Carbohydrate Polymers 95 (2013) 366–370

Contents lists available at SciVerse ScienceDirect

Carbohydrate Polymers

journal homepage: www.elsevier.com/locate/carbpol

Investigation of water-soluble inclusion complex of hypericin with β-cyclodextrin polymer

Wang Zhang[a], Xiangdong Gong[a], Yin Cai[a], Chuanli Zhang[b], Xing Yu[b], Jian Fan[b], Guowang Diao[a,*]

[a] College of Chemistry & Chemical Engineering, Yangzhou University, Yangzhou 225002, Jiangsu, PR China
[b] College of Medicine, Yangzhou University, Yangzhou 225002, Jiangsu, PR China

ARTICLE INFO

Article history:
Received 13 February 2013
Accepted 3 March 2013
Available online 13 March 2013

Keywords:
Hypericin
β-Cyclodextrin polymer
Inclusion complex
Characterization

ABSTRACT

A water-soluble inclusion complex of hypericin (HY) with β-cyclodextrin polymer (CDP) was achieved by supramolecular interactions between HY and CDP. The inclusion complex (HY-CDP) was characterized by ^{1}H NMR, FTIR, and UV–vis spectroscopies. Compared with HY, the water-solubility of HY-CDP was greatly enhanced because of the water-soluble CDP host. The mole ratio of β-cyclodextrin (β-CD) unit in CDP to HY was determined as 2:1. At 25 °C, the dissociated constant of HY-CDP was measured as 1.47×10^{-7} mol L^{-1} by UV–vis spectroscopy. In the formation of inclusion complexes, CDP could overcome the β-CD drawbacks – such as the poor water-solubility and the restriction of single cavity size, indicating it was able to use as a universal solubilizer for pharmaceutical application.

1. Introduction

β-Cyclodextrin (β-CD) is cyclic oligosaccharides including glucose unites linked by α-1,4-glucosidic bonds. Given the hydrophobic internal cavity and hydrophilic external surface, it can form supramolecular host–guest complexes with a variety of hydrophobic molecules (Szejtli, 1998). Through the distinct physical and chemical properties, β-CD is used in many fields, such as electroanalysis (Chen, Zhang, Jiang, & Diao, 2011), biotechnology (Yang, Lin, Chen, & Liu, 2009), environmental protection (Zhang, Chen, Zha, & Diao, 2012), and foodstuff (Hu, Wei, et al., 2012; Hu, Zhang, Song, Gu, & Hu, 2012). β-CD appears to be the best natural cyclodextrin for pharmaceutical applications because of its efficient drug complexation (Hu, Wei, et al., 2012; Hu, Zhang, et al., 2012), however, some limitations exist for the applications of β-CD: (1) the aqueous solubility of β-CD is low. (2) Although some chemical modifications have been prepared to improve the aqueous solubility of β-CD, adopted chemical routes usually use the toxic organic reagent. (3) Only several organic molecules having specific sizes can form inclusion complexes with CDs and the single and small size of CD cavities limit the CD use in pharmaceutical applications. β-Cyclodextrin polymer (CDP) has been obtained by reaction of the parent β-CD with a cross-linking agent, epichlorohydrin (EP) (Renard, Deratani, Volet, & Sebille, 1997). Especially, CDP as a highly water-soluble polymer is well known to selectively form inclusion complexes. Moreover, the features of CDP have been widely exploited, e.g., for solubilization and pharmaceutics (Li, Xiao, & Zhong, 2004; Zhang, Chen, & Diao, 2011; Zhao, Zhao, Zhu, Tian, & Shen, 2009).

Hypericin (HY) is a natural polycyclic quinone from *Hypericum perforatum*, commonly known as St John's wort (Agostinis, Vantieghem, Merlevede, & de Witte, 2002). The chemical structure of HY is shown in Fig. 1(a). This natural product is an effective antidepressant and anxiolytic (Lavie, Mazur, Lavie, & Meruelo, 1995; Vandenbogaerde et al., 2000), an antiretroviral agent (Lavie et al., 1995), and a potent photosensitizer suggested for use in photodynamic therapy of cancer (Saw, Olivo, Soo, & Heng, 2006). However, HY is very lipophilic and water-insoluble, which makes intravenous injection problematic and restrains its medical applications.

