International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

Formulation and development of clotrimazole nanoemulsion for topical delivery by (QBD) quality by design approach

Priya Patel*, Chainesh Shah, Mansi Patel, Jigar Vyas, Umesh Upadhyay
Sigma Institute of Pharmacy, Bakrol, Vadodara- 390019.

Submitted: 15-05-2023 Accepted: 30-05-2023

ABSTRACT:

Objective

Main objective of present study was to develop & optimized intranasal delivered Clotrimazole Nanoemulsion. Here, 3^2 full factorial design was employed by selecting independent variables such as Smix concentration (X1), Oil concentration (X2). Optimized average particle size was ranged in 100-400nm.

Experimental Work:

Preformulation studies, FTIR, DSC was carried out for identification of drug and to check interaction between drug & other excipients and to check drug compatibility. Nanoemulsion are prepared and 3^2 full factorial design employeded to study effect of independent variables, i.e., Conc. of Smix (X1) & Oil (X2) on dependent variables % Transmittance (Y1), Viscosity (Y2). Prepared formulation is evaluated for their physical parameters Particle size, Zeta potential, %Transmittance, Viscosity, pH,%loading efficiency, Drug content, in vitro drug release, Ex vivo Drug release. Release kinetic models are used to determine diffusion pattern of drug from Nes. Optimized formulation was subjected for stability studies.

Results & Discussion:

Main identification of drug presented that drug if pure. IR spectra of Clotrimazole exposed that function group of Clotrimazole present in sample shows their stretching in standard range. Thus, present sample was confirmed as Oxytocin with high purity grade. DSC study presented that drug is compatible with excipient. Results of 3^2 full factorial design shown that Smix & Oil significantly affected on dependent variables like % Transmittance (Y1), Viscosity (Y2).

Conclusion

Nanoemulsion of Clotrimazole were successfully formulated using 3^2 full factorial design. The overall result indicated that Concentration of Smix & Oil showed satisfactory % Transmittance, Viscosity, pH , Particle size, zeta potential Loading efficiency, % Drug release, in vitro drug release , Ex vivo drug release. So, Nanoemulsion of Clotrimazole may be good choice to enhance permeability.

Key words: Clotrimazole, Nanotechnology, Nanoemulsion, Design of Experiment.

I. INTRODUCTION

Nanotechnology

Nanotechnology has transformed into a trendy expression in drug sciences and endeavors are continuous to expand its applications in different surges of drug sciences. Nanotechnology has considerably inclined drug delivery investigate over last two periods and some nanoscale technologies have been and are existence discovered for getting better healing performance of drugs. Several ways via which nanoscale technologies can develop beneficial effectiveness of drugs are: Improving solubility of hydrophobic drugs; Improving permeability or transport of weakly permeable drugs class III and IV drugs as per Biopharmaceutical Classification System [BCS].

Nanoscale advances can be to a great extent arranged into: lipid-based nanocarriers, polymeric nanocarriers, inorganic nanocarriers, and medication nanoparticles or nanosuspensions.

Nanoemulsion Drug Delivery

An ideal medication conveyance framework satisfies the motivation behind expanding helpful impact as limiting harmfulness. With improvement in time and advances in science and innovation, dose structures incorporate developed from straightforward combinations and pills, to undeniably challenging frameworks, which are known as clever medication conveyance frameworks. One of the instances of original medication conveyance framework is Nanoemulsion. Nanoemulsion is defined as thermodynamically constant, isotopically clear dispersion of two immiscible liquids such as oil and water, stabilized in an interfacial layer of surfactant molecule. Nanoemulsion is emulsion by uniform and very minor droplet size in variety of

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

20-200 nm. Nanoemulsion do not form rapidly; to break bigger droplets into lesser ones, outer shear must be applied. As we compared to micro emulsion phases, rather little is known regarding creating and controlling Nanoemulsion. First Nanoemulsion is make in year 1940s, it very well may be of 3 kinds i.e., oil-in-water (O/W), water-in-oil (W/O), and bi-continuous. Nanoemulsion are similarly termed as mini emulsions, submicron emulsions and ultrafine emulsions. Study works verifies that as match up to additional transdermal drug delivery system Nanoemulsion is distant more able drug delivery system. Major variation connecting emulsion and Nanoemulsion is that the level though emulsion is kinetically stable but thermodynamically unbalanced, emulsion is cloudy and Nanoemulsion is very clear in physical look.

Materials & Methodology

Materials

Clotrimazole was a gift sample from IPCA laboratories, Maharashtra, India. Oleic acid, tween 80 were gifted by (Suvidh Nathlaboratories, Vadodara, India) and Propylene glycol were obtained from (Balaji pharmaceutical, SuratIndia). All other ingredients and reagents were of analytical grade.

Methodology

Method of Preparation Clotrimazole Nanoemulgel

HPMC was carefully weighed and dissolved in 100 mL of water for 2 hours of soaking with 600 RPM agitation, followed by the addition of a penetration enhancer to the formed gel to avoid drying. Slow agitation and continuous stirring will be used to introduce the triethanolamine. In the gel, Clotrimazole Loaded Nanoemulsion will be added.

Physical evaluation

It will be carried out to assess the gel's organoleptic properties, occlusiveness, and washability.

Measurement of pH

A digital pH meter will be used to assess the pH of the prepared gel.

Viscosity study

The produced gel will be stored in a 50 mL appropriate beaker, and the spindle Groove will be dipped in a Brookfield Viscometer at a set RPM. This will be repeated three times, with the mean computed from the collected observations.

Spreadability study

1 g of gel will be precisely weighed and put between two slides, where it remained for around 5 minutes. When no more spreading will be seen, the diameters of speed circles will be measured in cm and used as comparison values for Spreadability.

Homogeneity and grittiness

By squeezing the produced gel between the thumb and index finger, the consistency will be assessed. A little amount of gel will be applied to the back of the hand to check for homogeneity and grittiness.

Drug content

In a volumetric flask, 1 gm of each gel formulation will be dissolved in 20 mL of alcohol after 30 minutes of stirring. It will be then diluted and filtered. A further dilution to 10 mL alcohol will be prepared, and 1 mL will be removed from the above and diluted to 10 mL alcohol once more. In ultraviolet light, the absorbance will be measured at 296 nm.

Kinetics of Drug Release

In order to investigate the mechanism of drug release from Nanoemulgel of different ratios, the release data obtained from dissolution studies will be fitted to various kinetic equations.

The kinetics models used will be a

Zero order equation, ($\mathbf{Q_t = Q_0 - K_{0t}}$)

First order equation, ($\mathbf{\ln Q_t = \ln Q_0 - K_t}$)

Higuchi's equation ($\mathbf{Q_t = K_h\, t^{1/2}}$)

The following plots will be made,

Q_t Vs. t (Zero order kinetic model),

($Q_0 - Q_t$) Vs. t (First-order kinetic model)

And **Q_t vs. $t^{1/2}$**(Higuchi Model),

Where,

Q_t is the percent of drug released at time t,

Q_0 is the initial amount of drug present in the microspheres and

K_0, K and K_h are the constant of the equations of Zero order, First order and Higuchi respectively.

Accelerated stability studies of Nanoemulgel

Stability tests revealed that the quality of a medicine or dosage form can be influenced by changes in temperature, humidity, and light over time. It will be carried out for one month at room temperature for the chosen formulation. On the 0^{th}, 15^{th}, and 30^{th} days, samples will be taken and examined for physical appearance and drug content.

