Journal of Advanced Zoology

ISSN: 0253-7214

*Volume **44** Issue S-**7** **Year 2023** Page 1915**:1926***

Formulation Optimization And Evaluation Of Drug Loaded Nanosponges For Oral Drug Delivery

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Abstract:

Depression is treated with venlafaxine HCL, Additionally, it is used to treat panic disorder, social anxiety disorder, and generalised anxiety disorder. The main goal of this study was to prepare venlafaxine HCL loaded nanosponges into tablets for an oral purpose by using Box-Behnken design to increase the bio availability. The drug was placed into nanosponges utilising the hot-melt compression technique, three-dimensional colloidal nanosponges were prepared by using β-cyclodextrin and dimethyl carbonate as a cross-linker. The effect of three Independent variables such as β cyclodextrin, dimethyl carbonate and response on two dependent variables vesicle size and entrapment efficiency were investigated. With the help of box behnken design 15 formulations were prepared out of which formulation 10 was considered as optimized based on the drug EE. The nanosponge formulations were evaluated for particle size, percent drug entrapment, shape and surface morphology, drug content, and in vivo drug release studies. The prepared nanosponge had a particle size of 257.6 nm, a drug entrapment percentage of 97.42 percent, and was almost spherical in shape. Utilizing HPMC K4M, the formulations NA1 through NA15 matrix tablets were crushed into drug-loaded nanosponges for 24 hours.

Keywords: *β-cyclodextrin , drug release, optimization, drug entrapment, particle size*

Introduction:

The healthcare system makes use of nanomedicine, a multidisciplinary use of nanotechnology. It identifies distinct physical, chemical, and biological properties of therapeutic material at the nanoscale scale in order to diagnose, treat, or prevent sickness. A number of the many forms of nanomedicines being investigated, including nanoemulsion, nanosuspension, nanotubes, etc., have generated interest in nanosponges (NS), which are microscopic sponges having the capacity to incorporate a wide range of substances into their core. Long-term medication distribution and dose management issues have been a source of frustration for medical

researchers. The creation of novel, intricate molecules known as nanosponges presents a solution for these issues. An new class of materials called nanosponges is formed of nanometer-sized particles with interior chambers that may accommodate various chemicals. These particles may transport hydrophilic and lipophilic materials as well as increase the solubility of water-insoluble compounds

Everyone goes through periods of grief and misery throughout their life. However, compared to ordinary sadness or bereavement, clinical depression is more severe and lasts longer, making it difficult for a person to carry out everyday tasks. Loss of interest or pleasure in formerly pleasurable activities, significant changes in appetite (either significantly reduced or increased), sleep issues (sleeping too much or too little), fatigue, a sense of worthlessness or hopelessness, difficulties concentrating and making decisions, and suicidal thoughts are just a few of the symptoms of depression.

Major depression (or major depressive disorder) and dysthymic disorder are the two basic subtypes of depression. The symptoms listed above have been present in a person with major depression for more than two weeks. These signs might recur often.

Bipolar illness sufferers also exhibit depressive symptoms. The severe condition known as bipolar disorder causes mood swings between "up" and "down" states. Mania, the bipolar "up" state, is marked by an ecstatic (joyful, energetic) mood, hyperactivity, a positive, expansive attitude on life, grandiosity (a hyper-inflated sense of self-esteem), and the idea that everything is possible. One or more of the aforementioned depressed symptoms are present in a person who is experiencing the "down" phase of bipolar illness.

Depression, anxiety, panic attacks, and social anxiety disorder are all treated with venlafaxine (social phobia). Your attitude and energy may be lifted, and it could also help you regain interest in day-to-day activities. Additionally, it could lessen dread, worry, unwelcome thoughts, and panic attacks. Box-Bhenken A time-, effort-, and chemical-saving optimization technique called factororial design may be utilised to develop designs for formulae that are acceptable. A scientific strategy for evaluating components' relative weight and the cumulative impact of those factors on different responses is called a factorial design. Additionally, response surface characterization is a successful method for getting the right model without needing a lengthy testing time. Utilizing factorial design tools, the recipe for Venlafaxine hcl nanosponges was optimised in this study. The scaling up process is applied to the improved formula. By releasing the medicine, the solubility and therapeutic effectiveness may be enhanced. In these studies, the Box-Behnken design approach was used to create Venlafaxine hcl nanosponges and find the best formula.

2. EXPERIMENTAL MATERIALS AND METHODS

Materials

The supplies required for this project were sourced from a number of places. Venlafaxine hydrochloride was obtained from (HETERO LABS, India). Beta Cyclodextrine was purchased from (RESEARCH –LAB FINE CHEM INDUSTRIES Mumbai, India). Dimethyl carbonate and HPMC K80 are from (RESEARCH –LAB FINE CHEM INDUSTRIES Mumbai, India).

