

FORMULATION AND EVALUATION OF INDOMETHACIN MICROSPHERES FOR COLONIC DRUG DELIVERY SYSTEM

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ABSTRACT

Colon specific drug delivery has gained increased importance not just for delivery of drug for the treatment of local diseases, associated with the colon but also potential site for systemic delivery of therapeutic drug. Indomethacin is non-steroidal anti-inflammatory drug with short half life.

Indomethacin Microspheres were prepared by solvent evaporation method using Eudragit L-100 and Eudragit S-100 mixture [1:2, 1:3, 1:4, and 1:5] were used as pH-sensitive polymers. The prepared Microspheres were evaluated for various physicochemical parameters such as particle size, percentage yield, incorporation efficiency, drug polymer compatibility (IR study), scanning electron microscopy (SEM) and drug release of Microsphere (pH 6.8) for 12 hrs. Result shows that as the concentration of polymer increases it affects the particle size, percentage yield and drug release of micro capsules. The flow properties were found to be good and scanning electron microscopy confirmed their hollow structure with smooth surface. Formulation F1 shows excellent particle size, percentage yield (90.24%), incorporation efficiency (95.59%) and percentage drug release (95.58%) for a period of 12 hrs. Results of our present study suggest that Indomethacin Microspheres can be successfully designed to develop sustained drug delivery, which can improve compliance by reducing dosing frequency.

Keywords: Colon specific, Eudragit, Microspheres, Indomethacin.

INTRODUCTION

The word new or novel in the relation to drug delivery system is a search for something out of necessity. An appropriately designed sustained or controlled release drug delivery system can be major – advance towards solving the problem associated with the existing drug delivery systems^{1, 2}. In recent year, colon targeted drug delivery of both conventional and labile molecule is developed³. It is a drug delivery site for the treatment of local disease, associated with colon like crohn's disease, ulcerative colitis, irritable bowel syndrome, also it is a potential site for systemic delivery of therapeutic drugs^{4,5}. Colon is also found to be a promising site when delay in absorption is desirable from therapeutic point of view for the treatment of disease that have peak symptoms in early morning and that exhibit circadian rhythm, such as rheumatoid arthritis (RA), nocturnal asthma and angina pectoris^{6,7}. In case of rheumatoid arthritis, peak symptoms occur early in the morning due to the imbalance between anti-inflammatory effect by cortical and pro-inflammatory effects exerted by melaton⁸. The widely used approaches for colon specific targeting are bacterially triggered, pressure controlled, pH dependent and time dependent control drug delivery system³. The pH of colon is 6.5 and pH of intestine (7.00

to 8.00)⁹. Most commonly used pH dependent coating polymers such as Eudragit (L100/S100) which dissolves at pH 6.00 and 7.00 respectively.

Hence, none of these polymers are suitable to be used, alone for coating of dosage forms that would start release the drug at pH 6.5, although this has been generally accepted as the desired pH for colon targeted delivery³. Indomethacin [1, 1 – biphenyl] – 4-acetic acid, 2-fluro-alpha-methyl, is a important analgesic and non-steroidal anti-inflammatory drug (NSAID) also with anti-pyretic properties whose mechanism of action is the inhibition of prostaglandin synthesis. It is used in the therapy of rheumatoid disorders. Indomethacin is rapidly eliminated from the blood, its plasma elimination half-life is 3-6 hours and in order to maintain therapeutic plasma levels. The drug must be administered approximately 150-200mg daily by oral individual dosage¹⁰. The aim of this work was to develop Microspheres of Indomethacin by solvent evaporation technique. Indomethacin whose physicochemical properties and short half life make it suitable candidate for colonic drug delivery system.

MATERIALS AND METHODS

Indomethacin was obtained from Micro Lab Ltd. (India). pH sensitive meth acrylic acid co-polymers (Eudragit® L-100 and S- 100) were supplied as gifts by Degussa India Pvt. Ltd., Mumbai (India). Heavy liquid paraffin, Acetone, petroleum ether was obtained from S.D. fine Chem. Ltd., Mumbai (India), Span 80 were supplied from Ioba, Chemical, India, Ethanol were supplied from Jiangsu Huaxi International Trade Co. Ltd.(Made IN China), All other chemicals and reagent used in this study were of analytical grade.

Method of preparation

Microspheres were prepared by solvent evaporation method. Accurately weighted Eudragit L-100 and S-100 in different ratios were dissolved in 50ml of acetone to form a homogenous polymers solution. Core material, i.e. Indomethacin was dispersed in it and mixed thoroughly. This organic phase was slowly poured at 15°C into liquid paraffin (100 ml) containing 1% (w/w) of Span-80 with stirring at 1000 rpm to form a uniform emulsion. Thereafter, it was allowed to attain room temperature and stirring was continued until residual acetone evaporated and smooth-walled, rigid and discrete Microspheres were formed. The Microspheres were collected by decantation and the product was washed with petroleum ether (40– 60°C), four times and dried at room temperature for 3 hrs. The Microspheres were then stored in a desiccators over fused calcium chloride.

