INTERNATIONAL RESEARCH JOURNAL OF PHARMACY
www.irjponline.com
ISSN 2230 – 8407

Research Article

DESIGN OF A NOVEL COLON TARGETED MICROSPONGES LOADED WITH DICLOFENAC SODIUM USING THREE DIFFERENT POLYMERS

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Article Received on: 09/03/18 Approved for publication: 12/04/18

DOI: 10.7897/2230-8407.09454

ABSTRACT

The present investigation was done based on the many frequencies of drug taken orally per single day so, for altering the dose frequency of the drug delivery for the colonic diseases diclofenac sodium was loaded with the microsponges. The colonic targeted microsponges are prepared by acid resistant polymers such as eudragit L100, eudragit RS 100, eudragit EPO 100. These acid resistant polymers release the drug for controlled manner and site specific action. Diclofenac sodium was incorporated into microsponges which help in controlled release of the diclofenac sodium. The polymer used for the preparation of microsponges was useful in forming sponges in which drug was entrapped. FT-IR studies were performed and from the FT-IR spectra it was evident that there were no interactions between the diclofenac sodium and polymer. Formulations of microsponges were prepared by quasi-emulsion solvent diffusion technique using eudragit L100, eudragit RS 100 and eudragit EPO 100 in various ratios such as (1:0.5, 1:1, 1:2 and 1:2). By using the 3 polymers 12 formulations were prepared from the 12 formulations 3 formulations were selected as optimized formulations based on the production yield(%),drug loading(%), encapsulating efficiency and *in vitro* drug release studies (90.4.7%). Hence, it was proposed that diclofenac sodium microsponges having porous structure and drug release is controlled by predetermined rate. By comparing these three formulations (L2, S3, EPO1) L2 formulation containing EL100 gives the best results.

Keywords: Microsponges, patient compliance, Eutragit S 100, Eutragit L100, Eutragit EPO 100.

INTRODUCTION

The micro sponge technology was developed by Won in 1987 and the original patents were assigned to advanced polymer system, Inc. This company developed a large number of variations of the technique and applied to the cosmetic as well as over the counter (OTC) and prescription pharmaceutical products. At present, this technology has been licensed to Cardinal Health, Inc., for use in topical products[1].

The Microsponge Delivery System (MDS) is a patented polymeric system consisting of porous microspheres. They are tiny sponge like spherical particles that consist of a myriad of interconnecting voids within a no collapsible structure with a large porous surface through which active ingredient are released in a controlled manner. The size of the micro sponges ranges from 5-300µm in diameter and a typical 25 µm sphere can have up to 250000 pores and an internal pore structure equivalent to 10 feet in length, providing a total pore volume of about 1 mL/g for extensive drug retention. The surface can be varied from 20 to 500 mt /g and pore volume range from 0.1 to 0.3 cm/g.This results in a large reservoir within each micro sponge, which can be loaded with up to its own weight of active agent [2-4].

This approach for the new drug delivery designs has been termed as lamentation. Most drug compounds are not inherently long-lasting in the biological system and require multiple daily dosing to achieve the desired therapeutic results. The effects of pharmaceutical ingredients and formulation designs on the biological activity of the drug have been reviewed extensively in various scientific studies. An ideal dosage regimen in the drug therapy of any disease is the one which immediately attains the desired therapeutic drug concentration at the site of action in a constant manner. This is possible through the oral administration of conventional dosage forms in particular dosing intervals throughout the drug therapy. To overcome the inconvenience of multiple dosing, the controlled release or sustained release drug delivery systems have been increasingly gaining popularity in the treatment of various diseases. Such drug formulation designs offer the advantage of conveniently delivering the drug to the systemic circulation and also maintain the desired level of drug into the blood for an extended period of time with a single oral dose the controlled release dosage forms are not only capable of maintaining drug therapeutic levels with a narrow fluctuation range, but they make it possible to significantly reduce the frequency of drug administration. Consider the single dosing of a hypothetical drug that follows a simple pharmacokinetic model for disposition. Depending on the route of administration, a conventional dosage form of the drug, e.g., a solution, suspension, capsule and tablet. The short duration of action is due to the inability of conventional dosage forms to control temporal delivery.

MDS can provide increased efficacy for topically active agents with enhanced safety, extended product stability and improved aesthetic properties in an efficient manner. [5] To control the delivery

rate of active agents to a pre determined site in the human body has been one of the biggest challenges faced by pharmaceutical scientists.[6, 7].

MATERIALS AND METHODS

Diclofenac sodium, Eudragit S100, Eudragit L100, Eudragit EPO 100, were gift samples from Hetero labs, Hyderabad. Polyvinyl alcohol purchased from Loba Chemie Pvt. Ltd.

Fourier Transform Infrared (FTIR) Spectroscopy

The Fourier transform infrared (FTIR) spectra of samples were obtained using FTIR spectrophotometer (Perkin Elmer). Pure drug, individual polymers and optimized formulations were subjected to FTIR study. About 2–3 mg of sample was mixed with dried potassium bromide of equal weight and compressed to form a KBr disk. The samples were scanned from 500 to 4000 cm^{-1}.

