INTERNATIONAL JOURNAL OF PHARMACEUTICAL, CHEMICAL AND BIOLOGICAL SCIENCES

Available online at www.ijpcbs.com

Research Article

ENTERIC COATED 5-FLUOROURACIL CAPSULES DESIGNED TO ACHIEVE INTESTINAL TARGETING

Harpreet Kaur*, Saurav Kumar and MS. Rathore

CT Institute of Pharmaceutical Sciences, Shahpur, Jalandhar, Punjab, India.

ABSTRACT

The purpose of the present investigation was to achieve successful delivery especially to colon using shellac as coat over the hard gelatin capsule. The drug delivery system was based on the gastrointestinal transit time concept, assuming colon arrival time to be 6 h. Rapidly disintegrating capsules containing 50mg 5-FU were coated with different concentration of shellac by dipping method. In order to find the suitable formulation, various formulation factors were investigated through series of *in-vitro* dissolution studies in buffer solution at pH 1.2 for first 2 h, and at pH 6.8 for remaining hour. The results indicated that C4 is the most suitable formula in the approach of time dependent oral delivery system for colon targeting for achieving minimum release in the first five hours and maximum at the 24 th hour. The formulation C4 was then studied with different probiotics.. Gp1 shows a release of 12.14 % in first five hours but, on 24 hours Gp1 shows a maximum release of 98.75 %. So the capsule with probiotc was found to be more effective than without probiotics.

Keywords: Colon-targeted delivery system, 5-FU, guar gum, multiparticulate system, shellac.

1. INTRODUCTION

The principal goal of site specific delivery is to deliver the drug in the specific organ of body. The therapeutic advantages of targeting the drug to the diseased organ include reduced incidence of adverse side effects, lower conventional dose and delivery of drug as close as possible to the target site[1]. Orally administered dosage forms normally dissolves in the stomach fluid or intestinal fluid and absorb from these regions of the gastrointestinal tract (GIT) depends upon the physicochemical properties of the drug. It is a serious drawback in conditions where localized delivery of the drugs in the colon is required or in the hostile environment of upper GIT. Site-specific targeting of drugs to the colon has been attempted by several different approaches. Of these, utilisation of the bacterial population, existing almost exclusively in the colon, as a means of targeting offers considerable promise. Multiple-unit systems have been shown to spread out on entry to the colon and this may give improvements in drug absorption and local treatment. Additionally, the higher surface area of multiple unit systems should lead to a more rapid release of drug due to more rapid bacterial breakdown. The transit of dosage forms in the upper gastrointestinal tract is also a consideration in colonic delivery as delays expose the material to longer periods of time in a harsh environment. In this regard also, multiple-unit systems may empty from the stomach and traverse the ileo-caecal junction in a more reproducible manner than single units[2].

Delivery of drugs via the colon offers numerous therapeutic advantages. Various diseases of colon such as ulcerative colitis, Chron's disease, carcinoma and infections require local therapy. So, the development of locally acting colon targeted drug delivery systems may revolutionize the treatment of colonic diseases. Colon specific system could also be used in conditions in which a diurnal rhythm is evident e.g. asthma, rheumatic disease, ulcer diseases and ischemic heart disease[3].

Colon cancer is a disease of large intestine which begins at a structure called the caecum, located in the right lower quadrant of the abdomen, and continues through all portions of the abdomen to its junction with the rectum, located in the deep pelvis. Colon cancer begins when normal cells in the lining of the colon or rectum change and grows uncontrollably, forming a mass called a tumor. A tumor can be benign (non-cancerous) or malignant (cancerous, meaning it can spread

to the other parts of the body). These changes usually take years to develop; however, when a person has an uncommon inherited syndrome, changes can occur within months to years. Both genetic and environmental factors can cause the changes[4].

Several polysaccharides like, pectin and its salts, chondroitin sulphate, amylase and guar gum are being investigated as carriers for colon specific drug delivery. In pharmaceutical formulations, guar gum is used as a binder, disintegrant, suspending agent, thickening agent and stabilizing agent[5,6]. Guar gum and pectin are reported to be potential carriers for colon specific drug delivery. Colon specific drug delivery systems for 5-ASA and mebendazole have been developed using guar gum as a carrier[7, 8]. The guar gum matrix tablets of albendazole were found degraded by colonic bacteria of rat caecal contents and released about 44% of albendazole in simulated colon fluids at the end of 24h indicating the susceptibility of the guar gum formulation to the rat caecal contents[9].

