Original Research Article

# A PROMISING APPROACH TO PROVIDE APPROPRIATE COLON TARGET DRUG DELIVERY SYSTEMS OF METRONIDAZOLE: DEVELOPMENT AND EVALUATION

**Bijay Kumar Sahoo[*1], Sidheswar Prasad Pattajoshi[2], Sandhyarani Pattajoshi[2]**

[1]Dept of Pharmaceutics, IMT Pharmacy College, Gopalpur, Puri, Odisha

[2]Dept. of Pharmaceutics College of Pharmaceutical Sciences, Baliguali, Puri, Odisha

**Corresponding author:** Dr.Bijay Kumar Sahoo, Professor, IMT Pharmacy College, Puri, Odisha

**Conflict of interest statement:** No conflict of interest

## Abstract

The aim of present study was to develop colon targeted system for Metronidazole using guar gum and xanthan gum. Tablet matrices containing 10–60% of tablet weight of guar gum (F1–F6) were prepared by direct compression and subjected to *in vitro* release studies to explore their sustained release in the colon. Various release retarding synthetic and natural polymers, namely, hydrogenated castor oil, hydroxypropyl methyl cellulose, xanthan gum, and ethyl cellulose, Eudragit RL 100, were incorporated to modify the drug release rate from the guar gum matrix tablets. Matrix tablets were enteric coated with hydroxypropyl methyl cellulose phthalate as an enteric polymer. Various synthetic and natural polymers were incorporated to F6 to modify the drug release rate. Different 15 matrix tablet formulations (F6–F20) were enteric coated with hydroxypropyl methyl cellulose phthalate. The *in-vitro* drug release study was undertaken at 37±0.5°C in 0.1N HCl for 2 h; followed by pH 7.4 phosphate buffer (3h) finally in, simulated colonic fluid pH 6.8 phosphate buffer 20 h. The formulation F6, F13 and F20 showed promising sustained release results having median dissolution time (MDT) values: 8.25, 7.97, and 7.64, respectively. When studies were continued in colonic fluids, matrix tablets released almost 100% drug. whereas, Metronidazole enteric formulations did not release drug in stomach and small intestine, but delivered drug to the colon resulting in slow absorption of the drug and making drug available for local action in the colon.

**Keywords:** Colon Target Delivery, Guar gum, Metronidazole, Enteric coated, Tablet Matrices

## Introduction

Colon target drug delivery system (CDDS) is highly desirable for local treatment of a variety of bowel diseases such as ulcerative colitis, Crohn's disease, amoebiasis, colonic cancer, local treatment of colonic pathologies, and systemic delivery of protein and peptide drugs. The colon is believed to be a suitable absorption site for peptides and protein drugs for the following reasons: (i) less diversity and intensity of digestive enzymes, (ii) less proteolytic activity of colon mucosa leading to better protection from hydrolysis and enzymatic degradation in duodenum and jejunum, (iii) greater systemic bioavailability [**1-2**] and (iv) long colon residence time (5 days) and high responsiveness to absorption enhancers. The best alternative approach for colon specific drug delivery is the use of the carriers that are degraded exclusively by colonic bacteria. The microflora of the colon is in the range of 1011-1012 CFU/ml consisting mainly of anaerobic bacteria e.g. Bacteroides, Bifidobacteria, Eubacteria, Clostridia, Enterococci, Enterobacteria and Ruminococcus etc. Because of the presence of the biodegradable enzymes only in the colon, the use of the biodegradable polymers for colon specific drug delivery seems to be more site-specific approach as compared to the other approaches. These polymers shield the drug from the environments of stomach and small intestine and are able to deliver the drug to the colon. Natural polysaccharides are now extensively used for the development of solid dosage forms for colon drug delivery. A large number of polysaccharides have already been studied for their potential as colon-specific drug carrier systems, such as chitosan, pectin, chondroitin sulphate, cyclodextrin, dextrans, guar gum, inulin, amylose, and locust bean gum [**3-5**]. Guar gum and pectin are reported to be potential carriers for colon-specific drug delivery. These studies have shown the drug release retarding property of guar gum in the upper GIT and its degradation by the anaerobic bacteria in the colon [**6-7**]. It is effective for dracunculiasis, giardiasis, trichomoniasis, and amebiasis. It is an option for a first episode of mild-to-moderate *Clostridium difficile* colitis if vancomycin or fidaxomicin is unavailable.

For a formulation to act as an effective colon specific drug delivery system, the primary condition is that a minimum amount of drug should be released in the environment of the upper gastrointestinal tract, *i.e.*, in stomach and small intestine. The normal transit time in the stomach is 2 h (though this may vary), while in the small intestine it is relatively constant and is around 3 h .The usual colonic transit time varies from 20–30 h .This, for a dosage form to be effective as a colon drug delivery system, the drug release is required to be retarded in the upper GIT conditions. Thereafter, the drug release should be complete within the next 20–30 h [8-10].

Therefore, the present study is an approach to develop an appropriate sustained release colon target drug delivery tablets of this drug which would minimize its inactivation in the upper part of the gastrointestinal tract. These delayed release tablets are designed to improve the efficacy of the drug by concentrating the drug molecules where they are absorbed and thus would ensure lower dosing and less systemic side effects. In addition, CDDS of Metronidazole is suggested to be given to patients with Irritable bowel disease. Metronidazole matrix and enteric-coated tablets based on natural polysaccharide, namely, guar gum as a carrier, were formulated. Tablets matrix containing different concentrations of guar gum was prepared by direct compression method and subjected to *in vitro* release studies to find out the efficacy of guar gum in providing sustained release of the drug in the colon. Various release retarding synthetic and natural polymers, namely, hydrogenated castor oil, hydroxypropyl methyl cellulose, xanthan gum, ethyl cellulose, and Eudragit RL 100, were incorporated to modify the drug release rate from the guar gum matrix tablets. Matrix tablets were enteric coated with hydroxypropyl methyl cellulose phthalate as an enteric polymer.

