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Preparation And Evaluation Of Rectal Bilayer Suppositories Of Lidocaine And Aceclofenac For Proctology

Chaitali K. Gunjal, Sameer Shaikh, Naushin Deshmukh , Dr. S. D. Barhate, Swati Rathod.

Shree Sureshdada Jain Institute of Pharmaceutical Education and Research, Jamner (424206), Dist. Jalgaon, Maharashtra, India.

ABSTRACT:

The purpose of this research was preparation and evaluation of rectal bilayer suppositories of lidocaine and aceclofenac were prepared by fusion method (conventional mould). In that different concentrations of polymers like PEG 1000 and PEG 4000 was used. The formulation was optimized by using DOE and Central Composite Design was used for study. And the prepared bilayer suppository was evaluated by various parameters like Appearance, weight variation, content uniformity, drug content, Thickness, and Diameter, Hardness, Friability, melting range test, liquefaction time, Disintegration test, Dissolution study. From the result, S4 batch was optimized formulation because upto 8 hrs 97.54% drug was released. kinetic studies of the drug release for optimized formulation follows first order kinetics. Bi-layered suppository is beneficial technology than the single layered suppository. These immediate and sustained release bi-layered suppositories will be used for haemorrhoids and proctologic disease. In that Lidocaine as immediate release local anaesthetics and Aceclofenac as sustained release non-steroidal anti-inflammatory agent.

Keywords: Bilayer suppositories, Lidocaine, Aceclofenac, Proctology, DOE, Immediate release, Sustained release.

1. INTRODUCTION:

Suppositories are a medicated solid dosage form in tended into the body orifices. The term suppositories have its origin in Latin and means. “To place under’’. It is thought that suppositories were first used in nursing facilities to be administered to elderly patients who were not capable of receiving medication through more traditional delivery system. The rectum is an interesting area for drug absorption because it is buffered and has a neutral pH. It also has a very little enzymatic activity, the rectal mucus is more capable of tolerating various drug related irritations than the gastric mucosa. The ano-rectal physiological provides a sufficiently adequate surface area for drug absorption. The surface area is also permeable to non-ionized drugs. Suppositories formulations are rather efficient in variety of different base to increase absorption and reduce complications. The osmosis process allows the drug to transfer from the vehicle in the suppositories across the membrane of the rectum, and into the haemorrhoidal veins. The higher the concentration and the greater the solubility, the more efficient is the transfer of medication. The designing bi-layered suppositories is to administer fixed dose combinations of different drugs , to separate the incompatible drugs from each other and to control the delivery rate of either single or two different drugs. Most suppositories in this group are used to relive the pain and irritation of haemorrhoids. They contain local anaesthetics such as chinchocaine and benzocaine; astringents such as bismuth subgallate. Drug release from suppositories and subsequent

absorption through the rectum involves several stages, starting from suppository melting or softening at rectal temperature, followed by drug migration through the suppository mass and its transfer from the suppository surface to the rectal environment, and finally drug solubilisation in rectal fluids and drug permeation across the rectal membrane. Drug solubility, particle size of a dispersed drug, and excipient characteristics such as melting temperature, fusion rate, viscosity at rectal temperature, hydrophilic-lipophilic characteristics have a crucial role in release rate of a drug dose from suppositories and the rate of drug absorption. It has been shown that higher drug solubility in the vehicle results in slower drug release and reduced drug absorption from the dosage form. This is attributed to the tendency of the drug to be retained in the base. The same drug dose is therefore able to produce a different therapeutic response when included in excipients with different properties. Furthermore, the excipient properties can affect not only the rate, but also the extent of absorption, especially for drugs that undergo saturable presystemic metabolism. For such drugs, the magnitude of the first-pass effect could vary with the drug release rate from the suppositories. Bi-layered suppository has been specifically developed for many purposes such as providing of two different release rates or dual release of a drug from a single dosage form. Again a combination of the two drugs is feasible with bi-layered suppositories to maximize their individual therapeutic effect and minimize side effects. Various advances in bi-layered suppository technology.

2. EXPERIMENTAL:

2.1 Materials: Lidocaine and Aceclofenac were bought from Medley Pharmaceuticals Pvt. Ltd., Andheri; PEG 1000 was obtained from Encube Ethecal Pvt. Ltd., Andheri and PEG 4000 was obtained from Jinendra Scientific, Jalgaon.

2.2. Experimental Design (DOE) of Immediate and Sustained Release Bilayer Suppository[7,8]: -

The central composite design sampling method is widely used in response surface applications. By selecting corner, axial, and centre points, it is ideal solution for fitting a second - order response surface model. The CCD method also maintained the rotatability of the variation, which is helpful in maintaining the accuracy of model fitting.The central composite design was employed to systematically study the drug release profile to investigated the effect of two independent variables (factors) i.e., the amounts of Polyethylene glycol 1000 (X1) and Polyethylene glycol 4000 (X2) on dependent variable i.e., Percentage Drug release (Y1), Disintegration time (Y2) for both immediate and sustained release suppositories. In these study PEG 1000 (X1) and PEG 4000 (X2) were considered as formulation variable which varied, as required by experimental design and the amount of other excipients were kept constant. The percentage drug release (Y1), and disintegration time (Y2) were selected as response variables. all analysis were performed by using the Design – Expert 7.1.5 Software.

Table No.1: Formulation Variables with their Actual Coded Values for Immediate Release

Formulation Variables	Actual Coded Value	Levels				
		-α	-1	0	+1	+ α
Conc. of PEG 1000	X1	91.3787	92	93.5	95	95.6213
Conc. of PEG 4000	X2	1.37868	2	3.5	5	5.62132

Table No.2: Response Variables with their Actual Coded Values for Immediate Release

Response Variables	Actual Coded values	Unit
% Drug release	Y1	%
DT	Y2	Min.

Table No.3: Factor Combination as per Experimental Design for Immediate Release

Batches	**Conc. of PEG 1000 (X1)**		**Conc. of PEG 4000 (X2)**	
	Actual (%)	Coded	Actual (ml)	Coded
S1	92	-1	3.5	0
S2	95	1	3.5	0
S3	93.5	0	5	1
S4	**93.5**	**0**	**3.5**	**0**
S5	92	-1	2	-1
S6	92	-1	5	1
S7	93.5	0	2	-1
S8	95	1	5	1
S9	95	1	2	-1

Table No.4: Formulation Variables with their Actual Coded Values for Sustained Release

Formulation Variables	**Actual Coded Value**	**Levels**				
		-α	-1	0	+1	+ α
Conc. of PEG 1000	X1	62.9289	65	70	75	77.0711
Conc. of PEG 4000	X2	22.9289	25	30	35	37.0711

Table No.5: Response Variables with their Actual Coded Values for Sustained Release

Response Variables	**Actual Coded Values**	**Unit**
% Drug release	Y1	%
DT	Y2	Min.