Up to date, only a few studies focus on cyclodextrins and HY (de los Reyes & Koda, 2001; Falk, Sarhan, Tran, & Altmann, 1998; Sattler, Schaefer, Schneider, Hoelzl, & Lehr, 1997; Vandenbogaerde et al., 2000), but the water-solubility of HY is not remarkably improved with CDs and the host–guest interaction of HY with CDs is not explored carefully. In this paper, we used CDP as a solubilizing agent for HY, and studied the formation of a water-soluble inclusion complex between HY and CDP. In this preparation of HY-CDP, there was not any chemical synthesis in order to preserve the original structure of HY. The method would provide a convenient and efficient approach for obtaining HY with high water solubility, high bioavailability and low toxicity.

---

* Corresponding author. Tel.: +86 514 87975436; fax: +86 514 87975244.
E-mail address: gwdiao@yzu.edu.cn (G. Diao).

0144-8617/$ – see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.carbpol.2013.03.020

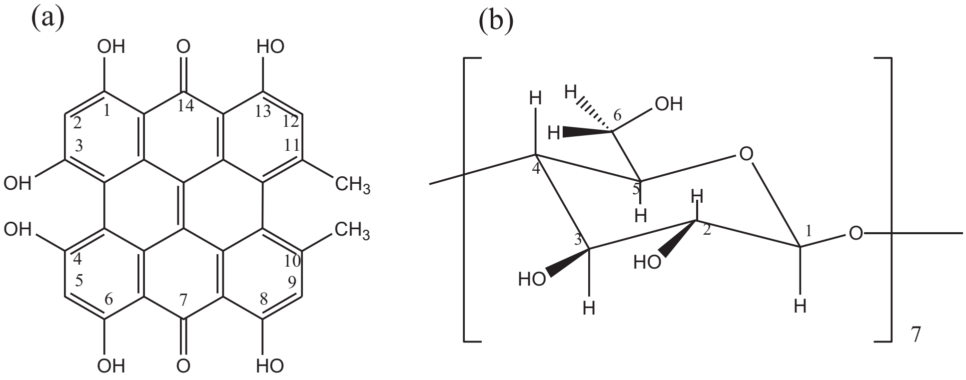

Fig. 1. Chemical structure of (a) HY and (b) CDP.

2. Experimental

HY was purchased from Aldrich. β-CD, epichlorohydrin (EP), ethylene glycol, and other reagents were all analytical purity and purchased from Sinopharm Chemical Reagents Company. Double distilled and sterilized water was used to prepare all solution.

2.1. Apparatus

The FTIR spectrum was used by the Tensor 27spectrophotometer (Bruker, Germany). The ^{1}H NMR spectra were conducted on a 600 MHz Bruker spectrometer (Bruker, Germany) at 303.1 K in deuterium oxide. The UV–vis spectrum was recorded on a UV-2550 double-beam spectra photometer (Shimazu, Japan) equipped with a stoppered quartz cell with 1.0 cm optical path length.

2.2. Preparation of CDP and HY-CDP inclusion complex

CDP was synthesized by cross-linking β-CD with EP under a strongly alkaline condition (33 wt% NaOH). The molar ratio of β-CD/EP was 1:7. The details of the synthesis and purification were shown as in the reference (Zhang et al., 2011).

The preparation of the inclusion complex of HY with CDP was shown as follow: it was prepared by dissolving 4 g CDP and 0.5 g HY in 100 ml water with sufficiently stirring for at least 24 h at room temperature. At the end of the reaction, a deep purple solution contained the inclusion complex was obtained after removing the residual HY. Then the deep purple product was obtained by pressure distillation, and it was dried in a vacuum oven at 65 °C for 48 h.

2.3. Aqueous solubility of HY-CDP

The saturated solubility of HY-CDP in water was measured at 25 °C according to the reference (Zhang et al., 2011). Firstly, an adequate amount of HY-CDP was added to 5 ml water to ensure the solution reaching saturation. The solution was mechanically shaken for 2 h at 25 °C. Secondly, the remaining solid in the solution was filtrated off using a 0.45 μm Nylon Cameo filter-syringe. Thirdly, the filtrated HY-CDP was dried in a vacuum oven for 24 h until a constant weight being reached. The solubility of HY-CDP in water could be calculated from the difference between the initial amount and the residual amount of HY-CDP. The measurement repeated three times. The saturated concentration of HY-CDP in water was also measured by UV–vis spectrophotometry.

