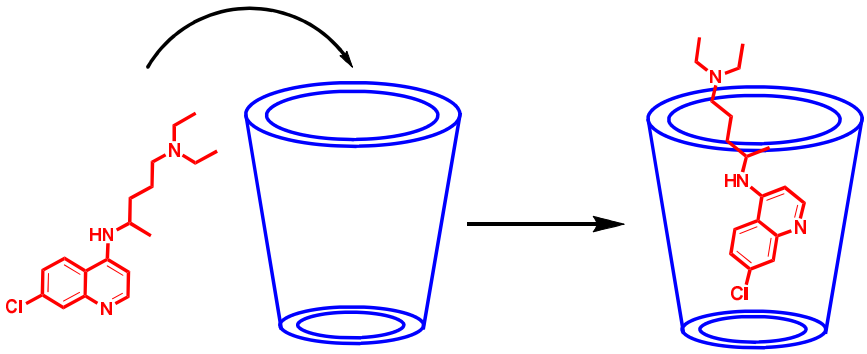
CHAPTER VII

Exploration of inclusion complexes of a vital drug with cyclodextrins

The main objective of this research article is the investigation of formation of inclusion complex of α and β-cyclodextrin with the drug chloroquine diphosphate. The solubility, versatility and the biological activity of the drug molecule is enhanced and some of its side effect is reduced after encapsulation by the CD molecule. The 1:1 stoichiometry of the inclusion complex was determined with Job's method by UV-Visible spectroscopy. Surface tension and conductance study also support this fact. ^{1}H NMR study, FT-IR spectra and HRMS spectra confirms the inclusion phenomenon. The binding constant was determined using the Benesi–Hildebrand method, while the thermodynamic parameters have been estimated with the help of van't Hoff equation for both the inclusion complexes. The negative values of G^{o} indicate that the formation of inclusion complexes is spontaneous. The association constant is found higher in case of β-cyclodextrin which was explained on the basis of their molecular structure.



VII.1 Introduction

Cyclodextrins (CD) are one of the most facinating host molecules in supramolecular chemistry.[1] They have the ability to form stable inclusion complexes (IC) with various guest molecules including drugs, amino acids, vitamins, ionic liquids, polymers, metal co-ordinated inorganic complexes etc.[2-7] The hydrophobic interactions between the cavity of cyclodextrin and hydrophobic part of the guest molecules are responsible for the formation of this type of stable host-guest inclusion complexes. The weak intermolecular forces acting between the host and guest molecules are van der waals force, dipole-dipole interaction, electrostatic and hydrogen bonding interactions.[8] They are extensively used in the pharmaceutical industry as the drug carrier to modify solubility, stability and bioavailability of biologically active molecules.[9] CDs are non-toxic and considered safe to humans. Cyclodextrin based inclusion is the best method to prepare inclusion complexes to improve the physicochemical properties of the drug molecule.[10] CDs are macro cyclic water soluble oligosaccharides composed of six (α-CD), seven (β-CD) and eight (γ-CD) glucopyranose units linked through α-(1-4) bond.[11] CD has a shape of truncated cone with hydrophobic interior and polar hydrophilic rims. Primary hydroxyl groups are situated at narrow end and secondary hydroxyl groups are placed around the wider end (Scheme VII.2).[12] This type of unique structure permits them to encapsulate a wide range of guest molecules.

The drug chloroquine diphosphate (CDP) is a medicine that is primarily used to prevent and treat malaria (Scheme 1). Certain types of complicated cases of malaria typically require additional medication. CDP is ocasionally used for amebiasis, rehumatoid artharitis and lupus erythematosus.[13,14] It is a member of drug class 4-aminoquinoline. It appears to be safe during pregnancy time. It works against the asexual form of malaria inside the red blood cell. This drug has some common side effects including mussel problems, loss of appetite, diarrhea and skin rash.[15] Controlled release of this drug using CD as the encapsulating agent can minimize these side effects. In order to have biological

activity drug molecule should be able to penetrate the lipophilic membrane. By forming inclusion complex CD can deliver the drug to the physiological target without losing any of its bioactivity.[9]

In the present study we attempt to ascertain the formation and nature of IC of α and β-CD with CDP in aqueous environment by spectroscopic and physicochemical studies. Our aim is to explore the formation, carrying and controlled release of this drug by forming IC with CD without any chemical and biological modification of the guest molecule. To the best of our knowledge no investigation concerning inclusion complex formation between α or β-CD and CDP has been performed so far.

VII.2. Experimental Section

VII.2.1. Source and Purity of Samples

Chloroquinediphosphate, α-cyclodextrin and β-cyclodextrin of puriss grade were purchased from SigmaAldrich, Germany and used in the experiment as it was. The mass fraction purity of CDP, α-cyclodextrin and β-cyclodextrin were ≥ 0.98, ≥ 0.99 and ≥ 0.98 respectively.

VII.2.2. Apparatus and Procedure

Triply distilled and degassed water has been used for checking the solubility of the drug CDP, α and β-CD. The drug and both the CD molecules are freely soluble in water. All the solutions are prepared by mass (weighed by Mettler Toledo AG-285 with uncertainty 0.0003g) at 298.15 K temperature. Solutions are made with proper care to avoid weight loss by evaporation.

^{1}H NMR spectra were recorded at 300MHz in D_2O solution using BRUKER ADVANCE instruments at 298K. Signals are quoted as δ values in ppm using residual protonated solvent signals as internal standard (D_2O: δ 4.79 ppm).

Surface tension experiment was carried out by using platinum ring detachment Tensiometer (K9, KRUSS; Germany) at 298.15 K. During experiment

the temperature of the system was kept constant by circulating auto thermostat water through double wall glass vessel containing the solution.

The specific conductance values of the solutions are measured by Mettler Toledo Seven Multi conductivity meter. The uncertainty of the result was approximately ±1.0 μS m^{-1}. The experiments were done in an thermostated water bath keeping the temperature constant at 298.15 K.

UV–visible spectra were taken by JASCO V-530 UV/VIS Spectrophotometer, with an uncertainty in wavelength as ±2 nm. A digital thermostat was used to keep the temperature constant.

Fourier transform infrared (FT-IR) spectra were taken on a Perkin Elmer FT-IR spectrometer applying the KBr disk technique. Samples were prepared as KBr disks with 1 mg complex and 100 mg KBr. The scanning range was 4000 400 cm^{1} at room temperature.

HRMS analyses were executed with Q-TOF high resolution instrument by positive mode electro-spray ionization.

Inclusion complexes [α-CD+CDP and β-CD+CDP] were also prepared in the solid state taking 1:1 molar ratio of the drug CDP and CDs. For this purpose 1.0 mmol CD was dissolved in 20 mL water and 1.0 mmol drug CDP was dissolved in 20 mL water. These two solutions are allowed to stir separately in a magnetic stirrer for 3 hours. The aqueous solution of the drug was added dropwise into the aqueous solution of the cyclodextrin. Then the mixture is constantly stirred for 4 days at 40-45 ^{o}C. The solution was filtered at that temperature, then cooled to 5^{o}C and kept for 24 hrs. The resulting suspension was filtered and the white polycrystalline powder was found, it was dried in air.

VII.3. Result and discussion

VII.3.1. ^{1}H NMR spectra analysis

The formation of inclusion complex and stoichiometry is confirmed by ^{1}H NMR study. Insertion of drug molecule into the hydrophobic cavity of CD molecules consequences the chemical shift of both the guest and host molecule.[16,17] Aromatic guest molecules results dimagnetic shielding of the aromatic protons as a result of interaction with the CD protons after inclusion. The location of different protons in the CD molecules are shown in scheme VII.3. H3 and H5 protons are located inside the cavity near the wider rim and narrower rim respectively. The other protons H1,H2 and H4 are situated outside the CD molecule.[18-20] The respective δ values of the drug CDP, α-CD, β-CD and inclusion complexes are reported in table VII.1. The protons of CD and aromatic protons of CDP show considarable upfield shift in 1:1 inclusion complex of the drug and CD (Figure VII.1, VII.2). Chemical shift indicates that the aromatic moiety of the drug interacts more with the H3 protons than H5 suggesting the drug molecule enters in the hydrophobic cavity from wider end. The shift in δ value of both the CD perhaps due to change of environment after inclusion complex formation. The upfield shift is basically due to formation of hydrogen bond of the guest with rim hydroxyl groups of CD. The shielding of H3 and H5 protons are due to ring current effect as the aromatic part of the ring was positioned perpendicular to the to the cyclodextrin molecule cavity.[2] These results indicate the formation of inclusion complex.