Evaluation of indomethacin microspheres

Micromeritic properties

Particle size:

Determination of average particle size of the Indomethacin Microspheres was carried out by the optical microscopy method. A minute quantity of Microspheres was spread on clean glass slide and average sizes of 100 Microspheres were determined in each batch.

Angle of repose:

Determination of angle of repose Indomethacin Microspheres were carried out by employing fixed funnel method.

Angle of repose $\theta = \tan^{-1} (H/R)$

H = Height of the pile

R = Radius of the pile

Percentage yield

The measured weight was divided by total amount of all non-volatile components which were used for the preparation of microsphere.

% yield = Total weight of excipient and drug / Actual weight of product x 100

Drug content uniformity

In 100ml volumetric flask 25mg of crushed microspheres were taken and dissolved with small quantity of ethanol and the volume was made up to mark with pH 6.8 and stirred for 12 hrs. After stirring the solution was filtered through whatman filter paper and from the filtrate appropriate dilutions were made and absorbance was measured at 247 nm by using Shimadzu 1700 UV spectrophotometer.

Infrared spectroscopy (FT-IR)

FT-IR spectra of the Indomethacin, F1 formulation, Eudragit L-100, Eudragit S-100 using KBr pellet technique, Samples were scanned over the 500-4000 cm^{-1} Spectral region at a resolution of 4 cm^{-1} . The ratio of the sample in KBr disc was 1% (shimadzu FT-IR spectrometer).

Scanning electron microscopy

The samples for SEM analysis were prepared by following method. Dry Microspheres brass stub an coated with gold in an ion sputter. Then picture of Microspheres were taken by random scanning of the stub. The SEM analysis or the Microspheres was carried out by using JEOL-6360A analytical scanning electron microscope (Physics Department pune University, pune (India)). The Microspheres were viewed at an accelerating voltage of 20KV.

Drug release

In vitro release studies: *In vitro* dissolution profile of each formulation was determined by employing USP XXIII rotating basket method (900 ml of pH 6.8-phosphate buffer, 100 rpm, 37±0.5 °C). Microspheres equivalent to 150 mg of Indomethacin was loaded into the basket of the dissolution apparatus. Five milliliters of the sample was withdrawn from the dissolution media at suitable time intervals and the same amount was replaced with fresh buffer. The absorbance of the filtrate was determined at wavelength of 247 nm by using UV-vis spectrophotometer, against pH 6.8 as blank.

RESULTS AND DISCUSSION

Preparative aspects of Eudragit Microspheres

To prepare pH dependent Microspheres the O/O (oil in oil) emulsion solvent evaporation technique was used since it yields more uniform particles. The method is correctly referred as O/O instead of W/O (water in oil) since a polymeric solution in organic solvent is considered as oil in micro encapsulation terminology. The organic phase containing pH dependent Eudragit L/S-100 mixture [1:2, 1:3, 1:4, 1:5] and dispersed Indomethacin was emulsified into an external oil phase. Pure acetone did not dissolve Eudragit; however acetone

with 2% water fitted the criterion well. Liquid paraffin was used as the dispersion media or external phase. Petroleum ether was used to clean the microparticle since it removes liquid paraffin without affecting the integrity of the microparticle.

Micromeritic properties

The arithmetic mean particle size of the formulations was determined by the optical microscope fitted with an ocular micrometer and stage micrometer. The average mean particle sizes

of the Microspheres were found to be between 164.62 - 249.92. The mean particle size of the Microspheres significantly increased with increase in polymer concentration due to high viscosity of medium at a higher polymer concentration resulting in enhanced interfacial tension and diminished shearing efficiency. The angle of repose of Microsphere ranges from 18° 62''- 23° 36'' The values of angles of repose indicate excellent flow properties.

Infrared spectroscopy

The FT-IR spectra study showed no change in the finger print of pure drug spectra, thus confirming absence of drug and polymer interaction.

Scanning electron microscopy (SEM)

Morphology of Microspheres was examined by scanning electron microscopy. The view of the Microspheres showed smooth surface morphology exhibited range of sizes within each batch. The outer surface of Microspheres was smooth and dense, while the internal surface was porous. The shell of Microspheres also showed some porous structure it may be caused by evaporation of solvent entrapped within the shell of Microspheres after forming smooth and dense layer.

Drug release

In vitro release studies were carried out using USP XXIII dissolution assembly. The release profile obtained for all the four formulations were shown in Fig. 4. It was observed that the drug release from the formulations decreased with increase in the amount of polymer added in each formulation. The release of drug from polymer matrix takes place after complete swelling of the polymer and as the amount of polymer in the formulation increase the time required to swell also increase thereby decrease in the drug release. However, the release showed a Bi-phases release with an initial burst effect At the end of first 30 min. drug release was 28.39%, 26.23%, 26.36% and 24.10% for F1 to F4 respectively. The mechanism for the burst release can be attributed to the drug loaded on the Microsphere or imperfect entrapment of drug. The mechanism for the burst release can be attributed to the drug loaded on the microsphere

or imperfect entrapment of drug. The overall cumulative % release for F1, F2, F3 and F4, were found to be 95.91%, 92.06%, 89.17%, and 87.81% at the end of 12th hour.

