# Formulation and Evaluation of a Novel Drug Delivery System of Aceclofenac for Colonic Drug Delivery

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## ABSTRACT

Colonic delivery microspheres of Aceclofenac were prepared by ion-gelation method with an aim of increasing absorption of drug and for controlled release. A polymeric mixture of Sodium alginate and cellulose acetate phthalate was used. Cellulose acetate phthalate used as coating agent. The solution was dropped to 4% calcium chloride solution. The prepared microspheres coated with cellulose acetate phthalate by solvent evaporation method. Colonic delivery microspheres were evaluated with respect to particle size distribution, drug content or entrapped efficiency, morphology and in vitro release study. Effect of polymer concentration and coating concentration ratio on the above mentioned parameters were evaluated and it was found that the sodium alginate and cellulose acetate phthalate had a pronounced effect on various parameters. The enhanced controlled release properties of cellulose acetate phthalate coated sodium alginate microspheres made them an excellent candidate for colonic delivery.

**Keywords:** Aceclofenac, Sodium alginate, Cellulose acetate phthalate, Colonic delivery microspheres.

## INTRODUCTION

Oral controlled release dosage forms are being developed for the past three decades due to their advantages. The design of oral controlled drug delivery system is primarily aimed at achieving more predictable and increased bioavailability, thereby obtaining a maximum therapeutic effect. However some of these systems don't work as planned due to several physiological difficulties, such as an inability to restrain and localize the drug delivery system within desired region of GI tract and highly variable nature of gastric emptying process. It can be anticipated that, depending upon the physiological state of subject and the design of pharmaceutical formulation, the emptying process can last from a few minutes to 12 hours. Rapid GI transit can prevent complete drug release in the absorption zone and reduce the efficacy of administered dose since the majority of drugs are absorbed in stomach or upper part of small intestine[1].

Thus placement of drug delivery system in a specific region of the GI tract offers a numerous advantages especially to the drugs having narrow absorption window, stability problem, and poor solubility in acidic pH, local irritation in stomach. In the colonic region absorption sites are more due to presence of microvillus and it increase the absorption of the drug and results increase the bioavailability of the drug. Therefore the design of a colonic delivery requires minimum drug release in stomach region and maximum drug release in the colonic region. Recently one of such systems has been reported as colonic

delivery dosage form. Such systems are delivered the drug to the specific site of the colon[2].

Aceclofenac is a Non steroidal anti-inflammatory agent used in treatment rheumatoid arthritis diseases has been taken as a model drug in the present investigation because of its low biological half-life (3-4h).

## MATERIALS AND METHODS

Aceclofenac was obtained as a gift sample from Mannet pharma Gujrat. Sodium alginate, cellulose acetate phthalate and calcium chloride, used were of analytical grade, purchased from Himedial Laboratries, CDH Mumbai respectively.

### Preparation of Cellulose acetate phthalate coated sodium alginate microspheres:[3-6]

Aceclofenac microspheres were prepared by ionic gelation method. The required amount of sodium alginate was dispersed in a specified volume of cold water allowed to swell for 2 hours. In another beaker required amount of drug was dispersed in phosphate buffer pH 7.4 solution. The drug solution was added to sodium alginate solution with stirring to produce a viscous form. The drug polymer solution was added drop wise by using syringe of 22 G in diameter form a height of about 5cms into a beaker containing 4% w/v solution of calcium chloride with continuous stirring by magnetic stirrer. Then the solution was containing microspheres was filtered by filter paper. The microspheres were allowed to dry at about 30 to $40^0$C and it is coated with cellulose acetate phthalate. Cellulose acetate phthalate was dissolve in 50% acetone and 50% ethanol solution and these solutions is used as a coating solution. Sodium alginate microspheres was poured in the coating solution by continuous stirring then the solution was evaporated and coated cellulose acetate phthalate microspheres was prepared.

### Process variables: [9]

The process variables were investigated (Bore diameter of the needle, concentration of sodium alginate, concentration of cellulose acetate phthalate, and concentration of calcium chloride and height of dropping) and different batches thus produced were analyzed for size, shape, ease of preparation, drug content and drug release.

### Measurement of Micromeritic properties:[7]

The flow properties of prepared microspheres were investigated by measuring the bulk density, tapped density and Carr's index. The bulk and tapped densities were measured in a 10 ml graduated measuring cylinder. The sample contained in the measuring cylinder was tapped mechanically by means of constant velocity rotating cam. The initial bulk volume and final tapped volume were noted from which, their respective densities were calculated.

