International Journal of Pharmacy and Pharmaceutical Sciences

ISSN- 0975-1491 Vol 2, Issue 4, 2010

Research Article

EFFECT OF PVP-K30 ON COMPLEXATION AND DISSOLUTION RATE OF NEVIRAPINE-β-CYCLODEXTRIN COMPLEXES

LOKAMATHA K M*[1], BHARATHI A[1], SHANTA KUMAR S M[2], RAMA RAO N[3]

[1]Department of Pharmaceutics, [2]Department of Pharmaceutical Chemistry, V. L. College of Pharmacy, Raichur-584103, [3]Department of Pharmaceutics, Chalapathi Institute of Pharmaceutical Sciences, Guntur- 522001. Email:lkmswamy@rediffmail.com

Received: 22 May 2010, Revised and Accepted: 26 Jun 2010

ABSTRACT

In the present study, attempts were made to improve the aqueous solubility and dissolution rate of Nevirapine (NVP), a poorly soluble drug via complexation with β-cyclodextrin (β-CD). Further, studies were carried out to investigate the effect of hydrophilic polymer (PVP-K30) on the complexing and solubilizing efficiency of β-CD. The complexation of NVP with β-CD was investigated by phase solubility studies in the presence and absence of PVP in pH 1.2 and pH 6.8 as the NVP exhibited pH dependent solubility. Ternary complexes were prepared by kneading method with addition of PVP (5%, 10% and 20% w/w of the solid complex) to NVP:β-CD (1:1M) systems. All solid complexes were characterized by performing dissolution studies in 0.1N HCl and pH 6.8 and by analytical techniques such as DSC, FT-IR, P-XRD and SEM. The phase solubility profiles in the presence and absence of PVP were classified as A_L-type, indicating the formation of 1:1 inclusion complex. Stability constants ($K_{1:1}$) calculated from the phase solubility diagrams were found to be pH dependent. Both binary and ternary systems exhibited higher dissolution rates in 0.1N HCl and pH 6.8 than their corresponding physical mixtures and pure drug. Ternary complexes reflect the vital role of PVP to improve the solubility and dissolution rate of NVP:β-CD systems both in gastric and intestinal pH, which could minimize the variable dissolution rates with increase in the oral bioavailability. Additionally, ternary inclusion complexes offers an advantage of using less amount of CD to be needed to solubilize a given amount of model drug in the development of pharmaceutical formulations. The release of drug from all the preparations was followed predominately first order kinetics compared to Hixson-Crowell's cube root law.

Keywords: Nevirapine, Antiretroviral, solid complexes, β-cyclodextrin, Polyvinylpyrrolidone -K30.

INTRODUCTION

Solubility plays an important role in drug disposition, since the maximum rate of passive drug transport across a biological membrane, is the product of permeability and solubility. According to the biopharmaceutics classification system (BCS), aqueous solubility and permeability are the most important parameters affecting drug bioavailability. Retrospective studies show that greater than 40% of drug failures in development can be traced to poor biopharmaceutical properties, mainly due to poor dissolution or poor permeability[1]. Thus improvement of aqueous solubility in such case is valuable goal to effectively formulate them into bioavailable dosage forms. Hence, great efforts have been made to improve oral bioavailability of poorly water soluble drugs by increasing their dissolution rate through various techniques[2,3], such as the formulation of amorphous solid form, nanoparticles, microemulsions, solid dispersions, melt extrusion, salt formation and formation of water-soluble complexes etc. Among them, the complexation with cyclodextrins is most frequently used. Cyclodextrins (CDs) are a group of structurally related natural products formed during bacterial digestion of cellulose and belongs to a family of cyclic oligosaccharides with a hydrophilic outer surface and a lipophilic central cavity. They are widely used as "molecular cages" in the pharmaceutical industry, as complexing agents to increase the aqueous solubility of poorly soluble drugs and to increase their bioavailability and also the stability[4].

Nevirapine is an antiretroviral drug that is currently used in the treatment of human immunodeficiency virus type 1 (HIV-1) infections[5,6]. The model drug belongs to Biopharmaceutical Classification System (BCS) class II (low solubility/high permeability), poses a challenge in achievement of optimal dissolution kinetics from the dosage form[7]. It is highly lipophilic and very slightly soluble in water (0.1 mg/ml) which gives rise to difficulties in the formulation of dosage forms and leads to variable dissolution rates with a resultant decrease in bioavailability[8]. Hence in the present work, inclusion complexes of NVP with β-cyclodextrin were tried, in order to achieve sufficient solubility along the whole gastro-intestinal tract, which is a crucial step in the development of NVP formulations. Cyclodextrins play an important role in improving the therapeutic efficacy of drugs with poor aqueous solubility through inclusion complexation. They can enhance the aqueous solubility of lipophilic drugs without changing their intrinsic ability to permeate biological membranes. Thus, through cyclodextrin complexation it is possible to move Class II drugs, and sometimes even Class IV drugs, into Class I. In such cases, the contribution of complexation with cyclodextrins will be highly appreciated which modifies the physico-chemical properties such as solubility, dissolution rate and bioavailability of the guest molecules[9,10]. β-CD appears to be the best natural cyclodextrin due to its efficient drug complexation, low cost, low toxicity and availability in the pure form.

For a variety of reasons, like toxicological considerations, formulation bulk, production cost, drug bioavailability and isotonicity, it is important to use as small amount of cyclodextrin as possible in pharmaceutical formulations[11]. In some cases, large amounts of CD are required to solubilize the small amounts of drug and thus making CD solubilization of drugs impractical. It is therefore important to develop methods which can be applied in order to enhance the complexation efficiency (CE) of CD. Various methods have been applied to enhance the complexation efficacy[12]. Therefore; it was further extended our investigation to enhance the complexing and solubilizing efficiency of β-CD by adding different concentrations of hydrophilic polymer, polyvinylpyrrolidone (PVP-K30) to NVP:β-CD 1:1 (molar ratio) system. Polymers are known to interact with CDs although the exact nature of the polymer: CD interaction is still not known. Various studies reported that, addition of small amount of water-soluble polymers to an aqueous complexation medium frequently results in an increase in the CE and, consequently, is less demanding on the formulation bulk[13,14].

