Journal of Cardiovascular Disease Research

ISSN: 0975-3583, 0976-2833 VOL 12, ISSUE 04, 2021

DEVELOPMENT OF AGOMELATIN LOADED NANOSUSPENSION UTILIZING STABILIZER AND SURFACTANT

[1]Mounika Kommuri[*], [2]Harekrishna Roy

Department of Pharmaceutics, Nirmala College of Pharmacy, Mangalagiri, Andhra Pradesh, India.

Corresponding author E Mail: mounikamouni12456@gmail.com

ABSTRACT:

The aim of the present work is to develop oral Nanosuspension of Agomelatin by solvent evaporation method using various Stabilizers and Surfactants such as PVP K30, Pluronic F127, Urea & SLS. Various formulation as well as process parameters were optimized in order to achieve desirable size and saturation solubility. Characterization of the prepared Nanosuspension was done with respect to particle size, zeta potential,

Saturation solubility, dissolution rate, morphology study (SEM), in-vitro dissolution study. Zetapotential value for the best formulation (F12) was found to -22mv which was found to be within theacceptable limits. Average particle size of nanosuspension of best formulations(F12) was found to be 118nm. From the invitro studies we can say that formulation F12 shows best drug releaseof 98.65% within 30 minutes where as all the other formulations didn't release the drug. The drug release from the Nanosuspension was explained by the using mathematicalmodel equations such as zero order, first order, and equation methods. Based on the regressionvalues it was concluded that the best formulationF12 follows first order kinetics.

Keywords: Agomelatin, PVP K30, Pluronic F127, Urea, SLS.

1. Introduction

The formulation of poorly water soluble drugs has always been a challenging problem faced by pharmaceutical scientists and it is expected to increase because approximately 40% or more of the new chemical entities being generated through drug discovery programs are poorly water-soluble. 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. Nanosuspension have revealed their potential to tackle the problems associated with the delivery of poorly water-soluble and poorly water-and lipid soluble drugs, and are unique because of their simplicity and the advantages they confer over other strategies. Solubility is the crucial factor for drug effectiveness, independence of the route of administration. Today the world is really facing a huge problem of poorly water soluble drugs. Many methods are there for increasing the solubility, but nanotechnology is one of the most prominent and latest technologies. It deals with the nanoparticles (having high surface area) which are useful for increasing the solubility of poorly water soluble drugs. Nanoparticles are the end products of a wide variety of physical, chemical and biological processes some of which are novel and radically different, others of which are quite commonplace. A nanometre is 1 x 10-9 m or one millionth of a millimetre. To give a sense of this scale, a human hair is of the order of 10,000 to 50,000 nm, a single red blood cell has a diameter of around 5000 nm, viruses typically have a maximum dimension of 10 to 100 nm and a DNA molecule has a diameter of 2 – 12 nm. The use of the term "nanotechnology" can be misleading since it is not a single technology or scientific discipline. Rather it is a multidisciplinary grouping of physical, chemical, biological, engineering, and electronic, processes, materials, applications and concepts in which the defining characteristic is one of size6. The formulation of poorly water soluble drugs has always been a challenging problem faced by pharmaceutical scientists and it is expected to increase because approximately 40% or more of the new chemical entities being generated through drug discovery programs are poorly water-soluble. 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. Nanosuspension have revealed their potential to tackle the problems associated with the delivery of poorly water-soluble and poorly water-and lipid-soluble drugs, and are unique because of their simplicity and the advantages they confer over other strategies. Nanoparticles can be prepared from a variety of materials such as proteins, polysaccharides and synthetic polymers. The selection of matrix materials is dependent on many factors including: (a) size of nanoparticles required; (b) inherent properties of the drug, e.g., aqueous solubility and stability; (c) surface characteristics such as charge and permeability; (d) degree of biodegradability, biocompatibility and toxicity. The major goals in designing nanoparticles as a delivery system are to control particle size, surface properties and release of pharmacologically active agents in order to achieve the site-specific action of the drug at the therapeutically optimal rate and dose regimen.[1]

Depression is a common mental disorder that affects thought, behaviour, feeling, and physical well-being of an individual. It is projected that by 2030 depression will be the foremost contributor among the worldwide burden of diseases. Despite introduction of several novel classes of antidepressant drugs, advances in understanding the psychopharmacology, and biomarkers of major

ISSN: 0975-3583, 0976-2833 VOL 12, ISSUE 04, 2021

depression, only 60%–70% of patients with depression respond to antidepressant therapy. Of those who do not respond, 10%–30% exhibit treatment-resistant symptoms with difficulties in occupational and social function, suicidal thought, decline of physical health, and increased health care utilization. Though they have different mechanisms of action, all present antidepressants ultimately produce the same final rates of response and remission [3]. Severe and intolerable side effects of available antidepressants and limited success rate (60–70%) of first-line monotherapy drugs have resulted in preference of potentiation or augmentation therapy in treatment for depression.[2-3]

Agomelatin is an antidepressant agent with a novel mechanism of action. It acts as a potent agonist at the melatonergic receptors MT_1, MT_2 and as an antagonist at the 5-H receptor. Agomelatin acts on melatonin receptors in the suprachiasmatic nucleus and normalizes circadian rhythms, thereby improving sleep and resynchronizing disrupted circadian rhythms. It enhances basal prefrontocortical dopaminergic and noradrenergic transmission as well as increasing basal noradrenaline release in the dorsal hippocampus by 5-H receptor antagonism. It also decreases stress-induced glutamate release in the prefrontal cortex (PFC) and increases brain-derived neurotrophic factor (BDNF) in hippocampus and PFC. Agomelatin is an effective antidepressant with a rapid onset of action not only in patients with Major Depressive Disorder (MDD) but also in patients with severe MDD, seasonal affective disorder, bipolar I disorder, and generalized anxiety disorder . Compared to other antidepressants, agomelatin does not cause sexual dysfunction or worsening of sleep disturbances and has fewer side effects. Agomelatin treatment in forced swim test resulted in significant reduction in the immobility period at 8 mg/kg dose.[2-3]

The main objective of this work is an attempt to overcome the poor solubility and dissolution rate of the model drug (agomelatin) by using Emulsification solvent evaporation method.Agomelatin was used as the model drug, PVP K30, Pluronic F127 and Urea as stabilizers, SLS as surfactant and methanol as solvent were used.

2. Materials and Methods:

Materials

Agomelatin is procured from B.M.R Chemicals, Hyderabad. Urea, Pluronic F127, Sodium Lauryl Sulphate, Polyvinyl Pyrrolidone K 30 are procured from Rankem, Mumbai. Methanol is procured from Narmada chemicals, Hyderabad.

