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Original Article

Formulation and *in vitro* Characterization of Metronidazole Loaded Polymeric Microspheres for Colon Specific and Sustained Drug Delivery

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ABSTRACT

Objective: Metronidazole stacked alginate microspheres have been utilized to drag out the gastric living arrangement time and enhance the neighbourhood impact of medication in the district of colon for the treatment of bacterial contaminations by expecting that the colonic microbes will enzymatically corrupt the polysaccharide into natural corrosive and by bringing down the pH condition with the goal that disintegration of corrosive dissolvable covering and arrival of medication occur at the same time. **Methods:** Metronidazole microspheres were made by ionic gelation method utilizing Guargum, Chitosan, Eudragit S-100 and sodium alginate at various proportions and utilized calcium chloride (4% w/v) as rigidizing operator. Microspheres were described for its molecule measure, medicate stacking, tranquilize capture, swelling file and medication discharge properties. **Results:** Microspheres are observed to be inside size extents from 36.77µm to 229.96µm with medication capture proficiency of 42–99% w/w. The microsphere kept up to maintain the medication discharge up to 12 h if there should be an occurrence of definition FG4. Medication discharge from the microspheres, swelling file and lightness establishes to be relies upon the grouping of chosen polymers in the polymer mixture. Definitions containing low groupings of Guargum, Chitosan, Eudragit S-100 and sodium alginate (1:1) demonstrating shorter drifting slack time and quicker medication discharge and the other way around. Accordingly, swelling record and rate of medication discharge seemed, by all accounts, to be balanced by the centralization of chose polymers in the polymer mix. The outcomes demonstrated that Glutaraldehyde (1ml) based upgraded plan FG4 could be helpful in the detailing of metronidazole microspheres for better treatment of bacterial contaminations in colon.

Key words: Metronidazole, Microspheres, Swelling index, *in-vitro* drug release, Sustained release, Colon specific.

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INTRODUCTION

Microspheres with molecule measure ranges from 1 to 1000 µm in width and it is a microparticle medicate conveyance framework.[1-3] As microspheres are simple regulated, so it is planned into single-unit measurements frames like filling them into hard gelatin containers or might be tablets.[4] Metronidazole is dissolvable in pH 1.2, pH of the terminal ileum and colon at pH 6.8 and pH 8.[5] The significant system of activity of Metronidazole to murders the trophozoites situated in the colon due its amoebicidal movement. Along these lines, assembling of metronidazole microspheres is required to exemplify the medication with appropriate pH reliant and deferred the arrival of medication at the site of colon at lower pH. Another issue related with the medication is its intense taste, which may prompts persistent resistance. Alginates utilized as colonic medication transporter and at colonic greenery it indicates nontoxicity,[6] biocompatibility[7] and biodegradability[8] and separated from these it additionally having defensive impact on the mucous layers of the upper GIT.[7] As dried alginate dots demonstrating swelling conduct at pH 7.4, so it shields the corrosive touchy medication from gastric squeeze and utilized for controlled discharge framework.[8] Sodium alginates utilized as a focusing on material for starch receptors (polysaccharide) present in the pimple mass of *E. histolytica* contains glycoproteins and focus on the arrival of medication at the site of contamination. Metronidazole is an immediate luminal amoebicidal tranquilize utilized securely and more powerful when contrasted with other luminal amoebicidal drugs.[9-11] The point of the present work was to plan and *in vitro* portrayal of metronidazole stacked polymeric microspheres for colon-particular and supported medication conveyance for the compelling treatment of amoebiasis.

MATERIALS AND METHODS

Materials

Metronidazole (MTZ) was obtained as a gift sample from Albert Devid Ltd., Kolkata, India. Guargum, Chitosan, Eudragit S-100 and Sodium alginate were purchased form Merck Specialities Private Limited, Mumbai. All other chemicals were used are of analytical grade.