Table No.6: Factor Combination as per Experimental Design for Sustained Release

Batches	**Conc. of PEG 1000 (X1)**		**Conc. of PEG 4000 (X2)**	
	Actual (%)	Coded	Actual (%)	Coded
S1	75	1.000	35	1.000
S2	65	-1.000	25	-1.000
S3	70	0.000	30	0.000
S4	**75**	**1.000**	**30**	**0.000**
S5	65	-1.000	35	1.000
S6	70	0.000	25	-1.000
S7	65	-1.000	30	0.000
S8	70	0.000	35	1.000
S9	75	1.000	25	-1.000

2.3 Preparation of Bi-layered Suppositories by Fusion Method[2,3,]: -

Prepare First Layer for Bi-layered Suppositories: Firstly, weigh all ingredients properly. Calculated amount of PEG 4000 and PEG 1000 were melted individually at 40 ^{0} C. Lidocaine was dispersed homogenously into melted base.These mixtures were poured into moulds. Moulds was left in ice bath at 4 ^{0} C. The first layer is prepared by partially filing the moulds with a first fraction of the mass in a predetermined volume. When it gets solidified.

Prepare Second Layer for Bi-layered Suppositories: Firstly, weigh all ingredients properly. Calculated amount of PEG 4000 PEG 1000 were melted individually at 40 ^{0} C. After that aceclofenac was added in melted base. Then these mixtures were poured into lubricated mould and mould was left in ice bath at 4^{0}C. The obtained suppositories were sealed in aluminium packaging coated inside with polyethylene. Suppositories were stored at 2-8 ^{0} C. The second fraction is added into the same mould to get a second layer and cooling them again to room temperature. An addition layer of inert base may also be included into separate two incompatible drugs to minimize the area of contact between the two layers.

2.4 Formulation of Optimized Batches of Bilayer Suppository: -

Sr. No.	Ingredients	Batches								
		S1	S2	S3	S4	S5	S6	S7	S8	S9
1	Lidocaine	0.050	0.050	0.050	0.050	0.050	0.050	0.050	0.050	0.050
2	PEG 4000	27.5	27.5	35.75	44	44	35.75	35.75	27.5	44
3	PEG 1000	522.50	522.50	514.25	506	506	514.25	514.25	522.50	506
4	Aceclofenac	0.100	0.100	0.100	0.100	0.100	0.100	0.100	0.100	0.100
5	PEG 4000	385	330	275	385	330	275	275	385	330
6	PEG 1000	715	770	825	715	770	825	825	715	770

- **All Ingredients are in mg**

2.5 Post-formulation Parameters of Bilayer Suppository:

1) General Appearance : The suppositories are examined with the naked eye (subjected evaluation) to assess the homogeneity of surface appearance and colour for the absence or presence of smoothness or gritty condition, fissuring, pitting, fat blooming, exudation and migration of the active ingredient [24,28].

2) Size and Shape: The width and length of the randomly selected suppositories (six suppositories from each batch) were measured for their physical dimension. After that the same number of suppositories were selected and cut longitudinally and the surface was examined with the naked eye for the homogeneity. Size was measured by vernier caliper and observation.

3) Weight Variation Test: Twenty suppositories were selected randomly; weighed individual suppository and the average weighed was calculated. There must be not more than 2 suppositories differ from the average weight by more than 5 % and no suppository differ from the average weight by more than 10% [26].

Avg. Wt. = Total weight of suppository / No. of suppository

4) Breaking Streangth (Hardness): The ability of the suppositories to withstand the hazards of packing, transportation and handling before usage depends on its mechanical strength (hardness). The hardness of randomly selected 6 suppositories from each batch can be measured by Monsanto hardness tester. The weight required for a suppository to collapse is recorded in kilograms [25,31].

5) Friability Test: Twenty suppositories were weighed and placed in the plastic chamber of Roches Fribilator. The chamber was then rotated for 4 minutes at 25 rpm (a total of 100 revolutions). After 100 revolutions suppositories were removed and weighed again. A loss of less than 1 % in weight is generally considered acceptable [16,17,18]. Percent friability (% F) was calculated as follows :

$$\% F = \frac{\text{Loss in weight (Initial weight} - \text{Final weight)}}{\text{Initial weight} \times 100}$$

6) Micro Melting Range Test: The melting time is a critical factor in the determination of the release rate of the active ingredient(s) from the suppository. This test is also known as macro melting range test. During this test, the time taken for the entire suppository to melt or disperse is measured when immersed in a water bath maintained at constant temperature (37±5 ^{0}C). The time required for the whole suppository to melt or disperse in the surrounding water was noted [29,33].

7) Disintegration Time: The disintegration time is the release rate of the active ingredients from the suppositories. The disintegration times can be recorded utilizing USP tablet disintegration test apparatus (Basket type). Randomaly Selected six formulated suppository placed in beaker of disintegrating test apparatus. 900 ml buffer of pH 7.4 was added in beaker. Temp. was kept constant at 37.5±0.5°C and also set time 60 mins. Then start disintegration process and time taken for disintegration of entire suppository was recorded in phosphate buffer (pH 7.2) [1,21,32].

8) Drug Content (%): The drug content was done by assay method. Firstly prepared suppository was dissolved in 10 ml phosphate buffer of pH 7.4. Prepared solution was filtered through Whatmann filter paper (# 42). Then from filtered solution 0.2 ml withdraw and dilute with 10 ml of phosphate buffer pH 7.4 (10 µg/ml). The drug content was determined at λ_{max} 287.2 nm by using UV spectrophotometer [22,23,27].

