



## SOLUBILITY ENHANCEMENT OF CYPROHEPTADINE BY FORMULATING INCLUSION COMPLEXES

Atul K. Gupta<sup>\*1</sup>, Susheel Kumar Sehrawat<sup>1</sup>, Ashok Kumar<sup>1</sup>, D.N. Mishra<sup>2</sup>

<sup>1</sup>M. M. College of Pharmacy, M. M. University, Mullana, Haryana, India.

<sup>2</sup>Department of Pharmaceutical Sciences, G. J. U. S & T., Hisar, Haryana, India.

\*Corresponding author's E-mail: [atul\\_mullana1979@yahoo.co.in](mailto:atul_mullana1979@yahoo.co.in)

Accepted on: 08-04-2011; Finalized on: 11-07-2011.

### ABSTRACT

The main object of this study was to enhance the solubility of cyproheptadine by formulating the inclusion complex (using  $\beta$ -Cyclodextrin). The prepared complexes were characterized by UV absorption spectroscopic studies and Fourier-transform infrared spectroscopy and does not shows any chemical changes in drug in physical mixture and solid dispersion. The phase solubility enhancement studies were done at different physiological pH (at gastric pH and at pH 6.8). The solubility of pure drug was found to be 2.09mg/ml at pH 6.8 and significantly decreases in acidic pH ( $0.72 \pm 0.01$ mg/ml) and become enhanced in solid dispersion as compare to the physical mixture and found to be increases linearly with  $\beta$ -Cyclodextrin ratio and become independent of the physiological pH at higher concentration of carrier (9.69mg/ml at pH 6.8, 9.12mg/ml at gastric pH). *In vitro* dissolution rate studies shows the drug releases from pure drug very slow (94% in 90min.) as compare to the solid dispersion in which more than 90% of drug dissolve within 5min.

**Keywords:** Cyproheptadine,  $\beta$ -cyclodextrin, solid dispersion, phase solubility.

### INTRODUCTION

Biopharmaceutics Classification System (BCS) is a fundamental guideline that is associated with the drug dissolution and absorption model, identifies the key parameters controlling the drug absorption and bioavailability and classified the drugs in four classes based solely on its solubility and intestinal permeability<sup>1</sup>. Class I drugs exhibit a high Absorption number and a high Dissolution number. *In-vivo* drug dissolution is then a rate-limiting step for absorption (except at very high Dose number)<sup>2</sup>. The absorption for Class II drugs is usually slower than Class I and occurs over a longer period of time. For Class III drugs, permeability is the rate-controlling step in drug absorption<sup>3</sup>. In class II, solubility enhancement is a genuine challenge for formulators. About 40% of marketed drugs are classified as "practically insoluble," according to Takagi<sup>4</sup>, one third of drugs in development are poorly soluble and two thirds of synthesized drugs have low solubility<sup>5</sup>. Many techniques have been developed to overcome these solubility issues and, by association, bioavailability when associated with good permeability. Cyclodextrin complexation is one of these techniques. The ability of cyclodextrins to form inclusion compounds through molecular encapsulation has been known for many years. Cyproheptadine hydrochloride (CPH) is 4-(5H-dibenzo [a,d] cyclohepten-5-ylidene)-1-methyl piperidine hydrochloride, potent antihistaminic, antimuscarine and antiserotonic drug and shows sedative with calcium-channel blocking along with stimulate appetite<sup>7,8</sup> and weight gain activity in children and adults and sometimes used in adjunct therapy in children who are taking human growth hormone<sup>9</sup>. Cyproheptadine is highly lipophilic drug, so its HCl salt is used which is slightly soluble in aqueous medium and the solubility decreases in acidic medium (stomach fluid) due

to counter ion effect. Hence this study aims to enhance the aqueous solubility and dissolution rate of cyproheptadine by forming an inclusion complex with  $\beta$ -Cyclodextrin and their physico-chemical properties were investigated.

