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Formulation and evaluation of controlled release microspheres containing acid resistant polymers

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ABSTRACT

In the present research work controlled release Microspheres Containing Acid Resistant Polymers of balsalazide were formulated. Balsalazide is delivered intact to the colon where it is cleaved by bacterial azoreduction with pH-dependent solubility. To achieve pH independent drug release of Balsalazide, pH modifying agents (buffering agents) were used. After a long process comprising preformulation study followed by subsequent formulation and evaluation pharmaceutically elegant and technologically competent controlled release system of chitosan and alginate polyelectrolyte complex microspheres were developed and further forwarded to propose a convenient oral solid dosage form to suffice the needful purpose of achieving site specific and sustainable delivery of balsalazide for treatment of colon related disorders. Thorough evaluation process related to performance of microsphere to show optimum swelling and mucoadhesive property with sufficient control over drug release pattern led to selection of suitable candidate and capacity of prepared enteric coated tablet to provide safe and optimum carriage of microspheres to colon where sustained release of balsalazide was expected.

KEY WORDS: Balsalazide, chitosan and Microspheres.

1. INTRODUCTION

Of several developed drug administration methods oral route has found its way to offer the greatest potential for more effective therapeutics, but they do not facilitate drug that easily cross mucosal surfaces and biological membranes; they are easily denatured or degraded, prone to rapid clearance in the liver and other body tissues and require precise dosing. The design of oral control drug delivery systems (DDS) should be primarily aimed to achieve more predictable and increased bioavailability. Control release implies the predictability and reproducibility to control the drug release, drug concentration in target tissue and optimization of the therapeutic effect of a drug by controlling its release in the body with lower and less frequent dose.

Chitosan microspheres are used to provide controlled release of many drugs and to improve the bioavailability of degradable substances such as protein, as well as to improve the uptake of hydrophilic substances across the epithelial layers. It has also been shown to enhance drug absorption via mucoadhesive mechanism. Chitosan based polyelectrolyte complexes have been developed for local or systemic administration of drugs and biodrugs. In particular recent trends in research has been focused on the study of different dosage forms for oral, buccal, nasal, vaginal and intravenous administration of drugs with unfavourable biopharmaceutical properties (peptide and protein, nucleic acids, antipsychotic substances and antihypertensive drugs). Eudragit products are pH-dependent methacrylic acid polymers containing carboxyl groups. Eudragit S 100 coatings have been used to target the anti-inflammatory drug of 5-aminosalicylic acid (5-ASA) in single-unit formulations on the large intestine. Present study comprises use of chitosan alginate polyelectrolyte complex for preparation of mucoadhesive microsphere as matrix tablet for colon targeting.

2. MATERIALS AND METHODS

Materials: Balsalazide Disodium was received as a gift sample from Cipla Research Laboratory. Chitosan was gifted by FMC Biopolymer (India). Alginate, Eudragit S100, SPAN 80 and Hydroxy propyl methyl cellulose (HPMC) was gifted by Chetan & Chetan (India). Lactose monohydrate, Microcrystalline cellulose, Magnesium stearate, Talc and Calcium chloride was gifted by Cabot Sanmer (India).

Methodology: Present study was designed and carried out being backed with earlier reported protocol with suitable modification thereof. Each step of investigation was accurately monitored to get best possible results.

Entire experiment can be briefed as follows:

Preformulation: Standard curve of balsalazide was prepared using U.V. Spectroscopy. Drug polymer interaction was studied with the help of Differential scanning calorimetry (DSC) and Fourier transform Infrared spectroscopy (FTIR).

Formulation and Evaluation: The solution of chitosan in acetic acid and alginate in water were prepared in separate tubes and 2 mM $CaCl_2$ solution was added into the tube with Chitosan solution and homogenized. Nine experimental formulations of Alginate Chitosan (ALG-CHI) Polyelectrolyte complex microspheres were produced in w/o emulsion, as described in earlier work using span80 as surfactant and $CaCl_2$ as cross linker using variable combinations different concentrations of two polymers.