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

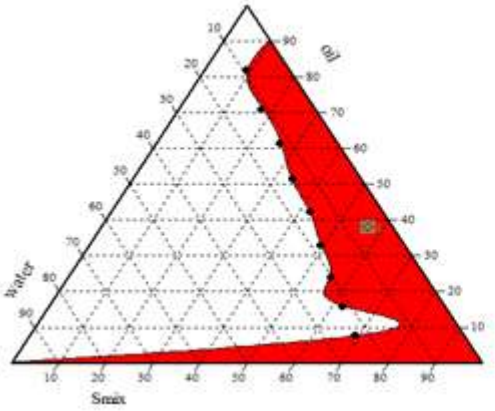
II. RESULTS & DISCUSSION

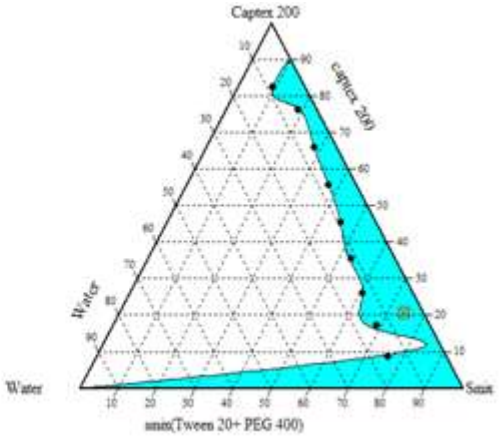
Selection of Surfactant and Co-surfactant:

Emulsificationcapabilityofsurfactantandcosurfactant

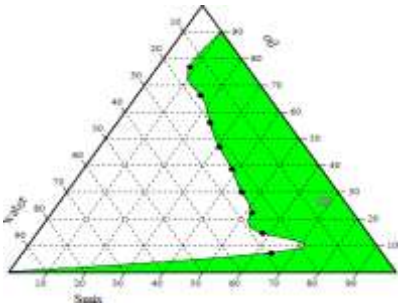
Surfactant	Maximumnumber ofInversion	% Transmittance	Cosurfactant	Maxim um Number ofinversion	% Transmittance
Labrasol	16	96.00	**Propylene Glycol**	5	98.2
Tween80	5	99.02	**Polyethylene Glycol400**	15	94.5
Tween20	18	89.3			

CONSTRUCTION OF PSEUDOTERNARY PHASE DIAGRAM:



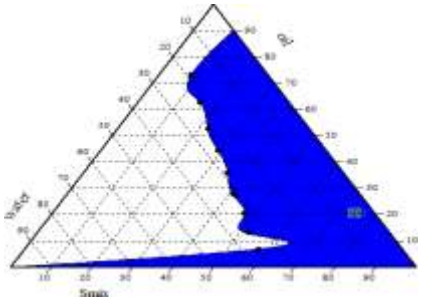


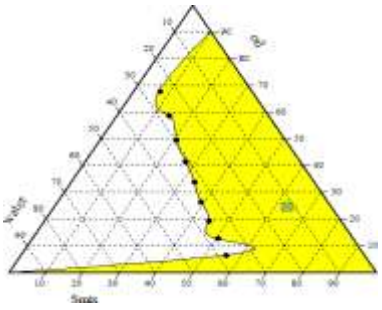
(A) **(B)**



(C)

(D) **(E)**





International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

Construction of Pseudo ternary phase diagram

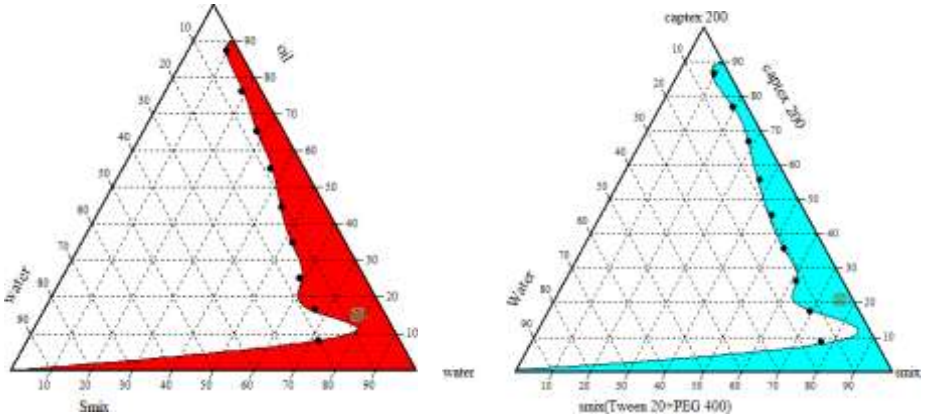
Composition for the screening of suitable ratio of Oleic acid, tween 80and Propylene glycol

SmixRatio	Oil (%w/w)	Smix (%w/w)	Water (%w/w)	%Transmittance
1:1	7.69	69.23	23.08	96
	15.62	62.50	21.88	95.1
	24	56	20	93.2
	32.79	49.83	17.38	91.7
	42.37	42.37	15.26	90.1
	51.28	34.19	14.53	77.4
	61.40	26.32	12.28	65.1
	70.80	17.70	11.5	53.2
	81.82	9.09	12.09	45.7
1:2	8.47	76.27	15.26	91.4
	17.24	68.97	13.79	87.6
	26.09	60.87	13.04	83.2
	30.08	60.15	9.77	50.0
	45.45	45.45	9.1	74.5
	55.56	37.04	7.4	65.7
	66.04	28.30	5.66	52.5
	76.14	19.05	4.81	48.9
	86.54	9.62	3.84	35.7
2:1	7.14	64.29	28.57	96.7
	14.60	58.39	27.01	95.8
	22.22	51.85	25.93	94.9
	30.08	45.11	24.81	93.5
	38.46	38.46	23.08	91.4
	46.88	31.25	21.87	89.7
	56	24	20	73.7
	6.12	10.53	17.35	65.7

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

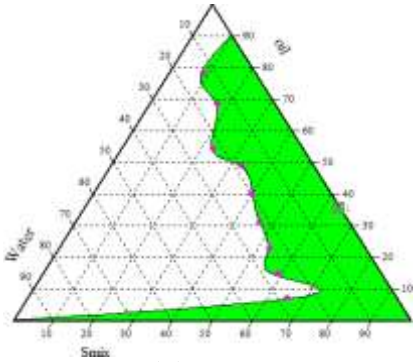
	76.92	8.55	14.53	58.5
3:1	6.45	58.06	35.49	98.4
	13.07	52.29	34.64	98.2
	20.41	49.62	31.97	95.8
	27.59	41.38	31.03	92.5
	35.71	35.71	28.58	92.1
	44.12	29.41	26.47	90.5
	52.63	22.56	24.81	77.7
	62.50	15.62	21.88	70.9
	73.17	8.13	18.7	60.5
4:1	6.25	56.25	37.5	99.5
	12.66	60.63	36.71	99.1
	19.93	64.87	35.9	98.7
	26.14	39.22	34.64	95.2
	33.78	33.78	32.44	94.9
	41.30	28.13	30.51	91.7
	49.30	21.13	29.57	81.1
	58.39	14.60	27.01	79.2
	67.67	7.52	24.81	72.2

CONSTRUCTION OF PSEUDOTERNARY PHASE DIAGRAM OF CLOTRIMAZOLE

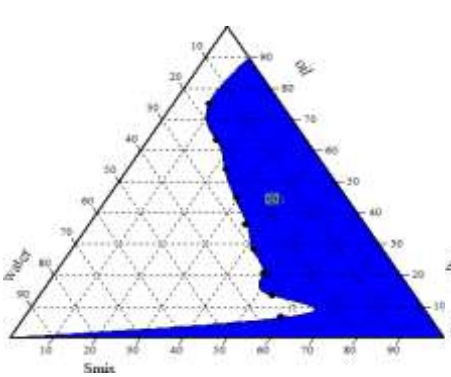


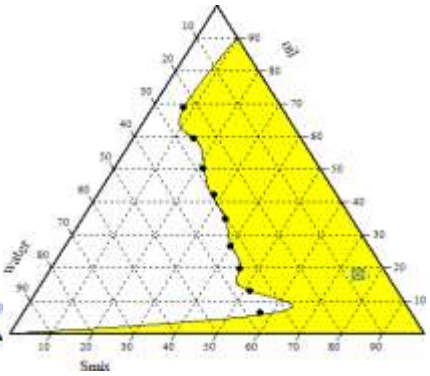
(A) **(B)**

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781



(C)





(D) (E)

Construction of Pseudo ternary phase diagram

1:1 **2:1**

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

3:1 4:1

Construction of Pseudo ternary phase diagram of Clotrimazole
Composition for the screening of suitable ratio of Oleic acid, tween 80 and Propylene glycolbypseudo ternaryphasediagramof Clotrimazole

Smixratio	%Oil (w/w)	% Smix(w/w)	Water (w/w)	% Transmittance
1:1	8	72	20	93.1
	16.67	66.67	16.66	92.6
	25.21	58.82	15.97	90.5
	34.78	52.17	13.05	87.4
	44.64	44.64	10.72	80.7
	55.05	36.70	8.25	72.9
	65.42	28.04	6.54	61.7
	76.19	19.05	4.76	52.9
	87.38	9.71	2.91	43.5
1:2	8.56	76.92	14.52	88.67
	17.39	69.57	13.04	83.4
	26.32	61.40	12.28	80.7
	35.71	53.57	10.72	77.5
	45.45	45.45	9.1	72.1
	55.56	37.04	7.4	63.9