Drug Content = Test absorption / Standard absorption x 100

9) In-Vitro Dissolution Profile: Domperidone maleate Sample of 5 ml were withdrawn at specific time intervals (0.5,1,2,…upto 8 hrs). Sample which has been withdrawn were replaced by fresh buffer solution. Drug concentration was analysed spectrophotometrically at 287.2.2 nm. The graph was plotted taking percentage drug dissolve along Y – axis against time X- axis and the dissolution pattern was observed [15,28,30].

$$\% \text{ DR} = \frac{\text{Test Abs.}}{\text{Std. Abs.}} \times \frac{\text{Std. dilution}}{\text{Test dilution}} \times \frac{\text{Purity}}{\text{Label claim}}$$

10) Release Kinetic Model Study: Data obtained from In vitro drug release studies were fitted to various kinetic models such as zero order, first order, Higuchi and Korsmeyer-Peppas to predict the drug release mechanism. The release rate constants (k), release exponent (n), and determination coefficients (R2) were calculated by means of a computer program (Microsoft Excel, 2019 version) [18,19,20].

3. RESULT AND DISCUSSION:

1) Identification of drugs by UV- spectroscopy:

a) UV Estimation of Lidocaine : **b) UV Estimation of Aceclofenac :**

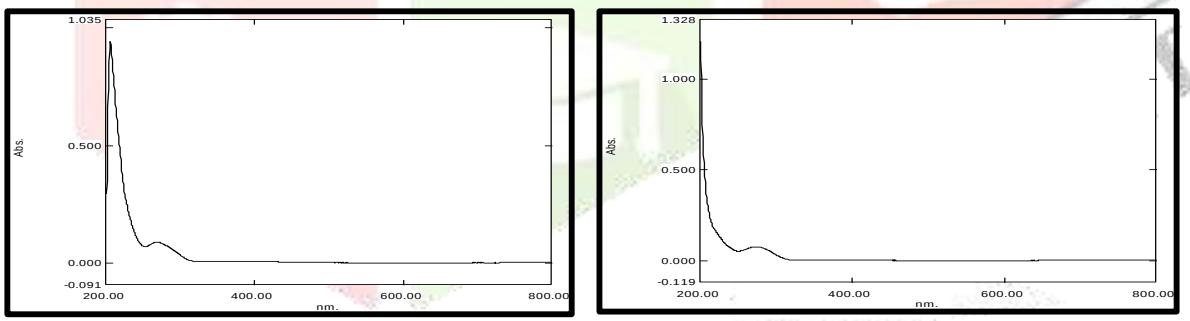


Fig.1 Wavelength Maxima of Lidocaine and Aceclofenac in Phosphate Buffer pH 7.4

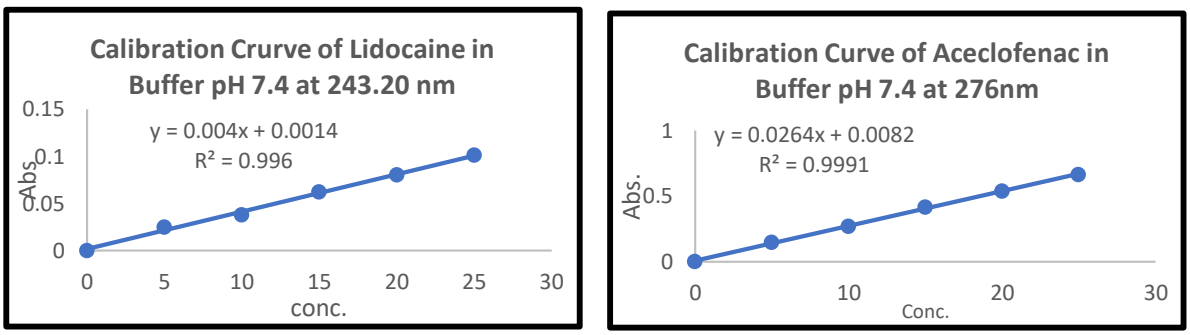
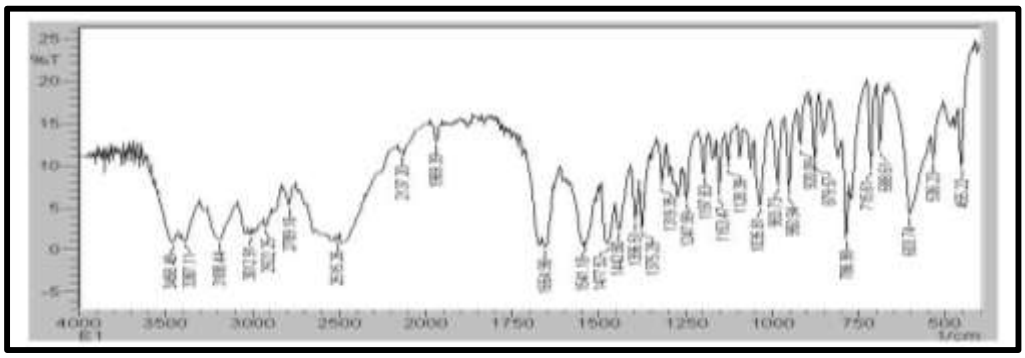


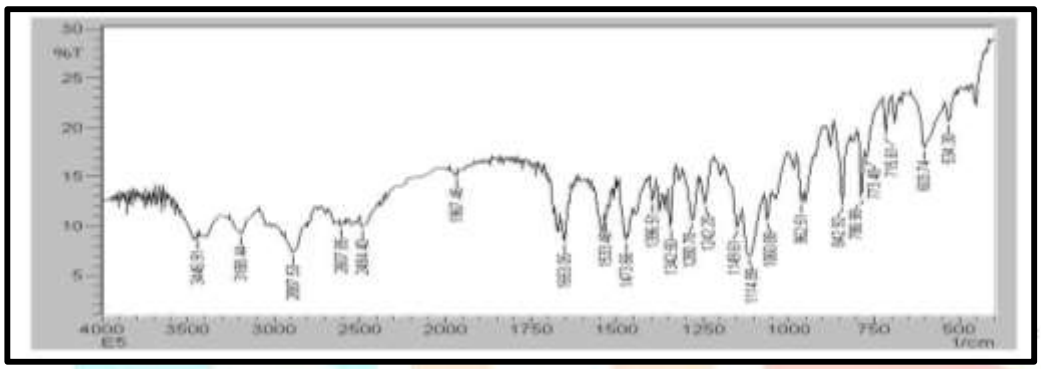
Fig.2 Calibration Curve of Lidocaine and Aceclofenac in Phosphate Buffer pH 7.4

2) Spectroscopic Study :

