International Journal of Pharmacy
Journal Homepage: http://www.pharmascholars.com

Research Article **CODEN: IJPNL6**

NANOCARRIER FOR INTRANASAL ADMINISTRATION

Pranita Savardekar*, Amrita Bajaj, Anupama Puntambekar

Department of Pharmaceutics, SVKM's Dr. Bhanuben Nanavati College of Pharmacy, Mumbai-400056, India

***Corresponding author e-mail:** pranita.savardekar@gmail.com

ABSTRACT

The objective of the present research work study was to design, optimize and characterize Nanoemulsion for improved brain transport of the drug. Drug nanoemulsion was optimized using Box Behnken Design. Particle size, zeta potential, and Polydispersity index were measured using Malvern Zetasizer. Morphology of nanoemulsion droplets was examined using scanning electron microscopy. Drug diffusion studies were performed and drug diffused was estimated using UV spectroscopic analysis. Stable nanoemulsions were formulated. The optimized nanoemulsion showed a uniform size distribution in the range of 40-100nm with zeta potential in the range of -18mv to -30 mV. The metered dose intranasal nanoemulsion sprays of Drug will prove as an alternative to conventional intranasal delivery for therapy of bipolar disorders and mania.

Keywords: Intranasal, nanoemulsion, Scanning electron microscope, Box-Behnken design

INTRODUCTION

Many drugs are difficult to being effectively and efficiently been delivered using conventional drug delivery approach to brain or central nervous system (CNS) due to complex structure. Intranasal administration is a promising approach for rapid-onset and delivery of medication to the CNS bypassing the BBB. Intranasal drug delivery is one of the major delivery options for brain targeting, as the nose and brain compartments are connected to each other via the olfactory route and via peripheral circulation. The drugs which are absorbed nasally via olfactory epithelium are found to enter in olfactory neurons and supporting cells and subsequently into the brain, with not only reduced systemic toxicity of centrally acting drugs but also enhanced therapeutic efficacy.

Intranasal drug delivery systems emerged as novel drug delivery systems because of their advantages such as absorption of drug is rapid via highly vascularised mucosa, large nasal mucosal surface area for dose absorption is available, onset of action is rapid, non-invasive route and ease of administration, bypass of the BBB, degradation of drug observed in GIT is avoided, hepatic first pass metabolism is absent, alternate to parenteral route especially for proteins and peptides, improved bioavailability, fewer side effects due to low dose, patient convenience and compliance is improved and self-administration is possible.[1]

Nanodelivery systems are fast becoming important approaches for delivering lipophilic compounds and improving physical and chemical stability of active ingredients within formulations. Nanoemulsions are transparent or translucent systems covering a size range of 10-100 nm. They are also referred to as submicron miniemulsions, ultrafine emulsions and emulsions.

The purpose of the present study was to design and develop was to develop a suitable drug delivery system to incorporate and improve the bioavailability of antipsychotic drug into brain tissue.[2] Drug nanoemulsion was optimized by Box- Behnken statistical method based on responses like globule size, zeta potential and viscosity, and invitro characterization was successfully established.[3]

MATERIALS AND METHODS

Materials: Oleic acid was purchased from S.D. Fine Chemicals, tween 20 and PEG 400 was received as a gift sample from Mohini Organics

Experimental work: Procurement & Standardization of drug and excipients

Formulation Development:

Development of microemulsion using ternary phase diagram: The oils, surfactants and cosurfactants were selected from preliminary studies. The pseudo-ternary phase diagrams were constructed using water titration method to determine the microemulsion region and to detect the possibility of making microemulsions with different compositions of oil, surfactant/ co-surfactant, and water. The ratios of surfactant to co-surfactants were chosen to be 1:1, 2:1; 3:1 mixtures were prepared. These mixtures (S/CoS) were mixed with the oil phase to give the weight ratios of 9:1, 8:2, 7:3, 6:4, 5:5, 4:6, 3:7, 2:8 and 1:9. Water was added drop by drop and stirred using vortex mixer until a homogeneous dispersion or solution was obtained. The end point of the titration was the point where the solution becomes cloudy or turbid. The quantity of the aqueous phase required to just make the mixture turbid was noted. [4]

Preparation of nanoemulsion: Oil and surfactants were weighed accurately in a clean glass beaker and stirred moderately. Weighed quantity of drug was dissolved in above oil-surfactant mixture. Sufficient quantity of water was added under stirring .It was then passed into high pressure homogenizer in order to reduce particle size.