In the present research work, a model drug 5-Fluorouracil was used. 5-Fluorouracil (5-FU) is one of the oldest anticancer drugs and is still used in the treatment of colorectal cancer. 5-Fluorouracil is an antimetabolite (pyrimidine base) used to treat breast, gastrointestinal, head and neck, and ovarian cancer. Due to its structure, 5-FU interferes with nucleoside metabolism and can be incorporated into RNA and DNA, leading to cytotoxicity and cell death[10]. With this information, it is planned to develop multiparticulate approaches in the form of granules of 5-Fu with guar gum with and without probiotic were prepared. The purpose of designing multiparticulate dosage form is to develop a reliable formulation that has all the advantages of single unit formulations.

2. MATERIAL AND METHODS

2.1. MATERIAL

Gift sample of 5-Fluorouracil, shellac, Lactobacillus acidophyllus, Lactobacillus sporogenes, Bifidobacterium bifidum were obtained from KEE GAD Biogen Pvt. Lmt., New Delhi, India. Guar gum was purchased from Central Drug House, New Delhi, India. All other ingredients used were of analytical grade.

2.2. Preparation of 5-Fluorouracil granules

Different batches of matrix granules of 5-fluorouracil were formulated by wet granulation method. All ingredients were weighed on digital weighing balance and passed through sieve no. 40. The sieved ingredients (except lubricants and glidants) were placed in mortar and pestle for mixing. Then distilled water was added for wet massing, to form a coherent mass. The obtained coherent mass was placed on the sieve no. 10 and forced out through a sieve screen where it was continuously formed into extrudates. Cylindrical shaped extrudates were then transferred and spreaded out on perforated trays for drying of product in hot air oven at $50^0C \pm 2^0C$ for 1.5-2 hours. Then dried cylindrical mass was forcefully passed through sieve no. 22 to get uniform sized granules. Lubricants (magnesium stearate) and glidant (talc) were added to improve flow properties.

Table 1: Composition of matrix granules of 5-fluorouracil containing guar gum and drug in different ratio from 1:1 to 1:5

S.No.	Ingredients	Formulation code				
		G1 (mg)	G2 (mg)	G3 (mg)	G4 (mg)	G5 (mg)
1	5-Fluorouracil	50	50	50	50	50
2	Guar gum	50	100	150	200	250
3	Lactose	385	335	285	235	185
4	Magnesium stearate	5	5	5	5	5
5	Talc	10	10	10	10	10

2.3. Evaluation of granules

Determination of bulk density and tapped density

An accurately weighed quantity of the granules (W) was carefully poured into the graduated cylinder and the volume (Vo) was measured, then the graduated cylinder was closed with lid, set for 100 taps and after that, the volume (Vf) was measured and continued operation till the two consecutive readings were equal. The bulk density, and tapped density were calculated using the following formulas.

Tapped density = W/Vf

Bulk density = W/Vo

Where, Vo = initial volume

Vf = final volume.

Compressibility index

The compressibility index and Hausner ratio may be calculated using measured values for bulk density (ρ bulk) and tapped density (ρ tapped) as follows:

Compressibility index =

$$\frac{\rho \text{ tapped} - \rho \text{ bulk}}{\rho \text{ tapped}} \times 100$$

Hausner ratio = $\frac{\rho \text{ tapped}}{\rho \text{ bulk}}$

Lower Hausner's ratio (<1.25) indicates better flow properties than higher ones, between 1.25 to 1.5 showing moderate flow properties and more than 1.5 poor flow.[11]

Angle of repose

The angle of repose of granules was measured by the funnel method. In brief, the accurately weighed granules were placed in the funnel. The height of the funnel was adjusted in such a way that the tip of the funnel just touched the apex of the heap of the granules. The granules were allowed to flow through the funnel freely onto the surface. The diameter of the powder cone was determined and then the angle of repose was calculated according to equation (1).[12]

$$\text{Tan } \theta = h/r \quad (1)$$

Where, h and r in (1) represent the height and radius of the powder cone.

Drug Content

An accurately weighed amount of powdered5-FU granules (100 mg) was extracted with 100 ml of distilled water and the solution was filtered. After suitable dilution (10 times or more), drug content in the filtrate was analyzed spectrophotometrically at 266.5 nm (Shimadzu-1700 UV–vis spectrophotometer) against a blank[13].

2.4. Capsule filling and coating

Size 00 capsules made from gelatin were filled by hand with 500 mg of matrix granules of 5-FU containing varying amount of guar gum. After filling the capsules were sealed with 5% (w/w) Ethyl cellulose in ethanolic solution[14].