Method

Method of preparation of microspheres

Microspheres were set up by ionic gelation method. Here, required measure of polymers, for example, guar gum, chitosan, Eudragit S-100 were scattered in a predefined volume of chilly water containing the medication and permitted to swell for 2 h. In another measuring glass reasonable measure of sodium alginate was taken and blended well with 10 ml of water. The polymeric arrangement containing the medication was added to sodium alginate arrangement with mixing to create a thick suspension. After total blending 1.0 ml of glutaraldehyde were added to the above scattering, trailed by consistent mixing at a speed of 500rpm. At that point polymermeric sedate arrangement was included drop astute by utilizing syringe with needle of 22 G in measurement from a tallness of around 5 cm into a container containing 4% w/v arrangement of calcium chloride with ceaseless blending by attractive stirrer. At that point the arrangement containing the framed microspheres was sifted by utilizing What man channel paper no-1. The microspheres were permitted to dry at around 30 to 40°C and put away in very much shut compartment for further examinations.[12,13]

API characterization

Bulk density

Bulk density or apparent density is defined as the ratio of mass of a powder to the bulk volume. The bulk density of a powder depends on particle size distribution, particle shape and the tendency of the particles to adhere on to one another.[14] Weighed accurately 25 g of drug, sifted through 20 # sieve and transferred into 100 ml graduated cylinder. Carefully levelled the powder without compaction and noted the unsettled apparent volume (Vo) and then calculated the apparent bulk density in g/ml by the following formula-1:

$$\text{Bulk density} = \frac{\text{Weight of the powder (M)}}{\text{Volume of the packing (VO)}} \quad (1)$$

Tapped density

The blend after determining the bulk density was subjected to mechanical tapping in the tapped density tester (USP I apparatus) that operates to tap at a distance of 14 ± 2 mm at a nominal rate of 300 drops per minute. Initially measuring cylinder was tapped for 500 times and tapped volume was determined as Va and after that tapping was continued up to 750 times and the tapped volume was determined as Vb. From the above two tapped volumes final tapped volume, Vf was determined from the difference values of the two volumes (Va-Vb).[15] Tapping was still continued up to 1250 taps, until the difference between succeeding measurements is less than 2%. The tapped density was determined in g/mL using the below formula-2:

$$\text{Tapped density} = \frac{\text{Weight of the powder (M)}}{\text{Tapped Volume of the packing (Vf)}} \quad (2)$$

Hausner's ratio

Hausner's ratio gives an idea regarding the flow of the blend. It is the ratio of tapped density to the apparent density.[16] Hausner's ratio was calculated by using the following formula-3:

$$\text{Hausner's ratio} = \frac{\text{Tapped density}}{\text{Bulk density}} \quad (3)$$

Compressibility index

The compressibility index determined for the powders to be compressed. The packing ability of drug was estimated from change in volume, which is due to rearrangement of packing occurred during tapping.[17] The Carr's compressibility index (CI) can be calculated using following formula-4:

$$\text{Compressibility index} = \frac{\text{Tapped density} - \text{Bulk density}}{\text{Tapped density}} \times 100 \quad (4)$$

Angle of repose

Irregular flow of powders from the hopper of a tablet machine may leads to production of tablets with non-uniformity in weights. Angle of repose (θ) is defined as the maximum angle that can be obtained between the free standing surface of a powder heap and the horizontal plane[18] ad can be calculated by using following formula-5 to find out the flow characteristics of the powder blend ready to be compressed:

$$\text{Tan}\,\theta = \frac{\text{Height of pile (h)}}{\text{Bulk density radius of the base of pile (r)}} \quad (5)$$

PARTICLE SIZE ANALYSIS

Optical microscope is used for the determination of particle diameter of the microspheres. The samples were suspended in dispersion and individual diameter of microspheres was measured using micrometre. A diameter of about 500 microspheres was measured and then mean particle diameter was calculated.[19]

Surface morphology

Scanning electron microscope (FEI Quanta-200 MK2, Netherlands) is used for the determination of morphology and size of the microspheres. The samples for the SEM analysis were prepared by sprinkling of the microspheres on one side of an adhesive stub. Then the microspheres were coated with gold before microscopy.[20]

DSC studies

Drug and polymers compatibility studies were performed by using Differential Scanning Calorimetry (DSC Q10 V9.0 Build 275). The pure drug along with individual excipients was analyzed for DSC to know the compatibility of excipients with drug. Differential scanning calorimeter is used to measure the specific heat and enthalpies of transition. Thermo grams were obtained by using a DSC at a heating rate of 15°C /min over a temperature range of 0 to 1000°C.[21]

Drug entrapment efficiency (DDE) and Drug loading (DL)

