Journal of Inclusion Phenomena and Macrocyclic Chemistry (2005) 52: 109–117
DOI 10.1007/s10847-004-6048-3

Cyclodextrin Solubilization of the Antibacterial Agents Triclosan and Triclocarban: Effect of Ionization and Polymers

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(Received: 21 June 2004; in final form: 10 November 2004)

Key words: ammonia, complexation efficiency, cyclodextrin, ionization, solubility, triclocarban, triclosan

Abstract

The natural β-cyclodextrin (βCD) and its complexes have limited solubility in aqueous solutions. This low aqueous solubility, as well as low aqueous solubility of the guest molecule (i.e. triclosan or triclocarban (TCC)), can result in low complexation efficiency (CE). The purpose of this study was to enhance the apparent intrinsic solubility (S_0) of the guest molecule and its βCD complexes through ionization and addition of auxiliary compounds such as polymers, amino acids and metal ions. Both triclosan (pK_a 7.9) and TCC (pK_a 12.7) are weak acids. Addition of ethanol to the complexation medium enhanced S_0 of both triclosan and TCC but at the same time ethanol lowered the stability constant (K_c) of their βCD complexes resulting in overall lowering of CE. Addition of small amount of water-soluble polymers enhanced the βCD solubilization of both guests, and addition lysine enhanced the solubilization of TCC. Ionization of triclosan resulted in significant enhancement of CE and enhanced triclosan release from tablets containing triclosan/βCD complex. The effect of ionization was not as pronounced in the case of TCC.

Introduction

In general, all drugs and all antibacterial agents have to possess some degree of aqueous solubility to be biologically active and most drugs and antibacterial agents have to be somewhat lipophilic to be able to permeate biological membranes via passive diffusion. How water-soluble a given drug or antibacterial agent needs to be is determined by its potency and type of formulation. Though complex formation cyclodextrins are able to enhance the aqueous solubility of lipophilic water-insoluble compounds without changing their intrinsic ability to permeate biological membranes. Enhanced aqueous solubility of the solid cyclodextrin complex, compared to the pure compound, will increase the dissolution rate of the drug in an aqueous environment. Faster dissolution and higher concentrations of dissolved drug (or antibacterial agent) will lead to faster onset and more potent action. However, cyclodextrin complexation will increase the formulation bulk, which can limit their usefulness as pharmaceutical excipient.

Triclosan (Irgasan DP 300 or cloxifenol) and triclocarban (TCC or 3,4,4′-trichlorocarbanilide) are used as antibacterial agents in a wide variety of consumer products. Lately it has been shown that triclosan also possesses anti-malaria [1] and anti-inflammatory activity [2], and these and other discoveries have resulted in increased interest in pharmaceutical applications of triclosan. Triclosan is lipophilic, low molecular weight compound that permeates the skin barrier [3]. Both triclosan and TCC are practically insoluble in water, which hampers their usage in water-soluble or aqueous based cosmetics and pharmaceutical products.

Cyclodextrins have recently been recognized as enabling pharmaceutical excipients. These are cyclic oligosaccharides with a hydrophilic outer surface and a lipophilic cavity in the center [4]. Cyclodextrins are able to form hydrophilic inclusion complexes with many lipophilic compounds. In aqueous solutions molecules bound within the inclusion complex are in a dynamic equilibrium with free molecules. Thus, cyclodextrins enhance the aqueous solubility of many lipophilic compounds without changing their intrinsic ability to permeate lipophilic membranes [5]. The natural β-cyclodextrin (βCD) and its inclusion complexes have limited aqueous solubility. Various water-soluble cyclodextrin derivatives have been synthesized. Cyclodextrin derivatives of current pharmaceutical interest include 2-hydroxypropyl-β-cyclodextrin (HPβCD), 2-hydroxypropyl-γ-cyclodextrin, sulfobutylether β-cyclodextrin, randomly methylated β-cyclodextrin(RMβCD), and some branched cyclodextrins such as glucosyl-β-cyclodextrin. Previously it has been shown that triclosan forms complexes with βCD, sulfobutylether β-cyclodextrin,

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HPβCD, RMβCD and γ-cyclodextrin, and that the complexation can enhance the bioavailability and antibacterial activity of the compound [6–9].

Including cyclodextrins in pharmaceutical formulations will increase the formulation bulk of solid dosage forms. Even under best conditions, cyclodextrin complexation will result in 4- to 10-fold increase in the formulation bulk [10]. This limits the use of cyclodextrins in solid oral dosage forms to potent drugs that possess good complexing properties. Cyclodextrin derivatives have greater molecular weight than their parent cyclodextrins and thus result in greater increase in the formulation bulk than the natural cyclodextrins, i.e. in the case of natural cyclodextrins the drug occupies greater fraction of the complex powder [11]. Most drug molecules (D) form 1:1 complexes with cyclodextrin molecules (CD) and the value of the stability constant ($K_{1:1}$) is most often between 50 and 2000 M^{-1} with a mean value of 129, 490 and 355 M^{-1} for α-, β- and γ-cyclodextrin, respectively [12–15]:

$$\mathrm{D} + \mathrm{CD} \overset{K_{1:1}}{\rightleftharpoons} \mathrm{D/CD\ Complex}$$

$$\text{or} \quad K_{1:1} = \frac{[\mathrm{D/CD}]}{[\mathrm{D}] \cdot [\mathrm{CD}]} = \frac{[\mathrm{D/CD}]}{S_0 \cdot [\mathrm{CD}]}$$