The core capsules obtained were coated with three layers of shellac of different concentration (5, 10 and 15%) by dipping method. In dipping method the core capsules obtained were pre-weighed in an analytical balance and the weight is noted. Each capsule was slightly pierced with a 21G hypodermic needle; this served to provide a firm support in order to avoid direct contact with the hand. The capsule was then dipped into the shellac coating solution for 3-5 sec and removed. It was dried under a fan (28^{0}C) and allowed to equilibrate for 24hrs and same procedure was repeated for the 2nd and 3rd coatings. After drying the needle was removed and the piercing spot sealed off with a little drop of shellac followed by 2nd and 3rd coat and again allowed to dry for another 24hrs[14].

Table 2: Capsules filled with granules with varying ratio of drug and guar gum and coated with different concentration of shellac solution

S.No	Formulation code	Drug:guar gum	% Shellac solution
1.	C1	1:1	5
2.	C2	1:2	5
3.	C3	1:3	5
4.	C4	1:4	5
5.	C5	1:5	5
6.	C6	1:1	10
7.	C7	1:2	10
8.	C8	1:3	10
9.	C9	1:4	10
10.	C10	1:5	10
11.	C11	1:1	15
12.	C12	1:2	15
13.	C13	1:3	15
14.	C14	1:4	15
15.	C15	1:5	15

EVALUATION OF COLON TARGETED DRUG CAPSULE

Weight variation

10 capsules were selected randomly from each batch and weighted individually to check for weight variation. The test requirements are met if none of the individual weights are less than 90% or more than 110% of the average.

Drug content and content uniformity

The drug content and uniformity of 5-FU was determined by UV-spectrophotometer. Distilled water was used to dissolve the drug for content uniformity assay. The content of 1 capsule was transferred to 100ml beaker. About 70 ml of water was added and stirred well. This solution was adjusted with distilled water to 100ml. The

above solution was used as stock solution and filtered using filter paper. The absorbance was measured at 266.5 nm after suitable dilution.

Disintegration test

Use the apparatus described under disintegration test (2.5.1), using one capsule in each tube. Operate the apparatus for 2 hours without the discs in 0.1 M hydrochloric acid. No capsule shows signs of disintegration or of rupture permitting the escape of the contents. Replace the medium in the vessel with mixed phosphate buffer pH 6.8, add a disc to each tube and operate the apparatus for a further 60 minutes. Remove the apparatus from the medium and examine the capsules. They pass the test if no residue remains on the screen or on the underside of the discs, or, if a residue remains, it consists of fragments of shell or of a soft mass with no palpable, unmoistened core[15].

***In- vitro* release study**

In vitro dissolution test was conducted in USP 2 apparatus at 75 rpm and a temperature of 37±0.5^{0}C. Sampling was done at predetermined time intervals and the same were estimated for drug content after suitable dilution by using double beam UV-VIS spectrophotometer (I.P., 2007). Initial drug release studies were conducted in 900 ml of 0.1N HCl for 2 hour followed by 900 ml of 7.4 potassium phosphate buffer solution for next 3 hours. Then, 900 ml of 6.8 potassium phosphate buffer solution for rest of the time[16].

2.5. Effect of probiotic on the drug release

***In vitro* digestion studies of guar gum by bacterial spores**

Accurately weighed 2gm of guar gum powder and transferred slowly in the beaker containing 200ml of warm distilled water. Above slurry was mixed on magnetic stirrer with magnetic bead for 30 minutes and kept at 37^{0}C ± 2^{0}C in incubator for 24 hours, so that gum gets fully swelled. After 24 hours, in each 200ml guar gum (1 percent w/v) slurry, different bacterial spores (*Bifidobacterium bifidum, Lactobacillus acidophilus* and *Lactobacillus sporogenes*) were added (1 percent w/v) in different beakers and incubated at 37^{0}C ± 2^{0}C in incubator. At different time interval the change in pH and viscosity was measured using calibrated pH meter and Brookfield viscometer with spindle no. 4 and 20 rpm, respectively. Controlled *in vitro* digestion studies were also performed using same above conditions except use of probiotics[17].