Drug entrapment efficiency (DDE) and drug loading (DL) capacities of the prepared microspheres were determined by taking equivalent weight of 200 mg of drug. Initially equivalent weight of microspheres were crushed in mortar and pestle and the powdered microspheres were dissolved in 100 ml of phosphate buffer (pH 7.4) solution and stirred for 15 min with an interval of 5 min and allowed to keep for 24 h. Then the solution was filtered through Whatman's (No.1) filter paper. Then the absorbance was measured using UV-Visible spectrophotometer at λmax 320nm against phosphate buffer of pH 7.4 solution as blank and concentrations were determined by employing simultaneous equation 1 to 3.[22]

$$Y = MX + C \quad \text{(Equ.1)}$$

$$\text{DEE}\,(\%) = \frac{\text{Experimental drug Content}}{\text{Initial drug Content int o the Formulation}} \times 100 \quad \text{(Equ.2)}$$

$$\text{DL}\,(\%) = \frac{Qm}{Wm} \times 100 \quad \text{(Equ.3)}$$

Where, W m = weight of the microspheres; Q m = quantity of the drug present in the microspheres

FT-IR studies

FT-IR spectrophotometer (Perkin Elmer Spectrum GX) were used for determination of functional groups of drug and to analysed the drug excipients compatibility of selected drug and its physical mixtures by selected the wavelength number of 4000 and 400cm^{-1} by using KBr pellet method.[23]

In vitro drug release study

In vitro drug release study were evaluated using USP Apparatus 1 (Basket Type) at a rotation speed of basket 100 rpm and temperature 37±0.5°C using dissolution media 0.1 N HCl (900ml) for first 2h and after 2h the pH of the dissolution media adjusted to 6.8 and performed the study for 4h. Finally the release study was continued up to 24h adjusting pH 7.4 phosphate buffer and 5ml samples were withdrawn and replaced by an equal volume of fresh medium to maintain the sink condition. Withdrawn samples were filtered, diluted and assayed at specified time interval for estimating the drug released at λ_{max} of 320 nm using

double beam UV-Vis spectrophotometer.[24] Drug release data were fitted in to different kinetic models such as zero order [Equation 4], first order [Equation 5], Higuchi matrix [Equation 6] and Korsmeyer-Peppa's [Equation 7] to find the equation with the best fit.

$$R = K0T \quad \text{(Equ.-4)}$$

$$\text{Log UR} = K1T/2.303 \quad \text{(Equ.-5)}$$

$$R = Kh\sqrt{T} \quad \text{(Equ.-6)}$$

$$R = KKP\ Tn^{*} \quad \text{(Equ.-7)}$$

Where, R and UR are the released and unreleased percentages respectively, at time [t]; K0, K1, Kh, K, Kp are the rate constants of zero order, first order, Higuchi matrix and Korsmeyer-Peppas respectively.

Swelling study

The prepared formulations were placed in distilled water, 0.1 N HCl and Phosphate buffer of pH 7.4 and allowed to swell to till the constant weight. The microspheres were removed, blotted with filter paper and their changes in weight were determined every 10 min time interval and the data was recorded.[25] The degree of swelling (a) was then calculated from the formula-6;

$$\text{Swelling index} = \frac{wg - wo}{wo} \times 100 \quad (6)$$

Where, Wo is the initial weight of the microspheres, Wg is the weight of the microspheres at equilibrium swelling in the medium.

RESULTS AND DISCUSSION

Process variables

The process variables were determined and the various formulations were determined for particle size, shape, ease of preparation, drug content and drug release characteristics. The process variables are described in Table 1 and 2.

MICROMETRIC PROPERTIES

The value of angle of repose of formulation found to be within the range of 25°C, indicating very good flow properties for the microspheres. The tapped density and Carr's Index values was found to be 0.758 to 0.796 g/cm^3 and 9.30 to 10.40% respectively, suggests excellent flow characteristics of the microspheres. Hausner's ratio found to be range from 1.10 to 1.19% indicates good flow property of microspheres and the results shown in Table-3.

PARTICLE SIZE ANALYSIS

The particle size analysis of the formulations were determined by sieve analysis method. Mean diameter of Glutaraldehyde based microspheres found to be 680 ± 0.68 to 768±0.38µm and increasing average particle size of microspheres due to increasing in polymer ratio along with crosslinking agent (Table 4).

Drug entrapment efficiency

The percent encapsulation efficiency found to be increased up to 72.95 ± 0.56% with increasing polymer concentration and the percent encapsulation efficiency was increased up to 75.21 ± 0.4 % with addition of cross linking agent Glutaraldehyde. So concentration of polymers and addition of cross linking agents found to be influencing the drug entrapment efficiency. The amount of cross linking agent i.e. Glutaraldehyde used is 1.0 ml. The results of percentages of encapsulation efficiency were placed in Table 4.

