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Formulation and Evaluation of Cefpodoxime Proxetil Fast Dissolving Film

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ABSTRACT

The purpose of this research work was to formulate fast dissolving film of cefpodoxime proxetil for oral delivery in order to improve oral bioavailability of drug with poor solubility. Cefpodoxime proxetil (CP) is the drug candidate belonging to BCS class IV with poor solubility and poor permeability is and limited oral bioavailability, an orally administered, extended spectrum, semi-synthetic β- lactum antibiotic of cephalosporin class. To improve oral bioavailability, cefpodoxime proxetil nanosuspension was prepared using solvent-antisolvent precipitation technique. Nanosuspension was characterized on the basis of drug concentration in organic phase, temperature, solvent-antisolvent ratio and the time period of stirring on the particle size systematically. Particle size and zeta potential of nanosuspension was observed at 755.6nm and −22.6mV, respectively. Solvent casting method be used in the formation of film, utilizing HPMC E50 as film former, PEG 400 as plasticizer and tween 80 as surfactant. The optimized fast dissolving film formulation F1 showed uniformity of weight (0.091mg), folding endurance (149) drug content uniformity (99.5%), surface pH (6.8) disintegration time (32 seconds in 6.8 PB) and *in- vitro* drug release 94.2% in 7 min. So, it is concluded from comparison studies between fast dissolving film (FDF) containing pure drug and nanosuspension, fast dissolving film containing cefpodoxime proxetil nanosuspension gives faster and high drug release.

Keywords: Nanosuspension, Bioavailability, Solubility, Fast Dissolving Film.

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INTRODUCTION

Oral route is the most convenient and commonly employed route for drug delivery due to its ease of administration, high patient compliance, cost effectiveness, least sterility constraints, and flexibility in the design of dosage form. About 50% of the drug compounds are facing the major challenge with the design of oral dosage forms that lies with their poor bioavailability. It depends on several factors including aqueous solubility, drug permeability, dissolution rate, first-pass metabolism, pre-systemic metabolism, and susceptibility to efflux mechanisms. Poor bioavailability in oral route has the consequence of more variability and poor controlled plasma concentration and drug effects to the patients. In recent years, much attention has been focused on nano technology for delivering of formulations, which is being applied to enhance the solubility bioavailability of lipophilic drugs[1]. The nano sizing of drugs has the potential to increase surface area therefore, enhance solubility, increase rate of dissolution, increase oral bioavailability, more rapid onset of therapeutic action[2]. For the effective formulation and to sort out the drug solubility related problem, one should have to be very familiar with the Biopharmaceutical Classification System (BCS). The introduction of the Biopharmaceutical Classification System (BCS) in FDA guidelines represents a major step forward in the regulation of oral drug products. The BCS groups poorly soluble compounds as Class II drugs features poor solubility, high permeability whereas class IV drugs features poor solubility and poor permeability. Class I drugs do not pose any problem in absorption (though its systemic availability may be low due to first pass metabolism) when solubility or permeability are considered, therefore efforts are made to change the properties of Class II, III, IV drugs with respect to dissolution and permeability in order to resemble Class I[3]. A pharmaceutical nanosuspension is defined as "finely divided biphasic colloidal dispersions of nano size drug particles which are stabilized by surfactants materials". The particle-size distribution of the solid particles in nanosuspensions is usually less than 1μm[4]. A surprisingly large proportion of new drug candidates emerging from drug discovery programmes are water insoluble, and therefore poorly bio-available, leading to abandoned development efforts. As per a recent report, 46% of the total New Drug Applications (NDA) filed between 1995 and 2002 were BCS class IV, while only 9% were BCS class I drugs, revealing that a majority of the approved new drugs were water insoluble. Cefpodoxime proxetil (CP) is the drug candidate belonging to class IV category is an orally administered, extended spectrum, semi-synthetic β- lactum antibiotic of cephalosporin class[5].

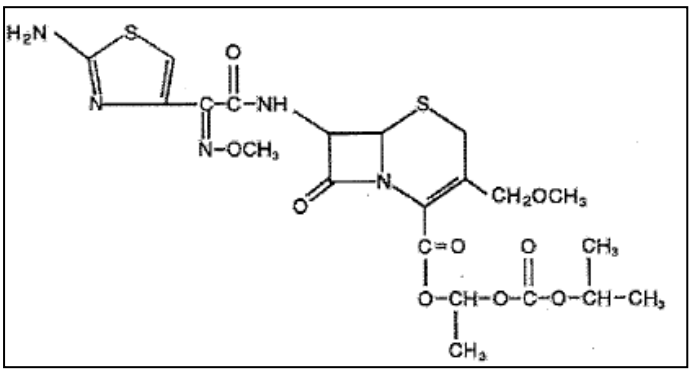

Figure 1: Structure of cefpodoxime proxetil[6]

Cefpodoxime proxetil is prodrug, its active metabolite is cefpodoxime. It is used in various conditions such as for the treatment of bacterial infection, like urinary tract infection, gonorrhoea, skin infection and upper and lower tract infection. It is active in vitro and in vivo against a wide range of Gram - positive and Gram - negative organism, including *Staphylococci ,Streptococci, Heamophillus, Influenza, Neisseria gonorrhoeae Escherichia coli, Klebsiella ,Pheumoniae.* Oral cefpodoxime proxetil treats bacterial infection by binding to penicillilin- binding proteins thereby causing abnormal bacterial cell wall synthesis and lysis[7]. After oral administration, cefpodoxime proxetil is absorbed from the gastrointestinal tract and de-esterifies to active metabolite cefpodoxime. Over the recommended dosing for adult, the usual dose is 100 mg (tablet or suspension) administered orally twice. 50% administered cefpodoxime dose was absorbed systemically. Its action is by binding to specific penicillin-binding proteins (PBPs) located inside the bacterial cell wall, it inhibits the bacterial cell wall synthesis. It is highly stable in the presence of beta - lactamase enzymes. Since it belongs to class IV it has poor solubility and poor permeability in water but can be improved by using novel technology i.e nanosuspension technology based on particle engineering. The elimination half-life of cefpodoxime proxetil is 2–2.5 hrs and its molecular weight is 557.59 g/mol which make it a suitable drug candidate for administering in the form of nanosuspension. It is useful for molecules with poor solubility, poor permeability or both, which poses a significant challenge for the formulators. The reduced particle size renders the possibility of intravenous administration of poorly soluble drugs without blockade of the blood capillaries. Oral route is most preferred route by medical practitioners and manufacturer for the administration of antibiotics due to highest acceptability of patients. About 60% of all dosage forms available are the tablets and capsules. Recently, fast dissolving films are gaining interest as an alternative novel drug delivery system designed for meeting current needs of the industry and consumer choice with respect to patient compliance. These films are designed to dissolve upon contact with a wet surface, such as the tongue, within a few seconds and the drug is

directly absorbed into systemic circulation, degradation in gastrointestinal tract and first pass effect can be avoided leading to increased bioavailability[8]. They provide quick onset of action within few seconds as the oral mucosal absorption of drug occurs directly from the site of administration to the systemic circulation to produce the desired effect[9]. So, the aim of the present study was to formulate cefpodoxime proxetil loaded nanosuspension in fast dissolving film for oral delivery system used for the treatments of mild to moderate respiratory tract infection, uncomplicated gonorrhea and urinary tract infection.