In a given aqueous complexation medium, saturated with the drug, the concentration of free drug ([D]) is constant and equal to the apparent intrinsic solubility (S_0) of the drug in the aqueous medium (i.e., drug solubility in absence of cyclodextrin). The complexation efficiency (CE) can be defined as [10]:

$$\mathrm{CE} = K_{1:1} \cdot S_0 = \frac{[\mathrm{D/CD}]}{[\mathrm{CD}]}$$

Increased CE can be obtained by increasing either $K_{1:1}$ or S_0, or by increasing both $K_{1:1}$ and S_0 simultaneously. Here it is assumed that both cyclodextrin and the drug/cyclodextrin complex (D/CD) are freely soluble in the aqueous complexation medium. However, the natural cyclodextrins and their complexes have limited solubility in aqueous solutions. For example, the aqueous solubility of βCD is only about 18.6 mg/ml at room temperature [16]. Enhancing solubility of the natural cyclodextrins during production of solid complex powder could enhance their CE. It has been shown that addition of small amounts of water-soluble polymers to the aqueous complexation medium increases CE of cyclodextrins [17–21] and that the polymers form ternary complexes with drug/cyclodextrin complexes [20, 22, 23]. Furthermore, it has been shown that cyclodextrins and cyclodextrin complexes self-associate to form aggregates and that those aggregates can act as solubilizers themselves [24–27]. There are some indications that the water-soluble polymers enhance the complexation efficiency by stabilizing these aggregates [17, 26, 28]. The aim of the present study was to prepare triclosan and TCC complexes with the natural βCD that possessed dissolution rates that are comparable to triclosan and TCC complexes of the more water-soluble βCD derivatives.

Experimental

Materials

Triclosan from Sigma (USA), triclocarban (TCC) from Colgate-Palmolive Co. (USA), βCD and RMβCD of degree substitution 1.8 from Wacker Chemie (Germany), HPβCD of molar substitution 0.6 from Roquette (France), hydroxypropyl methylcellulose 1000 (HPMC) from Norsk Medisinaldeport (Norway) and polyvinylpyrrolidone 40,000 (PVP) from Sigma (USA). Extra pure ammonia solution 32% was purchased from Merck (Germany). All other chemicals and solvents used in this study were commercial available products of analytical or special reagent grade.

Solubility studies

The solubility of triclosan and TCC was determined in aqueous complexation media containing 0–40% (w/v) HPβCD, 0–20% (w/v) RMβCD or 0–20% (w/v) βCD in solution or suspension, in pure water or aqueous polymer solutions containing from 0 to 10% (v/v) ammonia, or aqueous ethanolic solutions. An excess amount of triclosan or TCC was added to the aqueous cyclodextrin solution/suspension and the suspension formed heated in a sealed vial in ultrasonic bath (70 °C for 60 min). The suspension was heated to promote drug, and in some cases cyclodextrin, saturation of the aqueous complexation medium. After equilibration at room temperature (22–23 °C) over night the vials were opened, small amount of solid triclosan or TCC added to each vial and the aqueous suspensions allowed to equilibrate in the resealed vial at room temperature under constant agitation for additional six days. This was done to promote precipitation. Preliminary experiments showed that six days are more than enough time to reach solubility equilibrium. The chemical stability of the compounds was also monitored during heating and equilibration period and in all cases less that 0.5% degradation was observed. After equilibration the suspensions were filtered through 0.45-μm nylon membrane filters and the filtrate analyzed by HPLC. Phase-solubility profiles were obtained by plotting the solubility of triclosan or TCC versus the cyclodextrin concentration. The pK_a value of TCC was determined from the pH-solubility profile of the compound in aqueous 5% (w/v) HPβCD solution containing various amounts of ammonia [29]. The pH of the aqueous complexation media was determined at room temperature at the end of the 7-day equilibration period (Corning pH meter 24, UK).

The phase-solubility of naphthalene was determined in 0–20% (w/v) βCD solutions/suspensions in pure

water and aqueous 10% (v/v) ammonia solution. An excess amount of naphthalene was added to the aqueous βCD solution/suspension and the suspension formed shaken in a sealed container for 48 h in a mechanical shaker (Edmund Bühler KL 2, Germany). After equilibration the suspensions were filtered through 0.45-μm nylon membrane filters and the amount of dissolved naphthalene in the filtrate determined by UV (Perkin Elmer Lambda 35, UK) at 275 nm. Standard solutions of naphthalene were prepared in aqueous 20% HPβCD solution.

Determination of the stability constants

The stability constants ($K_{1:1}$) of triclosan/βCD 1:1 complex in pure aqueous solutions and ethanolic solutions containing 1% ammonia, was determined from the linear phase-solubility profiles [30]. In aqueous ethanolic solutions the stability constant of the unionized triclosan/βCD 1:1 complex was determined by UV spectrophotometry (Perkin Elmer Lambda 35, UK) monitoring changes in the molar absorptivity of triclosan at 283 nm and plotting the result according to the Scott equation [31].

Preparation of solid cyclodextrin complexes

HPβCD (20% w/v) or βCD (20% w/v) was dissolved or suspended in distilled water and equal molar amount of triclosan or TCC added to the solution/suspension. Then up to 30% (v/v) aqueous 32% ammonia solution was pipetted into the aqueous suspension (resulting in up to 10% (v/v) aqueous ammonia solution). The solutions were subsequently heated in an autoclave (10 min at 121 °C) to dissolve triclosan or TCC, and βCD. The solutions were then placed in a shaker for 1 h, during cooling, and the liquid subsequently evaporated in a Büchi 011 rotation evaporator (Switzerland) at 40 °C. Then the solid complex was dried for 12 h in a vacuum oven (Gallenkamp, UK) at 40 °C and 9 Torr. The solid powder was finally sieved through 300- μm sieve and 30–50 mg reserved for analysis. The amount of triclosan or TCC in the complex powder was determined by HPLC.