MATERIALS AND METHODS

Nevirapine was kindly supplied by Aurobindo Pharma Limited, Hyderabad, India. β-Cyclodextrin and Polyvinylpyrrolidone-K30 were purchased from HiMedia Laboratories Private Limited, and Rolex Chemical Industries, Mumbai, India, respectively. All other chemicals and solvents used were of analytical grade.

Solubility studies

The solubility of Nevirapine in distilled water, 0.1N HCl (pH 1.2), pH 4.6, pH 6.8 and pH 7.2 was determined. An excess amount of NVP was placed in glass bottles containing 20 ml of solvent. The bottles

were thoroughly shaken for 12 h and kept aside for 24 h at room temperature. At the end of this period, the solutions were filtered and filtrate was collected into dry containers. The solutions were suitably diluted and assayed for NVP content spectrophotometrically at 314 nm.

Phase solubility studies

Phase solubility studies were carried out in 0.1N HCl and pH 6.8 according to the method described by Higuchi and Connors[15]. Excess amount of NVP (50 mg) was added to 20 ml CD solution (ranging in concentration from 0.015 to 0.03M) prepared in 0.1N HCl and pH 6.8 buffer solution in a series of 100 ml stoppered conical flasks. Then, the suspensions were shaken on a rotary at 28^{o}C for 24 h. After equilibrium was achieved, 2 ml aliquots were filtered through 0.45 µm membrane filter and appropriately diluted. The concentration of the drug was determined spectrophotometrically at 314 nm. Shaking was continued until three consecutive estimations were the same. Phase solubility studies were conducted in each case with and without the addition of PVP-K30 at a concentration of 0.5% w/v to the solution containing CDs. The solubility experiments were conducted in triplicate. The blanks were performed in the same concentrations of CDs in 0.1N HCl and pH 6.8 buffer solutions in order to cancel any absorbance that may be exhibited by the CD molecules.

Preparation of NVP:β-CD solid complexes

The preparation of NVP:β-CD (1:1M) solid inclusion systems were performed by kneading method (table 1). The 1:1 molar ratio was based on the previous solubility studies. All preparations were passed through sieve no.120 and stored in dessicator for further evaluation.

Table 1: Physical mixtures and solid binary and ternary complexes of NVP

Sr. No	Batches	Drug	Cyclodextrin	Ratio	Method	PVP-K30
1	B1	NVP	β-CD	1:1M	PM	-
2	B2	NVP	β-CD	1:1M	KM	-
3	B3	NVP	β-CD	1:1M	PM	5%
4	B4	NVP	β-CD	1:1M	PM	10%
5	B5	NVP	β-CD	1:1M	PM	20%
6	B6	NVP	β-CD	1:1M	KM	5%
7	B7	NVP	β-CD	1:1M	KM	10%
8	B8	NVP	β-CD	1:1M	KM	20%

Physical mixtures (PM): The physical mixtures of NVP and β-CD in 1:1 molar ratio and physical mixtures of ternary complexes of NVP:β-CD:PVP (5%, 10% and 20% w/w of the solid complex) were prepared by mixing individual components that had previously been sieved through sieve no.120.

Kneading method (KM): The NVP and β-CD were triturated in glass mortar with small volume of methanol. The thick slurry was kneaded for 1h and then dried at 45^{0}C until dryness. The dried mass was pulverized and sieved through sieve no.120. Similarly, ternary complexes of NVP:β-CD:PVP were prepared by mixing in 1:1 molar ratio with the addition of PVP (K30) in different concentrations of 5%, 10% and 20% w/w of the solid complex.

Analysis of solid complexes in solid state

Fourier Transform Infrared Spectrometry (FT-IR): Solid samples were prepared by potassium bromide disc method and scanned for absorbance from 400–4000 cm−1. The spectra were obtained on a Perkin Elmer 1600 series, (USA) for NVP, β-CD, physical mixtures and all binary systems.

Differential scanning calorimetry (DSC): DSC Thermograms of NVP, physical mixtures and solid complexes were obtained by using differential scanning calorimeter (Perkin Elmer). Samples (2-5 mg) were sealed in aluminium pans and scanned at a heating rate of 10^{0}C/min over a temperature range of 30 to 300°C under a nitrogen gas stream.

Powder X-ray diffractometry (PXRD): The powder XRD patterns of pure drug, physical mixtures, solid binary and ternary systems were recorded using an X-ray diffractometer (Philips Analytical XRD). Samples were scanned over an angular range of 3-40°; 2θ at a scan rate of 0.01°/sec.

Scanning Electron Microscopy (SEM): The surface morphology of the raw materials and of the binary systems was examined by means of JSM-6400 (Jeol, Japan) scanning electron microscope. The samples were previously fixed on a brass stub using double-sided adhesive tape and were then made electrically conductive by coating with a thin layer of gold and palladium alloy (180-200 Å) using a fine coat ion sputter (JEOL, fine coat ion sputter JFC-1100). The pictures were taken at an excitation voltage of 20 kV and magnification in the range of 118 to 245X.

Analysis of solid complexes in solution state

Drug content uniformity: In each case, complex equivalent to 100 mg of NVP was accurately weighed and transferred to 100 ml volumetric flask and extracted in methanol. The volume was made up to 100 ml with 0.1N HCl. From this, 1ml was subsequently diluted with 0.1N HCl and assayed for NVP content by measuring at 314 nm using 0.1N HCl as blank.

***In-vitro* dissolution studies**

In vitro dissolution tests were performed for pure drug, physical mixtures and inclusion complexes using the dissolution test apparatus USPXXIV Type II (Electro Lab, Mumbai, India) by powder dispersion method. NVP (100 mg) or inclusion complex equivalent to 100 mg of NVP was used in each test. The dissolution studies were carried out using 900 ml of 0.1N HCl (pH 1.2) as well as in pH 6.8 solution maintained at 37 ± 0.5°C with paddle rotation maintained at 50 rpm (n=3). The release of NVP was measured by withdrawing 5 ml samples at regular intervals of time, filtered, suitably diluted and assayed spectrophotometrically at 314 nm. Fresh dissolution medium was added to maintain a constant volume after each sampling. Dissolution results of pure drug, physical mixtures and solid binary and ternary systems were computed by using dissolution software PCP DISSO V3.