MATERIALS AND METHOD

Cefpodoxime proxetil was obtained as a gift sample from Uni Speed Pharmaceuticals Pvt. Ltd. Baddi, India. Acetone, DMSO, was purchased from Qualigens Ltd Mumbai, India. Ethanol, Methanol was sourced from Changshu Yangyuan Chemical, China. Hexane, PEG 800, Hydrochloric acid, Potassium Dihydrogen Phosphate was purchased from Qualikems Fine Chem Pvt. Ltd. Oleic acid, Tween 80, Lactic acid were purchased from Molychem, India. PEG 400 was obtained from S.D. Fine Chem. Pvt Ltd. Glycerin, Ortho phosphoric acid, Triethylamine were sourced from Thomas Baker. Hydroxypropyl methylcellulose (E50) was obtained from Colorcon Asia Pvt. Ltd. Sodium hydroxide was obtained from Avarice Laboratories Pvt. Ltd. All chemicals and solvents were of analytical grade. Freshly double distilled water was used in the experiments.

Determination of organoleptic properties

The physical identification of cefpodoxime proxetil was done by checking its physical appearance i.e. colour, odour, taste and state. Weight quantity of cefpodoxime proxetil as drug was taken and viewed in well illuminated place. Very less quantity of drug was smelled to get the odour.

Determination of Melting point

Melting point of the drug was determined by using capillary method. Drug was filled into capillary tube by sealing its one end at the height of 3 mm from the closed end. The capillary was introduced into the digital melting point apparatus and the point at which the drug starts melting was noted until the entire samples get melted.

Identification of drug by FTIR and UV- Visible spectroscopy

Fourier transforms infrared spectral spectroscopy (FTIR): The pure drug was mixed with IR grade potassium bromide in a ratio of (1:100) and pellets were prepared by applying 10 metric ton of pressure in shimadzu hydrophilic press. The pellets were then scanned over range of 4000-400 cm^{-1} in FTIR spectrometer. FTIR spectrum of cefpodoxime proxetil showed the presence of the peaks which complies with the reference spectra.

UV- Visible spectroscopy: 100 mg of cefpodoxime proxetil was weighed and transferred to 100 ml volumetric flask. Drug was dissolved in 10 ml methanol and sonicated for 5 min. Final volume was made up to the mark with same solvent and strength of 1000 µg/ml was obtained. Further dilution was made with methanol to get 100 µg/ml solutions and scanned under 200 nm to 400 nm in UV-Visible Spectrophotometer[10].

Preparation of standard calibration curve of cefpodoxime proxetil

Preparation of stock solutions of cefpodoxime proxetil in methanol:

100 mg of cefpodoxime proxetil was weighed and transferred to 100 ml volumetric flask. Drug was dissolved in 100 ml methanol and sonicated for 5 min. Final volume was made up to the mark with same solvent and strength of 1000 µg/ml was obtained. From the above solution 10 ml of solution was transferred in 100 ml volumetric flask and volume was made up to 100 ml with methanol to prepare stock solution of 100µg/ml.

Preparation of serial dilutions

From the standard stock solution, a series of dilutions 2, 4, 6, 8, 10, 12, 14, 16, 18 and 20µg/ml were prepared by taking 0.2, 0.4, 0.6, 0.8, 1.0, 1.2, 1.4, 1.6, 1.8 and 2.0 ml of solution and was transferred into 10 ml volumetric flasks and volume was made up to 10 ml with methanol and absorbance was taken at 235 nm.

Preparation of stock solutions of cefpodoxime proxetil in phosphate buffer pH 6.8:

100 mg of cefpodoxime proxetil was weighed and transferred to 100 ml volumetric flask. 100 ml phosphate buffer was added and sonicated for 2 hrs. The volume was made up to the mark with phosphate buffer and the final strength obtained was 1000 µg/ml. From the above solution ,10 ml solution was transferred in 100 ml volumetric flask and volume was made up to 100 ml with to phosphate buffer prepare stock solution of 100µg/ml.

Preparation of serial dilutions

From the standard stock solution, a series of dilutions 2, 4, 6, 8, 10, 12, 14, 16, 18 and 20µg/ml were prepared by taking 0.2, 0.4, 0.6, 0.8, 1.0, 1.2, 1.4, 1.6, 1.8 and 2.0 ml of solution and was transferred into 10 ml volumetric flasks and volume was made up to 10 ml with phosphate buffer and absorbance was taken at 235 nm.

Determination of solubility of Cefpodoxime proxetil in different solvents:

Solubility studies were carried out in different solvents and surfactant like purified Water, Methanol, Ethanol, Acetone, Hexane, Tween 80, PEG 400, PEG 800, DMSO, Lactic acid, Oleic acid, 6.8 phosphate buffer and 0.1 N HCl. Excess amount of drug was added to each vial containing 2ml of solvents to saturate the solution. The drug solutions were shaken for 24 hrs in an

water bath shaker (Remi, Mumbai, India), maintaining the temperature 37±0.5°C and by providing shaking of 100 agitatation/min. Afterwards, solution were centrifuged at 10000 rpm for 15 min and then supernatant was filtered through membrane filter (0.22µm) to remove the remaining drug. The above samples of drug solution were taken and diluted suitably by methanol to observe the absorbance of drug by using UV-Visible spectrophotometer at λmax of 235 nm. The drug concentration was calculated with the help of standard calibration curve of drug in methanol and the graph was plotted between the concentrations vs absorbance[11].

Determination of partition coefficient:

Partition coefficient was determined by taking excess amount of cefpodoxime proxetil in 10 ml mixture of *n*-octanol and water (1:1) in a separating funnel. This system was shaken intermittently for 30 mins and kept undisturbed for overnight to achieve equilibrium. Then the two phases were separated and centrifuge at 10000 rpm for 15 minutes. After centrifugation, the concentration of cefpodoxime proxetil in both phases was determined by measuring the absorbance at 235 nm on UV-Visible spectrophotometer.

The partition coefficient is commonly determined by shake flask method and calculated by formula:

$$P\ (O/W) = C\ (oil)/\ C\ (water)$$

Where, C (oil) = Conc. of solute in organic phase.

C (water) = Conc. of solute in aqueous phase.

P ($^{O}/_{W}$) = Partition coefficient

Log P = log (O/W)

Determination of drug-excipients compatibility study

Drug and excipient compatibility studies were conducted to determine the compatibility of the excipients with the drug for the preparation of formulation. The FTIR spectrum was recorded by using FTIR after preparing potassium bromide disk. The finely ground drug powder and excipients powder were mixed with powdered potassium bromide and the mixture was pressed with a specific hydraulic compression. The prepared KBr pellet was then observed under Fourier transform infrared spectrometer (FTIR) and the spectrum of drug and excipients was recorded and compared.

Formulation Development

Preparation of Nanosuspension by Solvent Anti-Solvent Precipitation Method

Weighed amount of drug was taken and dissolved in solvent (ethanol/acetone) at room temperature to form a drug solution. HPMC E50 as stabilizing agent was first dissolved in small quantity of hot anti-solvent *i.e.* deionized water. Drug solution filled in syringe was quickly injected at a fixed flow rate (2-8 ml/min) into anti-solvent kept at a low temperature (4^0C) in an iced water bath.

During injection, the mixture was stirred continuously at 1000 rpm for 15 minutes. After precipitation, mixture was kept for 24 hrs for evaporation of volatile solvents. Final cefpodoxime proxetil nano particles were collected, filtered and dried at 40^0C in hot air oven[12].

Optimization of cefpodoxime proxetil Nanosuspension

Various formulations were prepared by using solvent anti-solvent precipitation method. Different concentration of drug (mg/ml) and solvent anti-solvent volume ratio was optimized.

