Development of Novel Floating In-Situ Gelling System for Stomach Specific Drug Delivery System of the Narrow Absorption Window Drug Loratadine

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Abstract

The present investigation deals with the formulation, optimization and evaluation of sodium alginate based in situ gel of Loratadine. Sodium alginate is used as polymer and calcium carbonate was used as a cross linking agent. In situ forming polymeric formulation drug delivery systems is in sol form before administration in the body, but once administered, undergoes gelation In-Situ to form a gel. The formulation of gel depends upon factors like temperature, pH, presence of ions and ultra-violet irradiation, from which drug gets released in sustained and controlled manner. The objective of this study to develop a novel In-Situ gel system for sustained drug delivery using natural polymer. The system utilizes polymers that exhibit sol-to-gel phase transition due to change in specific physico-chemical parameters. In-Situ gel was formed at a biological pH. In vitro release studies were conducted in simulated gastric fluid and cumulative amount of drug release was analyzed by spectrophotometry. From designed set of experiments, it was evident that formulation containing 1.5% of sodium alginate control the release of drug for longer duration. The In-Situ gel exhibited the expected viscosity, pH, drug content, in-vitro gelling capacity, in-vitro floating ability, water uptake ability and sustained drug release.

Key Words

In-Situ gel, Loratadine, pH, 0.1N HCl, polymer, simulated gastric fluid.

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Introduction:

In-Situ gel forming systems have been widely investigated as vehicles for sustained drug delivery. This interest has been sparked by the advantages shown by In-Situ forming polymeric delivery systems such as ease of administration and reduced frequency of administration, improved patient compliance and comfort. In-Situ gel formation occurs due to one or combination of different stimuli like pH change, temperature modulation and solvent exchange. So, In-Situ gelling system via different route such as oral, nasal, ophthalmic etc. can be formulated. Various natural and synthetic polymers such as gellan gum, alginic acid, xyloglucan, pectin, chitosan, poly (DL lactic acid), poly (DL-lactide-co-glycolide) and polycaprolactone are used for formulation development of In-Situ forming drug delivery systems. Gastro retentive In-Situ gelling system helps to increase bioavailability of drug compared to conventional liquid dosage form. The gels formed from In-Situ gelling system, being lighter than gastric fluids, floats over the stomach contents or adhere to gastric mucosa due to presence of bioadhesive nature of polymer and produce gastric retention of dosage form and increase gastric residence time resulting in prolonged drug delivery in gastrointestinal tract. This review attempts to discuss stomach specific In-Situ gelling system in detail including formulation factors to be considered in the development of In-Situ drug delivery system. Also, different types of smart polymers, their mechanisms of gel formation from the sol forms.[1]

The stomach floating In-Situ gel of environmentally sensitive can be affected by many stimulus, these are: temperature, ionic concentration, electrical field, inflammation, solvent concentration, and glucose concentration. According to the mechanism by which sol-gel phase transition occur, the following three types of systems can be recognized

1- pH triggered systems.
2- Temperature sensitive system.
3- Ion activated system.

From the point of view of patient compliance, a liquid dosage form that can sustain drug release and remains in contact for extended period of time, improving the bioavailability, decreasing the dose concentration and frequency may be achieved by In-Situ gelling formulations. Gelation of the orally administered liquid formulations (Ion activated system) was ensured by the inclusion of calcium ions in the formulation as a soluble complex designed to break down to release free calcium ions on encountering the acidic environment of the stomach. The gelation was takes place after the orally administered solution reached the stomach by complexing the calcium with sodium.[2]

Materials and Methods

Materials

Loratadine was obtained from Aarti Drugs Ltd, Sodium alginate, Calcium carbonate, Hydrochloric acid all are of analytical grade.

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Methods

Determination of λmax in 0.1 N Hydrochloric acid

Dilute solution ofLoratadine (10μg/ml) prepared from the above stock solution using solution and its maximum absorption identified through UV hydrochloric acid (0.1N) spectrophotometer by scanning within the wavelength region of 200–400 nm against hydrochloric acid (0.1N) blank. Obtained spectra showing the peak with a highest absorbance (Wavelength 275nm) is considered as absorbance maximum of the drug.