Construction of calibration curve for diclofenac sodium

Preparation of stock solution

100 mg of the drug was weighted and dissolve in small amount of ethanol and then made up to 100 mL with distilled water. This was noted as stock I. From stock I, 10 mL of solution was taken and made up to 100 mL with distilled water this was noted as stock II.From this 0.2 mL, 0.4 mL, 0.6 mL, 0.8 mL, and 1.0 mL and solutions was taken and made up to 10 mL with methanol to obtain 2 µg, 4 µg, 6 µg, 8 µg, and 10 µg concentrations respectively. The absorbance of the samples was seen at 275 nm in UV spectrophotometer [8].A calibration curve was constructed by plotting the absorbance against the concentration of diclofenac sodium. A regression equation was derived from the plot.

Microsponge preparation

The microsponges containing Diclofenac sodium were prepared by quasi emulsion solvent diffusion method using the different polymers amounts. In this method internal phase containing polymer such as Eudragit which is dissolved in ethyl alcohol. Then, the drug is slowly added to the polymer solution and dissolved, then plasticizer such as glycerin was added in order to aid the plasticity. The inner phase is then poured into external phase containing polyvinyl alcohol and distilled water with continuous stirring for 2 hours. Then, the mixture was filtered to separate the microsponges. The product (microsponges) was washed and dried in an air- heated oven at 40°C for 12 hr [9].Micro sponges were prepared as per formula in table 1.

Evaluation of micro sponge [10]

The prepared microsponges evaluated for production yield (%), drug content (%), encapsulating efficiency (%) was determined.

Production yield (%)

The production yield of the micro particles was determined by calculating accurately the initial weight of the raw materials and the last weight of the microsponge obtained.

Production yield = Practical mass of microsponges / Theoretical mass (polymer + drug) ×100

Drug content: 100 mg of diclofenac sodium loaded microsponges was taken and mixed with 100 mL of 6.8 pH phosphate buffer. Stirring was carried out for four hours then the mixture was filtered, then filtrate was taken for absorbance.

Encapsulation efficiency (%)

The loading efficiency (%) of the microsponges was calculated According to the following equation:

Encapsulation efficiency = Actual drug content in microsponge / Theoretical drug content × 100

Surface Morphology – SEM

Morphology scanning electron microscopy (SEM), SEM is used widely for which prepared Microsponges are coated with gold–palladium under an argon atmosphere at room temperature and then the surface morphology of the microsponges is studied.

In-vitro drug release study

In vitro dissolution of diclofenac microsponges was studied in USP XXIII dissolution apparatus (Electro lab) rotated at 100 rpm.900 ml of standard buffer pH 1.2 for two hours, followed by pH 6.8 for the next 4 hours and pH 7.4 phosphate buffer solutions for remaining hours were used as dissolution medium. The temperature of dissolution medium was maintained at 37± 0.5^{0C} throughout the experiment. One tablet was used in each test. Samples of dissolution medium (5ml) were withdrawn by means of syringe fitted with pre-filter at known intervals of time and analyzed for drug release by measuring the absorbance at 275 nm. The volume withdrawn at each time interval was replaced with fresh quantity of dissolution medium. The dissolution studies were carried out in triplicate. Cumulative percent drug released was calculated and plotted against time [11].

Dissolution test parameters
Rpm : 100
Time : 60min interval.
λ_{max} : 288 nm
Temperature : 37^0 C ± 0.5^0 C

In-vitro Release kinetics

Dissolution data of above formulations was fitted in Zero order, First order equations.
Zero-Order Kinetics:Zero order as cumulative amount of drug released vs. time,

$$C = K_0 t$$

Where K_0 is the zero-order rate constant expressed in units of concentration/time and t is the time in hours. A graph of concentration vs. time would yield a straight line with a slope equal to K_0 and intercept the origin of the axes [12].
First order kinetics: First order as log cumulative percentage of drug remaining vs. time,

$$Log\ C = Log\ C_0 - k\,t / 2.303$$

Where C_0 is the initial concentration of drug, k is the first order constant, and t is the time [12].

Higuchi Model:Higuchi's model as cumulative percentage of drug released vs. square root of time

$$Q = K\, t_{1/2}$$

Where K is the constant reflecting the design variables of the system and t is the time in hours. Hence, drug release rate is proportional to the reciprocal of the square root of time [13].

Korsmeyer Peppas equations:

To evaluate the mechanism of drug release from dosage form, data for the first 60% of drug release were plotted in Korsmeyer et al's equation log cumulative percentage of drug released vs. log time, and the exponent n was calculated through the slope of the straight line.

$$M\,t / M\,\infty = K\, t\, n$$

Where Mt/M∞ is the fractional solute release, t is the release time, K is a kinetic constant characteristic of the drug/polymer system, and n is an exponent that characterizes the mechanism of release of tracers. For cylindrical matrix tablets, if the exponent n = 0.45, then the drug release mechanism is Fickian diffusion, and if 0.45 < n < 0.89, then it is non-Fickian or anomalous diffusion. An exponent value of 0.89 is indicative of Case-II Transport or typical zero-order release [14].

RESULTS AND DISCUSSION

Diclofenac sodium was selected as the drug for the preparation of microsponges tablets using ethanolic solutions of eudragit, L100, eudagit RS 100, eudragit EPO which act as internal phase and 5 % w/v PVA as external phase for the preparation of microsponge tablets.