Calibration curve (λmax) using UV-visible spectrophotometer

In phosphate buffer with a pH of 6.8, a standard stock solution of venlafaxine (100 g/ml) was created. Venlafaxine solution at a concentration of 10 g/ml was generated by appropriately diluting a standard stock solution with phosphate buffer pH 6.8 and was scanned in the 200–400 nm wavelength range. Maximum wavelength was chosen from the drug's overlaid spectra for investigation.

Preparation of nanosponges:

β-Cyclodextrin (β-CD) was cross-linked using cross-linking polymer namely DMC to prepare the nanosponge. Specific polymer-to-cross linker molar ratios were used for nanosponge preparation. The polymer-to-cross linker mixture was taken and allowed to react for 5–7 h at around 90 °C. To collect the resulting solid, the reaction mixture was kept aside to cool, and then filtered. Subsequently, the formed solid particles were broken down by gentle grinding, and Soxhlet installed extraction using ethanol for around 30 min to remove unreacted cross-linkers and other impurities. The reaction was conducted with an excess DMC, and the resulting nanosponge was deposited at 25 °C after purification until further use.After the nanosponges were made and the drugs were loaded into them, three distinct formulations were made. Then, the formulations of loaded nanosponges were immediately crushed into tablets, utilising HPMC and microcrystalline cellulose as diluents and binders, and finally, the tablets were characterised and evaluated.

Preparation of Nanosponges loaded tablets:
Tablets with nanosponges loaded were made using a direct compression technique for the optimal formulation (F10). All of the materials were screened via sieve no. 40 after being precisely weighed in the requisite amounts of the complexed dispersion powder and excipients. Except for the lubricant and glidant, all the materials were put into a mortar and well mixed in a glass mortar with a pestle for 15 minutes. The lubricant and glidant were then added before compression and completely combined for an additional 2-3 minutes. These lubricating g mixtures were crushed utilising a rotating tablet compress machine and 11 mm flat facing punches. The dry powdered dispersion was put through a number of tests before compression.

Table 1: Formulation chart

Ingredients	Formulations(weight in mg)				
	NA1	NA2	NA3	NA4	NA5
Drug : polymer					
Venlafaxine hydrochloride	100	100	100	100	100
B-cyclodextrin(mg)	350	200	300	320	500
DCM (ml)	10	20	15	10	5
HPMC K100M	50	135	75	80	40
MCC	65	120	85	70	0
Mannitol	45	45	45	40	0
Magnesium stearate	22	22	22	22	22
Talc	8	8	8	8	8
Total tablet weight	650mg	650mg	650mg	650mg	650mg

Characterization of active ingredient:
Statistical Design
DOE is an effective statistical method for enhancing product/process designs and resolving production/process issues. Experiments are often employed when examining a process to determine which process inputs significantly affect the process output and what the goal level of the inputs should be to attain a desired outcome (output). Experimental Design (ED) and Design of Experiments (DOE) are other names for the same concept. Streamline the time it takes to create new goods and processes. Boost the efficiency of current procedures Boost product performance and dependability Reach robustness in your products and processes

Optimization
There were a total of 15 unique formulation compositions revealed by Box-Behnken Design, with only 3 shared formulas (as indicated in Table 2) (centre point). Particle size (Y1) and entrapment efficiency (Y2) of the generated formulations were evaluated. These experimental data were provided to the BBD software for analysis of other statistical factors. An expected response was found using polynomial equations of the second order. The usefulness of BBD was quantified by its coefficient of determinants, which rose to a high value in this case. A high value for the regression coefficient indicates that the independent variable significantly influenced the dependent variables (p 0.05). Counter surfaces and three-dimensional graphs were made for each response to show how cyclodextrin concentration, cross linker, and reaction duration affected vesicle size (Y1) and entrapment efficiency (Y2,). Because of this experimental and expected value was also constructed.

Table 2: Optimization of formulations revealed by Box-Behnken Design

Runs	Fator 1 A: βCyclodxtrine (mg) (X1)	Fator 2 B:DMC (ml) (X2)	Factr 3 C: Reaction time (X3)	Reponse-1 Particle size (nm) (Y1)	Response -2 EE% (Y2)
F1	350	15	7.5	323	87
F2	500	20	7.5	316	96
F3	350	15	7.5	319	83
F4	200	15	6	290	79
F5	200	10	7.5	331	81
F6	350	20	9	441	91
F7	500	15	9	457	98
F8	200	15	9	470	80

F9	350	15	7.5	329	85
F10	500	15	6	287	97
F11	350	20	6	292	88
F12	350	10	6	287	86
F13	350	10	9	437	83
F14	500	10	7.5	339	96
F15	200	20	7.5	321	79

Pre-formulation study:
The pre-formulation study was carried out to show the good flow property and meeting the pharmacopeia specifications.