Table 1: Optimization of drug loaded nanosuspension on the basis of different solvent anti-solvent ratio

Formulation code	Solvent	Anti-solvent	Drug (mg)	Solvent anti-solvent volume ratio (ml)	Stabilizing agent (HPMC E50) %(w/v)
K1	Acetone	water	40	1:10	0.1
K2	Acetone	water	40	1:20	0.1
K3	Acetone	water	40	1:30	0.1
K4	Acetone	water	40	1:40	0.1
K5	Acetone	water	40	1:50	0.1
K6	Acetone	water	40	1:60	0.1
K7	Ethanol	water	40	1:10	0.1
K8	Ethanol	water	40	1:20	0.1
K9	Ethanol	water	40	1:30	0.1
K10	Ethanol	water	40	1:40	0.1
K11	Ethanol	water	40	1:50	0.1
K12	Ethanol	water	40	1:60	0.1

Formulation K8 was selected on the basis of percentage yield (% yield) of nanosuspension formation by solvent-antisolvent precipitation method. Percentage yield was calculated on the basis of following formula

$$Percentage\ yeild = \frac{Practical\ yield}{Theoretical\ yield} \times 100$$

Table 2: Optimization of drug loaded nanosuspension on the basis of different drug concentration

Formulation code	Solvent	Anti-solvent	Drug(mg)	Solvent anti-solvent volume ratio
CK1	Ethanol	water	40	1:20
CK2	Ethanol	water	50	1:20
CK3	Ethanol	water	60	1:20
CK4	Ethanol	water	90	1:20

Formulation CK2 was selected having same solvent anti-solvent ratio but with different drug concentration on the basis of their saturation solubility in different solvents.

Determination of saturation solubility of cefpodoxime proxetil nanosuspension formulation

2 mg of cefpodoxime proxetil loaded nanosuspension was suspended in 2ml of water, 0.1N HCl and pH 6.8 phosphate buffers and shaken at 37°C for 24 hrs. From this, nanosuspension was taken into centrifugation tube and centrifuged at 10,000 rpm for 15 mins. The sample was filtered through 0.22 µm membrane filter and the filtrate was diluted appropriately and was analyzed spectrophotometrically using UV-Visible spectrophotometer at 235nm[13].

Evaluation of Optimized Cefpodoxime Proxetil Nanosuspension Formulation

Determination of particle size of cefpodoxime proxetil nanosuspension formulation

Particle size (in nanometers) of cefpodoxime proxetil nanosuspension was determined using a Beckman coulter instrument[14].

Determination of Zeta potential of cefpodoxime proxetil nanosuspension formulation

Zeta potential determines the physical stability of nanosuspenson. Measurement of zeta potential of the nanosuspension formulation was done by using a Malvern Nano Zeta Sizer instrument[15].

Determination of drug content of cefpodoxime proxetil nanosuspension formulation

The drug content was determined by calibration curve method[16]. 100 mg of nanosuspension was accurately weighed and dissolved into 100 ml of methanol followed by sonication and filtration through whatmann filter paper. The amount of drug was determined by UV-Visible spectrophotometer at 235 nm.

Preparation of Fast Dissolving Oral Film by Solvent Casting Method

Procedure for preparation of casting solution

HPMC E50 (water soluble polymer) was accurately weighed and soaked in distilled water for 24 hrs for the purpose of swelling. Glycerin and PEG 400 were mixed in polymer solution. Required quantity of loaded nanosuspension was dissolved in suitable solvent (1ml ethanol/ acetone) and incorporated into the above obtained solution. Both solutions were mixed thoroughly by mechanical stirring at the speed of 1000 rpm for 1 hrs and then sonicated for 30 mins to remove air bubbles from final solution.

Preparation of oral fast dissolving film

The casting solution was poured into glass petridish and dried at 40°C in a hot air oven for 24 hrs for solvent evaporation. The films were removed by peeling and cut into a square dimensions of 2×2cm ($4cm^2$). These films were kept in a desiccator for 2 days for further drying and wrapped in aluminium foil, and packed in self-sealing covers. Fast dissolving films were prepared by using different polymer concentrations while the concentration of the plasticizer, surfactant and sweetener was kept constant[17].

Calculation of dose of drug to be incorporated in fast dissolving films

Oral dose of cefpodoxime proxetil is 200 mg daily. In order to formulate a tablet and suspension of cefpodoxime proxetil, the total amount of drug to be incorporated would be 100 mg. Tablet or suspension given twice in daily total dose of drug is 200 mg. Cefpodoxime proxetil undergoes hepatic first pass metabolism following oral administration. Its oral bioavailability of is 50% since films are designed to dissolve upon contact with a wet surface, such as the tongue, within a few seconds and the drug is absorbed from oral cavity and directly reach into systemic circulation, degradation in gastrointestinal tract and first pass effect can be avoided leading to increased bioavailability. The dose incorporated would be one half of the oral conventional dose.

Therefore, the amount of cefpodoxime proxetil required for one day is

200×50/100 = 100 mg

Calculation of the amount of drug for circular cast film

Internal diameter of petridish = 9 cm.

Internal surface area of petridish = πr^2 = 3.14 x 4.5 x 4.5 = 63.585 cm^2

Diameter of fast dissolving film = 4cm

Area of fast dissolving film = πr^2

= 3.14 x 2 x 2

= 12.56 cm^2 [18].

Therefore the number of fast dissolving films from one circular cast film. = 63.585 /12.56 = 5.0 films

Since one film contained drug load = 20 mg.

Therefore one circular cast film drug load = 20 x 5

Therefore 100 mg of cefpodoxime proxetil is needed for one circular cast film.

Table 3: Amount of drug, polymer, plasticizer and other additives used in FDFF.

Components	Formulation F1	Formulation F2	Formulation F3
Nanosuspension loaded drug (mg)	100	100	100 (pure drug)
HPMC E50 (mg)	200	300	200
PEG 400 (ml)	0.2	0.2	0.2
Tween 80 (ml)	0.2	0.2	0.2
Glycerin (ml)	0.5	0.5	0.5
Distilled Water (ml)	10	10	10

Quantity of drug and weight was calculated as per the area of petridish, so that each film 2×2 cm^2 contained 20 mg of drug.

EVALUATION OF ORAL FAST DISSOLVING FILM

Appearance

The prepared films were inspected visually for clarity, colour and presence of any particle.

The test is important regarding patient compliance[19].

Surface pH

Surface pH of film was determined by digital pH meter, it is important as acidic or basic pH is liable to cause oral mucosal irritation. The pH value of a film was determined by putting the prepared film in a petri dish and moistened with 0.5 ml of distilled water and kept for 30s. The pH was noted by bringing the electrode of the pH meter in contact with the surface of the formulation and allowing equilibration for 1 min[17].

Weight uniformity

The weight uniformity was determined by digital weighing balance. For weight uniformity, each formulation taken was weighed individually on digital weighing balance and weight was calculated[20].

Folding endurance

Folding endurance value is number of times the film is folded without breaking. This also gives an indication about brittleness of the film. Film was subjected to folding endurance by folding the patch at the same place repeatedly for several times until a visible crack was observed and the values were reported[20].

Disintegration time

Disintegration apparatus mentioned in official pharmacopoeias is used for determining the disintegration time of a film. Normally, the disintegration time of film ranges from 5 to 30 s. Two methods are used for determining disintegration time of film i.e disintegration apparatus method and petridish method. A film is placed onto 2 ml distilled water taken in petridish. Time taken by the film to dissolve completely is considered as the disintegrating time[19].

Determination of drug content

The drug content was determined by flask shake method. A Fast dissolving film (2×2 cm^2) was transferred to a graduated flask containing 100 ml of methanol. The volumetric flask containing film was shaken for 2 hrs on mechanical shaker in order to get complete solubility of drug. This solution was filtered and absorbance was estimated by UV-visible spectrophotometer at wavelength 235 nm[21].