Calibration curve of diclofenac sodium

In the present investigation the calibration of diclofenac sodium was carried by using UV-spectrophotometry by measuring the absorbance at 275 nm. The results of absorbance vs. concentration are shown in Table 2 and the calibration curve is shown in Fig. 1. The absorbance vs. concentration obeyed the Beer's law in the range of 2-10 µg/mL. Good correlation was observed between the absorbance and concentration and the coefficient of determination was found to be 0.9979.

Evaluation of diclofenac sodium microsponges

The microsponges were prepared in drug-polymer ratios of L1(1:0.5), L2 (1:1), L3 (1:1.50), L4 (1:2), S1 (1:0.5), S2 (1:1), S3 (1:1.50), S4 (1:2) and EPO 1 (1:0.50), EPO2 (1:1), EPO 3 (1:1.50) and (1:2) by using eudragit L100, eudragit RS100 and eudragit EPO 100 as ethanolic solutions which acts as internal phase for diclofenac sodium and 5 % w/v PVA solution as external phase.

Production yield (%)

Product yield of all the formulations were found to be L1 (62.85±0.12), L2 (80.59±0.18), L3 (63.44±0.68), L4 (60.05±0.64), S1 (60.95±0.28, S2 (60.58±0.64), S3 (65.86±0.75) S4 (55.49±.98), EPO 1 (70.49±85), EPO 2 (64.94±0.85), EPO 3(63.94±0.29) and EPO 4 (55.99±0.57) In this, L2, S3 and EPO 1 showed higher product yield as shown in the Table-3.

Drug loading (%)

Drug loading of the formulations were found to be L1 (43.85±0.11), L2 (47.52±0.85), L3 (40.85±0.37), L4 (38.25±0.64), S1 (35.25±0.720), S2 (40.85±85), S3 (43.85±73) S4 (39.52±0.92), EPO 1 (40.89±0.34), EPO 2 (35.52±0.46), EPO 3(32.35±0.67) and EPO 4 (29.85±0.61). In this, L2, S3 EPO 1 showed higher drug load, in these 3 formulations S3 was found to be less then L2, EPO 1 was less then S3, are show on the Table-3.

Encapsulating efficiency (%)

The entrapment efficiency was found to be L1 (60.53±0.10), L2 (54.52±0.45), L3 (48.65±0.56), L4 (39.85±0.81), S1 (52.45±0.34), S2 (49.52±0.91), S3 (59.62±0.27) S4 (48.52±0.13), EPO 1 (60.52±0.67), EPO 2 (52.86±0.51), EPO 3(45.42±0.24) and EPO 4 (43.12±054). In this, L2, S3, EPO 1 showed higher encapsulating efficiency, in these 3 formulations L3 was found to be higher encapsulating then S3, and EPO 1 are show on the Table-3.

Drug release profiles of microsponges

Drug release studies indicated that microsponge formulations L1, L2, L3, L4, S1, S2, S3, S4, EPO1, EPO2, EPO3, and EPO4 released drug for about 12hrs. During this period, the amount of drug released by L2 is 90.15±0.49 at 12 hrs; S3 is 62.05±0.95 at 12 hrs, EPO1 59.51±0.54 is at 12hrs, among these 12 formulations, L1, S3, EPO 1 shows the maximum release for about 12hr and these are selected for the formulation of tablets.

Keeping the view of drug load (%), product yield (%) and encapsulating efficiency the microsponges are optimized and further preparation of tablets formulation. Results were shown in table no-4 to7 and figure no-2 to 9 From the above 12 formulations, from each polymer one optimized formulation were selected based on production yield, drug content , encapsulating efficiency. From the above tables, the drug release was observed to be about 12 hrs. Release kinetics followed zero order kinetics and the Release mechanism was observed as diffusion mechanism also the type of diffusion was super case –II transport, hence the prepared microsponge (L2, S3, and EPO1) could extend drug release as expected. When compared these three formulations L2 formulation release maximum % of the drug 82.17%.

FT-IR studies

The FT-IR spectra of pure drug diclofenac sodium, pure polymer eudragit L 100 and diclofenac sodium and eudragit L100 are shown in Fig. 10 , 11, and 12.The FT-IR spectrum of diclofenac sodium showed characteristic C–H stretch at 2998 cm^{-1}, aliphatic C=C at 1658 cm^{-1}.The FT-IR spectrum of eudragit L100 showed characteristic C-O stretch at and C=O at 1041 cm^{-1}.The FT-IR spectrum of both drug and polymer showed similar characteristics with minor shifts. The major peaks for the pure drug and the polymer are well in support with the theoretical prediction with respect to the functional groups. This indicates that there is no interaction between drug and the polymer used in the study. Hence FTIR spectral analysis proved the compatibility of the drug and polymer used in the study.