Angle of repose
A fixed funnel approach was used to calculate the angle of repose. The drug powder or its physical combination, which had been precisely weighed, was placed in a funnel after the physical mixes of the drug with various excipients had been made. The funnel was height-adjusted such that the tip of the funnel barely brushes the top of the pile of the medication powder. The funnel was left open, allowing the powder to freely pour out over the surface. The powder cone's height (h), diameter (d), and angle of repose were all measured and computed using the formula below:
Angle of repose (θ) = $\tan^{-1}$ (h/r)

Apparent bulk density
We measured and recorded the volume and weight of a pre-sieved drug excipient blend in a graduated cylinder to determine its apparent bulk density.

$$Density = \frac{Mass}{Volume}$$

Tapped density
A measured quantity of the powder combination was weighed and then tapped (100) in a graduated cylinder for a predetermined amount of time . Using this formula, we were able to calculate the tapped density.

$$Tapped\ density = \frac{weight\ of\ powder\ taken}{Tapped\ Volume}$$

Compressibility index
Using its apparent bulk density and tapped density, we were able to determine the powder mixture's compressibility as a percentage using the formula below.

$$Compressibilty\ Index = \frac{Tapped\ density - Bulk\ density}{Tapped\ density}$$

Hausner's ratio
The Hausner ratio provides an indirect measure of powder flowability. The formula for its calculation is as follows:

$$Hausner's\ ratio = \frac{Tapped\ density}{Bulkdensity}$$

If the Hausner ratio is less than 1.25, the flow qualities are better than if it is greater than that number.

Evaluation of drug loaded nanosponge:
Weight Variation test
Each batch of twenty pills was carefully weighed using a digital balance. The standard deviation and average weight were computed. Following is the formula that was used to determine the percentage of difference.

$$\%\ \text{Deviation} = \frac{\text{Individual weight} - \text{Average weight}}{\text{Average weight}} \times 100$$

Thickness test
Vernier Calipers were utilised to assess the thickness of 10 tablets from each batch. The standard deviation and mean thickness were both given to us.

Hardness test
6 tablets were taken from each batch and tested using a Pfizer hardness tester to get an average and standard deviation for the batch's hardness.

Friability test
This test evaluates the ability of tablets to withstand physical stress. The friability was tested in the following manner using a Roche friabilator: The friabilator was loaded with ten pre-weighed tablets. After being spun at 25 revolutions per minute for four minutes, the pills were analysed (100 rotations). At the conclusion of the test, the tablets were reweighed, and the percentage by which their weight had decreased served as the measure of friability.

$$\% \text{ Friability } = \frac{W1 - W2}{W1} \times 100$$

Where W_1 = Initial weight of 10 tablets
W_2 = Weight of the 10 tablets after testing

Assay of tablets
10 tablets were weighed and crushed to a fine powder in a mortar and pestle to get 1 mg of powder, and this powder was then dissolved in 100 ml of phosphate buffer, which has a pH of 6.8. Drug concentration was determined by filtering the solution through 0.45 m filter paper and then roughly diluting it with pH 6.8 phosphate buffer before measuring its absorbance at 226 nm using a UV-Vis spectrophotometer.

In vitro drug release studies
To record how much venlafaxine was released over the course of 24 hours from nanosponges-loaded venlafaxine matrix tablets, a USP type II dissolving test equipment was used. The release was performed at 37 0.5 °C at a rotational speed of 75 rpm. Initially, the dissolution experiments are conducted in a pH 1.2 buffer for 2 hours; then, the dissolving media is switched to a phosphate buffer of pH 6.8 for the final 2 hours of the research, which can last up to 24 hours. In order to keep the sink conditions constant, 5 ml samples were removed at regular intervals (up to 24 hours) and replaced with fresh medium. Before analysing the samples, they were filtered and diluted to the correct concentration.
Spectrophotometer for the visible and ultraviolet ranges set at 226 nm. The total amount of substance in all of the samples was determined.