CONCLUSION

The results obtained from the study of "Design and evaluation of chronic-therapeutic drug delivery system of Indomethacin" provide the following conclusion: The 1:2 ratio of Eudragit L-100 and S-100 are suitable for preparation of microspheres for colonic targeting. The particle size increased significantly as the amount of polymer increased. The flow properties of all the prepared microspheres were good as indicated by low angle of repose ($\theta < 40^\circ$). The good flow properties suggested that the microspheres produced were non-aggregated. As the entrapment efficiency was good in all the cases, suggest that optimized parameters were used in the method of preparations. The *In-vitro* drug release of microspheres exhibits two type release pattern for all microspheres with initial burst release effect, which may be attributed to the drug loaded onto the surface of the particles. On the basis of, particle size, drug content, Scanning Electron Microscopy, IR-study, *in-vitro* release studies and its kinetic data, F3 was selected as an optimized formulation for designing pulsatile device.

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Table 1: Formulation of Indomethacin Microspheres

Sl. No.	Ingredients	Formulation Code			
		F1	F2	F3	F4
1.	Drug (mg)	1000	1000	1000	1000
2.	Eudragit L-100 (mg)	166	333	500	666
3.	Eudragit S-100 (mg)	332	666	1000	1332
4.	Span-80 (ml)	0.5	0.5	0.5	0.5
5.	Acetone (ml)	50	50	50	50

Table 2: Micromeritic properties of Indomethacin Microspheres

Formulation code	Mean particles size(µm)	Angle of repose
F1	164.62 ± 1.04	18° 75" ± 2.89
F2	168.12 ± 1.21	18° 62" ± 1.79
F3	195.99 ± 2.69	22° 09" ± 2.61
F4	249.92 ± 1.91	23° 36" ± 2.99

Table 3: Percentage yield and Drug content of Indomethacin microspheres

Formulation code	Percentage yield	Drug content Uniformity
F1	89.61 ± 0.40	82.79% ± 0.68
F2	86.56 ± 0.27	88.23% ± 0.95
F3	91.54 ± 0.18	96.19% ± 0.28
F4	88.85 ± 0.22	85.09% ± 0.16

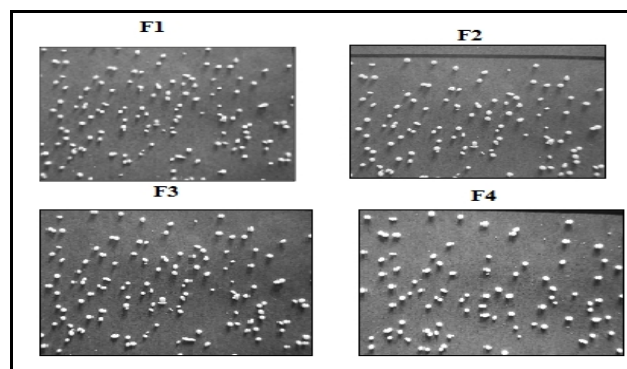


Fig. 1: Showing Microsphere of Indomethacin formulations(a) F1, (b) F2, (c) F3 (d) F4

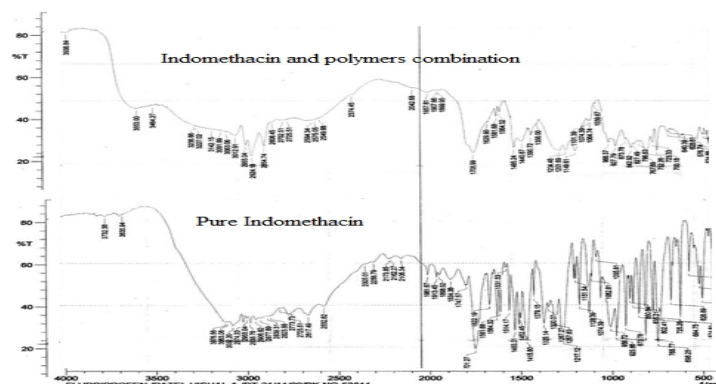


Figure 2: I.R. Spectrum of Pure Indomethacin and Indomethacin and polymers combination

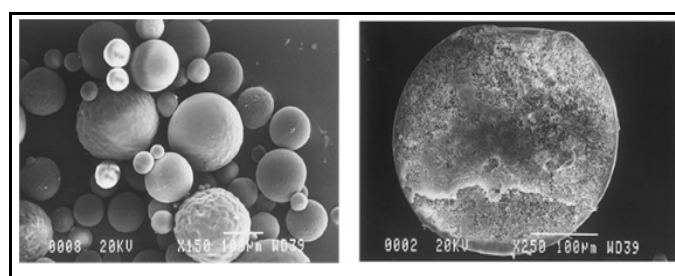


Figure 3: Scanning electron microphotographs of F3 formulation.

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