3. Results and discussion

3.1. Characterization of HY-CDP

Fig. 2 shows the FTIR spectra of (a) HY, (b) CDP, and (c) HY-CDP. The typical IR spectrum of HY was presented in Fig. 2(a), which was in good agreement with literature (Kapinus, Falk, & Tran, 1999). The carbonyl and aromatic stretching vibrations at 1595 cm^{-1} was exhibited in Fig. 2(a). Fig. 2(b) shows the typical CDP absorptions, such as the coupled C—O—C stretching vibrations at ~1160 cm^{-1}, the coupled C—O/C—C stretching vibrations at ~1030 cm^{-1}, CH$_2$ stretching vibrations at ~2920 cm^{-1}, CH bending vibrations at ~1456 cm^{-1}, and OH stretching vibrations at ~3330 cm^{-1}. As shown in Fig. 2(c), we found that the FTIR spectrum of HY-CDP exhibited the typical absorption features of CDP and HY. Compared with HY, the peak of the carbonyl and aromatic stretching vibrations shifted to 1554 cm^{-1} and the peak strength became much lower, indicating the formation of inclusion complex of HY with CDP.

The direct evidence for the formation of inclusion complex could be found from the ^{1}H NMR spectrum (Schneider, Hacket, Rüdiger, & Ikeda, 1998). In order to study the possible inclusion mode of HY-CDP, we compared the ^{1}H NMR spectra of HY in the absence and presence of CDP in Fig. 3. The chemical structure of β-CD unit in CDP was shown in Fig. 1(b). The ^{1}H resonances of CDP were assigned according to the reported method (Renard et al., 1997; Zhang et al., 2011). The typical ^{1}H NMR spectrum of HY was

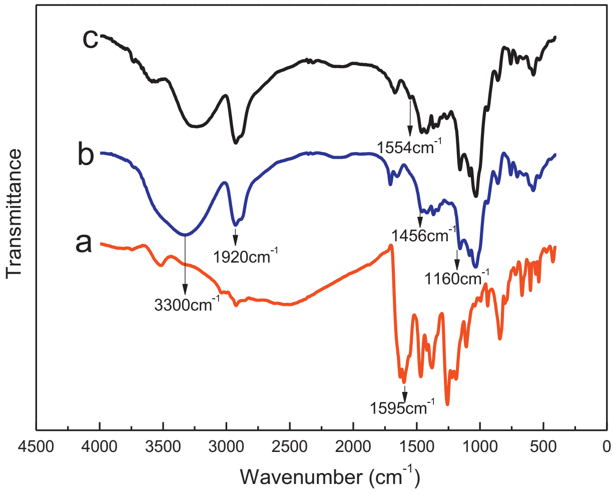

Fig. 2. FTIR spectra of (a) HY, (b) CDP, and (c) HY-CDP.

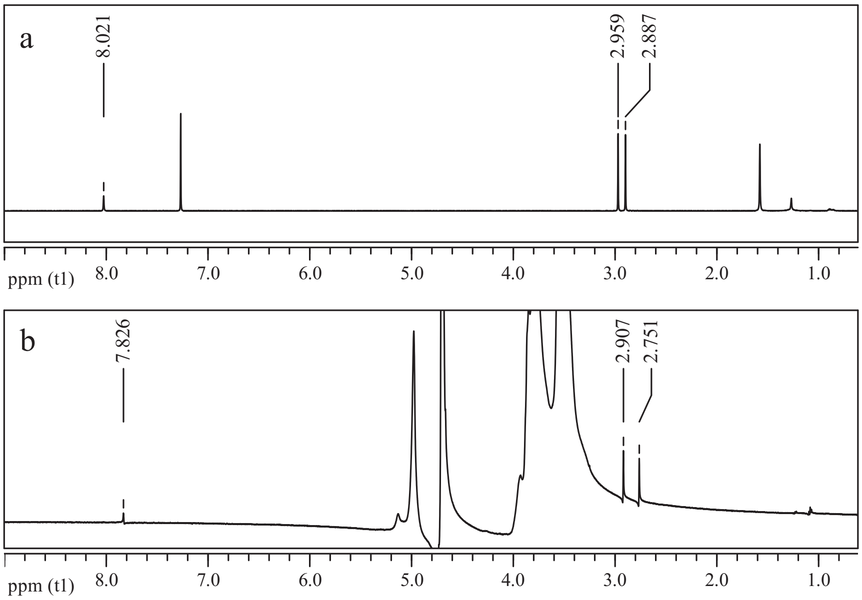

Fig. 3. ^1H NMR spectra of (a) HY in $CDCl_3$ and (b) HY-CDP in D_2O.