Table 3: Table showing formulations with different probiotic

S.No.	Ingredients	Formulation code		
		Gp1(mg)	Gp2(mg)	Gp3(mg)
1	5-Fluorouracil	50	50	50
2	Guar gum	200	200	200
3	B.bifidum	200	-	-
4	L.acidophyllus	-	200	-
5	L.sporogenes	-	-	200
6	Lactose	35	35	35
7	Magnesium stearate	5	5	5
8	Talc	10	10	10

Evaluation of CTDC filled with granules containing different probiotic

Weight variation and *in vitro* dissolution studies of coated capsules of 5-fluorouracil containing different probiotics was done by same method as given above.

3. RESULTS AND DISCUSSION

The granules of all the formulations were evaluated for angle of repose, bulk density, tapped density, and compressibility index and hausner ratio. The angle of repose was found to be 31°51' ±0.77 – 33°69' ± 0.74. It indicates that granules have a good flow property. The bulk density and tapped density was found to be in the range of 0.36 ± 0.00 - 0.41 ±0.01 gm/cc and 0.39 ± 0.01 - 0.48 ± 0.02 gm/cc respectively. The compressibility and hausner ratio was found to be 7.69 ±1.86 to 15.50 ± 1.74 and 1.08 ± 0.04 to 1.17 ± 0.04 indicating good flow character of the granules (table-4). All the results are within the prescribed limit.

Table 4: Evaluation of matrix granules of 5-FU

Formulation code	Bulk density* (g/cc)	Tapped density*(g/cc)	Carr's index* (%)	Hausner's ratio*	Angle of repose*(θ)	Drug content* (%)
G1	0.41±0.01	0.48± 0.02	15.50±1.74	1.17±0.04	33°14'±0.66	98.20± 1.33
G2	0.40±0.01	0.46±0.02	13.04±1.66	1.15±0.02	33°69'±0.74	100.87 ± 1.31
G3	0.38±0.05	0.42±0.01	9.52±1.27	1.10±0.02	30°30'±0.67	101.2 ± 1.89
G4	0.36±0.00	0.39±0.01	7.69±1.86	1.08±0.04	32°42'±0.59	95.99 ± 1.96
G5	0.36±0.01	0.40±0.01	10±2.01	1.11±0.03	31°51'±0.77	97.04 ± 2.13

***** All values are expressed as mean ± SD, n=3

Then the prepared granules were filled manually in size 00 hard gelatine capsules and evaluated for weight variation, drug content and content uniformity, disintegration test and *in vitro* dissolution studies. All the batches show a rapid release of drug in first five hours (i.e. upto 70 percent) in 0.1N HCl and 7.4 pH PBS. This is because of high solubility of 5-fluorouracil in water. To overcome this problem coating of capsules was done by using different concentrations of shellac. The prepared coated capsules were evaluated and results were shown in the table 5.

Table 5: Evaluation of coated capsules of 5-FU

S.NO	Formulation code	Evaluation Parameters		
		Weight variation (mg)**	Drug content (%)*	Disintegration time (hrs)*
1.	C1	575.67± 1.35	99.08 ± 1.33	3.4± 0.13
2.	C2	577.85±2.74	98.56 ± 0.86	3.3± 0.09
3.	C3	582.34±1.13	100.23 ± 1.34	3.4± 0.06
4.	C4	583.77±2.62	98.04 ± 1.06	3.5± 0.10
5.	C5	581.19±2.49	100.87 ± 1.31	3.4± 0.07
6.	C6	597.24±2.89	101.2 ± 3.89	3.8± 0.09
7.	C7	592.55±2.68	98.99 ± 3.96	3.6± 0.08
8.	C8	603.43±1.66	97.04 ± 2.33	3.9± 0.10
9.	C9	598.29±3.52	96.66 ± 1.83	3.8± 0.15
10.	C10	601.14±3.37	98.60 ± 0.60	3.7± 0.21
11.	C11	611.75±2.71	96.59 ± 3.93	4.2± 0.20
12.	C12	609.31±1.73	98.76 ± 3.32	4.2± 0.19
13.	C13	613.24±2.66	100.15 ± 1.73	4.3± 0.14
14.	C14	610.87±3.59	99.82 ± 2.78	4.1± 0.12
15.	C15	610.72±4.05	97.42 ± 1.93	4.2± 0.19

*Values are mean ± S.D, Number of determination=2
** Values are mean ± S.D, Number of determination=20

Table 6: *In vitro* release study of capsule containing matrix granules with varying concentration of guar gum, coated with 5% shellac solution