Kinetic analysis of drug release data
Various kinetic models have been developed to reflect the total drug release from various dosage formulations. The in vitro release data for venlafaxine hydrochloride from these nanosponges loaded tablets were explained using a number of different equations and kinetic models. The Higuchi model, the Korsmeyer-Peppas model, the zero-order equation, and the first-order equation were all employed to simulate the kinetics of the experiment. When trying to comprehend drug release from pharmaceutical matrices, the Higuchi model is the most common theoretical framework employed. When there is uncertainty about the release mechanism or when there are different types of releases, the Peppas model is often employed (Peppas 1985). For a deeper comprehension of the drug release mechanism from polymeric materials, the dissolution data was matched using the Korsmeyer-Peppas model (Korsmeyer et al., 1983).
$M_t / M_\infty = Kt^n$

Here, Mt is the amount of drug released at time t, K is the normal kinetic constant for the drug/polymer, and n is the transport/diffusion exponent that provides insight into the mechanism of drug release (Kuksal et al. 2006). In the Korsmeyer-Peppas equation, the zero-order release applies only if n = 1, while the super Case II transport applies if n > 1. (Pasa et al. 2012; Afaf et al. 2017). Anomalous transport (non-Fickian) transport is indicated due to drug diffusion and the concentration gradient if n=0.5 and n=1, respectively, representing the release mechanism as Fickian Diffusion (Higuchi matrix).

Surface pH study
In order to induce swelling, a bioadhesive tablet was exposed to 1 ml of distilled water at room temperature for 2 hours. After allowing the pH metre electrode to acclimatise for a full minute, the tablet's pH was measured (Bottenberg et al., 1991).

Stability studies.
Stability refers to how well a product retains the same qualities it had when it was first made, within specific limits, and during its entire storage and use duration. The new mixture was sealed and kept at 40 ° C with 75 % of total relative humidity for a month. Regular checks were performed on the formulation's colour, pH, drug purity, and in vitro drug release.

Pharmacodynamics studies for anti-depression activity
Forced Swim test:
Porsolt et al. were the ones who originally published the forced swim test (FST), which has grown to be the most well-liked behavioural paradigm for identifying antidepressant-like behaviour in mice. The identical procedure was followed as before. Individual mice were trained to swim in a fresh water tank that measured 25 x 15 x 25 cm and was kept at a temperature of 26 °C. Animals were unable to survive at this water level by resting their tails or rear feet on the side walls or chamber's floor. Since "used water" has been shown to alter behaviour, the water in the chamber was changed after each animal received FST. Each animal showed vigorous activity for the first two minutes of the experiment. The duration of immobility was manually recorded for the next four minutes of the six-minute testing period.
Mice were thought to be immobile when they stopped squirming and continued floating motionless in water, moving only enough to keep their heads above water. The mice were towel dried after their swimming session, then placed back in their environment.

Production yield (%)
All produced nanosponge compositions were accurately weighed and recorded. Next, the following equation was used to compute the nanosponge's output yield:.
Production yield (%) = practical mass of nanosponge x 100/theoretical mass (polymer + drug)

Determination of drug content and % Entrapment efficiency (EE%)
To ensure complete vesicle lysis, we diluted 10 mg of drug-loaded nanosponges in 30 ml of ethanol and raised the volume up to 100 ml with phosphate buffer at pH 6.8 in a volumetric flask. The transparent solution was then evaluated using a UV spectrophotometer. A mortar and pestle were used to crush drug-loaded nanosponges of known weight. The pH 6.8 phosphate buffer and 5 ml of ethanol were added to the standard 100 ml flask until it was full. After the mixture was left alone for an hour while being vigorously shaken, it was sonicated to ensure that all of the nanosponge vesicles had ruptured. After the solution was diluted appropriately, it was filtered to remove any trace of nanosponge particles, and its spectrophotometric properties were evaluated using a UV/VIS spectrophotometer at a maximum wavelength of 226 nm. The percentage of medication that was caught was determined by the following formula:

$$\% \text{ EE} = \frac{\text{Total drug content} - \text{unentraped amount of drug}}{\text{total drug content}} X100$$

$$\% \text{ Drug loading} = \frac{\text{Total drug content} - \text{unentraped amount of drug}}{\text{total weight of bilosomes}} \text{X}100$$

Vesicle size, polydispersity index (PDI) and zeta potential (ZP) analysis
Drug-loaded nanosponges were diluted in double-distilled water, and the samples were then put into a quartz cuvette for a zeta sizer to measure the vesicle size and PDI (Zetasizer Nano, Malvern, UK). With the use of a particular cuvette that included an electrode, the same sample was examined for the purpose of determining its zeta potential. After the samples were properly diluted, the mean vesicle size, PDI, and ZP of the generated formulas were determined using the dynamic light scattering method and Zetasizer. Distilled water was used to dilute about 1 ml of the dispersion to 10 ml. After shaking, the suspension was put into a typical cuvette to test the zeta potential. The sample was kept at a constant temperature of 25 °C. By examining the charged vesicles' electrophoretic mobility, the ZP determination was assessed.