showed in Fig. 3(a), which was consistent with the previous report (Tatsis, Exarchou, Troganis, & Gerothanassis, 2008). As illustrated in Fig. 3(b), it confirmed the existence of HY in the inclusion complex. Compared with the ^1H NMR spectrum of HY, the signal of protons of C10, C11–CH3 shifted from 2.958 and 2.887 ppm to 2.907 and 2.751 ppm, and the signal of protons of C9, C12-H shifted from 8.021 to 7.826 ppm. These great upfield shifts, which was similar to our previous studies in cyclodextrin inclusion complexes (Chen, Diao, & Zhang, 2006; Zhang et al., 2011), suggested HY monomer entered into the hydrophobic cavities of CDP. The results indicated that HY and CDP could be formed inclusion complex by supramolecular interactions. The ratio of HY to β-CD unit in the polymer inclusion complex was determined by the peak area of protons of HY and β-CD unit in inclusion complex. The number of the protons located at different positions was measured and listed in Table S1 (in the supporting information). The stoichiometry (1:2 molar ratio) of both HY and β-CD unit in CDP were confirmed by ^1H NMR spectroscopy.

3.2. UV–vis spectra and aqueous solubility of HY-CDP

Fig. S1 in the supporting information showed the UV–vis absorption spectrum of HY in ethanol. The polycyclic aromatic structure of HY resulted in a strong absorption throughout the UV spectrum with peaks at 546 and 595 nm, which was similar to the references (Chen et al., 2001; Huntosova et al., 2012; Smirnov et al., 1999). Fig. 4 showed the UV–vis absorption spectra of HY and HY-CDP saturated aqueous solutions. However, no absorption in water was observed for pure HY in the range of 400–700 nm as shown in Fig. 4(a), because of the poor water-solubility of HY. In Fig. 4(b), the two typical absorption peaks of HY in the inclusion complex were observed at 558 and 597 nm, which was very similar to that of HY in ethanol. However, the spectral profile of HY was markedly affected by formation of inclusion complex with HY. The main changes that we could observe in the complex are remarkable descends of the two peaks. Because CDP solution had no absorption in UV–vis spectrum, Fig. 4(b) could be only found the peaks of HY in the inclusion complex. These results indicated that CDP as a solubilizer improved the aqueous solubility of HY, and formed the inclusion complex of HY which might penetrate into β-CD cavities in CDP by hydrophobic interactions.

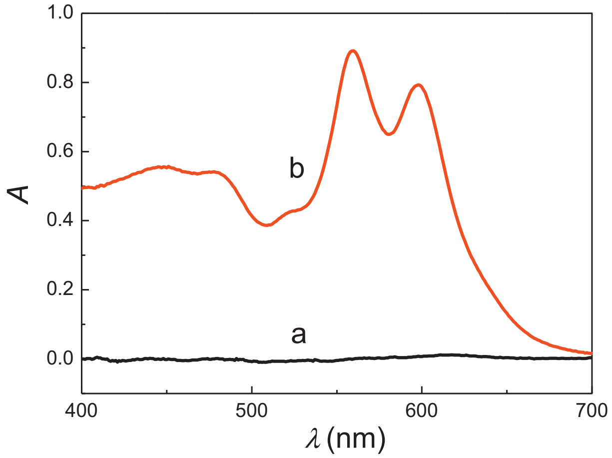

Fig. 4. UV–vis spectra of (a) HY and (b) HY-CDP saturated aqueous solutions.

Fig. 5(A) shows UV–vis spectra of HY-CDP with different concentrations in aqueous solution (pH = 7.0, 25 °C). It was found that the peak positions at 558 and 597 nm were independent of the concentrations of HY-CDP. However, the intensity of the peaks increased with the concentration of HY-CDP. Plotting the absorbance of HY-CDP at 558 nm vs. the concentration of HY-CDP, a straight line was obtained as shown in Fig. 5(B). According to the Lambert-Beer law, the absorption coefficient (ε) of HY-CDP in the aqueous solution (pH = 7.0, 25 °C) was evaluated as 0.478 L g^{-1} cm^{-1}. The solubilities of HY, CDP, and HY-CDP at 25 °C in water were listed in Table 1. Compared with HY, it was clear that the solubility of HY-CDP was

Table 1
Aqueous solubility of HY, CDP, and HY-CDP at 25 °C.

Sample	Aqueous solubility (g L^{-1})
HY	Insolubility
CDP	>100[a]
HY-CDP	1.864[b]
HY-CDP	1.794[a]

[a] The method by Section 2.3.
[b] Data taken from UV–vis spectrum.