	66.67	28.57	4.76	50.5
	76.92	19.23	3.85	45.7
	86.54	9.62	3.84	34.3
2:1	7.25	65.22	27.53	95.5
	14.81	59.26	25.93	93.7
	22.73	53.03	24.24	91.2
	31.01	46.51	22.48	90.4
	40.40	40.40	19.2	88.5
	49.18	32.79	18.03	87.4
	58.82	25.21	15.97	71.6
	68.38	17.09	14.53	64.7
	78.26	8.70	13.04	56.5
3:1	6.58	59.21	34.21	97.5
	13.42	53.69	32.89	97.9
	20.69	48.28	31.03	94.2
	28.17	42.25	29.58	91.0
	36.23	36.23	27.54	89.7
	44.78	29.85	25.37	88.0
	53.84	23.08	23.07	75.4
	63.49	15.87	20.64	66.9
	75.0	8.33	16.67	58.6
4:1	6.37	57.32	36.31	98.5
	12.90	51.61	35.49	98.0
	19.61	45.75	34.64	95.5
	26.67	40.00	33.33	94.7
	34.72	34.72	30.56	91.2
	42.25	28.17	29.58	89.7

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

	50.36	21.58	28.06	77.2
	59.26	14.81	25.93	63.1
	68.70	7.63	23.67	60.0

Preliminary Trial Batches Based on Pseudo ternary Phase Diagram:
preliminary trialbatchesbasedonpseudo ternarydiagram

Batch	**Composition ofNanoemulsion (%)**			**FormulaofNanoemulsion**		
	Oil (%)	**Smix (%)**	**Water (%)**	**Oil (w/w)**	**Smix (w/w)**	**Water (w/w)**
CLR1	6.37	57.32	36.31	1.274	7.242	11.48
CLR2	12.90	51.61	35.49	2.58	7.098	10.32
CLR3	19.61	45.15	34.64	3.922	9.03	7.04

Effect of composition of Nanoemulsion on %Transmittance, Viscosity,%DrugRelease(3hrs)

Batch	**%Transmittance**	**Viscosity**	**%DrugRelease (3hrs)**			
			Time (3hrs)			
			0	**1**	**2**	**3**
CLR1	97.7	132	0	10.99	14.37	29.40
CLR2	97.0	137	0	6.45	12.30	18.80
CLR3	96.0	142	0	4.30	11.85	16.95

Formulation and Development of Clotrimazole loaded Nanoemulsion using Design of Experiment [DoE] Approach:
Various batches ofClotrimazole Nanoemulsion prepared by DoEapproach werepreparedaccordingto3^2factorial designswhich areas follow:

Factorial Design

IndependentVariablesofFormulation			
Independent Variables	Low(-)	Medium(0)	High(+)
Oilconcentration(X1)	5%	10%	15%

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

Smixconcentration(X2)	50%	55%	60%
DependentVariables			
Y1=%Transmittance			
Y2=Viscosity			
Y3=%Drugrelease			

Compositions of Factorial Batches in Coded Form
VariousbatchesofClotrimazoleNanoemulsionwith4:1Smixratiowere preparedaccordingto 3^2factorialdesigns whichareas follow:

Compositions of Factorial Batchesin Coded Form

ClotrimazoleNanoemulsion3^2=9Batches		
BatchNo	**Variablelevelincodedform**	
	Oil concentration(X1)	**Smix Concentration (X2)**
F1	-1	-1
F2	-1	0
F3	-1	+1
F4	0	-1
F5	0	0
F6	0	+1
F7	+1	-1
F8	+1	0
F9	+1	+1

Composition of factorial batches in actual form:
Composition of factorialbatchesinactualform

BatchNo.	**ClotrimazoleNanoemulsion$3^{2=}$9Batches**
	ActualValue

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

	Oil Concentration(%) (X1)	Smix Concentration (%)(X2)	Amount ofOil(ml) (X1)	Amountof Smix(ml)(X2)
F1	5	50	1	10
F2	5	55	1	11
F3	5	60	1	12
F4	10	50	2	10
F5	10	55	2	11
F6	10	60	2	12
F7	15	50	3	10
F8	15	55	3	11
F9	15	60	3	12

Characterization of Batches F1-F9

Characterizationof BatchesF1-F9

BatchNo	% Transmittance(Y1)	Viscosity(Y2)	%Drug release(Y3)
F1	98.5	137	21.07
F2	98.6	134	24.08
F3	99.1	132	26.01
F4	98.1	143	15.04
F5	98.3	141	17.97
F6	99.0	138	20.6
F7	87.3	155	8.6
F8	93.3	151	10.98
F9	94.1	147	13.3

Effect on % Transmittance (Y1)- Surface Response Study:

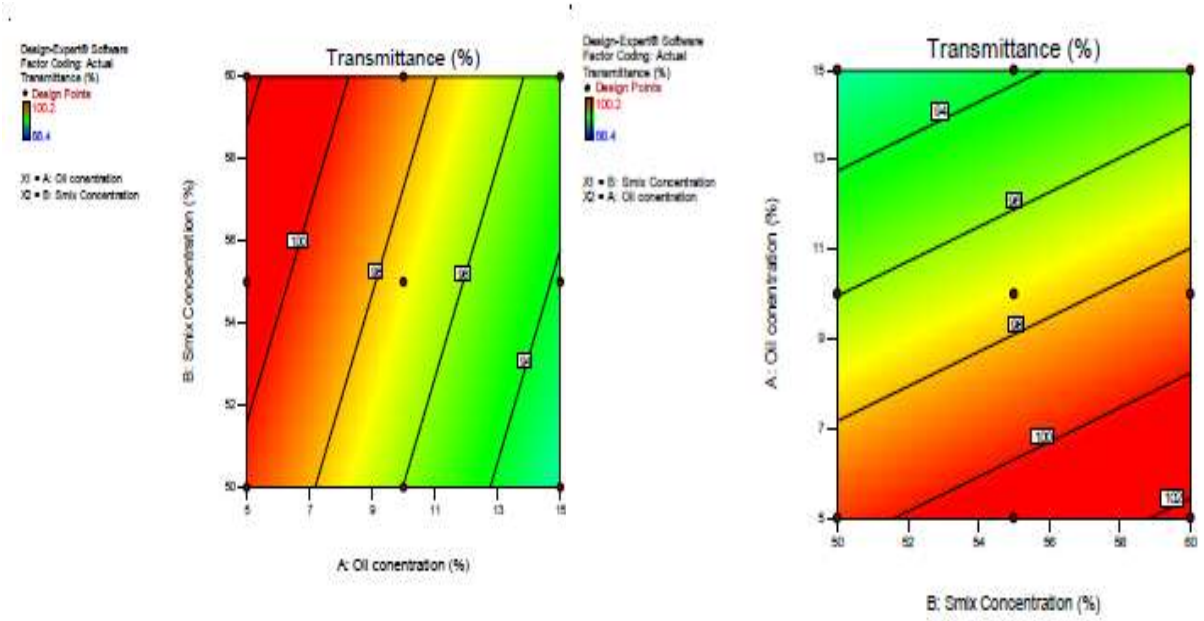
Negative value of coefficient of X1indicatesdecreasein response of Y1 i.e., %Transmittance. PositivevalueofcoefficientX2,S_{mix}Concentrationindicatesincreasein % Transmittance. It indicates linearity of surface response and contour plot as shown infigure.Fullmodelwasfoundsignificantanddetailed ANOVA,ResponseSurfaceCounterPlot and 3D plots areas follows:

