
Preparation, Characterization and *In Vitro* Release Kinetics of Ibuprofen Polystyrene Microspheres

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In the present study, an attempt has been made to develop polystyrene microspheres for controlled drug delivery. The microspheres were prepared by solvent evaporation technique with different polystyrene-ibuprofen ratio (1:1, 1:2 and 2:1). The microspheres were characterised by loading efficiency, percentage of entrapment, percentage of encapsulation, particle size distribution, scanning electron microscopy, infrared spectroscopy, differential scanning calorimetry and *in vitro* release studies. Depending upon drug polymer ratio, the loading, entrapment, and encapsulation was found to be 30-52 %, 84-90 % and 70-80 % respectively. The spheres were spherical, discrete, compact and free flowing. The particle size of microspheres was found to be in the range of 4-25 μm . Drug loaded spheres were found to be larger than unloaded one. Infrared spectrum and thermogram revealed the stability of ibuprofen in microspheres. The microspheres released ibuprofen more than 24 h. Depending on drug polymer ratio, polystyrene microspheres are able to deliver 30 to 80% of the loaded drug by the end of 24 h. The microspheres prepared with 1:2 polymer drug ratio shows better release pattern and control the drug release over a period of 24 h. The release followed Higuchi kinetics rather than first order kinetics, indicating diffusion controlled drug release.

Polystyrenes constitute one of the major class of polymers that has wide spread application¹⁻³. The earliest interest in the medical uses of polystyrene microspheres was to calibrate measuring instruments¹ such as the electron microscope and the coulter counter. Later these mono disperse particles were used in various serological tests¹. For example, diagnosis of rheumatoid arthritis, disseminated lupus erthematous and human pregnancy. Covalently bound antibodies on polystyrene spheres have also been used to locate cell surface antigens using scanning electron microscope. Piskin *et al.*² reviewed medical and biological applications of monosize beads based on polystyrene and their modified forms. Jani *et al.*³ described the disposition of polystyrene microspheres after oral and parenteral administration in male Wistar rats.

Although polystyrene microspheres are used in various biomedical applications, the development and application as a drug delivery system is relatively a new area. However a meagre amount of research work has been carried out in this area. Crosswell and Becker⁴ encapsulated paracetamol in polystyrene beads for sustained drug delivery. El-Gibaly *et al.*⁵ encapsulated ketoprofen by using cellulose acetate butyrate and polystyrene for sustained drug delivery. Polystyrene is a biocompatible^{6,8}, non-biodegradable polymer and has been used as drug carrier in implants and targeted drug delivery system. Tamilvanan and Sa⁹ studied *in vitro* release behaviour of indomethacin loaded polystyrene microparticles.

The objective of the present work is to study the feasibility of polystyrene microspheres for oral controlled drug delivery. Very few articles are published in this area and data are not available regarding the *in vitro* release kinetics

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of polystyrene microspheres. We attempted to develop polystyrene microspheres containing a model drug ibuprofen, as a drug delivery system by using emulsification solvent evaporation technique. The microspheres were characterised by loading efficiency, entrapment efficiency, encapsulation efficiency, optical microscopy, scanning electron microscopy, particle size analysis, FT-IR spectroscopy, differential scanning calorimetry and *in vitro* drug release. The data obtained from *in vitro* release were fit into various kinetic models to study the release mechanism and release kinetics. Ibuprofen is a non-steroidal antiinflammatory agent, which possesses analgesic and mild antipyretic action. Because of its shorter biological half-life and hazards of bronchospasm/gastric irritation, it is an appropriate candidate for controlled drug delivery.

MATERIALS AND METHODS

Polystyrene (Gppstryng-general purpose, marketed by Greaves Limited, Mumbai.) was obtained from Southern Polymers, Chennai. Polyvinyl alcohol (Lobachemie, Mumbai) was used as obtained. Ibuprofen was obtained from Cassel Research Laboratories, Chennai and used as obtained. All the other chemicals used were of analytical grade.