RESULTS AND DISCUSSION

Nevirapine is a BCS class II drug poses a challenge in the design of dosage form due to its low aqueous solubility. Hence complexation with β-CD was tried to overcome the drawback associated with the model drug. Solid binary inclusion complexes of NVP:β-CD (1:1M) were prepared by kneading method. In the next phase, ternary complexes were prepared by same method under similar set of conditions with the addition of different concentrations of PVP (5%, 10% and 20% w/w of the solid complex) to NVP:β-CD systems to investigate the effect of hydrophilic polymer on the complexation and solubilizing efficiencies of β-CD and on the dissolution rate of NVP from the β-CD complexes. All solid complexes prepared were found to be fine and free flowing. Characterization of binary and ternary complexes was performed by phase solubility studies, FT-IR, DSC, SEM and powder X-ray diffractometry. Dissolution results of pure drug, solid binary, ternary systems and their corresponding physical mixtures were computed by using dissolution software PCP DISSO V3.

Solubility studies

Solubility of drug was determined at 25 ± 0.5°C, in distilled water and wide range of pH solutions of 0.1 N HCl (pH 1.2), pH 4.6, pH 6.8 and 7.2. The solubility results are displayed in figure 1. Because NVP is weak basic drug (pKa 2.8), an increase in solubility was anticipated with decrease in pH. The pH solubility profile indicated a gradual decline in solubility with an increase in pH from 1.2 (1.703 mg/mL) to 4 .6 (0.252 mg/mL) and remained steady at pH 7 and 7.2 (0.1 mg/mL). The solubility of NVP decreased by approximately, 85% with an increase in pH from 1.2 to 4.6. These results revealed that the solubility of NVP is pH dependent, which is in accordance with the reported literature[16].

Drug content and percentage yield

The drug content and percentage yield were estimated to confirm that there is no degradation of the drug and expected amount of drug present in the product. The percentage yield of binary systems was in the range of 96.86 % -99.87 % of the initial amounts taken. The drug content was found to be in the range of 97.08 to 102.08 %. In case of ternary systems, the drug content and % yield was 89.51 to 99.68 % and 98.75 to 99.99% respectively. Low standard deviation (SD) and coefficient of variation (CV) values in the drug content of NVP:β-CD and NVP: β-CD:PVP systems indicated uniform drug distribution in all solid complexes and also ensured the applications of the present methods for the preparation of solid complexes with high content uniformity.

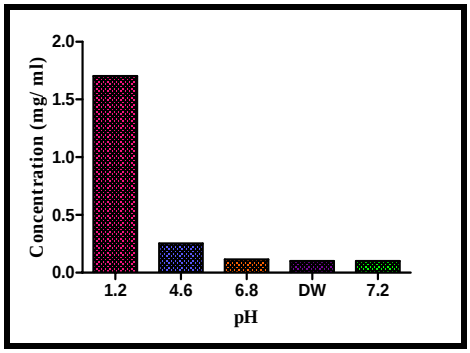


Fig. 1: Solubility profile of pure NVP in different pH solutions

Phase solubility studies

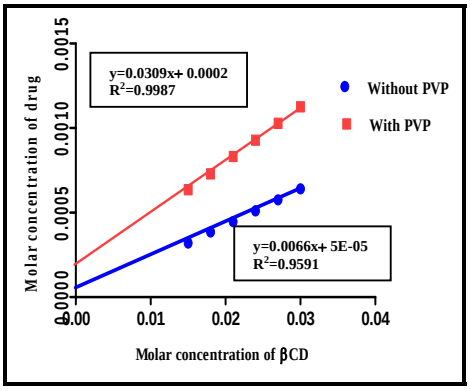
The phase solubility diagrams for the complex formation between NVP and β-CD in the presence and absence of PVP studied in 0.1N HCl and pH 6.8 are presented in figure 2 (a) & (b) respectively. These plots illustrated that the aqueous solubility of the drug increases linearly as a function of β-CD over the entire concentration range studied and can be classified as A_L-type according to Higuchi & Connors[15]. The linear host (β-CD)-guest (NVP) correlation coefficient with a slope less than 1 indicated the formation of 1:1 water soluble complex with respect to β-CD concentrations. The apparent stability constants ($K_{1:1}$) obtained from the slope of the linear phase solubility diagrams and solubilizing values are given in table 2. The values indicated that the complexes formed between NVP and β-CD was quite stable. The calculated $K_{1:1}$ value was found to be pH dependent and also the presence of hydrophilic polymer.

To evaluate the effect of hydrophilic polymer, the solubilizing efficiency of β-CD in the presence and absence of PVP was calculated. In each case solubilizing efficiency was calculated as the ratio between the drug solubility in pH 1.2 and pH 6.8 solutions of β-CD (0.021M), with and without hydrophilic polymers (0.5%), to the drug solubility in pH 1.2 and pH 6.8 solutions respectively. In pH 1.2, β-CD alone gave a 6.99 fold increase in the solubility of NVP, whereas in the presence of hydrophilic polymer, it gave a 13 fold increase. Similarly, in pH 6.8 solutions of β-CD; a 159.34 and 42.05 fold increase in solubility was observed in the presence and absence of PVP respectively. The values of stability constant ($K_{1:1}$) were found to be higher in the presence of hydrophilic polymer studied in pH 6.8, indicating the formation of a ternary complex, with increased complexation efficiency of CD. On the other hand, decrease in $K_{1:1}$ was observed in pH 1.2 in the presence of polymer might be due to decrease in drug-CD interaction. Such an effect can be explained on the basis of the higher initial drug solubility in the presence of hydrophilic polymer (PVP) with consequent less affinity to the cyclodextrin cavity[17].