The present study explores the comparative utility of the above polymers in developing a suitable dosage form, exhibiting a minimum drug release in the upper regions of the GIT in order to provide targeted drug delivery to the colon. For this purpose, varying concentrations of polymers were applied and the effect of the coating on drug release and site specifity was evaluated in vitro.

### Experimental

### Materials & Methods

Metronidazole IP was gift sample obtained from M/s J. B Chemical & Pharmaceutical Ltd, Mumbai. Microcrystalline Cellulose (AvicelTM PH-102), Guar gum and Xanthan gum were procured from S D Fine Chemical Ltd, Mumbai. Magnesium stearate and Talc was also incorporated in tablets as glidant and lubricant and was procured from S D Fine Chemical Ltd, Mumbai. Other excipients used to prepare tablets were of standard pharmaceutical grade and all chemical reagents used were of analytical grade.

**IR Study**.: IR spectra of physical mixtures (1: 1) of Metronidazole and various excipients, as well as the drug alone, were performed to find out any possible drug-excipients interaction by KBr pellet method using Perkin-Elmer FTIR series (model-1615) spectrophotometer between 4000 and 450 $cm^{-1}$.

### Preparation of Metronidazole matrix tablets:

Metronidazole sustained release tablets were prepared by direct compression technique using guar gum as the main matrix forming materials in different concentrations: 20, 30, 40, 50, and 60% w/w of tablet (300 mg)[11]. Other matrix forming materials, namely, HCO, HPMC, EC, Eud RL100, and XG, were added to the guar gum matrix to modulate the drug release. All formulations are listed in Table 1. The calculated amount of the drug and of each ingredient in the formulation was mixed thoroughly by geometric addition in a mortar and then compressed using single punch tablet machine (Erweka, Germany) using 10 mm flat punch under a pressure of 10 Kg. Enteric coating of the prepared matrix tablets was performed using 10% w/v solution of HPMCP in acetone: water (95 : 5 v/v) mixture by dipping methods and 15 coats were applied.

**Evaluation of Fabricated Matrix Tablets.** All prepared matrix tablets were evaluated for its hardness, friability, drug content, and thickness according to official methods [12-13].

### In Vitro Dissolution Study.

*In vitro* dissolution study of all tablet formulations was carried out using USP apparatus I (Electro lab,India). The test was performed in 500 mL of 0.1 N HCl for 2 hours then in phosphate buffer pH 6.8 at a temperature of 37 ± 0.5°C. The stirring speed was kept constant at 100 rpm. 5 mL samples were withdrawn at predetermined time intervals of 2, 3, 4, 5, 6, 8, and 24 hours and replaced with preheated fresh dissolution medium. The samples were assayed spectrophotometrically at $\lambda_{max}$ of 281 nm for drug content. All the dissolution tests were run in triplicate and the mean values ± standard deviation (SD) of the percentage cumulative drug release were plotted against time. The results were statistically analyzed using two-way analysis of variance (ANOVA) tests with Tukey's multiple comparison post hoc (Graphbad prism 6 program) to test the significance at a 5% significance level. Statistical difference dealing ($P < 0.05$) was considered. Excipients used in this study did not interfere in the spectrophotometric reading.

**Mathematical modeling of release kinetics:**

The *in vitro* drug release data were fitted to various release kinetic models *viz.*, zero order, first order  and Higuchi model. These models fail to explain the drug release mechanism due to swelling (upon hydration) along with gradual erosion of the matrix. Therefore the dissolution data was also fitted to the well known exponential equation *i.e.* Korsemeyer-Peppas model **[14].**

Zero order kinetic model: $Qt = Q0 + K0 . t$

First order model: $\ln Qt = \ln Q0 + K1 . t$

Higuchi model:   $Qt = K .\sqrt{t}$

*MechanismofDrug  Release.*  Korsmeyer  et  al. Korsemeyer-Peppas model:  $Mt/ M\infty = Kt^n$,

Where $Mt/M\infty$ is fraction of drug released at time $t$, $K$ is the release rate constant incorporating structural and geometric characteristics of the tablet, and $n$ is the release exponent. The    value is used to characterize different release mechanisms A plot of log cumulative % drug release versus log time was made. Slope of the line was $n$. The $n$ value is used to characterize different release mechanisms as given in Table 2, for the cylindrical shaped matrices. Case II generally refers to the erosion of the polymeric chain and anomalous transport. Non-Fickian refers to a combination of both diffusion and erosion controlled-drug release **[14].**

A value of n = 0.5 indicates case I (Fickian) diffusion or square root of time

kinetics, 0.5 < n < 1 anamolous (non-Fickian) diffusion, n = 1 indicates case II transport and n > 1 super case II transport.

**Table 1**: *Composition of Various Formulations of Metronidazole matrix tablets prepared by direct compression. Amount of ingredients in mg.*

| Formulation Code | Drug | GG | XG | HPMC | EC | HCO | Eudra.RL | MCC |
|---|---|---|---|---|---|---|---|---|
| F1 | 100 | 60 | – | – | – | – | – | 135 |
| F2 | 100 | 90 | – | – | – | – | – | 105 |
| F3 | 100 | 100 | – | – | – | – | – | 75 |
| F4 | 100 | 120 | – | – | – | – | – | 45 |
| F5 | 100 | 150 | – | – | – | – | – | 15 |
| F6 | 100 | 180 | – | – | – | – | – | 95 |
| F7 | 100 | – | 100 | – | – | – | – | 95 |
| F8 | 100 | 50 | 50 | – | – | – | – | 95 |
| F9 | 100 | 158 | – | 22 | – | – | – | 15 |
| F10 | 100 | 134 | – | 44 | – | – | – | 15 |
| F11 | 100 | 114 | – | 66 | – | – | – | 15 |
| F12 | 100 | 85 | – | 90 | – | – | – | 20 |
| F13 | 100 | 130 | – | – | – | 45 | – | 20 |
| F14 | 100 | 100 | – | – | – | 75 | – | 20 |
| F15 | 100 | 85 | – | – | – | 90 | – | 20 |
| F16 | 100 | 130 | – | – | 45 | – | – | 20 |
| F17 | 100 | 100 | – | – | 75 | – | – | 20 |
| F18 | 100 | 85 | – | – | 90 | – | – | 20 |
| F19 | 100 | 85 | – | – | – | – | 90 | 20 |
| F20 | 100 | 130 | – | – | – | – | 45 | 20 |

