Bioorganic & Medicinal Chemistry 26 (2018) 2287–2290

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry

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

Selective binding and controlled release of anticancer drugs by polyanionic cyclodextrins

Jian-Guang Cheng [a], Hua-Jiang Yu [a], Yong Chen [a], Yu Liu [a,b,*]

[a] College of Chemistry, State Key Laboratory of Elemento-Organic Chemistry, Nankai University, PR China
[b] Collaborative Innovation Center of Chemical Science and Engineering (Tianjin), Tianjin 300071, PR China

ARTICLE INFO

Article history:
Received 6 February 2018
Revised 8 March 2018
Accepted 8 March 2018
Available online 10 March 2018

Keywords:
Polyanionic cyclodextrin
Selective binding
Controlled release
Anticancer drugs
Supramolecular chemistry

ABSTRACT

The binding stoichiometry, binding constants, and inclusion mode of some water-soluble negatively charged cyclodextrin derivatives, i.e. heptakis-[6-deoxy-6-(3-sulfanylpropanoic acid)]-β-cyclodextrin (**H1**), heptakis-[6-deoxy-6-(2-sulfanylacetic acid)]-β-cyclodextrin(**H2**), mono-[6-deoxy-6-(3-sulfanylpropanoic acid)]-β-cyclodextrin (**H3**) and mono-[6-deoxy-6-(2-sulfanylacetic acid)]-β-cyclodextrin (**H4**), with three anticancer drugs, i.e. irinotecan hydrochloride; topotecan hydrochloride; doxorubicin hydrochloride, were investigated by means of ^{1}H NMR, UV–Vis spectroscopy, mass spectra and 2D NMR. Polyanionic cyclodextrins **H1**-**H2** showed the significantly high binding abilities of up to 2.6 × 10^{4}–2.0 × 10^{5} M^{-1} towards the selected anticancer drugs, which were nearly 50–1000 times higher than the corresponding Ks values of native β-cyclodextrin. In addition, these polyanionic cyclodextrins also showed the pH-controlled release behaviors. That is, the anticancer drugs could be efficiently encapsulated in the cyclodextrin cavity at a pH value similar to that of serum but sufficiently released at an endosomal pH value of a cancer cell, which would make these cyclodextrin derivatives the potential carriers for anticancer drugs.

1. Introduction

Recently, numerous effective anticancer drugs have been used for the treatment of various human and animal cancers. Among them, irinotecan hydrochloride (**CPT-11**), topotecan hydrochloride (**TPT**) and doxorubicin hydrochloride (**DOX**) are three prominent leader compounds. **CPT-11** and **TPT** are both water-soluble semisynthetic derivatives of the alkaloid camptothecin.[1,2] **CPT-11** exhibits remarkable antitumor activity in clinical trials against a variety of human tumors,[3–5] including colorectal cancer, lung cancer and malignant lymphoma.[6–8] **TPT** is used clinically in the treatment of relapsed ovarian, lung cancer, and cervical cancer.[9–12] **DOX** is a chemotherapeutic agent used for the treatment of a wide variety of human malignancies with an anthracycline structure, which consists of an aglycon, adriamycinone, combined with an amino sugar, daunosamine.[13–16] On the other hand, cyclodextrins (CDs), a class of cyclic oligosaccharides linked by 1,4-glucose bonds, are water-soluble, nontoxic, compounds commercially available at low price,[17–23] and their torus-shaped cavity can bind various inorganic/organic/biological molecules. This excellent property enables the wide application of CDs in fields of molecular recognition and molecular assembly. Among the CD family, the most used one is β-CD that contains 7 glucose units.[24–28] Nevertheless, the complex stability constants (*Ks*) between native β-CD and anticancer drugs (**CPT-11**, **TPT** and **DOX**) are very limited,[29] i.e. 2.6 × 10^{2} M^{-1} for β-CD/**CPT-11** pair, 8.8 × 10^{3} M^{-1} for β-CD/**TPT** pair, and 2.1 × 10^{2} M^{-1} for β-CD/**DOX** pair respectively, which greatly restricts the application of β-CD as carriers of anticancer drugs. Recently, the negatively charged CD derivatives have attracted more attention because of their potential applications in drug delivery. For example, Zhang et al. reported a negatively charged CD named ORG25969 as a good acceptor to give an extraordinarily high binding affinity towards rocuronium bromide (*Ks* up to10^{7} M^{-1}), and thus can be clinically used as a reversal agent in the post-operative recovery.[30] Wenz and Apostolakis et al. synthesized a series of negatively charged CDs and researched their binding behaviors with camptothecin. The result showed that the stabilities of camptothecin complexes obtained from solubility measurements of negatively charged CD derivatives were significantly higher than those of other reported CD derivatives.[31,32] Herein, we selected four negatively charged CD derivatives, i.e. heptakis-[6-deoxy-6-(3-sulfanylpropanoic acid)]-β-CD (**H1**), heptakis-[6-deoxy-6-(2-sulfanylacetic acid)]-β-CD (**H2**), mono-[6-deoxy-6-(3-sulfanylpropanoic acid)]-β-CD (**H3**) and mono-[6-