Optimization of nanoemulsion: The Box-Behnken statistical screening design was applied for optimizing the formulation ingredients to develop a stable drug nanoemulsion for brain delivery. A three factor two level Box-Behnken response surface method using design expert (version 9 Stat-ease) was used to optimize the required formulation. The high, medium and low values were selected from preliminary experimentation. The independant variables are concentration of PEG 400, Tween 20 and labrasol and the dependent variables used in this design are Globule size and Zeta potential.

Preparation of conventional drug solution: The drug solution meant for comparative evaluation of nanoemulsion based system was prepared by dissolving the drug in propylene glycol: ethanol mixture.

Evaluation of Developed Formulations:

Brain Deposition study: Male Sprague–Dawley rats weighing 200–250 g were anesthetized with an intraperitoneal injection of thiosol (40 mg/kg) and kept on a heating pad to maintain the body temperature. The trachea was cannulated with a polyethylene tube (PE 200) to allow free breathing. An incision was made in the skin over the occipital bone. The first layer of muscle was cut, and the atlanto-occipital membrane was exposed. All of the incisions were covered with wet gauze. For the intranasal administration, about 30 min after operation, 100 µl of the nasal formulation was administered via a PE 10 tube attached to a microlitre syringe inserted 1 cm into each nostril of rat at a dose of 4 mg/kg. For the i.m. administration, the drug solution was delivered (dose equivalent to 4 mg/kg) through the cranial thigh muscle, volumes were between 0.46 and 0.54 ml. At 15min, 30min, 60min, 120min, 180min, and 240min after the dose, the rats were euthanised brain tissue was withdrawn by cisternal puncture (Dahlin and Björk, 2000). Each brain tissue was quickly rinsed with saline and blotted up with filter paper to get rid of blood-taint and macroscopic blood vessels as much as possible. After weighing, the brain tissue samples, they were homogenized with Tissue Homogenizer (Remi Motors) at about 100 rpm for 10 min. Resulting mixture was centrifuged at 15,000 rpm (Eppendorf 5810 R, Rotor F-45-30-11) for 20 min (4^0C). The supernatant was filtered through 0.45um syringe filter and stored at -80^0C until the time of analysis. Samples were injected and the chromatographic separations were achieved on analytical column (Kromasil C-8 (25 x 0.46 mm, 5 µm). The mobile phase was prepared using methanol, acetonitrile and millipore water in the ratio of 40:20:40 v/v at a pH-3 and were filtered through a 0.45 µ filter. Flow rate of mobile phase was kept at 1.5 ml/min. Drug was detected at a wavelength of 260 nm. The concentration of Drug in the sample was analysed according to their area under curve and using respective straight line equation.

RESULTS AND DISCUSSIONS

Formulation development:

Pseudo-ternary Phase studies: The aim of the construction of pseudo-ternary phase diagrams was to find out the existence region of microemulsions. Pseudo-ternary phase diagrams were constructed to establish the optimum concentrations of oil, surfactant and co-surfactant used for the formulation of microemulsion. Based on solubility study Oleic acid was selected as oil, Tween 20 was selected as surfactant , Labrasol was selected as cosurfactant and

PEG 400 was selected as co-solvent. In pseudo-ternary phase diagram, the three axes represent oil phase, the aqueous phase and third represents fixed weight ratio of surfactant and co surfactant. The various Smix ratios were tried, such as 1:1, 2:1 and 3:1. Among these ratios, Smix 1:1 was found to be the best combination, where the microemulsion formed was less viscous, spontaneous in formation, had good drug loading capacity and best clarity as compared to all other formulations with different Smix ratios. So this oil, surfactant and co-surfactant ratio was used in preparation of microemulsion. [5]

Optimization of nanoemulsion:

Influence of factors on Globule size: (Y1) Globule size=+74.95+15.43*A+78.51*B-9.69*C+18.21*AB-11.05*AC-
44.98*BC+15.61*A2+105.63*B2+10.31*C2

For globule size concentration of Tween 20 was found to be significant model term. The predicted R^2 value for response globule size was 0.8379 and adjusted R^2 value was found to be 0.8341 which indicates the model has predicted the responses well.