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

%Transmittance=96.25-2.47*X1+1.27*X2

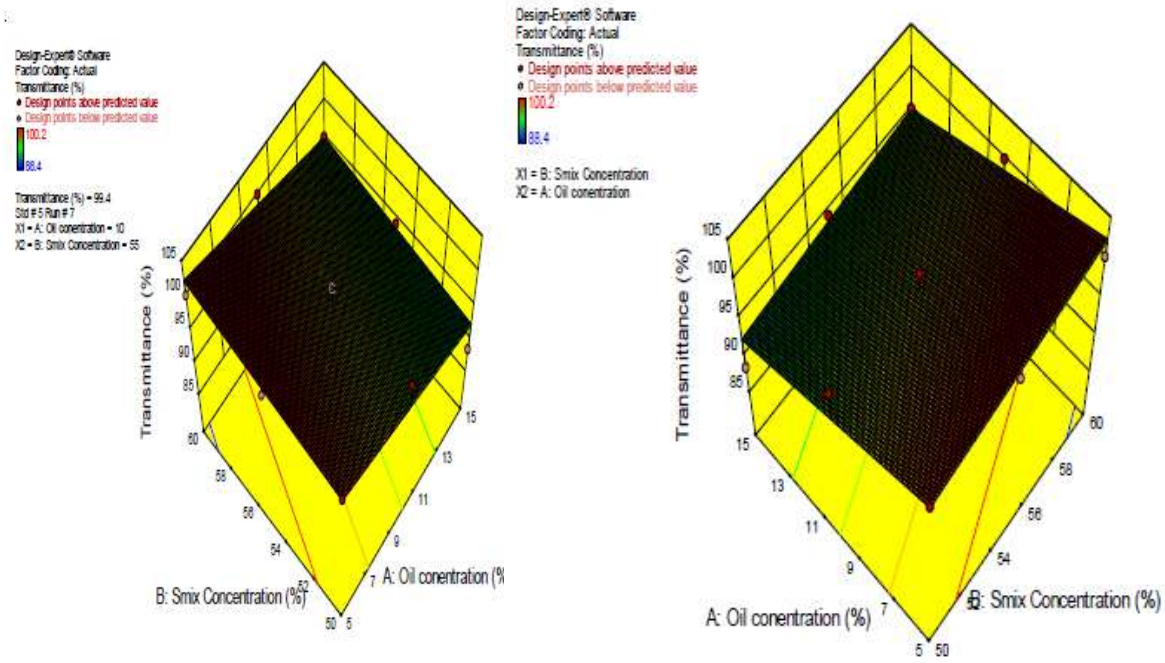
ANOVATable forResponseY1

Analysisof variancetable[Partialsumof squares-TypeIII]						
	Sumof		**Mean**	**F**	**p-value**	
Source	**Squares**	**df**	**Square**	**Value**	**Prob>F**	
Model	87.41	2	43.15	5.73	0.0272	significant
A-Oilconcentration	76.93	1	76.93	10.80	0.0125	
B-SmixConcentration	10.37	1	10.37	1.67	0.2300	
Residual	37.79	6	5.36			
CorTotal	126.21	8				



ResponseSurfacePlot

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781



3D SurfacePlot

5.6.2 Effect on Viscosity(Y2)- Surface Response Study:

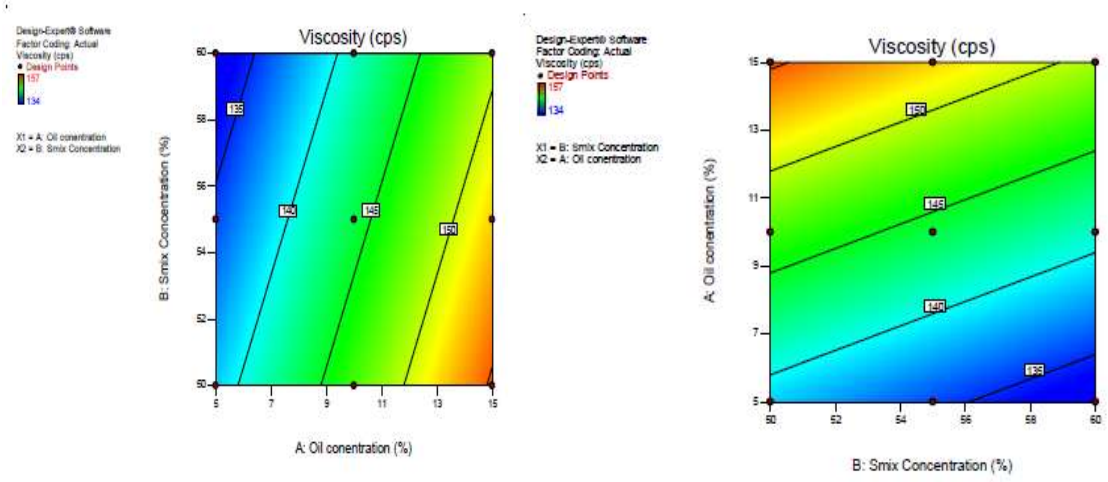
Positivevalue of coefficientof X1 indicatesincrease inresponse ofY2 i.e. %Viscosity. Negative value of coefficient X2, S_{mix} Concentrationindicates decrease inViscosity.Itindicates linearityof surfaceresponseandcontourplotasshowninfigure.Ful l model was found significantand detailed ANOVA, ResponseSurfaceCounter Plot and 3D plots areas follows:

Viscosity=133.0+7.22*X1-2.00*X2

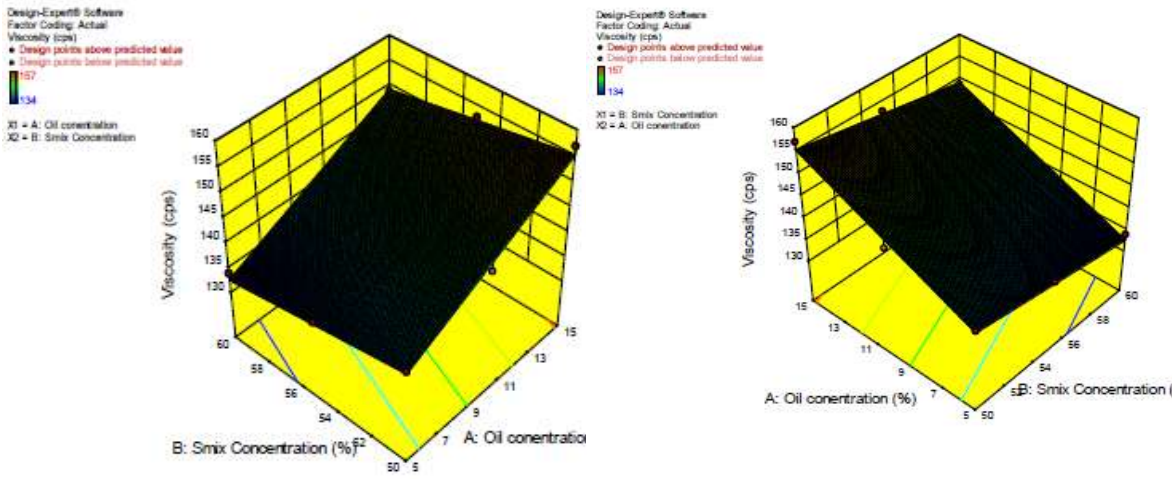
ANOVATable forResponseY2

ANOVATableforResponseY2 **Analysisof variancetable[Partial sumof squares-TypeIII]**						
	Sumof		**Mean**	**F**	**p-value**	
Source	**Squares**	**df**	**Square**	**Value**	**Prob>F**	
Model	469.56	2	224.22	113.48	<0.0001	significant
A-Oilconentration	415.56	1	405.56	219.48	<0.0001	
B-SmixConcentration	53.00	1	53.00	27.48	0.0017	
Residual	10.22	6	1.78			
CorTotal	471.00	8				

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781



Response Surfaceplot



3D SurfacePlot

Effect on %Drug Release (Y3) - Surface Response Study:

NegativevalueofcoefficientofX1indicatesdecreaseinresponseofY2i.e.% %DrugRelease. Positive valueofcoefficientX2,S_{mix}Concentrationindicatesincreasein%DrugRelease.Itindicates linearityof surfaceresponseandcontourplotas shown in figure.Full model was found significantand detailedANOVA,ResponseSurfaceCounterPlot and 3D plots areas follows.