Pre-formulation studies: [7-14]

Prior to the development of dosage form, it is essential that certain fundamentalphysical and chemical properties of the drug molecule alone and when combined withexcipients are determined. This first learning phase is known as pre-formulation. The overallobjective of the pre-formulation is to generate information useful to the formulator indeveloping stable and bioavailable dosage forms which can be mass produced. The goals ofpre-formulation studies are:

To evaluate the drug substance analytically and determine its necessary characteristics

To establish its compatibility with different excipients.

Spectroscopic study:

Identification of pure drug:

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 startsto melt and last particle completely melts is regarded as the range of melting point.

Solubility studies:

Solubility of Agomelatin was determined in Methanol, Ethanol, pH 1.2, pH 6.8 and pH 7.4 phosphate buffers. Solubility studies were performed by taking excess amount of Agomelatinin different beakers containing different solvents. The mixtures were shaken for 48 hrs inrotary shaker. The solutions were centrifuged for 10mins at 1000 rpm and supernatant wereanalyzed at 275nm by using UV Spectrophotometry.

Drug-Excipient Interactions Studies:

There is always possibility of drug- excipient interaction in any formulation due to theirintimate contact. The technique employed in this study is IR spectroscopy.IR spectroscopy is one of the most powerful analytical techniques, which offers possibility ofchemical identification. The IR spectrum was obtained by KBr pellet method. (Perkin-Elmerseries 1615 FT-IR Spectrometer).

Preparation of calibration curve of agomelatin:[15-19]

Procedure for standard curve in pH 6.8:

10 mg of Agomelatin was dissolved in 10 ml of pH 6.8 by slight shaking (1000 µg/ml). 1 ml of this solution was taken and made up to 10 ml with pH 6.8, which gives 100 µg/ml concentration (stock solution). From the stock solution, concentrations of 5, 10, 15, 20, 25& 30 µg/ml in pH 6.8 were prepared. The absorbance of diluted solutions was measured at 275nm and a standard plot was drawn using the data obtained.

Method of Preparation of Nanosuspension:

Preparation of Agomelatin Nanosuspension by Emulsification solvent evaporation method: Nanosuspension was prepared by the Emulsification solvent evaporation technique.Agomelatinwas dissolved in methanol at room temperature (organic phase). This

ISSN: 0975-3583, 0976-2833 VOL 12, ISSUE 04, 2021

solution isfollowed by its emulsification into water containing different stabilizers of PVP K30, SLS,Pluronic F127, and Urea maintained at room temperature. Addition of organic solvents bymeans of a syringe positioned with the needle directly into stabilizer containing water, andsubsequently stirred on magnetic stirrer to allow the volatile solvent to evaporate. Evaporation leads to precipitation of the drug.

Table 1:

Ingredients	F1	F2	F3	F4	F5	F6	F7	F8	F9	F9	F10	F11
Agomelatin (mg)	200	200	200	200	200	200	200	200	200	200	200	200
Urea	25	50	75	100	_	_	_	_	_	_	_	_
PVP K-30	_	_	_	_	25	50	75	100	_	_	_	_
Pluronic F 127	_	_	_	_	_	_	_	_	25	50	75	100
SLS	10	10	10	10	10	10	10	10	10	10	10	10
Methanol	5	5	5	5	5	5	5	5	5	5	5	5
Water(ml)	40	40	40	40	40	40	40	40	40	40	40	40

Evaluation parameters of Nanosuspension Agomelatin: [15-30]

The Nanosuspension was evaluated for various parameters:-

1. Entrapment efficiency
2. Particle size analysis
3. Zeta potential
4. In-vitro drug release studies
5. Scanning electron microscopy

Entrapment efficacy:

The freshly prepared nanosuspension was centrifuged at 20,000 rpm for 20 min at 5°C temperature using cool ultracentrifuge. The amount of unincorporated drug was measured by taking the absorbance of the appropriately diluted 5 ml of supernatant solution at 275nm using UV spectrophotometer against blank/control nanosuspensions. DEE was calculated by subtracting the amount of free drug in the supernatant from the initial amount of drug taken.

The entrapment efficiency (EE %) could be achieved by the following equation:

%Entrapment efficiency= Drug content *100/Drug added in each formulation

Scanning electron microscopy: The morphological features of Agomelatinnanosuspension are observed by scanning electron microscopy at different magnifications.

Particle size and shape: Average particle size and shape of the formulated nanosuspensions was determined byusing Malvern Zetasizer ZS using water as dispersions medium. The sample was scanned100 times for determination of particle size.

In vitro drug release study: In vitro dissolution study was performed by USP dissolution apparatus-type II using 900 ml of6.8pH buffer as a dissolution medium maintained at 37 ± 0.5°C and stirring speed (50 rpm).The freshly prepared nanosuspensions were added to the dissolution medium, five-milliliter samples were withdrawn at specific intervals of time, then filtered through a 0.45µm filter paper and analyzed for their drug concentrations by measuring at 275nmwavelength.

The results of in vitro release profiles obtained for the NDDS formulations were fitted intotwo models of data treatment as follows:

1. Cumulative percent drug released versus time (zero order kinetic model).
2. Log cumulative percent drug remaining versus time (first- order kinetic model).

ISSN: 0975-3583, 0976-2833 VOL 12, ISSUE 04, 2021

Table 2:

	n	**ollowing the Model**
der	ln Q o + K t is proportional to amount of drug ng	oluble drugs ıs matrix
	o t ndent of drug concentration)	rmal systems c systems

Where, ft = fraction of dose released at time‘t’
KH, K0, and Ks = release rate constants characteristic to respective models
Qo = the drug amount remaining to be released at zero hour
Qt = the drug amount remaining to be released at time‘t’
W0 = initial amount of drug present in the matrix
Wt = amount of drug released at time‘t’
1.Zero Order Kinetics: A zero-order release would be predicted by the followingequation.

$$At = A0 – K0t$$

Where:
At = Drug release at time‘t’
A0 = Initial drug concentration.
K0 = Zero-order rate constant (hr-1).
When the data is plotted as cumulative percent drug release versus time, if the plot is linearthen the data obeys zero-order release kinetics, with a slope equal to K0.
2. First Order Kinetics: A first-order release would be predicted by the followingequation

$$Log\ C = Log\ C0\text{-}Kt$$

Where:
C = Amount of drug remained at time‘t’
C0 = Initial amount of drug
K = First-order rate constant (hr-1).
When the data is plotted as log cumulative percent drug remaining versus time yields astraight line, indicating that the release follows First-order kinetics. The constant ‘K’ can beobtained by multiplying 2.303 with slope values.