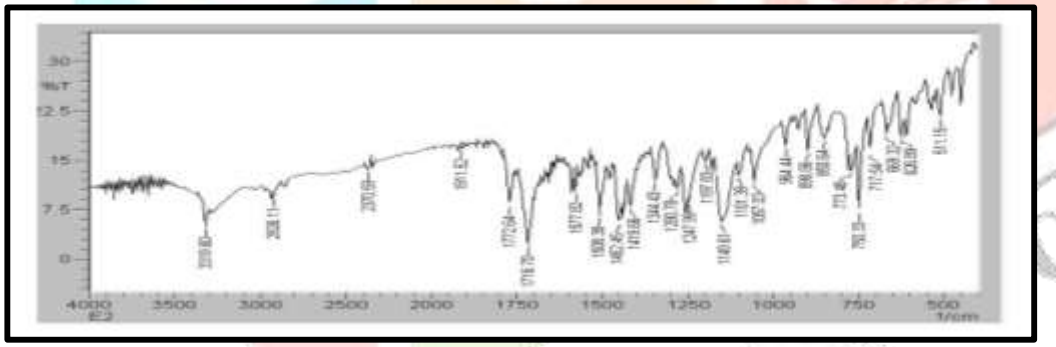
a) FTIR Spectrum of Lidocaine :



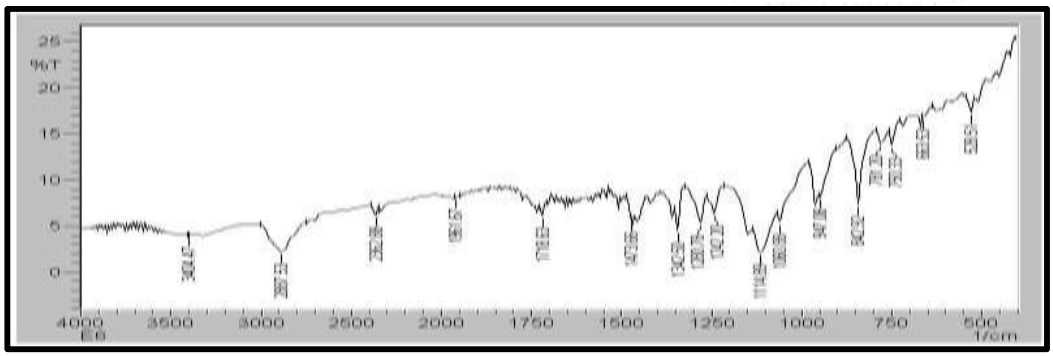
b) FTIR Spectrum Overlay Spectrum of Lidocaine and Excipient :



c) FTIR Spectrum of Aceclofenac :



d) FTIR Spectrum Overlay Spectrum of Aceclofenac and Excipient :



3) Differential Scanning Calorimetry (DSC) Thermograms :

a) DSC Thermogram of Lidocaine :

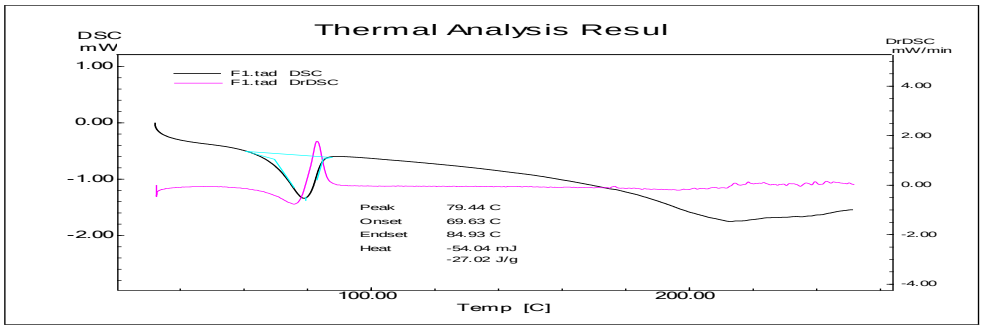


Fig.3: DSC Thermogram of Lidocaine

The DSC curve of lidocaine show sharp endothermic peak (72 ^{0}C) corresponding to its melting point, indicating its pure in nature. The melting point of lidocaine (72 ^{0}C) matches with the standard value, and that show no any interaction between drug and polymer.

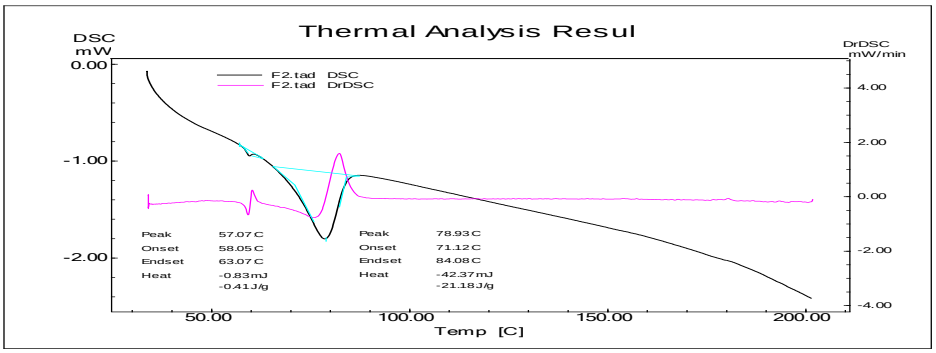


Fig.4: DSC Thermograms of Lidocaine and Excipient

b) DSC Thermogram of Aceclofenac :