TABLE 1: COMPOSITION OF MICROSPONGESFORMULATIONS

S.NO	FOR CODE	QTY OF DRUG mg	PVA mg	Glycerin mL	EUTRAGIT			Ethanol mL	Distilled water up to mL
					L 100 mg	S 100 mg	EPO 100 mg		
1	L-1	200	50	1	100	-	-	5	100
2	L-2	200	50	1	200	-	-	5	100
3	L-3	200	50	1	300	-	-	5	100
4	L-4	200	50	1	400	-	-	5	100
5	S-1	200	50	1	-	100	-	5	10
6	S-2	200	50	1	-	200	-	5	100
7	S-3	200	50	1	-	300	-	5	100
8	S-4	200	50	1	-	400	-	5	100
9	EPO-1	200	50	1	-	-	100	5	100
10	EPO-2	200	50	1	-	-	200	5	100
11	EPO-3	200	50	1	-	-	300	5	100
12.	EPO-4	200	50	1	-	-	400	5	100

TABLE.2: CONCENTRATION V/S ABSORBANCE MEAN±SD, (N=3)

Concentration (µg/mL)	Absorbance
2	0.057±0.0082
4	0.106±0.0051
6	0.151±0.0019
8	0.185±0.004
10	0.218±0.0043

TABLE.3: PRODUCT YIELD (%), DRUG LOADING (%) AND ENTRAPMENT EFFICIENCY (%)

Formulation code	Product Yield (%)	Drug loading (%)	Entrapment efficiency (%)
L1	62.85±0.12	43.85±0.11	60.53±0.10
L2	80.59±0.18	47.52±0.85	54.52±0.45
L3	63.44±0.68	40.85±0.37	48.65±0.56
L4	60.05±0.64	38.25±0.64	39.85±0.81
S1	60.95±0.28	35.25±0.720	52.45±0.34
S2	60.58±0.64	40.85±85	49.52±0.91
S3	65.86±0.75	43.85±73	59.62±0.27
S4	55.49±.98	39.52±0.92	48.52±0.13
EPO 1	70.49±85	40.89±0.34	60.52±0.67
EPO 2	64.94±0.85	35.52±0.46	52.86±0.51
EPO 3	63.94±0.29	32.35±0.67	45.42±0.24
EPO 4	55.99±0.57	29.85±0.61	43.12±054

All the values are expressed mean±SD, (n=3)

TABLE 4: DRUG RELEASE PROFILE OF EUDRAGIT L100

Time (mins)	L1	L2	L3	L4
15	0.099±0.75	0.204±0.54	0.188±0.78	0.799±0.58
30	3.894±0.34	3.278±0.16	2.63±0.28	4.368±0.18
60	11.756±0.85	20.52±0.45	18.76±0.19	8.796±0.37
120	18.641±0.64	26.8±0.65	20.58±0.18	25.11±0.48
180	32.046±0.42	46.22±0.37	34.65±0.25	36.31±0.28
240	42.498±0.35	56.37±0.18	41.98±0.45	44.9±0.75
360	48.498±0.75	63.75±0.34	44.98±0.27	47.19±0.18
480	50.625±0.19	71.46±0.48	45.05±0.29	60.2±0.48
540	53.152±0.85	76.62±0.19	57.95±0.37	68.6±0.27
720	70.75±0.75	90.15±0.49	71.40±0.67	71.5±0.27

All the values are expressed mean±SD, (n=3)

TABLE.5: DRUG RELEASE PROFILE OF EUDRAGIT S100 MICROSPONGES

Time (mins)	S1	S2	S3	S4
15	0.54±0.75	0.126±0.16	0.155±0.48	0.044±0.18
30	3.014±0.79	2.76±0.18	2.89±0.64	2.8±0.48
60	6.37±0.85	6.17±0.20	13.85±0.24	7.2±0.19
120	15.07±0.83	13.7±0.64	18.52±0.59	15.64±0.54
180	24.24±0.18	22.16±0.85	30.52±0.34	29.42±0.34
240	31.33±0.48	29.27±0.48	35.65±0.75	30.44±0.47
360	35.83±0.48	30.86±0.18	40.58±0.95	36.30±0.58
480	42.84±0.49	42.44±0.85	45.85±0.34	44.05±0.18
540	49.50±0.49	49.26±0.45	50.58±0.82	50.6±0.15
720	57.85±0.75	56.38±0.64	62.05±0.95	57.3±0.24

TABLE.6: DRUG RELEASE PROFILE OF EUDRAGIT EPO MICROSPONGES

Time (mins)	EPO 1	EPO 2	EPO 3	EPO 4
15	0.951±0.17	0.665±0.18	1.12±0.48	0.147±0.54
30	3.28±0.47	3.63±0.24	2.7±0.48	3.63±0.48
60	7.85±0.18	7.45±0.64	8.03±0.48	7.2±0.24
120	24.85±0.19	20.92±0.48	25.36±0.24	12.6±0.48
180	36.89±0.75	29.81±0.64	34.25±0.54	24.96±0.64
240	42.12±0.214	37.72±0.45	38.34±0.48	33.1±0.94
360	47.85±0.18	45.50±0.18	44.52±0.48	38.5±0.48
480	51.30±0.48	54.50±0.64	58.21±0.64	48.52±0.15
540	58.5±0.48	56.85±0.25	59.85±0.84	57.3±0.18
720	62±0.64	59.51±0.54	63.12±0.58	63.44±0.18

TABLE-7: MECHANISM OF DRUG RELEASE STUDIES ON OPTIMIZED FORMULATIONS DICLOFENAC LOADED MICROSPONGES

Tablet	Zero order		First order		Higuchi	Hixson	Korsmeyer-peppas	
	r	K_0 (%/h)	r	K_1(hr^{-1})	r	r	R	N
L2	**0.905**	0.048	0.832	0.0009	0.802	**0.856**	0.984	**1.36**
S3	**0.964**	0.038	0.930	0.0009	0.874	**0.968**	0.968	**1.78**
EPO 1	**0.977**	0.041	0.965	0.0009	0.897	**0.968**	0.968	**1.71**