VII.3.2. Surface tension study

Surface tension nicely explains the formation of IC.[21,22] CD being hydrophobic in nature do not show any considarable change of surface tension (γ) of aqueous solution.[23] The studied drug molecule CDP exsist as charged structure and behave like surfactant. Therefore when CDP forms ICs with the host molecules remarkable change in surface tension should be observed. The γ value of this drug is lower than pure water. In this study the γ value of a series of

solution of CDP with increasing concentration of α and β-CD have been measured at 298.15 K(Table VII.2,VII.3). The γ value regularly increases for both the CD molecules, perhaps due to the insertion of the surfactant like drug molecules from solution to the hydrophobic cavity of the host molecules (Scheme VII.4). There is a single distinguisable break in both the surface tension plots(Figure VII.3) which suggests the formation of IC.[24,25] The concentration of the CDs and CDP at the break point(Table VII.6) is approximately 1:1 which further confirms the stoichiometric ratio of the IC to be 1:1. More number of breaks in the surface tension plot indicates more complex stoichiometry of the IC.

VII.3.3. Conductivity study

Conductivity (κ) study provides us valuable information about the stoichiometry of the inclusion complex.[26,27] The aqueous solution of the drug CDP shows considarable conductivity due to the exsistence as a chaged atructure. For this purpose the conductivity of a series of solutions of this drug with increasing concentration of α and β-CD have been measured at 298.15K. With addition of CD solution the conductivity regularly decreases and after a sharp break point the conductivity value almost becomes constant (Table VII.4-VII.5). Similar results are obtained in case of both the host cyclodextrin molecules (FigureVII.4).The appearance of sharp break points suggest the formation of inclusion complex.[28,3] The decrease in the κ value probably due to the encapsulation of the guest drug molecules in the hydrophobic cavity of CD. The values of κ and corresponding concentration of the host CD molecules are reported in table VII.6 which suggests that the ratio of concentration of CDP and the CD at the break point is almost 1:1. This confirms the stoichiometry of the IC to be 1:1.

The break point indicates certain concentration where maximum number of drug molecules are inserted in CD molecule ever before.[18] A dynamic equilibrium exsists between the guest drug and host CD molecules.

Drug + Cyclodextrin Inclusion complex

Maximum inclusion takes place at break point, after it the concentration of CD is higher than the concentration of CDP and the equilibrium shifts more towards the formation of IC.

VII.3.4. Job plots confirm the stoichiometric ratio

Job plot method can be employed to predict the stoichiometry of the inclusion complexes formed [29,30]. Job plots are constructed by the continuous variation method with the help of UV-Visible spectroscopy. For this purpose a series of solutions for the drug CDP and both the CDs was prepared with the variation of the mole fraction of the drug molecule (Table VII.7-VII.8). A× R was plotted against R to obtain these plots where A is the difference in the absorbance of the drug without and with the presence of host CDs and R= [CDP]/ ([CDP] + [CD]).[31,21] Absorbance values are recorded at the respective λ_{max} for each of the solutions at 298.15K. The stoichiometry of the inclusion complex was found from the R values at maximum deviation that is the ratio of host and guest is R=0.5 for 1: 1 complexes; R=0.33 for 1:2complexes; R=0.66 for 2: 1 complexes. [8] From fig VII.5, it has been found that the maxima is R=0.5 for each plot which confirms the 1:1 stoichiometry of the IC.

VII.3.5. Association constant and thermodynamic parameters

Association constants have been determined for both the CDP-CD ICs from UV-Visible spectra.[32] When the drug molecule is encapsulated in the CD cavity there is a change from polar aqueous environment to the apolar hydrophobic cavity. As a result the molar exticton coefficient (∈) of the drug molecule changes after inclusion. The difference in absorbance intensity (A) of the drug is observed as a function of concentration of both the CD molecules to determine the K_a value (Table VII.9,VII.10, VII.11). Double reciprocal plots have been generated on the basis of Benesi–Hildebrand method for 1:1 host–guest ICs [33,8](Figure VII.6 ,VII.7,VII.8 equation 1)

$$\frac{1}{A} = \frac{1}{\varepsilon[CDP]K_a} \times \frac{1}{[CD]} + \frac{1}{\varepsilon[CDP]} \qquad (1)$$

The above mentioned equation is a linear equation and K_a value is calculated dividing the intercept by the slope from double-reciprocal plots for both the ICs.[34]

The thermodynamic parameters can be also evaluated using the K_a values found at various temperatures on the basis of above method using Van't Hoff Equation mentioned below [35]

$$\ln K_a = \frac{H^o}{RT} + \frac{S^o}{R} \qquad (2)$$

Linear relationship exsists between lnKa and 1/T. The H° value depends upon the equilibrium constant at different temperatures. The thermodynamic parameters are listed in table VII.11. The negative value of G for both the systems indicates that formation of inclusion complex is spontaneous. The negative H value suggests that the process is exothermic.[31] The decrease in S value implies that the process is entropy controlled but not entropy driven. The effect of negative value of S is adverse for the process and overcome by more negative value of G. the overall process is thermodynamically favorable.

VII.3.6. FT-IR spectra analysis

FT-IR spectral study helps to determine the interaction between the host and guest in the IC.[36,37] The drug CDP and the CDs form solid inclusion complexes by non covalent interactions such as hydrophobic interactions, Vanderwaals interaction and hydrogen bonding. After encapsulation of the drug molecule by the CDs, the resulting absorption bands from the included part of the guest molecule are shifted in their position.[38,39] The FT-IR spectrum of pure CDP, α-CD,β-CD and the two inclusion complexes are shown in the figure VII.9 and VII.10. The various frequencies of the above mentioned compounds are reported in table VII.12. The spectrum of CDP is charecterised by peaks of –N-H, -C=N, -C-N,-C-Cl, aromatic –C=C, -C-H of $-CH_2$ and $-CH_3$ etc. The broad –O-H stretching frequency for α-CD and β-CD was observed at 3412.10 and 3349.84 cm^{-1} respectively. In the ICs the –O-H frequency shifted to lower regeion i.e.;

3384.08 and 3329.76 cm^{-1} for α-CD and β-CD respectively. This shift occurs probably due to involvement of the –O-H groups of both the CDs in hydrogen bonding with the guest drug molecule. The peaks of -C=N, -C-N, aromatic –C=C of the drug CDP are shifted in the spectra of the ICs. The changes in the FT-IR spectra of ICs are due to the restriction of the vibration of free CDP molecules as the quinoline part of the drug with Cl groups is encapsulated in the cavity of CD molecules.[40,41] No additional peaks are found in the spectra of the ICs. This fact again confirms that only non covalent interaction exsists between the host and guest, only Vanderwaal's interaction are present.

VII.3.7. HRMS study

ESI–MS study is a widely accepted technique to characterize host–guest IC.[42,43] The result of ESI–MS study can be easily compared with other experiments such as Job Plot titration obtained from UV-Visible spectra. The solid ICs synthesized by the method mentioned in the experimental section are used for MS study. Figure VII.11,VII.12 shows the MS spectra of the two CDP-CD inclusion complexes. The intense peaks at m/z 1293.54 and 1454.61 refers to the proton adduct of CDP-α-CD IC and CDP-β-CD IC Since no other intense peaks are observed at higher values the ESI-MS experiment verifies the 1:1 stoichiometry of the ICs.[44,45]

VII.4. Conclusion

The present study confirms that chloroquine diphosphate form inclusion complex with both α and β-CD in aqueous medium and in solid state. The two IC's can be used for controlled release of this drug. ^{1}H-NMR study confirms the inclusion phenomenon whereas surface tension, conductivity and Job's plot variation suggest the 1:1 stoichiometry of the IC's. FT-IR spectra and mass spectra also supported the formation of IC. The binding constants and thermodynamic parameters have been evaluated from UV-Visible spectroscopy which is higher for β-CD. The overall inclusion process is thermodynamically

favorable. These two IC's have potential application in the pharmaceutical industries and biomedical fields.