Melting point determination
Using Thiel's melting point apparatus, the melting point of the drug may be determined by inserting a little amount of the medication in a capillary tube with a closed end and heating the tube to the point where the drug melted.

Fourier-transform infrared spectroscopy (FTIR).
FTIR spectroscopic analysis was carried out for pure drugs, polymers used, and their physical mixture for the compatibility evaluation of drugs and polymers. FTIR spectrophotometer was used to obtain the spectra.

Particle size determination

Using a Zetasizer, we were able to calculate the polydispersity index and average particle size of nanparticulate dispersions. The dispersion sample required to be diluted with water for this test. Prior to particle size measurement, the material was filtered using a membrane filter with a 0.45 mesh size.

Surface Morphology studies of the nano particles

The following methods may be used to determine the morphology, size, and dispersion of SLN particles.

Scanning electron microscopy (SEM)

Nanosponges' surface morphology was reported using SEM. On an aluminium stub with double-sided sticky tape, appropriate tests were placed. First, the sample powder was evenly spread throughout the surface of the stub after the tape had been securely fastened to it. The stub containing the material was then sputter-coated with a tiny coating of gold to keep the specimens conductive

***In Vivo* Permeation of Venlafaxine hydrochloride through rat epithelium membrane from optimized drug loaded nanosponges**

The in vivo permeability of drug-loaded nanosponges for the ideal formulation across the membrane of the rat epithelium was investigated. The intactness of the rat epithelial membrane was sustained by placing it in a phosphate-saline buffer solution at pH 7.4. A Franz diffusion cell with an internal diameter of 2.1 cm and a membrane placed over it were sandwiched between the recipient and donor compartments. The donor compartment was inserted with the optimised nanosponges formulation (4 mg/ml), which was securely clamped to prevent two compartments from coming loose from the epithelial membrane. In the receptor compartment, 25 cc of phosphate buffer with a pH of 7.4 were added. Temperature was maintained at 370 C and the complete setup was put over a magnetic stirrer. At predefined intervals, 2 ml samples from the receptor compartment were taken and replaced with an equivalent amount of buffer. A UV-Vis spectrophotometer was then used to measure the absorbance at 226 nm in order to assess how much of the medicine had penetrated through the rat skin mucosa from the improved formulation.

Table 3:

S.no	Touching 3 Walls	Swim/struggle	Immobility (sec)	Defecation
1	12	1.5	60	0
2	13	2.4	68	1
3	27	1.95	23	0
4	20	1.21	26	1
5	33	1.24	8	0
6	30	1.30	6	0
7	34	1.72	4	0

RESULTS AND DISCUSSION:

Determination of absorption maximum values

The maximum concentration (10 g/ml) of venlafaxine in a phosphate buffer with a pH of 6.8 was determined using a UV- spectrophotometer. A UV-Visible Spectrophotometer measured its highest absorbance at 226 nm after scanning it between 200 and 400 nm. The highest wavelength is the one researchers use.