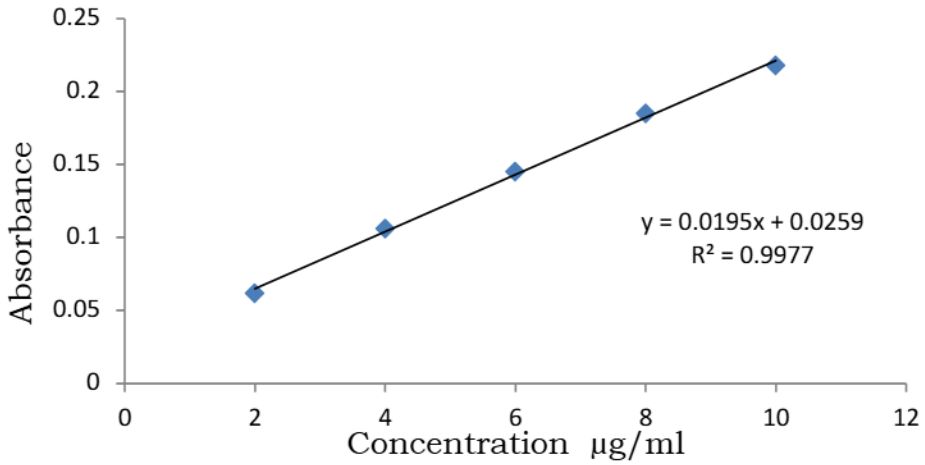


FIG.1: CALIBRATION CURVE OF DICLFENAC SODIUM

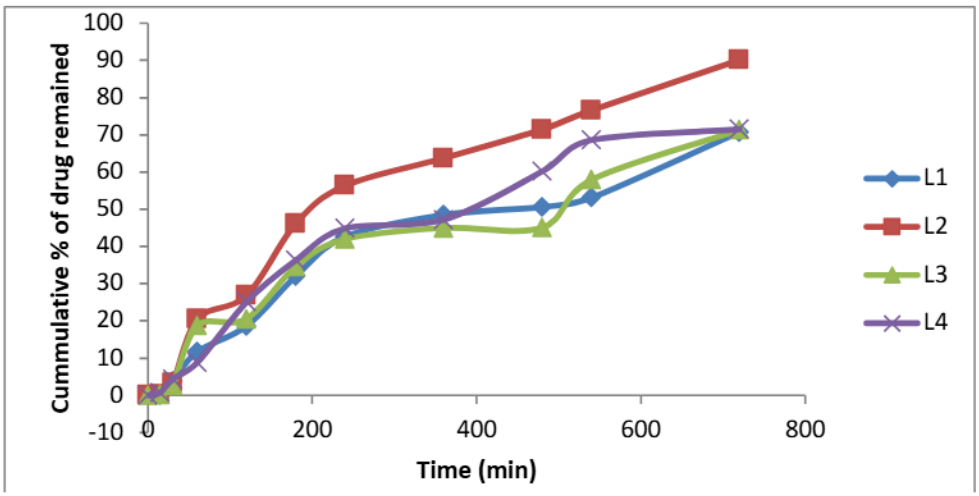


FIG.2: DRUG RELEASE PROFILE OF EUDRAGIT L100 MICROSPONGES

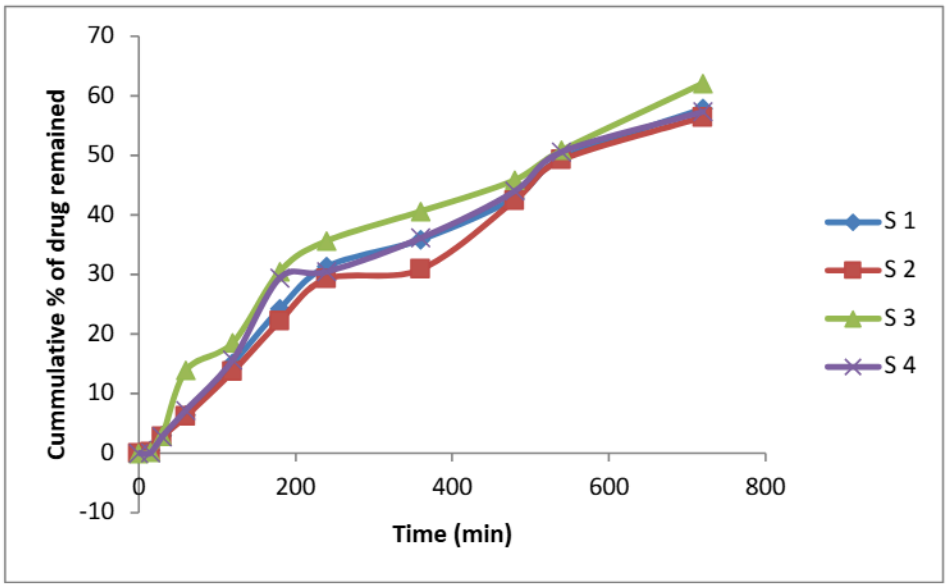


FIG.3: DRUG RELEASE PROFILE OF EUDRAGIT S100 MICROSPONGES