Tables

Table VII.1. ^{1}H NMR data of CDP, α-CD, β-CD and inclusion complexes

α-Cyclodextrin: (500 MHz, Solv: D_2O) δ=3.48-3.51 (6H, t, *J*= 9.00 Hz), 3.53-3.56 (6H, dd, *J*=10.00, 3.00 Hz), 3.74-3.83 (18H, m), 3.87-3.91 (6H, t, *J* = 9 Hz), 4.96-4.97 (6H, d, *J* = 3 Hz)
β-Cyclodextrin: (500 MHz, Solv: D_2O) δ=3.49-3.54 (6H, t, *J* = 9.2 Hz), 3.57-3.60 (6H, dd, *J* = 9.6, 3.2 Hz), 3.79-3.84 (18H, m), 3.87-3.92 (6H,t, *J* = 9.2 Hz), 5.00-5.01 (6H, d, *J* = 3.6 Hz)
CDP: (300MHz, Solv: D_2O) δ=1.11-1.16(6H, t, *J* = 6 Hz), 1.29-1.31(3H,d, *J* = 6 Hz),1.72(4H,m), 3.04-3.11(4H,q), 6.74-6.76(1H,d, *J* = 6 Hz),7.51-7.54(1H,m),7.74-7.75(1H,d, *J* = 3 Hz),8.10-8.13(1H,d, *J* = 9 Hz),8.17-8.19(1H,d, *J* = 6 Hz).
CDP-α-CD: (1:1 molar ratio, 300 MHz, Solv: D_2O) :δ=1.10-1.15(6H, t, *J* = 6 Hz), 1.29-1.31(3H,d, *J* = 6 Hz), 1.72(4H,m), 3.04-3.11(4H,q), 3.45-3.51(6H, t, *J* = 9.2 Hz),3.54-3.55(6H, dd, *J* = 9.6, 3.2 Hz),3.75-3.78(18H, m),3.82-3.88((6H,t, *J* = 9.2 Hz),4.97-4.98(6H, d, *J* = 3.6 Hz), 6.72-6.74(1H,d, *J* = 6 Hz),7.49-7.52(1H,m),7.71-7.72(1H,d, *J* = 3 Hz),8.08-8.11(1H,d, *J* = 9 Hz),8.14-8.16(1H,d, *J* = 6 Hz).
CDP-β-CD: (1:1 molar ratio, 300 MHz, Solv: D_2O) :δ=1.11-1.16(6H, t, *J* = 6 Hz), 1.29-1.31(3H,d, *J* = 6 Hz), 1.72(4H,m), 3.04-3.12(4H,q), 3.43-3.48(6H, t, *J* = 9.2 Hz),3.52-3.57(6H, dd, *J* = 9.6, 3.2 Hz),3.74-3.77(18H, m),3.81-3.87((6H,t, *J* = 9.2 Hz),4.96-4.97(6H, d, *J* = 3.6 Hz), 6.71-6.73(1H,d, *J* = 6 Hz),7.48-7.51(1H,m),7.70-7.71(1H,d, *J* = 3 Hz),8.07-8.10(1H,d, *J* = 9 Hz),8.13-8.15(1H,d, *J* = 6 Hz).

Table VII.2. Data for the surface tension study of CDP-*α*-CD system (concentration of stock solution of CDP= 10mM, concentration of stock solution of *α*-CD = 10mM) at 298.15K[a]

Volm of α-CD (mL)	Total volm (mL)	Conc of CDP (mM)	Conc of α-CD (mM)	ST (mN m^{-1})
0	10	10.000	0.000	62.5
1	11	9.091	0.909	63.8
2	12	8.333	1.667	64.9
3	13	7.692	2.308	65.9
4	14	7.143	2.857	66.8
5	15	6.667	3.333	67.6
6	16	6.250	3.750	68.2
7	17	5.882	4.118	68.8
8	18	5.556	4.444	69.4
9	19	5.263	4.737	69.8
10	20	5.000	5.000	70.3
11	21	4.762	5.238	70.45
12	22	4.545	5.455	70.53
13	23	4.348	5.652	70.64
14	24	4.167	5.833	70.75
15	25	4.000	6.000	70.86
16	26	3.846	6.154	70.94
17	27	3.704	6.296	71.03
18	28	3.571	6.429	71.10
19	29	3.448	6.552	71.15
20	30	3.333	6.667	71.20

[a] Standard uncertainties in temperature (T) = 0.01 K.

Table VII.3. Data for the surface tension study of CDP-*β*-CD system (concentration of stock solution of CDP = 10mM, concentration of stock solution of *β*-CD = 10mM) at 298.15K[a]

Volm of *β*-CD (mL)	Total volm (mL)	Conc of CDP (mM)	Conc of *β*-CD (mM)	ST (mN m^{-1})
0	10	10.000	0.000	62.5
1	11	9.091	0.909	63.9
2	12	8.333	1.667	65.1
3	13	7.692	2.308	66.2
4	14	7.143	2.857	67.1
5	15	6.667	3.333	67.9
6	16	6.250	3.750	68.6
7	17	5.882	4.118	69.2
8	18	5.556	4.444	69.7
9	19	5.263	4.737	70.2
10	20	5.000	5.000	70.7
11	21	4.762	5.238	70.9
12	22	4.545	5.455	71.0
13	23	4.348	5.652	71.2
14	24	4.167	5.833	71.2
15	25	4.000	6.000	71.4
16	26	3.846	6.154	71.4
17	27	3.704	6.296	71.5
18	28	3.571	6.429	71.6
19	29	3.448	6.552	71.7
20	30	3.333	6.667	71.8

[a] Standard uncertainties in temperature (T) = 0.01 K.

Table VII.4. Data for the conductivity study of CDP-*α*CD system (concentration of stock solution of CDP = 10mM, concentration of stock solution of *α*-CD = 10mM) at 298.15K[a]

Volm of *α*-CD (mL)	Total volm (mL)	Conc of CDP (mM)	Conc of *α*-CD (mM)	Conductuvity (mS cm^{-1})
0	10	10.000	0.000	1.090
1	11	9.091	0.909	0.989
2	12	8.333	1.667	0.902
3	13	7.692	2.308	0.824
4	14	7.143	2.857	0.761
5	15	6.667	3.333	0.702
6	16	6.250	3.750	0.659
7	17	5.882	4.118	0.614
8	18	5.556	4.444	0.578
9	19	5.263	4.737	0.539
10	20	5.000	5.000	0.508
11	21	4.762	5.238	0.502
12	22	4.545	5.455	0.495
13	23	4.348	5.652	0.488
14	24	4.167	5.833	0.483
15	25	4.000	6.000	0.478
16	26	3.846	6.154	0.473
17	27	3.704	6.296	0.470
18	28	3.571	6.429	0.467
19	29	3.448	6.552	0.464
20	30	3.333	6.667	0.460

[a] Standard uncertainties in temperature (T) = 0.01 K.

Table VII.5. Data for the conductivity study of CDP-*β*-CD system (concentration of stock solution of CDP = 10mM, concentration of stock solution of *β*-CD = 10mM) at 298.15K[a]

Volm of *β*-CD (mL)	Total volm (mL)	Conc of CDP (mM)	Conc of *β*-CD (mM)	Conductuvity (mS cm^{-1})
0	10	10.000	0.000	1.090
1	11	9.091	0.909	0.978
2	12	8.333	1.667	0.891
3	13	7.692	2.308	0.814
4	14	7.143	2.857	0.745
5	15	6.667	3.333	0.689
6	16	6.250	3.750	0.641
7	17	5.882	4.118	0.595
8	18	5.556	4.444	0.552
9	19	5.263	4.737	0.516
10	20	5.000	5.000	0.491
11	21	4.762	5.238	0.484
12	22	4.545	5.455	0.477
13	23	4.348	5.652	0.471
14	24	4.167	5.833	0.464
15	25	4.000	6.000	0.458
16	26	3.846	6.154	0.453
17	27	3.704	6.296	0.450
18	28	3.571	6.429	0.447
19	29	3.448	6.552	0.445
20	30	3.333	6.667	0.443

[a] Standard uncertainties in temperature (T) = 0.01 K.

Table VII.6. Values of surface tension (γ) and conductance (κ) data the break point with corresponding concentration of α and β-CD at 298.15 K[a]

CDP			
α-CD		**β-CD**	
Surface Tension Concentration /mM	γ[a] /mNm^{-1}	Surface Tension Concentration /mM	γ[a] /mNm^{-1}
5.08	70.34	5.07	70.75
Conductivity Concentration /mM	κ[a] /mSm^{-1}	Conductivity Concentration /mM	κ[a] /mSm^{-1}
5.01	0.512	5.0	0.490

[a] Standard uncertainties (u): temperature: u(T) = ±0.01 K, surface tension: $u(\gamma)$ = ±0.1 mNm ^{1}, conductivity: $u(\kappa)$ = ±0.001 mSm^{-1}.