Drug excipient compatibility study
FTIR
Compatibility study was carried out by using Fourier transform infrared spectrophotometer (Shimadzu). FTIR study was carried on pure drug. Physical mixture of drug and polymers were prepared and samples kept for 1 month at 40^0C. The infrared absorption spectrum of physical mixture of drug and polymerswas recorded using KBr disc over the wave number 4000 to 650 cm^{-1}.[4,14]

Preparation of floating gel of loratadine [19,20,21,22]
Sodium alginate suspension of fixed concentration were prepared by adding ultrapure water containing sodium citrate heated to 60^0C with constant stirring on magnetic stirrer. Allowed to cool below 40^0C. Then an appropriate amount of calcium chloride was added with stirring. In another beaker drug was dissolved in 0.1 N hydrochloric acid this is added to above suspension. Calcium carbonate was also added. Methyl paraben is used as preservative. To neutralize above suspension 0.1 N sodium hydroxide was added.

Formulation optimization
3^2full factorial design was applied to the formulation that showed the satisfactory results. To see the effect of concentration of variables Sodium alginate (X1) and Calcium carbonate (X2) on various responses like % drug release. For Sodium alginate lower, middle and higher level were 0.5,1.5 and 2.5 gm respectively. Similarly for the Calcium carbonate lower, middle and higher level were 0.16, 0.33 and 0.50 gm respectively. Composition of all batches is shown in table no.1[9,16]

Formulation code / Ingredient	F1	F2	F3	F4	F5	F6	F7	F8	F9
Loratadine (mg)	50	50	50	50	50	50	50	50	50
Sodium alginate (gm)	0.5	1.5	2.5	0.5	1.5	2.5	0.5	1.5	2.5

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Sodium citrate (gm)	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Calcium chloride (gm)	0.016	0.016	0.016	0.016	0.016	0.016	0.016	0.016	0.016
Methyl paraben (gm)	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18
NaOH (0.1N)	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.
Distilled water (up to ml)	50	50	50	50	50	50	50	50	50

Table 1: Composition of formulation

Evaluation of floating gel

1. Appearance:

The developed formulation met all the pre-requisite to become an In-Situ gelling floating system, gelled and floated instantaneously at the pH condition of the stomach.   [8]

2. pH:

The pH of the in situ solution of Loratadine was measured using calibrated digital pH meter at 37ºC. All measurement of pH was made in triplicate.[6]

3.Viscosity Determination:

The viscosity of the prepared hydrogel formulations were measured at room temperature by Brookfield viscometer (DV-II +) attached with spindle 61. The spindle was rotated at varying rpm and readings were recorded to study the effect of shearing stress on viscosity.[11]

4. Determination of Drug content[3,20]:

The In-Situ solution was dissolved in HCL (0.1N) under sonication and filtered. The drug content was assayed using UV-spectrophotometer (V-630, Shimadzu Co Ltd., Japan) at 275 nm after suitable dilution with 0.1NHCl. Percent drug content was determined using formula

$$\text{Percent Drug Content} = \frac{\text{Actual Drug Content}}{\text{Total Drug Amount Taken}} \times 100$$

5. In vitro floating duration:

The in vitro floating study was determined using USP dissolution apparatus II having 900 ml of simulated gastric fluid (pH 1.2). The medium temperature was kept at 37^0 C. 10 ml prepared In-Situ gel formulations were drawn up using disposable syringe and placed into the petri dish (4.5mm internal diameter) and finally petri dish containing formulation was kept in the dissolution vessel containing medium without much disturbance. The time the formulation took to emerge on the medium surface (floating lag time) and the time the formulation constantly floated on the dissolution medium surface (duration of floating) were noted.[10]

6. Floating lag time:

The floating lag time is defined as time taken by the gel to reach the top from bottom of the dissolution flask. The floating lag time is determined by visual inspection a USP (Type II) dissolution test apparatus containing 900 ml of 0.1N HCl at 37^0C.[7]