Dissolution studies

Tablets (200 mg) containing dry complex of triclosan or TCC and βCD or HPβCD (corresponding to 19.8 mg triclosan or 17.0 mg TCC), 40 mg Avicel and glucose ad 200 mg, were prepared in an IR-press (Specac Ltd, UK) under 900 ton/sqi pressure for 1 min.

The dissolution profiles of the tablets were obtained using paddle type dissolution apparatus (Ph. Eur., 4th Ed., 2002, p. 194) from Prolabo (France), operated at ambient temperature (22 °C). The dissolution medium (500 ml) consisted of aqueous 0.01% (v/v) hydrochloric acid solution containing 1% (w/v) HPβCD (triclosan), or water containing 8% (w/v) HPβCD (TCC). The paddle was rotated at 100 rpm. At various time points 1 ml samples were withdrawn from the dissolution media, filtered through a 45-μm nylon membrane filter and quantified undiluted on HPLC.

Quantitative determinations

The amount of triclosan and TCC in the solid complex powder and in the aqueous solutions was determined on a high performance liquid chromatographic (HPLC) component system, consisting of ConstaMetric 3200 solvent delivery system operated at 1.5 ml/min, a SpectroMonitor 3200 UV/VIS variable-wavelength detector operated at 283 nm, a Merck-Hitachi AS-2000A autosampler, Merck Hitachi D-2500 Chromato-Integrator and a Phenomex ODS 5-μm (150 × 4.6 mm) column. The mobile phase for determination of triclosan consisted of acetonitrile and water (74:26), and the retention time was 2.9 min. The mobile phase for determination of TCC consisted of acetonitrile and water (70:30), and the retention time was 2.6 min.

An enzymatic assay was used for quantitative determination of ammonia in the dry complex powder (cat. no. 11 112 732 035, Boehringer Mannheim, Germany).

Results and discussion

The intrinsic solubility (S_0) of triclosan and TCC in pure water at room temperature was estimated to be about or less than 1 μg/ml and less than 50 ng/ml, respectively. The pK_a value of triclosan was obtained from the literature but that of TCC was estimated from the pH-solubility profile (Figure 1) to be about 12.7 in aqueous 5% (w/v) HPβCD solution at room temperature. The

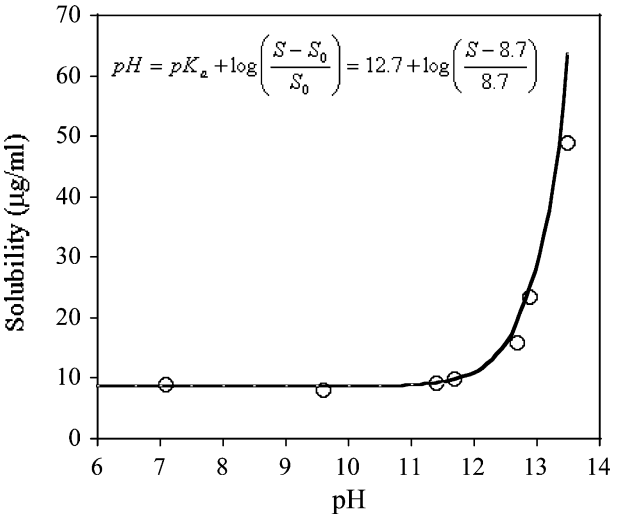
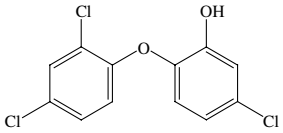
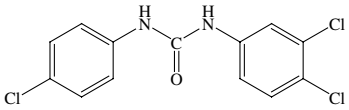


Figure 1. Plot of solubility (S) of TCC in aqueous ammonium solutions containing 5% (w/v) HPβCD as a function of pH at room temperature (22–23 °C). The pKa of TCC was estimated to be 12.7 and the intrinsic solubility of TCC in the aqueous cyclodextrin solution (S_0) to be 8.7 μg/ml. The experimental data was fitted to the equation shown (the solid line).

Table 1. Structure and physicochemical properties of triclosan and triclocarban (TCC). Based on references [32–34] and data obtained during this present investigation

	Triclosan	Triclocarban
		
Molecular weight	289.5 Da	315.6 Da
Melting point	54–57 °C	255–256 °C
Log $P_{(octanol/water)}$	4.8	4.9
pK_a	7.9	12.7[a]
Solubility at room temperature:		
Water	≤1 μg/ml	<50 ng/ml
Acetone	Very soluble	40 mg/ml
Propylene glycol	Very soluble	10 mg/ml
MIC (μg/ml) for:		
S. aureus	0.1	0.5
E. Coli	5	>5
P. aeruginosa	>300	>5

[a]In aqueous 5% (w/v) HPβCD solution.

physicochemical properties and antibacterial activities of triclosan and TCC are shown in Table 1.