Table VII.7. Data for the Job plot performed by UV-Vis spectroscopy for CDP-α-CD system at 298.15K[a]

CDP (mL)	α-CD (mL)	CDP (μM)	α-CD (μM)	[CDP]/([CDP]+[α-CD])	Absorbance (A)	ΔA	ΔA×[CDP]/([CDP]+[α-CD])
0.0	1.0	0	100	0.0	0.0000	0.2789	0
0.1	0.9	10	90	0.1	0.1841	0.0948	0.00948
0.2	0.8	20	80	0.2	0.1852	0.0937	0.01874
0.3	0.7	30	70	0.3	0.1899	0.0890	0.0267
0.4	0.6	40	60	0.4	0.2003	0.0786	0.03144
0.5	0.5	50	50	0.5	0.2111	0.0678	0.0339
0.6	0.4	60	40	0.6	0.2258	0.0531	0.03186
0.7	0.3	70	30	0.7	0.2379	0.0410	0.0287
0.8	0.2	80	20	0.8	0.2509	0.0280	0.0224
0.9	0.1	90	10	0.9	0.2638	0.0151	0.01359
1.0	0.0	100	0	1.0	0.2789	0.0000	0

[a] Standard uncertainties in temperature (T) = 0.01 K.

Table VII.8. Data for the Job plot performed by UV-Vis spectroscopy for CDP-*β*-CD system at 298.15K[a]

CDP(mL)	β-CD (mL)	CDP (μM)	β-CD (μM)	[CDP]/([CDP]+[β-CD])	Absorbance (A)	ΔA	ΔA×[CDP]/([CDP]+[β-CD])
0.0	1.0	0	100	0.0	0.0000	0.2767	0
0.1	0.9	10	90	0.1	0.1818	0.0949	0.00949
0.2	0.8	20	80	0.2	0.1829	0.0938	0.01876
0.3	0.7	30	70	0.3	0.1874	0.0893	0.02679
0.4	0.6	40	60	0.4	0.1947	0.0820	0.0328
0.5	0.5	50	50	0.5	0.2062	0.0705	0.03525
0.6	0.4	60	40	0.6	0.2221	0.0546	0.03276
0.7	0.3	70	30	0.7	0.2352	0.0415	0.02905
0.8	0.2	80	20	0.8	0.2496	0.0271	0.02168
0.9	0.1	90	10	0.9	0.2618	0.0149	0.01341
1.0	0.0	100	0	1.0	0.2767	0.0000	0

[a] Standard uncertainties in temperature (T) = 0.01 K.

Table VII.9. Data for the Benesi-Hildebrand double reciprocal plot performed by UV-Vis spectroscopy for CDP-*α*-CD system

Temp /K[a]	[D] /μM	[*α*-CD] /μM	Ao	A	ΔA	1/[*α*-CD] /M^{-1}	1/ΔA	Intercept	Slope	K_a /M^{-1}
288.15	50	30	0.2540	0.2661	0.0121	33333	82.6	4.6293803	0.0023255	1990.70
	50	40		0.2704	0.0164	25000	61.0			
	50	50		0.2730	0.0190	20000	52.6			
	50	60		0.2768	0.0228	16667	43.9			
	50	70		0.2809	0.0269	14286	37.2			
293.15	50	30	0.2540	0.2648	0.0108	33333	92.6	4.5265640	0.0026346	1718.12
	50	40		0.2685	0.0145	25000	69.0			
	50	50		0.2710	0.0170	20000	58.8			
	50	60		0.2744	0.0204	16667	49.0			
	50	70		0.2783	0.0243	14286	41.2			
298.15	50	30	0.2540	0.2636	0.0096	33333	104.2	4.4876567	0.0029782	1506.84
	50	40		0.2671	0.0131	25000	76.3			
	50	50		0.2689	0.0149	20000	67.1			
	50	60		0.2721	0.0181	16667	55.2			
	50	70		0.2762	0.0222	14286	45.0			

303.15	50	30	0.2540	0.2625	0.0085	33333	117.6	4.384832	0.00342	1281.40
	50	40		0.2652	0.0112	25000	89.3			
	50	50		0.2672	0.0132	20000	75.8			
	50	60		0.2697	0.0157	16667	63.7			
	50	70		0.2742	0.0202	14286	49.5			
308.15	50	30	0.2540	0.2614	0.0074	33333	135.1	4.301798	0.0039588	1086.64
	50	40		0.2637	0.0097	25000	103.1			
	50	50		0.2656	0.0116	20000	86.2			
	50	60		0.2676	0.0136	16667	73.5			
	50	70		0.2718	0.0178	14286	56.2			

[a] Standard uncertainties in temperature (T) = 0.01 K.

Table VII.10. Data for the Benesi-Hildebrand double reciprocal plot performed by UV-Vis spectroscopy for CDP-*β*-CD system

Temp /K[a]	[D] /μM	[β-CD] /μM	Ao	A	ΔA	1/[*β*-CD] /M^{-1}	1/ΔA	Intercept	Slope	K_a /M^{-1}
288.15	50	30	0.2540	0.2675	0.0135	33333	74.1	4.9652165	0.0020842	2382.31
	50	40		0.2711	0.0171	25000	58.5			
	50	50		0.2761	0.0221	20000	45.2			
	50	60		0.2792	0.0252	16667	39.7			
	50	70		0.2825	0.0285	14286	35.1			
293.15	50	30	0.2540	0.2661	0.0121	33333	82.6	4.9571858	0.0023206	2136.17
	50	40		0.2698	0.0158	25000	63.3			
	50	50		0.2741	0.0201	20000	49.8			
	50	60		0.2768	0.0228	16667	44.0			
	50	70		0.2798	0.0258	14286	38.8			
298.15	50	30	0.2540	0.2647	0.0107	33333	93.5	5.0125621	0.0026445	1895.47
	50	40		0.2680	0.0140	25000	71.4			
	50	50		0.2719	0.0179	20000	55.9			
	50	60		0.2739	0.0199	16667	50.3			
	50	70		0.2772	0.0232	14286	43.1			
303.15	50	30	0.2540	0.26330	0.0093	33333	107.5	5.0482032	0.0030714	1643.62
	50	40		0.26630	0.0123	25000	81.3			
	50	50		0.26900	0.0150	20000	66.7			
	50	60		0.27143	0.0174	16667	57.4			
	50	70		0.27482	0.0208	14286	48.0			
308.15	50	30	0.2540	0.26210	0.0081	33333	123.5	5.0434197	0.0035440	1423.09
	50	40		0.26480	0.0108	25000	92.6			
	50	50		0.26710	0.0131	20000	76.3			
	50	60		0.26924	0.0152	16667	65.6			
	50	70		0.27234	0.0183	14286	54.5			

[a] Standard uncertainties in temperature (T) = 0.01 K.

Table VII.11. Association constant (K_a) and thermodynamic parameters H^o, S^o and G^o of different CDP-cyclodextrin inclusion complexes

	CDP				
	Temp (K)[a]	$K_a \times 10^{-3}$ (M^{-1})[b]	H^o ($kjmol^{-1}$)[b]	S^o ($Jmol^{-1}K^{-1}$)[b]	G^o ($kJmol^{-1}$)[b] (298.15 K)
CDP+α-CD	288.15	1.99	-22.19	-13.78	-18.08
	293.15	1.71			
	298.15	1.50			
	303.15	1.28			
	308.15	1.08			
CDP+β-CD	288.15	2.38	-19.06	-1.35	-18.65
	293.15	2.13			
	298.15	1.89			
	303.15	1.64			
	308.15	1.42			

[a] Standard uncertainties in temperature *u* are: *u*(T) = ±0.01 K.

[b] Mean errors in K_a = ±0.02×10^{-3} M^{-1}; ΔH^o = ±0.01 kJ mol^{-1}; ΔS^o = ±0.01 J $mol^{-1}K^{-1}$, ΔG^o = ±0.01 kJ mol^{-1}.