7. In vitro drug release study:

The release of Loratadine from sustained release suspension was determined using XXIV dissolution apparatus I (basket covered with muslin cloth) at 50 rpm.This speed was slow enough to avoid the breaking of gelled formulation and was maintained the mild agitation conditions believed to exist in-vivo.The dissolution medium used 900 ml of 0.1 N HCl, and temperature was maintained at 37^oC.A sample (1 ml) of solution was withdrawn from the dissolution apparatus at 0 min.,30 min.,1hr, 2hr,3hr,4hr,5hr,6hr,7hr,8hr of dissolution. The samples were filtered through whatman filter paper and analyzed using UV method. Cumulative % of drug release was calculated and observed.[12]

8. Drug release kinetics:

To examine the drug release kinetics, the release data were fitted to models representing zero order, first order, Higuchi's square root of time kinetics and KorsemeyerPeppas kinetics. The coefficient of determination (r^2) values were calculated from the plots of CDR vs. t for zero order, log %CDR remaining vs. t for first order, %CDR vs. $t^{1/2}$ for Higuchi model and log %CDR vs. log t for Korsemeyer Peppas model, where %CDR is the amount of drug released at time t. The data obtained from study of diffusion kinetics of the optimized formulation was studied to obtain the best fit model. The best fitted model is the one which gives the highest R^2 value and least slope value.

9. Stability studies[13,15]:

Test conditions for stability studies are shown in table no.2

Test Conditions	
Duration of study	3 months
Temperature conditions	40°C± 2°C
Relative humidity conditions	75%RH ± 5%RH
Frequency of testing	1 month, 2 month, 3 months

Table 2: Test conditions for stability studies

The formulations were evaluated mainly for their physical characteristics at 1, 2 and 3 months. Physical appearance in terms of appearance, pH, viscosity and drug content were evaluated.

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Results and discussion
Compatibility study
FTIR
The IR spectra of loratadine, polymer and physical mixture were generated. The IR absorption bands observed in the IR spectrum of drug and polymers resembles with that of found in the physical mixture proves compatibility of drug with polymers.

 Appearance:
The developed formulation met all the pre-requisite to become an In-Situ gelling floating system, gelled and floated instantaneously at the pH condition of the stomach.[17]

Fig. 1: Floating In-Situ gel formulation

pH
The pH of the in situ solution of Loratadine was measured using calibrated digital pH meter at 37⁰C. All measurement of pH was made in triplicate.[6]

Sr. no.	Formulation code	Observed pH (±SD)
1.	F1	7.7 ± 0.10
2.	F2	7.6 ± 0.17
3.	F3	8.1 ± 0.15
4.	F4	7.9 ± 0.10
5.	F5	8.1 ± 0.15
6.	F6	8.1 ± 0.07
7.	F7	7.8 ± 0.26
8.	F8	7.7 ± 0.26

9.	F9	8.1 ±0.20

Table 3: pH values of formulations

The pH of all the formulations from F1 to F9 was found to be in the range of 7.6 to 8.1.

Viscosity Determination

The viscosity values of formulations are shown in table no.4

Rpm	Viscosity (cp) at Room Temperature								
	Formulation code								
	F1	F2	F3	F4	F5	F6	F7	F8	F9
10	448.8	1120	1200	1201	840.1	1132	1342	3301	4780
20	309.2	1123	1144	1133	809.8	1023	948.2	2724	3659
50	300.4	1055	1094	992.4	777.3	948.8	663	2252	2456