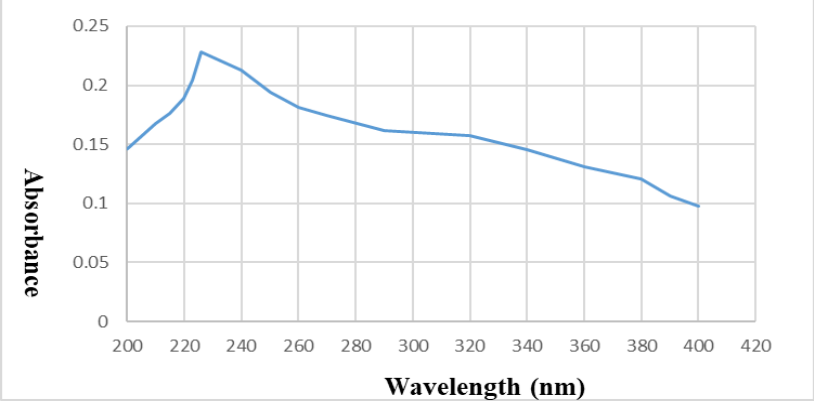

Figure 1: **UV absorption spectrum of Venlafaxine in phosphate buffer pH 6.8**

S. NO	Concentration µg/ml	Absorbance
1	2	0.236
2	4	0.416
3	6	0.61
4	8	0.778
5	10	0.942

Table 4: Concentration versus absorbance values

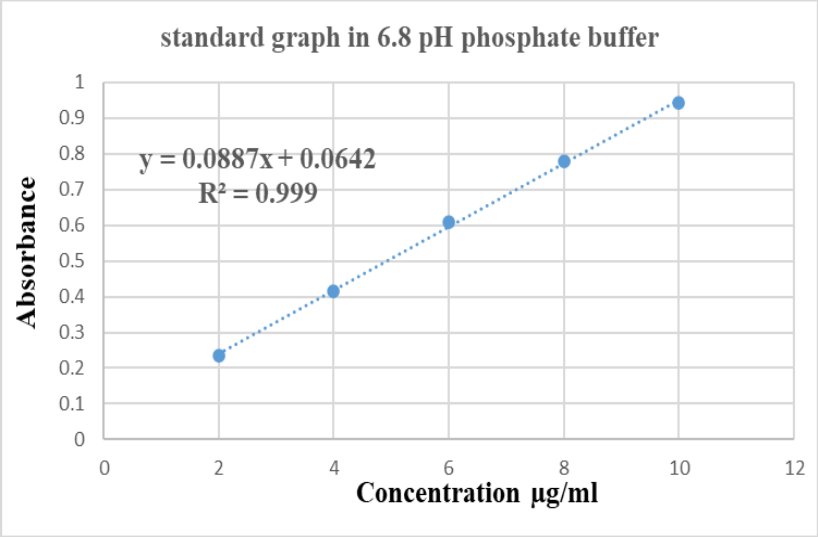

Figure 2: Standard graph of venlafaxine in pH 6.8 phosphate buffer

Solubility and melting point determination

In the first stage of our research, the drug's solubility and melting point were tested together with other preformulation factors. The solubility findings were shown in the following table. The sample's melting point was determined to be 105 0C.

Table 5: Drug solubility in different solvents

S.NO	SOLVENTS	SOLUBILITY
1	Water	Completely soluble
2	Chloroform	Completely soluble
3	Methanol	Completely soluble
4	Dimethyl sulfoxide	Completely soluble
5	6.8 pH buffer	Completely soluble
6	1.2 pH buffer	Completely soluble
7	7.4 pH buffer	Completely soluble

Production yield

The production yield for all venlafaxine loaded nanopsponges was calculated. Formulation F2 was showed 93% of yield which is less among all the formulations. In formulations F3, F8 the yield is 100% but vesicle size was high and entrapment efficiency low compared to Formulation F12. In F12, size of vesicle is optimum but entrapment efficiency is less. Among all, F10 have less vesicle size, more entrapment efficiency and high percentage yield, This was determined using an equation involving the theoretical weight and actual weight of the product.

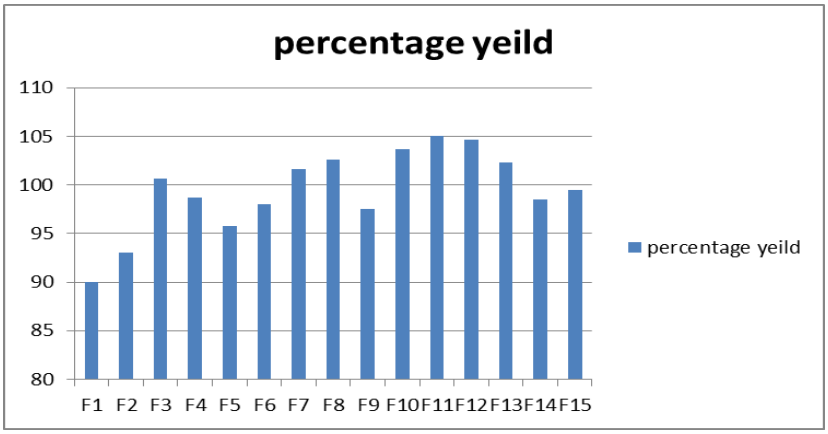

Figure 3: **Percentage yeild graph**

Vesicle Size, PDI, and Zeta Potential

Preparation of nanosponges yielded vesicle sizes ranging from 287 (F10) to 470 (F7). The vesicle size value was 248.6 nm for the optimal composition (F10) (Figure). Distribution homogeneity was indicated by a PDI value of 0.494, which is below the accepted threshold of 0.5. With a zeta potential of +24.1mV, we know that the nanosponges we generated are quite stable and that the vesicles did not clump together during preparation. ZP is a measure of physical stability since it provides a rough indication of the amount of repulsion between neighbouring vesicles. For formulation F7 the average vesicle size is PDI, Zeta potential was found to be 251.7nm, 0.466, 17.8mV respectively. Comparing to above formulations F10 is considered as optimized.