* Corresponding author at: College of Chemistry, State Key Laboratory of Elemento-Organic Chemistry, Nankai University, PR China.
E-mail address: yuliu@nankai.edu.cn (Y. Liu).

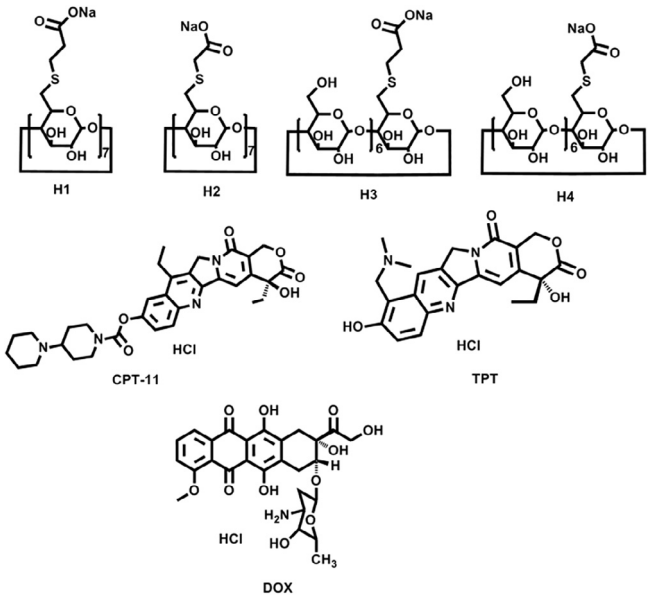
https://doi.org/10.1016/j.bmc.2018.03.013
0968-0896/

deoxy-6-(2-sulfanylacetic acid)]-β-CD (**H4**),[33] and investigated their selective binding and controlled release behaviors towards anticancer drugs **CPT-11**, **TPT** and **DOX** (Scheme 1). Significantly, with binding abilities much stronger than those of most previously reported CD derivates, these polyanionic CDs exhibited the pH-responsive release of drug in a cancer cell environment. That is, the polyanionic CD/anticancer drug complex was stable in a biological environment such as serum (pH 7.2), but efficiently released the encapsulated anticancer drug at pH 5.7 (endosomal pH values of a cancer cell).

2. Results and discussion

2.1. Job plots and binding constants of *H1*-*H4* and anticancer drugs

UV−vis spectroscopy was employed to determine the host–guest binding stoichiometry. As shown in Fig. 1, the Job plot of **H1**/**CPT-11** in water gave a maximum at molar fraction of 0.5, indicating that **H1** formed stoichiometric 1:1 inclusion complex with **CPT-11**. Moreover, the mass spectrum measurements (Figs. S31–S33) also demonstrated the formation of 1:1 inclusion complexes between cyclodextrin hosts and anticancer drugs. The quantitative investigation on the molecular binding behavior of **H1** with **CPT-11** was examined by means of UV−vis spectral titration, wherein the UV–vis spectra of a series of solutions containing the same amounts of **CPT-11** and different amounts of **H1** were measured to determine the binding constant between **CPT-11** and **H1**. As can be seen from Fig. S20, with the addition of **H1**, the absorbance maximum of **CPT-11** slightly decreased, accompanied by the appreciable red shift of maximum wavelength. By using the non-linear least-squares method,[34] the stability constants (*Ks*) values could be calculated as $(1.7 \pm 0.2) \times 10^5\ M^{-1}$ according to the sequential changes of absorbance intensity of **CPT-11** with the different concentrations of **H1**. Similar 1:1 binding stoichiometry was also found in the association of hosts **H1**-**H4** with anticancer drugs **CPT-11**, **TPT** and **DOX**, and the corresponding stability constants (*Ks*) were determined (Fig. 2) and listed in Table 1. Moreover, we also tried to use isothermal titration calorimetry to determine the binding constants. However, the isothermal titration calorimetry experiment required the higher concentrations, and the inclusion complex formed precipitate under such a concentration.