Preparation of polymer-drug solution:

Polymer solution was prepared by adding 1.2 g of polystyrene to 30 ml of dichloromethane in a 50 ml beaker. The beaker was closed and the contents were stirred with magnetic pellet for 6 h at room temperature to dissolve polystyrene. To this solution, required quantity of ibuprofen was added and dissolved by stirring as per the formula given in Table 1.

Preparation of 4% w/v polyvinyl alcohol (PVA) solution containing 0.1% w/v Tween 80:

Polyvinyl alcohol solution was prepared by adding 9.6 g of PVA to 240 ml of water. Then the contents were stirred and heated (90 to 100°) to get a solution, after cooling the volume was corrected to 240 ml with water. To this 240 mg of tween 80 was added and mixed well.

Preparation of microspheres:

Polyvinyl alcohol solution (240 ml) was taken in a 500 ml beaker. To this 30 ml of polystyrene-ibuprofen solution was added drop by drop. The mixture was stirred (approximately at 500 rpm) with help of magnetic pellet to get a stable emulsion. The stirring was continued (at minimum speed) for 12 h at 50 to 60° to get microspheres. The microspheres formed were collected by filtration by using Whatman filter paper (No. 542, pore size 2.7 µm). The filtered microspheres were washed with 3x20 ml of phosphate buffer (pH 7.4) in order to remove unencapsulated ibuprofen and polyvinyl alcohol. Finally, the microspheres were washed with 2 x 20 ml of water and dried at 40° for 24 h to remove residual solvents. The same procedure was followed to prepare three more batches of polystyrene microspheres with different ibuprofen content. The amount of polystyrene and ibuprofen used were given in the Table 1.

Determination of drug content in microspheres:

The amount of ibuprofen present in polystyrene microspheres was determined by hot extraction method. The ibuprofen from the microspheres was extracted¹⁰ with phosphate buffer (pH 7.4) for 60 min by refluxing at 100°. It was then cooled to room temperature and filtered. Suitable dilu-

TABLE 1: SHOWING THE CHARACTERISTICS OF POLYSTYRENE MICROSPHERES

Batch no	Polymer/ drug ratio	Amount of poly styrene used (g)	Amount of ibuprofen used (g)	Yield (g)	Percentage loading		% Entrap- ment	% Encapsu- lation	Average particle size (µm)
					Theoreti- cal	Actual			
1	2:1	1.2	0.6	1.6	33.5	30	89.6	80	13.7
2	1:1	1.2	1.2	2.0	50	42	84	70	14.9
3	1:2	1.2	2.4	3.2	66.6	52	76.5	70	15.6
4	1:0	1.2	—	0.8	—	—	—	—	6.7

Each data shows average of three determinations.

tion of the filtrate was prepared and estimated¹¹ at 228 nm using UV/Vis spectrophotometer (Shimadzu 2100 S).

Determination of drug loading and encapsulation efficiency:

The ibuprofen loading in microspheres was estimated using the formula, $L = Q_m/W_m \times 100$, where L is the percentage loading of microspheres, W_m is the weight of microspheres in g and Q_m is the quantity of ibuprofen present in W_m g of microspheres. The amount of ibuprofen encapsulated in the microspheres was determined using the formula, $E = Q_p/Q_t \times 100$, where E is the percentage encapsulation of microspheres, Q_p is quantity of drug in g encapsulated in microspheres, Q_t is quantity of drug in g added for encapsulation and Q_p is the product of drug content per g of microspheres and yield of microspheres in g.

Particle Size Analysis and Scanning Electron Microscopy (SEM):

Particle size analysis was carried out by using optical microscopy¹². About 200 microspheres were selected randomly and their size was determined using optical microscope fitted with a standard micrometer scale. SEM analysis of microspheres was done using a Jeol JSM 5300 scanning electron microscope.

FT-IR and Differential Scanning Calorimetric (DSC) Analysis:

Infrared spectra of polystyrene microspheres loaded with ibuprofen were taken by using carbon tetrachloride as solvent and were recorded on a Bomem MB-II FT-IR spectrometer. DSC scans of ibuprofen and polystyrene microspheres loaded with ibuprofen were performed using Perkin-Elmer DSC-7 model calibrated with indium.