### MATERIALS AND METHODS

#### Materials

Cyproheptadine HCl, beta-cyclodextrin was obtained as a gift sample from Himalayan Laboratories, H.P., INDIA). All other materials were of analytical grade.

#### Preparation of Physical mixtures

The physical mixture of Cyproheptadine HCl with beta cyclodextrin in different ratios (Table 1) were prepared by mixing of pulverized and sieved (#100) drug with weighed amount of BCD in a modified rotary flask shaker at 30 rpm (inclined an angle of 50°) for 2 hours followed by analysis for phase solubility enhancement<sup>10</sup>.

#### Preparation of solid dispersion

Solid dispersion of Cyproheptadine HCl with  $\beta$ -Cyclodextrin was prepared by solvent evaporation method. Accurately weighed amount of Cyproheptadine HCl (table 1) and  $\beta$ -cyclodextrin were dissolved in 10ml of ethanol. The prepared dispersions were poured into the petridish and solvent was evaporated in vacuum oven at 600mm Hg at  $60^\circ\text{C} \pm 0.5^\circ\text{C}$  for 30min. Dispersion was collected from petridish with help of spatula. The dried mass was pulverized and sieved through mesh #100 for evaluation.



## Evaluation

### Fourier-transform infrared (FTIR) spectroscopy

Physical mixture and solid dispersion of cyproheptadine and  $\beta$ -Cyclodextrin in 1:2w/w ratio was used for FTIR study using a FTIR Spectrophotometer (Shimadzu, Tokyo, Japan). The procedure consisted of dispersing samples and compressing into discs by applying a pressure of 5t for 5 min in a hydraulic press. The pellet was placed in the light path and the spectrum was recorded<sup>11</sup>.

### UV Spectroscopy Studies

UV absorption spectroscopy studies of Cyproheptadine HCl and  $\beta$ -Cyclodextrin complexes were carried out using UV-VIS scanning spectrophotometer (Shimadzu UV-2101PC, Japan)<sup>12</sup>. UV absorption spectra were recorded using pure Cyproheptadine HCl (conc. of 30mcg/ml in 0.1N HCl) and compared with the  $\beta$ -Cyclodextrin physical mixture and solid dispersion (ratio 1:6w/w).

### Phase solubility studies

Phase solubility studies were carried out by adding known excess amount of cyproheptadine HCl with  $\beta$ -Cyclodextrin in different ratios (physical mixture and solid dispersion separately) in 3ml of gastric fluid (0.1N HCl) and placed overnight in water bath incubator shaker (30rpm,  $37^{0\pm 0.5^0}C$ )<sup>13</sup>. The solution was filtered through micro-syringe filters (#0.22micron), diluted and studied by UV-VIS spectroscopic method (Shimadzu UV-2101PC, Japan) at 286nm. The same study was repeated in 3ml of Phosphate buffer at pH 6.8.

### In-Vitro Dissolution Rate Study

*In-vitro* dissolution studies of pure drug, physical mixture and complexes were carried out in 900 ml of 0.1 N HCl (pH 1.2), using USP 8-stage dissolution rate apparatus (USP XXI/XXII, TDT08L ELECTROLAB, Mumbai, INDIA). Samples equivalent to 500 mg of cyproheptadine HCl was tied in muslin cloth and kept in basket and rotated at 50rpm<sup>14</sup>. Samples at different time intervals were taken, filtered with micro-syringe filter (0.22mm) and analyzed.