S.No.	Time (hours)	C1 (%)	C2 (%)	C3 (%)	C4 (%)	C 5(%)
1.	2	2.76±2.71	2.72±0.35	1.10±1.12	0.44±1.77	0±0.32
2.	5	56.74±0.94	39.87±3.21	25.10±0.54	11.37±2.24	9.88±2.47
3.	7	64.48±1.08	57.41±2.49	26.30±2.64	32.93±2.69	25.24±2.35
4.	9	68.37±2.16	64.13±4.02	37.26±2.38	45.04±1.53	37.12±3.17
5.	12	76.85±0.87	76.85±2.30	46.45±4.21	52.11±2.98	49.19±2.98
6.	15	78.62±3.02	82.16±0.92	58.83±1.05	55.89±4.23	52.20±2.77
7.	18	83.22±1.38	84.99±3.42	73.32±3.62	63.42±2.43	58.47±1.49
8.	21	85.69±1.76	90.99±1.57	78.98±1.83	75.44±1.29	67.18±2.76
9.	24	88.17±2.75	95.24±2.25	87.81±2.45	84.63±2.78	72.61±2.56

*****Values are mean ± S.D, Number of determination=3

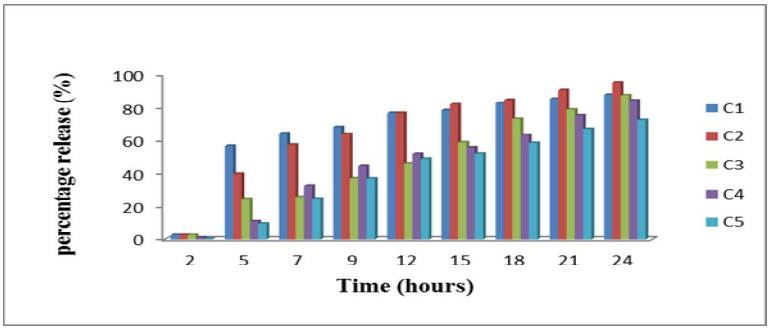

Fig. 1: *In vitro* release study of capsule coated with 5% shellac

Table 7: *In vitro* release study of capsule containing matrix granules with varying concentration of guar gum, coated with 10% shellac solution

S.No.	Time (hours)	C6 (%)	C7 (%)	C8 (%)	C9 (%)	C10 (%)
1.	2	0±1.27	0±0.97	0±1.45	0±1.03	0±0.79
2.	5	0.57±1.41	0.45±1.41	0.39±1.79	0.23±1.94	0.20±2.31
3.	7	2.27±1.76	2.19±3.56	2.06±2.53	1.95±2.65	1.83±1.96
4.	9	4.82±0.68	4.79±1.39	4.71±2.86	4.58±2.39	4.49±1.54
5.	12	7.54±2.54	7.45±4.04	7.38±1.96	7.29±1.86	7.25±2.49
6.	15	13.41±2.35	13.38±2.67	13.24±2.45	13.15±2.82	13.09±2.37
7.	18	20.58±1.57	20.43±1.31	20.36±3.21	20.28±1.99	20.17±4.21
8.	21	33.42±3.07	33.36±2.78	33.17±1.19	33.09±1.68	32.97±3.83
9.	24	42.91±2.13	42.85±2.66	42.77±2.30	42.15±2.15	42.11±1.62

***Values are mean ± S.D, Number of determination=3

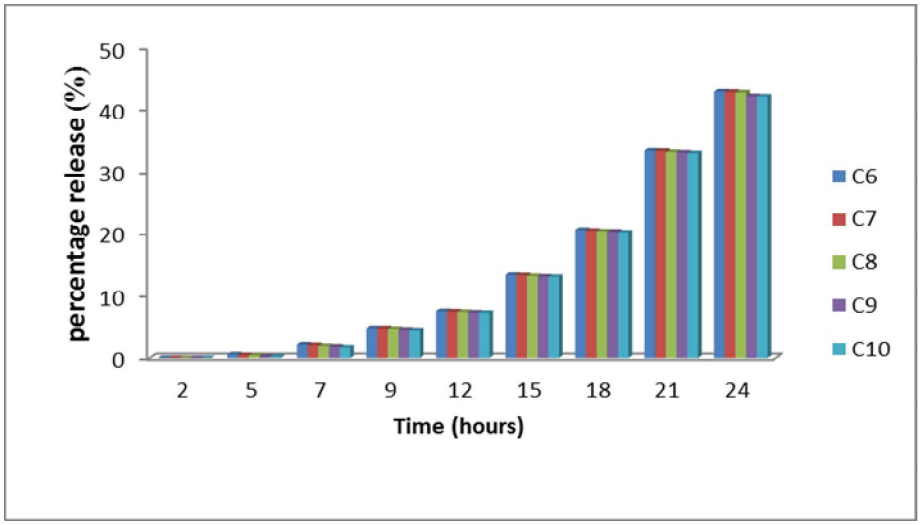

Fig. 2: *In vitro* release study of capsule coated with 10% shellac

Table 8: *In vitro* release study of capsule containing matrix granules with varying concentration of guar gum, coated with 15% shellac solution