*Mixture of magnesium stearate (Mg. st) and talc in the ratio of 2:1 was used as a lubricant equivalent to 5 mg.*

**Mean Dissolution Time.**

Due to the difference in drug release kinetics, the constant $k$, though one of the measures of release rate, should not be used for comparison. Therefore, to characterize the drug release rates in different experimental conditions, another dissolution parameter used for comparing the formulations was mean dissolution time (MDT). This is calculated from the amount of drug released to the total cumulative drug.MDT is a measure of the rate of the dissolution process: the higher the MDT, the slower the release rate. The mean dissolution time (MDT) is the mean time for the drug to dissolve under *in-vitro* dissolution conditions. MDT is a model-independent method and is suitable for dosage forms having different mechanisms

of drug release **[15-16].** This parameter helps to characterize the drug release profile and enables comparison of drug release rates from the various formulation and is calculated using following Equation

$$MDT = \frac{\sum_{j=1}^{j=n} t_j \triangle M_j}{\sum_{j=1}^{j=n} \triangle M_j}$$

where $j$ = Dissolution sample number; $t_j$ = midpoint of the jth time period (easily calculated with ((t + t-1)/2)

and $\triangle M_j$ = additional amount of drug dissolved between $t_j$ and $t$-1.

## Results and Discussion

Since guar gum can be compressed directly in the presence of directly compressible materials such as MCC, Metronidazole tablets were prepared by direct compression technique for its simplicity.

### IR Spectroscopic Studies.

*The* IR spectra of pure drug and physical mixtures of the drug and different excipients are shown in Figures 1 and 2. All the characteristics spectral bands of the drug were not significantly affected in the physical mixture of the drug and excipients. They were retained at their respective positions in the IR spectra of drug-excipient physical mixtures. No significant shift in the position of the characteristics bands was observed indicating absence of interaction between Metronidazole and the selected tablet excipients in the physical mixtures.

### Evaluation of Physicochemical Parameters of Prepared Tablets.

The physical properties of different batches of developed matrix tablets were studied. The thickness of the tablets ranged from 5.20 to 5.40 mm. The hardness of the tablets of all the formulations ranged from 8.2±0.5 to 9.8± 0.1 kg/cm$^2$. Friability test indicated that the percentage loss was less than 1% (0.63 ± 0.00 to 0.76 ± 0.017). The results of hardness and friability tests denoted that the tablets were hard enough to withstand tablet handling during the study. Drug content was in the range of 99.03 ± 1.0 – 99.70±0.39%. Weight variation before coating ranged from 298.7±1.75 to 289±1.04 mg while after coating ranged from 358.6±1.52 to 359±1.56 mg.

### In Vitro Release Study.

The present study was aimed at developing novel matrix tablet of Metronidazole for colon targeting using guar gum as a matrixing agent. The release of drug depends not only on the nature of matrix but also upon the drug polymer ratio. The percentage of drug released from guar gum matrix tablets reduced in the physiological environment of stomach and small intestine. Majority of

drug was released in the physiological environment of colon.
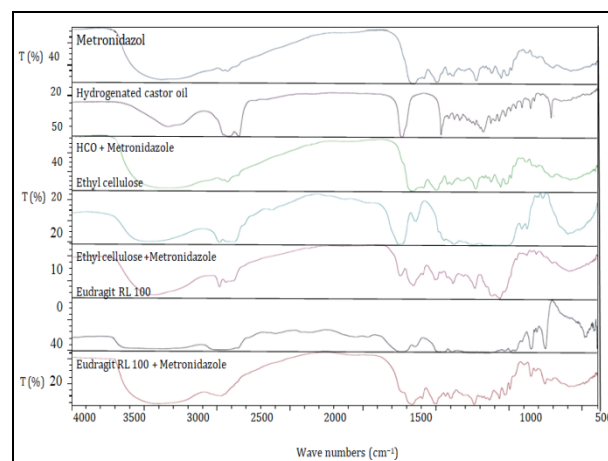


**Figure 1**: *IR spectra of Pure drug ( Metronidazole), HCO, EC, Eudragit RL 100, and 1:1 physical mixtures of the drug and each excipient*.
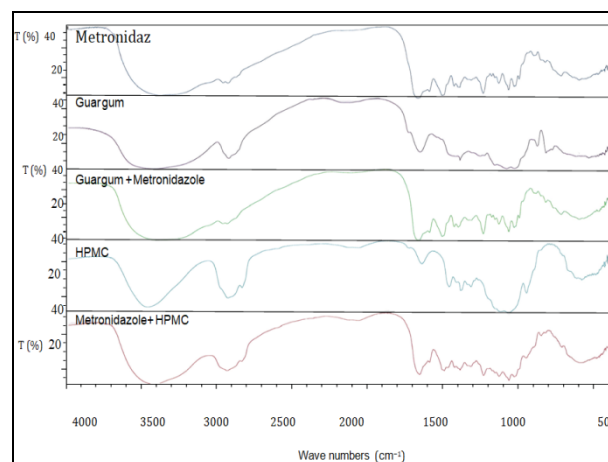


**Figure 2**: *IR spectra of Metronidazole, GG, HPMC, and 1:1 physical mixtures of the drug and each excipient.*

When matrices containing swellable polymers are exposed to dissolution medium, tablet surface becomes wet and hydrated to form a gel layer. The initial release of drug from these matrices occurs by the drug dissolution in the water penetrated into the matrix. The overall drug release from these matrices is governed by hydration, gel layer formation, and drug diffusion into the gel layer and to the dissolution media [**17-18**]. Polymer erosion also plays a major role in releasing drug from these matrices. These considerations indicate that hydrophilic polymers have the potential to sustain the release of drug from matrix tablets. During the dissolution process, a general trend was observed in all the formulations; that is, an increase in polymer concentration resulted in the reduction in amount of drug released [**19**]. An increase in the polymer concentration causes increase in viscosity of the gel as well as the formulation of gel layer with longer

diffusional path. This could cause a decrease in effective diffusion coefficient of drug and therefore a reduction in drug release rate. The *in vitro* cumulative percent drug released versus time profiles of all the tablet formulations is shown in Figures 3, 4, 5, 6, 7, 8, and 9.