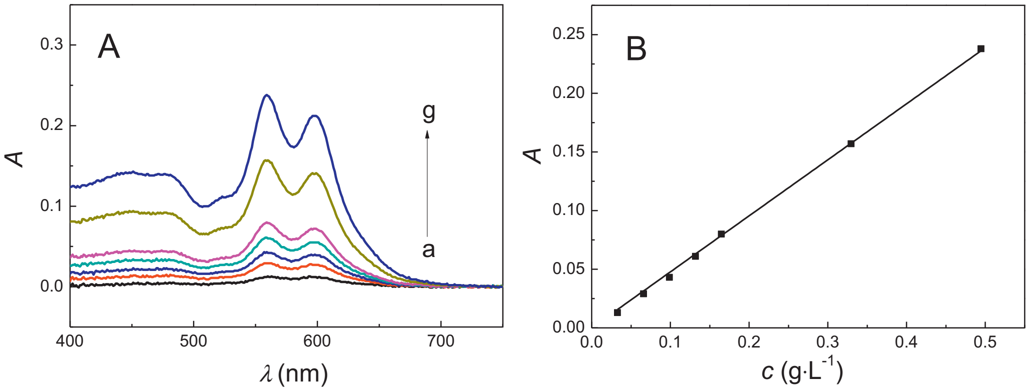

Fig. 5. (A) UV–vis spectra of HY-CDP with different concentrations (g L^{-1}) in aqueous solution (pH = 7.0, 25 °C): (a) 0.033; (b) 0.066; (c) 0.099; (d) 0.132; (e) 0.165, (f) 0.330, (g) 0.495. (B) A plot of absorbance ratio of HY-CDP at 558 nm vs. the concentration of HY-CDP. Date taken from (A).

much higher than that of HY. These results revealed that the water-soluble host CDP improved the aqueous solubility of HY remarkably by the formation of HY-CDP inclusion complex.

3.3. Dissociated constant of inclusion complex

Because of the insolubility of HY in water, ethylene glycol, a suitable solvent for HY and CDP, was chosen to study the dissociated constant of the inclusion complex. The formation of HY-CDP in ethylene glycol could be confirmed by the UV–vis spectrum as shown in Fig. 6(A). The peak position was independent of the addition of CDP. However, the peak intensity decreased with CDP. Assuming the ratio of HY to β-CD unit in CDP was 1:2, the formation of the inclusion complex could be described as follows:

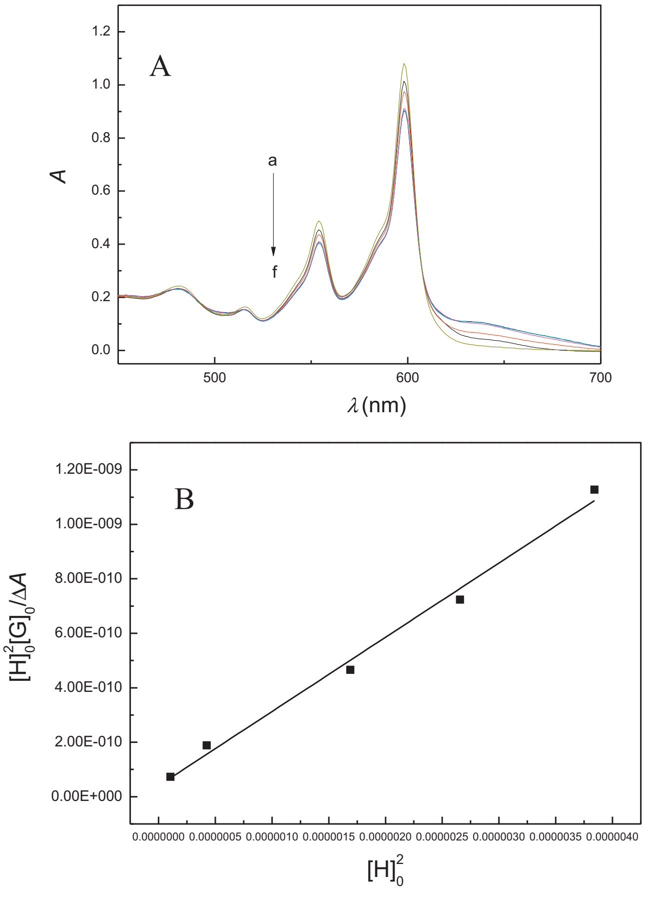

Fig. 6. (A) At 25 °C, UV–vis spectra of 2.26 × 10^{-4} M HY in ethylene glycol with different concentrations of β-CD unit in CDP (mM): (a) 0, (b) 0.326, (c) 0.652, (d) 0.978, (e) 1.63, (f) 1.96. (B) The plot of $[H]_0^2[G]_0/\Delta A$ vs. $[H]_0^2$. Data taken from (A).