Drug Loading

The results of drug loading found to be increased from 13.72 ± 0.48% to 25.04 ± 0.39% of microsphere with increasing the amount of polymer as well as using the cross linking agent. So, concentration of polymers and addition of cross linking agent Glutaraldehyde (1.0 ml) found to be influencing the drug loading efficiency. The drug loading values are described in Table 4.

DRUG-POLYMER COMPATIBILITY STUDIES

FTIR studies

The reported peaks (cm^{-1}) of metronidazole by FTIR spectroscopy of -OH, -C-CH, -N-O, -C-O and -C-N assignments were 3230 cm^{-1}, 3105 cm^{-1}, 1538 cm^{-1} and 1375 cm^{-1}, 1078 cm^{-1} and 830 cm^{-1}, respectively, whereas the observed peaks (cm^{-1}) were found to be 3228.09 cm^{-1}, 3096.03 cm^{-1}, 1538.76 cm^{-1} and 1372.41 cm^{-1}, 1074.87 cm^{-1} and 818.59 cm^{-1}, respectively. The FT-IR spectral analysis of the physical mixtures of drug and selected polymer shows that there were no significant interaction between drug and polymers as shown in Figure 1 and 2.[26]

Differential Scanning Calorimetry (DSC) studies

The sample of metronidazole was scanned at 10°C/min from 30°C to 305°C and thermal behavior of the drug was studied by recording the thermo gram. The DSC thermo gram of metronidazole exhibited sharp endothermic peak shown in Figure 3.[27]

Table 1: Formula of prepared microspheres by ionic gelation technique

Ingredients (mg)	Formulation Code							
	Fl	F2	F3	F4	FG1	FG2	FG3	FG4
Drug	400	400	400	400	400	400	400	400
Guargum	80	160	240	320	80	160	240	320
Chitosan	100	200	300	400	100	200	300	400
Eudragit S-100	120	240	360	480	120	240	360	480
Sodium Alginate	100	200	300	400	100	200	300	400
Total Polymers	400	800	1200	1600	400	800	1200	1600
Ratio	1:1	1:2	1:3	1:4	1:1	1:2	1:3	1:4
Glutaraldehyde (ml)	-	-	-	-	1	1	1	1

Table 2: Process parameters for preparation of microspheres

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 to 40^{0}C for 4hrs.
Polymers concentration	30% w\v dispersion.
Sodium alginate concentration	10% w\v dispersion.
Calcium chloride concentration	4% w\v solution
Concentration of glutaraldehyde	1.0ml
Bore diameter of the needle	22G

Table 3: Flow properties of pure drug and prepared microspheres

Formulation code	Angle of repose (Degree)	Carr's index	Hausner's ratio
Pure drug	28.52±0.09	29.87±0.346	1.405±0.12
Fl	18± 0.68	9.45 ± 0.34	1.11 ± 0.48
F2	20± 0.57	10.35 ± 0.42	1.10 ± 0.64
F3	22± 0.61	9.15 ± 0.28	1.11 ± 0.84
F4	24±0.29	9.95 ± 0.56	1.19 ± 0.73
FG1	20± 0.64	10.28 ± 0.47	1.14 =t 0.63
FG2	18±0.38	9.34 ± 0.69	1.18 ± 0.68
FG3	18±0.52	10.18 ± 0.48	1.19 ± 0.54
FG4	21±0.34	10.21 ±0.41	1.14 =t 0.39

Data presented as Mean ± S.D. (n=3), n is the number of observation

Table 4: Particle size analysis, drug entrapment efficiency and drug loading

Formulation Code	Mean Particle Size(um) (±S.D)*	DEE %*	DL %*
Fl	680± 0.68	47.45 ± 0.34	13.72 ± 0.48
F2	733± 0.57	53.65 ± 0.42	17.88 ± 0.64
F3	692± 0.61	61.15 ± 0.28	19.28 ± 0.84
F4	723±0.29	72.95 ±0.56	24.59 ±0.73
FG1	710± 0.64	49.48 ± 0.47	14.74 ± 0.63
FG2	768±0.38	56.34 ± 0.69	18.78 ±0.68
FG3	708±0.52	65.18 ± 0.48	21.29 ± 0.54
FG4	758±0.34	75.21 ± 0.41	25.04 ± 0.39

Data presented as Mean ± S.D. (n=3), n is the number of observation

SCANNING ELECTRON MICROSCOPY (SEM)

Surface morphology for the prepared microspheres was performed by using Scanning electron microscopy (FEI Quanta 200 MK2, Netherlands) without drug as well as drug before dissolution and after dissolution. The results depicted in Figure 4.[28]

Swellability study

Microspheres found to be swelled in water, 0.1N HCl, pH 6.8 phosphate buffer and phosphate buffer 7.4. The result of swellability index was described in Table 5.