As seen from the regression equation, positive coefficient of A and B indicates globule size increases with increase in concentration of PEG 400 and Tween 20. Negative coefficient of C indicates decrease in globule size with increase in concentration of PEG 400.Results are shown in

Influence of factors on Zeta potential:

Final equation in terms of coded factors: Zeta potential=-21.29+5.16 *A - 1.91 * B - 1.97 * C+ 3.41 * AB + 0.19 * AC – 7.33 * BC-1.68 *A^2- 6.20 * B^2+ 0.042 * C^2

For zeta potential concentration of PEG 400 was found to be significant model term. The predicted R^2 value for response zeta potential was 0.9979 and adjusted R^2 value was found to be 0.9943 which indicates the model has predicted the responses well.

As seen from the regression equation, positive coefficient of A indicates zeta potential increases with increase in concentration of PEG 400. Negative coefficient of B and C indicates zeta potential decreases with increase in concentration of Tween 20 and Labrasol respectively. Negative coefficients of A^2 and B^2 indicate zeta potential decreases with increase in concentration of PEG 400 and tween 20 respectively.[7]Results are shown in the **Figure.3-5.**

Brain Deposition Studies: To evaluate brain targeting efficiency, after nasal dosing, statistical differences between intramuscular conventional solution, intranasal conventional solution and intranasal nanoemulsio were assessed by comparison between Concentration vs AUC plots after administration of formulation intranasally. Male Sprague Dawley rats were given Intramuscular injections of Conventional drug Solution, intranasal spray of conventional solution and intranasal spray of nanoemulsion. The rats were Euthanised at Time Points of 15min, 30min, 60min, 120min, 180min, and 240min. The brains were isolated and drug separated by liquid liquid extraction was measured by Bioanalytical HPLC. [6].As seen from the **Figure 6**, the drug concentration in brain for intranasal nanoemulsion is 2 times more as compared to conventional intramuscular administration of Thienobenzodiazepine.

Conclusion: The delivery of drug molecules across the nasal mucosa opens a new hope for the systemic delivery of medicaments. Selection of appropriate oil, surfactant/co surfactant is vital factor to develop an efficient nanoemulsion formulation with optimum drug loading. Oleic acid was found to be suitable oil for the drug. Anti-psychotic drug loaded nanoemulsion was successfully prepared and *in vitro* characterization was established. The formulation was free from nasal ciliotoxicity. Formulations were also characterized for Scanning Electron Microscopy to determine shape and size of particles. The *in-vitro* studies demonstrated the potential of nanoemulsion for intranasal delivery of drug. Drug delivery through nanoemulsions is a promising area for continued research with the aim of achieving sustained release with enhanced bioavailability and for drug targeting to various sites in the body.

Acknowledgement: The authors are thankful to SVKM'S Dr. Bhanuben Nanavati College of Pharmacy for providing the necessary facilities for the preparation of the paper.