Prepared batches of microspheres were characterized monitoring several parameters like micromeritic properties (particle size, bulk and tapped density, angle of repose, Hausner's ratio and compressibility index), General properties (surface morphology by SEM), Zeta potential by Malvern Zeta sizer, % yield, drug entrapment, swelling index and mucoadhesive property by *in vitro* wash off test) *in vitro* drug release study and corresponding kinetic modelling and stability study to select superior batch.

Selected batch of microspheres were coated with enteric polymer Eudragit S 100 using conventional pan coating method.

Preparation of matrix tablet of microspheres by wet granulation method using Lactose as diluents, HPMC as binder, MCC as disintegrant, magnesium stearate and talc as lubricant.

Prepared tablet was evaluated for all official parameters like weight variation, drug content by assay, friability by Roche friabilator, mechanical strength by Monsento Hardness tester, disintegration time by USP tablet disintegration apparatus and dissolution study using USP dissolution apparatus II in variable simulated physiological pH conditions and corresponding phase wise kinetic interpretation to investigate capacity of enteric coating on improvement of mechanical strength and site specific drug release pattern and stability profile.

3. RESULTS AND DISCUSSION

Standard curve of Balsalazide:

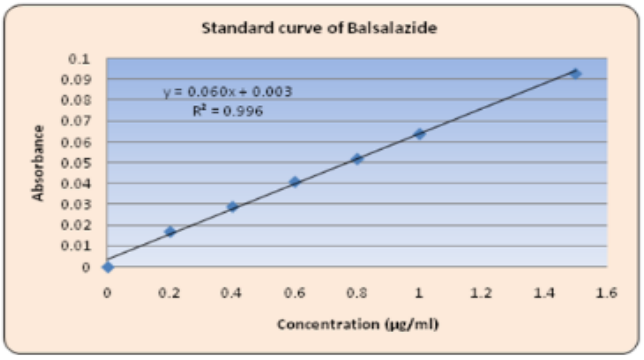

Figure.1. Standard Curve of Balsalazide

Drug Polymer Interaction: The FT-IR and DSC spectra of Balsalazide, chitosan, alginate and their complex forming microspheres were shown in Figure 2 and 3 respectively. In PEC complex showed all identifying peaks characterizing presence of drug, alginate and chitosan with a favorable interaction to form stable microspheres.

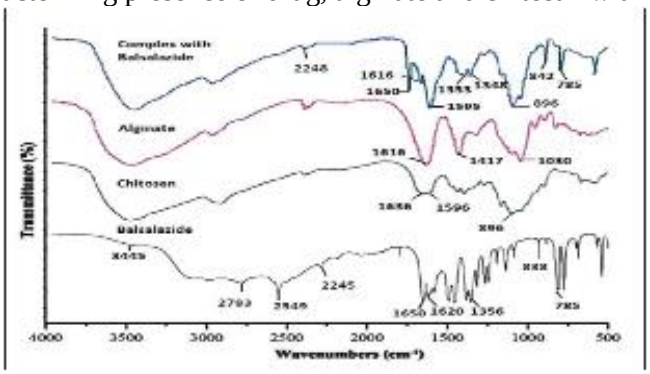

Figure.2. FT-IR Spectra

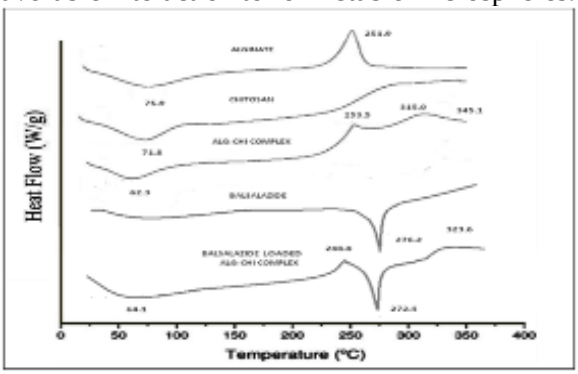

Figure.3. DSC Thermogram

Preparation of Microspheres: As per earlier report in present study Polyelectrolyte Complex (PEC) microspheres were formed as a result of the electrostatic interactions between the protonated amino group cations of chitosan and the carboxylate anions of alginate. Calcium chloride as ionic and covalent crosslinkers was added to the ALG-CHI system for improving the properties and thereby causing a reduction in the hydrogel porosity.