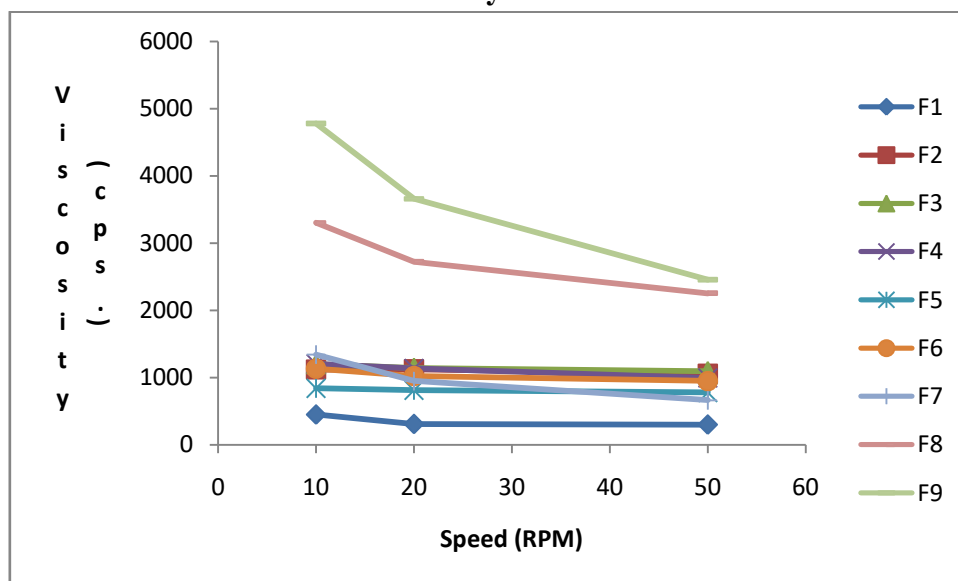**Table 4: Viscosity of formulations**

Fig. 2: Viscosity profile of formulations

Viscosity v/s rpm plots for all formulations shows decrease in viscosity as shear rate (rpm) was increased. Concentration of Sodium alginate was a major factor affecting viscosity of formulations.

Drug content

The Drug content of formulations is shown in table no.5.

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Sr. no.	Formulation code	Drug content (%) (±S.D.)
1.	F1	99.60± 0.00015
2.	F2	99.96 ± 0.00022
3.	F3	98.15 ± 0.00010
4.	F4	99.48 ± 0.00012
5.	F5	99.00± 0.00022
6.	F6	99.24 ± 0.00012
7.	F7	98.76 ± 0.00022
8.	F8	100.2 ± 0.00020
9.	F9	99.48 ± 0.00010

Table 5: Drug content of floating gel

The percentage drug content of all prepared formulations was found to be in the range of 98-100 %.

In vitro floating duration

Sr.no.	Formulation code	Floating time
1.	F1	>8 hr
2.	F2	>8 hr
3.	F3	>8 hr
4.	F4	>8 hr
5.	F5	>8 hr
6.	F6	>8 hr
7.	F7	>8 hr
8.	F8	>8 hr
9.	F9	>8 hr

Table 6: In vitro floating duration of floating gel

Floating lag time

Sr.no.	Formulation code	Floating lag time
1.	F1	>2 min
2.	F2	<1 min
3.	F3	<1 min
4.	F4	>3 min
5.	F5	<2 min

6.	F6	<2 min
7.	F7	>3 min
8.	F8	<2 min
9.	F9	<2 min

Table 7: Floating lag time of floating gel

In-vitro Drug Release Study

Time	Cumulative Drug Release (%) (±S.D.)				
	F1	**F2**	**F3**	**F4**	**F5**
0 min.	0	0	0	0	0
30 min.	17.53 ±0.00007	18.61 ± 0.0001	17.53 ± 0.0001	17.53 ± 0.00007	18.61 ± 0.0001
1 hr	20.56±0.00007	20.68 ± 0.0001	20.56 ± 0.0001	21.64 ± 0.0001	27.17 ± 0.0001
2 hr	23.70 ±0.0001	24.90 ±0.0001	24.79 ± 0.0001	29.24 ± 0.00007	32.84 ± 0.0002
3 hr	28.05 ±0.00007	31.55 ± 0.0001	31.43± 0.0001	35.27 ± 0.0001	39.47 ± 0.0001
4 hr	36.98 ±0.0001	39.74 ± 0.0001	39.63 ± 0.012	48.17 ± 0.00007	53.82 ± 0.0001
5 hr	54.20 ±0.0001	49.63 ± 0.0001	52.43 ± 0.0001	55.65 ± 0.00015	62.85 ± 0.0001
6 hr	68.66 ± 0.00017	52.66 ± 0.0001	52.43 ± 0.0001	71.06 ± 0.00007	73.47 ± 0.0001
7 hr	83.11 ±0.0001	75.05 ± 0.0001	72.66 ± 0.0001	79.02 ± 0.0001	82.13 ± 0.0001
8 hr	90.95 ±0.0001	89.74 ± 0.0001	87.11 ± 0.0001	91.53 ± 0.00026	96.72 ± 0.00007