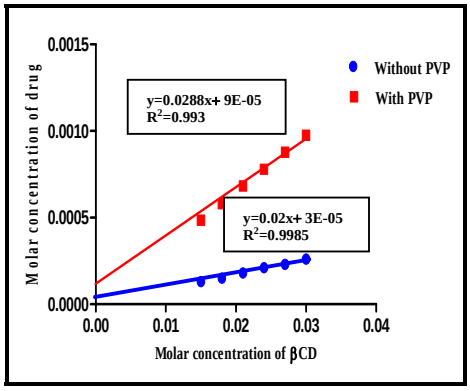
Table 2: Effect of PVP on stability constant and solubilizing efficiency of β-CD

Solutions	Sample	$K_{1:1}$ (M^{-1})	Solubilizing efficiency*
0.1N HCl	NVP: β-CD	339.48	6.99
pH 6.8	NVP: β-CD	213.98	42.05
0.1N HCl	NVP:β-CD:0.5% PVP	170.83	13.00
pH 6.8	NVP:β-CD:0.5% PVP	292.57	159.34

*Ratio between the drug solubility in 0.1N HCl and pH 6.8 solutions of CDs (with or without PVP) and drug solubility in 0.1N HCl and pH 6.8 solutions



(a)



(b)

Fig. 2: Phase solubility profile of NVP in the presence and absence of PVP studied in (a) 0.1N HCl and (b) pH 6.8

Fourier transform-IR studies

To study the possible interactions between the NVP and β-CD/PVP in the solid state, IR spectra of binary and ternary systems were compared with the corresponding physical mixtures and with the drug alone. The IR spectrum of NVP exhibited characteristic peaks for amide group at 3186.40 cm^{-1} and 1647.21 cm^{-1} due to N-H and C=O streching respectively. The IR spectrum of NVP: β-CD complex (1:1M) prepared by kneading method showed peaks at 3194.12 cm^{-1} and 1649.14 cm^{-1} due to N-H and C=O streching of amide group of the drug. Shifts of peaks from 3186.40 cm^{-1} to 3194.12 cm^{-1} of N-H and 1647.21 cm^{-1} to 1649.14 cm^{-1} of C=O indicated a weak interaction between the drug and β-CD (figure 3).

The IR spectra of ternary systems i.e., B6, B7 and B8 prepared by kneading method clearly corresponded to that of the major component of the mixture; i.e., β-CD. Therefore the spectra of the ternary systems were compared with the corresponding ones of the NVP and NVP:β-CD complexes. In the IR spectra of all ternary systems, it was observed that N-H streching of amide group of the drug is strongly shifted towards higher wavelength. Shift of peak from 3186.40 cm^{-1} to 3350.35 cm^{-1}, indicated complete interaction

during complexation of NVP with β-CD in the presence of hydrophilic polymer, PVP (figure 3).

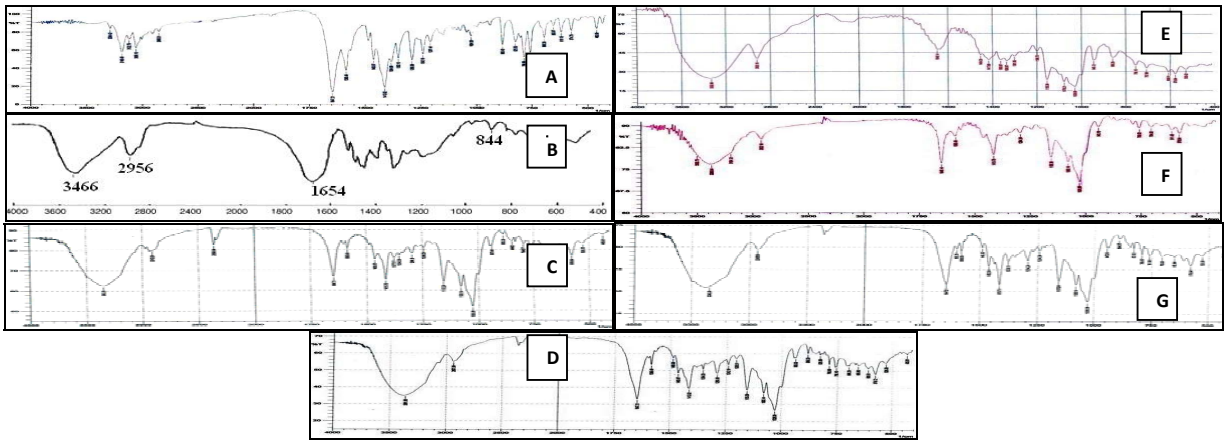


Fig. 3: Comparison between FTIR spectra of (A) NVP, (B) PVP, (C) βCD, (D) NVP:βCD (KM), (E) NVP:βCD:PVP 5% (KM), (F) NVP:βCD: PVP 10% (KM), and (G) NVP:βCD: PVP 20% (KM)

DSC studies

DSC thermogram of pure NVP, β-CD and its binary and ternary solid systems (B6) are presented in figure 4. The DSC thermogram of NVP exhibited an endothermic peak corresponding to its melting point (t_{onset} = 245.44 °C, t_{peak} = 249.04 °C, ΔH = 241.0185 J/g). β-CD showed broad endothermic peak due to loss of water. In the thermogram of NVP:β-CD prepared by kneading method, the intensity of the endothermic peak at 249 °C was reduced and indicated an interaction between NVP and β-CD. DSC thermograms of pure NVP and NVP:β-CD solid binary complexes were compared with the thermogram of NVP:β-CD:PVP 5% solid ternary complexes to analyze the effect of hydrophilic polymer on the inclusion complexation. Thermograms clearly showed that the endothermic peak at 249 °C with the solid ternary complex of NVP:β-CD:PVP 5% (figure 4, E) was completely disappeared which indicated the absence of crystallinity of drug and also the formation of true complex of NVP with β-CD in the presence of hydrophilic polymer.