Scheme 1. Chemical structures of host and guest.

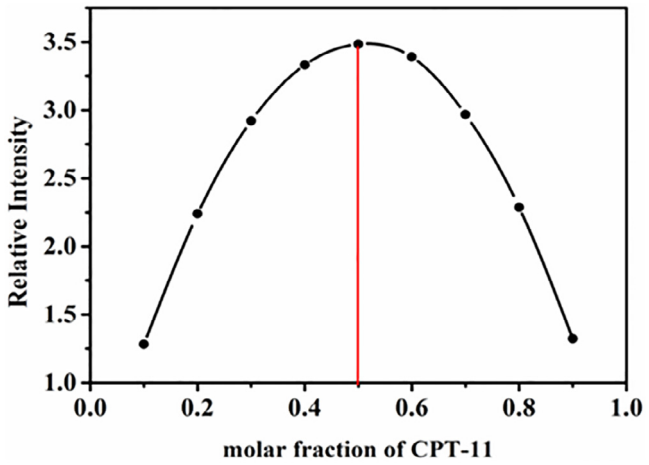


Fig. 1. Job plot for the binding of **H1** with **CPT-11** in water at 25 °C, indicating a 1:1 stoichiometry. The changes of absorbance were measured at 369 nm, and the total concentration was maintained at 0.1 mM.

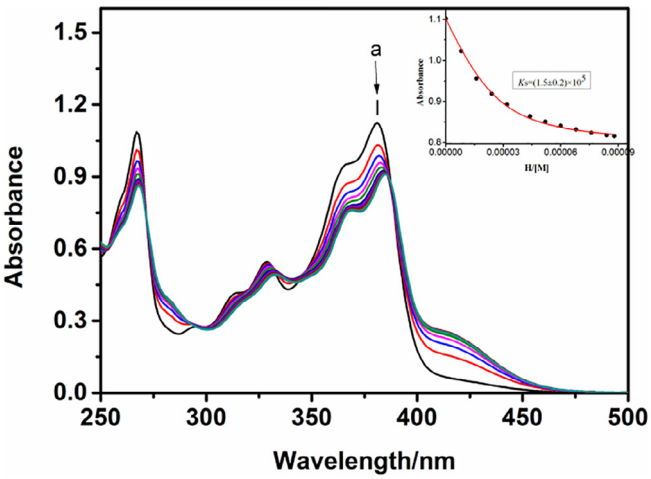


Fig. 2. UV–vis spectral titration of **TPT** upon addition of **H1** in H_2O at 25 °C. The nonlinear least-squares analysis (inset) of the differential absorbance to calculate the complex stability constant. The changes of absorbance were measured at 381 nm. ([**TPT**] = 0.05 mM, **H1** = 0.00, 0.01, 0.02, 0.04, 0.06, 0.08, 0.10, 0.12, 0.14, 0.16, 0.18, 0.20, 0.22 mM from a to l).

Accordingly, the encapsulation and loading efficiency of anticancer drugs by hosts were calculated and listed in Table 1. As seen in Table 1, the native β-CD only showed very poor binding ability towards the selected anticancer drugs. Possessing an anionic side arm on the β-CD rim, host **H3** or **H4** showed the moderate binding ability (1.02×10^3–$1.7 \times 10^4\ M^{-1}$) towards anticancer drugs owing to the electrostatic interactions between the anionic side arm of host and the cationic guest. However, host **H1** or **H2** showed a significantly increased binding ability towards anticancer drugs up to 2.6×10^4–$2.0 \times 10^5\ M^{-1}$, which was nearly 50–1000 times higher than the corresponding *Ks* values of native β-CD. A possible reason may be that the seven anionic side arms on **H1** or **H2** (either of **H1** or **H2** possesses 7 negative charges) gave the greatly strengthened electrostatic interactions with the cationic guest. Moreover, the extended cavity formed by seven side arms may also provide the additional van der Waals and hydrophobic interactions towards the accommodated drug. As a result, host **H1** exhibited the fairly high encapsulation efficiency (>75%) and loading efficiency (>18%) towards the selected anticancer drugs when the concentrations of anticancer drugs and hosts were fixed at 0.1 mM, which enables it as a good candidate of anticancer drug carriers. The anti-