Zeta potential:
There are three ways by which a solid particle (colloid) dispersed in a liquid media canacquire a surface charge. First, by the adsorption of ions present in the solution. Second, bythe ionization of functional groups on the particle’s surface. Third, due to the difference indielectric constant between the particle and the medium. Attention should be paid to theformation of electric double layer at the solid-liquid interface. The zeta Potential is definedas the difference in potential between the surface of the tightly bound layer (shear plane)and the electro-neutral region of the solution. The potential gradually decreases as thedistance from the surface increases.As the concentration of electrolyte increases in the medium, the zeta potential falls offrapidly due to the screening effect of the counter ions (Figure 2). The zeta potential cannotbe measured directly; however, it can be calculated using theoretical models and fromexperimentally determined electrophoretic mobility data. The theory is based onelectrophoresis and can be expressed as:

$$\mu = \zeta\varepsilon/\eta$$

Where (μ) is the electrophoretic mobility, (ε) is the electric permittivity of the liquid, (η)
Is the viscosity and (ζ) us the zeta potential.

ISSN: 0975-3583, 0976-2833 VOL 12, ISSUE 04, 2021

3. Results and Discussion

Determination of melting point: The melting point of agomelatin was found to be in range of 108° C which was determinedbycapillarymethod.

Saturation Solubility: Saturation solubility was carried out at 25^0C using Methanol, Ethanol, 0.1N HCL, 6.8 phosphate buffer, and 7.4pH buffer.

Table3: Solubility data

Solvent	Solubility(mg/ml)
Ethanol	2.30
Methanol	2.72
CL	0.37
phosphate buffer	0.82
phosphate buffer	0.74

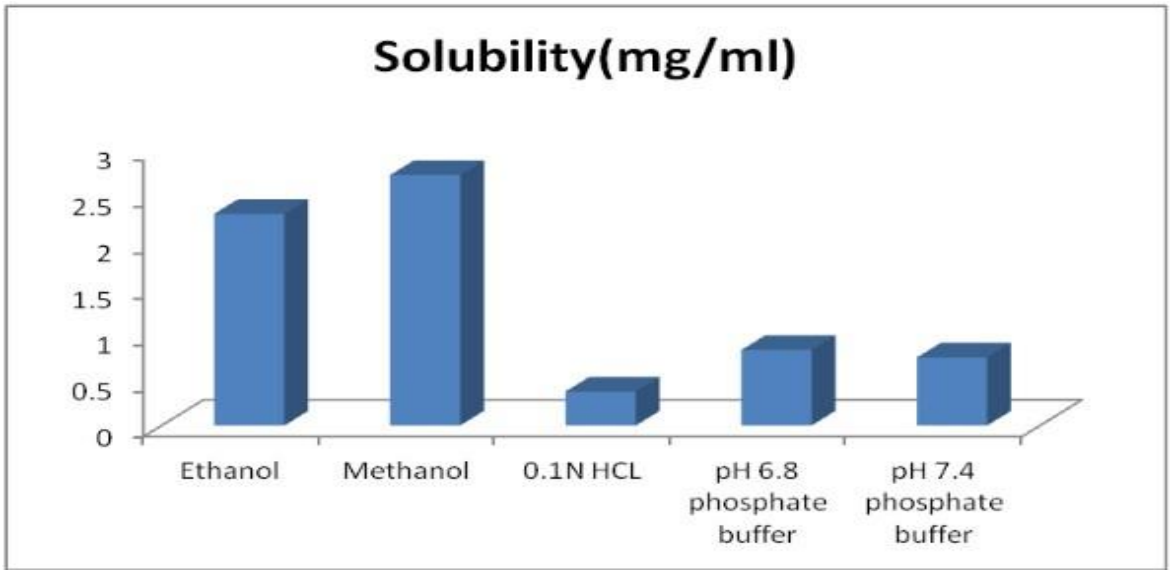

Fig:1Solubility studies of Agomelatin

Discussion: From the above conducted solubility studies in various buffers we can say that 6.8 phosphate buffer has more solubility when compared to other buffer solutions. So Ph 6.8 buffer is used as dissolution medium,based upon the solubility studies on organic solvents methanol has more solubility than others so methanol was used in the nanosuspension formulation.

ISSN: 0975-3583, 0976-2833 VOL 12, ISSUE 04, 2021

Determination of absorption maximum (λmax):
Determination of Agomelatin λ-max was done in pH 6.8 buffer medium for accuratequantitative assessment of drug dissolution rate.

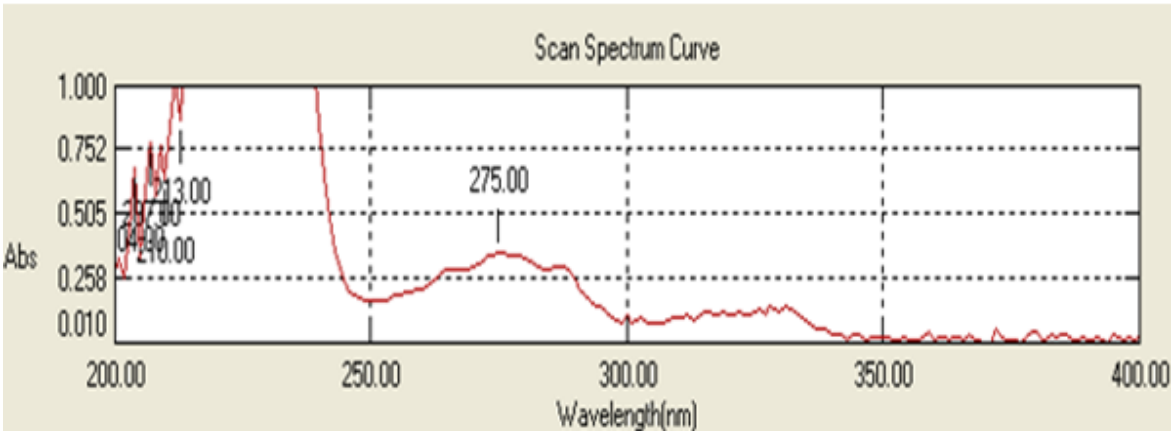

Fig 2:UV spectrum of Agomelatin

Table 4: Standard graph ofAgomelatinpH 6.8(λmax275 nm)