Time (hrs)	Cumulative Drug Release (%) (±S.D.)			
	F6	**F7**	**F8**	**F9**

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0min.	0	0	0	0
30 min.	18.61 ± 0.0001	15.35 ± 0.00007	17.53 ± 0.00007	15.35 ± 0.00007
1 hr	28.26 ± 0.0001	20.31 ± 0.00015	21.64 ± 0.00015	21.39 ± 0.0001
2 hr	32.25 ± 0.00017	35.36 ± 0.00035	32.48 ± 0.0003	24.66 ± 0.0001
3 hr	39.61 ± 0.0001	43.21 ± 0.00015	47.53 ± 0.00007	32.39 ± 0.00015
4 hr	51.78 ± 0.0063	51.52 ± 0.0003	55.26 ± 0.00015	47.21 ± 0.0001
5 hr	62.75 ± 0.00017	65.75 ± 0.0002	68.75 ± 0.0001	51.33 ± 0.0001
6 hr	74.44 ± 0.0001	75.62 ± 0.0001	77.79 ± 0.0001	69.14 ± 0.00026
7 hr	84.68 ± 0.0001	84.92 ± 0.0001	87.96 ± 0.00007	78.05 ± 0.0001
8 hr	96.58 ± 0.0001	95.64 ± 0.0007	97.04 ± 0.00025	89.51 ± 0.00015

Table 8: Cumulative Drug release of formulations

From the dissolution study it can be said that maximum release is shown by F8 formulation. The data also suggests that gel formulations are capable to produce linear drug release for a longer period of time. The optimized formulation F8 gel shows 97.04% drug release respectively in 8hrs.

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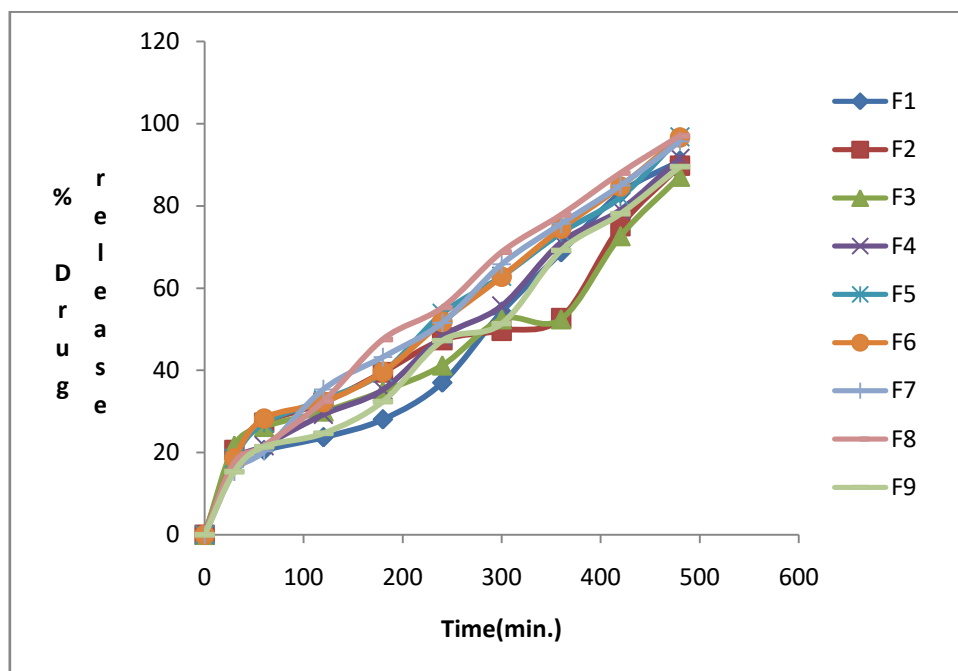System of the Narrow Absorption Window Drug Loratadine

Fig. 3: In-vitro drug release profile of formulations F1 to F9

Drug release kinetics

The classical zero order release curve was found to be linear. The curves plotted according to first order and Higuchi release model were also found to be linear. For the Korsemeyer-Peppas release curves r^2 was found to be ≥ 0.75 for all 9 formulations and n value was found to be ≥ 0.5 which indicates that all the formulations show anomalous (non-Fickian release i.e. swellable matrix).The drug release occurs probably by diffusion and erosion.[18]