The release profiles of formulations F0 to F6 are presented in Figure 3. All formulations released their drug content in acidic pH to various extents depending on guar gum concentration. Increasing the guar gum concentration from 20% w/w (F1) to 60% w/w (F6) reduced significantly the drug release in pH 1.2 from 58% to 22%, respectively, after 2 hrs (p < 0.0001). The obtained results can be explained as when the guar gum matrix tablets of Metronidazole come into contact with the dissolution medium, they take up water and swell, forming a gel layer around the matrix. Then the dissolved drug diffuses out of the swollen guar gum matrix at a rate determined by the amount and viscosity of guar gum in the tablet formulation. Sustained release was displayed by all formulations in phosphate buffer (pH 6.8). The percentage of drug release at the end of 24 hrs of dissolution test (pH 6.8) ranged from 82% (F1) to 28% (F6) with p < 0.0001. The release retarding, matrix forming gum was succeeded to sustain drug release over a period of 24 hrs. Formulations F3, F7, and F8 were prepared to study the influence of the hydrophilic xanthan gum on the release of the water soluble of drug. F3 and F7 matrix tablets contained 33.33% w/w of guar gum or xanthan gum, respectively (Figure 4), whereas F8 containing guar and xanthan gums in the ratio of 1 : 1 was prepared to examine the synergistic or antagonistic effect of both polymers on drug release. Formulation F7 released about 89.96% of its drug content, whereas F3 released 44.89% of the drug at the end of 2 hrs of drug dissolution test (p < 0.0001). Since XG is present predominantly in an unionized state at low pH, this results in absence of charged molecules which prevented hydration. Consequently, intermolecular and intra-molecular attraction was suppressed leading to inhibition of xanthan hydrogel network formation. Thus increased drug release in pH 1.2 could be explained by the prevention of gel formation of xanthan gum [20].

Guar gum gives pH-independent drug release due to its nonionic nature. it is not affected by ionic strength or pH. GG had the potential as a release retardant for the water soluble drug as xanthan gum due to gel formation. In addition the release profile of F8 denoted the synergistic effect of the two gums. This could be attributed to the stronger hydrogen bonding between the carboxyl groups of the Xanthan and the hydroxyl groups of guar gum, leading to stronger physical cross-linking between the polymers. To overcome the problem of drug release in the acidic pH, the tablet matrix was coated with the enteric polymer HPMCP. Figure 5

demonstrates the release profiles of coated and uncoated tablet matrices of F4, F5, and F6. It can be noticed that the release of the Metronidazole from coated tablets was completely blocked in pH 1.2 followed by its faster release in pH 6.8 compared to the uncoated tablets (Figure 5). These formulations were selected for further studies because they showed the slowest drug *in vitro* release rates. Drug release rate could be expected to increase *in vivo* as a result of biodegradation of guar gum by the bacteria present in colon. Many studies reported that the drug release in rat cecal content could be increased to the two or fourfold of its value in presence of the colon bacteria [21-22]. Based on this consideration, formulation for colon target that showed the slowest drug release *in vitro* would show a reasonable sustained release *in vivo*. Therefore formulation F6 was used for further study. Although coated tablet matrix of formulation F6 succeeded to sustain drug release over a period of 24 hrs, yet it failed to comply with the USP official limits of sustained drug release.

The drug release rate was above the official limits at the specified time intervals. After 1 and 4 hours of dissolution test, the sustained release matrix released 45% and 48% of its drug content, respectively. These drug release percentages were above the official limits which are not more than 25% and 40% after 1 and 4 hours of dissolution test, respectively. Therefore, combination of release retarding polymer (guar gum) and release modifying agents in the formulation of matrix tablets was recommended to modulate the drug release from 60% w/w guar matrix tablet (F6). All the tablets containing different concentrations of each release modifier (F9–F18) were coated with HPMCP polymer to prevent the release of the drug in acidic pH.

HPMC is hydrophilic cellulose ether, which is used as a retarding polymer in swellable matrices .Figure 6 shows the drug release from guar matrix tablets containing different concentrations of HPMC K4M. It is evident that, in pH 6.8 as the HMPC concentration increased from 7.3 to 20% w/w in F9 to F11, respectively, the drug release extent decreased significantly (p < 0.0001) due to faster water absorption capacities. The high water absorption capacities led to a more rapid swelling resulting in the formation of a gel layer with a longer diffusion path and high gel strength which could cause a decrease in the diffusion coefficient of the drug. Therefore a reduction in the drug release was observed.