Micromeritic Characterization: Result for comparative evaluation of all micromeritic parameters revealed a reasonable range as in Particle size (71.07±1.35 to101.41±1.33 µm), Bulk density (0.292±0.19 to 0.388±0.09 gm/cc), Tapped density (0.329±0.19 to 0.458±0.09 gm/cc), Hausner's Ratio (1.09±0.07 to 1.224±0.03), Angle of repose (12.76±0.55 to 24.71±0.44°) and Carr's Index (6.26±0.22 to 18.33±0.21) and suggested spherical smooth surfaced, free flowing and compressible formulations within official limit showing with more satisfactory result obtained in M6 due to proper polymer composition.

General Characterization of Microspheres: As a primary observation percentage yield was between 69.49± 0.64 % to 89.97±0.66% of all the formulations with highest value for M6 that was symmetrical with earlier results. The percentage of drug entrapment was increased with the increase in polymer concentration with special reference to the optimum batch i.e. M6 that showed a value of 78.83±0.52% as with earlier data. Swelling index was found to

be a variable depending on the nature of polymer used, their surface charges, degree of interaction to form complex, available porosity after swelling etc. with a satisfactory value of 0.701±0.02 for the batch M6 having well conformity with earlier work. Mucoadhesive property described comparative aspects of % of mucoadhesion after 10 hours in colon pH. It was found to have a range of 68.43±1.11 for the batch CM to 89.63±0.82 % for the batch M6 that differed to attach more to the mucosal tissue from other batches as in earlier findings. Zeta potential had a range of -45.15±0.97 mv found in batch AM whereas maximum charge of +54.09 ± 0.73 mv was found for batch M6 that was found optimal supporting analogous finding.

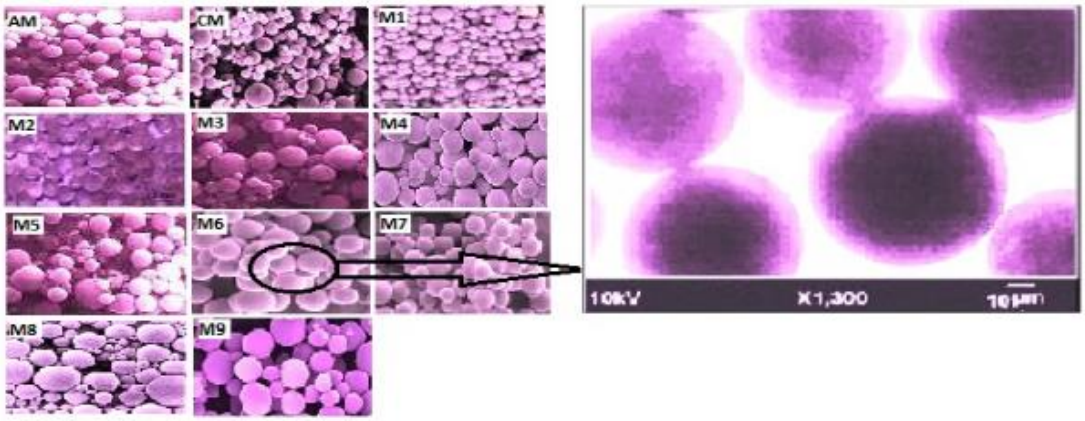

Figure.4. SEM of prepared batches of microsphere with an enlarged image of selected batch M6

In result of Surface Morphology obtained from SEM studies (Figure 4) confirmed the porous and spherical structure of microspheres that revealed the absence of drug crystals on the surface of microsphere, indicating uniform distribution of the drug within the microspheres with no event of aggregation resulted from controlled surface charges yielding optimum zeta potential.