S.No.	Time (hours)	C11 (%)	C12 (%)	C13 (%)	C14 (%)	C15 (%)
1.	2	0±0.12	0±0.03	0±0.05	0±0.15	0±0.09
2.	5	0±0.14	0±0.21	0±0.19	0±0.06	0±0.07
3.	7	0.19±0.06	0.12±0.03	0.05±0/08	0±0.01	0±0.02
4.	9	1.39±0.07	1.25±0.13	1.17±0.02	1.04±0.21	1.03±0.08
5.	12	3.86±0.24	3.73±0.32	3.52±0.45	3.19±0.32	3.10±0.27
6.	15	5.14±0.89	5.03±1.04	4.96±1.23	4.81±1.72	4.54±0.99
7.	18	7.68±1.20	7.53±1.21	7.46±1.14	7.38±0.86	7.27±2.35
8.	21	12.81±0.69	12.69±0.75	12.57±1.35	12.43±0.55	12.36±1.87
9.	24	25.50±0.98	25.26±1.74	24.57±1.53	24.15±1.59	24.11±0.54

***Values are mean ± S.D, Number of determination=3

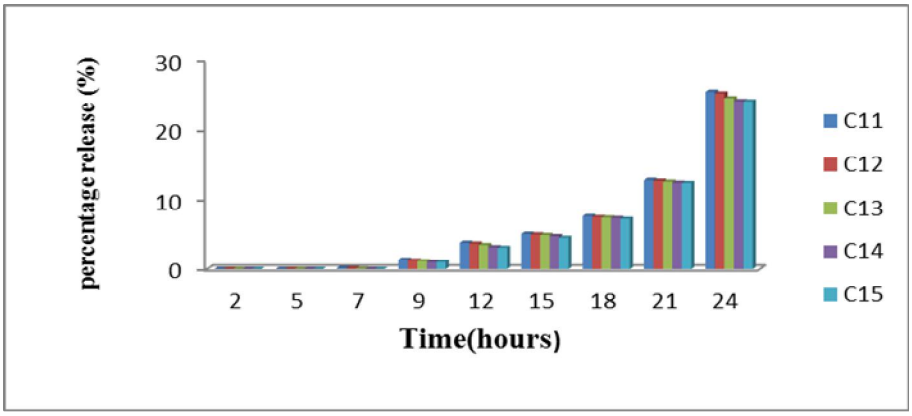

Fig. 3: *In vitro* release study of capsule coated with 15% shellac

Prepared colon targeted drug capsule formulations were subjected to preliminary *in - vitro* release studies. Dissolution was carried in two media, namely simulated gastric fluid (acidic buffer, pH 1.2) for the first two hours, and simulated colonic fluid (phosphate buffer pH 6.8) for the subsequent hours.

After ingestion of the capsule, there was no drug release in the stomach due to the acid resistibility of the polymeric layer with all the formulation, indicating the efficiency of shellac as enteric coating polymer. This polymer proved that the drug was not released in the stomach. The release profile of the various formulations coated with 5, 10 and 15% shellac is given in table 6, 7, 8 and figure 1, 2, 3 respectively. The drug release was slow in first 4 hours followed by spread over 24 hour and depends upon concentration of shellac.

The 5-FU release was more in the case of capsules with 5% shellac coat (C1) and by the end of lag time 79% of release was observed. When 10% of shellac coating was done, C6, the release at the end of lag time was about 14%. C11 capsules which were coated with 15% shellac released 6%drug at the end of lag time. Of all the different formulae prepared C4 is the most suitable formula in the approach of time dependent oral delivery system for colon targeting for achieving minimum release in the first five hours and maximum at the 24 th hour.