Table VII.12. Frequencies at FTIR spectra of CDP, α-CD, β-CD and solid inclusion complexes

	Wave Number / cm^{-1}	**Group**
CDP	3433.7	Stretching of –N-H
	2950.73	Stretching of -C-H from – CH_3
	2798.02	Stretching of -C-H from – CH_2
	1615.58	Stretching of -C=N
	1459.23	Stretching of aromatic -C=C
	1213.34	Stretching of -C-N
	815.76	Stretching of –C-Cl
α-Cyclodextrin	3412.10	stretching of O-H
	2930.79	stretching of –C-H from – CH_2
	1406.76	bending of –C-H from – CH_2 and bending of O-H
	1154.39	bending of C-O-C
	1030.39	stretching of C-C-O
	952.36	skeletal vibration involving α-1,4linkage
β-Cyclodextrin	3349.84	stretching of O-H
	2921.52	stretching of –C-H from – CH_2
	1412.36	bending of –C-H from – CH_2 and bending of O-H
	1157.57	bending of C-O-C

	1033.51	stretching of C-C-O
	938.53	skeletal vibration involving α-1,4linkage
CDP+α-CD	3384.08	Stretching of –O-H of α-CD
	2924.03	Stretching of –C-H from –CH_2 of α-CD
	1633.96	Stretching of –C=N of CDP
	1439.46	Stretching of aromatic –C=C of CDP
	1223.51	Stretching of –C=N of CDP
	1144.53	Bending of –C-O-C of α-CD
	1021.64	Stretching of –C-C-O of α-CD
CDP+β-CD	3329.76	Stretching of –O-H of β-CD
	2911.86	Stretching of –C-H from –CH_2 of β-CD
	1623.96	Stretching of –C=N of CDP
	1449.73	Stretching of aromatic –C=C of CDP
	1211.87	Stretching of –C-N 0f CDP
	1025.34	stretching of C-C-O of β-CD
	945.32	

FIGURES

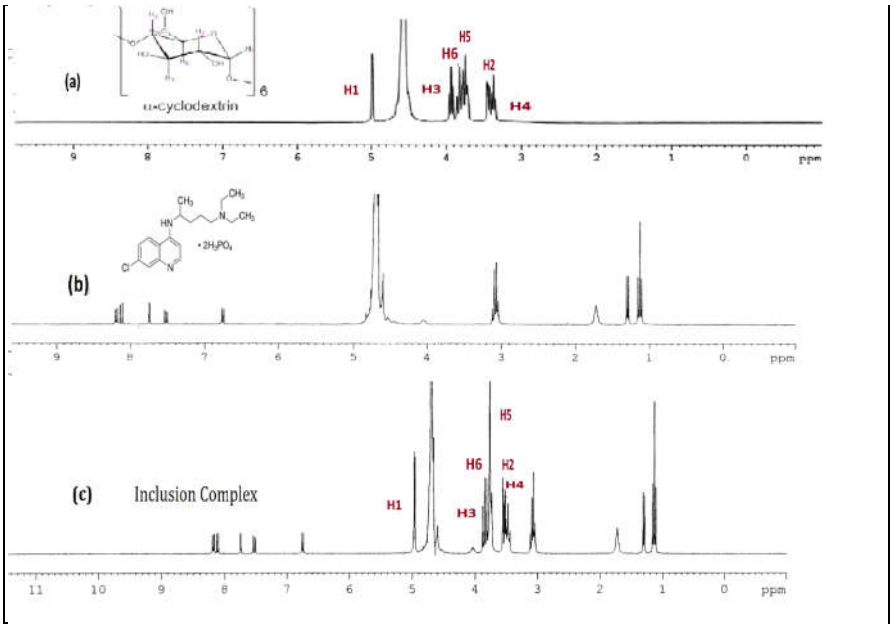


Figure VII.1. ^{1}H NMR Spectra of (a) α-CD (b) CDP and (c) 1:1 molar ratio of α-CD + CDP in D_2O in 298.15 K.

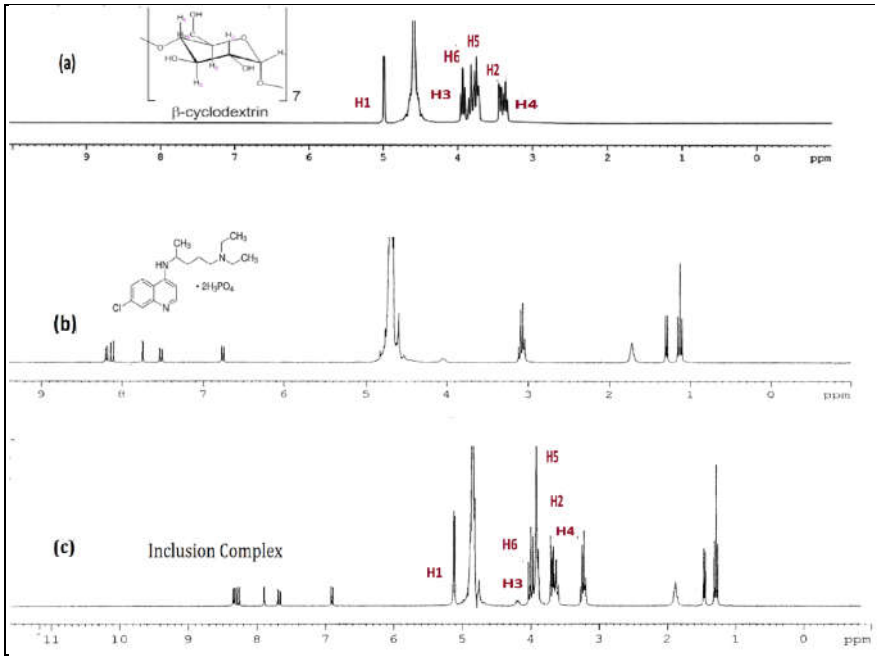


Figure VII.2. ^{1}H NMR Spectra of (a) β-CD (b) CDP and (c) 1:1 molar ratio of β-CD + CDP in D_2O in 298.15 K.

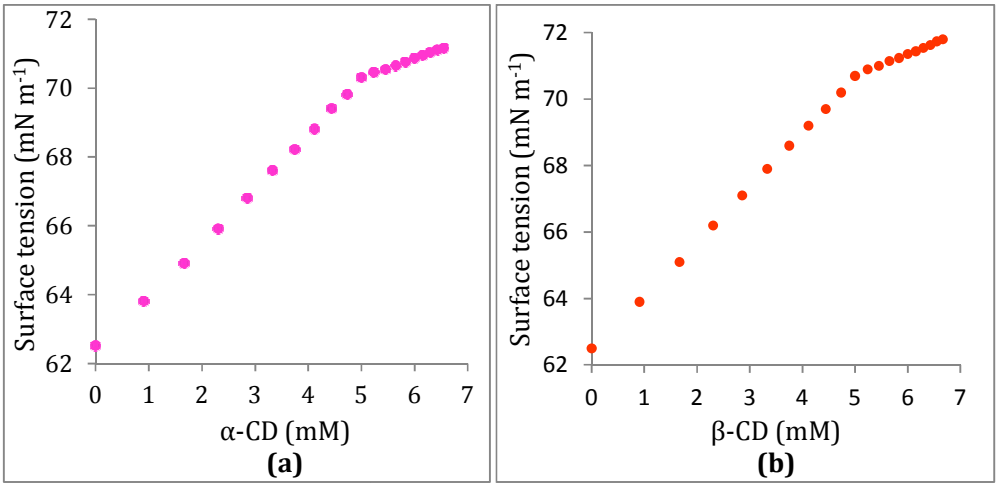


Figure VII.3. variation of surface tension of aqueous CDP solution with increasing concentration of (a) α-CD (b) β-CD respectively at 298.15K.

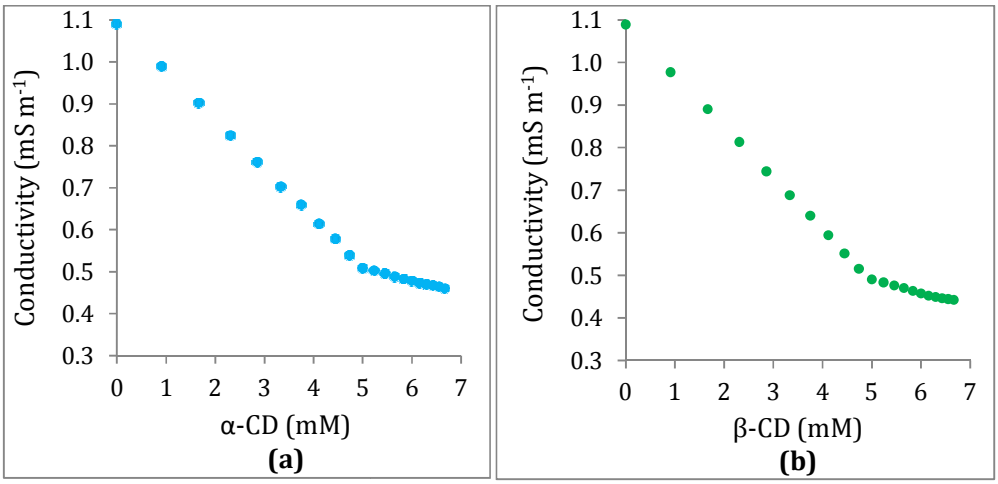


Figure VII.4. variation of conductivity of aqueous CDP solution with increasing concentration of (a) α-CD (b) β-CD respectively at 298.15K.