ntration (µg/ml)	**bance**

ISSN: 0975-3583, 0976-2833 VOL 12, ISSUE 04, 2021

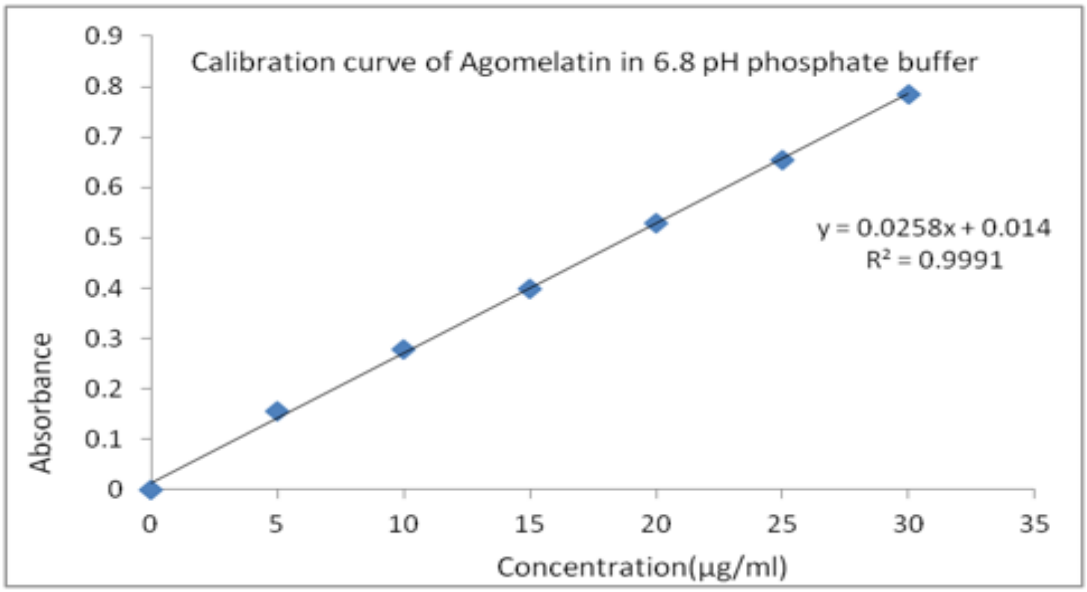

Figure 3: Standard calibrationcurve of AgomelatinpH 6.8

Discussion:

The linearity was found to be in the range of 5-30 µg/ml in acetone, pH 6.8 buffer. The regression value was closer to 1 indicating the method obeyed Beer-lamberts' 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.

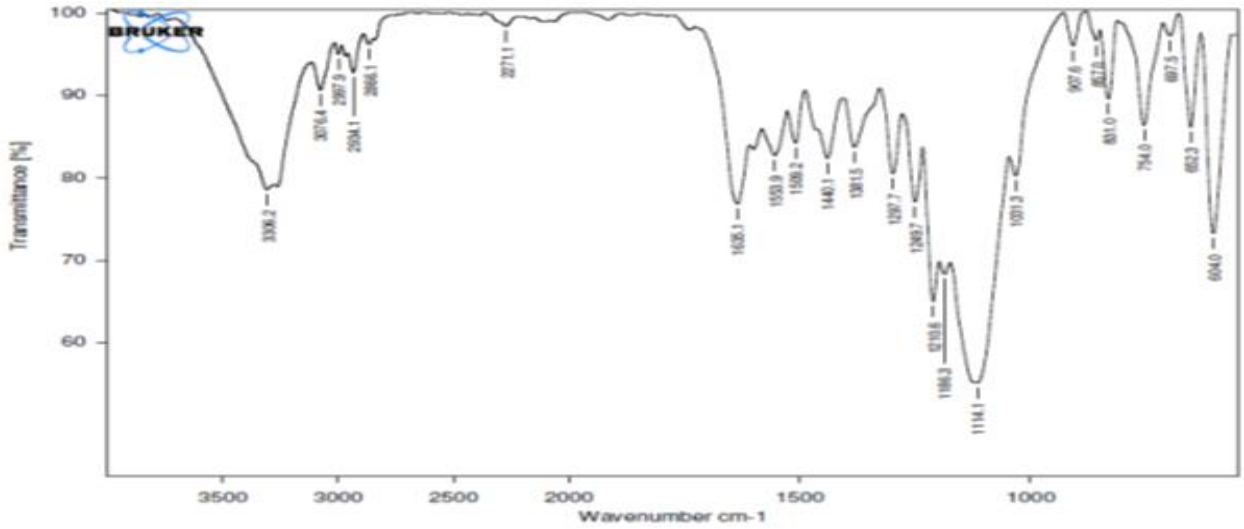

Figure 4: IR spectrum of Agomelatin

ISSN: 0975-3583, 0976-2833 VOL 12, ISSUE 04, 2021

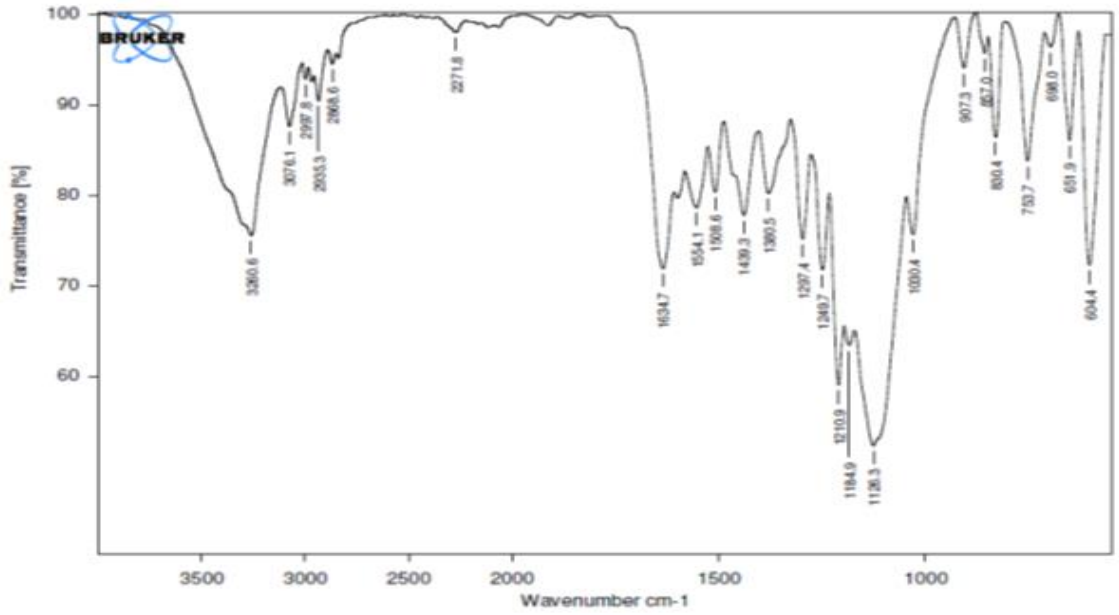

Figure 5: IR spectrum of Agomelatin Best Formulation

Discussion: The sample of best formulation F12 was triturated and mixed well with IR grade potassium bromide in 1:100 ratio. Then this mixture is introduced into the sample holder of FT-IR instrument and scanned in the range of 4000-400cm^{-1} to obtain spectrum.Form the drug excipient compatibility studies we observe that there are no interactions between the pure drug (Agomelatin) and best formulation (Agomelatin+ excipients) which indicates there are no physical changes.