**Table1:** Composition of Batches Containing Cyproheptadine and  $\beta$ -Cyclodextrin

Batches for Physical Mixture	Cyproheptadine (mg)	$\beta$ -Cyclodextrin (mg)	Drug Carrier Ratio (w/w)	Batches for Solid dispersion
PM1	25	50	1:2	SD1
PM2	25	100	1:4	SD2
PM3	25	150	1:6	SD3
PM4	25	200	1:8	SD4

**Table 2:** Phase solubility studies at different physiological pH

Ratio (w/w)	Solubility (mg/ml)			
	at Gastric pH		at pH 6.8	
	P.M. of BCD	S.D. of BCD	P.M. of BCD	S.D. of BCD
Pure Drug	0.72 $\pm$ 0.01	0.72 $\pm$ 0.02	2.09 $\pm$ 0.09	2.09 $\pm$ 0.04
1:2	1.93 $\pm$ 0.06	2.73 $\pm$ 0.04	3.34 $\pm$ 0.07	4.33 $\pm$ 0.09
1:4	3.49 $\pm$ 0.11	5.19 $\pm$ 0.08	4.42 $\pm$ 0.05	6.49 $\pm$ 0.08
1:6	4.95 $\pm$ 0.10	7.32 $\pm$ 0.11	5.73 $\pm$ 0.11	8.72 $\pm$ 0.11
1:8	6.63 $\pm$ 0.14	9.12 $\pm$ 0.13	7.45 $\pm$ 0.12	9.69 $\pm$ 0.15

Number of trial (n) = 6

**Table3:** *In Vitro* Dissolution Rate studies of solid dispersion in comparison to pure drug

Time (min)	Percentage Cumulative Drug Release				
	Pure Drug	SD1	SD2	SD3	SD4
2	18.35 $\pm$ 0.03	62.45 $\pm$ 0.02	74.33 $\pm$ 0.03	80.09 $\pm$ 0.05	80.45 $\pm$ 0.04
5	26.56 $\pm$ 0.05	74.68 $\pm$ 0.03	80.69 $\pm$ 0.04	90.81 $\pm$ 0.06	91.52 $\pm$ 0.03
10	46.27 $\pm$ 0.06	82.85 $\pm$ 0.05	91.16 $\pm$ 0.06	95.54 $\pm$ 0.07	98.06 $\pm$ 0.06
20	64.72 $\pm$ 0.08	91.71 $\pm$ 0.07	96.62 $\pm$ 0.08	98.08 $\pm$ 0.09	99.00 $\pm$ 0.07
40	81.17 $\pm$ 0.07	96.98 $\pm$ 0.08	98.99 $\pm$ 0.09	99.90 $\pm$ 0.09	99.9 $\pm$ 0.09
60	92.78 $\pm$ 0.10	98.99 $\pm$ 0.07	99.90 $\pm$ 0.07	99.91 $\pm$ 0.08	99.91 $\pm$ 0.08
90	94.47 $\pm$ 0.12	99.9 $\pm$ 0.09	99.91 $\pm$ 0.09	99.91 $\pm$ 0.09	99.91 $\pm$ 0.09



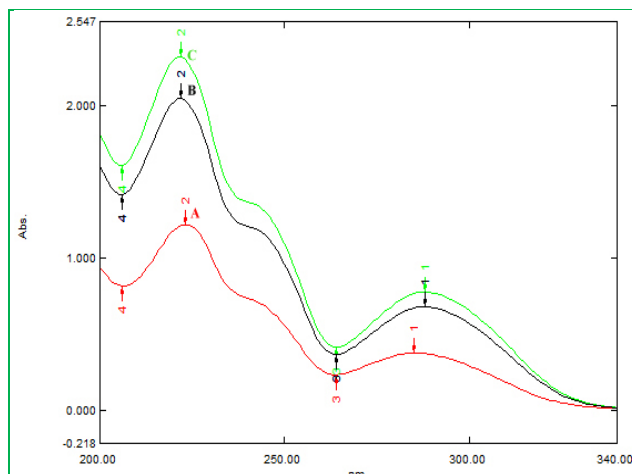
## RESULTS AND DISCUSSION

### Fourier-transform infrared (FTIR) spectroscopy

From FTIR spectra, the characteristic absorption peaks of Cyproheptadine was found at 3556  $\text{cm}^{-1}$  (N–H asymmetrical stretch), 3286  $\text{cm}^{-1}$  (N–H symmetric stretch), 1238  $\text{cm}^{-1}$  (C–N stretch), 1589, 1060 and 603  $\text{cm}^{-1}$  (aromatic C–H stretch). In physical mixture and solid dispersion, the peaks of Cyproheptadine were not prone and intensity also reduced with no change in peaks in case of PM and SD showing inclusion type of complex formed.