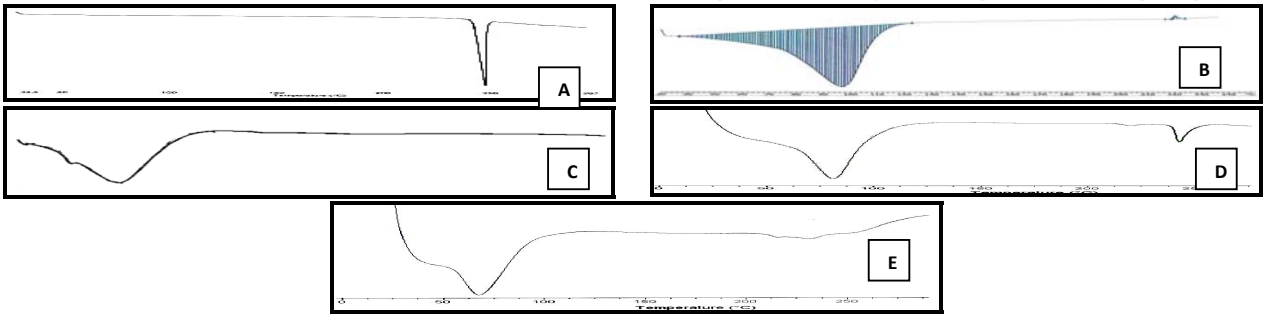


Fig. 4: Comparison between DSC thermograms of (A) NVP, (B) β-CD, (C) PVP (D) NVP: β-CD (KM), (E) NVP: β-CD:PVP 5% (KM)

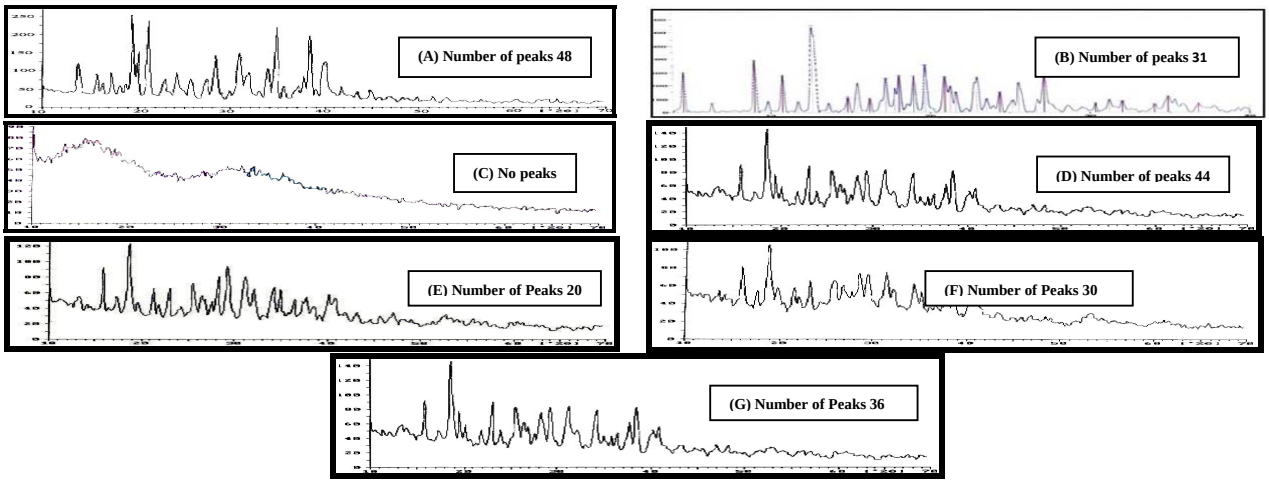


Fig. 5: Comparison between XRD spectra of (A) NVP, (B) β-CD, (C) PVP, (D) NVP: β-CD (KM), (E) NVP:β-CD:PVP 5% (KM), (F) NVP:β-CD:PVP 10% (KM) and (G) NVP:β-CD:PVP 20% (KM)

Powder X-ray diffractometry

The XRD pattern of pure NVP showed more number of peaks that were intense and sharp, indicating its crystalline nature. The spectrum of PVP was characterized by complete absence of any diffraction peak. The diffraction pattern of NVP:β-CD binary systems also showed other peaks of NVP with decrease in the intensity of peak indicating the reduction in crystallinity. X-ray diffractograms of NVP, β-CD and ternary complexes of NVP:β-CD:PVP 5%, 10% and 20% prepared by kneading method in comparison with the NVP:β-CD binary systems are shown in figure 5. The XRD pattern of ternary complexes differs significantly from that of binary systems. Also, the effect of polymer concentration on the complexation was evident from the XRD studies. It was clear from the XRD patterns of ternary systems that intensity and number of peaks were decreased as the concentration of PVP was decreased as depicted in figure 5(E), (F) and (G).

SEM analysis

Morphological features of the NVP solid complexes were examined by scanning electron microscopy. Figures 6 shows the scanning electron microscopic pictures of (A) NVP alone, (B) β-CD, (C) PVP, (D) solid binary complex of NVP:β-CD, and (E) ternary system NVP:β-CD:PVP 5%. NVP existed as irregular shaped aggregates and β-CD appeared as large crystalline particles, whereas the PVP were seen as amorphous or pieces of spherical particles. SEM photographs of binary systems clearly depicted the reduction in NVP drug particle sizes and adhered onto the surface of β-CD. This observation suggested the existence of interaction between NVP and β-CD. In case of ternary system, the original morphology of all three components disappeared and existed as small aggregates of irregular amorphous pieces.

Therefore, the reduced drug particle size, increased surface area and also close contact between hydrophilic polymer (PVP-K30) and NVP might be responsible for the increased solubility and dissolution rate of the prepared solid complexes.

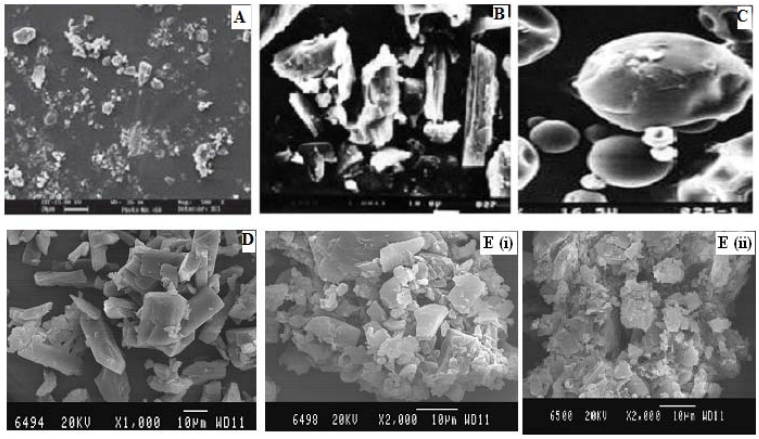


Fig. 6: SEM photographs of (A) NVP, (B) β-CD, (C) PVP, (D) NVP: β-CD 1:1M (KM) and (E) NVP: β-CD:PVP 5%