***In-vitro* dissolution studies**

The dissolution studies of immediate release films of cefpodoxime proxetil was carried out in a beaker containing 30 ml of the phosphate buffer pH 6.8 as a dissolution medium, maintained at 37 ± 0.5°C. The medium was stirred at 50 rpm. Aliquots 1 ml of the dissolution medium was withdrawn at 1, 2, 3, 4, 5, 6, and 7 mins time interval and the same amount was added with the fresh medium in order to maintain the sink conditions. Samples were assayed spectrophotometerically at 235 nm[20].

RESULTS AND DISCUSSION

Cefpodoxime proxetil (drug sample) was observed for organoleptic properties like physical appearance, odor, and melting point. The drug was identified with the help of UV and FTIR and exhibited absorption maxima at 235 nm when methanol was used as solvent as mentioned in literature.

Melting point analysis

The melting range of cefpodoxime proxetil was observed to be 111°C -112°C which complies with reported melting range *i.e.* 111°C -113°C.

Identification of drug by Fourier transforms infrared spectral spectroscopy

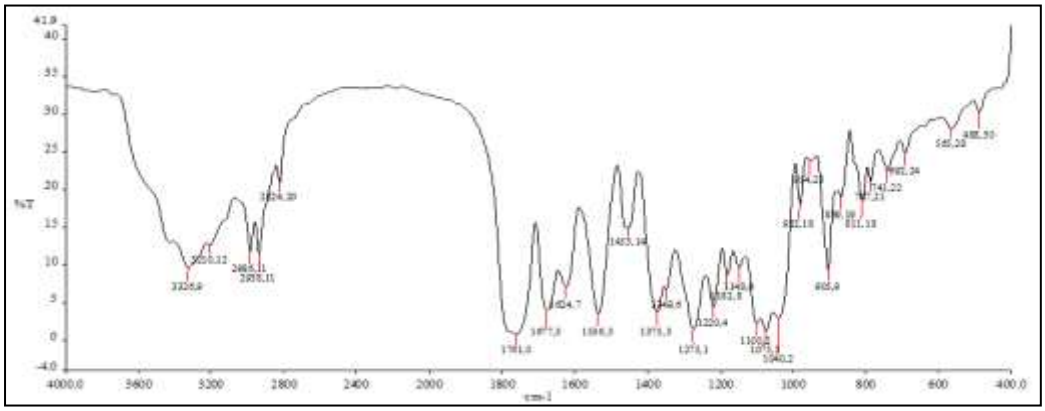

Figure 2: FTIR spectrum of cefpodoxime proxetil as pure drug

Table 4: Interpretation of infrared spectrum bands of cefpodoxime proxetil

Functional groups stretching and bending	Observed value(cm^{-1})
NH_2	3326, 3210
C-H	2986, 2938
C=O (ester)	1761
C=O (amide)	1677
C=N	1624
CH_3 bending	1453, 1375
C-O	1275
=C-H	906

The IR spectrum of drug sample have shown identical peaks as reported into reference sample of cefpodoxime proxetil.

Identification by UV- Visible spectroscopy

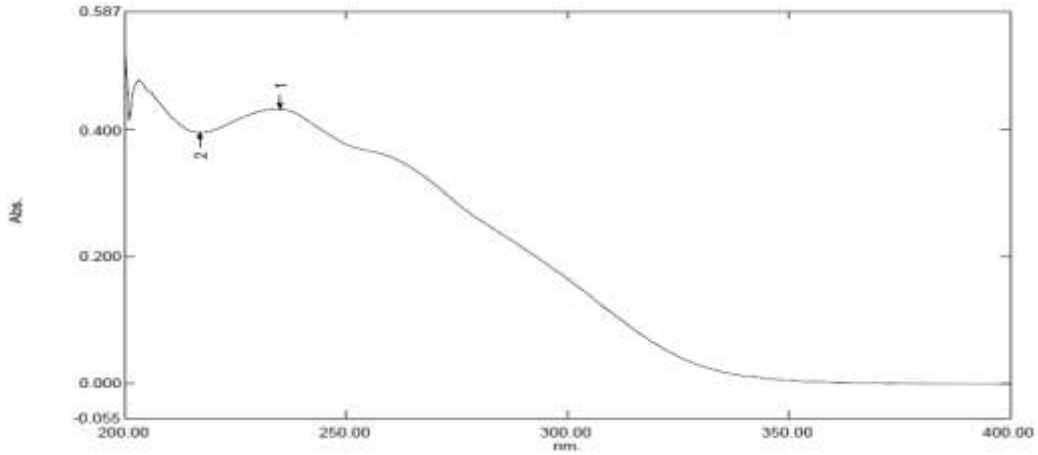

Figure 3: UV- Visible spectroscopy of cefpodoxime proxetil at 235nm

Standard calibration curve of cefpodoxime proxetil in methanol

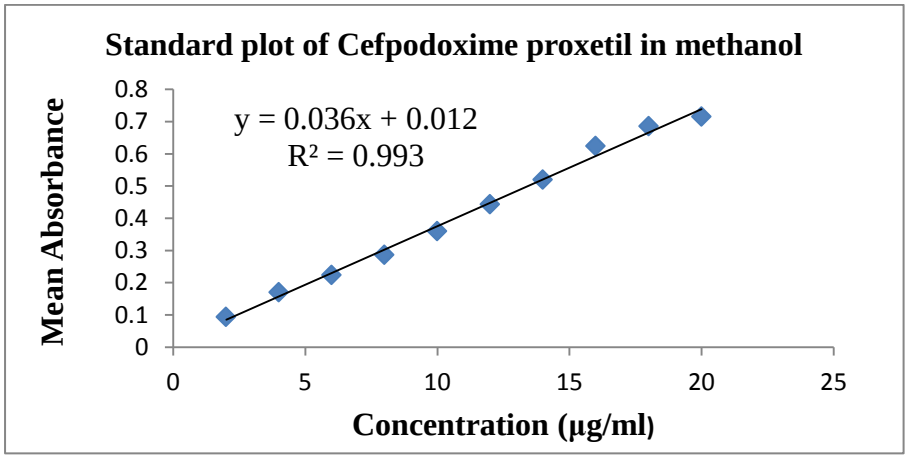

Figure 4: Standard plot curve of cefpodoxime proxetil in methanol at 235 nm

Preparation of standard curve of cefpodoxime proxetil in pH 6.8 phosphate buffer(PB)

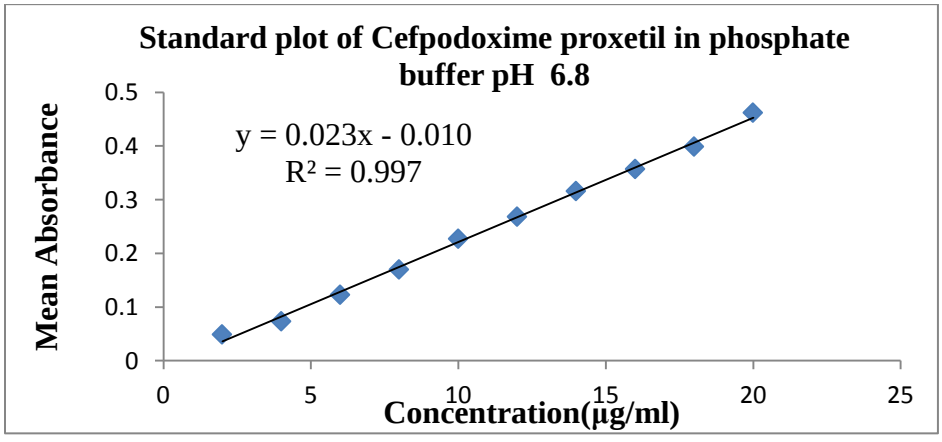

Figure 5: Standard curve of cefpodoxime proxetil in pH 6.8 PB at 235 nm

Determination of partition co-efficient

Table 5: Partition co-efficient of cefpodoxime proxetil in water and octanol

Solvent	Abs.	Conc. µg/ml	DF µg/ml	Conc. mg	Conc. mg/ml	partition co-efficient	Log P
Water	0.35	9.3888	938.888	0.9388	9.38888	18.284	1.2620
n-Octanol	0.63	17.166	17166.6	17.1666	171.666		