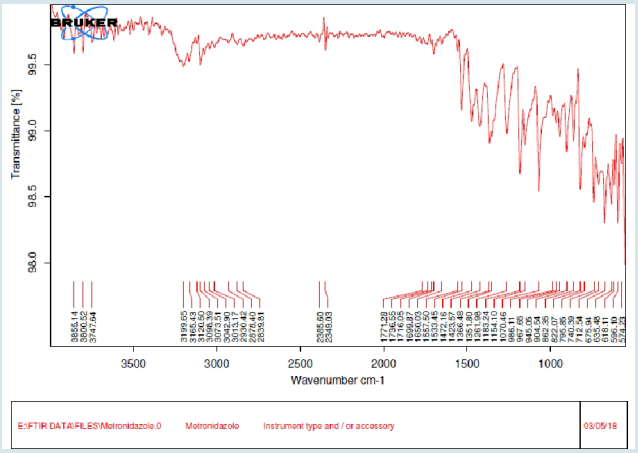


Figure 1: FT-IR Spectra of metronidazole pure drug.

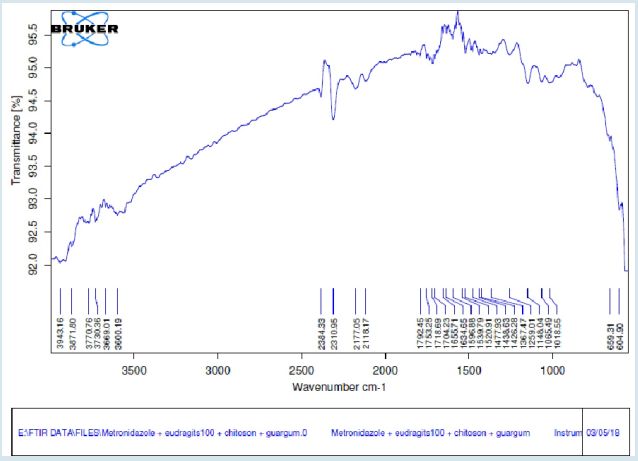


Figure 2: FT-IR Spectra of final formulation.

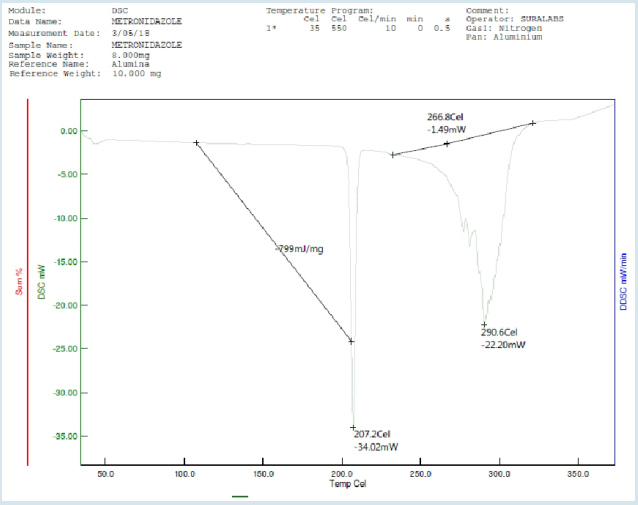


Figure 3: Differential scanning calorimetric spectra of pure drug.

Table 5: Swelling study of the prepared microspheres

Nature of the solvents	Swelling percentage of Formulations							
	Fl	F2	F3	F4	FG1	FG2	FG3	FG4
Distilled water	110	160	310	390	92	120	220	360
0.1NHC1	80	110	200	320	65	90	175	2 80
Phosphate buffer pH6.8	100	180	320	311	89	174	210	300
Phosphate buffer pH7.4	120	200	350	450	100	170	300	400