In- vitro Drug release studies of microsphere:

Table.1. *In vitro* drug release study for microspheres

Time Hour	AM % CDR	CM % CDR	M1 % CDR	M2 % CDR	M3 % CDR	M4 % CDR	M5 % CDR	M6 % CDR	M7 % CDR	M8 % CDR	M9 % CDR
0	0	0	0	0	0	0	0	0	0	0	0
0.25	7.56	9.91	4.21	6.36	4.14	4.14	4.14	3.37	4.99	7.56	4.09
0.5	18.78	21.44	10.98	13.09	8.29	5.39	5.39	7.18	11.31	17.78	10.32
0.75	26.34	34.19	29.43	20.11	14.75	18.22	18.22	11.25	21.09	21.34	14.38
1	39.29	45.51	42.43	28.42	25.03	27.31	27.31	16.19	28.26	26.49	22.09
2	56.56	53.33	51.88	34.93	37.22	42.29	38.29	22.53	37.44	31.56	34.16
3	68.56	65.63	60.25	52.16	48.86	57.93	47.93	30.84	40.71	33.56	42.23
4	77.78	78.09	76.11	68.88	61.49	64.41	54.41	39.18	53.28	40.78	55.04
6	90.18	86.74	88.37	75.37	68.31	79.02	59.02	48.04	65.53	45.18	61.9
8	98.64	93.23	95.42	82.02	80.04	84.66	64.66	56.39	78.2	58.64	77.11
10	99.46	95.17	97.29	88.43	87.48	90.17	76.17	66.26	90.41	79.87	89.03
12	99.88	99.08	99.06	90.28	91.17	97.25	88.25	70.91	92.13	95.67	96.14
22	99.89	99.41	99.84	98.11	99.21	99.18	99.18	88.99	94.08	97.14	96.36
23		99.93	99.87	98.79	99.48	99.43	99.43	92.14	94.11	97.49	97.13
24				99.24	99.67	99.79	99.49	95.08	94.64	98.06	97.68

Result shown in Table 1 and Figure 5 revealed at the end of the 24 hr of testing within the test batches (M1-M9) drug release rate was more efficiently controlled and sustained as found in the batch M6 showing more uniform rate of drug release that was maintained uninterruptedly up to 24 hours as a more or less continuous pattern showing 48.04% after 6 hours, 66.26% after 10 hours, 88.99% after 22 hours and finally liberated 95.08% after 24 hours to possess enormous potential to release drug in a predetermined, controlled and reproducible way. Even kinetic modelling resulted accordingly (Figure 6) stating similar evidence of release profile following Higuchi model supporting diffusion related drug release followed Higuchi model with highest correlation co-efficient of 0.992 describing presence of both Fickian and non-Fickian drug release mechanism that corroborated well with earlier results.

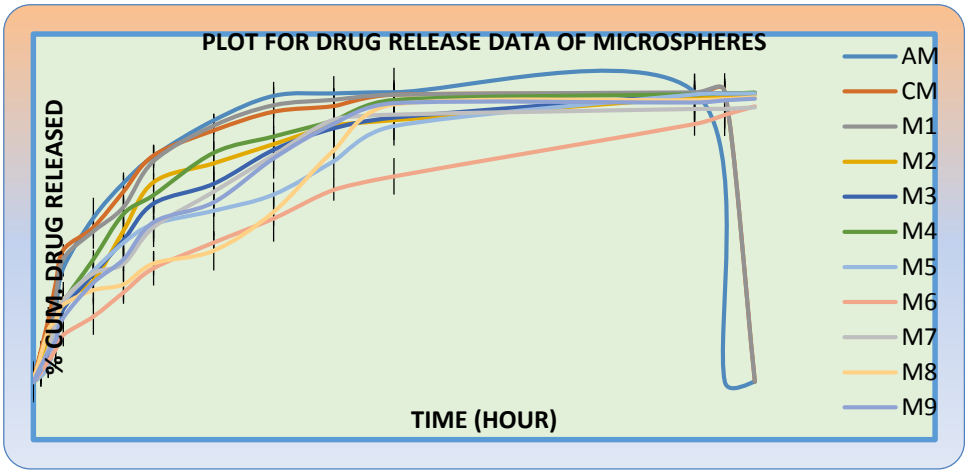

Figure.5. Plot *In vitro* Drug release profile of different batches of microsphere