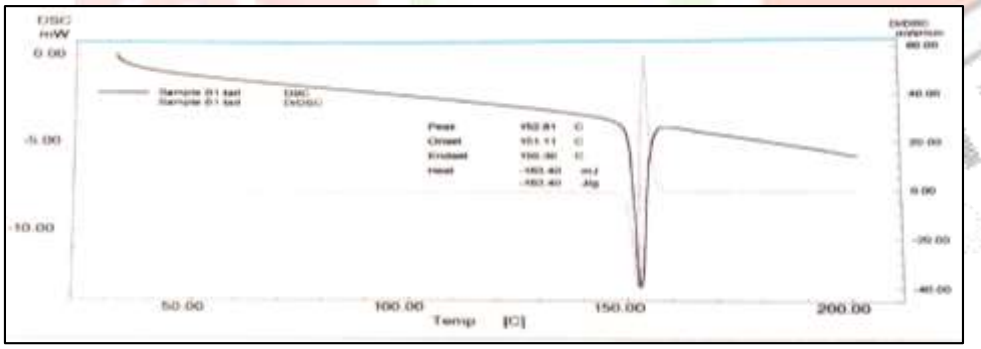


Fig.5: DSC Thermograms of Aceclofenac

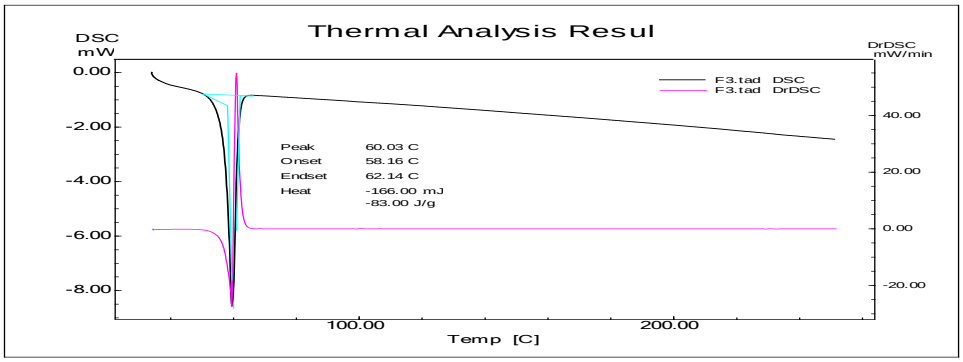


Fig.6: DSC Thermograms of Aceclofenac and Excipient

4) Evaluation of Post formulation Parameters:

1) Appearance, Size and Shape:

Table 7. Appearance Size and Shape of Optimized Batches of Bi-layered Suppositories

Sr. No.	Formulation	Colour and Appearance		Surface Texture
		First Layer	Second Layer	
1	LABS1	White	Off – White	Smooth
2	LABS2	White	Off – White	Smooth
3	LABS3	White	Off – White	Smooth
4	**LABS4**	**White**	**Off – White**	**Smooth**
5	LABS5	White	Off – White	Smooth
6	LABS6	White	Off – White	Smooth
7	LABS7	White	Off – White	Smooth
8	LABS8	White	Off – White	Smooth
9	LABS9	White	Off – White	Smooth

Table 8: Evaluation of Optimized Batches of Bi-layered Suppositories: -

Evaluation Parameters	Batches								
	LABS 1	**LABS 2**	**LABS 3**	**LABS 4**	**LABS 5**	**LABS 6**	**LABS 7**	**LABS 8**	**LABS 9**
Weight Variation (gm)	1.55	1.46	1.66	**1.62**	1.59	1.68	1.68	1.54	1.60
Melting range time (min)	31.82	33.5	32.9	**33.7**	34.6	32.5	33.4	36.8	35.2
Hardness (kg/cm) ^{2}	2 .20	2.25	2.22	**2.33**	2.33	2.25	2.15	2.12	2.20
Liquefaction Time (min)	8.50	8.15	9.15	**9.55**	8.40	8.20	8.26	8.25	9.80
Disintegration Time (min)	24.55	23.12	24.21	**23.23**	21.22	23.45	23.45	22.15	25.5
Drug Content (%)	96.6	93.5	95.5	**98.2**	99.5	95.8	95.4	98.7	94.6
Thickness (mm)	9.65	9.25	9.45	**9.55**	9.22	9.23	9.65	9.84	9.66
Invitro drug release	116	86.43	100.32	**95.23**	81.25	85.61	87.41	92.52	90.02

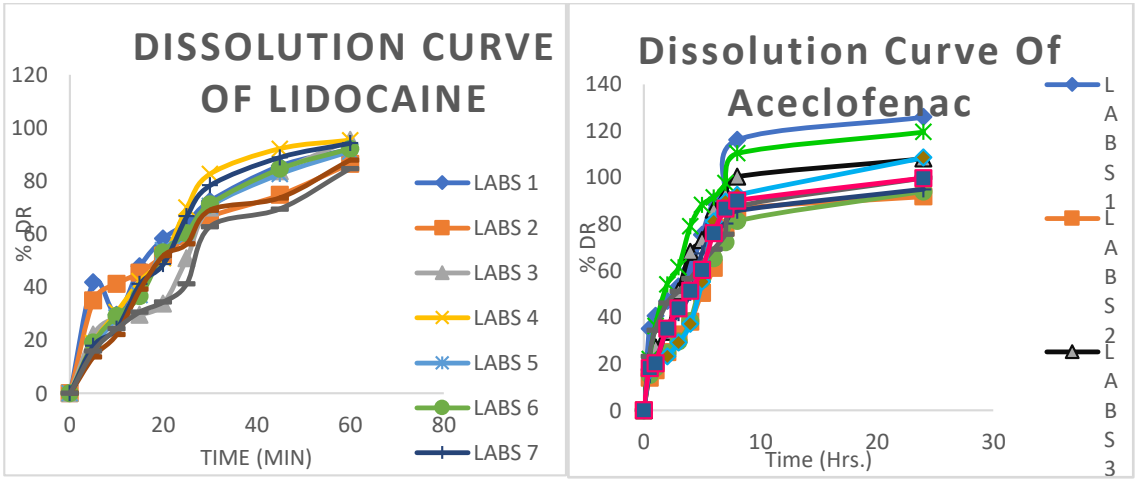
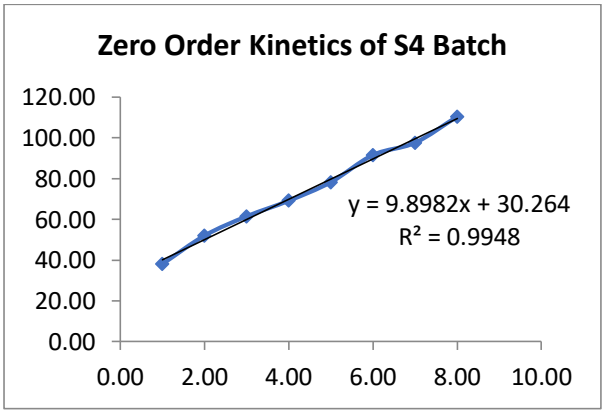


Fig. 7 and 8 In – Vitro Drug Release study of LABS 1 to LABS 9 Batch for Immediate Release and SR Layer of Bi-layered Suppositories

3) Drug Release Kinetics Model Fitting of the Dissolution Data of Optimized Batches :