### UV Spectroscopy Studies

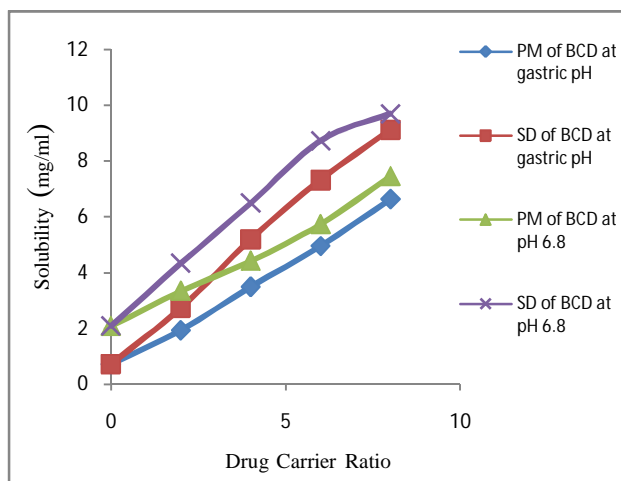
UV Spectroscopy Studies of pure drug, Drug BCD Physical mixture and Solid dispersion are shown in figure 1 and shows two dominant peaks (peak 1 at 286nm, peak 2 at 222nm). Peak 1 of physical mixture and solid dispersion shows negligible shift in wavelength from 286nm to 288nm and peak 2 shows no shift in wavelength, showing no chemical interaction with drug. Physical mixture and solid dispersion show more absorbance as compare to pure drug, which indicate that the solubility of drug is being increased with BCD in physical mixture and solid dispersion as well.



**Figure 1:** UV Spectroscopy Studies, A. spectra of pure drug, B. spectra of physical mixture and C. spectra of solid dispersion

### Phase solubility studies

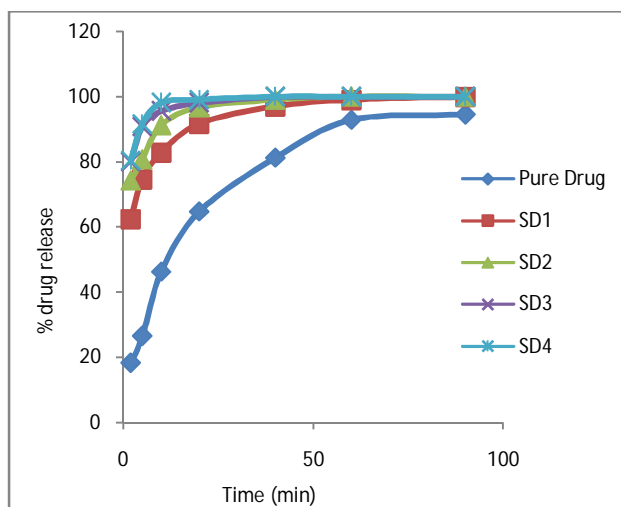
The solubility of pure drug was found to be 2.09mg/ml at pH 6.8 and significantly decreases in acidic pH ( $0.72 \pm 0.01 \text{mg/ml}$ ) due to counter ion effect (Fig. 2). By forming inclusion complex with  $\beta$ -Cyclodextrin in different ratios, the solubility was much more enhance in solid dispersion as compare to the physical mixture and found to be increases linearly with  $\beta$ -Cyclodextrin and become independent of the physiological pH at higher concentration (9.69mg/ml at pH 6.8, 9.12mg/ml at gastric pH).



