

**FORMULATION AND EVALUATION OF ROSUVASTATIN NANOSUSPENSIONS**S Raja Shekhar\*<sup>1</sup> and P Vijaya Lakshmi<sup>2</sup><sup>1</sup>CMR College of Pharmacy, Kandlakoya (v), Medchal road, Hyderabad- 501401.<sup>2</sup>Sri Datta Institute of Pharmacy, Sheriguda(v) Ibraimpatnam(M), Nagarjuna Sagar road, Rangareddy Dist-501510.**\*Corresponding author e-mail:** [s.rajashekhar2@gmail.com](mailto:s.rajashekhar2@gmail.com)**ABSTRACT**

The objective of the present study was to formulate and evaluate nanosuspensions of rosuvastatin, a poorly soluble drug in order to enhance its solubility and dissolution characteristics. Rosuvastatin is a Biopharmaceutical Classification System (BCS) Class II drug having very low solubility therefore low oral bioavailability. In this study rosuvastatin nanosuspensions were prepared by precipitation technique followed by high frequency sonication by using a combination of stabilizers like PVP K90 and LUTROL F127 in different ratios. The formulated nanosuspensions were characterised by Scanning Electron Microscope (SEM) and FTIR. The formulations were evaluated for drug content, entrapment efficacy, Zetapotential and In-Vitro dissolution. SEM results showed the particle size of the formulated nanosuspensions in nanosize. FTIR spectrum revealed that there are no interactions between drug and carriers. The effect of particle size was found to be significant on the saturation solubility of the drug and *in-vitro* drug release studies showed significant increase in the dissolution rate of nanosuspensions as compared with pure drug.

**Key Words:** Nanosuspensions, High Pressure Homogenizer, bioavailability, freeze drying, PVP K90, dissolution.**INTRODUCTION**

Nanosuspensions are colloidal dispersions of nanosized drug particles stabilized by surfactants. They can also be defined as a biphasic system consisting of pure drug particles dispersed in an aqueous vehicle in which the diameter of the suspended particle is less than 1 $\mu$ m in size<sup>[1]</sup>. Reduction of drug particles to nanometer range leads to an enhanced dissolution rate not only because of increased surface area but also because of saturation solubility. The increase in the saturation solubility and solution velocity of nanoparticle is due to increase of vapour pressure of the particles. More than 40 percent of the drugs coming from High-throughput screening are poorly soluble in water<sup>[2]</sup>. Obviously poorly water-soluble drugs show many problems in formulating them in conventional dosage forms. One of the critical problems associated with poorly soluble drugs is too low bioavailability and or erratic absorption<sup>[3]</sup>. Nanosuspensions are promising

strategy for the efficient delivery of hydrophobic drugs.

**Potential Benefits of Nanosuspension Technology for Poorly Soluble Drugs**

- Reduced particle size, increased drug dissolution rate, increased rate and extent of absorption, increased bioavailability of drug, onset time, peak drug level, reduced variability and reduced fed/fasted effects.
- Nanosuspensions can be used for compounds that are water insoluble but which are soluble in oil. On the other hand, Nanosuspensions can be used in contrast with lipidic systems, successfully formulate compounds that are insoluble in both water and oils<sup>[4]</sup>.
- Nanoparticles can adhere to the gastrointestinal mucosa, prolonging the contact time of the drug and thereby enhancing its absorption.
- A pronounced advantage of Nanosuspension is that there are many administration routes for

Nanosuspensions, such as oral, parenteral, pulmonary, dermal and ocular<sup>[5]</sup>.

