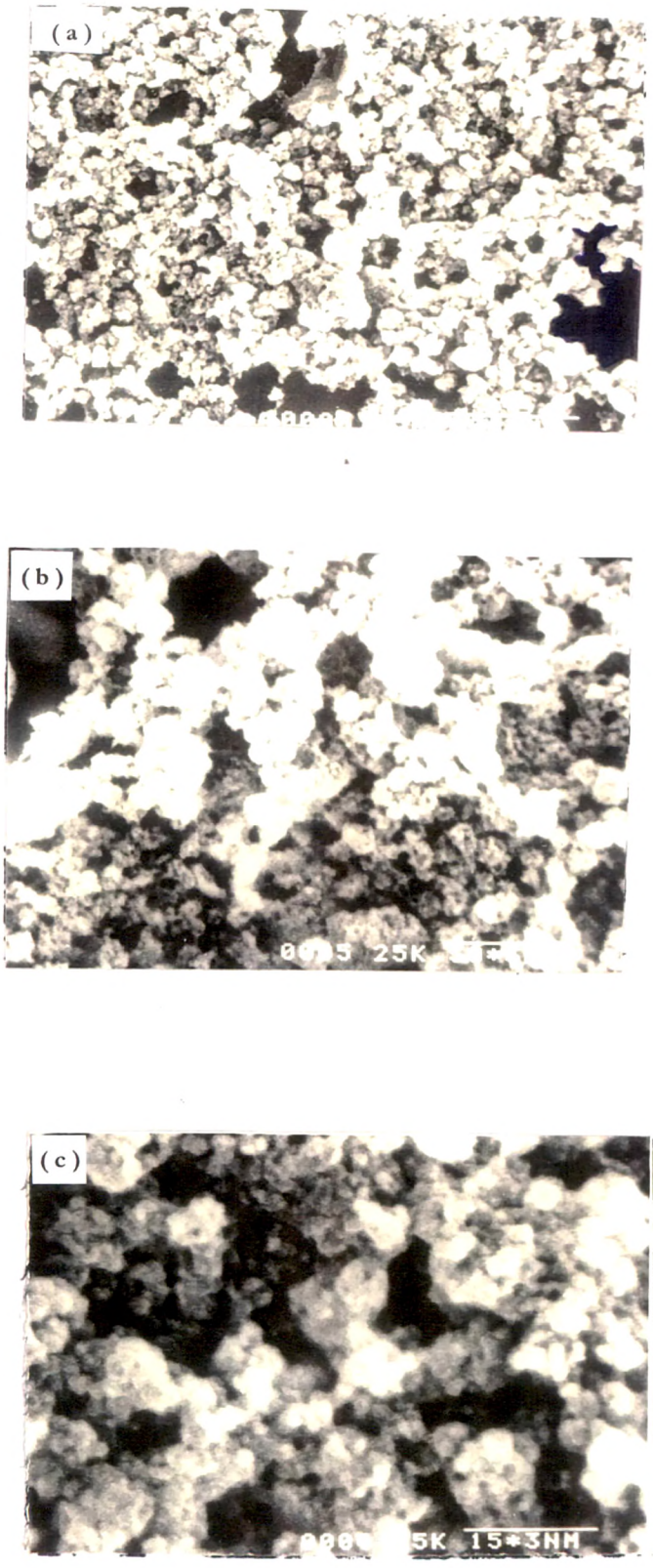


Fig 5 . 1 : SEM shows the variation in copolymer microstructure on increasing the % of HEMA in feed. [a] 90 / 10 (MMA : HEMA) [b] 80 / 20 (MMA : HEMA) [c] 70 / 30 (MMA : HEMA).

[5 .4 .3] Effect of crosslinker concentration

Cross linker ethylene glycol dimethacrylate [EGDMA] was used at 1, 2 and 3 % concentration to increase the rigidity of polymer matrix resulting into an increase in % entrapment of the drug [Table 5.1].

The particle size of the polymerized latex for 90 / 10 [MMA / HEMA] system was analyzed by dynamic light scattering. Incorporation of the drug resulted into an increase in particle size up to 100 nm from 78 nm without the drug. The particle size distribution after drug incorporation is given in Fig 5. 2. IR analysis further confirmed the drug entrapment in the polymeric matrix. The necessary peaks are discussed in the characterization chapter [Fig 5.3 and 5.4].

[5 . 5] In vitro Drug Release

According to US pharmacopia 23, the dissolution medium reported for the study of release profile of carbamazepine is 1 % SDS solution. Since the emulsion had been initially formulated with 1 % SDS, the reaction mixture was used as such for the study of drug release at various time intervals at pH of 7.2 . The release pattern over a period of 60 hrs showed a slower release initially due to very little solubility of the drug in water [Fig 5. 5]. A faster diffusion of the entrapped drug molecules for 90 / 10 [MMA : HEMA] compared to 70 / 30 [MMA : HEMA] might be due to a more discrete structure of the polymeric matrix resulting into a greater specific surface area of the polymer exposed to the reaction medium.