Optimization

A 3^2 full factorial design was selected and the 2 factors were evaluated at 3 levels, respectively. The percentage of Sodium alginate (X_1) and Calcium carbonate (X_2) were selected as independent variables and the dependent variable was % drug release. The data obtained were treated using Design expert version 8.0.7.1 software and analyzed statistically using analysis of variance (ANOVA). The data were also subjected to 3-D response surface methodology to study the interaction of Sodium alginate (X_1) and Calcium carbonate (X_2) on dependent variable. Table no 32 shows ANOVA for the dependent variable % drug release. The values of X_1 and X_2 were found to be significant at **p <0.05**, hence confirmed the significant effect of both the variables on the selected responses.From this data optimum concentration of Sodium alginate 2%w/v and Calcium carbonate 3% w/v was found.[5]
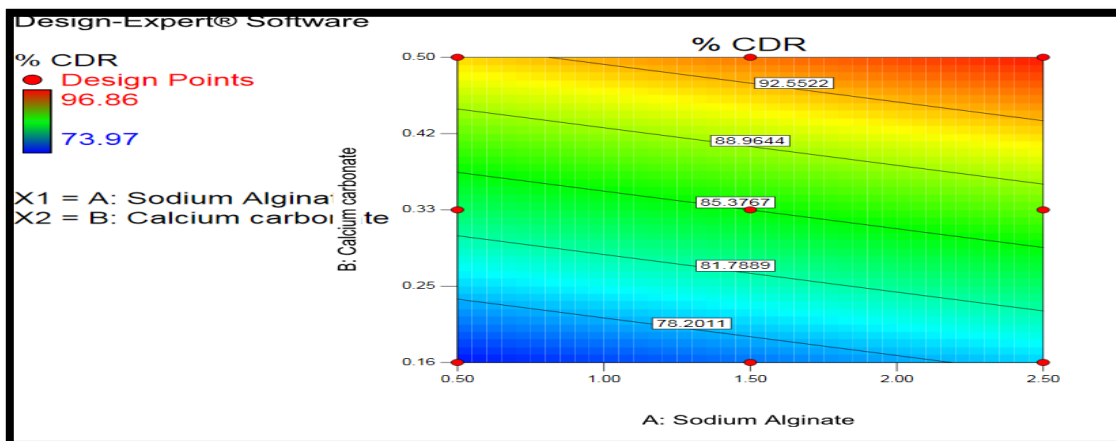
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Fig.4: Counter plot of % drug release for 8hrs

In the above Fig. 4 the contour plot shows that as the concentration of sodium alginate increases % CDR decreases. Hence it can be concluded that the two factorssodium alginate and Calcium carbonate have a combined effect on % CDR
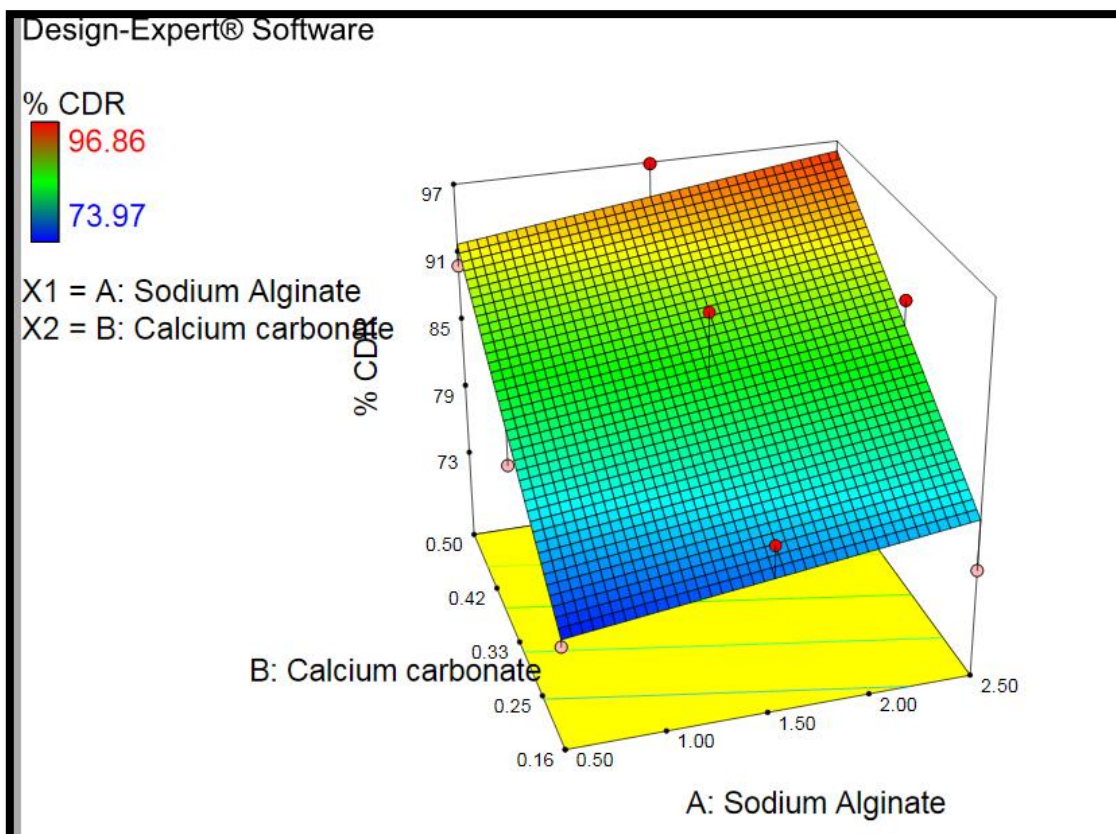