$$2\mathrm{H} + \mathrm{G} = \mathrm{H_2G} \tag{1}$$

where H represented the host, β-CD unit in the polymer, G was the guest, HY, and H_2G was the inclusion complex. The stability of HY-CDP was described in terms of its formation (K_F) or dissociation ($K_D = 1/K_F$) constants. If the initial concentrations of H and G were $[H]_0$ and $[G]_0$ respectively, and $[H]_0 \gg [G]_0$, the concentration of H_2G at equilibrium was x, then K_D was calculated according to the Benesi–Hildebrand method (Tamaki, 1984):

$$K_D = \frac{([G]_0 - x)([H]_0 - 2x)^2}{x} \tag{2}$$

Taking account of the optical path length, $l = 1.0$ cm, the UV–vis absorbance at a given wavelength could be described as follows:

$$A = \varepsilon_H([H]_0 - 2x) + \varepsilon_G([G]_0 - x) + \varepsilon_{H_2G}x \tag{3}$$

where ε_H, ε_G, and ε_{H2G} were molar absorption coefficients of H, G, and H_2G, respectively.

Because $[H]_0 \gg [G]_0$, therefore, $[H]_0 \gg 2x$. Combining Eq. (2) with Eq. (3), we had

$$\frac{A - \varepsilon_H[H]_0 - \varepsilon_G[G]_0}{\varepsilon_{H_2G} - \varepsilon_G - \varepsilon_H} = \frac{[H]_0^2[G]_0}{K_D + [H]_0^2} \tag{4}$$

There was almost no absorption when wavelength ranges from 400 to 750 nm for CDP, ε_H could be negligible comparing to ε_G, and ε_{H2G}. Let $\Delta A = A - \varepsilon_H[H]_0 - \varepsilon_G[G]_0$, $\Delta\varepsilon = \varepsilon_{H2G} - \varepsilon_G$, Eq. (4) could be rearranged as:

$$\frac{[H]_0^2[G]_0}{\Delta A} = \frac{[H]_0^2}{\Delta\varepsilon} + \frac{K_D}{\Delta\varepsilon} \tag{5}$$

Plotting $[H]_0^2[G]_0/\Delta A$ vs. $[H]_0^2$, a straight line was obtained. The experimental result was shown in Fig. 6(B). The linear relationship between $[H]_0^2[G]_0/\Delta A$ and $[H]_0^2$ proved that the assumption of 1:2 ratio of HY to β-CD unit in CDP was correct. According to the slope and the intercept of the line, K_D of the inclusion complex was evaluated as 1.47×10^{-7} mol L^{-1}.

The stoichiometry of the inclusion complex was also confirmed by the continuous variation Job's method (Ge, Huang, Tian, Huang, & Zeng, 2012; Job, 1928; Liu, Yang, Chen, & Ding, 2005; Negi & Singh, 2013). The sum of the concentrations of both components was kept constant ([HY] + [β-CD unit] = 4 × 10^{-4} M). The Job's plot of the complex between HY and β-CD unit in CDP was shown in Fig. 7, where the molar fraction of HY, R (R = [HY]/([HY] + [β-CD unit]) varied from 0.0 to 1.0 and plotted against absorbance difference ($\Delta A \times$ mole fraction). As shown in Job's plot the maximum

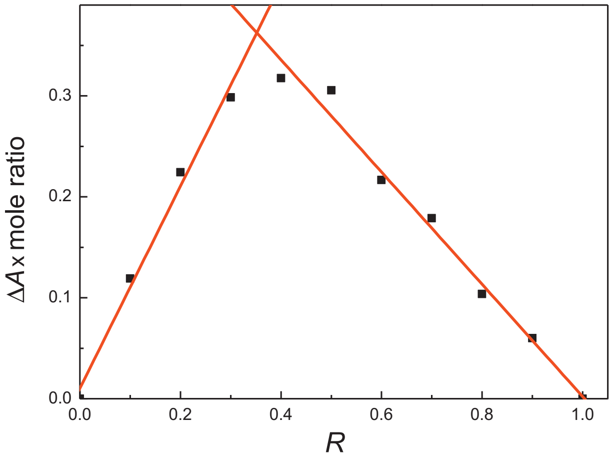

Fig. 7. Job's plot for different molar ratio of HY and β-CD unit in CDP.

peak was obtained at $R = 0.35$, which indicated the formation of 1:2 inclusion complex between HY and β-CD unit in CDP.