Matrix tablet formulations F9, F10, and F12 containing 7.3, 14.7, and 22% of HPMC, respectively, showed a faster drug release from 2 to 3 hrs of the release experiment, followed by a slower release from 3 to 24 hrs. Sucha biphasic release pattern may be beneficial in providing the initial therapeutically effective plasma

concentration followed by an extended plasma concentration. The drug present on the surface of the matrix tablet might have resulted in the initial fast release of the water soluble drug Metronidazole from the formulation. In addition the faster water uptake by HPMC polymer on the surface, leading to formation of loose gel which eroded quickly and increase the diffusion coefficient of the drug from the guar matrix tablets. When the HPMC gel layer on the surface of the tablet eroded, the porosity of tablet increased and facilitated the access of further penetration of the dissolution medium within the tablet [23]. Thus the presence of low concentration of HPMC K4M (7.3% w/w) increased the drug release rate compared to F6. Further increase in concentration of HPMC to 14.7 and 20% w/w in F10 and F11, respectively, reduced the drug release rate compared to F9 ($p < 0.05$) but still higher than F6 ($p < 0.05$). Increasing the concentration of polymer to 30%w/w resulted in a slower drug release rate compared to F6 at the initial stage of dissolution test up to 5 hours, and then the drug release was increased exceeding that from F6 ($p < 0.05$). This can be explained by the following: at the initial stage of dissolution test, in presence of high concentration of HPMC, the fast water uptake capacity leads to rapid formation of a strong gel layer with a longer diffusion path which could cause a reduction in the drug release . Thus HPMC acted as a synergistic gel forming agent which increased the drug release retarding effect of guar gum up to 5 hrs. Then the loose gel of HPMC underwent faster erosion than that of guar gum leading to increased diffusion coefficient and drug release rate [24].

The effect of hydrogenated castor oil on the drug release from the guar matrix tablet (F13 to F15) is shown in Figure 7. HCO is extremely hydrophobic in nature with lower wet- tability. It is obvious that increasing the concentration of the hydrophobic polymer in the guar based matrix tablets resulted in a significant decrease in the drug release rate. The hydrophobic nature of the HCO decreased the wettability of the tablet and thus decreased the release of drug present on the tablet surface. In addition HCO being hydrophobic acted as a barrier to water penetration into the tablets, leading to retardation in water absorption, swelling, hydration, and gel formation by guar gum. The extent of retardation in gel formation depended on the concentration of HCO. The release profiles (Figure 7) indicated that increasing the concentration of HCO from 0 to 30% w/w, (F6 to F15) respectively, reduced the drug release rate from 45 (F6) to 34.16% (F15) after 3 hrs ($p < 0.05$). However, increasing the concentration of HCO from 15% w/w (F13) to 30% w/w (F15) increased the drug release rate from 29.16 to 34.16%, respectively. This can be explained by the following: as the concentration of HCO increased, the extent of hydropho-bicity of the matrix increased,

leading to decrease in the rate and intensity of gel formed by guar gum. Thus, the rate and strength of the gel formed in F13 containing the least concentration of HCO (15% w/w) were higher than those in F14 and F15, containing 25 and 30% w/w HCO, respectively.

Similar results were obtained using ethyl cellulose polymer as a drug release modifier (Figure 8). It can be seen that incorporation of EC in guar matrix tablets F16, F17, and F18 resulted in reducing the drug release rate from guar matrix tablet F6. The obtained results could be due to the hydrophobic nature of EC and its erosion characteristics.The decrease in drug release rate may be attributed to the net result of increased hydrophobicity of the matrix and slow erosion of polymeric content of the matrix tablets .

Incorporation of Eudragit RL 100 in the drug-guar gum matrix (F19 and F20) resulted in a significant decrease in the drug release rate ($p < 0.05$) as shown in Figure 9. Eudragit RL 100 is cationic copolymer of methacrylate with quaternary ammonium groups. It is inert resins and insoluble at physiologic pH but have swelling properties. It is compressible and erodible and due to the presence of 10% quaternary ammonium group the Eudragit matrix is permeable. Thus when the matrix tablet was placed in the dissolution medium the presence of Eudragit RL facilitated the permeation of the dissolution medium into the matrix tablet containing guar gum. The gum rapidly hydrated forming a gel layer inside the matrix and on the matrix surface. The hydrogelation of the gum slowed down the drug release rate from the matrix [25].

Based on drug release rate studies, the polymers used as release modifiers can be arranged, according to their release retarding efficacy, in ascending order as XG < EC < HPMC < HCO < Eudragit RL 100.
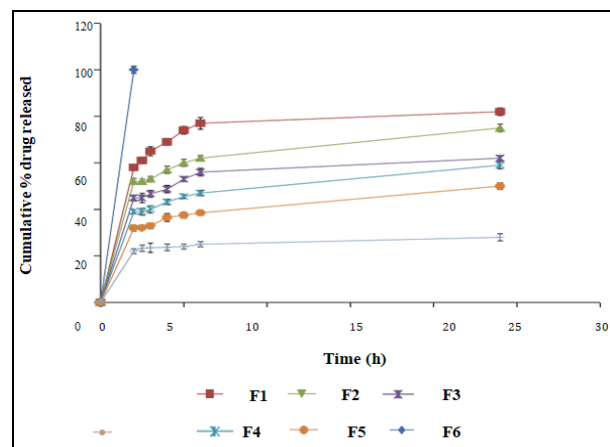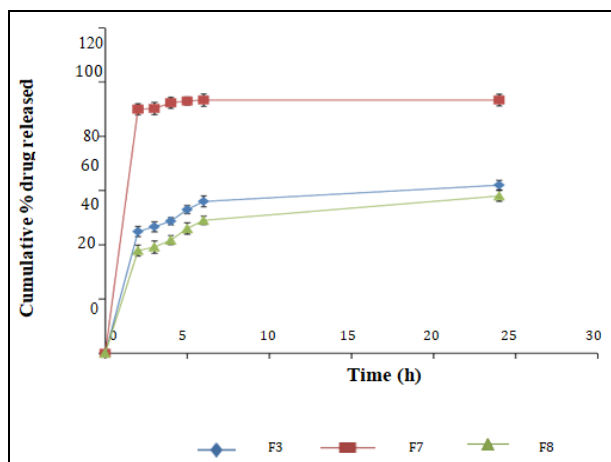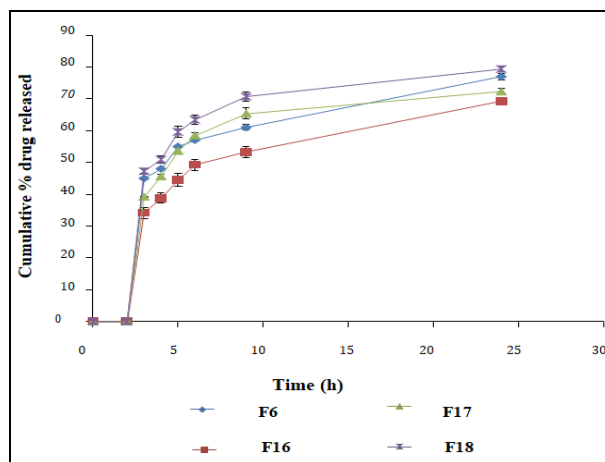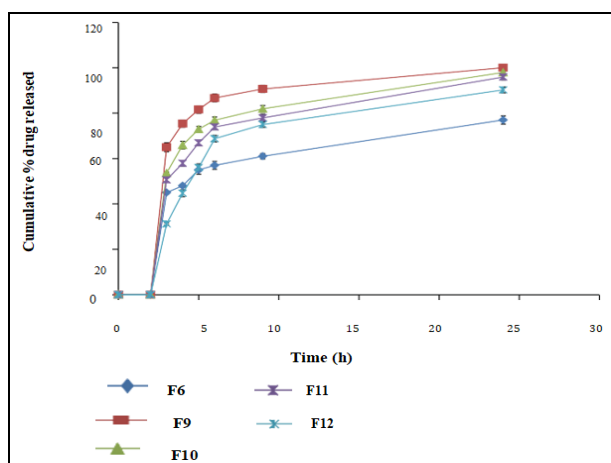