Evaluation of CTDC filled with granules containing different probiotic

Table 9: *In vitro* digestion studies of guar gum

Digestion source	Time (hours)	pH ± Standard deviation	viscosity ± Standard deviation
Bifidobacterium bifidum	0	6.60 ± 0.00	2760.00 ± 0.00
	24	5.23 ± 0.03	783.33 ± 17.66
Lactobacillus acidophillus	0	6.60 ± 0.00	2760.00 ± 0.00
	24	5.10 ± 0.06	1376.67 ± 38.48
Lactobacillus sporogenes	0	6.60 ± 0.00	2760.00 ± 0.00
	24	5.27 ± 0.03	1040.00 ± 32.18
Control	0	6.60 ± 0.00	2760.00 ± 0.00
	24	5.97 ± 0.05	2656.67 ± 6.67

Maximum difference in pH and viscosity was found in *Bifidobacterium bifidum.*

The extent of breakdown of guar gum was influenced by the bacterial enzymes present as well as the chemical structure of guar gum.a fall in viscosity can be related to a reduction polysaccharide chain length i.e. destruction of (1,4)-β-linked D-mannan backbone by microbial enzymes. A fall in pH is caused by the production of short chain fatty acids as well as generation of CO_2, which indicates that fermentation has occurred. Thus the data suggested that a mixed population of colonic bacteria can produce extracellular enzymes that reduce the chain length of guar gum.

Viscosity and pH reducing capacity was better in case of *Bifidobacterium bifidum* than all others spores used. The order of pH reduction by all probiotics was *Lactophilus acidophilus* > *Bifidobacterium bifidum* > *Lactobacillus sporogenes*. The order was selected on the basis of differences in the initial to the final pH. The order of viscosity reduction by all probiotics was *Bifidobacterium bifidum* > *Lactobacillus sporogenes* > *Lactophilus acidophilus*. The order was selected on the basis of differences in the initial to the final viscosity.

Table 10: Weight variation of CTDC filled with granules containing different probiotic coated with 5% shellac solution

S.No	Formulation code	Average weight (mg)*	Percentage weight variation (%)
1.	Gp1	579.77	- 2.35 to + 2.71
2.	Gp2	584.15	- 1.74 to + 2.28
3.	Gp3	582.34	- 2.13 to + 2.16

Table 11: *In vitro* dissolution studies of CTDC filled with granules containing different probiotic coated with 5% shellac solution

S .No	Time (in hrs)	Formulation code		
		Gp1 (%)	Gp2 (%)	Gp3 (%)
1.	2	1.49±0.23	0.86±1.27	1.31±1.06
2.	5	12.14±2.31	12.18±1.87	11.65±1.59
3.	7	31.72±1.69	31.69±3.56	32.54±2.79
4.	9	44.33±3.78	45.04±1.67	28.42±1.34
5.	12	61.29±1.45	55.29±2.59	41.50±3.45
6.	15	69.78±4.23	59.18±3.65	49.28±2.54
7.	18	78.26±2.38	62.01±1.16	55.29±0.49
8.	21	89.98±0.46	71.55±2.92	68.37±3.52
9.	24	98.75±1.33	87.46±2.06	82.86±1.64

*Values are mean ± S.D, Number of determination=3

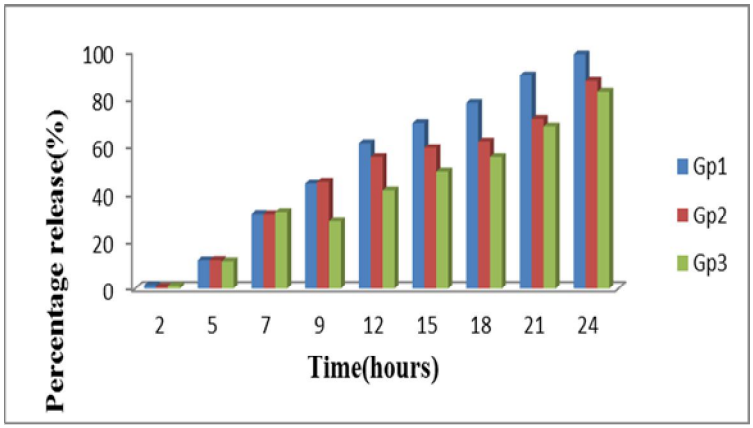

Fig. 4: *In vitro* release study of capsule containing different probiotic coated with 5% shellac solution

The formulation C4 was studied with different probiotics. The CTDC with probiotic were evaluated for weight variation and *in-vitro* dissolution profile. The results are shown in table 10, 11 and figure 4 respectively. Gp1 shows a release of 12.14 % in first five hours but, on 24 hours G5 shows a maximum release of 98.75 %. So the capsule with probiotc was found to be more effective than without probiotics. The Stability of enteric coated capsules was found stable in presence of the excipients used, under conditions of temperature and humidity. Physical visualization of enteric coated capsules showed no change in appearance. No major change in amount of drug release was observed during the storage conditions which reflected the stability of formulated capsule.