**Particle size Analysis:**[7]

Microspheres were separated into different size fractions by sieving for 10 minutes using a mechanical shaker (Labtech, Indore, Co. India) containing standard sieves # 22 to # 44 and mean particle sizes of microspheres were calculated.

**Percentage encapsulation efficacy of microspheres:**

One hundreds milligram of formulations was dissolved in 10 ml of phosphate buffer of pH 7.4. The samples were assayed for drug content by UV-spectrophotometer (UV-1800) at 274 nm and the drug content was calculated[5].

***In vitro* drug release study:**

The drug release rate from microspheres was carried out using Tablet dissolution test apparatus. A weight of floating microspheres corresponding to 100 mg of drug was filled into a capsule and placed in basket. Dissolution media was 900 ml simulated gastric fluid and simulated intestinal fluid maintained at $37 \pm 2^0$ and stirred at 50 rpm. Samples (5 ml) were withdrawn at suitable interval of time and volume was adjusted. It was then assayed spectrophotometrically at 274 nm[4,8].

***In vitro* drug release study in rat cecal content:**

Rat cecal content was prepared by the method reported by Van den Mooter et al[4,8]. Four albino rats, (Sprague-Dawley strain) of uniform body weight (150-200 g) with no prior drug treatment, were used for all the present in vivo studies; they were weighed, maintained on normal diet, and administered 1 ml of 2% dispersion of sodium alginate in water, and this treatment was continued for 7 days for polymer induction to animals. Thirty minutes before starting the study, each rat was humanely killed and the abdomen was opened. The cecal were traced, legated at both ends, dissected, and immediately transferred into phosphate buffered saline (PBS) pH 6.8, which was previously bubbled with CO2. The cecal bag was opened; the contents were weighed, homogenized, and then suspended in PBS (pH 7.4) to give the desired concentration (2%) of cecal content, which was used as simulated colonic fluid. The suspension was filtered through cotton wool and ultrasonicated for 10 minutes in an ice bath at 40%voltage frequency using a probe sonicator at 4-C to disrupt the bacterial cells. After sonication, the mixture was centrifuged at 2000 rpm for 20 minutes.

Microspheres (100 mg) were placed in 200 mL of dissolution media (PBS, pH 7.4) containing 2% wt/vol rat cecal content. At different time intervals, the samples were withdrawn and replaced with fresh PBS. The withdrawn samples were pipette into a series of 10 ml volumetric flasks, and volumes were made up to the mark with PBS and centrifuged. The supernatant was filtered through 0.45-µm membrane filter (Millipore Corp, Billerica, MA) and the filtrate analyzed for aceclofenac at 274 nm using UV method.

**Fourier Transform Infrared Spectroscopy [FTIR] Study:**

Drug-polymer interactions were studied by FTIR spectroscopy. The spectra were recorded for pure drug, pure polymer and drug-loaded microspheres using FTIR. The scanning range was 400-4000$cm^{-1}$ and the resolution was 2$cm^{-1}$.

**Surface morphology:**

The sample for the Scanning Electron Microscopy analysis was prepared by sprinkling the microspheres on one side of a adhesive stub. Then the microspheres were coated with gold before microscopy. Finally the microspheres were observed with the scanning electron microscope.

**Statistical Analysis:**[4]

The mean percentage of Aceclofenac released in SGF (at different pH) and in presence of 2% cecal content of rats from cellulose acetate phthalate coated sodium alginate microspheres was prepared by using various drug:polymer ratios and compared. The ANOVA test was used to find the statistical significance. A value of P less than .05 was considered statistically significant.

## RESULTS AND DISCUSSION

**Evaluation of preparation method:**

In this project attempts have been made to prepare the cellulose acetate phthalate coated sodium alginate microspheres by ionotropic gelation method. Here the microspheres were performed with increased the sodium alginate concentration and coating concentration increases drug entrapment efficiency increased.