Table 6: *In vitro* drug release profiles of the prepared microspheres

HC1 PH 1.2	Time	Cumulative % Drug Release							
	(Hr)	Fl	F2	F3	F4	FG1	FG2	FG3	FG4
	0	0	0	0	0	0	0	0	0
	1	0	0	0	0	0	0	0	0
	2	0	0	0	0	0	0	0	0
PBS PH 6.8	3	1.59	1.98	0.97	0.92	1.58	2.19	1.51	0.14
	4	2.34	3.59	4.87	2.16	3.96	4.13	3.21	2.13
	5	8.72	9.89	10.21	8.06	9.06	10.02	10.33	3.41
	6	12.12	12.46	13.65	10.17	10.19	12.1	14.12	4.15
	7	14.12	16.32	17.56	13.17	14.2	15.12	18.13	6.1
PBS PH 7.4	8	16.14	18.09	19.44	17.8	19.6	21.48	24.83	11.76
	12	18.9	20.07	21.25	22.3	26.33	28.63	34.52	17.82
	16	65.46	72.42	78.28	57.9	67.81	76.15	70.2	60.44
	20	76.06	84.98	86.3	68.95	73.81	85.55	88.72	67.46
	24	79.9	86.43	88.81	79.1	82.12	91.82	93.26	75.35

Table 7: *In vitro* drug release kinetics analysis of the prepared microsphere

Formulation	Zero order		First order		Higuchi order		Korsmeyer	
code	Model		Model				Pappa	's
							Model	
	r^2	k_0	r^2	k_1	r^2	kh	r^2	n
Fl	0.914	9.23	0.865	0.114	0.861	34.61	0.959	1.221
F2	0.933	9.11	0.899	0.104	0.861	33.83	0.966	1.294
F3	0.946	9.19	0.902	0.109	0.865	33.96	0.970	1.291
F4	0.953	9.25	0.917	0.107	0.858	33.92	0.974	1.410
FG1	0.960	8.53	0.942	0.084	0.871	31.42	0.972	1.298
FG2	0.967	8.45	0.937	0.080	0.859	30.77	0.976	1.412
FG3	0.859	8.41	0.982	0.081	0.966	30.74	0.988	1.320
FG4	0.976	8.26	0.939	0.076	0.860	29.96	0.985	1.456

In-vitro drug release study

Drug release study was performed using USP dissolution rate test apparatus (Apparatus 1, 100 rpm, 37±0.5°C) for first 2h in 0.1 N HCl (900ml) and next 2h in pH 6.8 phosphate buffer and then in 7.4 Phosphate buffer up to 24h. The drug release is found to be 75.35% to 93.26% at the end of 24 h (Table 6, 7 and Figure 5). The rate of drug release rate found to be followed zero order kinetics and numerical data fitted into Koresmeyer-Peppa's model, whereas the value of n reaches above 1. This represents the case II and super case II transport, which indicates that the release is following zero order.[29]

In-vivo Studies

GIT distribution study

For GIT dissemination consider 100-150 weigh of pale skinned person rodent creatures were chosen. Chosen creatures were kept in an all around divided ventilated enclosures and kept up to solid with legitimate eating routine. The creatures were similarly partitioned into four gatherings and every gathering contain six creatures. The first aggregate utilized as the control. The second gathering got plain medication (dosage Figured according to the body weight of the rabbit). Third gathering creatures were taken as microspheres arranged without glutaraldehyde

and fourth gathering creatures will get microspheres arranged with glutaraldehyde. Computed creature measurements were controlled orally with the assistance of cannula. After 2, 4, 6 and 8 h, the creatures were sacrificed and stomach, small digestive tract and colon were secluded and homogenized with little measure of phosphate cushion pH 7.4 and after that centrifuged at 10,000 rpm for 5 min and supernatant was isolated. In the isolated supernatant, 1 mL of acetonitrile was included and kept for 30 min and filtered. The filtrates were examined for medication content by estimating the absorbance at most extreme wavelength of 320 nm (UV 1601, Shimadzu, Japan) against individual clear arrangement.

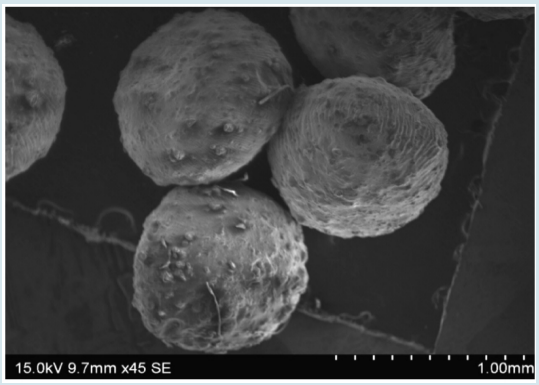


Figure 4: SEM study of selected formulation (FG4) with higher magnification.