- Nanosuspensions overcome delivery issues for the compounds by obviating the need to dissolve them, and by maintaining the drug in a preferred crystalline state of size sufficiently small for pharmaceutical acceptability.
- Nanosuspension of nanoparticles (NPs) offers various advantages over conventional ocular dosage forms, including reduction in the amount of dose, maintenance of drug release over a prolonged period of time, reduction in systemic toxicity of drug, enhanced drug absorption due to longer residence time of nanoparticles on the corneal surface, higher drug concentrations in the infected tissue, suitability for poorly water-soluble drugs and smaller particles are better tolerated by patients than larger particles, therefore increased resistance to hydrolysis and oxidation, increased physical stability to settling<sup>[6]</sup>.
- Nanosuspension has low incidence of side effects by the excipients.
- Reduced administration volumes, essential for intramuscular, subcutaneous, ophthalmic use. Finally, Nanosuspensions can provide the passive targeting. In the present study nanosuspensions of Rosuvastatin was prepared. Rosuvastatin is a antihyperlipidemic drug which comes under BCS class II. **HMG-CoA reductase inhibitors (statins)** are a class of drugs used to lower cholesterol levels by inhibiting the enzyme HMG-CoA reductase, which plays a central role in the production of cholesterol in the liver. Increased cholesterol levels have been associated with cardiovascular diseases (CVD), and statins are therefore used in the prevention of these diseases<sup>[7]</sup>.

## MATERIALS AND METHODS

Simvastatin was collected as a gift sample from Spectrum labs, Hyderabad, India. Pvpk-90 and lutrolF127 from S.d. Fine chem Mumbai. All other chemicals and solvents are of analytical graded.

### Preformulation studies

**Solubility studies:** Solubility of Rosuvastatin was carried out in different solvents like- distilled water, methanol, ethanol, DMSO and Di methyl formamide(DMF).Saturated solutions were prepared by adding excess drug to the vehicles and shaking on the shaker for 48 hr. at 25°C under constant vibration. Filtered samples (1ml) were diluted appropriately with 0.1N Hcl buffer and solubility of rosuvastatin was determined spectrophotometrically at 246nm.

The solubility of rosuvastatin in methanol and ethanol is approximately 1mg/ml and approximately 5mg/ml in DMSO and DMF.

**Melting Point:** The temperature at which the first particle of the substance completely melts is regarded as melting point of the substance. The temperature at which the first particle starts to melt and last particle completely melts is regarded as the range of melting point. Melting point of the drug was determined by capillary tube method and found to be 156-160°C.

**Organolectic properties:** The color, odor and taste of the drug were recorded using descriptive terminology and found to be white to off-white crystalline powder, tasteless and odorless.

**Drug-Excipient Interactions Studies:** There is always possibility of drug excipient interaction in any formulation due to their intimate contact. The technique employed in this study is IR spectroscopy. IR spectroscopy is one of the most powerful analytical techniques, which offers possibility of chemical identification. The IR spectra of Rosuvastatin, Pvp K-90 and optimised formulation (F4) were obtained by KBr pellet method using Perkin-Elmer series 1615 FTIR Spectrometer.

**Preparation of nanosuspensions:** All the ingredients including drug, polymer and excipients were weighed accurately according to the batch formula (Table-1). The required amount of polymer (carrier) and stabilizer were accurately weighed and added to required measure of H<sub>2</sub>O in a beaker. The drug was dissolved in solvent (methanol) and added to the above mixture in a drop wise manner using a syringe while on stirring Magnetic stirring for 1 hour and then ultrasonication for 2 hours.

### Evaluation of Nanosuspension Rosuvastatin:

The following evaluations were done to the formulated nanosuspensions:

**A. Drug content uniformity:** 10ml of each formulation was taken and dissolved in 10ml isotonic solution and kept overnight. 10 mg (similar as in formulation) of drug was taken and dilution was made to 10µg/ml. The dilutions were filtered and analyzed using UV for their content uniformity. The absorbance of the formulations were read using one cm cell in a UV-Vis spectrophotometer. The instrument was set at 245 nm. The drug content in each formulation was calculated based on the absorbance values of known standard solutions.