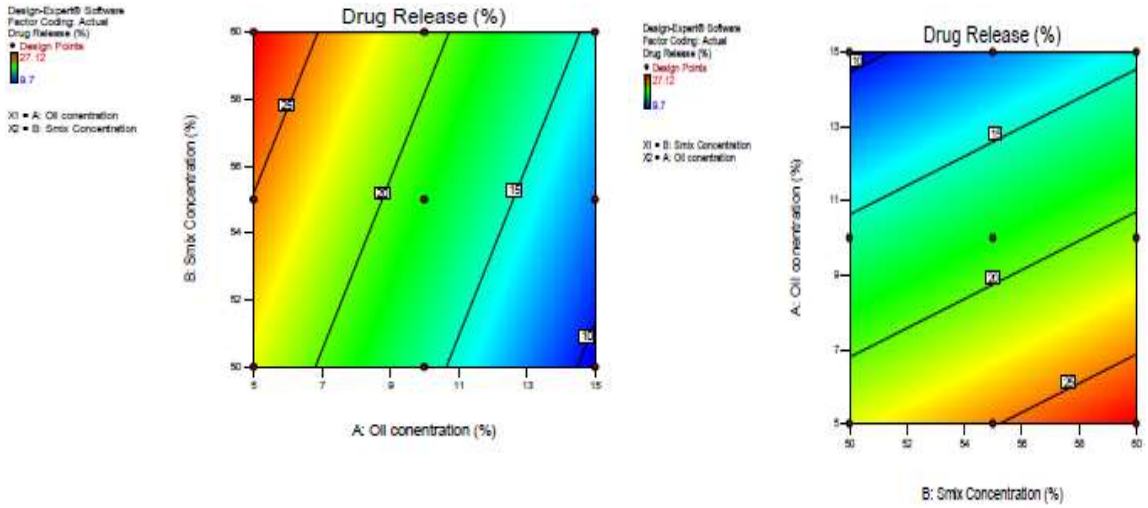
DrugRelease=17.27-5.40*X1+1.44*X2

ANOVATable forResponseY3

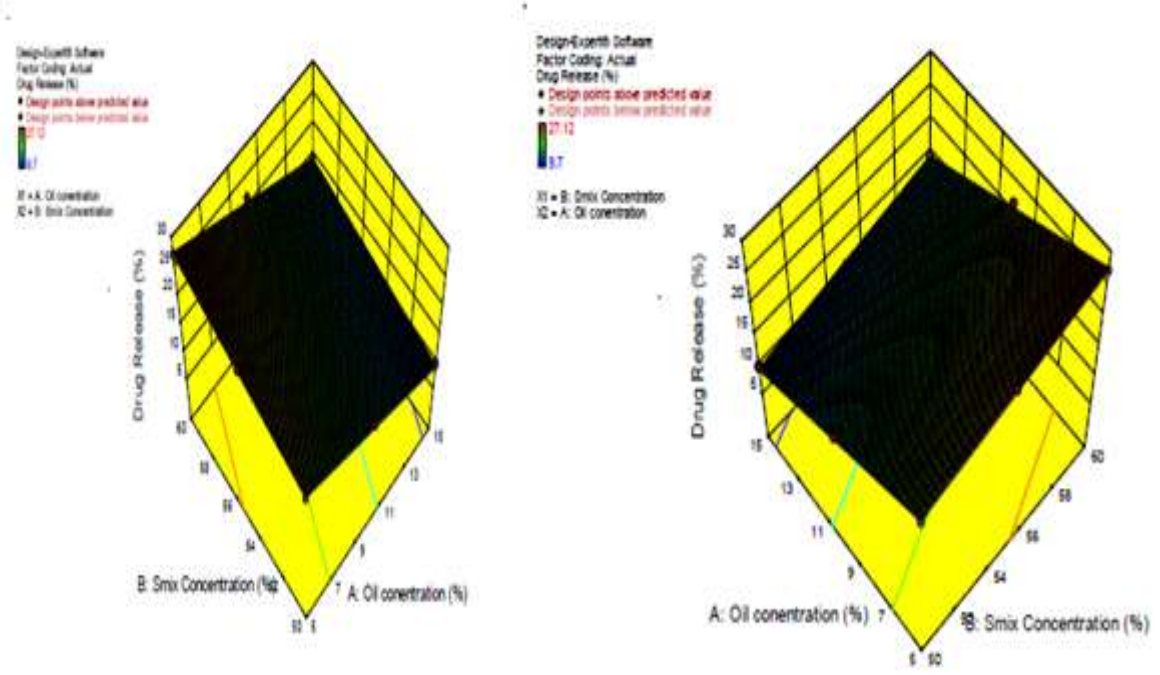
ANOVAforResponseSurfaceLinearmodel						
Analysisof variancetable[Partial sumof squares-TypeIII]						
	Sumof		**Mean**	**F**	**p-value**	
Source	**Squares**	**df**	**Square**	**Value**	**Prob>F**	
Model	282.50	2	135.79	500.61	<0.0001	significant
A-Oilconcentration	243.43	1	243.43	857.08	<0.0001	

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

B-SmixConcentration	38.06	1	38.06	122.14	<0.0001	
Residual	1.65	6	0.29			
CorTotal	284.26	8				



ResponseSurfacePlot



3D SurfacePlot

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

Check point analysis of Validation Batches:
F10&F11formulationwasmadeforcheckpointanalysisandpredictedandexperimentalvalues werecompared.

Validationof BatchesF10& F11:PredictedResponse

Batch No	OilConcentration (X1)	SmixConcentration (X2)	% Transmittance(Y1)	Viscosity (Y2)	% Drugrelease (Y3)
F10	5.83	53.83	100.01	137.75	23.2
F11	6.98	53.55	99.11	139.83	21.6

ValidationBatchesF10&F11:ActualResponse

BatchNo	OilConcentration (X1)	SmixConcentration (X2)	% Transmittance(Y1)	Viscosity (Y2)	% Drugrelease (Y3)
F10	5.83	53.83	99.8	135.4	20.1
F11	6.98	53.55	96.5	142.9	27.6

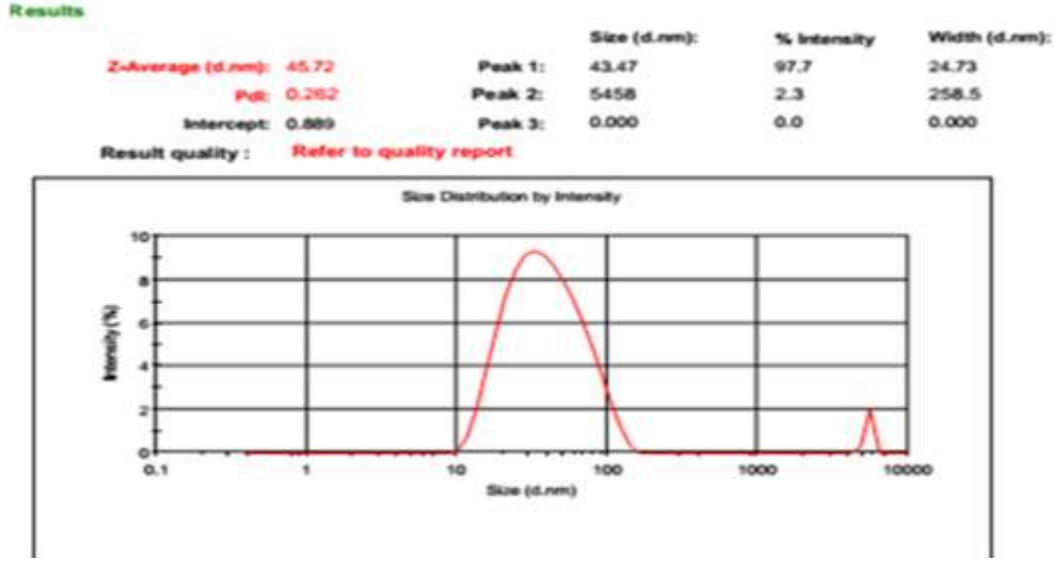
Selection of Optimized Formulation:
F10wasselectedasvalidatedoptimizedbatchandfurtherconsidered forformulationinto Gel which was having %Transparency 99.8, Viscosity 135.4, % Drug Release20.6.

OptimizedNanoemulsionformulationformula

Composition	Concentration (%)	Actual value ofNanoemulsion in20ml
Oil	5.83	1.166
Smix	53.83	10.766
Water	40.34	8.068

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

Droplet Size Analysis of optimized batch:



Dropletsizeanalysisof Optimized Batch
Dilution test of Nanoemulsion Formulation.
DilutiontestofNanoemulsion

Dilution	Observation
10	Notfound phaseseparation
50	Notfound phaseseparation
100	Notfound phaseseparation

The prepared Nanoemulsion formulation was diluted in 1:10,1:50,1:100 ratiowith distill water the system doesn't show any sign of separation and found to beclear.soits confirm thatprepared nanoemulsion is o/wtype.

Measurements of Zeta Potential:

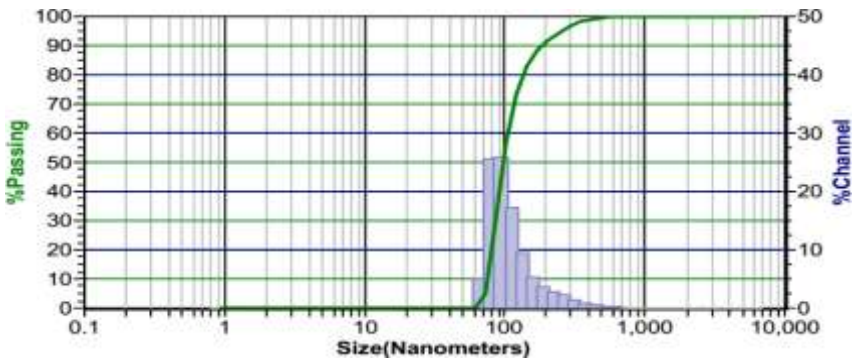
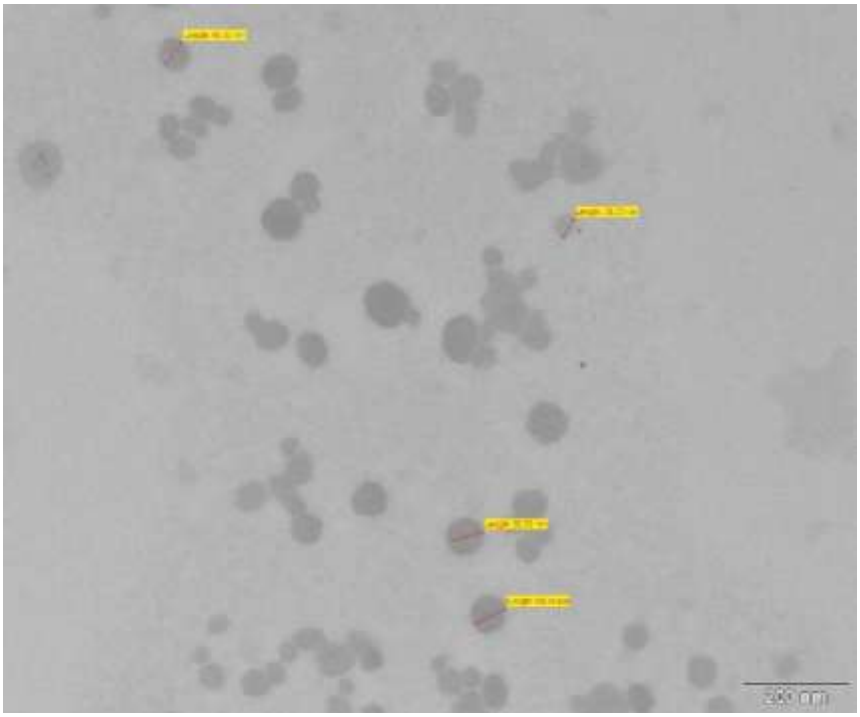