Phase-solubility studies and the effect of ionization

The phase-solubility diagram of triclosan in aqueous βCD solution/suspension (Figure 2) is of type B_S, reaching maximum triclosan solubility of 0.08 mg/ml at 1% βCD concentration but the solubility decreases at βCD concentration greater than 5% [30]. The phase-solubility diagram of triclosan in aqueous HPβCD solution is of type A_P suggesting formation of higher order complexes with respect to HPβCD or formation of complex aggregates [26]. Triclosan is a lipophilic, weak acid with pK_a of 7.9 (Table 1). It is possible to enhance the apparent intrinsic solubility of triclosan from about 1 μg/ml to about 7 mg/ml by fully ionizing the compound in the aqueous complexation media (i.e. by keeping the pH above 10). Ionizing triclosan resulted in sharp increase in the overall triclosan solubility in 20% cyclodextrin solution/suspension (Figure 3), reaching maximum solubility at 2% (v/v) ammonia in the HPβCD solution but at 4% (v/v) ammonia in the βCD suspension. The effect of ammonia on the phase-solubility of triclosan in aqueous βCD media is shown in

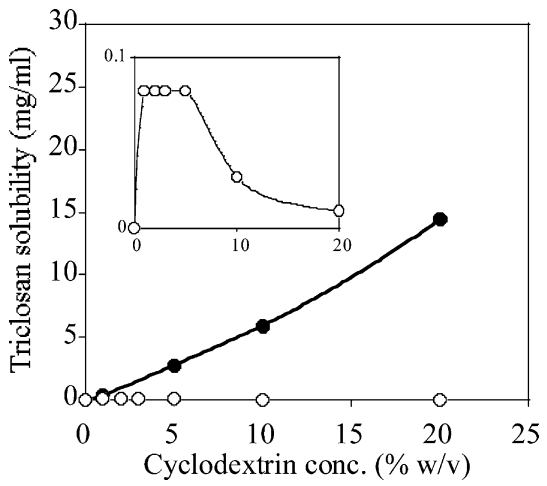


Figure 2. Phase-solubility plot of triclosan in aqueous βCD solution/suspension (○) and aqueous HPβCD solution (●).

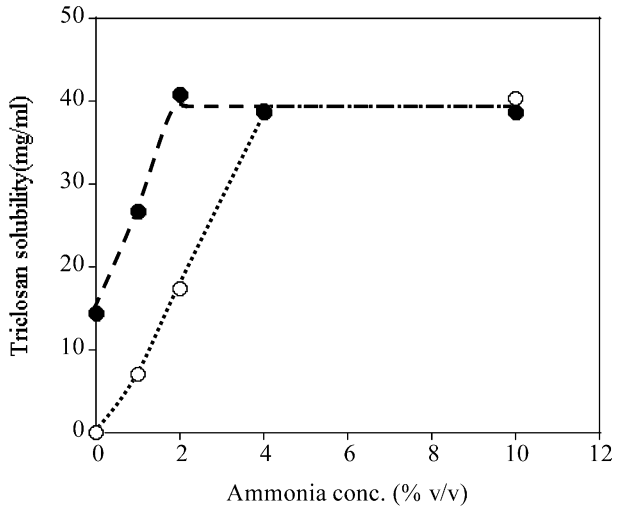


Figure 3. The effect of ammonia on triclosan solubility in aqueous 20% (w/v) βCD suspension (○) or in aqueous 20% (w/v) HPβCD solution (●). The pH in aqueous 1.0% (v/v) ammonia solution was determined to be 10.2 ± 0.4 (mean ± standard deviation, $n = 3$), it was 11.2 ± 0.2 in the 4% (v/v) solution and 11.4 ± 0.0 in the 10% (v/v) solution.

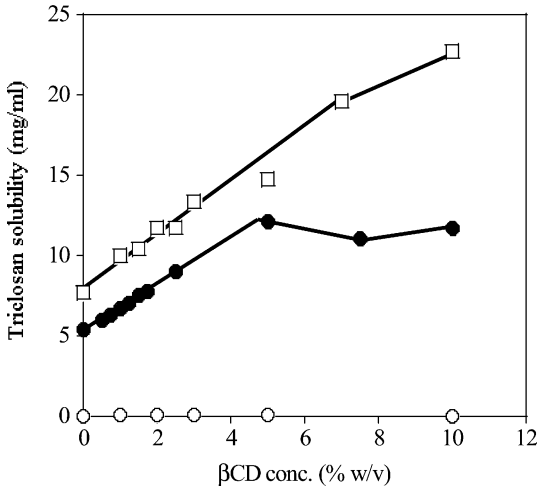


Figure 4. The effect of ammonia on the phase-solubility of triclosan in aqueous βCD solutions/suspensions; no ammonia present in the aqueous complexation media (pH 6.8 ± 0.2, mean ± standard deviation) (○), 1% (v/v) ammonia (●) and 4% (v/v) ammonia (pH 11.2 ± 0.2) (□).

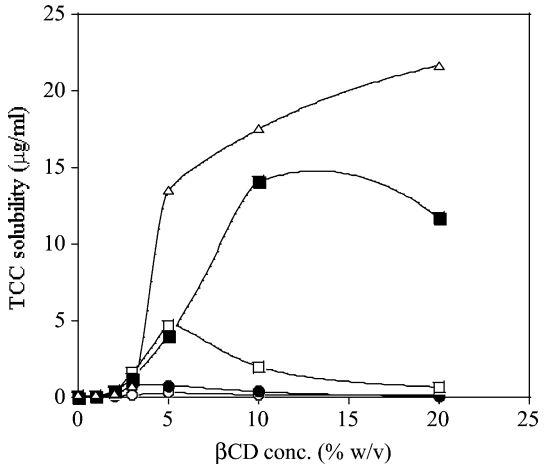


Figure 6. The effect of ammonia on the phase-solubility diagram of TCC in aqueous βCD solution/suspension; no ammonia, pH 6.3 ± 0.2 (○), 1% (v/v) ammonia, pH 11.6 ± 0.1 (●), 4% (v/v) ammonia, pH 12.1 ± 0.1 (□), 10% ammonia, pH 12.1 ± 0.1 (■), and 10% ammonia and 0.25% (w/v) PVP, pH 11.7 ± 0.2 (Δ).

Figure 4. The results indicate that at least 4% (v/v) ammonia is needed to fully solubilize the triclosan/βCD complex.