***In vitro* release studies:**

The *In vitro* release studies of drug loaded microspheres were carried out at 37° using phosphate buffer pH 7.4. Microspheres containing 25 mg of ibuprofen was added to 250 ml of phosphate buffer pH 7.4 in Erlenmeyer flasks¹³. The flasks were shaken (60 oscillations/min) in an incubator (Remi make) at 37°. One ml of samples was withdrawn at regular time intervals and same volume of phosphate buffer was replaced. After suitable dilution ibuprofen content in phosphate buffer (pH 7.4) was estimated at 228 nm by using UV/Vis (Shimadzu 2100 S) spectrophotometer.

Release kinetics:

Data obtained from *in vitro* release studies was fitted to

various kinetic equations to find out the mechanism of ibuprofen release from polystyrene microspheres. The kinetic models used were a zero order equation¹⁴, first order equation¹⁵ and Higuchi release¹⁶. The following plots were made: Q_t vs. t (zero order kinetic model); $\log(Q_0 - Q_t)$ vs. t (first order kinetic model) and Q_t vs. square root of t (Higuchi model). Where Q_t is the amount of drug released at time t and Q_0 is the initial amount of drug present in microspheres.

RESULTS AND DISCUSSION

We have tried different types of emulsifying agents such as polyvinyl alcohol, carboxy methylcellulose, tween 80, sodium lauryl sulphate, gelatin, and acacia in order to make a stable dispersion of dichloromethane globules containing ibuprofen and polystyrene in water. A solution of 4% w/v of polyvinyl alcohol and 0.1% w/v tween 80 was found to be the effective emulsifying system. This was used to prepare polystyrene microspheres. The obtained microspheres were discrete, free flowing and spherical. The selection of the emulsifying agent is an important factor for obtaining spheres of uniform size without aggregation. Tween 80 was found to reduce aggregation among polystyrene microspheres. Minimum speed was used to prepare microspheres in order to prevent foam formation on the surface of emulsion, which may influence the rate of evaporation of dichloromethane and formation of microspheres.

We have tried organic solvents like dichloromethane, trichloromethane, xylene, and alcohol based on solubility parameters to extract ibuprofen from polystyrene microspheres, but hot extraction with phosphate buffer was found to be simple and reproducible. Since the softening point of polystyrene is 98°, it is possible to get complete extraction of ibuprofen. In order to find out any drug degradation might have taken place during extraction of ibuprofen from microspheres, a standard ibuprofen solution was prepared and exposed to extractive conditions. The quantity of ibuprofen present was compared with unexposed standard solution. No drug decomposition was observed and ibuprofen was found to be stable during the extraction process.

Higher percentage of loading was obtained by increasing the amount of ibuprofen with respect to polystyrene. Nevertheless, the percentage of entrapment was decreased in case of higher drug loading (Table 1). This indicates the wastage of drug in higher proportions. The encapsulation process was found to be good and 70 to 80% of the drug employed in the process was encapsulated by the microspheres. The percentage of encapsulation was higher (80%) in 2:1 polymer to drug ratio. This may be due to pres-

ence of higher amount of polystyrene with respect to ibuprofen.

The particle size of ibuprofen loaded microspheres were analysed by optical microscopy. All the batches of microspheres show uniform size distribution as shown in fig. 1. The average particle size of unloaded microspheres was smaller than the drug-loaded sphere. The particle size of unloaded microspheres was found to be in the range of 4-16 μm and ibuprofen loaded was found to be in the range of 4-24 μm . The prepared microspheres had good spherical geometry as evidenced by the SEM photographs. Microspheres with high drug content shows some irregular