4. Conclusion

In the present work, the water-soluble host, CDP improved the physical and chemical properties of the HY guest by forming HY-CDP inclusion complex. This work involved the characterization of HY-CDP, aiming to gain high solubility, and stable electro-activity in water. The inclusion complex between HY and β-CD unit in CDP was formed with a stoichiometry of 1:2 and the dissociated constant K_D was obtained by UV–vis spectroscopy. All the results suggested that the CDP complexation technique was a promising strategy to improve the water-solubility of HY.

Acknowledgments

The authors acknowledged the financial support from the National Natural Science Foundation of China (Grant Nos. 21273195, 20973151, 20901065), the Project Funded by the Priority Academic Program Development of Jiangsu Higher Education Institutions, the Foundation of the Educational Committee of Jiangsu Provincial General Universities Graduate Student Scientific Research Invention Plan, and the Foundation of Jiangsu Provincial Key Program of Physical Chemistry in Yangzhou University.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.carbpol.2013.03.020.

References

Agostinis, P., Vantieghem, A., Merlevede, W., & de Witte, P. A. M. (2002). Hypericin in cancer treatment: More light on the way. *The International Journal of Biochemistry & Cell Biology*, *34*(3), 221–241.

Chen, B., Xu, Y., Roskams, T., Delaey, E., Agostinis, P., Vandenheede, J. R., et al. (2001). Efficacy of antitumoral photodynamic therapy with hypericin: Relationship between biodistribution and photodynamic effects in the RIF-1 mouse tumor model. *International Journal of Cancer*, *93*(2), 275–282.

Chen, M., Diao, G., & Zhang, E. (2006). Study of inclusion complex of beta-cyclodextrin and nitrobenzene. *Chemosphere*, *63*(3), 522–529.

Chen, M., Zhang, W., Jiang, R., & Diao, G. (2011). Development of glucose amperometric biosensor based on a novel attractive enzyme immobilization matrix: Amino derivative of thiacalixarene. *Analytica Chimica Acta*, *687*(2), 177–183.

de los Reyes, G. C., & Koda, R. T. (2001). Development of a simple, rapid and reproducible HPLC assay for the simultaneous determination of hypericins and stabilized hyperforin in commercial St. John's Wort preparations. *Journal of Pharmaceutical and Biomedical Analysis*, *26*(5–6), 959–965.

Falk, H., Sarhan, A.-E.-W. A. O., Tran, H. T. N., & Altmann, R. (1998). Synthesis and properties of hypericins substituted with acidic and basic residues: Hypericin tetrasulfonic acid – A water soluble hypericin derivative. *Monatshefte für Chemie/Chemical Monthly*, *129*(3), 309–318.

Ge, X., Huang, Z., Tian, S., Huang, Y., & Zeng, C. (2012). Complexation of carbendazim with hydroxypropyl-β-cyclodextrin to improve solubility and fungicidal activity. *Carbohydrate Polymers*, *89*(1), 208–212.

Hu, L., Zhang, H., Song, W., Gu, D., & Hu, Q. (2012). Investigation of inclusion complex of cilnidipine with hydroxypropyl-beta-cyclodextrin. *Carbohydrate Polymers*, *90*(4), 1719–1724.

Hu, X., Wei, B., Li, H., Wu, C., Bai, Y., Xu, X., et al. (2012). Preparation of the beta-cyclodextrin-vitamin C (beta-CD-Vc) inclusion complex under high hydrostatic pressure (HHP). *Carbohydrate Polymers*, *90*(2), 1193–1196.

Huntosova, V., Buzova, D., Petrovajova, D., Kasak, P., Nadova, Z., Jancura, D., et al. (2012). Development of a new LDL-based transport system for hydrophobic/amphiphilic drug delivery to cancer cells. *International Journal of Pharmaceutics*, *436*(1-2), 463–471.

Job, P. (1928). Job's method of continuous variation. *Annali di Chimica*, *10*, 113–203.