Dissolution behaviour of solid complexes

All solid binary and ternary complexes and their corresponding physical mixtures were tested for dissolution properties and compared with the pure NVP. Since NVP exhibited pH dependent solubility (figure 1), dissolution studies of all solid complexes were carried out in both simulated gastric fluid (0.1N HCl i.e., pH 1.2) and simulated intestinal fluid (pH 6.8). It is reported that, the absorption of NVP is excellent from the small intestine[18]. Dissolution data were evaluated on the basis of cumulative percentage drug release, dissolution efficiency and correlation coefficient (r). The percentage of NVP dissolved at 30 min (DP_{30}) and dissolution efficiency at 10 min (DE_{10}), at 30 min (DE_{30}) and at 60 min (DE_{60}); and the characteristic time for 30% and 50% dissolution of NVP (T_{30} &T_{50} min respectively) were calculated for all solid complexes. The dissolution efficiency of all preparations was calculated by the method mentioned by Khan[19].

Solid binary systems

In vitro dissolution data of NVP, physical mixtures, solid binary as well as ternary complexes studied in 0.1N HCl and pH 6.8 are presented in table 3 and 4 respectively. *In vitro* dissolution profiles of NVP alone, NVP:β-CD physical mixture (B1) and binary system (B2) studied in 0.1N HCl and pH 6.8 are shown in figure 7(a) and 7(b) respectively. The dissolution of drug alone was incomplete even after 120 minutes in both the media studied. Binary complexes exhibited higher rates of dissolution and dissolution efficiency values than the physical mixtures and pure drug. The slight increase noted in the dissolution rate from the physical mixtures of binary systems as compared with the pure drug might be due to the 'microenvironment effect'. The value of T_{50} of solid binary system was much lower than NVP alone. The mean percent drug release from the solid complexes at 30 minutes in pH 1.2 was 1.12 (B1), 4.95 (B2) fold higher when compared to pure drug (table 6). Improved dissolution rate and dissolution efficiency observed in this case might be due to the formation of solid inclusion complexes, with better interaction of drug and CD during the process. Similarly mean percent drug release from the solid complexes at 30 minutes (DP_{30}) in pH 6.8 was 1.17 (B1), 3.26 (B2) fold higher when compared to pure drug (table 7). As expected, all preparations showed little higher percentage of dissolved drug in pH 1.2 than in pH 6.8, probably due to the favourable solubility of the drug in the gastric juice. Overall, the *in vitro* results have shown an enhanced dissolution rate of NVP in both gastric and intestinal media from prepared drug-β-CD complexes.

Solid ternary systems

Dissolution profiles of ternary systems studied in 0.1N HCl and pH 6.8 are displayed in figure 8(a) and 8(b) respectively. The slight increase noted in the dissolution rate from the physical mixtures of ternary systems as compared with the pure drug might be due to the ability of the hydrophilic polymer to increase drug wettability. All ternary systems (B6, B7 and B8) exhibited a significant increase in dissolution rate with respect to the physical mixtures (B3, B4 and B5) and the reference drug. The increase in dissolution rate with these systems in both media of 0.1N HCl (pH 1.2) and pH 6.8 were found in the following manner,

B8 (PVP 5%) > B9 (PVP 10%) > B10 (PVP 20%)

The mean percent drug release from the solid ternary complexes at 30 minutes was 8.44 (B8), 7.49 (B9) and 7.31 (B10) fold higher;

and 9.08 (B8), 8.80 (B9) and 8.63 (B10) fold higher in pH 1.2 (table 6) and pH 6.8 (table 7) respectively when compared to pure drug. The slight increase in drug release at 30 minutes in pH 6.8 could be attributed to the marked increase in solubilizing efficiency of PVP in pH 6.8 than in pH 1.2 which was in accordance with the phase-solubility studies (table 2). Further, it was noted that the dissolution was progressively increased with decreasing concentration of PVP in the ternary systems. Ternary system with PVP 5% showed the most significant effect on the dissolution properties of drug. An amount of 83%, 93% and 99.9% of drug was released at the end of 30, 60 and 90 minutes of dissolution respectively. This significant result might be due to the increase in drug wettability, mechanical treatment and enhancing the complexation of drug with β-CD in the presence of lower concentration of hydrophilic polymer. These results were further confirmed by their XRD patterns (figure 5) and also by the absence of NVP endothermic peak in DSC thermogram of NVP:β-CD:PVP 5% ternary system (figure 4). Among the three concentration of PVP studied, PVP 5% was identified as best concentration to enhance the solubilizing and complexation efficiency of β-CD. Comparatively, DE_{10}, DE_{30} and DE_{60} and DP_{30} values of NVP:β-CD:PVP 5% ternary system were found to be much higher when compared with binary system (B2) prepared by kneading method. Also, the value of T_{50} of ternary systems was lower compared to the binary systems and pure drug. Thus a low amount of CD can be used to obtain the desired dissolution rate and also the efficiency of drug.

Dissolution profiles of all solid complexes were analysed according to Hixson-Crowell's cube root law and first order kinetics. The correlation coefficient (r) values of the first order kinetics were found to be slightly higher to the 'r' values of Hixson-Crowell's cube root model (table 5 and 6). Hence the release of the drug from the preparations followed predominantly first order kinetics compared to Hixson-Crowell's cube root law.

Table 3: *In vitro* dissolution data of NVP, binary and ternary systems in 0.1N HCl