**FIGURE 3**: *Release profiles of Metronidazole from formulations F1 to F6 containing different concentrations of guar gum (0 to 60% w/w)*
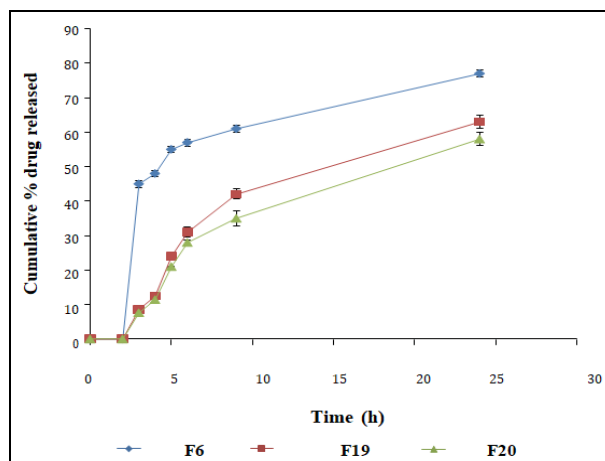
**Figure 4**: *Release profiles of Metronidazole from coated tablet formu- lations F3 (guar gum matrix), F7 (xanthan gum matrix), and F8 (containing mixture of guar and xanthan gums 1:1).*
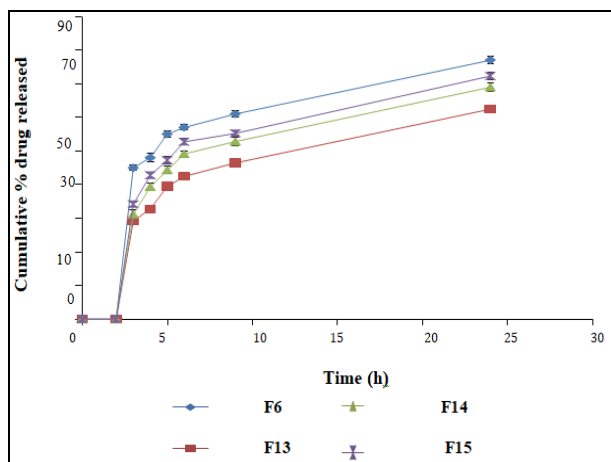


**Figure 5***: Release profiles of Metronidazole from coated tablet formulations F6, F9–F12 containing different ratios of guar gum and HPMC*



**FIGURE 6**: Release profiles of Metronidazole from coated tablet formulations F6, F13–F15 containing different ratios of guar gum and hydrogenated castor oil.



**FIGURE 7**: *Release profiles of Metronidazole from coated tablet formulations F6, F16–F18 containing different ratios of guar gum and EC*



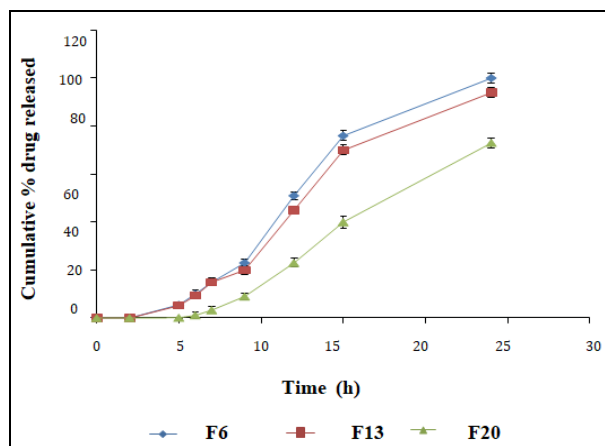**FIGURE 8**: *Release profiles of Metronidazole from coated tablet formulations F6, F19, and F20 containing different ratios of guar gum and Eudragit RL 100.*



**FIGURE 9**: *Release profiles of Metronidazole from coated tablet formulations F6, F13, and F20 containing different ratios of guar gum and Eudragit RL 100 in pH 1.2 for 2 hours, in pH 7.4 for further 3 hours, and then in pH 6.8 till the end of 24 hours.*

TABLE 2: Results of kinetics study.