**B. Entrapment efficacy:** Entrapment efficacy was calculated by following formula:  
%Entrapment efficiency= Drug content \*100/Drug added in each formulation

**C.%Transmittance:** %Transmittance was measured by U.V spectroscopy at a wavelength of 245nm. A graph for %particle range vs. formulations was plotted.

**D.pH measurement:** The pH values were measured at 25 °C using a pH digital meter at  $20 \pm 1$  °C. The formulation was brought in contact with the electrode of pH meter and equilibrated for 1 min. This method was done in triplicate and mean was calculated along with standard deviation.

**F.Particle size and shape:** Particle size and shape of the formulated microcapsules was determined by using Optical Microscope.

**G.In vitro drug release study:** This is carried out in USP XXIII dissolution test apparatus-II (Electrolab TDT-06N), employing paddle stirrer at 50 rpm and 200 ml of pH 6.8 phosphate buffer as dissolution medium. The release study is performed at  $37 \pm 0.5$ °C. Samples of 5 ml are withdrawn at predetermined time intervals and replaced with fresh medium. The samples were filtered through 0.22 µm membrane filter disc (Millipore Corporation) and analyzed for Rosuvastatin after appropriate dilution by measuring the absorbance at 245 nm.

## RESULTS AND DISCUSSION

**Calibration curve using 6.8 phosphate buffer:** The linearity was found to be in the range of 5-25mcg/ml in distilled water, 6.8 phosphate buffer. The regression value was closer to 1 indicating the method obeyed Beer-lambert's law.

**Drug excipient compatibility:** Drug and excipient compatibility was confirmed by comparing spectra of FT-IR analysis of Pure drug with that of various excipients used in the formulation.

**Drug content:** The drug content of the formulated Nanosuspension was found in the range of 93.86 to 99.87 percent.

**Entrapment efficacy:** The entrapment efficacy of the formulated Nanosuspension was found to be in the range of 55.4%-96.7% respectively.

**%Transmittance measurement:** UV-Visible spectrum of pure Nanosuspension was recorded in range of 200-400 nm.

**Zeta Potential:** The measurement itself is a particle electrophoresis, the particle velocity is determined via the doppler shift of the laser light scattered by the moving particles. The field strength applied was 20 V/cm. The electrophoretic mobility was converted to the zeta potential in mV using the Helmholtz-Smoluchowski equation. At standard measuring conditions (room temperature of 25 °C, water) this equation can be simplified to the multiplication of the measured electrophoretic mobility (µm/cm per V/cm) by a factor of 12.8, yielding the ZP in mV.

The Zeta potential for the optimised formulation was found to be 12.5mv. When compared to the standard zeta potential values the optimised F<sub>4</sub> formulation was stable.

**Particle size:** The optimized batch (f4) had a average particle size of 300.3nm with 0.218 poly dispersivity index which indicate the particles are in uniform distribution. The particle size distribution pattern of the optimized nanosuspension formulation is given in figure 4.