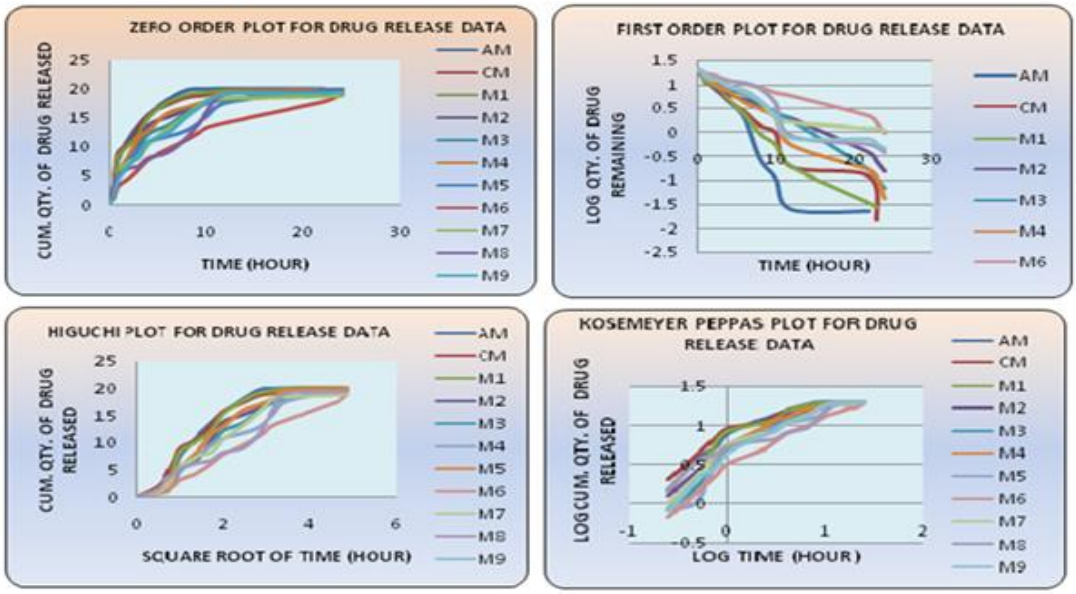

Figure.6. *In-vitro* drug release kinetic of microsphere

Stability Analysis for Selected Batch (M6): Result of such investigation revealed that M6 maintained drug content 99.87±1.01% to 98.05±0.95%, mucoadhesion 89.18±0.88% to 87.95±0.85 %, T50% 6.02±0.08 to 6.11±0.09, T80% 10.04±0.13 to 10.84±0.05 and T90%22.41±0.11 to 22.07± 0.06 hours respectively after 180 days showing very slow rate of decrease with insignificant changes which made symmetry with previous experimental outcome.

Enteric coating of microsphere: The optimized chitosan microspheres were coated with Eudragit S – 100 by emulsion solvent evaporation method.

Preparation of Tablet with coated microsphere (M6): Stemming from previously reported results wet granulation method was adopted for preparation of matrix tablet using microsphere of selected batch M6 with one control batch with uncoated microsphere of same batch. Moreover drug quantity was adjusted in such way that 30% of drug remained outside microsphere to facilitate immediate release and remaining 70% was kept inside microspheres to achieve sustained release at colon satisfying targeting purpose. Three times consecutive coating with Eudragit S100 and sufficient time provided for drying rendered microspheres enough protection from gastric environment and facilitate easy and complete disintegration in colon sufficing effective colon targeting.

Evaluation of tablets: In general evaluation all parameters relating generalized evaluation of uncoated tablet was presented to monitor tablet appearance, size, shape, weight and strength eligible for coating. Weight variation test was conducted as per I.P and the results are shown in Table 2. Average weight of tablet was 99.54±1.28 mg. Proper size distribution of granules and uniform size of the microspheres were found to be responsible for uniform weight as mentioned earlier.

The measured mean hardness of the tablets was found to be 5.5±0.11 Kg/cm^2 and the results are shown in Table 2. This value was found optimum to withstand wear and tear during further handling due to HPMC used as binder in the formula. The results of the friability test were tabulated in Table 2 and it presented as 0.35±0.02 %.The data indicated that the friability was less than 1% ensuring their mechanical stability rendered by binder selection. Thickness of the tablets was found to be almost uniform in tablet. The mean thickness was found to be 2.11±0.09

mm with no sign of significant deviation. Drug content showed a mean value of 98.78±1.42 % indicating the percentage of drug content to be in the official range 95.00% to 101.00% complying with the acceptable limits as per Indian Pharmacopoeia i.e.± 5% probably due to spherical size of microsphere loaded granules and their proper size distribution.