Fig.5: 3D Response curve of % cumulative drug release for 8 hrs

Optimized formula

After generating model, equations relating main effects and responses various gel formulations containing loratadine were optimized based on in vitro drug release at 8 hours. (The optimal values for responses were obtained by numerical analysis based on the criteria

of desirability and optimal batch was selected. Optimized batch (F8) having highest drug release. This revealed that mathematical model obtained by factorial design to produce optimized responses was well fitted.

Accelerated stability study

Results of the stability studies showed that there is no change in the physical parameters of the formulation. Drug content of the formulation was found to be same as that before stability testing.

Conclusion

By preparing the in situ gel of Loratadine the effect of different variables on gel was studied.pH of all the formulations were found to be in between the range (7.7-8.1).The viscosities of all the formulations were determined and it was observed that viscosity was increased due to increasing concentrations of Sodium alginate.The prepared gel was also evaluated for floating lag time, floating duration, % drug content, %CDR (In vitro drug release.)It is one of novel dosage formulations that delivers the drug slowly into the GI tract and provide desired therapeutic effect for long period of time.

References

1. Doshi Nidhi R. In-Situ Gel: A Novel Approach of Gastroretentive drug delivery. Asian Journal of Pharmaceutical Sciences and Research 2013; 3(3):1-14.

2. P. S. Dongare, A. B. Darekar et al. Floating drug delivery system: A better approach. International Journal of Pharmacy and Biological Sciences 2013; 3(4): 72-85.

3. Vinay Wamorkar, M. Mohan Varma et al. Formulation and Evaluation of Stomach Specific In-Situ Gel of Metoclopramide using Natural, Bio-Degradable polymers. International Journal of Research in Pharmaceutical and Biomedical Sciences 2011; 2(1): 193-201.

4. Shital Uttarwar Formulation and Development of In-Situ Gelling System for Nasal Administration for an Antiemetic Drug Ondansetron Hydrochloride by using Pluronics 127P and Pluronics 68. International Journal of Research in Pharmaceutical and Biomedical Sciences 2012; 3(3): 1103-1118.

5. Mowafaq M. Ghareeb et al. Preparation and Characterization of Cinnarazine floating oil entrapped calcium alginate beads. International Journal of Pharmaceutical Sciences and Research 2011; 3(2): 501-508.

6. Shivraju et al Preparation and Evaluation of Ornidazole In-Situ Gelling System for Gastroretentive drug delivery. International Journal of Pharmacy 2013; 3(2): 62-69.

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7. Rama Sarrof et al Sodium alginate based oral In-Situ floating gel of Metformin Hydrochloride. Research Journal of Pharmaceutical, Biological and Chemical Sciences 2012; 3(1): 890-897.