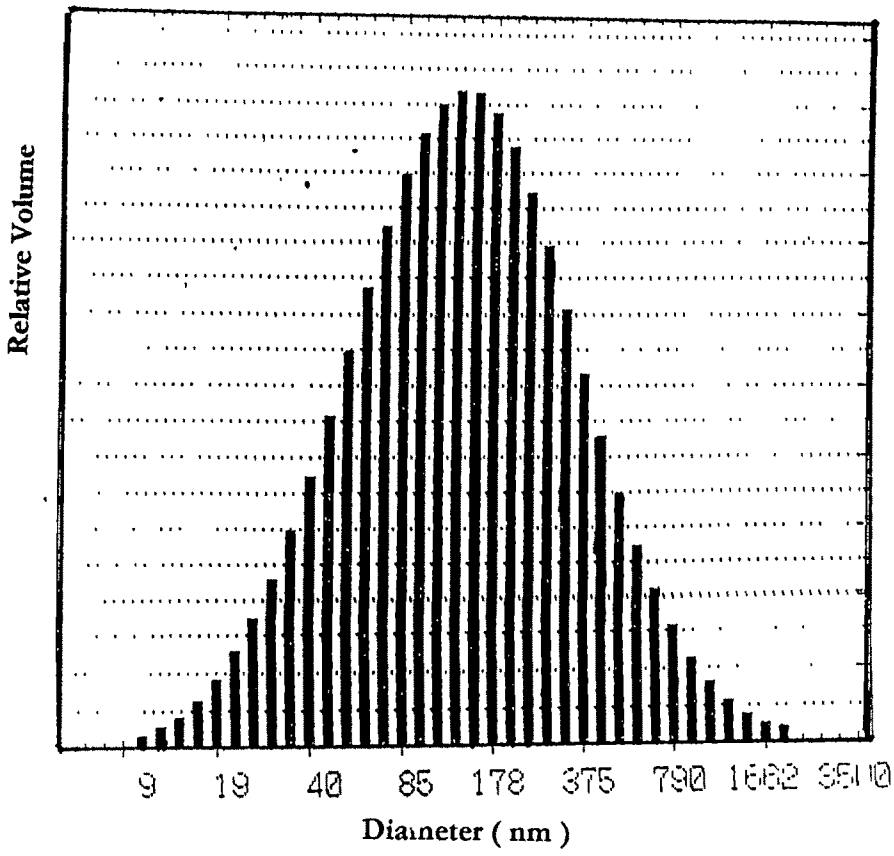


Fig 5 . 2 : Particle size distribution of MMA – HEMA containing the entrapped drug.

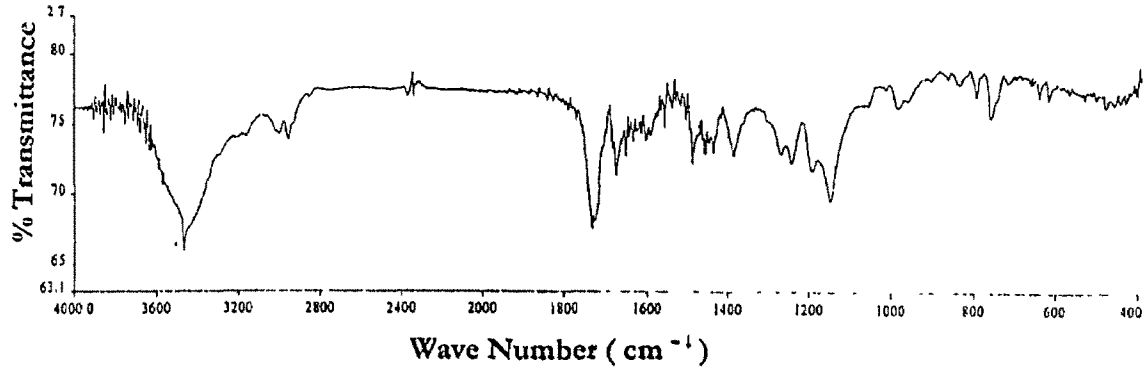


Fig 5.3 : IR spectra of MMA – HEMA copolymer containing the entrapped drug.

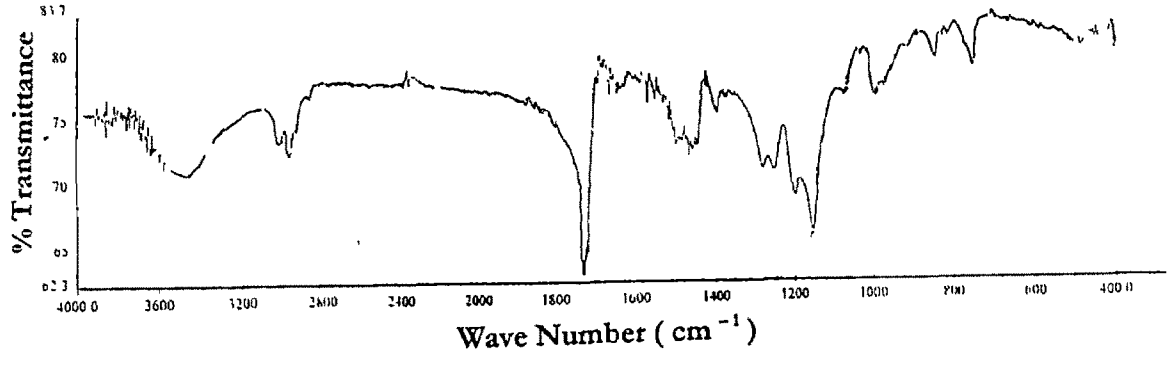
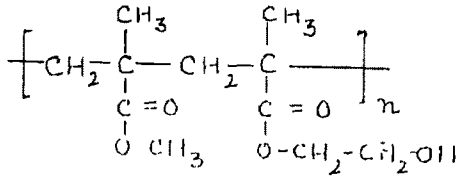