Solubility studies

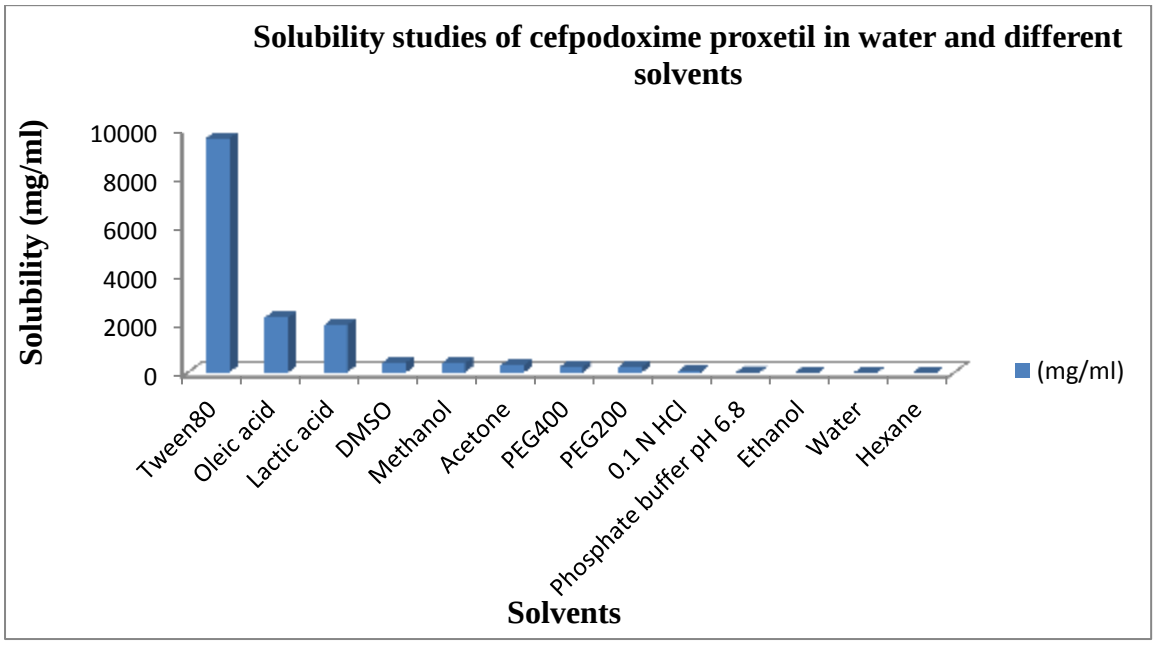

Figure 6: Solubility studies of Cefpodoxime proxetil in water and different solvents

Compatibility study of drug with polymer

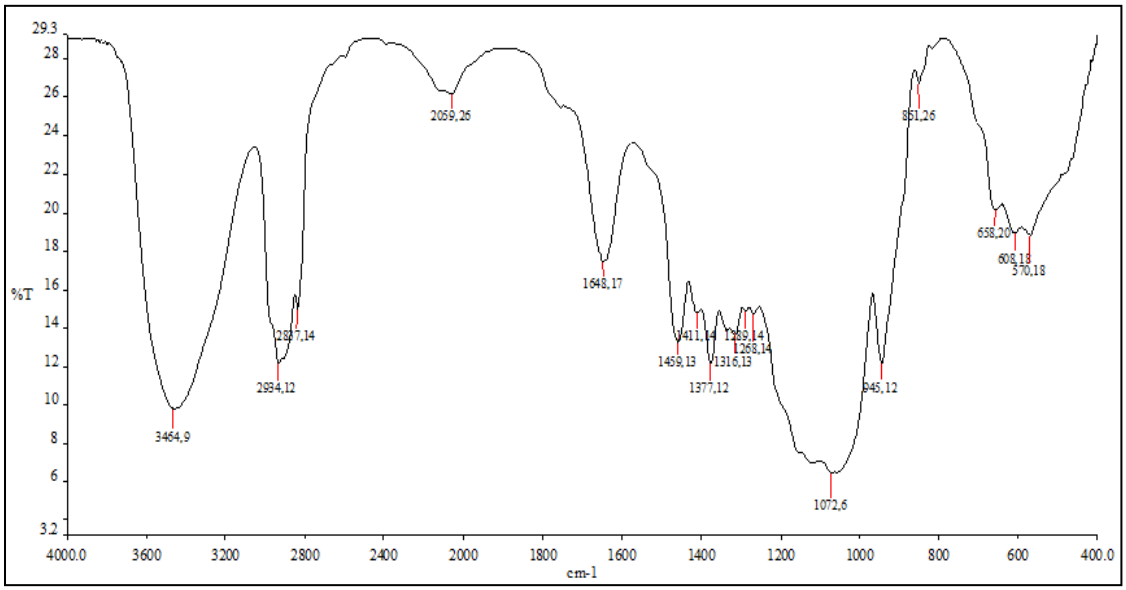

Figure 7: FTIR spectrum of HPMC E50

Table 6: Interpretation of infrared spectrum bands of HPMC E50

Functional groups stretching and bending	Observed value(cm^{-1})
O-H	3464
C-H	2934, 2837
CH_3	1459, 1377
C-O	1072

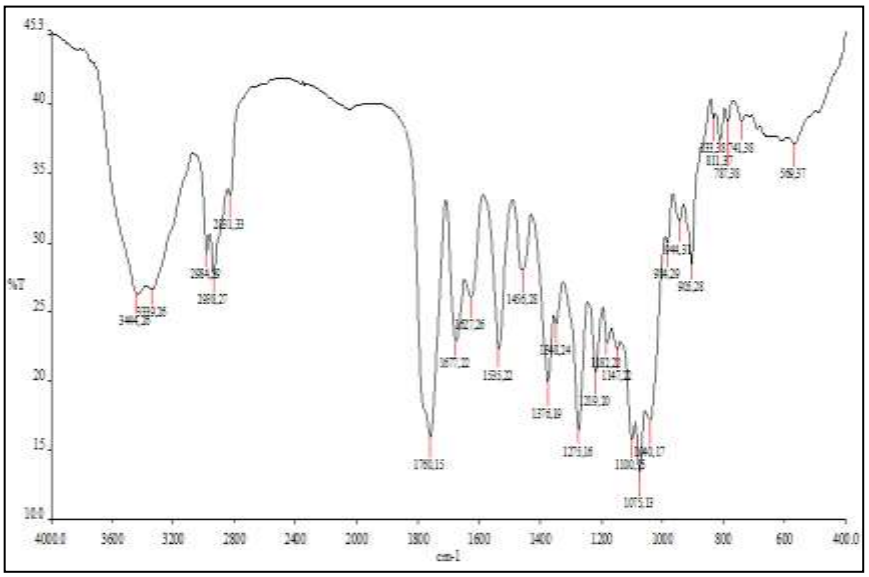

Figure: 8. FTIR spectrum of cefpodoxime proxetil and HPMC E50

Drug polymer interaction studies are carried out to eliminate the possibility of interaction between drug and polymer used with analytical method of drug estimation. The FTIR spectra of pure drug and pure polymer are shown in figure no.2, figure no.7 and Physical mixtures of drug with excipient (HPMC E50) are shown in figure no.8 respectively. The peaks observed in FTIR of mixture of cefpodoxime proxetil and excipients at 3339 cm^{-1}, 2884, 2938 cm^{-1}, 1760 cm^{-1} 1777 cm^{-1}, 1627 cm^{-1}, 1456, 1376 cm^{-1}, and 1275 cm^{-1}, 905 cm^{-1}. Drug shows characteristic peak and there be no significant changes in the position of the characteristic peak of drug when mixed with excipients which indicate compatibility of drug with polymer.