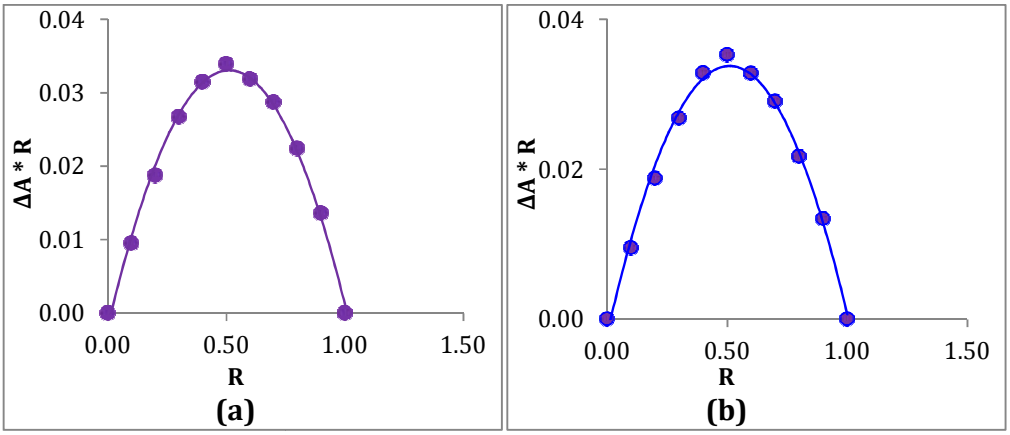
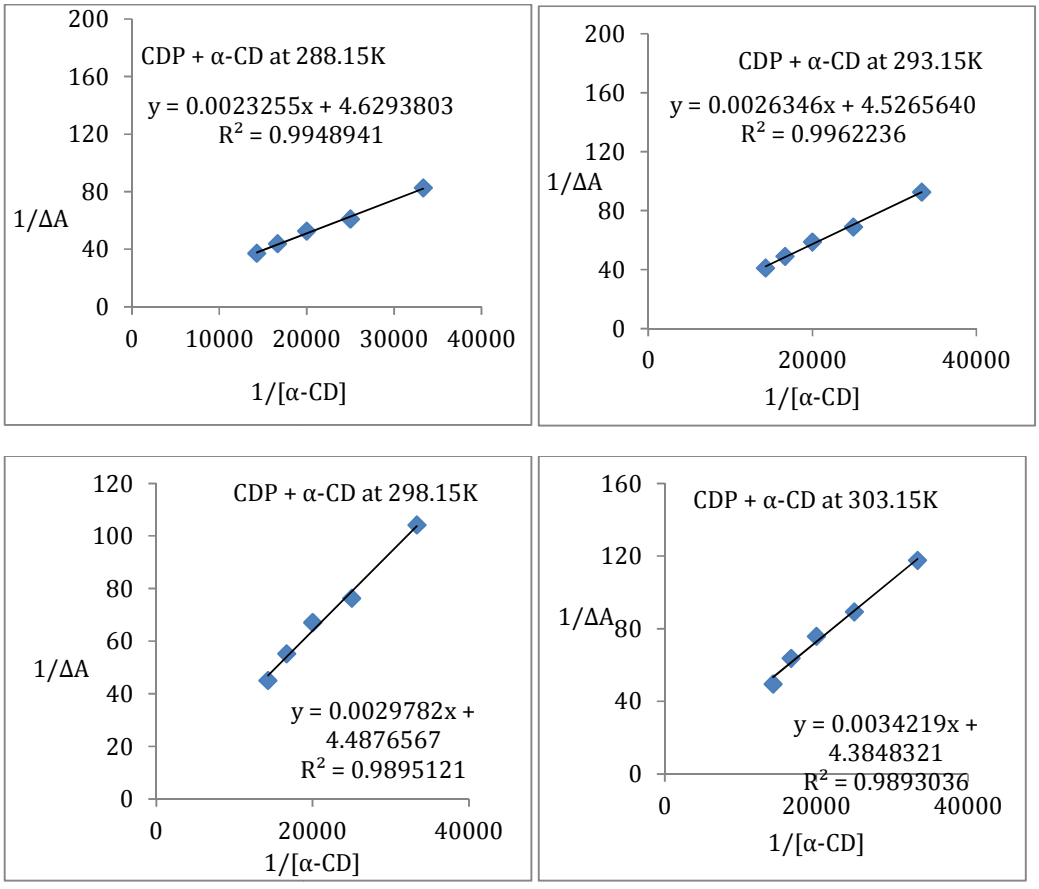


Figure VII.5. Job plot of (a) CDP-α-CD system at λ_{max}=254 nm (b) CDP-β-CD system at λ_{max}=254 nm at 298.15 K. R= [CDP]/ ([CDP] + [CD]), A=absorbance difference of CDP without and with CD.



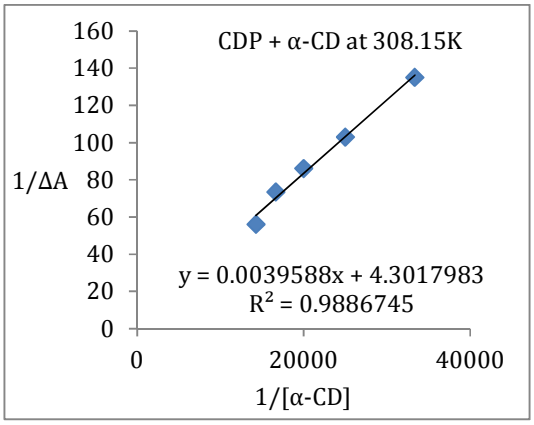
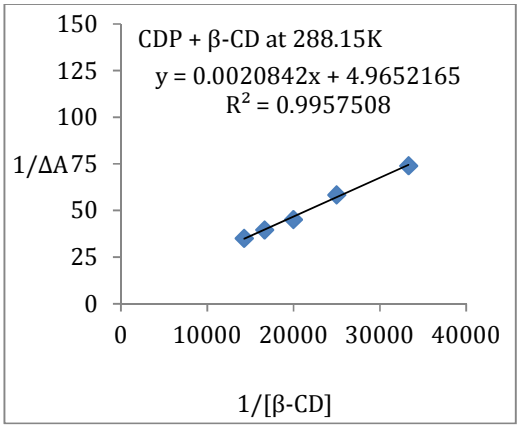
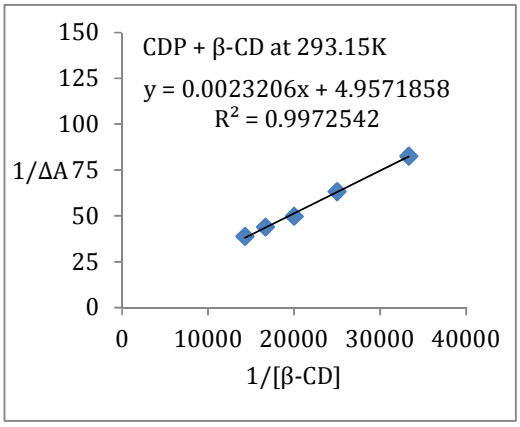
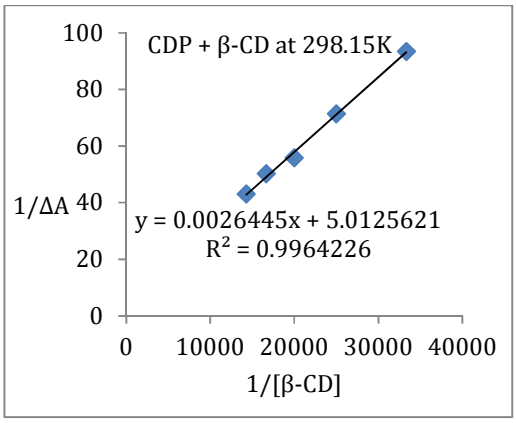
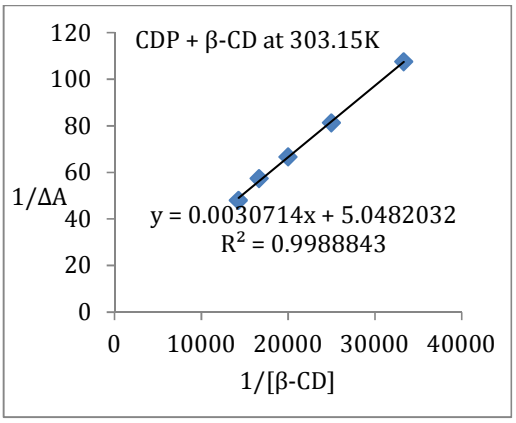


Figure VII.6. Benesi-Hildebrand double reciprocal plot for the effect of *α*-CD on the absorbance of CDP (254 nm) at different temperatures (X axis = 1/[*α*-CD] and Y axis = 1/ΔA).