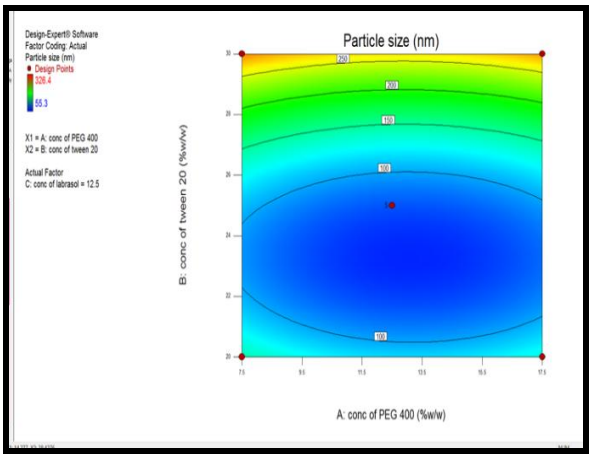

Figure 1: Contour plot showing effect on Tween 20 and PEG 400 on globule size

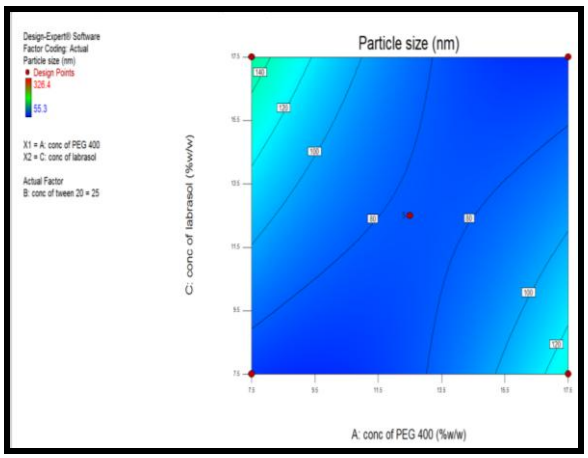

Figure 2: Contour plot showing effect on Labrasol and PEG 400 on globule size

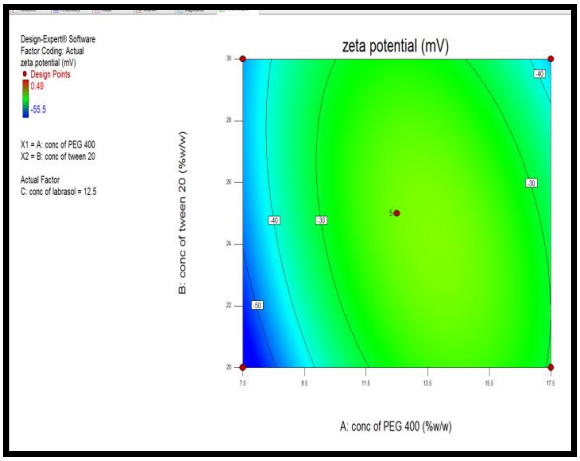

Figure 3: Contour plot showing effect on PEG 400 and Tween 20 on Zeta potential

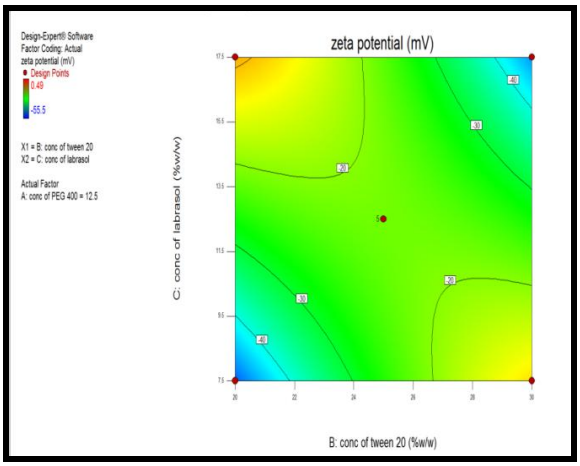

Figure 4: Contour plot showing effect on Tween 20 and Labrasol on Zeta potential

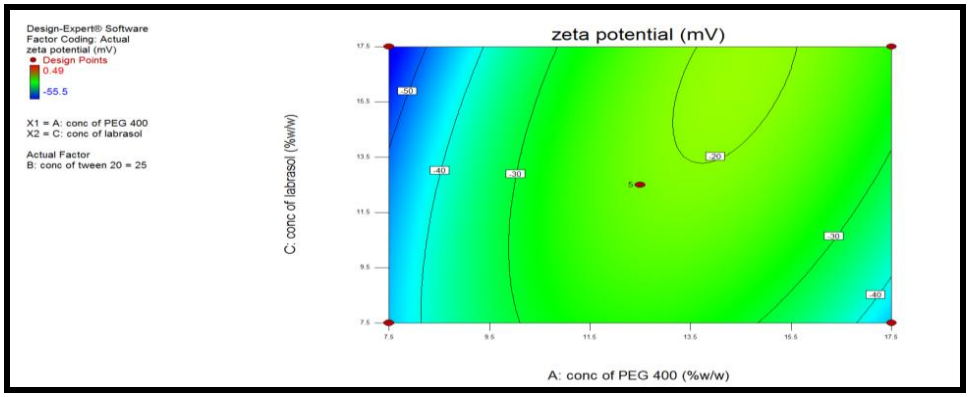

Figure 5: Contour plot showing effect on PEG 400 and labrasol on zeta potential

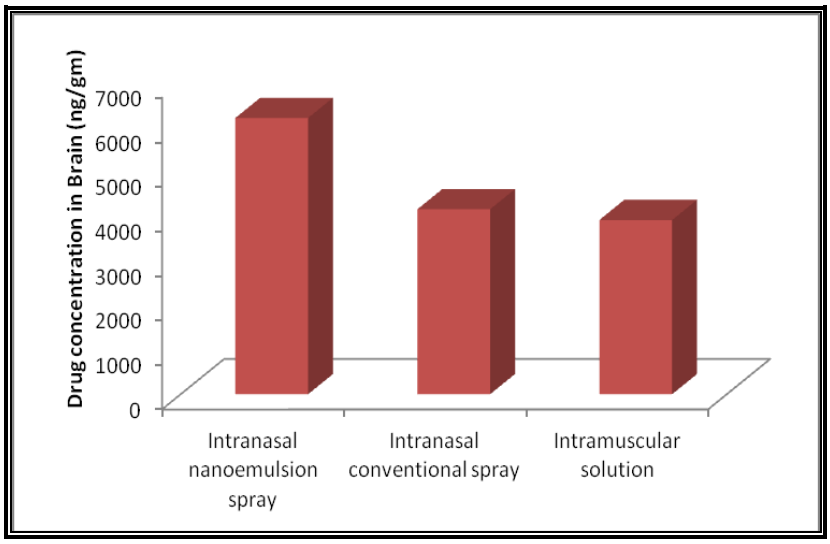

Figure 6: Comparitive evaluation of Drug concentration in whole brain tissue following intranasal administration of nanoemulsion, intranasal administration of conventional solution and intramuscular administration of conventional solution

Table 1: Box-Behnken Design

Std	Run	Factor 1 A:Conc of PEG 400(%w/w)	Factor 2 B:Concof Tween20 (%w/w)	Factor 3 C:Concof Labrasol (%w/w)	Response 1:Particle size	Response 2:Zeta potential