Table 9 : Results of Drug Release Kinetics Model of Optimized Formulation

Batch	Zero order		First order		Higuchi model		Korsemeyer-peppas	
	K	R^2	K	R^2	K	R^2	K	R^2
LABS1	8.1982	0.9455	0.121	0.9177	31.842	0.9387	0.5414	0.9486
LABS2	10.108	0.979	0.2125	0.9917	38.375	0.9287	0.9221	0.9707
LABS3	10.921	0.9775	0.168	0.8931	42.692	0.9831	0.7714	0.9783
LABS4	**9.8982**	**0.9948**	**0.123**	**0.9917**	**38.318**	**0.9812**	**0.5383**	**0.9803**
LABS5	9.574	0.9808	0.2112	0.9607	36.618	0.9443	0.9322	0.9724
LABS6	9.611	0.9765	0.1591	0.9442	37.584	0.9828	0.7099	0.9772
LABS7	6.8874	0.986	0.1037	0.9968	26.598	0.9679	0.4448	0.9534
LABS8	12.128	0.944	0.2567	0.9542	45.936	0.8913	1.1263	0.9546
LABS9	10.124	0.9878	0.1634	0.9789	39.239	0.9767	0.7168	0.9786



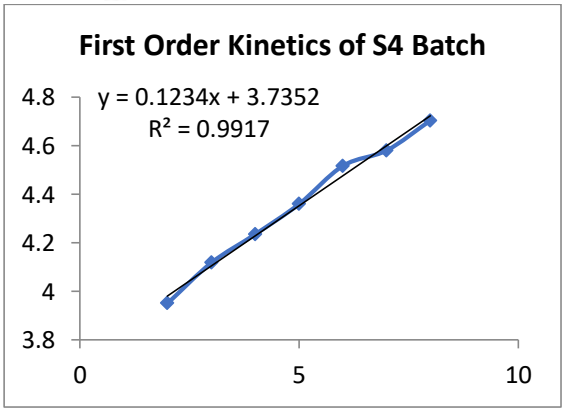


Fig. No. 9: Zero Order Kinetics S4 Batch

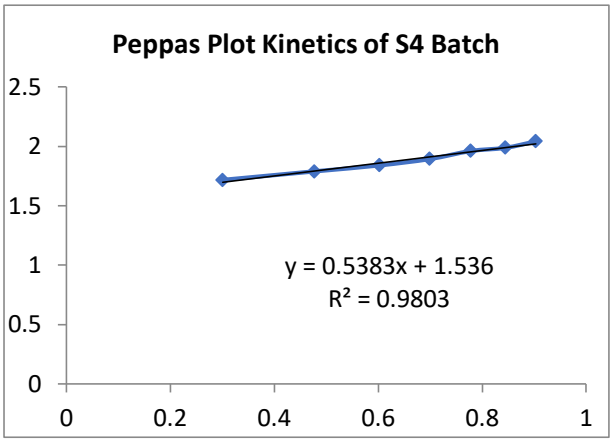


Fig. No. 10: First Order Kinetics S4 Batch

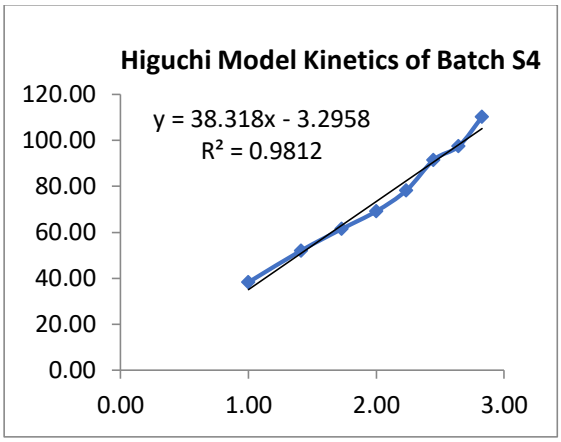


Fig. No. 12: Peppas Plot Kinetics S4 Batch

Fig. No. 11: Higuchi Model Kinetics S4 Batch

3.4 Optimization and Data Analysis :

The central composite design was applied to optimize the immediate release lidocaine suppositories the response surface methodology was used to illustrate the quantitative effect of variable on response. The data of in-vitro drug release, disintegration time and % friability were gathered after the study of preliminary trial batches (S1-S9) and optimized batches (LABS 1-LABS 9) immediate release lidocaine bilayer suppositories was used to target the response Y1, Y2 and disintegration time minimizing and the drug release maximized.

First Layer: - Lidocaine

Table No.10: Result of Optimization Batches by Central Composite Design.

Variable Level	Actual Coded Value	Levels				
		-α	Low (-1)	Medium (0)	High (+1)	+ α
Conc. of PEG 1000	X1	91.3787	92	93.5	95	95.6213
Conc. of PEG 4000	X2	1.37868	2	3.5	5	5.62132

Run	Batch	Factor 1 X1 Conc. of PEG 1000 (%)	Factor 2 X2 Conc. of PEG 4000 (%)	Dependent Variables	
				Drug Release (%) Y1	DT (min) Y2
1	S1	2	3.5	91.89	22.52
2	S2	95	3.5	86.48	22.19
3	S3	93.5	5	95.52	28.61
4	**S4**	**93.5**	**3.5**	**95.27**	**28.82**
5	S5	92	2	91.04	21.85
6	S6	92	5	92.09	26.62
7	S7	93.5	2	94.21	29.24

8	S8	95	5	87.86	20.83
9	S9	95	2	84.63	26.94

Regression Equations for the quadratic model

% Drug release of Lidocaine (Y1) = +95.21 -2.67X1 +0.9317X2 +0.5450X1X2 -6.00$X1^2$ -0.3217$X2^2$

Disintegration Time (Y2) = +28.11 -0.1717X1 -0.3283X2 -2.72X1X2 -5.40 $X1^2$ +0.3217$X2^2$

Regression Equations for % Drug Release of Lidocaine

% Drug release of Lidocaine (Y1) = +95.21 -2.67X1 +0.9317X2 +0.5450X1X2 -6.00$X1^2$ -0.3217$X2^2$

The % drug release of lidocaine at 20 min varied from 84.63 % to 95.52 %. The results of multiple regression analysis showed that negative value of X1 indicate inverse relationship with response Y1 (% drug release). That is % drug release increases with decrease in conc. of PEG 1000 and increase with increases in conc. of PEG 4000.