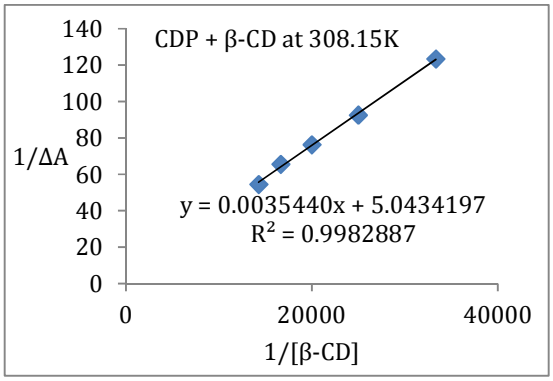


Figure VII.7. Benesi-Hildebrand double reciprocal plot for the effect of *β*-CD on the absorbance of CDP (254 nm) at different temperatures (X axis = 1/[*β*-CD] and Y axis = 1/ΔA).

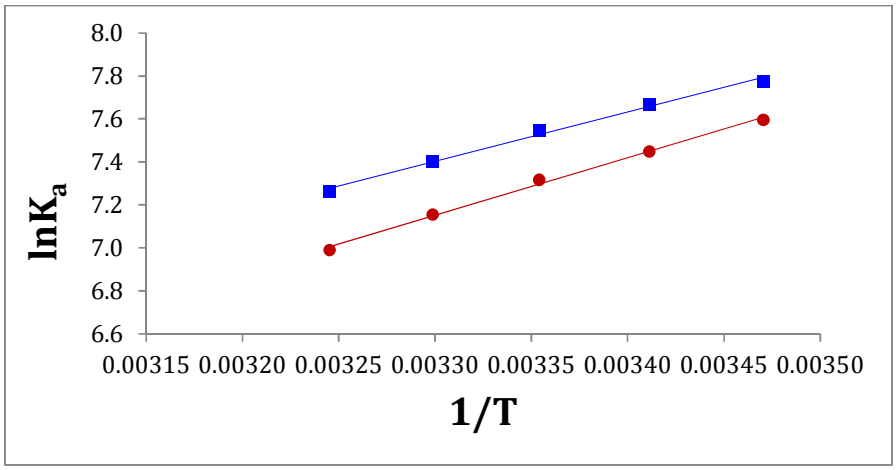


Figure VII.8. Plot of $\ln K_a$ vs 1/T for the interaction of CDP with *α*-CD (●) and *β*-CD ( ).

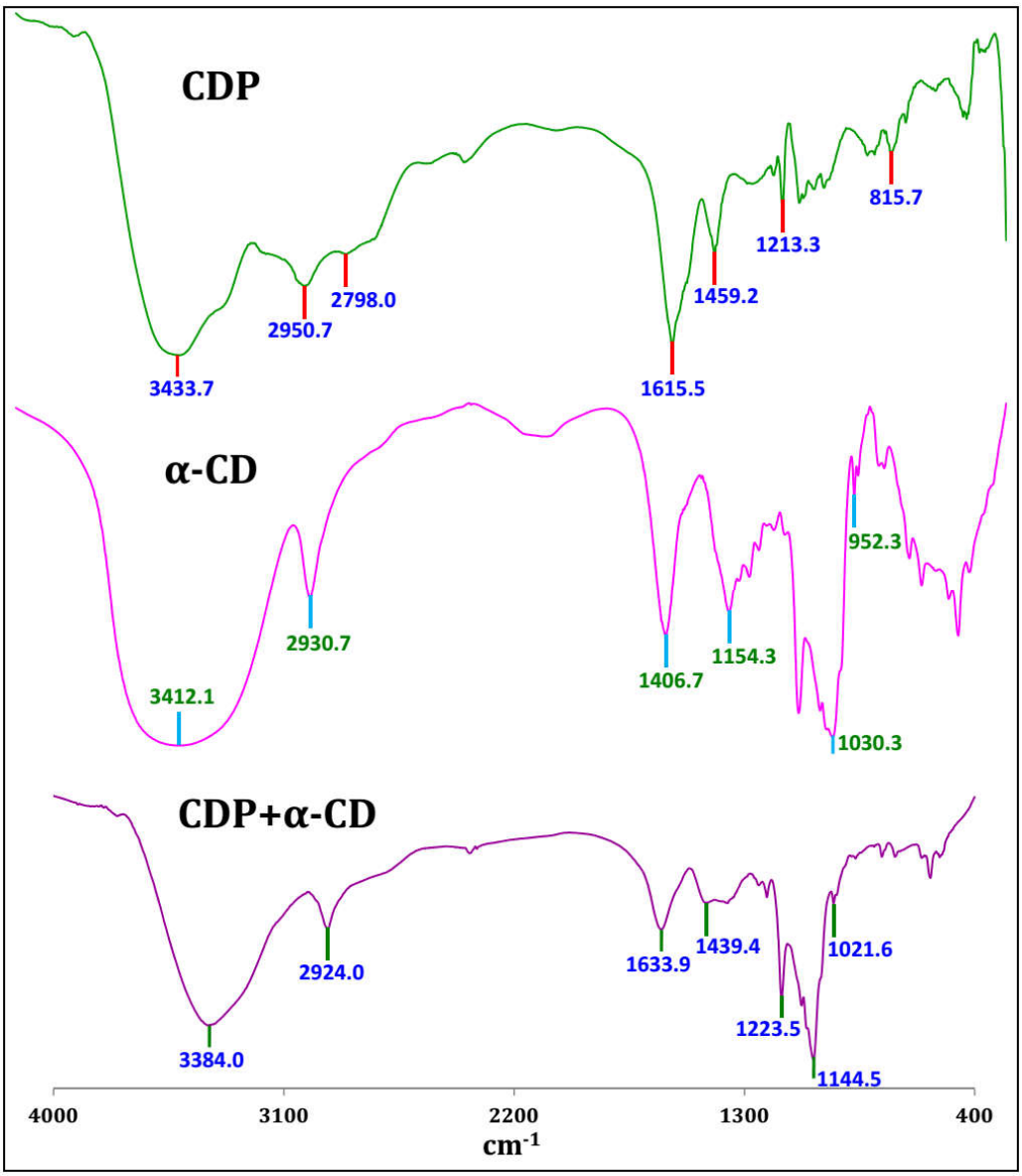


Figure VII.9. FTIR spectra of (a) CDP (b) α-CD and (c) CDP-α-CD inclusion complex.