Entrapmentefficacy: The entrapmentefficacy of theformulatedNanosuspensionwasfoundto beinthe rangeof82.46%-98.52%respectively.

Table 5: Entrapment efficiency of formulated Nanosuspensions

lation code	% entrapment efficiency

Discussion: The entrapmentefficacy of formulation F1 was found to be 83.23%, formulation F2 was found to be 85.15%, formulation F3 was found to be 87.45%, formulation F4 was found to be 89.63%, formulation F5 was found to be 88.23%, formulation F6 was found to be 97.14%, formulation F7 was found to be 93.26%, formulation F8 was found to be 95.15%, formulation F9 was found to be 97.26%, formulation F10 was found to be 82.46%, formulation F11 was found to be 98.52%, formulation F12 was found to be 94.27%.

ISSN: 0975-3583, 0976-2833 VOL 12, ISSUE 04, 2021

Scanning Electron Microscopy:SEM photographs were taken for the best formulation using a scanning electron microscope at specified magnification in room temperature.

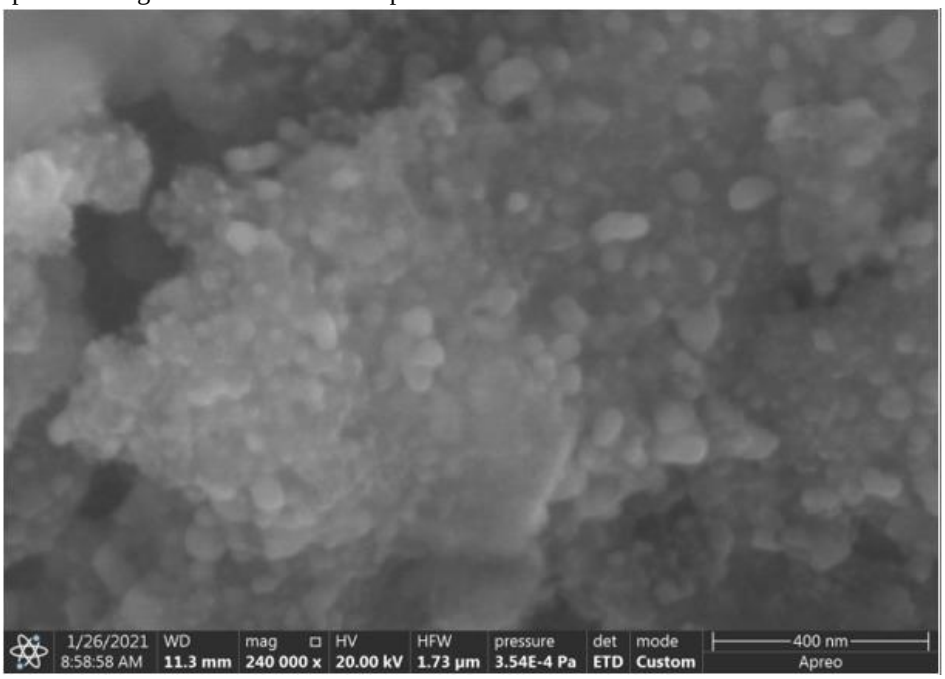

Figure No 6: Scanning Electron Microscopy ofBest Formulation

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.

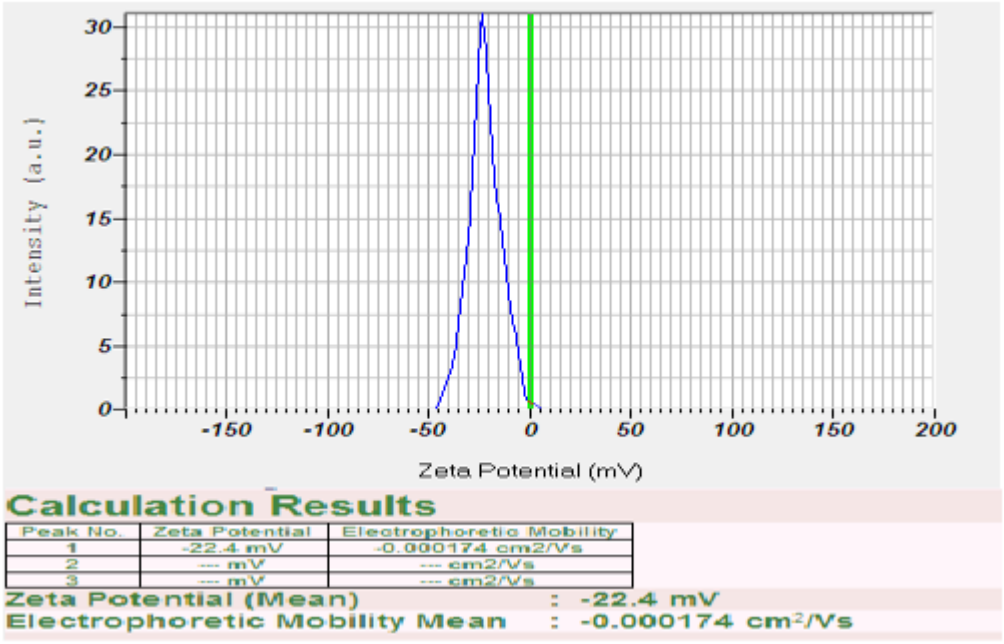

Figure:7: Zeta potential value forthe best formulation (F12)

ISSN: 0975-3583, 0976-2833 VOL 12, ISSUE 04, 2021

Discussion: The stability of colloidal dispersion depends on the zeta potential. Higher the zeta potential value indicates the greater charge on particle surface, resulting in repulsion of particles, reducing the aggregation of particles. Zeta potential value for the best formulation (F12) was found to be -22.4mV as shown in the above figure.