Correlation coefficients ($r^2$)

| Formula | Zero order | First order | Hixson | Higuchi's model | Korsemayer'sslope ($n$) | $T70\%$ (hr) | MDT ( hr) | DR % | DE9h % after 9 hrs |
|---------|-----------|-------------|--------|-----------------|------------------------|-------------|-----------|------|--------------------|
| F6 | 0.952 | 0.537 | 0.933 | 0.982 | 0.43 | 21.82 | 8.25 | 61.00 | 29.71 |
| F9 | 0.820 | 0.507 | 0.833 | 0.883 | 0.52 | 3.72 | 4.96 | 90.67 | 34.00 |
| F10 | 0.882 | 0.529 | 0.847 | 0.932 | 0.50 | 4.80 | 6.28 | 81.91 | 31.36 |
| F11 | 0.902 | 0.540 | 0.870 | 0.947 | 0.51 | 5.68 | 6.74 | 77.91 | 28.16 |
| F12 | 0.826 | 0.583 | 0.771 | 0.889 | 0.56 | 8.40 | 4.83 | 74.99 | 31.16 |
| F13 | 0.947 | 0.572 | 0.917 | 0.978 | 0.60 | >24 | 7.97 | 46.45 | 27.89 |
| F14 | 0.923 | 0.565 | 0.932 | 0.962 | 0.57 | >24 | 7.19 | 52.82 | 28.68 |
| F15 | 0.926 | 0.558 | 0.887 | 0.961 | 0.56 | 23.25 | 7.40 | 55.21 | 28.66 |
| F16 | 0.940 | 0.559 | 0.912 | 0.974 | 0.60 | >24 | 7.95 | 53.32 | 28.85 |
| F17 | 0.825 | 0.535 | 0.795 | 0.890 | 0.54 | 23.23 | 4.37 | 65.32 | 33.87 |
| F18 | 0.864 | 0.528 | 0.839 | 0.920 | 0.52 | 8.92 | 5.05 | 70.76 | 32.97 |
| F19 | 0.923 | 0.552 | 0.841 | 0.963 | 0.69 | >24 | 6.80 | 42.00 | 25.00 |
| F20 | 0.940 | 0.553 | 0.859 | 0.945 | 0.58 | >24 | 7.64 | 23.00 | 22.63 |

**Kinetic Studies.**

The values of the release exponent ($n$), mean dissolution time, zero-order, first-order, Higuchi release models, and time of 70% drug release for different formulations are presented in Table 2. In the present study the release profiles were not linear suggesting that the drug release from the formulations was not zero order that was confirmed by $R^2$ values of 0.820 to 0.947.The release did not fit to first-order model that was also ensured by the low $R^2$ values of 0.507 to 0.583. Hixon-Crowell model showed $R^2$ values in the range of 0.771 to 0.933. It was observed that the *in vitro* release profiles of drug from all these formulations can be best expressed by Higuchi equation as the correlation coefficients showed the higher values ($R^2$: 0.883 to 0.982) (Table 2). Higuchi's kinetics explains why the drug diffuses at a comparatively slower rate (0.048–0.100) as the distance for diffusion increases. To confirm the diffusion mechanism the data was fitted into Korsemeyer-Peppas equation. All the formulations showed slope ($n$) values ranging from 0.30 to 0.69. The $n$ values for formulation F6 was 0.43 indicating quasi-Fickian diffusion. The other formulations showed $n$ values higher than 0.45 indicating anomalous diffusion or non-Fickian diffusion. Anomalous diffusion or non-Fickian diffusion refers to a combination of both diffusion and erosion controlled-drug release.

The release rate and $T_{70\%}$ values of these formulations can be considered as a function of the type and concentration of the retarding polymer used. The differences in drug release rate and $T_{70\%}$ among the different formulations are confirmed from their MDT data. MDT value is used to characterize the drug release rate from the different formulation and the retarding efficacy of the polymers. It is obvious that guar gum in 60% w/w concentration showed the higher value of MDT indicating high polymer retarding efficacy. In general, polymers used as release modifiers in this study can be arranged as an efficient polymer based on MDT as HCO >HPMC > Eud RL 100 > EC. It was also observed that on using the same modifier MDT values varied according to the concentration and accordingly the ratio between the release retarding and release modifier polymers, for example, in case of HPMC as the concentration increased the MDT increased except for F12 of the highest concentration of polymer, showed the least value of MDT. The similar results were observed in case of EC (Table 2).

These observations may be explained by the net mechanism of drug release influenced by guar gum and the modifier type and ratio. Formulations F6, F13, and F20 were selected for further study depending on their MDT values 8.25, 7.97, and 7.64, respectively. They also showed promising results as sustained release formulations. They were subjected to further examination of the drug release in different pHs along the passage of the formulations through the GIT. The drug release rate was determined in pH 1.2 for 2 hrs

followed by pH 7.4 for further 3 hrs then in pH 6.8 up to 24 hrs. The drug release was blocked in pH 1.2 due to HPMCP coating. After 5 hours of the release study, the drug released in pH 7.4 was 5.5, 5.2%, and 0.1% from F6, F13, and F20, respectively (Figure 10). In pH 6.8, formulations F20 showed significant reduction in drug release rate compared to formulations F6 and F13 ($P < 0.05$). The three formulations showed sustained release characteristics over 24 hours.

## Conclusion

The present investigation was carried out to develop colon targeted drug delivery system for Metronidazole using guar gum and Xanthan gum as carrier for an effective of safe therapy of amoebiasis. Matrix tablet of Metronidazole containing various proportions of guar gum and Xanthan gum were prepared and subjected to in-vitro drug release studies. The results of this work revealed that among 20 prepared Metronidazole tablets, only three formulations showed promising sustained release *in vitro*.

These three formulations showed sustained release characteristics over 24 hours. So the selected tablets could be evaluated in -vivo in animals. However, the further studies are planned to assess the utility of these colon targeted drug delivery system of Metronidazole in patients suffering from amoebiasis.

## References

1. A. K. Philip, S. Dabas, and K. Pathak, Optimized prodrug approach: a means for achieving enhanced anti-inflammatory potential in experimentally induced colitis, *Journal of Drug Targeting*, vol. 17,. 3, 235–241, 2009.