Table 1: Formulation for aceclofenac microsphers

| Sl. No. | Formulation code | Aceclofenac (Ratio) | Sodium alginate (Ratio) | Cellulose acetate pthalate (%) |
|---|---|---|---|---|
| 1 | $MF_1$ | 1 | 1 | 5 |
| 2 | $MF_6$ | 1 | 1.50 | 9 |
| 3 | $MF_9$ | 1.25 | 1.50 | 7 |
| 4 | $MF_{10}$ | 1.25 | 0.75 | 7 |
| 5 | $MF_{11}$ | 1.25 | 2 | 7 |
| 6 | $MF_{12}$ | 1.25 | 2 | 9 |
| 7 | $MF_{13}$ | 1 | 1 | 7 |
| 8 | $MF_{14}$ | 1 | 1 | 9 |

**Process optimization:**

The process variables were investigated and different batches thus produced were analyzed for size, shape, ease of preparation, drug content and drug release. Optimized process variables are described by Table No.2.

Table 2: Optimized process variables data

| Process variable parameters | Optimized data |
|---|---|
| Bore diameter of the needle | 22G |
| Height of dropping | 5 cm from the level of $CaCl_2$ solution |
| Drying time and temperature | $30^0$ to $40^0$ c for 4 hrs |
| Sodium alginate concentration | 2% |
| Cellulose acetate phthalate concentration | 9% |
| Calcium chloride concentration | 4% |

**Micromeritics properties:**

Particle size was determined by sieve analysis method. The most of microspheres were obtained in the size range of 355-710 µm (Table 3). The variation in particle size is due to the variation in polymer concentration and coating concentration.

Table 3: Micromeritics properties of aceclofenac microspheres

| Formulation Code | Weight % retention in different ASTM Sieve | | |
|---|---|---|---|
| | 710µm | 500 µm | 355 µm |
| $MF_1$ | 91.11±0.54 | 6.66±0.09 | 2.22±0.04 |
| $MF_6$ | 98.17±0.42 | 1.09±0.05 | 0.72±0.01 |
| $MF_9$ | 95.37±0.35 | 3.70±0.06 | 0.92±.001 |
| $MF_{10}$ | 97.23±.085 | 2.20±0.02 | 0.55±0.02 |
| $MF_{11}$ | 96.79±0.56 | 1.28±0.01 | 1.92±.001 |
| $MF_{12}$ | 90.57±0.87 | 6.52±0.06 | 2.89±0.02 |
| $MF_{13}$ | 97.59±0.98 | 1.20±0.05 | 1.20±0.03 |
| $MF_{14}$ | 96.66±0.45 | 2.66±0.02 | 0.67±0.05 |

± S.D (n=3)

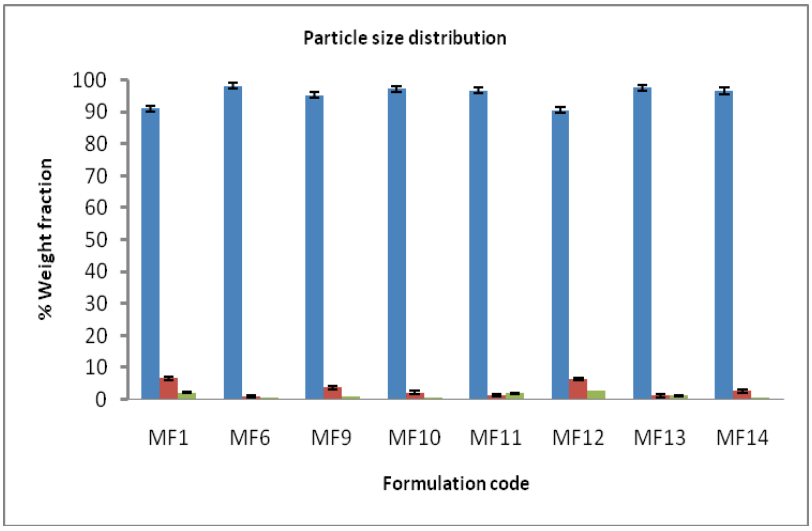


Fig 1: Particle size distribution curve

**Percentage drug entrapment efficiency:**

The percentage drug entrapment efficiency was increased with increasing the sodium alginate concentration. The results were shown in Table No.4.