Formulation Development

Effect of solvent anti-solvent volume ratio

Percentage yield of different cefpodoxime proxetil nanosuspensions formulations.

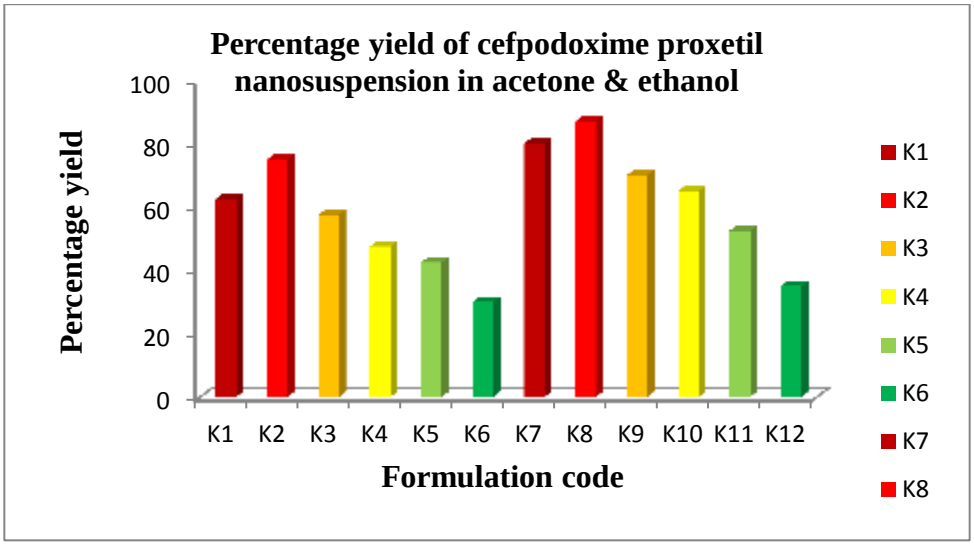

Figure 9: Percentage yield of different nanosuspension formulations

Formulation K7, K8, and K1 showed highest percentage yield as compared to other. The maximum percentage yield of nanosuspension formulation was found to be 87% for K8 formulation containing solvent anti-solvent in the ratio of 1:20. So, it was concluded that ethanol produce more precipitation as compared to acetone. Simultaneously on increasing solvent anti-solvent ratio percentage yield was reduced.

Effect of drug concentration

Table 7: Effect of drug concentration on solubility and percentage yield in nanosuspension formulation

Formulation code	Solvent anti-solvent volume ratio (ml)	Drug (mg)	Solubility study in methanol (mg/ml)			%Yield
			Water	0.1N HCl	6.8 buffer	
CK1	1:20	40	5	47	22	87
CK2	1:20	50	19	72	24	95
CK3	1:20	60	22	21	22	78
CK4	1:20	90	21	24	26	73

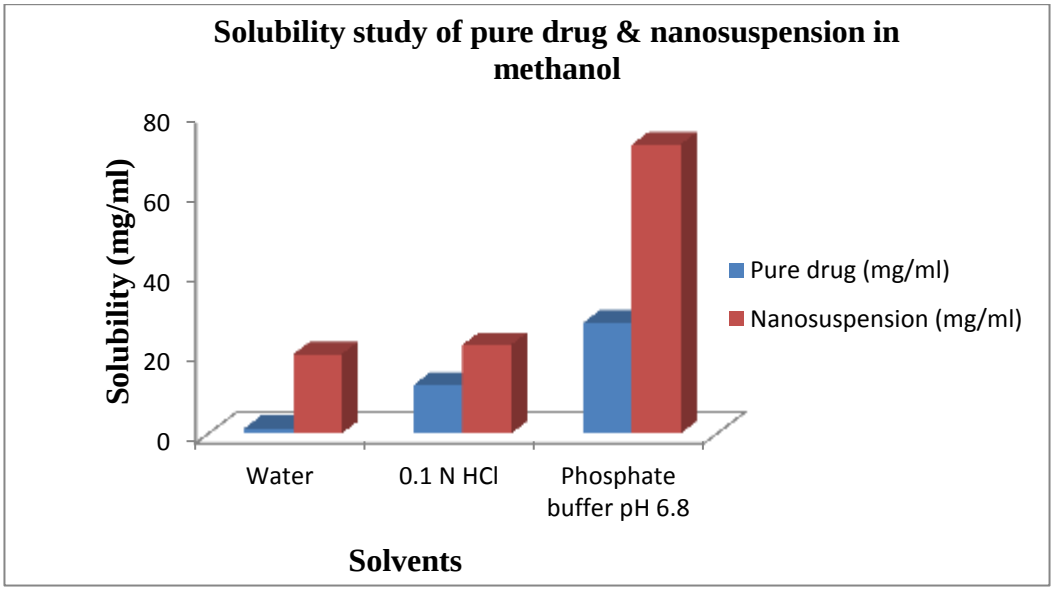

Figure 10: Solubility of Cefpodoxime proxetil loaded nanosuspension in methanol

An optimum solvent/antisolvent ratio was requisite to obtain the higher percentage yield. Use of a different drug concentration above optimal level increases percentage yield. But while the drug concentration is increased, the nucleation rate too increased owing to higher super saturation solubility in different solvent is decrease. From table 7 it was concluded that formulation CK2 was selected for final formulation.

Particle size

Particle size analysis of selected cefpodoxime proxetil nanosuspension formulation (CK2) was measured by Dynamic Light Scattering phenomenon using a Beckman coulter instrument.

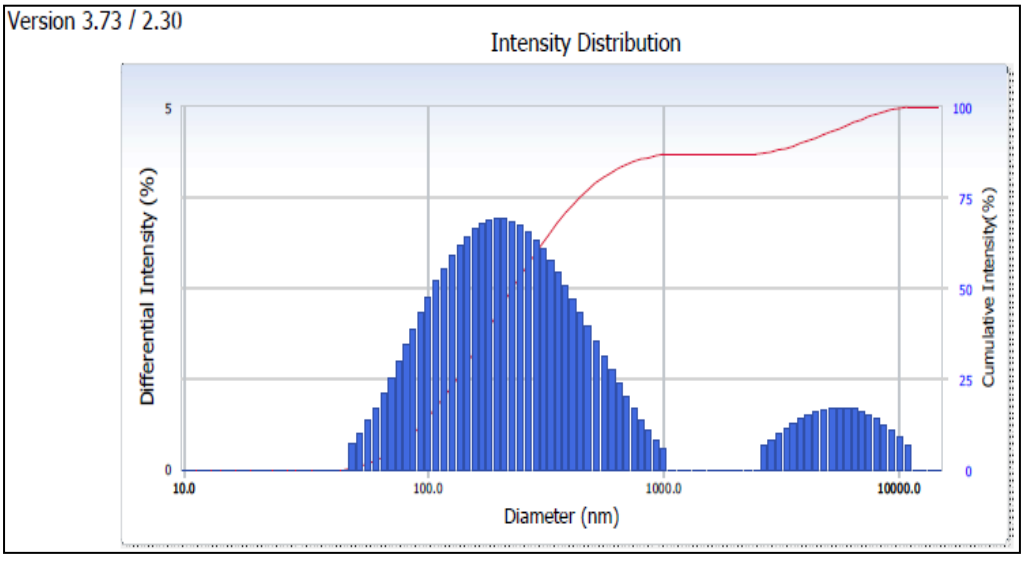

Figure 11: Particle size analysis of cefpodoxime proxetil nanosuspension formulation