Time in minutes	Cumulative percent of drug released (± SD, n=3)								
	NVP	B1	B2	B3	B4	B5	B6	B7	B8
10	05.32	00.76	11.87	6.46	3.046	4.75	48.60	42.91	39.78
	(0.02)	(0.21)	(0.19)	(0.42)	(0.29)	(0.62)	(0.48)	(0.47)	(0.48)
20	07.59	04.46	24.94	14.13	10.15	9.30	65.33	59.64	59.07
	(0.03)	(0.17)	(0.22)	(0.25)	(0.40)	(0.30)	(0.69)	(0.73)	(0.53)
30	09.85	07.30	48.78	18.38	14.97	12.70	83.16	73.78	72.08
	(0.02)	(0.18)	(0.21)	(0.46)	(0.53)	(0.37)	(0.26)	(0.39)	(0.63)
45	14.67	16.94	60.08	25.17	21.76	18.64	90.44	81.93	81.93
	(0.06)	(0.27)	(0.28)	(0.72)	(0.47)	(0.67)	(0.54)	(0.40)	(0.67)
60	21.46	23.16	69.08	30.53	26.84	24.57	93.46	87.79	87.79
	(0.01)	(0.26)	(0.30)	(0.51)	(0.63)	(0.72)	(0.29)	(0.47)	(0.61)
90	27.94	29.64	72.40	39.27	34.17	32.76	99.92	93.07	93.07
	(0.01)	(0.11)	(0.27)	(0.63)	(0.52)	(0.74)	(0.53)	(0.39)	(0.56)
120	33.29	37.53	85.90	48.84	41.21	40.92	----	98.34	96.08
	(0.02)	(0.25)	(0.26)	(0.42)	(0.68)	(0.54)		(0.36)	(0.72)

Table 4: *In vitro* dissolution data of NVP, binary and ternary systems in pH 6.8

Time in minutes	Cumulative percent of drug released (± SD, n=3)								
	NVP	B1	B2	B3	B4	B5	B6	B7	B8
10	03.33	03.61	06.17	4.47	3.90	3.61	42.34	39.21	36.64
	(0.45)	(0.56)	(0.39)	(0.48)	(0.58)	(0.48)	(0.52)	(0.43)	(0.46)
20	04.75	05.88	14.13	9.58	7.02	5.88	59.36	61.92	62.20
	(0.63)	(0.70)	(0.36)	(0.38)	(0.57)	(0.35)	(0.37)	(0.54)	(0.70)
30	08.15	09.57	26.62	12.98	10.14	9.00	74.07	71.79	70.37
	(0.32)	(0.57)	(0.46)	(0.27)	(0.70)	(0.47)	(0.64)	(0.25)	(0.41)
45	12.40	13.25	43.90	18.36	14.38	12.96	81.65	81.65	81.36
	(0.37)	(0.38)	(0.39)	(0.58)	(0.34)	(0.58)	(0.36)	(0.65)	(0.53)
60	15.79	16.64	52.64	24.29	19.47	17.20	88.36	87.51	85.81
	(0.28)	(0.39)	(0.38)	(0.48)	(0.26)	(0.57)	(0.44)	(0.70)	(0.62)
90	21.15	23.41	69.85	33.61	27.38	24.26	93.36	92.79	92.22
	(0.71)	(0.63)	(0.48)	(0.35)	(0.64)	(0.64)	(0.66)	(0.63)	(0.42)
120	27.06	29.89	77.98	39.51	34.70	32.72	98.06	95.80	95.51
	(0.64)	(0.48)	(0.28)	(0.28)	(0.27)	(0.59)	(0.73)	(0.64)	(0.34

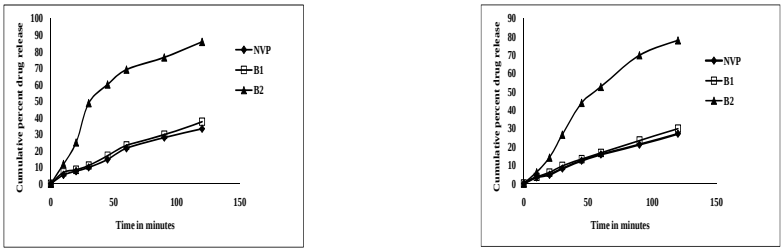
Table 5: Various dissolution parameters and the best model fitting curve values of NVP and solid complexes in 0.1N HCl

Batches	DE_{30} (%)	DE_{60} (%)	DP_{30}	T_{50} (min)	RDR_{30}	MDT_{30}	First order rates $K_1 \times 10^2$ (min^{-1})		Hix.Crow $K_{HC} \times 10^2$ ($mg^{1/3}.min^{-1}$)	
							R	K_1	R	K_{HC}
NVP	5.95	10.56	9.9	195.0	1	11.90	0.9937	-0.0036	0.9912	-0.0011
B1	2.96	9.52	7.3	177.1	1.12	17.83	0.9889	-0.0039	0.9889	-0.0039
B2	20.40	39.96	48.8	41.8	4.95	17.45	0.9776	-0.0166	0.9551	-0.0044
B3	9.93	17.37	18.4	120.3	1.86	13.79	0.9936	-0.0058	0.9877	-0.0018
B4	6.90	14.12	15.0	147.8	1.51	16.18	0.9917	-0.0047	0.9870	-0.0015
B5	6.80	12.72	12.7	155.5	1.28	13.93	0.9991	-0.0045	0.9972	-0.0014
B6	51.84	70.61	83.2	13.7	8.44	11.30	0.9928	-0.0507	0.9226	-0.0101
B7	46.48	63.92	73.8	20.5	7.49	11.10	0.9833	-0.0339	0.8956	-0.0071
B8	44.97	62.95	11.2	22.9	7.31	11.47	0.9625	-0.0303	0.8570	-0.0067

Table 6: Various dissolution parameters and the best model fitting curve values of NVP and solid complexes in pH 6.8

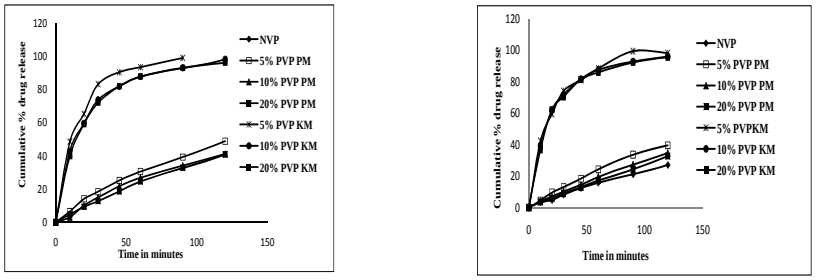
Batches	DE_{30} (%)	DE_{60} (%)	DP_{30}	T_{50} (min)	RDR_{30}	MDT_{30}	First order rates $K_1 \times 10^2$ (min^{-1})		Hix.Crow $K_{HC} \times 10^2$ ($mg^{1/3}.min^1$)	
							R	K_1	R	K_{HC}
NVP	4.05	8.12	8.2	257.7	1	15.09	0.9976	-0.0027	0.9962	-0.0009
B1	4.76	8.97	9.6	231.1	1.17	15.07	0.9988	-0.0030	0.9975	-0.0010
B2	11.21	11.21	26.6	54.8	3.26	17.37	0.9946	-0.0126	0.9938	-0.0035
B3	6.85	12.67	13.0	157.8	1.59	14.17	0.9971	-0.0044	0.9945	-0.0014
B4	5.33	9.97	10.1	194.9	1.24	14.23	0.9998	-0.0036	0.9994	-0.0011
B5	4.67	8.85	9.0	216.4	1.10	14.45	0.9987	-0.0032	0.9993	-0.0010
B6	46.25	63.84	74.1	20.7	9.08	11.27	0.9834	-0.0334	0.8902	-0.0071
B7	45.68	63.16	71.8	23.2	8.80	10.91	0.9559	-0.0298	0.8441	-0.0067
B8	44.68	62.21	70.4	23.9	8.63	10.95	0.9577	-0.0290	0.8508	-0.0066