## CONCLUSION

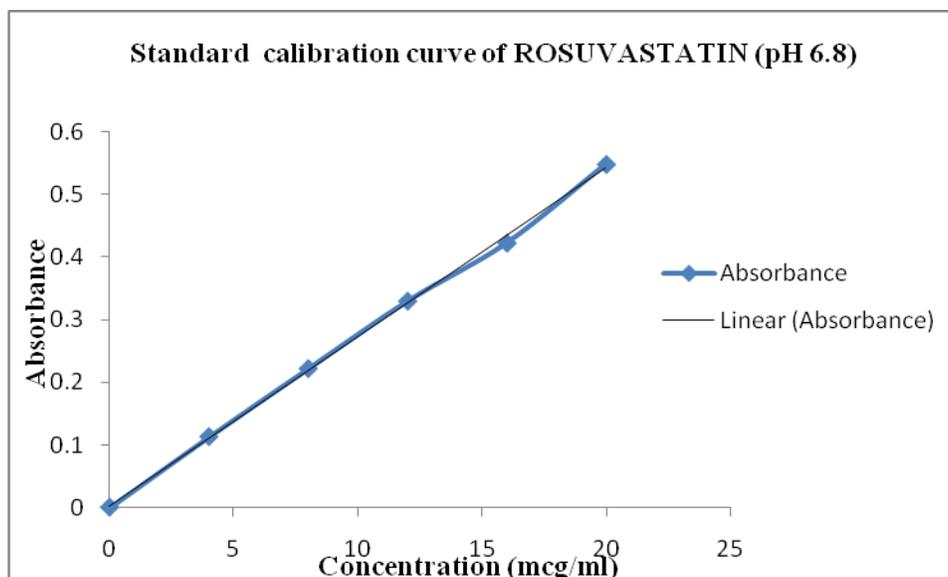
Rosuvastatin an anti hyperlipidic drug is a BCS class II drug, which has very poor solubility. Nanosuspensions were prepared by precipitation technique using stabilizers like PVP K90 and LUTROL F127 in different ratios. Formulation F4 which has 15% PVP K90 and 2% LUTROL F127 showed desired particle size and dissolution parameters and thus found to be optimised formula. SEM, Zetapotential and Particle size distribution studies of the optimised formula was found to be satisfactory.

**Table No. 1: Formula of nanosuspensions batch F1 to F6.**

INGREDIENTS	F1	F2	F3	F4	F5	F6
ROSUVASTATIN	25mg	25mg	25mg	25mg	25mg	25mg
PVP K 90	5%	5%	10%	15%	10%	15%
LUTROL F127	-	2%	2%	2%	4%	4%
METHANOL	2ml	2ml	2ml	2ml	2ml	2ml
WATER	Q.S to 50ml					

**Table No.2: *In vitro* drug release data of formulation F1to F6.**

Time(min)	F <sub>1</sub>	F <sub>2</sub>	F <sub>3</sub>	F <sub>4</sub>	F <sub>5</sub>	F <sub>6</sub>
0	0	0	0	0	0	0
5	14.1	20	28.3	34.5	38.4	39.7
10	25.23	28.5	35.2	48.2	70.1	70.1
15	38.85	45.2	49.7	79.8	85.1	85.1
20	49.43	67.1	75.6	86.27	96.25	96.25
25	56.25	88.3	86.3	98.5	-	-
30	69.83	99.5	100.5	-	-	-
35	78.4	-	-	-	-	-
40	82.8	-	-	-	-	-
45	98.85	-	-	-	-	-
50	-	-	-	-	-	-