Particle size analysis:

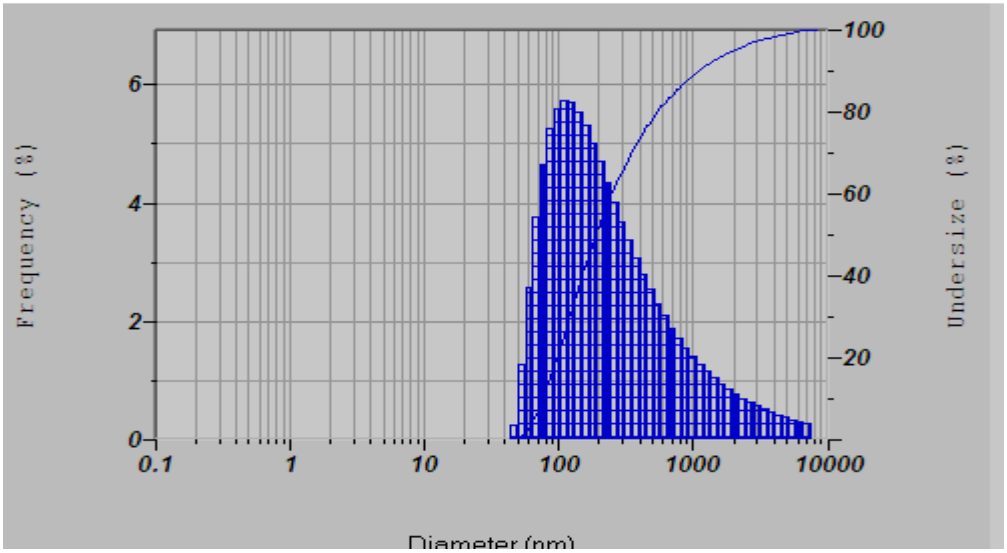

Figure 8: Particle Size Analysis of Best Formulation

Discussion: Averageparticlesizeof nanosuspensionof bestformulations (F12)wasfoundtobe having maximum particles at a range of 118 nm. Lower the particle size is suitable as it increases physical stability of nanosuspension. Lower the particle size increases the surface area of particle leading to increased drug dissolution.

Dissolution results:

Table 6: In-vitro drugrelease data of formulation F1 to F12

ISSN: 0975-3583, 0976-2833 VOL 12, ISSUE 04, 2021

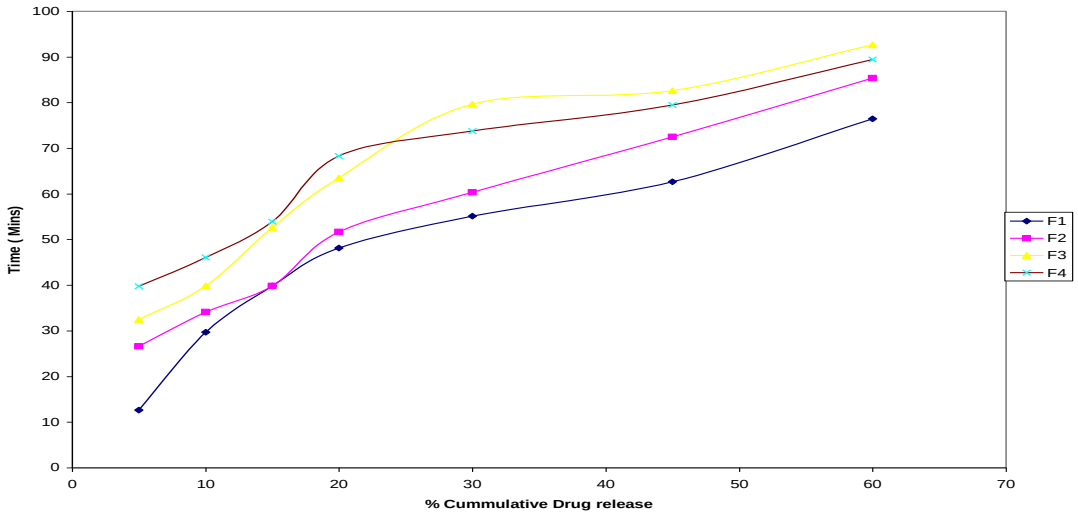

Figure9: Dissolution parameters forthe formulations F1-F4

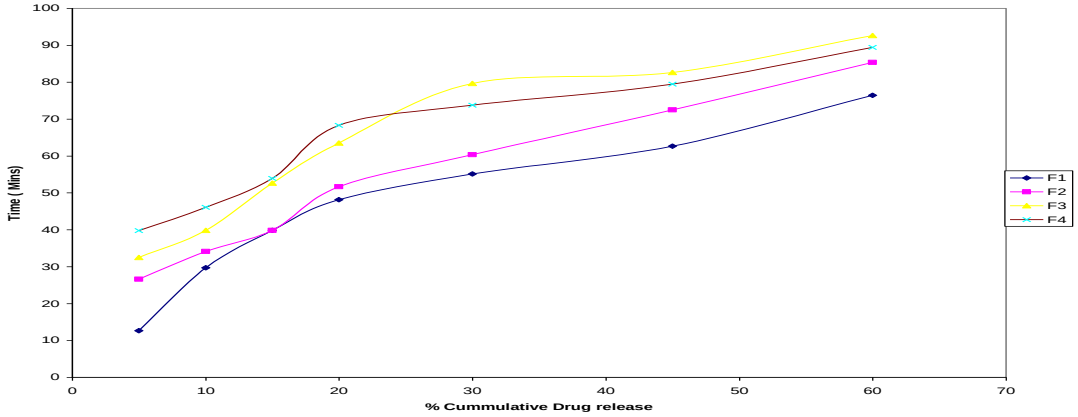

Figure 10: Dissolution parameters forthe formulations F5-F8

ISSN: 0975-3583, 0976-2833 VOL 12, ISSUE 04, 2021

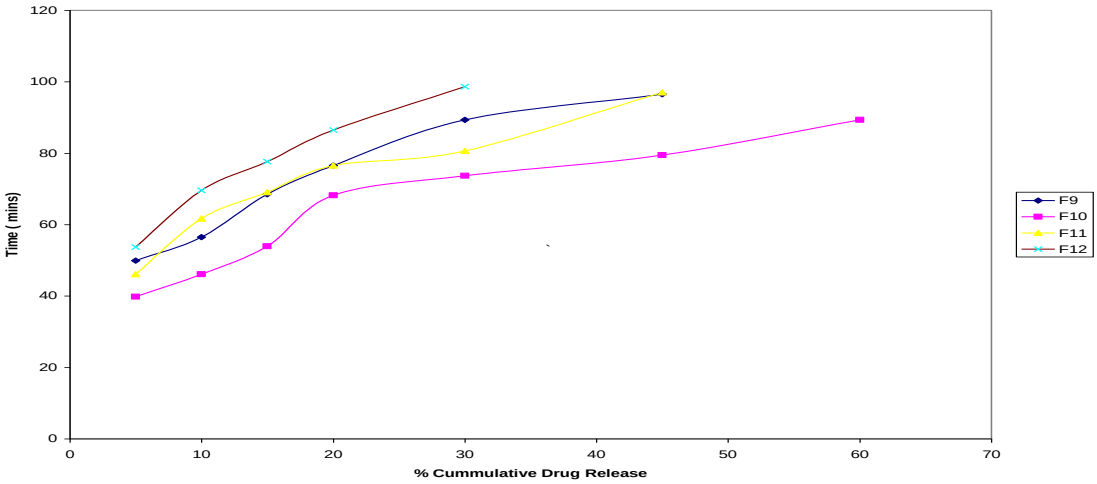

Figure 11: Dissolution parameters forthe formulations F9-F12

Discussion: From the above in-vitro studies we can say that increase in the polymer concentration of polymers decrease in the dissolution time of all the formulations.
From the above in-vitro studies we can say that at low polymer concentrations the drug release time was increased. SoF12 is considered as best formulation as it shows drug release with in 30mins.
Among all the four stabilizers we have used F12 containing Pluronic F127 at 1.0% concentration releases maximum drug release at the end of 30 minutes when compared to the formulations prepared by using PVP K30 and Urea.
Increase in the stabilizer concentration of Pluronic F127 shows 98.65% of drug release, so the formulations prepared by using Pluronic F127 releases more drug release at the end of 30mins than the other stabilizers.

Table 7: Kinetic data of the formulation F12

	of kinetics	**rder**	**rder**

Discussion: ssion
The drug release from the Nanosuspension was explained by using mathematical model equations such as zero order, first order, and equation methods. Based on the regression values it was concluded that the best formulation F12 follows first order kinetics, indicating concentration dependent drug release.

4. Summary and Conclusion:

In present investigation Nanosuspensions of Agomelatin was prepared by Emulsification solvent evaporation method. The nanosuspensions are novel promising target and controlled released dosage form which is gaining importance because of ease of manufacturing and diversified applications. The present trend of pharmaceutical research lies in the usage of biodegradable polymer because of its availability and low toxicity. Nanosuspensioncontaining drugwas preparedby emulsification solvent evaporation method byusingcombinationsofUrea, Pluronic F127, PVP-K30, SLS, methanol and quantity sufficient water. EstimationofAgomelatin was carried out spectrophotometrically at 275 nm. The Nanosuspension were evaluated for parameters such as drug content uniformity, scanning electron microscopy, particle size analysis, zeta potential, in-vitro release, drugexcipientinteractions (FTIR). Thestabilitydata was also subjected to statistical analysis.The melting point of Agomelatin was found to be in range of 108°C which was determinedbycapillarymethod. Saturation solubility was carried out at 25^0C using 0.1N HCL, 6.8 phosphate buffer, 7.4 pH buffer, methanol & ethanol.Form the drug excipient compatibility studies we observe that there are no interactions between the pure drug (Agomelatin) and best formulation (Agomelatin+ excipient) which indicates there are no physical changes.The entrapmentefficacy of formulation F1 was found to be 83.23%, formulation F2 was found to be 85.15%, formulation F3 was found to be 87.45%,

ISSN: 0975-3583, 0976-2833 VOL 12, ISSUE 04, 2021

formulation F4 was found to be 89.63%, formulation F5 was found to be 88.23%, formulation F6 was found to be 97.14%, formulation F7 was found to be 93.26%, formulation F8 was found to be 95.15%, formulation F9 was found to be 97.26%, formulation F10 was found to be 82.46%, formulation F11 was found to be 98.52%, formulation F12 was found to be 94.27%.Zeta potential value forthe best formulation (F12) was found to -7mv which was found to be -22.4mV within theacceptable limits. Averageparticlesizeof nanosuspensionof bestformulations (F12)wasfoundtobe118nm.From the in-vitro studies we can say that formulation F12 shows best drug release of 98.65% within 30 minutes when compared with other formulations.The drug release from the Nanosuspension was explained by the using mathematical model equations such as zero order, first order, and equation methods. Based on the regression values it was concluded that the best formulation F12follows first order kinetics.