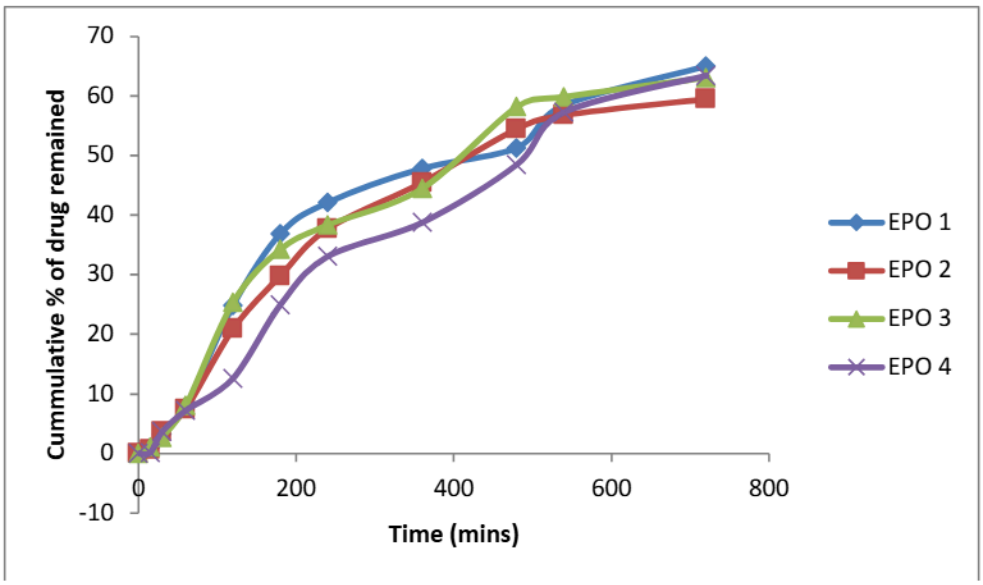


FIG.4: DRUG RELEASE PROFILE OF EUDRAGIT EPO 100

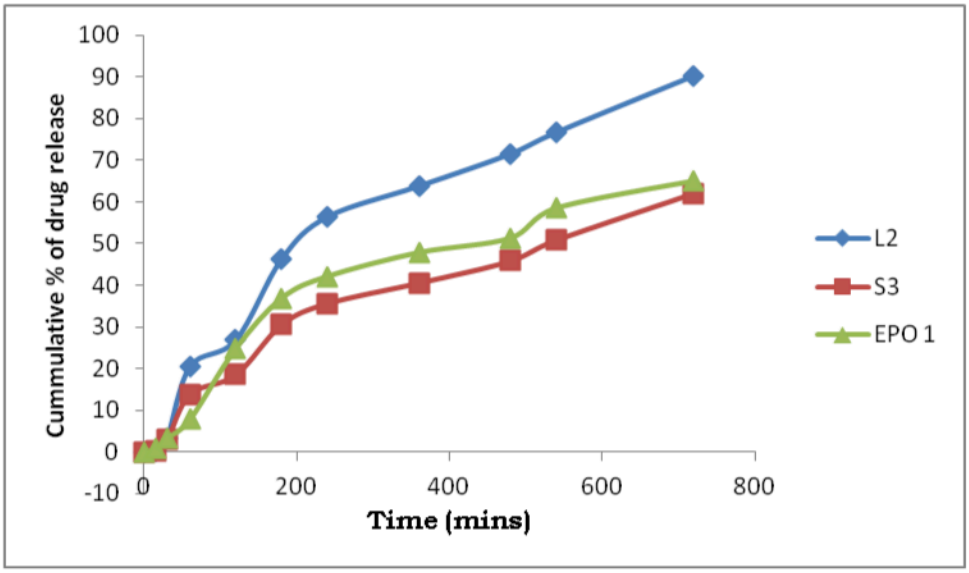


FIG.5: DRUG RELEASE OF OPTIMIZED MICROSPONGES L2, S3, EPO1

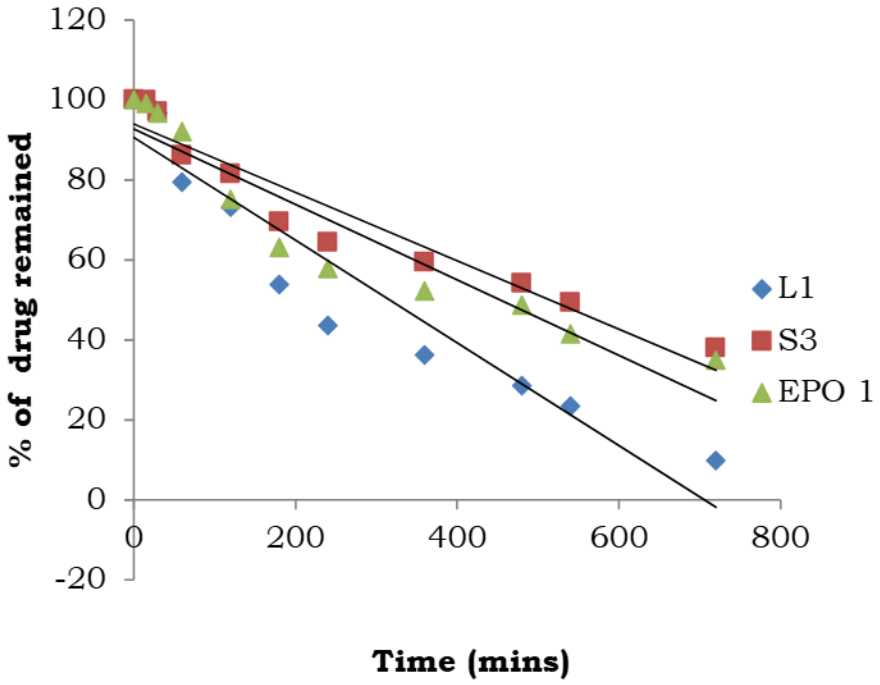