In Disintegration Time data cited in Table 2 shown the mean value of 43.22 ±1.32 minutes demonstrating the performance of simple tablet in variable biological environment. Result of Dissolution Rate Analysis of Tablet of Enteric Coated microspheres was depicted in Figure 7 and Table 3. Result demonstrated that unlike tablets with uncoated microspheres enteric coating of Eudragit S100 facilitated tablet to restrict drug release showing insignificant extent compared to control (p=0.5). After being introduced to SIF enteric coating shown minimal but significant erosion of coating releasing substantial amount (14.29±0.26%) of drug from the region exterior to microspheres in tablet mass for immediate release in comparison to uncoated counterpart that showed more than 50% release in next 4 hours (Table 3) keeping symmetry with previous research. Kinetic interpretation of these release profile as depicted in Figure 6 revealed first order drug release profile was most conveniently adopted in both cases showing higher correlation calculated all three phases. Possibility of higher rate of sustained and controlled drug release resulted from fast disintegration of coating on microspheres followed by controlled swelling and sustained residence of mucoadhesive microspheres in colon providing long standing.

Table.2. Evaluation of Tablet

Parameters evaluated	Observed Value
Avg. Weight(mg)	99.54±1.28
Thickness (mm)	2.11±0.09
Hardness (Kg/cm^2)	5.5±0.11
Friability (%)	0.35±0.02
Assay (%)	98.78±1.42
Disintegration Time	43.22 ±1.32 minutes

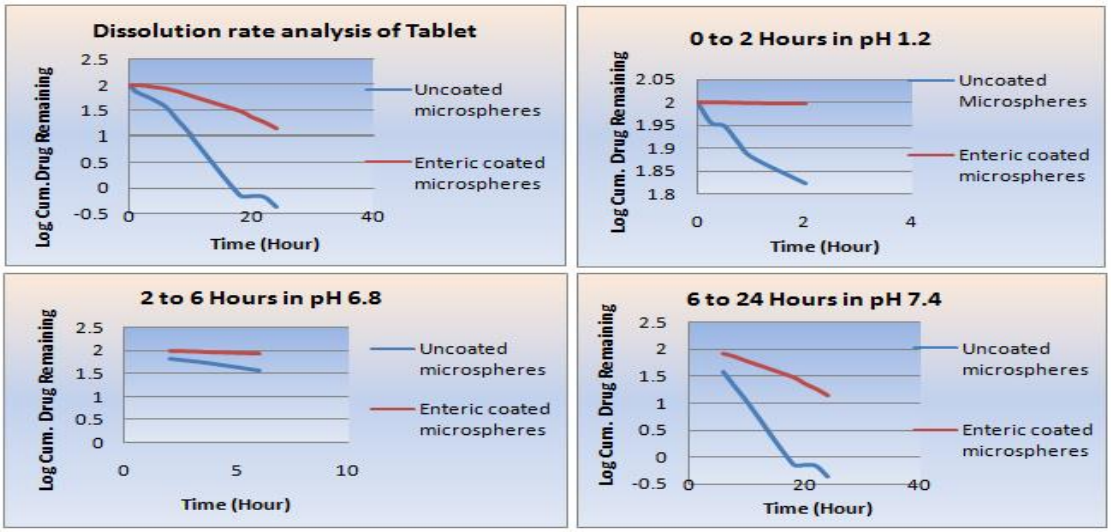

Figure.7. Dissolution rate analysis of tablets of uncoated and enteric coated microspheres

Table.3. Kinetic analysis of tablet dissolution data

Batch	Dissolution Medium	Time period (total 24 hours)	Maximum % CDR	Regression Equation (First order plot)	R^2
Tablet of Uncoated microspheres	SGF (pH1.2)	First 2 Hours	33.56±0.83	Y=-0.086X+1.984	0.958
	SIF (pH 6.8)	Next 4 Hours	62.64±1.02	Y=-0.065X+1.956	0.994
	SCF (pH7.4)	Next 18 Hours	99.57±0.91	Y=-0.111X+2.159	0.967
Tablet of EudragitS100 Coated microspheres	SGF (pH1.2)	First 2 Hours	0.86±0.08	Y=-0.002X+2.000	0.956
	SIF (pH 6.8)	Next 4 Hours	14.29±0.26	Y=-0.016X+2.030	0.993
	SCF (pH7.4)	Next18 Hours	86.18±1.01	Y=-0.043X+2.217	0.991

