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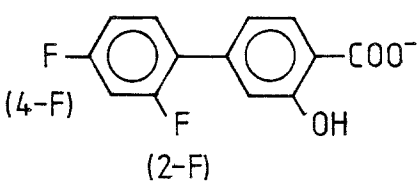
THE INCLUSION OF THE DRUG DIFLUNISAL BY ALPHA- AND BETA- CYCLODEXTRINS. A NUCLEAR MAGNETIC RESONANCE AND ULTRAVIOLET SPECTROSCOPIC STUDY.

Stephen F. Lincoln, John H. Coates, Bruce G. Doddridge and Andrea M. Hounslow
Department of Physical and Inorganic Chemistry,
University of Adelaide,
Adelaide, South Australia, 5001.

ABSTRACT. ^{19}F nmr and ultraviolet spectroscopic studies show that the inclusion of the anion of the drug diflunisal (DF) by alpha- and beta-cyclodextrins (αCD and βCD) in water produces the complexes: DF.αCD, DF.βCD and DF.$(\beta CD)_2$ characterized by stability constants of 17, 1.81×10^5 and 3.07×10^3 dm^3mol^{-1} respectively.

INTRODUCTION

The cyclodextrins are α-1,4-linked cyclic oligomers of D-glycopyranose which form inclusion complexes with a wide range of substrates.[1] Amongst such substrates are drug molecules whose cyclodextrin inclusion complexes are potentially valuable in drug delivery systems. As part of our studies[2] in this area we have used ultraviolet and ^{19}F nmr spectroscopy to investigate the inclusion, by alpha- and beta-cyclodextrin (αCD and βCD) of the anionic form of the anti-inflammatory drug diflunisal[3] (DF):



RESULTS

The spectroscopic studies were carried out in 10% D_2O KH_2PO_4/Na_2HPO_4 buffer solution of ionic strength 0.1 at pH 7.00 at 298.2 K. The DF (1.713×10^{-5} mol dm^{-3}) ultraviolet spectrum exhibited little variation at its 250 nm absorbance maximum in the [αCD] range 0-0.124 mol dm^{-3} consistent with the interaction between DF and αCD being weak. In contrast, DF exhibited a substantial variation in its spectrum as [βCD] varied in the range $(1.312\text{-}337.5) \times 10^{-5}$ mol dm^{-3}

consistent with the formation of 1:1 and 1:2 complexes shown in eqns (1) and (2):

$$DF + \beta CD \underset{}{\overset{K_1}{\rightleftharpoons}} DF.\beta CD \qquad (1)$$

$$\beta CD + DF.\beta CD \underset{}{\overset{K_2}{\rightleftharpoons}} DF.(\beta CD)_2 \qquad (2)$$

and the K_1 and K_2 derived in the range 240-255 nm are given in Table 1.

In solutions containing either of the cyclodextrins, a ^{19}F broad band ^{1}H decoupled doublet resonance (J_{F-F} = 7.20 Hz) is observed for each of the 2-F and 4-F of DF consistent with exchange of DF between the free and included states being fast on the nmr timescale. The variation of the observed ^{19}F chemical shift (δ) of DF (4.81×10^{-3} mol dm^{-3}) with total [αCD] is shown in Figure 1, and is consistent with the formation of DF.αCD only in an equilibrium analogous to eqn (1). The K_1 derived from the simultaneous fit of δ for 2-F and 4-F to eqn (3), in which δ_0 and δ_1 are the ^{19}F chemical shift of DF and DF.αCD, is given in Table 1.

$$\delta = \frac{\delta_0[DF] + \delta_1[DF.\alpha CD]}{[DF] + [DF.\alpha CD]} \qquad (3)$$

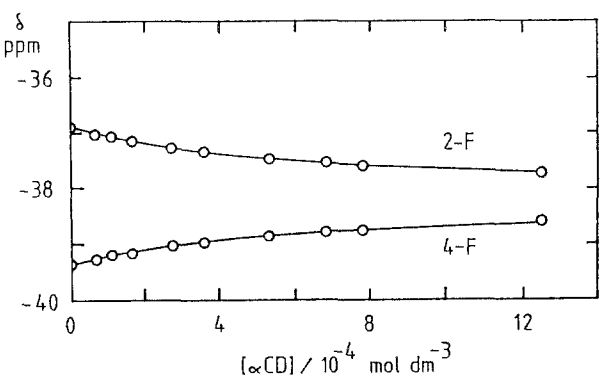


Figure 1. Variation of the ^{19}F chemical shift (δ) of DF (4.81×10^{-3} mol dm^{-3}) with total [αCD]. The negative shifts signify upfield shifts from a 2% sodium trifluoroacetate solution in D_2O external reference which is assigned a shift of zero. The solid curves represent the best fits of these data to eqn (3).

The variation of the observed ^{19}F chemical shift (δ) of DF (5.00×10^{-3} mol dm^{-3}) with total [βCD] is shown in Figure 2, and is consistent with the formation of DF.βCD and DF.$(\beta CD)_2$ as shown in eqns (1) and (2).