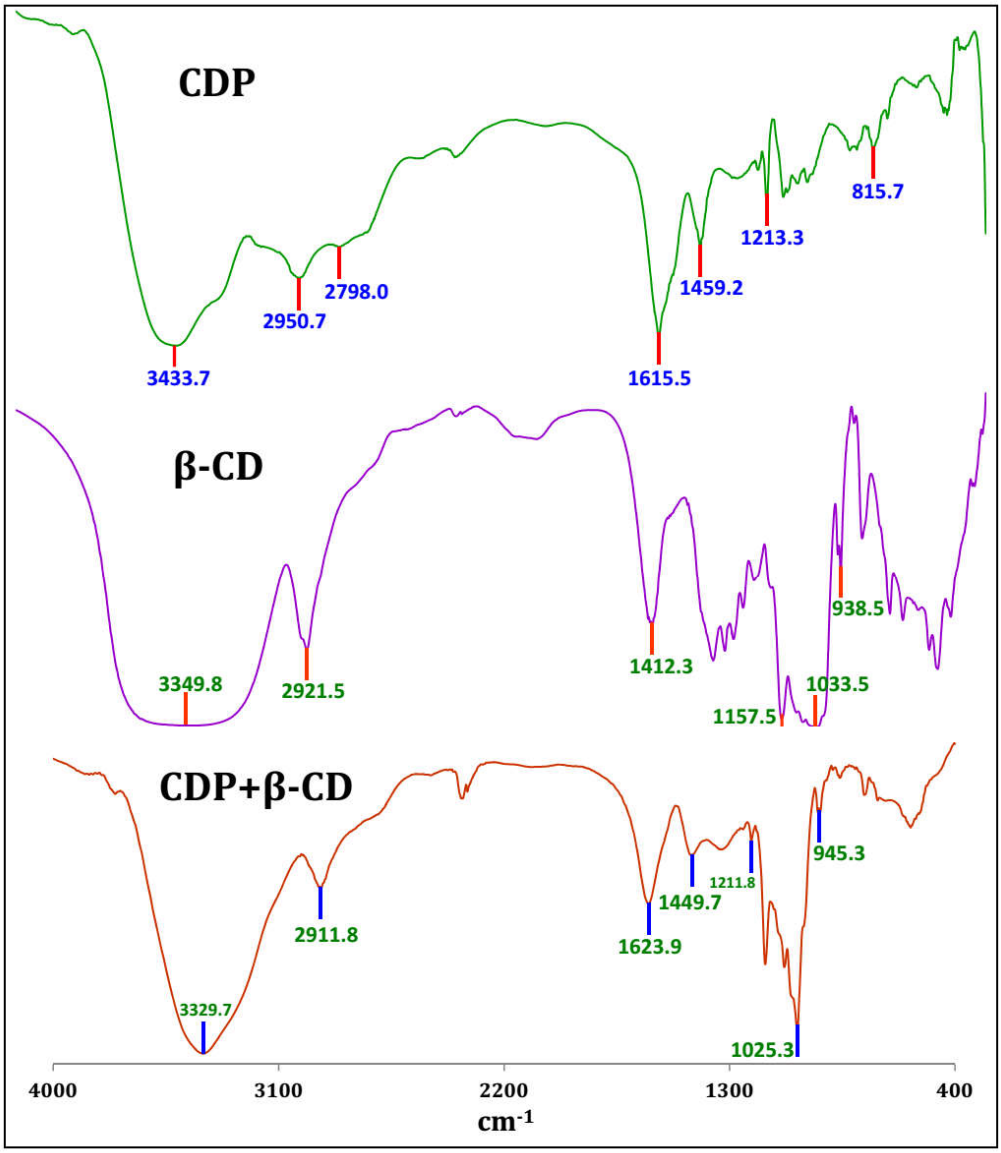


Figure VII.10. FTIR spectra of (a) CDP (b) β-CD and (c) CDP-β-CD inclusion complex.

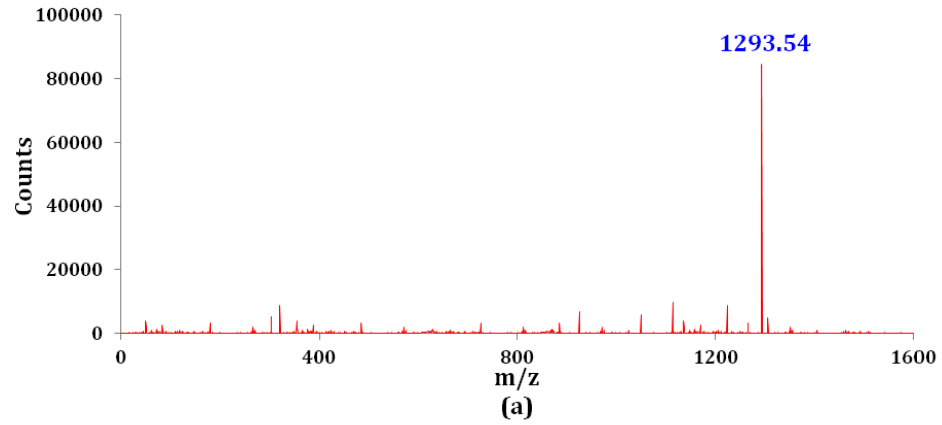


Figure VII.11. ESI mass spectra of CDP-α-CD inclusion complex.

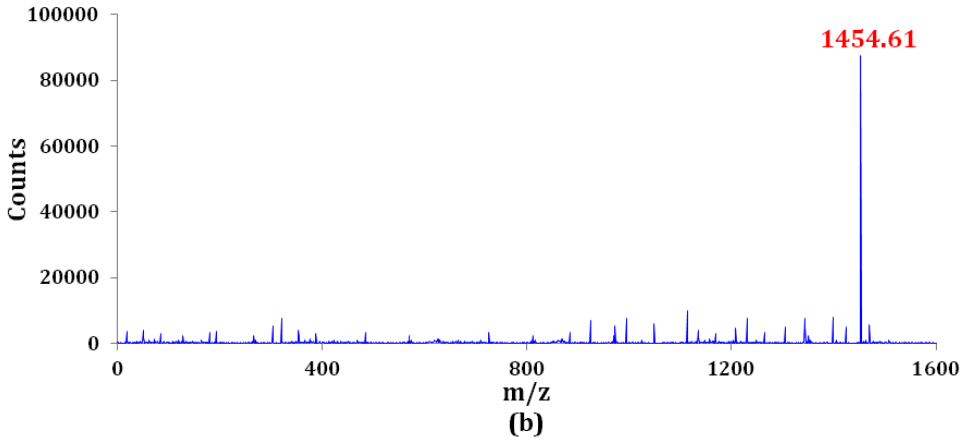
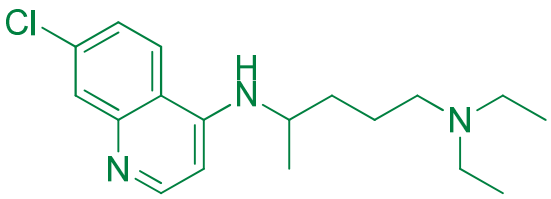
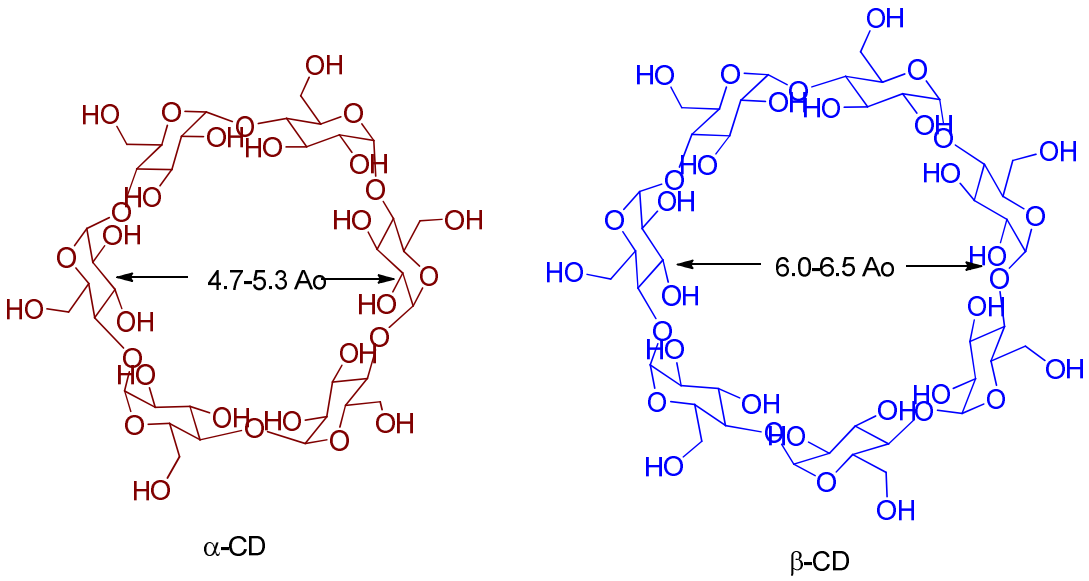


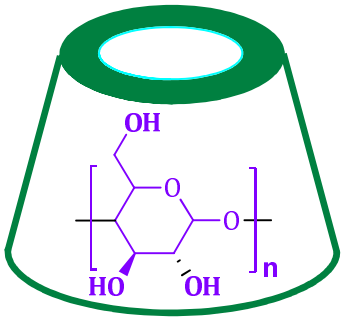
Figure VII.12. ESI mass spectra of CDP-β-CD inclusion complex.

SCHEMES



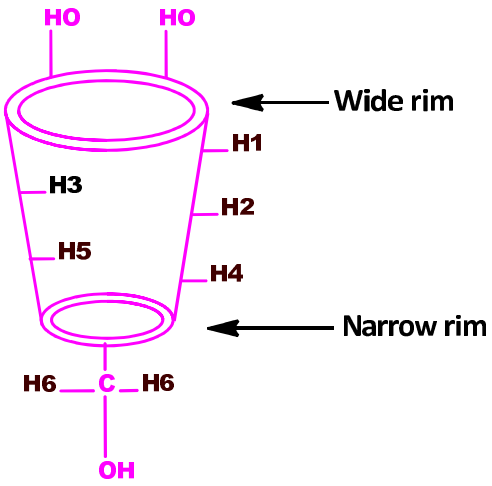
Scheme VII.1: Molecular structure of chloroquine.





Cyclodextrin
(n = 6, 7 for α-CD and β-CD respectively)

Scheme VII.2: Structure of cyclodextrin molecules.



Scheme VII.3. Truncated conical structure of cyclodextrin.