Kapinus, E. I., Falk, H., & Tran, H. T. N. (1999). Spectroscopic investigation of the molecular structure of hypericin and its salts. *Monatshefte für Chemie/Chemical Monthly*, *130*(5), 623–635.

Lavie, G., Mazur, Y., Lavie, D., & Meruelo, D. (1995). The chemical and biological properties of hypericin—A compound with a broad spectrum of biological activities. *Medicinal Research Reviews*, *15*(2), 111–119.

Li, J., Xiao, H., & Zhong, Y. (2004). Drug carrier systems based on water-soluble cationic beta-cyclodextrin polymers. *International Journal of Pharmaceutics*, *278*(2), 329–342.

Liu, Y., Yang, Y. W., Chen, Y., & Ding, F. (2005). Efficient fluorescent sensors of oligopeptides by dithiobis(2-benzoylamide)-bridged bis(beta-cyclodextrin)s: Structure in solution, binding behavior, and thermodynamic origin. *Bioorganic & Medicinal Chemistry*, *13*(4), 963–971.

Negi, J. S., & Singh, S. (2013). Spectroscopic investigation on the inclusion complex formation between amisulpride and γ-cyclodextrin. *Carbohydrate Polymers*, *92*(2), 1835–1843.

Renard, E., Deratani, A., Volet, G., & Sebille, B. (1997). Preparation and characterization of water soluble high molecular weight β-cyclodextrin-epichlorohydrin polymers. *European Polymer Journal*, *33*(1), 49–57.

Sattler, S., Schaefer, U., Schneider, W., Hoelzl, J., & Lehr, C.-M. (1997). Binding, uptake, and transport of hypericin by Caco-2 cell monolayers. *Journal of Pharmaceutical Sciences*, *86*(10), 1120–1126.

Saw, C. L., Olivo, M., Soo, K. C., & Heng, P. W. (2006). Delivery of hypericin for photodynamic applications. *Cancer Letter*, *241*(1), 23–30.

Schneider, H.-J., Hacket, F., Rüdiger, V., & Ikeda, H. (1998). NMR studies of cyclodextrins and cyclodextrin complexes. *Chemical Reviews*, *98*(5), 1755–1786.

Smirnov, A. V., Das, K., English, D. S., Wan, Z., Kraus, G. A., & Petrich, J. W. (1999). Excited-state intramolecular H atom transfer of hypericin and hypocrellin A investigated by fluorescence upconversion. *The Journal of Physical Chemistry A*, *103*(40), 7949–7957.

Szejtli, J. (1998). Introduction and general overview of cyclodextrin chemistry. *Chemical Reviews*, *98*(5), 1743–1754.

Tamaki, T. (1984). Reversible photodimerization of water-soluble anthracenes included in γ-cyclodextrin. *Chemistry Letters*, *13*(1), 53–56.

Tatsis, E. C., Exarchou, V., Troganis, A. N., & Gerothanassis, I. P. (2008). ^{1}H NMR determination of hypericin and pseudohypericin in complex natural mixtures by the use of strongly deshielded OH groups. *Analytica Chimica Acta*, *607*(2), 219–226.

Vandenbogaerde, A., Zanoli, P., Puia, G., Truzzi, C., Kamuhabwa, A., De Witte, P., et al. (2000). Evidence that total extract of hypericum perforatum affects exploratory behavior and exerts anxiolytic effects in rats. *Pharmacology Biochemistry and Behavior*, *65*(4), 627–633.

Yang, B., Lin, J., Chen, Y., & Liu, Y. (2009). Artemether/hydroxypropyl-beta-cyclodextrin host–guest system: Characterization, phase-solubility and inclusion mode. *Bioorganic & Medicinal Chemistry*, *17*(17), 6311–6317.

Zhang, W., Chen, M., & Diao, G. (2011). Preparation and electrochemical behavior of water-soluble inclusion complex of ferrocene with β-cyclodextrin polymer. *Electrochimica Acta*, *56*(14), 5129–5136.

Zhang, W., Chen, M., Zha, B., & Diao, G. (2012). Correlation of polymer-like solution behaviors with electrospun fiber formation of hydroxypropyl-beta-cyclodextrin and the adsorption study on the fiber. *Physical Chemistry Chemical Physics*, *14*(27), 9729–9737.

Zhao, D., Zhao, L., Zhu, C., Tian, Z., & Shen, X. (2009). Synthesis and properties of water-insoluble β-cyclodextrin polymer crosslinked by citric acid with PEG-400 as modifier. *Carbohydrate Polymers*, *78*(1), 125–130.