The phase-solubility diagram of TCC in aqueous βCD solution/suspension (Figure 5) is of type B_S reaching maximum solubility of 0.5 μg/ml at 5% βCD concentration. The phase-solubility diagrams of TCC in aqueous HPβCD and RMβCD solutions are of type A_P indicating formation of higher order complexes with respect to cyclodextrin or formation of some complex aggregates. For TCC RMβCD was found to be significantly better solubilizer than HPβCD. TCC is a lipophilic compound with pK_a of 12.7 and, thus, it is possible to increase the apparent intrinsic solubility (i.e. S_0) of TCC by increasing the pH of the aqueous complexation media. This resulted in enhanced TCC solubilization in the aqueous βCD media (Figure 6). However, the pK_a of the hydroxy groups on the βCD molecule have been determined to be about 12.2 [35] and, thus, raising the pH above 11 will also result in some ionization and enhanced solubility of the natural βCD. For example, the aqueous solubility of naphthalene/βCD complex is equal or less that the solubility of free naphthalene and, thus, the aqueous solubility of naphthalene was not increased by βCD complexation in pure water (pH about 7.0). Naphthalene cannot be ionized in aqueous solutions but increasing the pH of the aqueous complexation medium to about 12.0 enhanced significantly the naphthalene solubility in the aqueous βCD complexation medium (Figure 7). Thus, it is possible that some of the solubilizing effects seen in Figure 6 are due to ionization of βCD. However, the results indicate that at least 10% (v/v) ammonia is needed to fully solubilize the TCC/βCD complex.

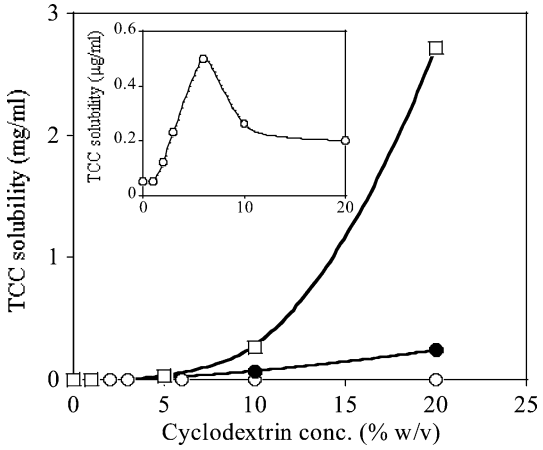


Figure 5. Phase-solubility plot of TCC in aqueous βCD solution/suspension (○), aqueous HPβCD solution (●) and aqueous RMβCD solution (□).

Effect of ethanol on the phase-solubility

It is possible to increase the apparent intrinsic solubility (S_0) of lipophilic water-insoluble compounds by decreasing the dielectric constant of the aqueous complexation media, for example by addition of ethanol. However, addition of ethanol to the aqueous complexation media resulted in notable decrease in triclosan

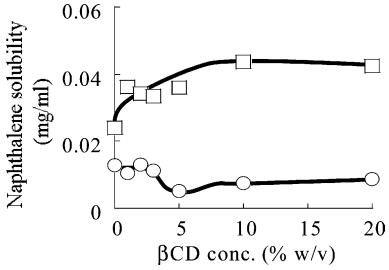


Figure 7. Phase-solubility of naphthalene in aqueous βCD solution/suspension; pure water, pH 7.0 ± 0.2 (mean ± standard deviation) (○); 10% (v/v) ammonium solution, pH 12.0 ± 0.1 (□).

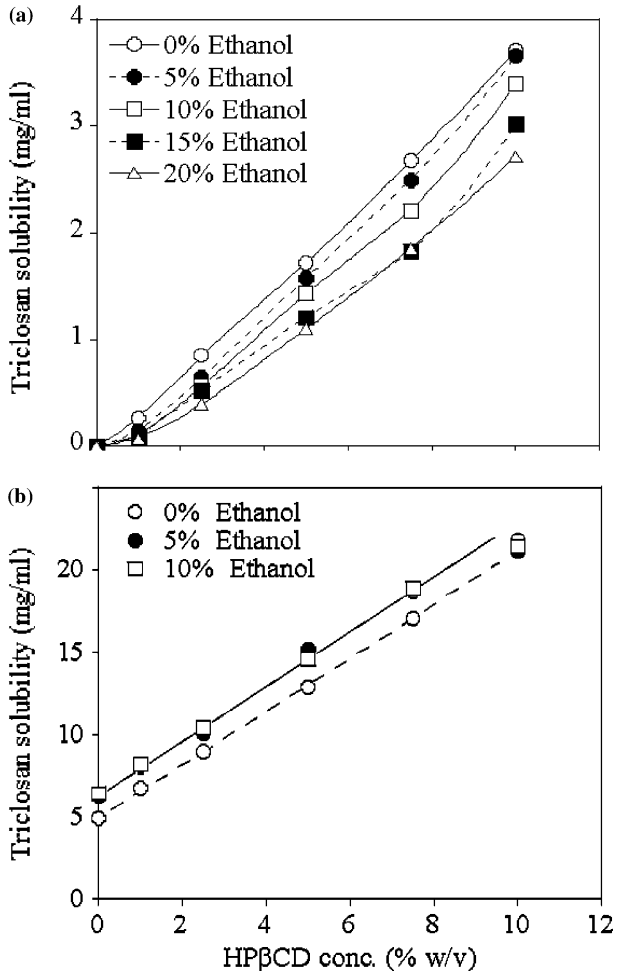


Figure 8. The effect of ethanol on the HPβCD solubilization of triclosan in aqueous complexation media. The complexation media consisted of water containing from 0 to 20% (v/v) ethanol; pure aqueous ethanol solution (a) and aqueous ethanol solution containing 1% (v/v) ammonia (b).