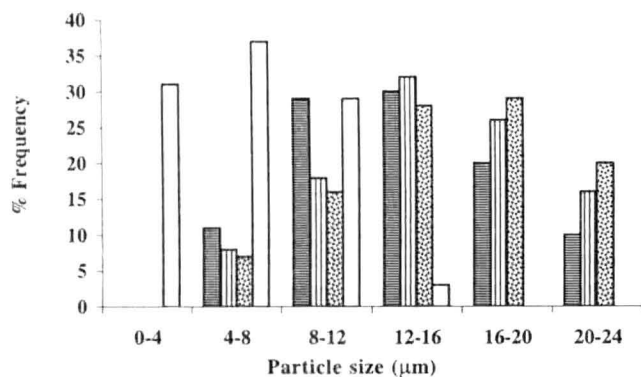


Fig. 1: Size distribution of polystyrene microspheres. Particle size distribution of polystyrene/ibuprofen microspheres prepared with different ratios, 2:1 [▨], 1:1 [▩], 1:2 [▤] and unloaded microspheres [□].

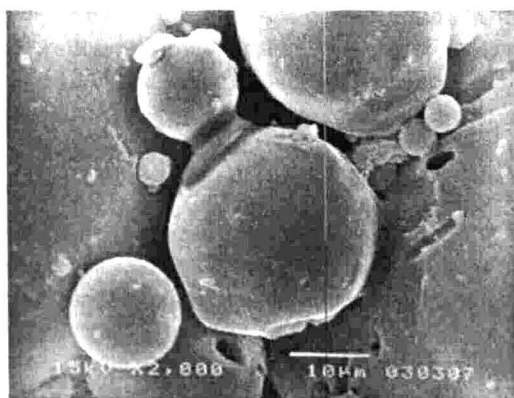


Fig. 2: SEM photomicrograph of ibuprofen-loaded polystyrene microspheres. SEM photomicrographs of a) showing spherical nature of polystyrene microspheres and b) showing the drug present on the surface of microspheres.

shapes may be due to adsorption of drug on the surface of microspheres. Unloaded spheres were smaller than the loaded ones. The microspheres were discrete, spherical and uniform. SEM analysis indicates the smooth external surface of the microspheres with spherical geometry and is compact in nature as seen in fig. 2a and 2b.

Infrared spectrum of ibuprofen loaded polystyrene microspheres showed characteristic absorption peaks¹⁷ of carboxylic acid groups at 1705 cm^{-1} and identical with the reference spectrum given in Indian Pharmacopoeia. This clearly indicated the stability of drug during microencapsulation process and revealed the absence of drug polymer interaction. This was further supported by DSC studies. DSC of ibuprofen showed a sharp endothermic peak at 74.1°. Ibuprofen in the polystyrene microspheres also showed similar characteristic peak (fig. 3) in comparison with pure drug, indicating crystalline nature of drug in microspheres. Evaporation of solvent during the microsphere preparation results in concentration of ibuprofen above the saturation and induces formation of ibuprofen crystals in microspheres. The intensity of the drug peak is increased as the drug loading is increased, this may be due to increase in degree of ibuprofen crystallinity in the microspheres at higher drug loading.

The cumulative percent release of ibuprofen from different batches is shown in fig. 4. In all batches, the release was very fast for the first 3 h. Then the release was slow from the 3 h and sustained over 24 h, depending upon polymer/drug ratio. By the end of 24 h release studies, 1:2, 1:1, and 2:1 (polymer/drug) polystyrene microspheres released