In Stability Analysis of Tablet enormous studies were undertaken in the investigation of stability of solid oral dosage forms to support post formulation strategies as per ICH guide line. From the results of the accelerated

stability study of tablet of enteric coated microspheres for 6 months, it was concluded that with storage conditions no significant changes were found in the sample monitored with several parameters such as hardness(5.5±0.05 to 6.0±0.11 kg/cm^2), Disintegration time (2.11±0.16 to 2.16±0.21 hours), Drug content(98.81±1.25 to 97.75±0.99%) and T80 (12.18±0.29 to12.06±1.12 hours) after 180 days suggesting its reasonable capacity to withstand accelerated condition that corroborated well with previous researches.

4. CONCLUSION

After a long process comprising preformulation study followed by subsequent formulation and evaluation pharmaceutically elegant and technologically competent controlled release system of chitosan and alginate polyelectrolyte complex microspheres were developed and further forwarded to propose a convenient oral solid dosage form to suffice the needful purpose of achieving site specific and sustainable delivery of balsalazide for treatment of colon related disorders. Thorough evaluation process related to performance of microsphere to show optimum swelling and mucoadhesive property with sufficient control over drug release pattern led to selection of suitable candidate and capacity of prepared enteric coated tablet to provide safe and optimum carriage of microspheres to colon where sustained release of balsalazide was expected.

Entire study indicated possibility for this design to industrial approach with necessity of more effort toward sophisticated design, vivid statistical analysis for optimization and additional accuracy in formulation and evaluation method in order to achieve a prominent goal to develop potential oral tablet dosage form targeted to deliver optimized dose of drug at a controlled rate while residing for significant period of time in colon and/or its nearest possible region. Present work was aimed to provide additional valuable information to support future research in this ever popular field of colon targeted drug delivery system and contribute little more area for extended scientific and robust critical aspects due for more refinement, development and growth.

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REFERENCES

Brahmaiah B, Desu P.K, Nama S, Khalilullah S, SatishBabu S, Formulation and evaluation of simvastatin microsphere, Int J Pharm Biomed Res, 4 (1), 2013, 57-64.

Gawde P, Agrawal S, and Jain P, Development of Mucoadhesive Microsphere for Colon Delivery, International Journal of Pharmaceutical & Biological Archives, 3 (3), 2012, 440-442.

Jadhav K.R, Pawar A.Y, & Talele G.S, Bioadhesive Drug Delivery System: An Overview, Asian J Pharm Clin Res, 6 (3), 2013, 225-230.

Kurnool A.N, Harika J, and Akshatha R.S, Preparation And Evaluation Of Microspheres Of Natural Gums Containing An Anti-Viral Drug World, Journal of Pharmacy and Pharmaceutical Sciences, 4 (7), 2015, 804-816.

Ofokansi K.C, and Kenechukwu F.C, Formulation Development and Evaluation of Drug Release Kinetics from Colon-Targeted Ibuprofen Tablets Based on Eudragit RL 100-Chitosan Inter polyelectrolyte Complexe, ISRN Pharmaceutics, 2013.

Pandey S, Mishra A, Raval P, Patel H, Gupta A, Shah D, A review on Pharmaceutical application of cyclodextrins, J Young Pharm, 5 (4), 2013, 160–166.

Rathore S.B.S, Sharma A, Garg A, Sisodiya D.S, Formulation and evaluation of enteric coated tablet of Ilaprazole, International Current Pharmaceutical Journal, 2 (7), 2013, 126-130.

Sharma S.K, Mohan S, Jaimini M, Formulation and In-Vitro Evaluation Of Modified Release Tablets Of Gliclazide (Antidiabetic Drug), Int J Pharm Pharm Sci, 6 (2), 2014, 259-261.