**Fig.no.1: Standard calibration curve of Rosuvastatin in pH 6.8 phosphate buffer**

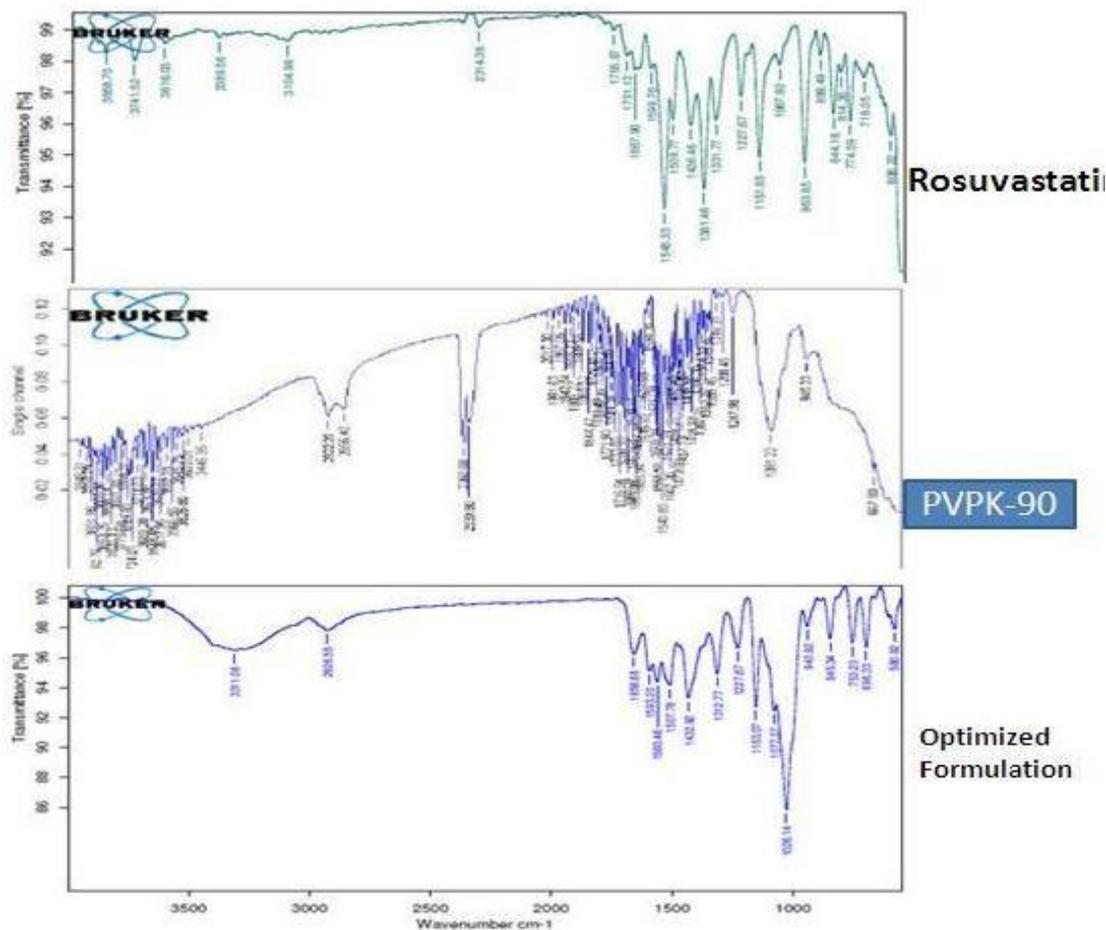


Fig.no.2: FTIR spectra of Rosuvastatin, PVP K90 and optimised formulation (F4).