The medication content in various parts of GIT was estimated at various time interims. All creature contemplate conventions were appropriately affirmed by the Samskruti College of Pharmacy, Kondapur Village, Ghatkesar Mandal, Medchal District, T.S., Approval No-CPCSEA/IAE/EXP/29/409/2017/EXP/82. Microspheres arranged with glutaraldehyde, were accepted to stay unblemished in upper GIT, i.e. the physiological condition of stomach and small digestive system, yet once they came to in the colon, they were followed up on by the bacterial pectinase chemical, which brought about the corruption of the frameworks and accordingly discharged the medication from the microspheres. These microparticulate frameworks have observed to be biodegradability property in the colon. *In vivo* considers show that most extreme grouping of medication (57.9%) was seen in stomach after oral organization and in consequent h, less measure of medication achieved the small digestive system and no medication was found in the colon. Just 26.4% of aggregate medication heap of ordinary measurements frame achieved the colon after 8 h (Figure 6). After 6 h of orally directed microspheres without glutaraldehyde indicated greatest medication content (41.6%) in the small digestive tract because of filtering of medication from microspheres on its swelling in gastrointestinal fluids and 37.5% into the colon after 8 h of its organization which is higher than the customary measurements shape (Figure 7). The microspheres arranged from glutaraldehyde were watched generally flawless in the upper piece of GIT (Figure 8). Around 1– 5% of the aggregate medication stack was discharged amid its travel through upper GIT (1– 5 h) and demonstrated the most extreme measure of medication in the colon (67.9%) after 8 h. The most extreme medication which is seen in the colon from microspheres arranged from glutaraldehyde could be

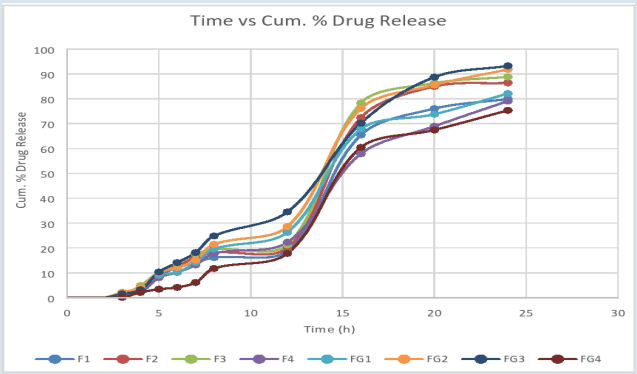


Figure 5: *In vitro* drug release study of prepared microspheres.

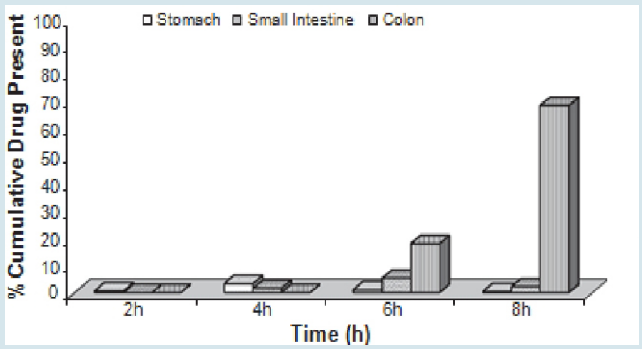


Figure 8: Bio distribution of during in various organs after oral administration of microspheres prepared with Glutaraldehyde.

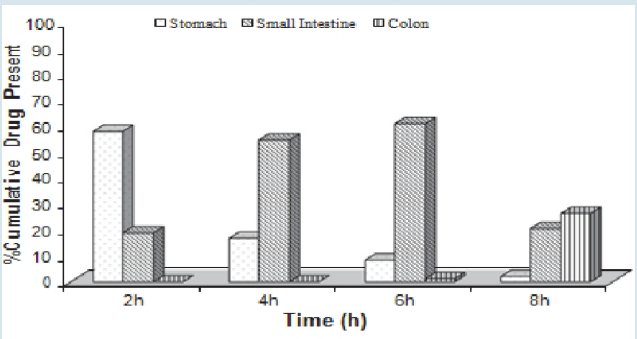
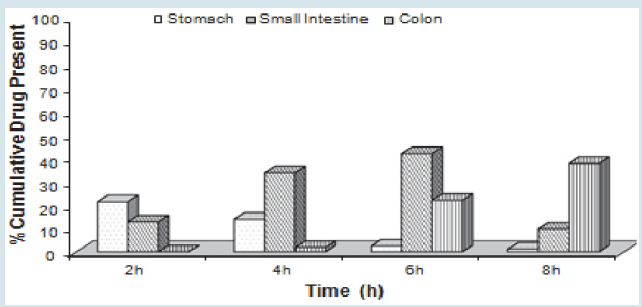


Figure 6: Bio-distribution of drug in various organs after oral administration of pure drug.