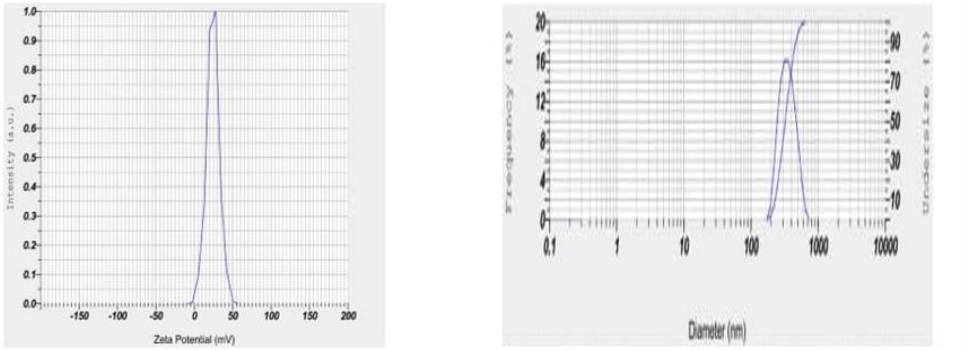

Figure 4: Graph representing zeta potential vesicle sizes

SEM

The optimal surface's morphology formula of venlafaxine loaded nanosponges (F10) is studied by scanning electron microscopy (SEM) in (figure 5). SEM analysis of optimized formulation revealed spherical particles with smooth surfaces and no aggregation, no vesicles with irregular surfaces.

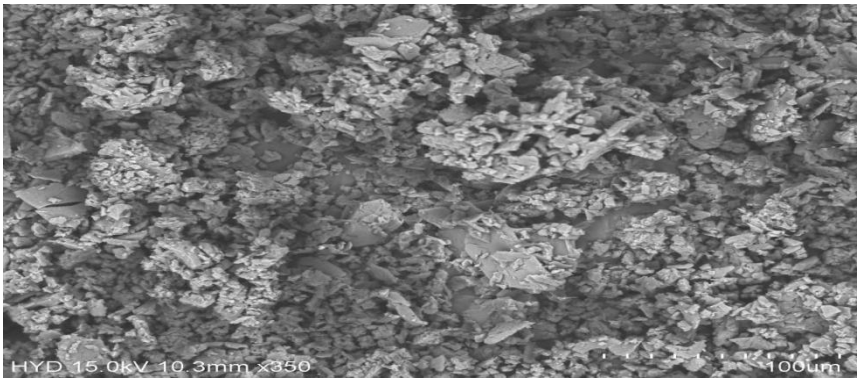

Figure 5: **Scanning electron microscopy image of venlafaxine loaded nanosponges**

***In Vivo* Permeation of Venlafaxine hydrochloride through rat epithelium membrane from optimized drug loaded nanosponges**

The F10 formulation was chosen for in vivo drug permeation experiments based on the vesicle particle size, Entarpment efficiency, and % yield of all formulations. Venlafaxine was released from the formulation and passed across the membrane as shown by the findings of drug permeation from Venlafaxine-loaded nanosponges via the rat skin epithelium. According to the findings, the drug permeability was gradual and consistent for the first four hours. Venalfaxine might penetrate the epidermal barrier for up to 24 hours using improved formulations F10. The table and picture displayed the overall proportion of medicine from optimised nanosponges that had entered the epithelium.

Table 6: Formulation composition of venlafaxine loaded nanosponges and data of responses

Time (hrs)	Cumulative % of drug permeated from optimized formulation F10
1	6.78%
2	12.95%
3	23.26%
4	31.785%
8	49.245%
12	68.87%
16	79.25%
20	89.24%
24	97.89%

Table 7: Physical parameters of optimized nanosponges loaded tablets

Formulation code	Weight variation (mg)	Thickness (mm)	Hardness (kg/cm^2)	Friability (%)	Assay (%)	Surface pH
NA1	648±1.06	4.82±0.02	6.1±0.2	0.14	97.52	6.54
NA2	654±1.43	4.89±0.03	6.5 ± 0.4	0.16	98.73	6.68
NA3	662±3.78	4.83±0.02	6.2 ± 0.3	0.24	99.66	6.82
NA4	653±2.1	4.76±0.04	5.8 ± 0.5	0.13	97.34	6.92
NA5	649±2.25	4.90±0.05	6.2 ± 0.3	0.18	96.58	6.97

In vitro drug dissolution studies

The formulations NA1 to NA5 matrix tablets were formulated by using HPMC K4M, and compressed into drug loaded nanosponges for 24 hrs. NA1 released more quickly than the other formulations in this formulation. Because the percentage of medication release decreased from 96.54% to 80.71% in 24 hours with an increase in polymer concentration from NA1 to NA5, for example. Only the NA3 formulation has shown a 24-hour drug release rate greater than 96.54%.