Graphical Representation

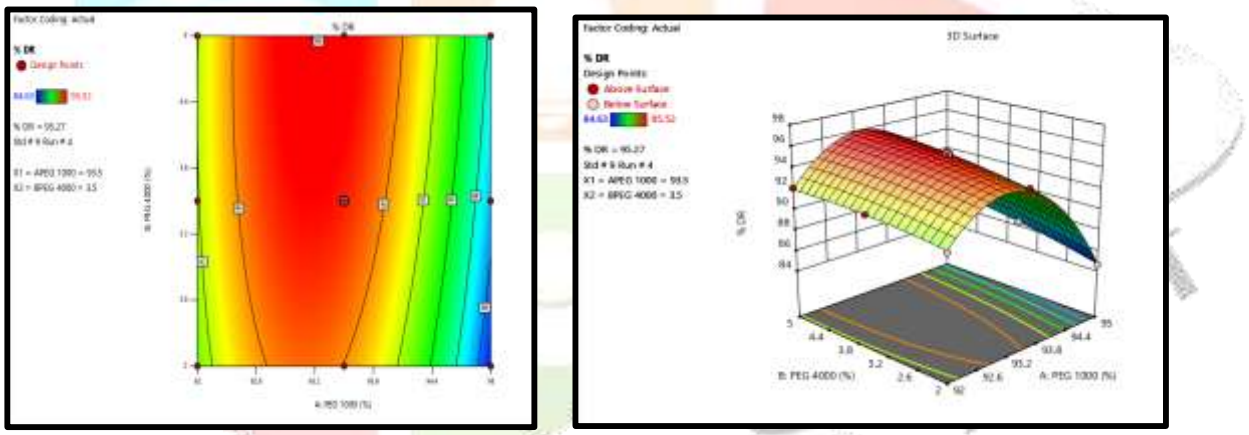


Fig. No. 13 and Fig. 14: Response Surface Contour Graph and 3-D Surface Graph Showing the Influence of Conc. of PEG 1000 (X1), Conc. of PEG 4000 (X2) on % Drug Release

Table No. 11: Summary of Results of Analysis of Variance

1) ANOVA for Quadratic Model : Response 1: % Drug Release (Y1)

Source	Sum of Squares	DF	Mean Square	F-Value	p-Value	
Model	121.58	5	24.32	304.86	0.0003	Significant
A-PEG 1000	42.93	1	42.93	538.29	0.0002	
B-PEG 4000	5.21	1	5.21	65.30	0.0040	
AB	1.19	1	1.19	14.90	0.0307	
A^2	72.04	1	72.04	903.22	< 0.0001	
B^2	0.2069	1	0.2069	2.59	0.2056	
Residual	0.2393	3	0.0798			

Cor Total	121.82	8				

Regression Equations for Disintegration Time of Lidocaine

Disintegration Time (Y2) = +28.11 -0.1717X1 -0.3283X2 -2.72X1X2 -5.40 $X1^2$ +0.3217$X2^2$

Concerning the disintegration time, the results of multiple linear regression analysis showed that the coefficients X1 and X2 bear a negative sign. Therefore, increase in the concentration of X1 and X2 is expected to decrease the disintegration time.

Graphical Representation

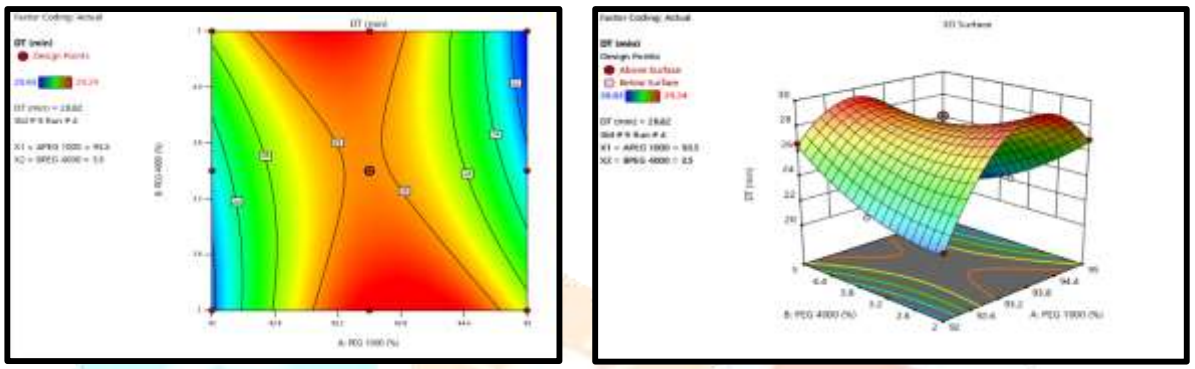


Fig. No. 15 and Fig.16: Response Surface Contour Graph and 3-D Surface Graph Showing the Influence of Conc. of PEG 1000 (X1), Conc. of PEG 4000 (X2) on Disintegration Time

Table No.12 : Summary of Results of Analysis of Variance

1) ANOVA for Quadratic Model

Response 1: % Disintegration Time (Y2)

Source	Sum of Squares	DF	Mean Square	F-Value	p-Value	
Model	91.45	5	18.29	48.20	0.0046	Significant
A-PEG 1000	0.1768	1	0.1768	0.4659	0.5438	
B-PEG 4000	0.6468	1	0.6468	1.70	0.2828	
AB	29.59	1	29.59	77.98	0.0031	
A^2	58.28	1	58.28	153.59	0.0011	
B^2	2.75	1	2.75	7.24	0.0744	
Residual	1.14	3	0.3795			
Cor Total	92.59	8				

Second Layer: - Aceclofenac

Variable Level	Actual Coded Value	Levels				
		-α	Low (-1)	Medium (0)	High (+1)	+ α
Conc. of PEG 1000	X1	62.9289	65	70	75	77.0711

Conc. of PEG 4000	X2	22.9289	25	30	35	37.0711

Table No.13: Result of Optimization Batches by Central Composite Design

Run	Batch	Factor 1 X1 Conc. of PEG 1000 (%)	Factor 2 X2 Conc. of PEG 4000 (%)	Dependent Variables	
				Drug Release (%) Y1	DT (min) Y2
1	S1	75	35	125.87	499.43
2	S2	65	25	91.82	470.43
3	S3	70	30	108.7	493.92
4	S4	**75**	**30**	**119.54**	**499.86**
5	S5	65	35	94.02	480.22
6	S6	70	25	94.86	479.95
7	S7	65	30	99.79	483.25
8	S8	70	35	108.53	492.18
9	S9	75	25	99.68	484.61

Regression Equations for the quadratic model

% Drug release of Aceclofenac (Y1) = $+108.62 +9.91X1 +7.01X2 +6.00X1X2 +1.09X1^2 -6.88X2^2$