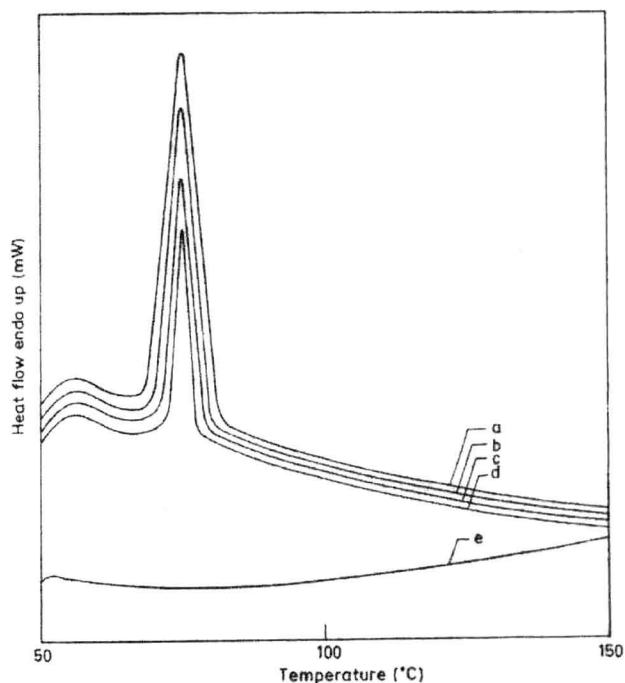


Fig. 3: Thermogram of polystyrene microspheres. Thermograms of ibuprofen (a), microspheres prepared with polystyrene/ibuprofen ratios of 1:2 (b), 1:1 (c) 2:1 (d) and that of polystyrene alone (e).

82, 69, and 30.1% of loaded drug, respectively. The microspheres 2:1 and 1:1 (polymer/drug) could not able to release above 30.1 and 69% of its loaded drug, respectively. This may be due to hydrophobic and plastic nature of the polymer polystyrene. The polymer drug ratio 1:2 shows better loading, encapsulation efficiency and release pattern. It controls the drug release over 20 h and was found to be the most suitable among other batches prepared in this work for oral controlled drug delivery.

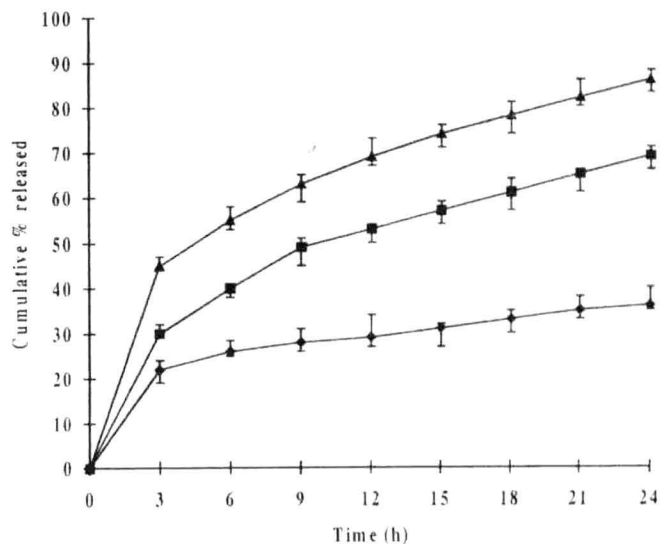


Fig. 4: *In vitro* release of ibuprofen from polystyrene microspheres.

Dissolution profiles of ibuprofen from polystyrene microspheres (n=3, mean±SD) prepared with polystyrene/ibuprofen ratio 2:1 (-●-), 1:1 (-■-) and 1:2 (-▲-).

The *in vitro* release data were applied to various kinetic models to predict the drug release mechanisms. The best fit with highest correlation coefficients (r^2) was shown (Table 2) by the Higuchi rather than first order and zero order models. The plot of cumulative percentage release versus square root of time produces high linearity. This clearly indicates that the release followed Higuchi model. Furthermore, a plot of log percentage release against log time (Table 2) revealed¹⁸ a high level of linearity, which conforms ibuprofen release from polystyrene microsphere, is diffusion-controlled.

TABLE 2: COEFFICIENTS AND RELEASE RATE OF IBUPROFEN FROM POLYSTYRENE MICROSPHERES

Batch no	Polymer drug ratio	Time for 25 % release (h)	Time for 50 % release (h)	First order model $\log(Q_0 - Q_t)$ vs. t		Higuchi model Q_t vs. square root of t		Log $c/\log t$
				r^2	Release rate (k_1) h^{-1}	r^2	Release rate (k) $h^{-1/2}$	r^2
1	2:1	12	-	0.962	0.722	0.983	12.87	1
2	1:1	1.5	12	0.989	0.432	0.995	11.98	0.978
3	1:2	0.5	5	0.998	0.140	0.999	4.32	0.978

Where r^2 is correlation co-efficient. The release rate constants k_1 and k was calculated from the slopes of the respective plots.

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