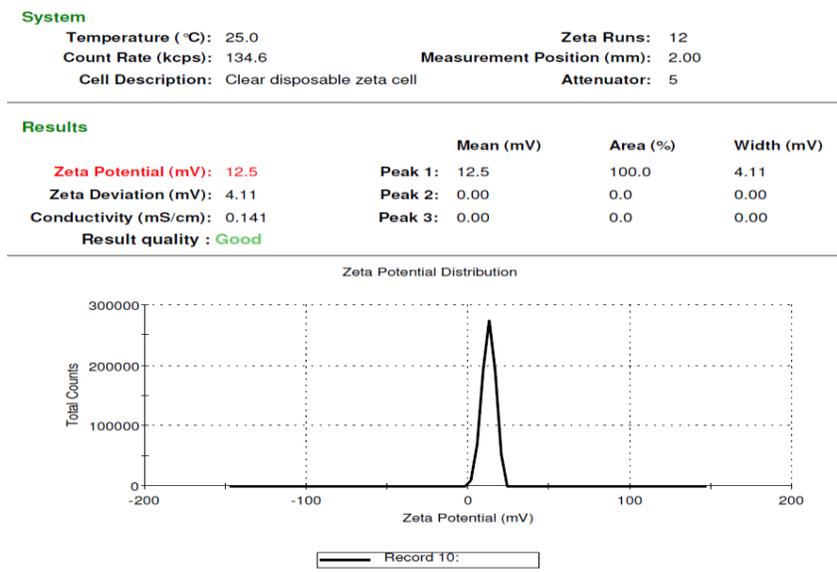


Fig.no.3: ZetaPotential For F<sub>4</sub> formulation

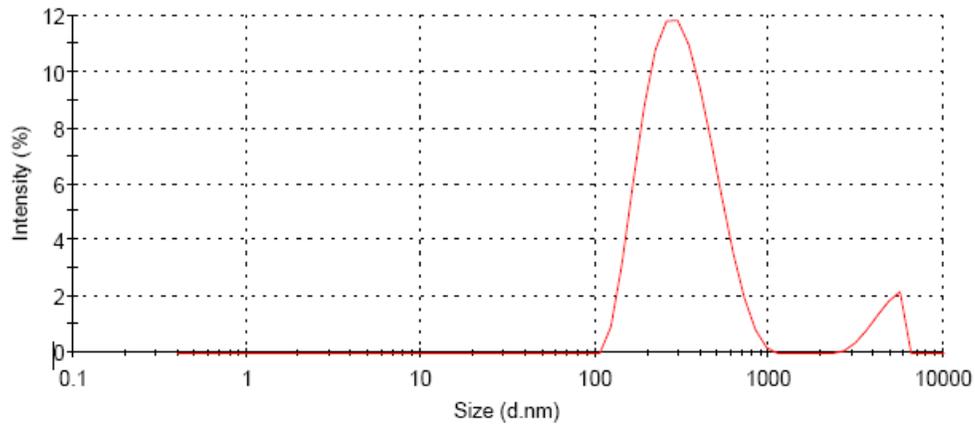


Fig.no.4: particle size graph for optimized formulation F<sub>4</sub>

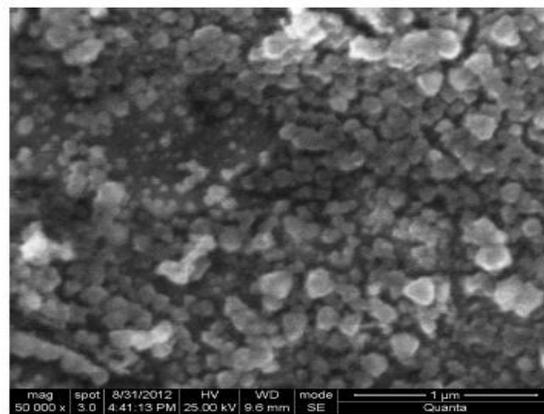


Fig. No.5: SEM picture of optimised formulation F<sub>4</sub>.

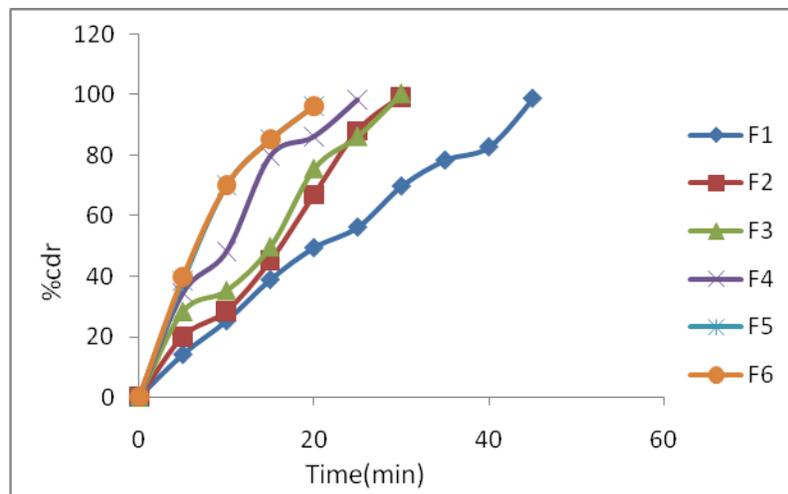


Fig. No.6: Percentage drug release vs. time graph of formulations F<sub>1</sub> to F<sub>6</sub>.

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