Fig .6: ZERO ORDER PLOT FOR OPTIMIZED MICROSPONGES L2, S3, EPO1

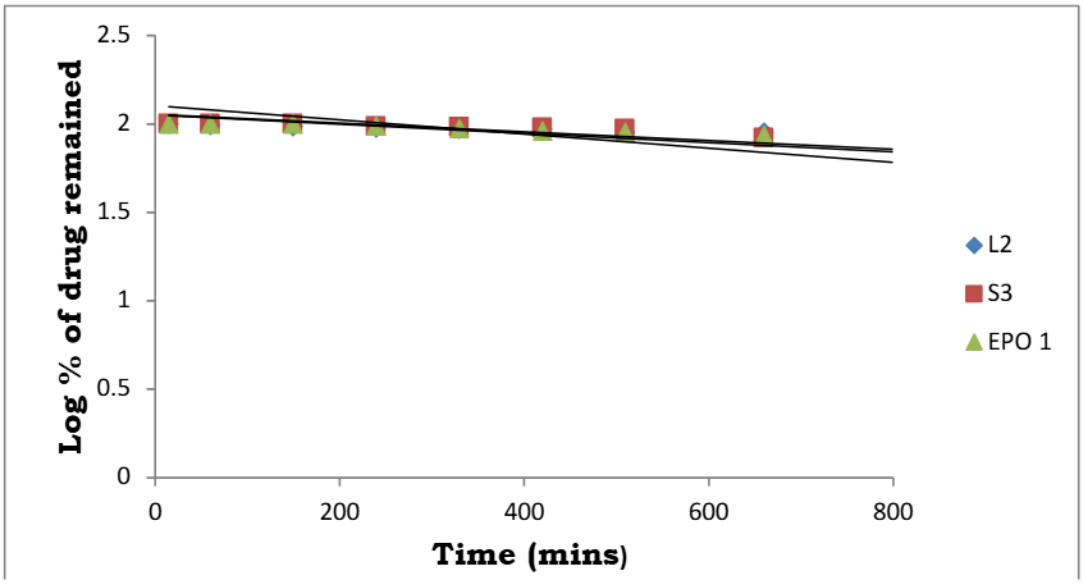


FIG .7: FIRST ORDER RELEASE OF OPTIMIZED FORMULATIONS (L2, S3, EPO1)

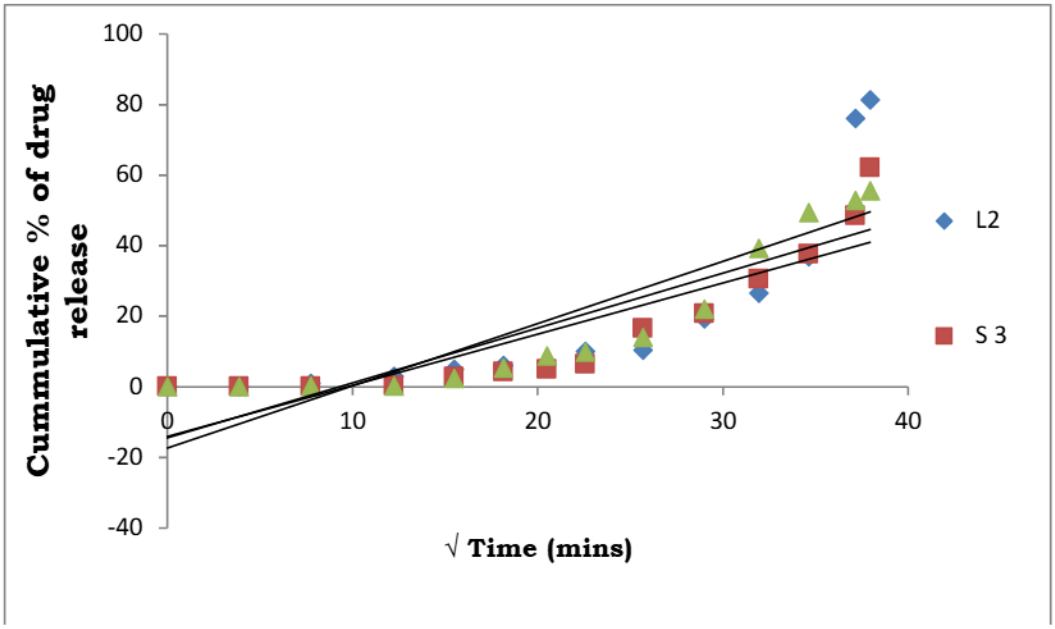


FIG.8: HIGUCHI PLOT OF OPTIMIZED FORMULATIONS (L1, S3, EPO 1)