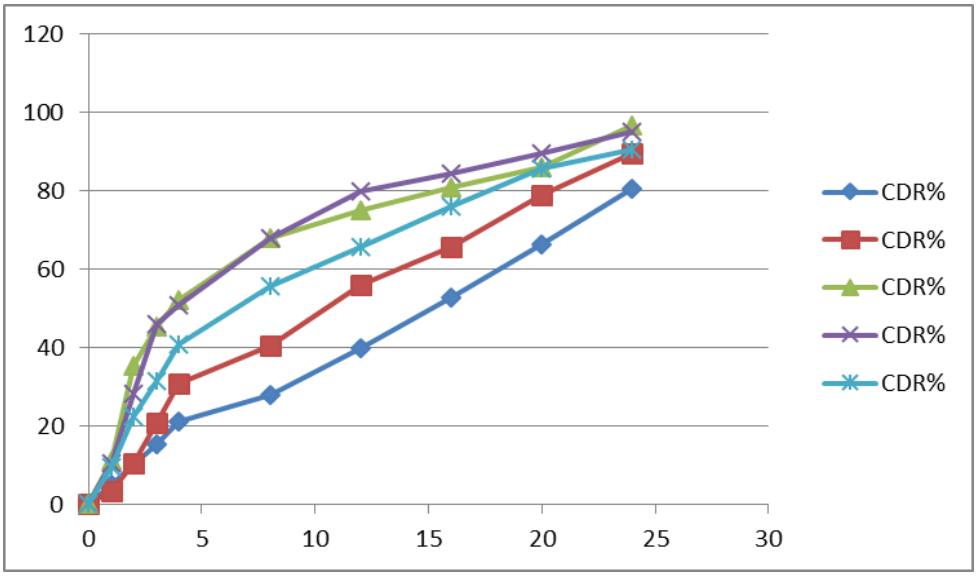

Figure 6: **Percentage medication release of NA1 to NA5 formulations**

Table 8: Kinetic parameters for the *in-vitro* release of Venlafaxine hydrochloride from different formulations of nanospongesloaded tablets

Formulation	Zero-order	First-order	Higuchi	Korsmeyer-Peppas	
	R^2				**N**
NB1	0.979	0.842	0.981	0.915	0.731
NB2	0.957	0.789	0.955	0.898	0.543
NB3	**0.947**	**0.893**	**0.988**	**0.952**	**0. 64**
NB4	0.944	0.913	0.994	0.943	0.832
NB5	0.961	0.923	0.988	0.928	1.127

Stability studies.

For stability testing, the venlafaxine nanosponges loaded tablet formulation NA3 was selected since it was thought to be the best formulation. The stability investigations were carried out for a month at 40°C and 75°RH. The acquired findings revealed no significant differences between parameters measured before and during the research period, showing that the formulation NA3 displayed a uniform colour, no colour change, and no flaws. The optimized formulation showed intact surface pH of 6.5 and 96.84% drug content, hence considered as stable formulation and passed the stability test.

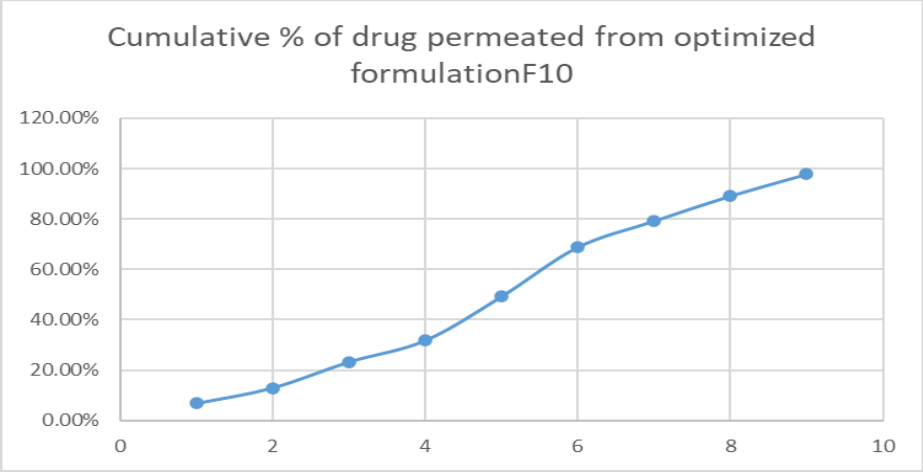

Figure7: Graph of cumulative percentage of drug of formulation F10

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