Table 4: Characteristic of aceclofenac microspheres

| Sl. No. | Formulation code | Drug:Polymer | Percentage Encapsulation Efficiency (%) |
|---|---|---|---|
| 1 | $MF_1$ | 1:1 | 88.72 ± 0.85 |
| 2 | $MF_6$ | 1:1.50 | 58.79±0.92 |
| 3 | $MF_9$ | 1.25:1.50 | 74.42±0.87 |
| 4 | $MF_{10}$ | 1.25:0.75 | 64.01±0.78 |
| 5 | $MF_{11}$ | 1.25:2 | 61.58±.086 |
| 6 | $MF_{12}$ | 1.25:2 | 45.79±0.79 |
| 7 | $MF_{13}$ | 1:1 | 51.27±0.99 |
| 8 | $MF_{14}$ | 1:1 | 50.92±0.85 |

± S.D (n=3)

***In vitro* drug release study:**

Drug release was performed using USP dissolution rate test apparatus(Apparatus 1,50 rpm, 37± 0.5$^0$C) for first 2 hrs in simulated gastric fluid pH 1.2 (900 ml) and further release was performed in simulated intestinal fluid pH 6.4 (900 ml). The effect of sodium alginate concentration and cellulose acetate phthalate on drug release was also calculated; it was found that with increase in

amount of cellulose acetate phthalate the drug content and drug release increases (Table 5and 6). This might be attributed to the increase in coating thickness on sodium alginate microspheres.

Table 5: Comparative percentage drug release profile of various formulations in SGF

| Drug release profile of various formulations in SGF (%) | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Time(hrs) | $MF_1$ | $MF_6$ | $MF_9$ | $MF_{10}$ | $MF_{11}$ | $MF_{12}$ | $MF_{13}$ | $MF_{14}$ |
| 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 0.5 | 1.26±0.02 | 1.77±0.21 | 1.33±0.01 | 0.92±0.02 | 1.12±0.03 | 1.62±0.07 | 0.75±0.04 | 0.91±0.04 |
| 1 | 1.70±0.02 | 1.98±0.02 | 1.44±0.06 | 1.23±0.02 | 1.35±0.04 | 1.75±0.13 | 1.06±0.04 | 1.05±0.04 |
| 1.5 | 2.18±0.02 | 2.25±0.01 | 1.63±0.02 | 1.37±0.12 | 1.69±0.02 | 2.13±0.02 | 1.19±0.01 | 1.18±0.03 |
| 2 | 2.27±0.04 | 2.41±0.02 | 1.77±0.02 | 1.65±0.02 | 2.25±0.02 | 2.46±0.06 | 1.46±0.02 | 1.37±0.02 |

± S.D (n=3)

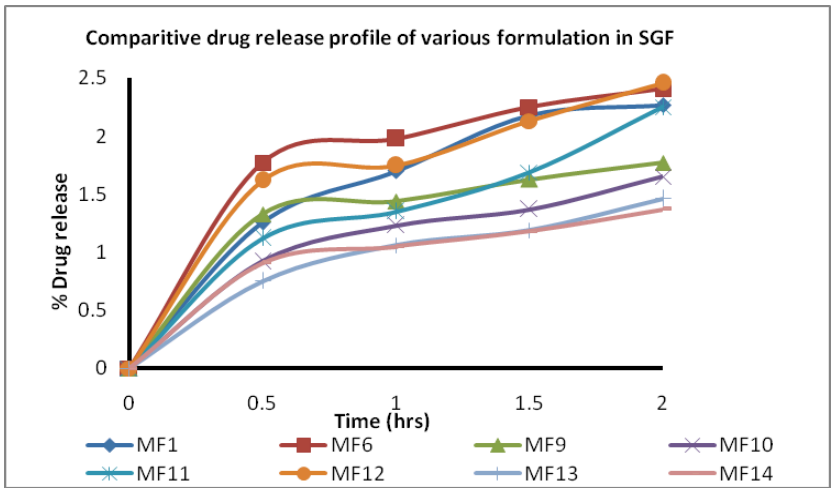


Fig 2: Comparative percentage drug release profile of various formulations in SGF