5	1	7.5	25	7.5	55.3	-25.36
7	2	7.5	25	17.5	86.79	-34.14
4	3	17.5	30	12.5	106.4	-26.0
15	4	12.5	25	12.5	104.1	-21.99
8	5	17.5	25	17.5	83.1	-19.51
12	6	12.5	30	17.5	101.9	-27.6
17	7	12.5	25	12.5	44.45	-28.72
13	8	12.5	25	12.5	65.4	-13.89
14	9	12.5	25	12.5	67.3	-16.39
3	10	7.5	30	12.5	87.7	-23.57
1	11	7.5	20	12.5	91.7	-18.91

11	12	12.5	20	17.5	125.1	-28.1
2	13	17.5	20	12.5	106.4	-24.21
10	14	12.5	30	7.5	99.5	-27.8
6	15	17.5	25	7.5	60.1	-33.85
16	16	12.5	25	12.5	67.1	-20.60
9	17	12.5	20	7.5	88.6	-26.0

REFERENCES

1. Pagar SA, Shinkar DM, Saudagar RB. J. Adv. Pharm. Educ. Res, 2013; (3): 4.
2. Botner S, Amnon CS. Pharmacol. Pharm, 2011; (2): 180-88.
3. Shailaja M, Divan PV, S. Ramakrishna, G. Ramesh, ReddyK.H, RaoY.M. Int. J. Pharm. Sci. Nanotechnol, 2012 ;(5)1.
4. Bhanushali RS, Gatne MM, Gaikwad RV, Bajaj AN, Morde MA. Indian J. Pharm. Sci, 2009; 71(6); 707-09.
5. Mishra D, Kumar A, Rakesh R, Chaturvedi A. Bull. Pharm. Res; 2013; 3(1): 34-9.
6. Dahlin M, Björk E. Int. J. Pharm, 2000; 195: 197–05.
7. Sood Sumeet, Jain Kunal, Gowthamarajan K. Colloids Surf. B, 2014; 113:330–37.