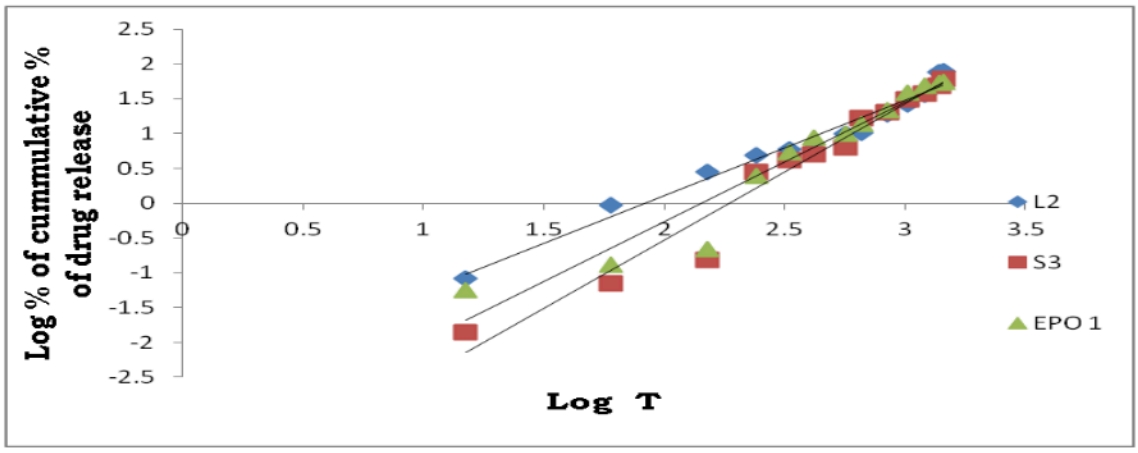


FIG 9: KROSMEYER - PEPPAS PLOT OF OPTIMIZED FORMULATIONS (L2, S3, EPO 1)

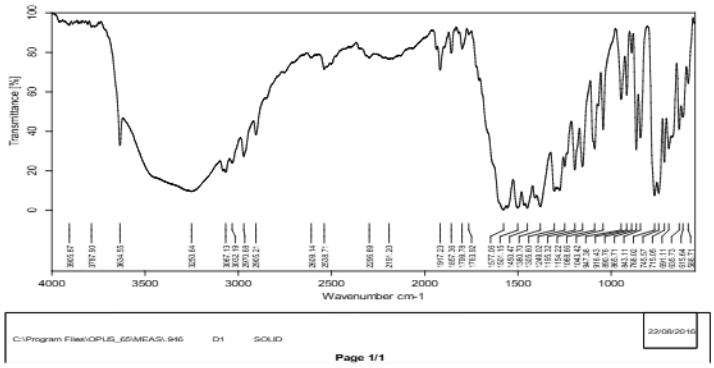


FIG.10: STRUCTURE OF DICLOFENAC SODIUM

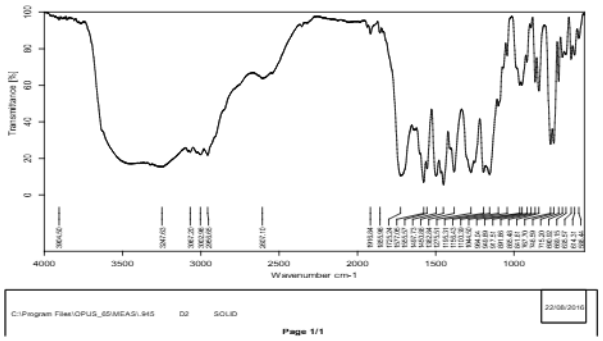


FIG.11: STRUCTURE OF EUDRAGIT L100

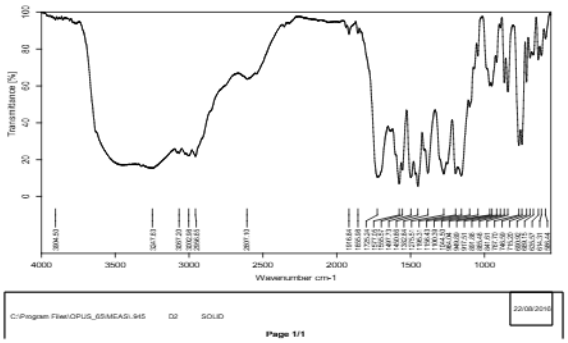


FIG.12: SPECTRA OF DICLOFENAC SODIUM AND EUDRAGIT L100

CONCLUSION

The present work was focused on development of colon targeted diclofenac sodium loaded microsponges which serve the purpose of increasing the drug release by the use of ethanol and eudragit as internal phase and 5% w/v PVA as external phase. Among 12 formulations 3 formulations were optimized formulations among the three formulations L2 gives best release rate of the drug, so it was seen that drug release follows zero order release kinetics and diffusion mechanism with case II transport mechanism.Thus, by this work, I could conclude that microsponges can be used as efficient means of formulation to enhance drug delivery thus helping to enhance the bioavailability of the drug and as efficient carrier through the colonic targeting.

ACKNOWLEDGEMENTS

The author is thankful to DST, Women scientist-A program for sponsoring the project.

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Cite this article as:

S.Janakidevi and K.V.Ramanamurthy. Design of a novel colon targeted microsponges loaded with diclofenac sodium using three different polymers. Int. Res. J. Pharm. 2018;9(4):10-19 http://dx.doi.org/10.7897/2230-8407.09454

Source of support: DST, Women scientist-A program, Conflict of interest: None Declared

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