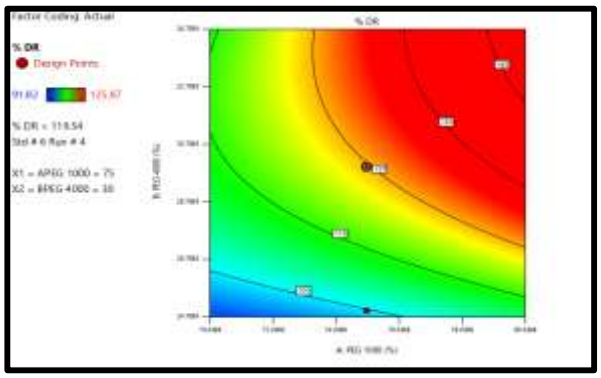
Disintegration Time (Y2) = $+493.93 +8.33X1 +6.14X2 +1.26X1X2 -2.38X1^2 -7.87X2^2$

Regression Equations for % Drug release of Aceclofenac

% Drug release of Aceclofenac (Y1) = $+108.62 +9.91X1 +7.01X2 +6.00X1X2 +1.09X1^2 -6.88X2^2$

The % drug release of aceclofenac at 8 hrs. varied from 91.82 % to 125.87 %. The results of multiple linear regression analysis showed that the coefficients X1 and X2 bear a positive sign. Therefore, increase in the concentration of X1 and X2 is expected to increase the % drug release.

Graphical Representation



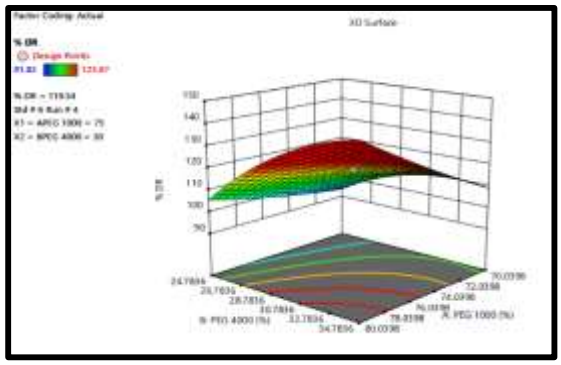


Fig. No. 17 and Fig. No.18: Response Surface Contour Graph and 3-D Surface Graph Showing the Influence of Conc. of PEG 1000 (X1), Conc. of PEG 4000 (X2) on % Drug Release

Table No. 14: Summary of Results of Analysis of Variance

1) ANOVA for Quadratic Model

Response 1: % Drug Release (Y1)

Source	Sum of Squares	DF	Mean Square	F-Value	p-Value	
Model	1125.01	5	225.00	6071.59	< 0.0001	Significant
A-PEG 1000	589.25	1	589.25	15900.57	< 0.0001	
B-PEG 4000	294.84	1	294.84	7956.12	< 0.0001	
AB	143.88	1	143.88	3882.53	< 0.0001	
A^2	2.38	1	2.38	64.12	0.0041	
B^2	94.67	1	94.67	2554.59	< 0.0001	
Residual	0.1112	3	0.0371			
Cor Total	1125.13	8				

Regression Equations for Disintegration Time of Aceclofenac

Disintegration Time (Y2) = $+493.93 +8.33X1 +6.14X2 +1.26X1X2 -2.38X1^2 -7.87X2^2$

Concerning the disintegration time, the results of multiple linear regression analysis showed that the coefficients X1 and X2 bear a positive sign. Therefore, increase in the concentration of X1 and X2 is expected to increase the disintegration time.

Graphical Representation

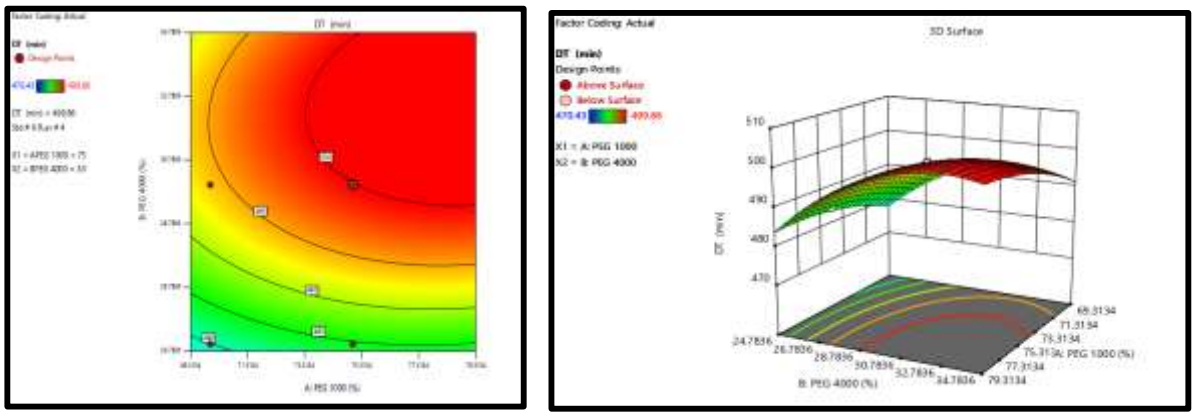


Fig. No.19 and Fig. No.20: Response Surface Contour Graph and 3-D Surface Graph Showing the Influence of Conc. of PEG 1000 (X1), Conc. of PEG 4000 (X2) on Disintegration Time

Table No. 15: Summary of Results of Analysis of Variance

1) ANOVA for Quadratic Model

Response 1: % Disintegration Time (Y2)

Source	Sum of Squares	DF	Mean Square	F-Value	p-Value	
Model	784.53	5	156.91	1.019E+05	< 0.0001	Significant
A-PEG 1000	416.67	1	416.67	2.706E+05	< 0.0001	
B-PEG 4000	226.20	1	226.20	1.469E+05	< 0.0001	
AB	6.33	1	6.33	4107.78	< 0.0001	
A^2	11.36	1	11.36	7377.87	< 0.0001	
B^2	123.98	1	123.98	80515.37	< 0.0001	
Residual	0.0046	3	0.0015			
Cor Total	784.53	8				

5. Conclusion:-

Bi-layer suppositories is improved beneficial technology when compared to single layered suppository. It provides one of the important design approaches where two or more incompatible drugs can be incorporated into a single unit. Bi-layer suppository is suitable for sequential release of two drugs with different indication, in combination and also for sustained release suppositories in which one layer is immediate release as initial dose and second layer is maintenance dose. On the basis of above result the formulation of immediate and sustained release bi-layer suppositories will be used for haemorrhoids and proctologic disease, lidocaine as local anaesthetics and aceclofenac non-steroidal anti-inflammatory agent.

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