solubility when no ammonia was present and did not increase the slope of the linear phase-solubility (A_L-type) diagrams obtained in presence of 1% (v/v) ammonia (Figure 8). When no ammonia was present the apparent stability constant ($K_{1:1}$) of the triclosan/HPβCD 1:1 complex was estimated from the linear phase-solubility diagram (0% ethanol) but spectrophotometrically when ethanol was present (aqueous 5 and 10% v/v ethanol solutions) at and found to be about 10^4, 400 and 330 M^{-1} at ethanol concentration of 0, 5 and 10% (v/v), respectively. When 1% (v/v) ammonia was present in the aqueous complexation medium (pH about 10) $K_{1:1}$ was determined by the phase-solubility method to be 240, 130 and 120 M^{-1} and the complexation efficiency to be 4.0, 2.9 and 2.6 at ethanol concentration of 0, 5 and 10% (v/v), respectively. As expected, the more hydrophilic ionized form of triclosan has lower $K_{1:1}$ value than the lipophilic unionized form. Addition of ethanol decreased the value of $K_{1:1}$, perhaps by competing with triclosan for a space in the lipophilic cyclodextrin cavity. Although addition of ethanol resulted in some increase in S_0 this increase was not sufficient to compensate for the decrease in $K_{1:1}$ and, consequently, the complexation efficiency decreased when ethanol was added to the aqueous complexation media.

Addition of ethanol did not enhance HPβCD solubilization of TCC in aqueous solutions (Figure 9). Addition of 5% ethanol resulted in 60% decrease in TCC solubility in aqueous 10% HPβCD solution, and 55% decrease in aqueous 40% HPβCD solution. Less solubility decrease was observed in the solution containing 4% ammonia, and less decrease was observed in the 45% ethanolic solutions than in the 5% solutions. This was most probably due to changes in S_0. The solubility of TCC in pure water as well as in aqueous 5% ethanolic solution was determined to be less than 0.05 μg/ml, 0.05 μg/ml in aqueous 5% ethanolic solu-

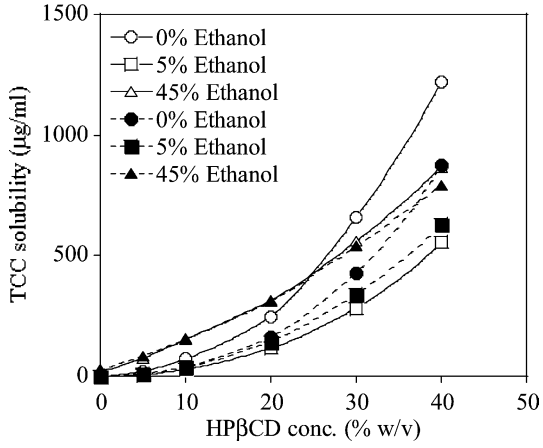


Figure 9. The effect of ethanol on the HPβCD solubilization of TCC in aqueous complexation media. The complexation media consisted of water containing from 0 to 45% (v/v) ethanol. Open marks: pure aqueous ethanol solution (pH about 6.5); closed marks: aqueous ethanol solution containing 4% (v/v) ammonia (pH about 12).

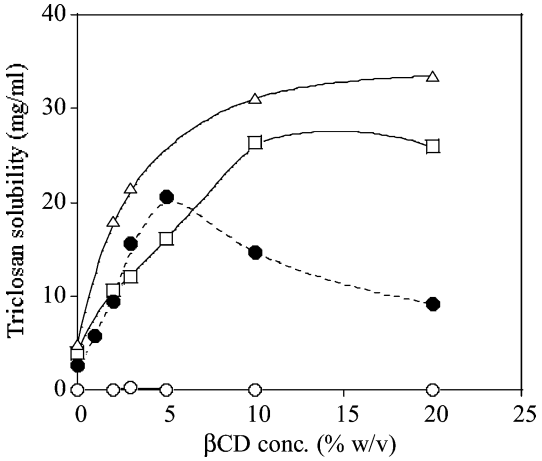


Figure 10. The effect of addition of 0.25% (w/v) PVP (○), 4% (v/v) ammonia (□), 4% (v/v) ammonia and 0.25% (w/v) PVP (Δ), or 4% (v/v) ammonia and 50 mM lysine (●) on triclosan solubility in aqueous βCD solutions/suspensions.

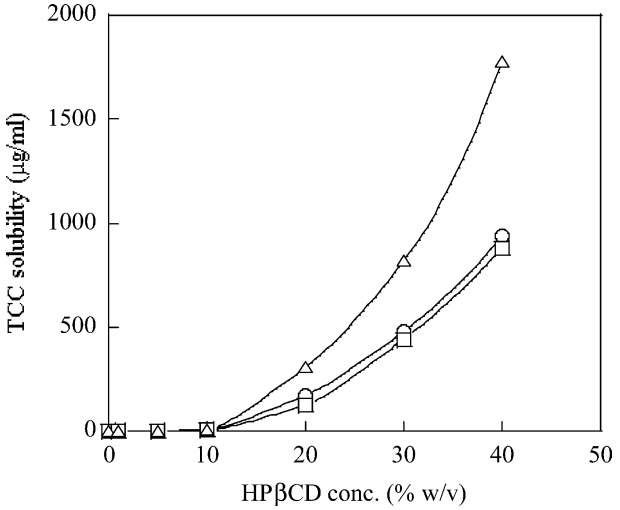


Figure 11. The effect TCC ionization and polymer on TCC solubility in HPβCD solutions; pure water, pH 8.0 ± 0.4 (○); aqueous 10% (v/v) ammonia, pH 11.6 ± 0.1 (□); aqueous 10% (v/v) ammonia and 0.25% (w/v) PVP, pH 11.7 ± 0.2 (Δ).