Figure 5. 1Zetapotentialof OptimizedBatch

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

Transmission Electron Microscopy:

TEMimageofValidatedoptimizedBatchNanoemulsion



Characterization of Nanoemulsion:
CharacterizationofNanoemulsion

Batch	(Mean±S.D) (n=3)				
	PolydispersityIndex	RefractiveIndex	pH	DrugContent	Conductivity
F10	0.251	1.22±0.004	6.3±0.01	94.6±0.162	62.4±0.24

Batch	(Mean±S.D) (n=3)	
	%Transmittance	Viscosity
F10	99.8±0.1	135.4±1.2

Thermodynamic Stability
ThermodynamicStabilitystudy

Batch	Heating coolingcycle	Centrifugation	FreezThaw Cycle
F10	Yes	Yes	Yes

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

Nanoemulsion are thermodynamically stable systems and are formed at particular concentration of oil, Surfactant, and water, making them stable and not subject to phase separation, creaming, or cracking.it is thermostability that differentiates nano or microemulsion from emulsion that have kinetic stability and eventually phase separate. The result in table revealed that the nanoemulsion formulations passed thermodynamic stability tests.

In-Vitro Drug Release study:
In-VitroDrugReleasestudy

Time (hrs.)	**% DrugRelease (Mean ±S.D.) (n =3)**
0	0
1	10.69±1.32
2	21.82±1.65
3	32.86±1.09
4	43.07±1.56
5	49.11±1.46
6	54.82±1.96
7	62.3±1.76
8	68.21±1.87
9	74.42±1.76
10	81.24±1.54
11	86.64±1.76
12	93.45±1.24

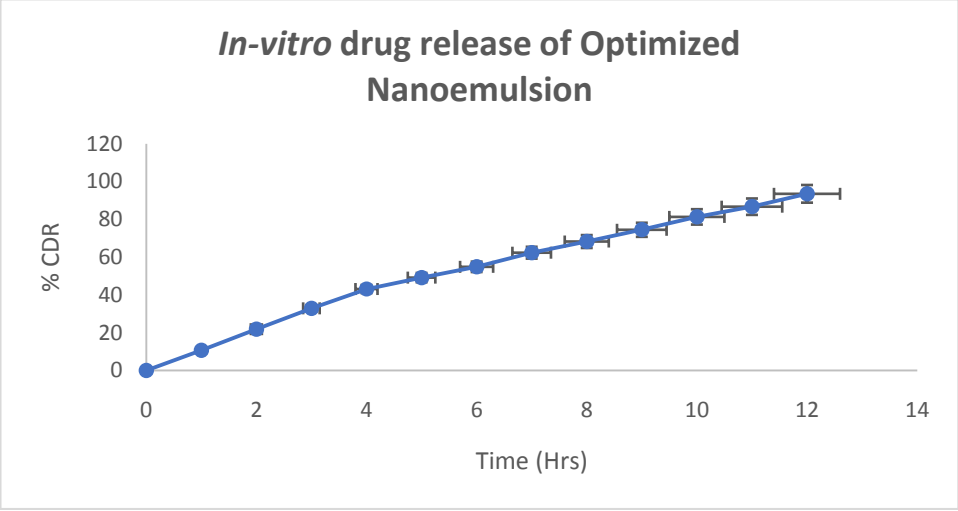


Figure 5. 2In-vitrodrugreleaseof Optimized Nanoemulsion

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

Dose Calculation for Loading Drug containing Nanoemulsion into topical Gel

Dosecalculationfor loadingCTRNE intotopicalgel
100 gm marketed CTR gel formulation contain 2.5 % w/w CTR as a drug

So, in20 gm of Nanoemulsion carbopol gel contains (?)
In 100 gm formulation =2500 mg (2.5 gm) ClotrimazoleSo,20 gm formulation=?
2500*20/100=500mgClotrimazoleisrequiresin20ml Nanoemulsion gel

Preliminary Trial batches

FormulationDesignofTopicalGeltrialBatches

Ingredient	**CTRG 1**	**CTRG 2**	**CTRG 3**	**CTRG 4**
HPMC (%w/v)	**1**	1.5	2	2.5
Propyleneglycol (mL)	**5**	5	5	5
Methylparaben	**0.1**	0.1	0.1	0.1
Propylparaben	**0.05**	0.05	0.05	0.05
Triethanolamine (mL)	**0.25**	0.25	0.25	0.25
Water (mL)	**100**	100	100	100

ResultofEvaluationof HPMCgel

Batch code	**Colour**	**Odour**	**pH (Mean ±S.D.)(n=3)**	**ViscositySpindleno:61 (Mean ±S.D.)(n=3)**	**Spreadability(gm.cm/sec) (Mean ± S.D.)(n=3)**
CTRG 1	**Colorless**	**Odourless**	**6.3±0.01**	**9222±46**	**12.34±0.89**
CTRG2	Colorless	Odourless	6.1±0.07	9527±50	10.17±1.23
CTRG3	Colorless	Odourless	6.2±0.02	12508±56	9.2±1.35
CTRG4	Colorless	Odourless	6.3±0.03	14439±29	11.24±1.76

CTRG1*Formulationwastakenasoptimizedformulation

The **CTRG**1 shows good Spreadability and viscosity. Therefore, it was taken asoptimized formula for further formulation of promising alternative Nanoemulsionloadedgel.

Formula for Nanoemulsion gel

FormulaforNanoemulsionGel(20gm)

Sr.no	**Ingredients**	**Quantity**
1	Oil	0.583ml
2	Smix	5.383ml
3	Water	4.03ml

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

4	Drug (Clotrimazole)	500 mg

Characterization

Result of CTRTopicalGel

Parameter	Puredrug Gel	Marketed ClotrimazoleGel	OptimizedCTR NanoemulsionGel
Dose	500 mg	2.5%	500mg
Strength	20gm	20gm	20 gm
Clarity	Transparent	Transparent	Transparent
Odour	Odourless	Odourless	Odourless
pH	6.1±0.69	6.7±0.01	6.3±0.01
Spreadability	9.7±0.80	10.30±1.02	12.34±0.89
Viscosity	9580±0.80	9784±34	9222±46
% Drug Content	88.23±0.89	96±1.47	93±0.92

in-vitro release study:

In-vitroreleasestudy

Time(hr)	%DrugReleasestudy		
	% CDR ofOptimized CTRNanoemulsion(Mean ± S.D.)(n=3)	% CDR ofOptimized CTRNanoemulsionGel (Mean±S.D.) (n=3)	%CDR ofPuredruggel (Mean ± S.D.)(n=3)
0	0	0	0
1	7.74±1.54	12.15±1.45	13.48±1.19
2	13.10±1.74	22.30±1.01	24.92±1.43
3	19.23±1.34	33.43±1.07	35.97±1.87
4	31.96±1.02	41.99±1.13	46.18±1.34
5	41.68±1.87	48.82±1.54	52.22±1.24
6	50.81±1.34	58.43±1.43	65.93±1.74
7	56.56±1.74	66.32±1.85	73.49±1.54
8	62.77±1.84	72.90±1.34	79.32±1.67
9	70.55±1.51	77.31±1.63	84.92±1.54