Table 6: Comparative percentage drug release profile of various formulations in SIF

| Drug release profile of various formulations in SIF (%) | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Time(hrs) | $MF_1$ | $MF_6$ | $MF_9$ | $MF_{10}$ | $MF_{11}$ | $MF_{12}$ | $MF_{13}$ | $MF_{14}$ |
| 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 0.5 | 74.30±0.94 | 24.16±0.37 | 48.17±0.30 | 61.98±0.62 | 53.66±0.38 | 22.68±0.35 | 55.36±0.21 | 31.97±0.63 |
| 1 | 85.22±1.0 | 45.65±0.33 | 61.15±0.03 | 67.14±0.49 | 59.31±0.42 | 32.41±0.70 | 64.22±0.86 | 45.68±0.65 |
| 1.5 | 91.78±0.74 | 51.02±0.65 | 72.26±00.56 | 72.31±1.11 | 81.91±0.84 | 45.37±0.58 | 73.08±0.36 | 59.38±0.32 |
| 2 | 96.15±0.57 | 56.39±0.09 | 79.68±0.70 | 79.19±0.43 | 90.38±0.79 | 51.85±0.41 | 81.94±0.48 | 73.09±1.02 |
| 2.5 | 98.33±0.84 | 64.45±1.31 | 85.24±0.78 | 91.25±0.41 | 93.21±0.30 | 61.58±0.50 | 90.80±0.98 | 82.22±0.26 |
| 3 | | 77.87±1.49 | 98.21±0.44 | 99.86±0.36 | 98.86±0.15 | 84.26±0.82 | 99.66±0.38 | 91.36±0.15 |
| 3.5 | | 96.67±0.47 | | | | 93.99±0.68 | | 93.64±0.23 |
| 4 | | | | | | 97.23±0.43 | | 98.21±0.16 |

± S.D (n=3)

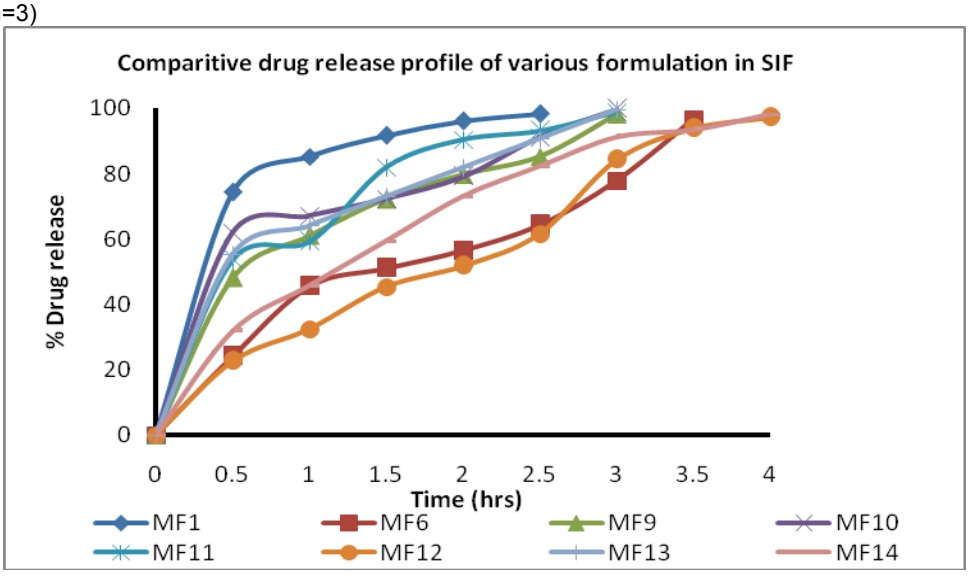


Fig 3 : Comparative percentage drug release profile of various formulations in SIF

### *In vitro* drug release study in rat cecal content:

The *in-vitro* drug release of aceclofenac microspheres in presence of 2% rat cecal content in simulated colonic fluid showed faster drug release at different time periods when compared with release study without rat cecal content (Table 7 & 8). This finding could be attributed to the various anaerobic bacteria present in cecal content and responsible for degradation of sodium alginate in order to release drug from microspheres.

Table 7: In Vitro Drug Release Study in the Presence and absence of Rat Cecal Content ($MF_{12}$)

| Time(hrs) | % Drug release(Without rat cecal content) | % Drug release (With rat cecal content) |
|---|---|---|
| 0 | 0 | 0 |
| 0.5 | 39.91±0.21 | 48.78±0.20 |
| 1 | 57.65±0.91 | 62.09±1.15 |
| 1.5 | 70.96±0.43 | 97.57±0.02 |
| 2 | 86.48±0.21 | |
| 2.5 | 99.79±0.27 | |

± S.D (n=3)