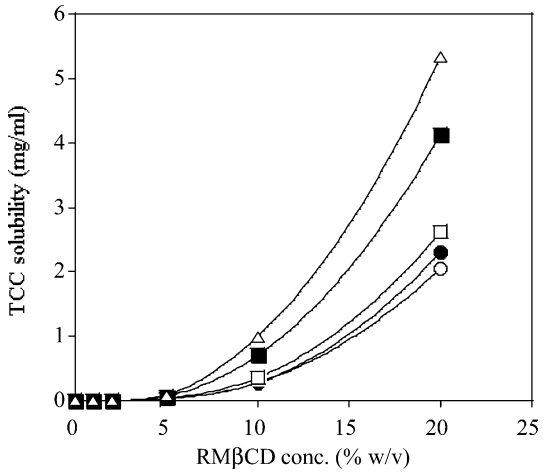


Figure 12. The effect of PVP, magnesium ions (Mg^{2+}), ascorbic acid and lysine on the RMβCD solubilization of TCC. Pure aqueous RMβCD solutions (○), aqueous 0.25% (w/v) PVP solution (●), aqueous 0.25% (w/v) PVP solution containing 50 mM Mg^{2+} (□) aqueous 0.25% (w/v) PVP solution containing 50 mM Mg^{2+} and 50 mM ascorbic acid (■), and aqueous 0.25% (w/v) PVP solution containing 50 mM Mg^{2+} and 50 mM lysine (Δ).

tion containing 4% ammonia, 20 μg/ml in aqueous 45% ethanolic solution and 27 μg/ml in aqueous 45% ethanolic solution containing 4% ammonia. However, this increase in S_0 was not sufficient to compensate for the decrease in the apparent stability constant of the TCC/HPβCD complex and again decreased complexation efficiency was observed when ethanol was added to the aqueous complexation media. The results indicate that addition of ethanol to the aqueous complexation media will not enhance the complexation efficiency of triclosan and TCC.

The effects of polymers, metal ions and lysine

Addition of small amount of polymers to the aqueous complexation media enhanced the solubility of both triclosan and TCC, and both of the unionized and the ionized forms of the compounds. Figure 10 shows the effect of PVP and lysine on the βCD solubilization of triclosan. The polymer enhances the βCD solubilization of triclosan but lysine does not. Similar results were obtained in aqueous HPβCD solutions where polymers, such as PVP and HPMC, enhanced the solubilization but lysine had little or no effect. However, these same additives enhanced the cyclodextrin solubilization of TCC. Partly ionization of TCC in the aqueous complexation medium increased significantly the solubility of TCC in aqueous βCD solutions/suspensions (Figure 6) and aqueous HPβCD solutions (Figure 11), especially when 0.25% PVP was present in the solution. However, largest solubilization was obtained in aqueous solutions containing RMβCD and 0.25% PVP. Preliminary studies showed that addition of polymers (0.10–0.25% w/v) to the aqueous complexation medium enhanced the solubility of TCC in 20% (w/v) RMβCD from 2.0 to 2.3 mg/ml, and that addition of metal ions (50 mM $MgCl_2$, $ZnCl_2$ or $FeCl_3$) enhanced it even further to 2.6 mg/ml. Including both metal ions and vitamins (50 mM of ascorbic acid or nicotine amide) in the same complexation media enhanced the TCC solubility even further or up to 4 mg/ml. However, the largest solubilization was obtained when amino acids (such as tryptophan, cysteine, leucine, threonine or glutamic acid) were added to the complexation media. Significant enhancement was also observed when adipic acid was included in the complexation media, resulting in TCC solubility of about 5 mg/ml. Figure 12 shows the effect of PVP, $MgCl_2$, ascorbic acid and lysine on the RMβCD solubilization of TCC. Other investigators have shown that metal ions and amino acids, as well as other acids and bases, can enhance cyclodextrin solubilization of water-insoluble compounds. For example, Yamakawa and Nishimura [19] have shown that Mg^{2+} ions have synergic effect on HPβCD solubilization of a quinolone and that PVP further enhanced the solubilization. Mura et al. [36] have shown that various amino acids enhance HPβCD solubilization of naproxen and several investigators have shown that hydroxy acids can enhance cyclodextrin solubilization of basic drugs and that organic bases can enhance cyclodextrin solubilization of acidic drugs [37, 38]. Finally, in our previous work we have shown that salts of organic acids, such as sodium acetate, and quaternary ammonium compound, such as benzalkonium chloride, can enhance βCD solubilization of uncharged compounds, such as hydrocortisone, and that addition of water-soluble polymer to the complexation medium enhances the overall solubilizing effect [28]. There are some indications that these auxiliary compounds do not participate directly in the inclusion complex formation but interact with complex aggregates formed in the aqueous complexation media [17].