Where, DE= Dissolution efficiency after 10, 30 and 60 min, DP= percent of drug dissolved after 30 min (DP), T_{50} = time necessary to dissolve 50% drug, RDR = relative dissolution rate, r = Coefficient of correlation; K_1, K_{HC} = release rate constants for First order and Hixson Crowell's model respectively



(a) **(b)**

Fig. 7: *In vitro* dissolution profiles of NVP alone, NVP:β-CD physical mixture (B1) and NVP:β-CD binary complex (B2) in (a) 0.1N HCl and (b) pH 6.8



(a) **(b)**

Fig. 8: **Comparative *in vitro* dissolution profiles of NVP:β-CD:PVP ternary systems in (a) 0.1N HCl and (b) pH 6.8**

CONCLUSION

Both binary and ternary solid complexes exhibited higher dissolution rates in pH 1.2 and pH 6.8 than their corresponding physical mixtures and also the pure drug. Ternary system with PVP 5% showed the most significant effect on the dissolution properties of the model drug. Thus, inclusion complexes of NVP:β-CD:PVP systems are suitable to achieve sufficient solubility along the whole gastro-intestinal tract, which is a crucial step in the development of NVP formulations. The present results suggest that the prepared solid complexes reflect the vital role of β-CD to improve the solubility and dissolution rate of NVP both in acidic and intestinal pH via complexation process, which could minimize the variable dissolution rates with increase in oral bioavailability. Further, ternary complexes offer an advantage of using less amount of CD to be needed to solubilize a given amount of model drug in the development of pharmaceutical dosage formulations.

ACKNOWLEDGEMENT

The authors are thankful to Academy of Medical Education, V. L. College of pharmacy, Raichur, Karnataka, INDIA for its generous financial support to this research work.

REFERENCES

1. Davis ME, Brewster ME. Cyclodextrin based pharmaceutics: Past, present and future. Nat Rev Drug Discovery 2004; 3:1023-1035.
2. Craig DQM. The mechanisms of drug release from solid dispersions in water-soluble polymers. Int J Pharm 2002; 231:131-144.
3. Serajuddin ATM. Solid dispersions of poorly water soluble drugs: Early promises, subsequent problems and recent breakthroughs. J Pharm Sci 1999;88(10):1058-1066.

4. Loftsson T, Duchene D. Cyclodextrins and their pharmaceutical applications. Int J Pharm 2007; 329:1-11.
5. Nelson M, Waters L, John L. Non-nucleoside reverse transcriptase inhibitors: a review. Int J Clin Pract 2007; 61(1):105-118.
6. Kusum VD, Roopa SP. Antiretrovirals: Need for an Effective Drug Delivery. Ind J Pharm Sci 2006**;** 68(1):1-6.
7. Kasim NA, Whitehouse M, Ramachandran C, Bermejo M, Lennernas H, Hussain AS, et al. Molecular properties of WHO essential drugs and provisional biopharmaceutical classification. Mol Pharm 2004; 1(1):85-96.
8. Lamson MJ, Sabo JP, MacGregor TR, Pav TW, Rowland L, Hawi A. Single dose pharmacokinetics and bioavailability of nevirapine in healthy volunteers. Biopharm Drug Dispos 1995; 20:285-291.
9. Szejtli J, Smolen F, Ball L, Eds., Molecular Entrapment and Release Properties of Drugs by Cyclodextrins and Congeners. Controlled Drug Bioavailability. 1985; 5:365-420.
10. Pitha, Szente L, Bruck SD, Eds., Molecular Encapsulation by Cyclodextrins and Congeners. Controlled Drug Delivery, 1983; 1:125-148.
11. Loftsson T. Increasing the cyclodextrin complexation of drugs and drug bioavailability through addition of water-soluble polymers. Pharmazie 1998; 53:733-740.
12. Loftsson T, Masson M, Sigurjonsdottir JF. Methods to enhance the complexation efficiency of cyclodextrins. STP Pharma Sci 1999; 9:237-42.
13. Siguroardottir AM, Loftsson T. The effect of polyvinylpyrrolidone on cyclodextrin complexation of hydrocortisone and its diffusion through hairless mouse skin. Int J Pharm 1995; 126:73-78.
14. Loftsson T, Masson M. The effects of water-soluble polymers on cyclodextrins and cyclodextrin solubilization of drugs. J Drug Del Sci Tech 2004; 14:35-43.
15. Higuchi T, Connors KA. Phase Solubility Techniques. Adv Anal Chem Instrum 1965; 4:117-212.
16. Hawi A, Bell G. Preformulation studies of nevirapine, a reverse transcriptase inhibitor. Pharm Res 1994;11(Suppl):236.
17. Mura P, Faucci, MT, Manderioli A, Bramanti G. Multicomponent system of econazole with hydroxyacids and cyclodextrins. J Incl Phenom Macro 2001; 39:131-138.
18. Macha S, Yong C, MacGregor T, Castles M, Quinson A, Rouyrre N, Wilding I. Assessment of nevirapine bioavailability from targeted sites in the human gastrointestinal tract. J Clin Pharmacol 2009; 49:1417-1425.
19. Khan KA. The concept of dissolution efficiency. J Pharm Pharmacol 1975; 27:48-49.