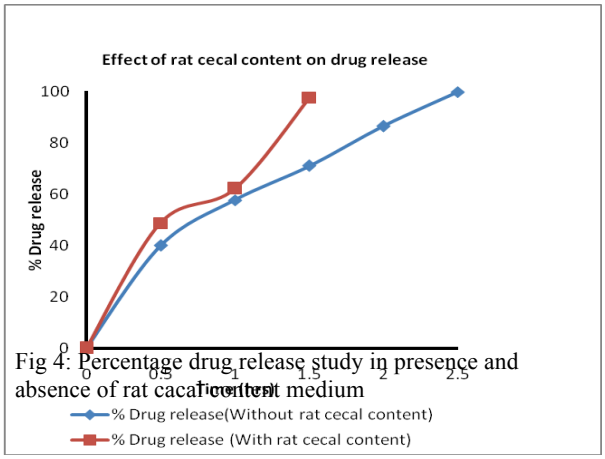


Fig 4: Percentage drug release study in presence and absence of rat cacal content medium

Table 8: In Vitro Drug Release Study in the Presence and absence of Rat Cecal Content ($MF_{14}$)

| Time(hrs) | % Drug release(Without rat cecal content) | % Drug release (With rat cecal content) |
|---|---|---|
| 0 | 0 | 0 |
| 0.5 | 38.81±0.25 | 42.19±1.01 |
| 1 | 43.75±1.10 | 65.63±0.34 |
| 1.5 | 68.76±0.7 | 87.51±0.56 |
| 2 | 79.70±0.29 | 95.33±0.84 |
| 2.5 | 93.76±0.43 | |
| 3 | 96.89±0.30 | |

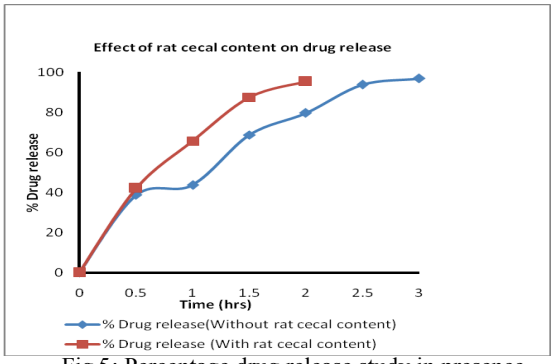


Fig 5: Percentage drug release study in presence absence of rat cacal content medium and

### Fourier Transform Infrared Spectroscopy [FTIR] Study:

Drug polymer interference was studied between drug and polymer and it was shown in figure 6, 7 and 8 was found that there is no chemical interaction between drug and polymer.