8. Appa Rao Potu et al In Vitro and In Vivo Characterization of Sodium alginate based In-Situ gelling system of Meloxicam for stomach specific drug delivery. Journal of Pharmaceutical and Scientific Innovation 2014; 3(2): 125-134.

9. Vora Vilpul et al Formulation and characterization of novel floating In-Situ gelling system for controlled delivery of ramipril. International Journal of Drug Delivery 2013; 5(1):43-55.

10. Jayvadan K. Patel et al Floating In-Situ gel based on alginate as carrier for stomach-specific drug delivery of Famotidine: Research paper. International Journal of Pharmaceutical Sciences and Nanotechnology 2010; 3(3): 1092-1104.

11. K. S. Parekh et al Formulation development and evaluation of oral In-Situ floating gel of Domperidone. An International Journal of Pharmaceutical Sciences 2013;4(1): 228-246.

12. Swaroop R. Lahoti et al pH triggered sol-gel transition system of ofloxacin for prolonged gastric retention. Pelgia Research Der Pharmacia Sinica 2011; 2(5): 235-250.

13. Sanket Chaniyara et al Formulation and evaluation of floatable In-Situ gel for stomach specific drug delivery of ofloxacin. American Journal of Advanced Drug Delivery 2013; 1(3): 285-299.

14. S. Parthiban et al Formulation and evaluation of gastroretentive drug delivery of Ornidazole In-Situ gelling system using gellan gum. International Journal of Research in Pharmaceutical and Nano Sciences 2013; 2(6): 747-756.

15. Prasad Garepally et al Design, Development and Evaluation of stomach specific In-Situ Gel for Antibiotics: Cefdinir. International Journal of Pharmacy and Biological Sciences 2014; 4(1): 128-137.

16. Rishad R. Jivani et al Development of a Novel floating In-Situ gelling system for stomach specific drug delivery of the narrow absorption window drug loratadine. Indian Journal of Pharmaceutical Research 2010; 9(4): 359-368.

17. Vishvadeep Patel et al A Review on In-Situ gelling system: Novel approach for stomach specific drug delivery. International Journal of Pharmaceutical Research and Bio-Science 201; 3(2): 466-480.

18. Moin K. Modasiya et al Sodium alginate based In-Situ gelling system of Famotidine: Preparation and In-Vivo characterizations. E-Journal of Science and Technology; 27-42.

19. USPNF the Official Compendia of Standards Asian Ed. U. S. Pharmacopoeia the Standards of Quality, Volume-II; USP-31 NF-26 (A-L): 2547-49.

20. British Pharmacopoeia-2009, Published by- MHRA London, Volume-II: 1238-40.

21. http://www.drugbank.ca/drugs/db00455

22. Anthany C., Moffat M David, Osselton and Brian Widdop, Clarks: - Analysis of Drugs and Poisons, 3rdEd .Published by- Pharmaceutical Press, Volume-II: 1186-87.

23. Sehgal.P, Kumar.B, Sharma.M, Salameh A.A, Kumar.S, Asha.P (2022), Role of IoT In Transformation Of Marketing: A Quantitative Study Of Opportunities and Challenges, Webology, Vol. 18, no.3, pp 1-11

24. Kumar, S. (2020). Relevance of Buddhist Philosophy in Modern Management Theory. Psychology and Education, Vol. 58, no.2, pp. 2104–2111.

25. Roy, V., Shukla, P. K., Gupta, A. K., Goel, V., Shukla, P. K., & Shukla, S. (2021). Taxonomy on EEG Artifacts Removal Methods, Issues, and Healthcare Applications. Journal of Organizational and End User Computing (JOEUC), 33(1), 19-46. http://doi.org/10.4018/JOEUC.2021010102

26. Shukla Prashant Kumar, Sandhu Jasminder Kaur, Ahirwar Anamika, Ghai Deepika, MaheshwaryPriti, Shukla Piyush Kumar (2021). Multiobjective Genetic Algorithm and Convolutional Neural Network Based COVID-19 Identification in Chest X-Ray Images, Mathematical Problems in Engineering, vol. 2021, Article ID 7804540, 9 pages. https://doi.org/10.1155/2021/7804540