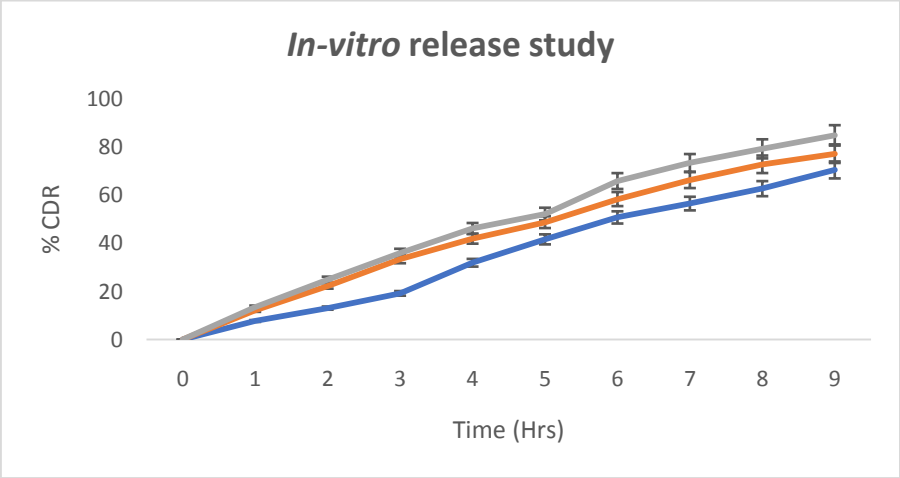


Figure 5. 3comparisonofin-vitrodrugreleasestudy

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

J-flux & Permeability Co-efficient

J-flux&PermeabilityCo-efficient

Time(hrs.)	FluxJ ($mg/cm^2/hr$)	Permeability co-efficient (Kp)
0	0.0000	0
1	0.1626	0.000820
2	0.0767	0.000390
3	0.2416	0.001225
4	0.1869	0.000946
5	0.4315	0.002174
6	0.5946	0.00208
7	0.4102	0.00216
8	0.7010	0.00362
9	0.1522	0.0008

Release Kinetic

Release Kinetic of Clotrimazole Nanoemulsion &Nanoemulsiongel

Model	Parameter	OptimizedNanoemulsion	OptimizedNanoemulsion gel
ZeroOrder	**R^2**	0.9853	0.9948
	Slop	10.424	9.4064
	Intercept	1.6343	1.6989
FirstOrder	**R^2**	0.9822	0.9945
	Slop	-0.0711	-0.065
	Intercept	2.0172	2.0096
HiguchiModel	**R^2**	0.9953	0.9981
	Slop	3.739	3.739
	Intercept	10.041	10.041
HixonCrowell	**R^2**	0.9819	0.9948
	Slop	0.2163	0.1886
	Intercept	0.0242	0.0091

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 1727-1752 www.ijprajournal.com ISSN: 2249-7781

CorsmeyerPeppas	**R^2**	0.9549	0.9642
	Slop	69.262	63.181
	Intercept	4.6352	4.29

By plotting the values for Higuchi model, near straight lines with parallel positiveslopes were obtained indicating that, thebest fit model for the formulations wasHiguchimodel.

Stability Analysis.
StabilityAnalysisof CTRG 10 atRoomTemperaturefor1Months

PARAMETER	**OptimizedClotrimazole (CTRG 10)loadedGel**			
	RoomTemperature			
	0Day	10 Day	20 Day	30 Day
Clarity	Transparent	Transparent	Transparent	Transparent
Odour	Odourless	Odourless	Odourless	Odourless
pH	6.2	6.3	6.2	6.3
Spreadability	12.34±0.08	12.49±0.06	12.34±0.01	12.92±0.05
Viscosity	9222±25	9221±39	9225±29	9222±04
%Drugcontent	93±0.92	94±0.80	93±0.50	93±0.27

III. CONCLUSION:

Clotrimazole Nanoemulsion topical drug delivery system to avoidance of related side effects and these DDS is directly and targetability to affected area of the skin. The major objective behind this formulation is delivery of hydrophobic drugs via skin. Nanotechnology with the use of Nano sized particle have large surface area may be succeeded in overcoming skin barrier and hence Nano sized emulsion can easily penetrate the pores of the skin and reach the systemic circulation thus getting channelized for effective delivery which can be maximize efficacy, reduce dose and dose frequency & hence increase patient Compliance.

REFERENCES:

1. Kanojia N, "Explicating the Applications of Quality by Design Tools in Optimization of Microparticles and Nanotechnology Based Drug Delivery Systems." Biointerface research in applied chemistry. **2021**, 12(4), 4317 – 4336.
2. Kalkate S, "QbD in pharmaceutical manufacturing: a systematic review of current status, challenges and future perspectives." World journal of pharmaceutical research. **2021,** 10(10), 358-371.
3. S. Cunha, "Using the quality by design (QbD) approach to optimize formulations of lipid nanoparticles and nanoemulsions: A review." Nanomedicine: Nanotechnology, Biology, and Medicine. **2020**, 27, 102-119.
4. Pallagi E, "Adaptation of the quality by design concept in early pharmaceutical development of an intranasal nanosized formulation." Int. J. of Pharm.**2015**, 491, 384–392.
5. "ICH guideline Q9 on quality risk management" **2006,** ICH Q8/Q9/Q10 Training material, ICH Q8/Q9/Q10 Points to consider
6. Gandhi A, "Quality by Design (QbD) in Pharmaceutical Industry: Tools, Perspectives and Challenges." Pharma Tutor. **2016**, 4(11) 12-20.
7. Nadpara A, "Quality by design (QbD): a complete review." **2012,** Int. J. Pharm. Sci. Rev. Res.**2012,** 17(2), 20-28.
8. Abhinandana A and Nadendla R, "Application of Quality by Design and its Parameters for Pharmaceutical Products."

Int. J. of Pharma and Chemical Research. **2017**, 3(3), 505-515.
9. KumarV,**"**RecentAdvancesInNdds(Novel DrugDeliverySystem)ForDeliveryOfAnti-HypertensiveDrugs."Int.DrugDev. &Res.**2011**,3(1),252-259.
10. ManivannanRandKugalurGP,"RecentAdvancesInNovelDrugDeliverySystem."Int.J.Res.Ayur.andpharm.**2010**,1(2),316-326.
11. MartinhoN,"RecentAdvancesinDrugDelivery Systems."J.ofBio.andNanobiotech.**2011**,2,510-526.
12. ShahA, "Applicationofnoveldrugdeliverysysteminthepharmacotherapyofhyperlipidemia."J. ofChem.andPharma. Sci.**2013**,6(3),138-146.
13. DhruvinT,"PharmacokineticsinDrugDevelopmentandNovelDrugDeliverySystem." M.pharmQA, Parul collegeofpharmacy limda, Waghodia,1-80.
14. ChhotalalK,"Nanoemulsionbasedemulgel: ANoveltopicaldrugdeliverysystem."Asian.J. ofTopicalDisease.**2014**,4(1),S27-S32.
15. SwathiB,"Emulgelasatopicaldrugdeliverysystem."Int. J. Rev. Life. Sci.**2013**,3(3),58-65.
16. Singla V,"Emulgel:Anew platformfortopicaldrugdelivery."Int. J.Pharm.andBioSci.**2012**,3(1),485-498.
17. JoshiB,"Emulgel: acomphrehensivereviewontherecentadvances inthetopicaldelivery."Int.Res.J.Pharm.**2011,**2(11),66-70.
18. M Mangale, "Nanoemulsion: As Pharmaceutical Overview." Int. J. Pharm. Sci. Rev. Res. **2015**, 33(1), 244-252.
19. Himesh S, "Current Update on Nanoemulsion: A Review." Scholars International Journal of Anatomy and Physiology. **2021**, 4(1), 6-13.
20. Kumar S, "Nanoemulsions: increasing possibilities in drug delivery." Eur. J. Nanomed. **2013**, 5(2), 97-110.
21. B Chatterjee, "Targeted drug delivery to the brain via intranasal nanoemulsion: Available proof of concept and existing challenges." International Journal of Pharmaceutics. **2019**, 565, 258-268.
22. G Kumar, "Nanoemulsion Based Targeting in Cancer Therapeutics." Med chem. **2015**, 5(5), 272-284.
23. Charles Lovelyn, "Current State of Nanoemulsions in Drug Delivery." Journal of Biomaterials and Nanobiotechnology. **2011**, 2, 626-639.
24. Nor Azrini and Nadiha Azmi**,** **"**Nanoemulsions: Factory for Food, Pharmaceutical and Cosmetics." MDPI. **2019**, 7, 617.
25. Manish Kumar, "Techniques for Formulation of Nanoemulsion Drug Delivery System: A Review." Preventive Nutrition and Food Science. **2019**, 24(3), 225-234.
26. ShahA,"Emulgel:ATopicalPreparationforHydrophobicDrugs."PhTecMed.**2013**,2(5),370-376.
27. HymaP,"Emulgel: Review." Int.J.Pharm.Archive.,**2014**,3(3),1-11.
28. Sonaje S**,**"GellifiedEmulsion:ANewBornFormulationforTopicalDeliveryofHydrophobicDrugs."Worl.J.Pharm.andPharma.Sci. **2013**,3(1),233-251.
29. HaneefaK,"Emulgel:AnAdvancedReview."J. PharmaSci.&Res.**2013**,5(12),254–258.
30. https://www.loudouncenterforplasticsurgery.com/skin-anatomy
31. DebjitB,"RecentAdvancesinNovelTopicalDrugDeliverySystem."ThePharma.Inno.**2012**,1(9), 12-31.
32. VatsS,"EmulsionBasedGelTechnique:Novel Approachfor EnhancingTopicalDrugDeliveryofHydrophobicDrugs."Int.J.Pharma.Res.School.**2014,**3(2),649-660.
33. BenameurH."Lipid-basedDosageForms-Anemergingplatformfordrugdelivery."https://www.aaps.org/uploadedFiles/Content/Sections_and_Groups/Sections/Formulation_Design_And_Development_Section/FDDTechCornerJan2012.pdf
34. ShresthaH,"Lipid-BasedDrugDeliverySystems."J.Pharma.**2014**, 1-10.
35. SinghP,"Emulgel: ANovel Approach toBioavailabilityEnhancement."Int.J.Rec.Adv.InPharma.Res.**2014**,4(2), 35-47**.**
36. https://go.drugbank.com/drugs/DB00257
37. Pradeep K, "Clotrimazole Loaded Ufosomes for Topical Delivery: Formulation Development and In-Vitro Studies." Molecules. **2019**, 24, 3139.
38. Renuka M, "Formulation, development and characterization of mucoadhesive film for treatment of vaginal candidiasis." International Journal of Pharmaceutical Investigation. **2016,** 6(1), 47-55.
39. Umme H, "Formulation Design and Evaluation of A Novel Vaginal Delivery