Fig 5. 4 : IR spectra of MMA – HEMA copolymer.



MMA HEMA

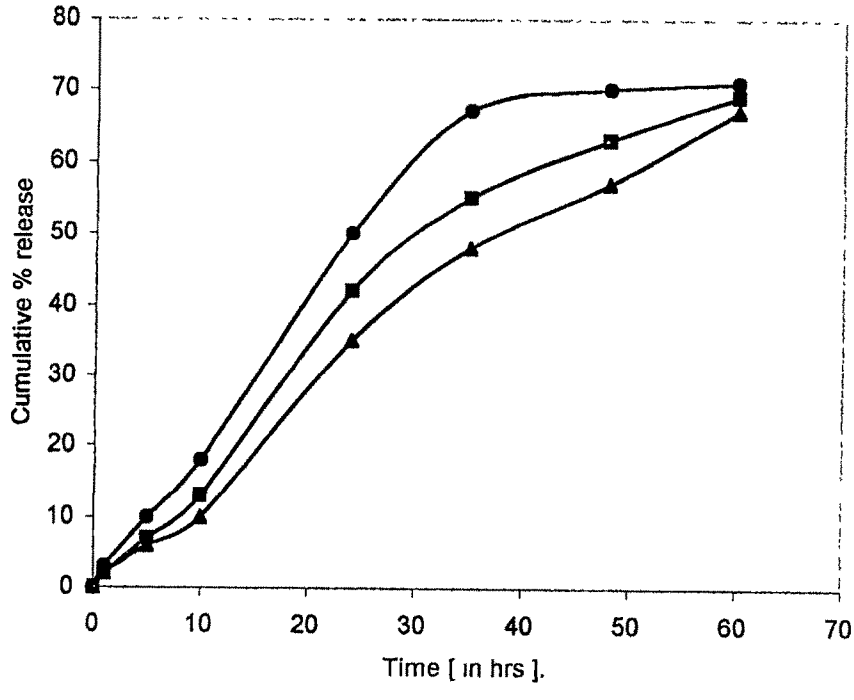


Fig 5. 5 : Release pattern of the of the drug from the polymeric network over a period of 70 hrs at pH 7. 2.
(▲) 70 /30 MMA : HEMA , (■) 80 / 20 MMA : HEMA ,
(●) 90 /10 MMA : HEMA.

[5 . 6] Characterisation

[5 . 6 . 1] SEM analysis

The polymer samples were analysed by ISI SX – 40 Scanning Electron Microscope (SEM) to study the pore morphology with variation in MMA · HEMA ratio. Prior to analysis, the polymerized samples were coated with gold with uniform thickness of 2 mm. The necessary details are discussed in the text. [Section : 5 . 4 . 2].

[5 . 6 . 2] Particle size analysis

Particle size of the polymer particles synthesized through emulsion polymerisation measured using Malvern 4700 instrument in dynamic mode was found to be 78 and 100

nm for a 90 / 10 [MMA : HEMA] sample before and after the drug entrapment. Increase in particle size was observed on incorporation of the drug. Drug incorporation was further confirmed by IR analysis.

[5 . 6 . 3] IR analysis

Infrared spectrum of the synthesised polymers in a KBr pellet was recorded on a Perkin Elmer FT-IR spectrophotometer RX1.

IR spectra shows $\rangle C{=}O$ [str] due to both MMA and HEMA units in the copolymer at 1734 cm ^{1}. Whereas the $\rangle C = O$ [str] due to amide group from carbamazepine arises at 1676 cm $^{-1}$ [amide 1] and-NH_2 (def.) [amide II] at 1620 cm^{-1}. A broad peak around 3676 cm ^{1} arises due to – OH [str] from HEMA. While a sharp absorbance at 3466 cm^{-1} is due to $\rangle N - H$ [str] for carbamazepine. CH_3, CH_2 [Str] vibration appeared around 2953 cm^{-1}. Peak assignments were made by checking the reference spectra of MMA – HEMA copolymer and the drug.

[5 . 7] Conclusion

1. Increase in % of HEMA in the comonomer mixture changed the polymer microstructure from a more discrete to a complete network like structure with a corresponding increase in the % entrapment of the drug. Increase in % of the cross linker made the polymeric matrix more rigid resulting into slower diffusion of the drug molecule and an increase in drug entrapment.
2. In vitro release studies carried out in 1 % SDS solution shows a slower release of the drug initially.

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