2. O. A. Odeku and J. T. Fell, In-vitro evaluation of khaya and albizia gums as compression coatings for drug targeting to the colon, *Journal of Pharmacy and Pharmacology*, vol. 57, 2, 163–168, 2005.

3. M. K. Chourasia and S. K. Jain, Pharmaceutical approaches to colon targeted drug delivery systems, *Journal of Pharmacy and Pharmaceutical Sciences*, vol. 6, 1, 33–66, 2003.

4. A. Basit and J. Bloor, Prespectives on colonic drug delivery: business briefing, *Pharmaceutical Technology*, 185–190, 2003.

5. Krishnaiah YSR, Reddy PBR, Satyanarayanan V, Karthikeyan RS. Studies on development of oral colon targeted drug delivery system for metronidazole in treatment of amoebiasis. Int. J. Pharm.; 23 6:43-45, 2000.

6. Vanden Mooter G, Kinget R. Oral Colon specific drug delivery: A review. Drug Delivery. 2:8,19-31, 1995.

7. Chourasia MK, Jain SK. Pharmaceutical approaches to colon targeted drug delivery system. J. Pharm. Pharm. Sci.;6(1):33-66, 2003

8. Sinha VR, Kumria R. Coating polymers for colon specific drug delivery: A comparative in vitro evaluation. Acta Pharm.; 53:41-47, 2003.

9. H. Maier, M. Anderson, C. Karl, K. Maqnuson, and R.L. Whistler, "Industrial Gums," in *Polysaccharides and their Derivatives*, Academic Press, New York, NY, USA, 3rd edition, 1993.

10. G. L. Simon and S. L. Gorbach, "Intestinal flora in health and disease," *Gastroenterology*, vol. 86,. 1, 174–193, 1984.

11. Krishnaiah YSR, Devi S, Rao NL, Satyanarayanan V, Karthikeyan RS, Bhaskar PR. Guar gum as a carrier for Colon-specific delivery: Influence of Metronidazole and Tinidazole on in vitro release of albendazole from guar gum matrix tablets. J. Pharm. Pharmaceutical Sci.;4(3):235-243,2001.

12. T. P. Hadjiioannou, G. D. Christian, and M. A. koupparis, "Quantitative calculations," in *Pharmaceutical Practices and Research*, 345–348, NY- VCH, New Dehli, India, 1993.

13. Krishnaiah YSR, Satyanarayanan S, Ramaprasad YV, Rao N. Evaluation of guar gum as a compression coat for drug targeting to the colon. Int. J. Pharm.; 171:137-146, 1998.

14. B.K. Sahoo, K.V. Gowda, U. Mandal, A. Das, U. Chakrabarty, T.K. Pal Sustained Delivery of Ranitidine HCl from Floating Matrix Tablets: Formulation and *in vitro* Evaluation: *Asian Journal of Chemistry:* Vol. 21, 2, 1350-1358, 2009.

15. Bijay Kumar Sahoo, Sidheswar Prasad Pattajoshi, Sandhyarani Pattajoshi, Formulation and Evaluation of Bimolar release of Ciprofloxacin HCl from Bilayer Gastro-retentive Floating System.**;** *Asian Jour. Of Pharma Res.* Vol.8, NO.02, 2018.

16. P. Apparao, J. V. Prabhakarreddy, J. Raju, and B. Shashidher, "Formulation and evaluation of gum based matrix tablets of Lamivudine," *Der Pharmacia Sinica*, vol. 2, no. 3, 176–192, 2011.

17. Y. W. Chien, "Novel Drug Delivery Systems," in *Fundamentals of Rate-Controlled Drug*, Marcel Dekker, New York, NY, USA, 1992.

18. Bijay Kumar Sahoo, Jayanti Mukherjee, Tapan Kumar Pal, Development and Validation of a High-Performance Liquid Chromatographic Method for Bioanalytical Application with Ranitidine HCl, Int. Jour. Pharm.& Pharma Sc.,Vol 3.No.2 ,2011.

19. B. M. Al-Taani and B. M. Tashtoush, "Effect of microen-vironment pH of swellable and erodable buffered matrices on the release characteristics of diclofenac sodium," *AAPS Pharmaceutical Science Technology*, 4, 3, article E43, 2003.

20. B. Abrahamsson, M. Alpsten, B. Bake, A. Larsson, and J. Sjögren, "In vitro and in vivo erosion of two different hydrophilic gel matrix tablets," *European Journal of Pharmaceu- tics and Biopharmaceutics*, 46, 1, 69–75, 1998.

21. Sinha VR, Kumria R. Polysaccharides matrices for microbially triggered drug delivery to the colon. Drug Dev. & Ind. Pharm.;3:(2):143-150, 2004

22. A. M. Cerdeira, P. Goucha, and A. J. Almeida, "Hydrox- ypropyl methylcellulose phthalate beads containing a model non-steroid anti-inflammatory drug," *International Journal of Pharmaceutics*, 164, 1-2, 147–154,1998

23. P. W. S. Heng, L. W. Chan, M. G. Easterbrook, and X. Li, "Investigation of the influence of mean HPMC particle size and number of polymer particles on the release of aspirin from swellable hydrophilic matrix tablets," *Journal of Controlled Release*. 76. 1-2. 39–49, 2001.

24. T. Gafourian, A. Safari, K. Adibkia, F. Parviz, and A. Nokhod- chi, "A drug release study from hydroxypropylmethylcellulose (HPMC) matrices using QSPR modeling," *Journal of Pharma- ceutical Sciences*, 96, 12,. 3334–3351, 2007.

25. Kadria A. Elkhodairy,Samar A. Afifi, Azza S. Zakaria:An approach to provide appropriate colon target drug delivery systems of vancomycin hcl: Pharmaceutical and Microbiological studies*, Bio Med Research International*,10,1,182-197,2014.