The variation of δ with [βCD] anticipated for equilibria (1) and (2) is given by eqn (4), in which δ_0, δ_1 and δ_2 are the ^{19}F chemical shifts of DF, DF.βCD and DF.$(\beta CD)_2$ respectively.

$$\delta = \frac{\delta_0[DF] + \delta_1[DF.\beta CD] + \delta_2[DF.(\beta CD)_2]}{[DF] + [DF.\beta CD] + [DF.(\beta CD)_2]} \quad (4)$$

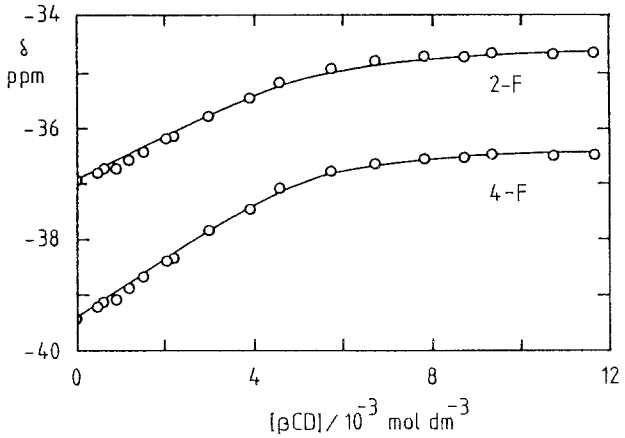


Figure 2. Variation of the ^{19}F chemical shift (δ) of DF (5.00×10^{-3} mol dm^{-3}) with total [βCD]. The negative shifts signify upfield shifts from a 2% sodium trifluoroacetate solution in D_2O external reference which is assigned a shift of zero. The solid curves represent the variation of δ predicted by eqn (4) using the K_1 and K_2 values determined from the ultraviolet spectrophotometric data.

A simultaneous non-linear least squares fit of the 2-F and 4-F data to eqn (4) yields: $K_1 = (2.17 \pm 9.16) \times 10^5$ dm^3mol^{-1} and $K_2 = (5.1 \pm 20.0) \times 10^3$ dm^3mol^{-1}, in which the large errors are a consequence of [DF] and [βCD] being very small compared to [DF.βCD] and [DF.$(\beta CD)_2$] at the total [DF] = 5.00×10^{-3} mol dm^{-3}. These K_1 and K_2 are in qualitative agreement with the more accurate values (Table 1) derived at much lower total [DF] using ultraviolet spectrophotometric methods. When K_1 and K_2 are set equal to the values derived from the ultraviolet spectrophotometric data, and the ^{19}F chemical shift data are again fitted to eqn (4), the best fit curves are seen to reproduce closely the experimental data (Figure 2). The corresponding δ_1 and δ_2 values are given in Table 1.

Table 1. Equilibrium constants and ^{19}F chemical shifts[a] for the diflunisal anion/cyclodextrin systems (298.2 K)

$K_1/10^3$ /dm^{3}mol^{-1}	$K_2/10^3$ /dm^{3}mol^{-1}		δ_0 ppm	δ_1 ppm	δ_2 ppm
		α-cyclodextrin			
0.0170±0.0009[b]	-	(2-F)	-36.89±0.01	-38.18±0.03	-
		(4-F)	-39.37±0.01	-38.27±0.03	-
		β-cyclodextrin			
181±20[c]	3.07±0.25[c]	(2-F)	-36.92±0.01	-34.91±0.05	-34.52±0.05
		(4-F)	-39.40±0.01	-36.71±0.05	-36.34±0.05

[a] A negative shift signifies an upfield shift from a 2% CF_3COONa solution in D_2O external reference which is assigned a shift of zero. The δ_0 values vary slightly with diflunisal concentration, and hence different values appear in the table for the αCD and βCD systems. The digital resolution was 0.007 ppm.

[b] Determined from ^{19}F shift data.

[c] Determined from ultraviolet spectrophotometric data.

DISCUSSION

The higher stability of DF.βCD by comparison with that of DF.αCD (Table 1) is consistent with DF fitting the βCD annulus (diameter 7-8 Å) better than the αCD annulus (diameter 5-6 Å). This probably also explains the high stability of DF.$(\beta CD)_2$, which contrasts with the absence of DF.$(\alpha CD)_2$ at detectable concentrations. The differing variations of the DF ^{19}F chemical shifts characterizing DF.αCD and DF.βCD (Figures 1 and 2, and Table 1) indicate differing structural features in these inclusion complexes. It has been suggested that downfield ^{19}F shifts indicate transfer to a hydrophobic environment.[4] On this basis both 2-F and 4-F of DF encounter a hydrophobic environment in DF.βCD, whereas only 4-F experiences a hydrophobic environment in DF.αCD. Space-filling models indicate that if the fluorinated end of DF enters the cyclodextrin annulus first, both 2-F and 4-F in DF.βCD can interact with the hydrophobic regions of the annulus, as can 4-F in DF.αCD. However the steric hindrance of the smaller annulus of DF.αCD leaves 2-F in the hydrophilic region at the annulus entrance. Similar changes in environment may be invoked to explain the variation in ^{19}F chemical shifts accompanying the formation of DF.$(\beta CD)_2$.

This study demonstrates that annular size is a major determinant of the stoichiometry and stability of cyclodextrin inclusion complexes; and the inclusion of other drugs is now being studied.

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