4.CONCLUSION

On the basis of above study it may be concluded that the formulation containing probiotics ensures the major portion of drug (more than 95%) to be released at colon even in absence of colonic microflora. The guar gum matrix granules formulation developed with probiotic filled in capsules holds tremendous potential to deliver a variety of drugs in colon diseases (viz anticancer drugs) specifically at colon and ensures maximum drug concentration at colon.

The present study confirmed the idea of providing excellent evidence of enteric protection for the coated capsules.

5. ACKNOWLEDGEMENT

The authors are sincerely thankful to CT Institute Pharmaceutical Sciences, Shahapur, Jalandhar for providing us infrastructure facilities and moral support to carry out this research work.

6. REFERENCES

1. Watts PJ and Illum L. Colonic Drug Delivery. Drug Development and Industrial Pharmacy. 1997; 29: 893–913.
2. Devi USK and Thiruganesh R. Preparation and characterization of pectin pellets of Aceclofenac for colon targeted drug delivery. Journal of Chemical and Pharmaceutical Research. 2010;2:1: 361-374.
3. Kumar KGB and Yadav NS. Design and Evaluation of a new Capsule-type Dosage form for Colon-Targeted Delivery of Etoricoxib. International Journal of Pharmaceutics Science. 2013;3:1:147-151.
4. Yeatman TJ. Colon cancer. Encyclopedia of life sciences. Macmillan Publishers Ltd, Nature Publishing Group. 2001; 261-264.
5. Lachman L, Lieberman HA and Kanig JL. The theory and practice of industrial pharmacy. India 1990; 592.
6. Goldstein AM, Alter EN, Seaman JK and Guar Gum. In: Whistler RL [Ed]. Industrial Gums, Polysaccharides and their derivatives. Academic press, New York. 1993;303-321.
7. Krishnaiah YSR, Veer RP, Kumar DB, Bhaskar P and Satyanarayana V. Development of colon targeted drug delivery systems for mebendazole, Journal of Controlled Release. 2001;77:87–95.
8. Krishnaiah YSR, Satyanarayana V, Dinesh Kumar B, Karthikeyan RS and Bhaskar P. European Journal of Pharmaceutical Sciences. 2003;19:355–362.
9. Badmapriya D and Rajalakshmi AN. Guar Gum Based Colon Targeted Drug Delivery System: In-Vitro Release Investigation. Research Journal Of Pharmaceutical, Biological And Chemical Sciences. 2011;2:3:899-903.
10. Calabresi P and Chabner BA. Chemotherapy of neoplastic diseases. In: Hardman JG, Limbird LE, Perry BM and Raymond WR. (Eds.), Goodman and Gilman's The Pharmacological Basis of Therapeutics, 9th Edition 1996. McGraw-Hill, New Delhi, 1225-1232.
11. Lachman L, Lieberman HA and Kanig JL. The theory and practice of industrial pharmacy. India, third edition 1990;183-184.
12. Wenwu Z and Zhongcai F. Development and pharmacokinetic evaluation of once-daily sustained-released matrix capsules of nifedipine using solid dispersion technique. African Journal of Pharmacy and Pharmacology. 2013;7:12:658-665
13. Antesh K Jha and Bhattacharya A. Formulation And In Vitro Evaluation Of Sustained Release Matrix Tablets of Metoprolol Succinate Using Hydrophilic Polymers. International Journal of PharmTech Research. 2009;1:4:972-977.
14. Kiran Kumar GB and Naresh Yadav S. Design and Evaluation of a new Capsule-type Dosage form for Colon-Targeted Delivery of Etoricoxib. International Journal of Pharmaceutics Science. 2013;3:1:147-151
15. Indian Pharmacopoeia. The Indian pharmacopoeia commission, Ghaziabad. (I.P.), 2007.
16. Sinha VR, Mittab BR and Rachna K. In vivo evaluation of time and site of disintegration of polysaccharide tablet prepared for colon specific drug delivery. International Journal of Pharmaceutics. 2005;289:79-85.
17. Ghosh PK, Gupta VB, Gondoliya B and Rathore MS. Probiotic-assisted colon specific delivery of diclofenac sodium from guar gum matrix tablets: In vitro evaluation. Asian Journal Pharmaceutics. 2010;4:173-178.