5. References

1. Debashishborah, shivanand kalyanappa and shantha kumar gs, formulation and evaluation of nanosuspension of lercanidipine hcl.world j pharm sci. 2015; 3(2): 308-317.
2. Jegan Sakthivel Nadar et al., Potentiation of Antidepressant Effects of Agomelatin and Bupropion by Hesperidine in Mice. Neurology Research International. Vol 2018. 7 pages.
3. Mahesh Shinde, Pramod Salve, Shahadev Rathod. Development and Evaluation of Nanoparticles Based Transdermal Patch of Agomelatin for the Treatment of Depression. Journal of Drug Delivery & Therapeutics. 2019; 9(4-s):126-144.
4. Ahmed M Fatouh, Ahmed H Elshafeey, Ahmed Abdelbary. Intranasal agomelatin solid lipid nanoparticles to enhance brain delivery: formulation, optimization and in vivo pharmacokinetics. Drug Des Devel Ther. 2017; 11: 1815–1825.
5. Chetan S. Urade, Sunil M. Mahakalkar, Prashant G. Tiple. A comparative study of the clinical efficacy and safety of agomelatin with escitalopram in major depressive disorder patients: A randomized, parallel-group, phase IV study. Journal of Pharmacology and Pharmacotherapeutics. 2015; 6[4].
6. Srikrishna Nukala ,Ravi Babu Komaram , Jayasree Palla and Murthy KSN. Efficacy and Safety of Agomelatin in treatment of major depressive disorder - A pilot study. IJPSR, 2016; 7(8): 3517-3521.
7. Randa M. Zaki, Adel A. Ali SFEMAAAB. Formulation and In-Vitro Evaluation of Diacerein Loaded Niosomes. Int J Pharm pharmacuetical Sci 2014; 6(2): 515–21.
8. D, Pan H, He F, Wang X, Li J, Yang X, et al. Effect of particle size on oral absorption of carvedilol nanosuspensions: In vitro and in vivo evaluation. Int J Nanomedicine. 2015; 10: 6425–34.
9. Lai F, Pini E, Angioni G, Manca ML, Perricci J, Sinico C, et al. Nanocrystals as tool to improve piroxicam dissolution rate in novel orally disintegrating tablets. Eur J Pharm Biopharm. 2011; 79(3): 552–8.
10. Kakran M, Shegokar R, Sahoo NG, Shaal L Al, Li L, Müller RH. Fabrication of quercetin nanocrystals: comparison of different methods. Eur J Pharm Biopharm. 2012; 80(1):113–21.
11. Li W, Yang Y, Tian Y, Xu X, Chen Y, Mu L, et al. Preparation and in vitro/in vivo evaluation of revaprazan hydrochloride nanosuspension. Int J Pharm. 2011; 408(1–2): 157–62.
12. Pardeike J, Strohmeier DM, Schrödl N, Voura C, Gruber M, Khinast JG, et al. Nanosuspensions as advanced printing ink for accurate dosing of poorly soluble drugs in personalized medicines. Int J Pharm. 2011; 420(1): 93–100.
13. Jawahar, N., Eagappanath, E., Venkatesh, N., Jubie, S., Samanta, M.K. 2009. Preparation and characterisation of PLGA-Nanoparticles containing an anti-hypertensive agent. Int J Pharm Tech Res. 1(2): 390-93.
14. Kalpesh, S.W., Patil, S.K. Nanosuspension - a new approach of bioavailability enhancement. International Journal of Pharmaceutical Sciences Review and Research. 2011; 8(2): 61-5.
15. Mukherjee, S., Ray, S., Thakur, R.S. Solid lipid nanoparticles: A modern formulation approach in drug delivery system. Indian J Pharm Sci. 2009; 4: 349-58.
16. Müller RH, Peters K. Nanosuspensions for the formulation of poorly soluble drugs: I. Preparation by a size-reduction technique. Int. j. pharm. 1998;229–237.
17. Ning Z, et al. Pharmacokinetics of cyadox and its major metabolites in swine after intravenous and oral administration. J. Integ. Agri. 2013; 12: 495–501.
18. Sattar A, et al. Pharmacokinetics and Metabolism of Cyadox and Its Main Metabolites in Beagle Dogs Following Oral, Intramuscular, and Intravenous Administration. Front. pharmacol. 2016; 7: 236.
19. Cutler D. Theory of the mean absorption time, an adjunct to conventional bioavailability studies. J.Pharm. Pharmacol. 1978; 30: 476–478.
20. S. S. Vasava, N. P. Chotai and H. K. Patel. Formulation and Evaluation of Nanosuspension drug delivery system of Etoricoxib. Pharma Science Monitor. 6(1); 2015: 10-27.
21. Sundar V.D, Divya P, Sridevi P, Akhila K, Dhanaraju, M. D. Design, Formulation and Evaluation of Nanosuspension for drug delivery of Celexocib. International Journal of Pharmaceutical Research. 2019; 11[1].
22. Suvarna S. Vadje , Rajendra K. Surawase, Santosh S. Surana. Formulation and Evaluation of Nanosuspension Drug

Delivery System of Furosemide Produced by Nanoprecipitation Method. Int. J. Pharm. Sci. 2020; 65(2): 50-55.

23. Dr. Yasmin Begum, M., Saisree, R., Harshitha, P., Shwetha, A. and Dr. Sudhakar, M. Preparation and Evaluation of Nanosuspension of Nifedipine. international Journal of Current Research. 2017; 9[9]: 57091-57098.
24. Kiran G. Sonkambale, Dr. Ramesh G. Katedeshmukh, Amol B. Kumbhar, Shivraj V. Mane, Prashant A. Pawar, Jyoti N. Kadam, Anita V. Malusare. Formulation and evaluation of nanosuspension for enhancing the solubility of poorly soluble Antihyperlipidemic Drugs. 2021; 8[3].
25. Omkar A. PATIL, Indrajeet S. PATIL , Rahul U. MANE , Dheeraj S.RANDIVE, Mangesh A. BHUTKAR , Somnath D. BHINGE. Formulation optimization and evaluation of Cefdinir nanosuspension using 2^3 Factorial design. Marmara Pharm J. 2018; 22(4): 587-598.
26. M. Santhosh Raja, K. Venkataramana. Formulation and Evaluation of Stabilized Glipizide Nanosuspension prepared by Precipitation Method. Research J. Pharm. and Tech. 2020; 13(11): 5145-5150.
27. Mangesh Ramesh Bhalekar, Prashant Girish Upadhaya, Satish Reddy, Sanjay J. Kshirsagar, Ashwini Raghvendra Madgulkar. Formulation and evaluation of acyclovir nanosuspension for enhancement of oral bioavailability. Asian Journal of Pharmaceutics. 2014.
28. Harekrishna Roy, S Balaiah, Vinay Kumar T. Design and in-vitro evaluation of cefuroxime axetil floating microbeads for the treatment of acute bacterial caused chronic bronchitis. Research Journal of Pharmacy and Technology 2018; 11(6): 2276-2282.
29. Vinayak Shivamurthi Mastiholimath, Bhagat Ankita Rajendra, Vinodkumar Shirshalappa Mannur, Panchaxari Mallappa Dandagi, Anand Panchaxari Gadad, Pukar Khanal. Formulation and Evaluation of Cefixime Nanosuspension for the Enhancement of Oral Bioavailability by Solvent-Antisolvent Method and its Suitable Method Development. Indian Journal of Pharmaceutical Education and Research. 2020; 54[1].
30. Roy H, Nayak BS, Nandi S. In-Silico Factorial Screening and Optimization of Chitosan Based Gel for Urapidil Loaded Microparticle Using Reduced Factorial Design. Combinatorial Chemistry & High Throughput Screening. 2020; 23(10): 1049-1063.
31. Roy H, Nayak BS, Nandi S. Chitosan Anchored Nanoparticles in Current Drug Development Utilizing Computer-Aided Pharmacokinetic Modeling: Case Studies for Target Specific Cancer Treatment and Future Prospective. Current Pharmaceutical Design. 2020,26(15): 1666-1675.
32. Gaikwad D. D., Bankar Pavan V., Miss Gandhi Anuja S. Dr. Jadhav S. J., Dr. Gadhave M. V. Formulation and Evaluation of Nanosuspension of Antidiabetic Drug. Ijppr.Human. 2016; 8 (1): 256-268.
33. Harshil M Patel, Urvashi B. Patel, Chainesh Shah and Bhavesh Akbari. Formulation and Development of Nanosuspension as an Alternative Approach for Solubility and Dissolution Enhancement of Aceclofenac. IJAP. 2018; 7[5].

ISSN: 0975-3583, 0976-2833 VOL 12, ISSUE 04, 2021