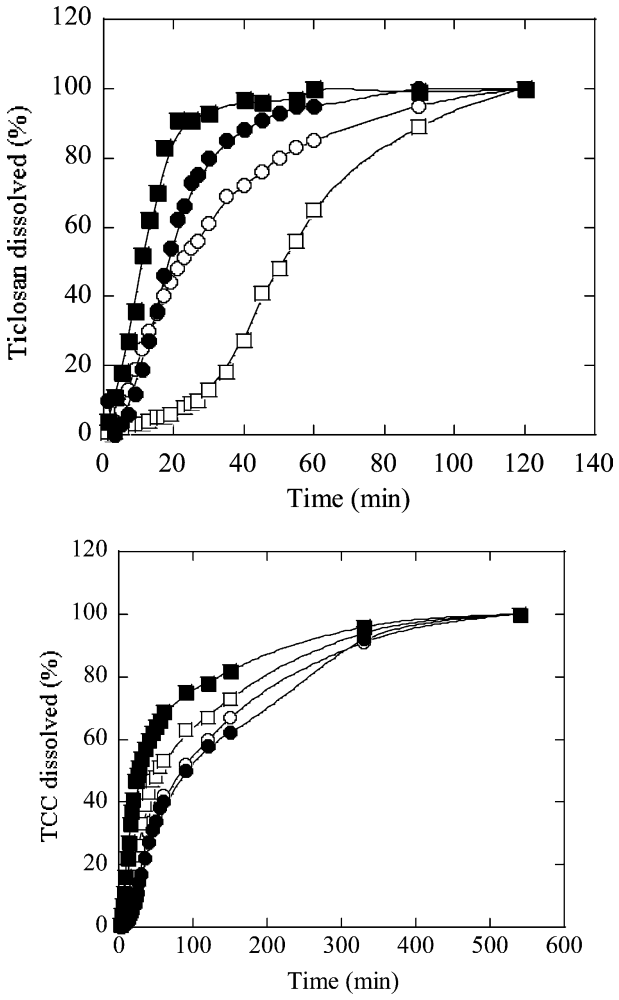


Figure 13. The dissolution profiles of triclosan and TCC in aqueous solutions (and mole ratio in dry complex); tablets containing triclosan/HPβCD (0.7:1.0) and TCC/HPβCD (0.7:1.0) complex prepared in water (■), triclosan/βCD (1.2:1.0) and TCC/βCD (0.6:1.0) complex prepared in aqueous 10% (v/v) ammonia solution (●), triclosan/βCD (0.9:1.0) and TCC/βCD (0.8:1.0) complex prepared in 4% ammonia (□), and equivalent amount of uncomplexed triclosan and TCC in a cyclodextrin free tablet (○). The data points represent the mean of three experiments.

Dissolution studies

Addition of ammonia to the aqueous complexation medium (i.e. increasing the apparent S_0 through ionization) enhanced the complexation efficiency, increasing the triclosan/βCD mole ratio from 0.7:1.0 in pure water to 1.2:1.0 in aqueous 10% ammonia solution. The pK_a value of TCC was determined to be 12.7 compared to 7.9 for triclosan and, thus, addition of ammonia had much less effect on the complexation efficiency of TCC. In fact decreased complexation efficiency was observed in aqueous 10% ammonia solution (TCC/βCD mole ratio 0.6:1.0) compared to 4% ammonia solution (TCC/βCD mole ratio 0.8:1.0). This decrease in efficiency could partly be due to ionization of the βCD molecule in the 10% ammonia solution and electrostatic repulsions between the anionic TCC and anionic βCD. The amount of ammonia in the dried complex powders was in all cases negligible or less than 0.001 mole per mole triclosan or TCC. The dissolution profiles (Figure 13) show that cyclodextrin complexation enhances the dissolution of the lipophilic antibacterial agents. The profiles show also that addition of ammonia during preparation of the complex powder can enhance the complexation efficiency that results in enhanced dissolution. However, the enhancement is not as significant as one would expect based on the solubility data shown in Figures 3–6. Furthermore, previous study had indicated that cyclodextrin complexation of triclosan enhanced significantly dissolution of triclosan from solid complex powder without any additives [39]. In the present study the tablets contained the complex, Avicel and glucose compressed into a tablet. In previous study the dissolution medium did not contain any solubilizer. In this present study the triclosan dissolution medium consisted of aqueous 1% (v/v) hydrochloric acid containing 1% (w/v) HPβCD, and the solubility of triclosan in this medium was determined to be 0.3 mg/ml or total 150 mg in the 500 ml medium. Each triclosan tablet contained 20 mg triclosan or only 13% of the amount of triclosan in the saturated medium. In previous study triclosan was fully dissolved within 3–5 min resulting in supersaturated triclosan solution. In this present study it took from 60 to 120 min for triclosan to be fully dissolved. The dissolution of TCC was even slower taking about 9 h to be fully dissolved from the tablets. The TCC dissolution medium consisted of 8% (w/v) HPβCD in water, and the solubility of TCC in the medium was determined to be 0.05 mg/ml or total 25 mg in the 500 ml medium. Each TCC tablet contained 17 mg TCC or 70% of saturated dissolution medium.

Conclusion

The antibacterial agents triclosan and TCC are lipophilic compounds with a log $P_{(\text{octanol/water})}$ value of about 5. The melting point of triclosan is about 55 °C and that of TCC is about 255 °C, which could explain why triclosan is about 20 times more soluble in water than TCC (i.e. its intrinsic solubility (S_0) is about 20 times greater). Both compounds form complexes with βCD and the water-soluble βCD derivatives. However, since triclosan has much greater S_0 its solubility in aqueous HPβCD solutions is in the mg/ml range while that of TCC is in the μg/ml range. Ionization of triclosan (by including ammonia in the aqueous complexation media), and consequent increase of its apparent S_0, enhanced the complexation efficiency that resulted in enhanced rate of dissolution from tablets containing lyophilized triclosan/βCD complex. Since the pK_a of TCC is comparable to that of βCD ionization of TCC (by addition of ammonia) did not result in as significant enhancement in complexation efficiency and rate of dissolution.

Acknowledgement

This study was supported by a grant from the University of Iceland Research Fund.

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