Table 8: Particle size of cefpodoxime proxetil nanosuspension formulation by Beckman coulter Instrument

Formulation code	Particle size (nm)	Polydispersity index	Diffusion constant (cm^2/sec)
CK2	755.6	0.327	6.510e-009

Zeta potential of nanosuspension formulation

Zeta potential of sonicated cefpodoxime proxetil nanosuspension was determined by Malvern nano zetasizer instrument**.**

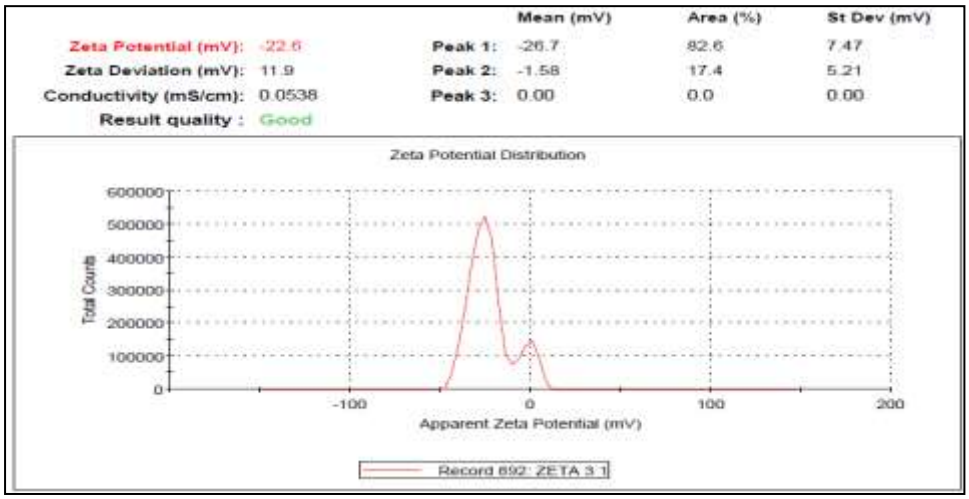

Figure 12: Zeta potential analysis of nanosuspension.

Drug content

Table 9: Drug content of cefpodoxime proxetil nanosuspension formulation

Solvent	Absorbance	Conc. (µg/ml)	DF (x100) (µg/ml)	Conc. (mg)	% drug content= (initial-final)/initial×100
Methanol	0.257	6.805555556	680.555556	0.68055556	99.31944444

Table 10: Physicochemical characteristics of oral fast dissolving film

Oral fast dissolving film formulations			
Characteristics	**Formulation F1**	**Formulation F2**	**Formulation F3**
Weight (mg)	0.091	0.099	0.090
Surface pH	6.8	6.7	6.5
Folding endurance	149	163	132
Disintegration time (sec)	9-32	30-35	32-36
Percentage drug content	99.52	98.9	96.4

Nanosuspension loaded fast dissolving films of cefpodoxime proxetil be prepared by the solvent casting method on glass petridish, using HPMC E50 as polymers. Propylene glycol was used as a plasticizer and glycerin as a sweetener. Distilled water was used as a solvent for HPMC. The effect

of the concentration ratio of polymers was studied by preparing various formulations of fast dissolving films. In all these formulations, a constant amount of drug (100 mg) was maintained. The casting solution (10 ml) was poured into petridish, so that each square centimeter contains approximately 20 mg of the drug. Polymers were used in different concentration and the concentration of other ingredients such as plasticizer and sweetener were kept constant. Fast dissolving films of cefpodoxime proxetil was evaluated for various parameters. In the present study, three formulations were prepared by varying the polymer concentration. Effects of the polymer concentration different formulations (F1 and F2) were prepared using HPMC E50. Table 3 and table 10 involve studying the effect of polymer concentration on the physicochemical properties of the film. The physical appearances of films were evaluated. All the films prepared with different polymer concentrations be found to be flexible, smooth, transparent, non-sticky, and homogeneous. For evaluation purposes 2×2 cm^2 areas was cut from it. Variation in the weights of the formulations was determined by weighing 2×2 cm^2 section of each film on a digital balance and then calculating the average weight. From the results shown in table 10 it was observed that all the batches were uniform in weight with no significant difference in the weight of the individual formulation from the average value. Weight variation was found to be in the range of 0.091 to 0.099 mg for films prepared. The surface pH was found to be in the range of 6.2-7.08 which is close to the neutral pH which indicates that films may have less potential to irritate to the mucosal lining of the oral cavity, and hence, more acceptable by the patients. The folding endurance was measured manually. It measures the ability of the film to withstand rupture. The results indicated that the endurance increases on increasing polymer content in the film. It varied from 149-163 in the films formulated as shown in table 10. Drug content of all the formulations was determined using UV-Visible spectrophotometer. The result showed good uniformity of drug content throughout the films without any significant variation as shown in table 10 drug content was found to vary from 99.52% and 98.9% indicated good content uniformity.

Table 11: *In-vitro* dissolution profile data of film formulation (F1 & F2)

Time (min.)	% Cumulative Drug release (F1 Film formulation)	% Cumulative Drug Release (F2 Film Formulation)
1	59.0869565	36.1304347
2	64.1869565	40.5956521
3	67.8260869	45.1695652
4	76.9956521	48.1565217
5	86.1391304	51.9782608
6	90.2913043	60.0521739
7	94.6260869	65.9956521

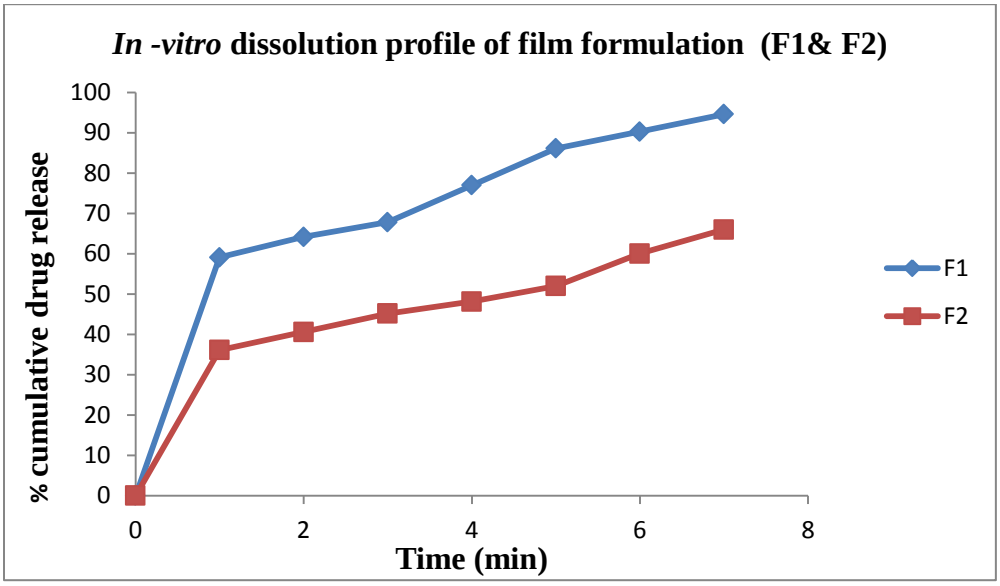

Figure 13: Comparison of *in-vitro* dissolution profile of film formulation F1 and F2

In-vitro dissolution study was performed for 7 mins. Release studies showed that drug get rapidly released from all formulations. When the concentration of the polymer increased, the drug release was found to be decreased due to the increase in the time required for wetting and dissolving the drug molecules present in the polymer matrices. The drug release was found to be in the following order: F1 > F2. Among the two formulations (F1, F2) prepared formulations F1was found to be the best formulations in terms of drug release.