Figures 7: Bio distribution of drug in various organs after oral administration os microspheres having physically entrapped drug without Glutaraldehyde.

because of substance official of medication to polymer framework, which was debased by compounds discharged from colonic small scale flora and in this manner discharged the medication.[26]

CONCLUSION

Microspheres containing Metronidazole were prepared by ionic gelation method using Eudragit S 100, Chitosan, Guargum and Sodium alginate polymers. All the formulations were subjected for the drug content estimation and loading efficiency. The drug content was found to be uniform and reproducible in all the formulations. From the FT-IR analysis it was concluded that there is no significant drug and selected polymers showed any interaction. FG4 formulation showed maximum drug release that is up to 93.26% at pH 7.4. Therefore, the prepared microspheres formulations have significant improvement in absorption of drug in colon for successful treatment of the bacterial infection. It was found that a sufficient quantity of drug was delivered into colon from microspheres prepared with combination of drug with total polymers ratio 1:4 (FG4) when compared to that of the plain drug or other conventional formulations. So conjugation of drug with selected polymers (FG4 with 1:4 ratio) will be an effective delivery of the drug targeting to site specific with sustained delivery at the colonic area.

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CONFLICT OF INTEREST

The authors confirm that the content of the article has no conflicts of interest.

ABBREVIATIONS

CI: Carr's compressibility index; **DEE:** Drug entrapment efficiency; **DL:** Drug Loading; **FTIR:** Fourier infrared spectroscopy; **GIT:** Gastro intestinal tract; **HCl:** Hydrochloric acid; **MTZ:** Metronidazole; **SEM:** Scanning electron microscopy; **Vo:** Apparent volume.

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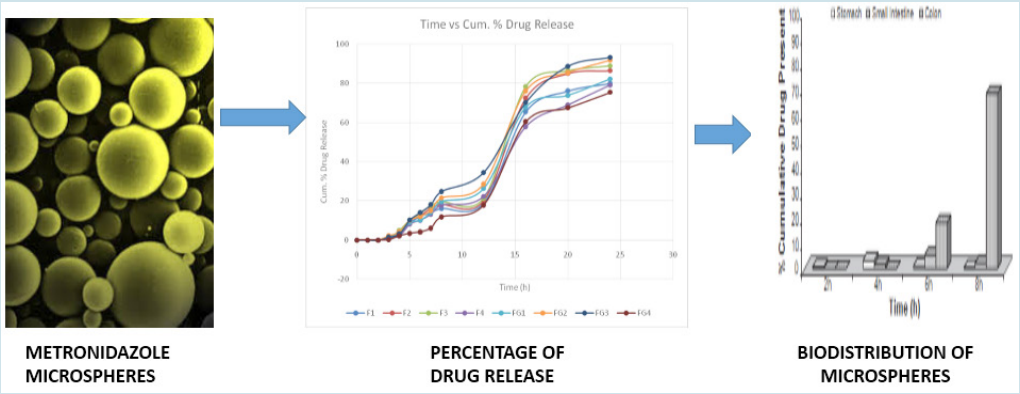
PICTORIAL ABSTRACT



SUMMARY

- Metronidazole microspheres were design by ionic gelation technique.Obtained microspheres particle size ranges from 36.77µm to 229.96µm with medication capture proficiency of 42– 99% w/w. Metronidazole microsphere kept up to maintain the medication discharge up to 12 h if there should be an occurrence of definition FG4. Medication discharge from the microspheres, swelling file and lightness establishes to be relies upon the grouping of chosen polymers in the polymer mix. Definitions containing low groupings of Guargum, Chitosan, Eudragit S-100 and sodium alginate (1:1) demon¬strating shorter drifting slack time and quicker medication discharge and the other way around. FG4 formulation showed maximumdrug release that is up to 93.26% at pH 7.4. Therefore, the preparedmicrospheres formulations have significant improvement in absorption of drug in colon for successful treatment of the bacterial infection.

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