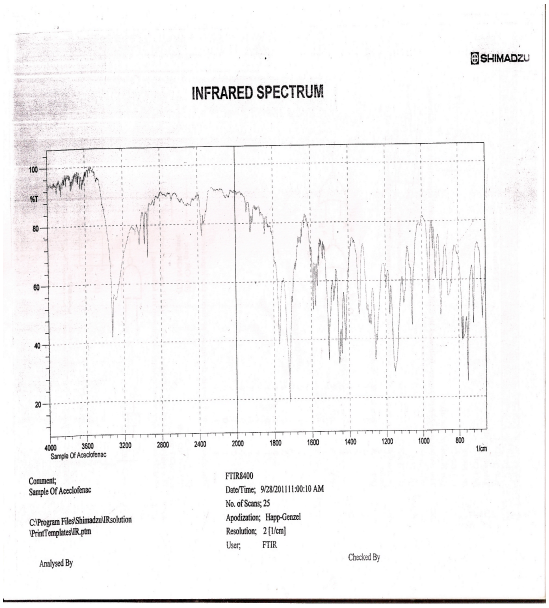


Fig 6: FTIR spectra of aceclofenac

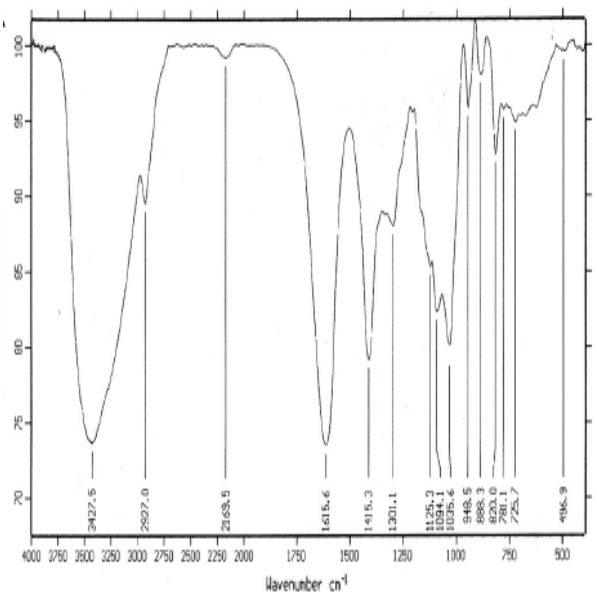


Fig 7: FTIR spectra of sodium alginate

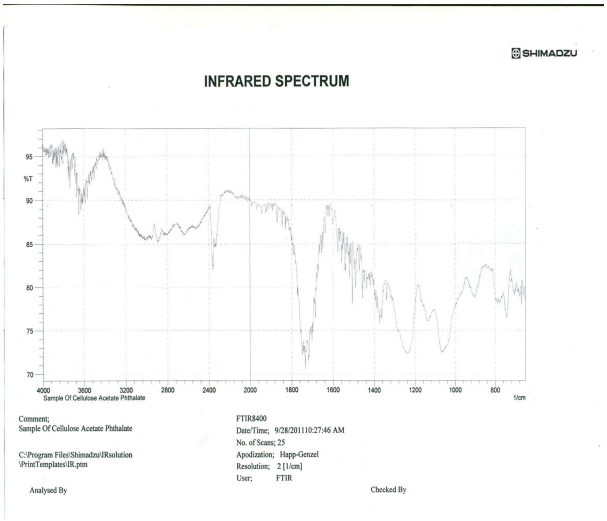


Fig 8 : FTIR spectra of cellulose acetate phthalate

**Surface morphology**:

Scanning electron microscopy was used to observe the surface morphology of cellulose acetate phthalate coated sodium alginate microspheres with drug. The scanning electron microscopy shows smooth surface of the

microspheres and particle size was found to be 500 µm. The image of drug loaded microspheres was shown in figure 8.

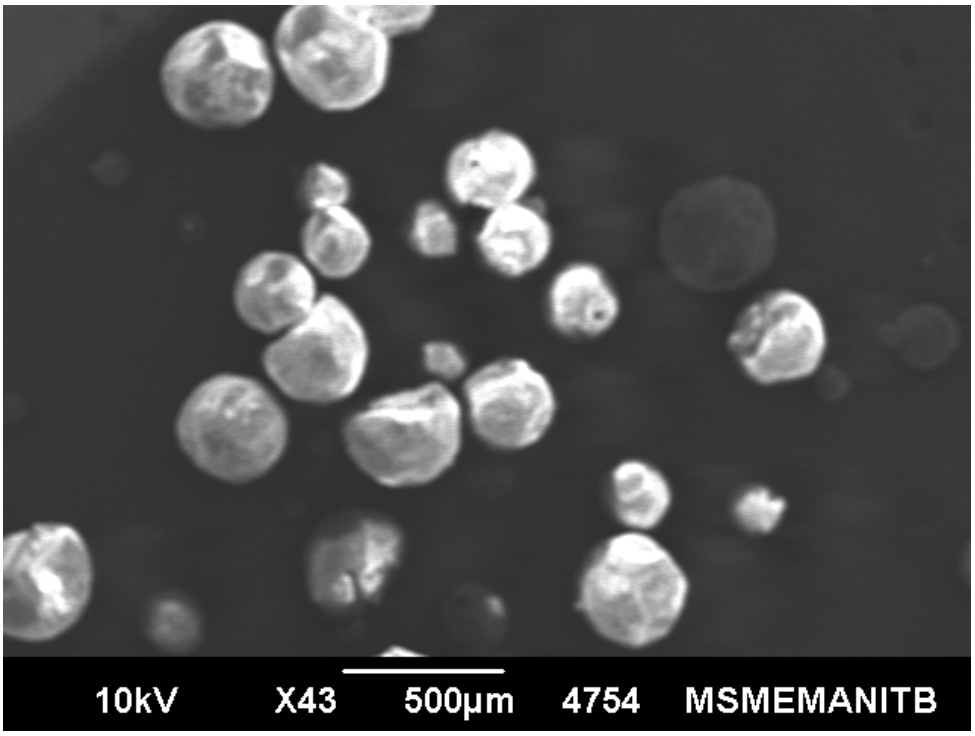


Fig 9: Scanning Electron Microscopy image of $MF_1$ formulation

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