System of Clotrimazole." IJPSR. **2014**, 5(1), 220-7.
40. Crowley P, "Clotrimazole as a pharmaceutical: past, present and future." Journal of Applied Microbiology. **2014,** 117, 611-7.
41. Surendran S, "Formulation, Evaluation and Optimization of Clotrimazole Solid Dispersion Incorporated Gels." Journal of Medical Pharmaceutical and Allied Sciences. **2014**, 4, 42-51.
42. Fahima M, "Formulation, Characterization, and Clinical Evaluation of NanoemulsionContaining Clotrimazole for Topical Delivery." AAPS Pharm. Sci. Tech. **2011**, 12(3), 879-86.
43. Kamalesh tripathi, "development and in vitro assessment of buspirone loaded in situ Nanoemulsion gel." International journal of pharmaceutical applied. **2021**, 13(4), 142-149.
44. Jasjeet Narang, "Formulation and Evaluation of Tocopheryl Polyethylene Glycol (TPGS) Stabilised Nanoemulsion of Curcumin for Topical Application." International Journal of Current Research and Review. **2021**, 13(7), 149-155.
45. Suraj mandal, "formulation and evaluation of carica papaya Nanoemulsion for treatment of dengue and thrombocytopenia." Plant archives. **2021**, 21(1), 1345-1354.
46. Abdhesh and Sanjiv G, "Formulation and Evaluation of Nanoemulsion Based Nanoemulgel of Aceclofenac." Journal of pharmaceutical science and research. **2020**, 12(4), 524-532.
47. Lígia N. and M. Ribeiro, "Functional Hybrid Nanoemulsions for Sumatriptan Intranasal Delivery." Sumatriptan in Hybrid Nanoemulsions. **2020**, 8, 1-11.
48. Shaikh M, "Formulation and Evaluation of Nanoemulsion for Topical Application." Journal of Drug Delivery & Therapeutics. **2019**, 9(4-s), 370-375.
49. Bharti Gaba, "Vitamin E Loaded Naringenin Nanoemulsion via Intranasal Delivery for the Management of Oxidative Stress in a 6-OHDA Parkinson's Disease Model." BioMed Research International. **2019**, 1-21.
50. Niyaz Ahmad, "Preparation of a novel curcumin Nanoemulsion by ultrasonication and its comparative effects in wound healing and the treatment of inflammation." RSC Adv. **2019**, 9, 20192–20206.
51. Hira Choudhury, "Formulation development and evaluation of rotigotine mucoadhesive Nanoemulsion for intranasal delivery." Journal of Drug Delivery Science and Technology. **2019**, 1-33.
52. Sally galal, "Intranasal tadalafil Nanoemulsion: formulation, characterization and pharmacodynamic evaluation." Pharmaceutical development and technology. **2019**, 1-13.
53. Ramsha Iqbal, "Design and development of letrozole Nanoemulsion: A comparative evaluation of brain targeted Nanoemulsion with free letrozole against status epilepticus and neurodegeneration in mice." International Journal of Pharmaceutics. **2019**, 565, 20-32.
54. Jyotsana suyal, "Formulation and evaluation of Nanoemulsion for enhanced bioavailability of itraconazole." International journal of pharmaceutical sciences and research. **2018**, 9(7), 2927-2931.
55. Sagar savale, "Formulation and evaluation of intranasal Nanoemulsion containing rutin." Asian journal of research in pharmaceutical sciences and biotechnology. **2017**, 5(3), 49-52.
56. Ebtsam M Abdou, "Brain targeting efficiency of antimigrain drug loaded mucoadhesive intranasal Nanoemulsion." International Journal of Pharmaceutics. **2017**, 529, 667-677.
57. Mithila Boche, "Quetiapine Nanoemulsion for Intranasal Drug Delivery: Evaluation of Brain-Targeting Efficiency." AAPS pharmaceutical science technology. **2016**, 18(3), 1-11.
58. Mrunali Patel, "Preparation and in vitro/ex vivo evaluation of Nanoemulsion for transnasal delivery of paliperidone." Applied Nanoscience. **2016**, 6(8), 1095-1104.
59. Yogendra Pandey, "Intranasal delivery of paroxetine Nanoemulsion via the olfactory region for the management of depression: formulation, behavioural and biochemical estimation." Nanotechnology. **2015**, 27(2), 1-15.
60. Hetal Thakkar, "Formulation and evaluation of Itraconazole Nanoemulsion for enhanced oral bioavailability." Journal of microencapsulation. **2015**, 32(6), 559-569.
61. Hitendra Mahajan, "Nanoemulsion-based intranasal drug delivery system of saquinavir

mesylate for brain targeting." Drug delivery. **2014**, 21(2), 148-154.
62. Hiren Patel, "Formulation and evaluation of o/w Nanoemulsion of ketoconazole." An international journal of pharmaceutical sciences. **2013**, 4(4), 338-351.
63. Prajapati, "Nanoparticles Containing Gel Formulation for the Treatment of Psoriasis."Int. J. Pharm. Investigation. **2021**. 11(1), 76-81.
64. Prem Samundre, "TOPICAL GEL." International journal of creative thoughts.**2020**, 8(3).04-9.
65. Kashyap, "Formulation and Evaluation of Transdermal Topical Gel of Ibuprofen." Journal of Drug Delivery & Therapeutics.**2020,** 10(2), 20-25.
66. Patil S, "A Review on Topical Gels as Drug Delivery System." journal of drug delivery and therapeutics. 2019, 5(2) 09-13.
67. Krishna Sailaja, "An Overall Review on Topical Preparation of Gel." Innovate International Journal Of Medical & Pharmaceutical Sciences. **2016**, 1(1)557-582.
68. Ruchi Sharma, Gurvinder Kaur and Deepak N Kapoor, "Fluconazole Loaded Cubosomal Vesicles for Topical Delivery." International Journal of Drug Development and Research. **2015**, 3(2)144-62.
69. K. Rodrigues, "QBD approach for the development of hesperetin loaded colloidal nanosponges for sustained delivery: In-vitro, ex-vivo, and in-vivo assessment." OpenNano. **2022**, 100045, 1-12.
70. Tejashree Waghule, "Quality by design (QbD) in the formulation and optimization of liquid crystalline nanoparticles (LCNPs): A risk based industrial approach." Biomedicine & Pharmacotherapy. **(2021) ,**141; 111940
71. Vishal R, "Systematic implementation of quality-by-design (QbD) to develop NSAID-loaded nanostructured lipid carriers for ocular application: preformulation screening studies and statistical hybrid-design for optimization of variables." Drug Development and Industrial Pharmacy**(2020)**,1-13.
72. Sarwar Beg, "QbD-Based Development of Cationic Self-Nanoemulsifying Drug Delivery Systems of Paclitaxel with Improved Biopharmaceutical Attributes." AAPS PharmSciTech, **(2019)** 20: 118,1-13.