**Figure 2:** Phase solubility studies at different physiological pH

### In-vitro Dissolution Rate Study

*In-vitro* dissolution rate studies show that the drug releases from pure drug was slow (94% in 90min) as compare to the solid dispersion in which more than 90% of drug dissolve within 5 to 20min (Fig. 3). Drug release was found to very fast with increases in  $\beta$ -Cyclodextrin ratio (In SD4 more than 90% drug releases within 5 minutes).



**Figure 3:** *In Vitro* Dissolution Rate studies of solid dispersion in comparison to pure drug

## CONCLUSION

Inclusion complex with  $\beta$ -Cyclodextrin successfully enhanced the solubility of the cyproheptadine HCL and it is much more enhance by preparing solid dispersion as compare to the physical mixture and this solubility enhancement was also independent of the physiological pH. So inclusion complexes prepared by solid dispersion technique can be used for the water soluble salt drugs whose solubility decreases in gastric fluid.

## REFERENCES

1. Amidon GL, Lennernas H, Shah VP, Crison JR. A theoretical basis for a biopharmaceutic drug classification: The correlation of *in vitro* drug product dissolution and *in vivo* bioavailability. *Pharmaceutical Research*. 12(3); 1995: 413-419.
2. Young D, Devane JG, Butler J. *In Vitro-In Vivo Correlations*, New York: Plenum press, 82(2); 1997: 132-137.
3. Amidon GL, Robinson JR, Williams RL. (Eds.). *Scientific Foundations for Regulating Drug Product Quality*, American Association of Pharmaceutical Scientists, Alexandria, Virginia: AAPS Press, 36; 1997: 187–203.
4. Sweetman SC. *Martindale-The Complete Drug Reference*. Edition 33<sup>rd</sup>. Pharmaceutical Press, London. 2002: 414.
5. Reynolds JEF. *Martindale: The Extra Pharmacopoeia*. Edition 28<sup>th</sup>. The Pharmaceutical Press, London. 1982: 293-294.
6. Stiel JN, Liddle GW, Lacy WW. Studies on mechanism of cyproheptadine-induced weight gain in human subjects *Metabolism*. 19; 1970: 192-200.
7. Lefkowitz JB. Cyclooxygenase 2-specificity and its clinical implications. *Am. J. Med*. 106; 1999: 43.
8. T.K. Murthy, M.N. Reddy, D.G. Shankar, Several analytical methods for the estimation of tizanidine, *Ind. J. Pharm. Sci*. 63; 2001, 521.
9. Joanna D, Hanna H, Cyproheptadine ion-selective electrodes and their applications in some pharmaceutical formulations, *Desalination*, 163:2004, 119-125
10. Swati R, Sanjay KJ, Solubility enhancement of celecoxib using  $\beta$ -cyclodextrin inclusion complexes. *European Journal of Pharmaceutics and Biopharmaceutics*. 57; 2004: 263–267.
11. Ravikumar, Sachin RP, Patil MB, Mahesh SP, Mahalaxmi R. Design and characterization of aceclofenac mouth dissolving tablets by effervescent formulation approach. *Der Pharmacia Lettre*, 2 (1); 2010: 220-236.
12. Katrina G, Ljiljana K, Angel S, Kristina M. Drug dissolution profiles and physico-chemical stability evaluation of controlled-release solid dispersion granules. *Bulletin of the Chemists and Technologists of Macedonia*. 17 (1); 1998: 33–39.
13. Higuchi T, Connors KA. Phase-solubility techniques. *Adv. Anal. Chem. Instrum*. 4; 1965: 117–212.
14. Hazem AH, Ali HA, Baboucarr J, Alaa AH, Gaber AR, Enhancement of dissolution amount and *in vivo* bioavailability of itraconazole by complexation with  $\beta$ -cyclodextrin using supercritical carbon dioxide. *Journal of Pharmaceutical and Biomedical Analysis*. 45; 2007: 243–250.

\*\*\*\*\*