In-vitro dissolution studies of pure drug loaded in fast dissolving film.

Table 12: *In-vitro* dissolution profile of pure drug loaded film formulation (F3)

Time (min.)	% Cumulative Drug Release (F3 Pure Drug loaded in Film Formulation)
1	30.13043478
2	36.87391304
3	41.2
4	43.41304348
5	46.56956522
6	48.74347826
7	51.85652174

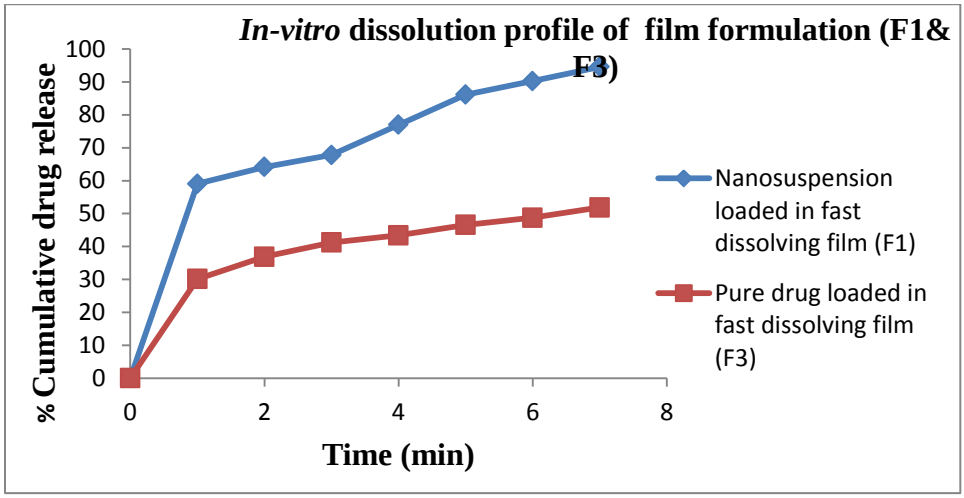

Figure 14: Comparison of *in-vitro* dissolution profile of film formulation F1 and F3

The release profile of fast dissolving films (F1) was compared with that of pure drug of fast dissolving film (F3) and it was observed that the drug release from nanosuspension loaded in fast dissolving films was much faster than that from raw drug of fast dissolving film.

CONCLUSION

Oral films intended for the application in the oral cavity are an innovative and promising dosage form especially for use in pediatric and geriatric patients. A part of the research work focused on the development of dosage form for pediatric and geriatric use with an appropriate active substance while the other part aims in the development of adequate analytical methods for the characterization as well as improving already existing approaches for solubility and bioavailabilty enhancement. So, it might be concluded that the nanosuspension loaded fast dissolving film act as a promising delivery for the treatment of upper respiratory tract bacterial infections with higher drug release when compared to film containing pure drug.

FUTURE ASPECTS

The present study only evaluates different nanosuspension components for their suitability to obtain an optimal formulation with required physico-chemical parameters and dissolution study. However, further studies including pharmacokinetic and pharmacodynamic profiling would be required to actually establish the formulation as a product for the treatment of bacterial infection. The work may be taken up as an extension of the present study.

REFERENCES

1. Savjani KT, Gajjar AK and Savjani JK: Drug solubility: Important and enhancement techniques, ISRN Pharm 2012.

2. Shafiq-un-Nabi S, Shakeel F, Talegaonkar S, Ali J, Baboota , Ahuja, A, Khar RK and Ali M: Formulation development and optimization using nanoemulsion technique: A technical note. AAPS Pharm. Sci. Tech 2007; 8:1-6.
3. Khadka P, Jieun R, Kim R, Kim I, Kim, JI, Kim H, Cho JM, Yum G and Lee J: Pharmaceutical particle technologies: An approach to improve drug solubility, dissolution and bioavailability. Asian J Pharma Sci 2014; 9:304-316.
4. Geetha G, Poojitha Uand Ahmed Khan A: Various techniques for preparation of nanosuspension. A Review Int J Pharma Res 2014; 3(9):30-37.
5. Shete RS, Gadhave MV: Nanosuspension, A promising drug delivery system. World J Pharma Res 2014; 4(2):560-573.
6. http://www.drugbank.ca/drugs/DB01416, Accessed on 18. 01. 2015.
7. Borin TM: A review of the pharmacokinetics of cefpodoxime proxetil. Clinical pharmacokinetic unit Upjohn laboratories 1991; 3:13-21.
8. Martindale: The extra pharmacopoeia. Published by pharmaceutical press, London, Edition 36th 2009: 238-239.
9. Bala R, Pawar P, Khanna S and Arora S: Orally dissolving strips: A new approach to oral drug delivery system. Int. J. Pharm 2013; 3(2):67-76.
10. Mundhe B, Kadam V, Jadhav S, Zamiruddin Md, Bharkad: A short review on fast dissolving oral film. World J Pharm Pharma Sci 2014; 3(3): 463-475.
11. Swamy MSS, Shetty SK and Kumar A: UV-visible spectrophotometric methods for the estimation of cefpodoxime proxetil in bulk drug and pharmaceutical dosage form. Int J Pharma. Tech. Res 2012; 4(2):750-756.
12. Patel DJ, Patel JK, Pandya VM, Patel RD: Effect of formulation variable on nanosuspension containing famotidine prepared by solvent evaporation technique. Int J Pharma Sci Nanotechnology 2010; 2(4): 707-713.
13. Kakrana M, Sahooa NG, Lia L, Judeh Z, Wang Y, Chong K, Loh L: Fabrication of drug nanoparticles by evaporative precipitation of nanosuspension. Int J Pharm 2010; 383:285-292.
14. Sandhya J, Pavani R and Reddy R: Formulation and evaluation of nanosuspension of nisoldipone. Int J Pharm. 2014; 24(1):177-181.
15. Thadkala K, Kumari NP, Rambabu B, Sailu C, Aukunuru B: Preparation and characterization of amorphous for enhancement of oral bioavailibity. Int J Pharma 2014; 4(3): 131-137.

16. Lakshmi P and Kumar GA: A review nanosuspension technology. Int J Pharm Pharma Sci 2010; 2(4):35-40.
17. Attari Z, Bhandari A, Jagadish PC and Lewis S: Enhanced ex vivo intestinal absorption of olmesartan medoxomil nanosuspension: Preparation by combinative technology. Saudi Pharma J 2015.
18. Prabhu P, Malli R, Koland M, Vijaynarayana K, Souza UD, Harish NM, Shastry CS and Charyulu RN: Formulation and evaluation of fast dissolving films of levocitirizine dihydrochloride. Int J Pharma Investigation 2011; 1(2): 99-104.
19. Yatin DK, Dipen A, Trambadiya AV and Patel VP: Formulation and evaluation of fast dissolving sublingual film of metoprolol succinate. Int J Pharma Sci 2013; 4(3):140-154.
20. Irfan MR, Sumeira BQ, Qadir MI, Jabeen F and Khan A: Orally disintegrating films: A modern expansion in drug delivery system. Saudi Pharma J 2015: 1-10.
21. Bansal S, Bansal M, and Garg G: Formulation and evaluation of fast dissolving film of an antihypertensive drug. Int J Pharma Chemical and Biological Sci 2013; 3(4): 1097-1108.
22. Keshari A, Sharma PK: Fast dissolving oral film: A novel and innovative drug delivery system. Int J Pharma Sci Res 2014; 5(3):92-95.