UNDERSTANDING pH-DEPENDENT INCLUSION COMPLEXATION OF CINNARIZINE WITH β-CYCLODEXTRIN *via* NMR SPECTROSCOPY

Franjo Sakoman[1,*], Ana Čikoš[1], Nives Galić[2]

[1] NMR Centre, Ruđer Bošković Institute, Bijenička cesta 54, 10000 Zagreb, Croatia
[2] Department of Chemistry, Faculty of Science, University of Zagreb, Horvatovac 102a, Zagreb, Croatia
* fsakoman@irb.hr

Introduction

Cinnarizine (CIN) is a piperazine-based antihistamine used for vestibular disorders and motion sickness. As a Biopharmaceutical Classification System (BCS) Class II drug, it shows high permeability but low aqueous solubility and pH-dependent dissolution, complicating formulation.[1,2]
Cyclodextrins (CDs) are cyclic oligosaccharides with a hydrophobic cavity and hydrophilic exterior that can encapsulate hydrophobic drugs to improve solubility. β-Cyclodextrin (β-CD) is the most widely studied due to its suitable cavity size and low cost.[3]

In our previous work[4], we confirmed CIN:β-CD complex formation through chemical shift perturbations, examined CIN orientation within the central β-CD cavity using ROESY, and performed an acid titration to determine the pH conditions that optimize solubility without reducing stability.

Here we extend the study by determining the percentage of complexation using DOSY NMR and investigate how varied pH influences the stability and stoichiometry of the inclusion complex.

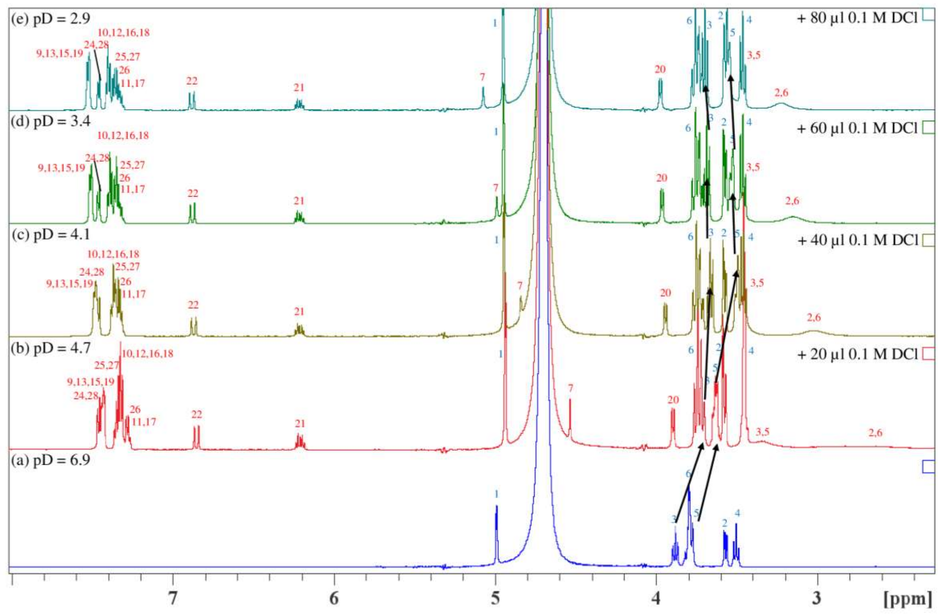


Fig.4. CIN: β-CD complex titration with DCl

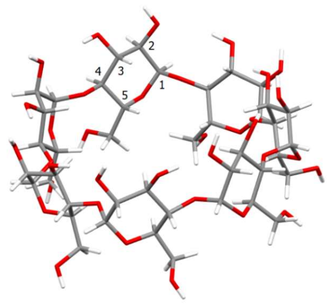


Fig.1. β-cyclodextrin (CSD: BCDEXD)

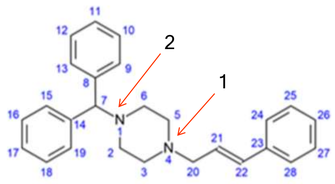


Fig.2. Cinnarizine with points of protonation marked with arrows

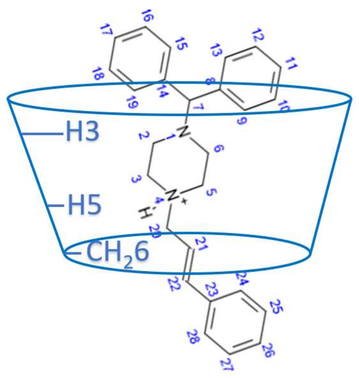
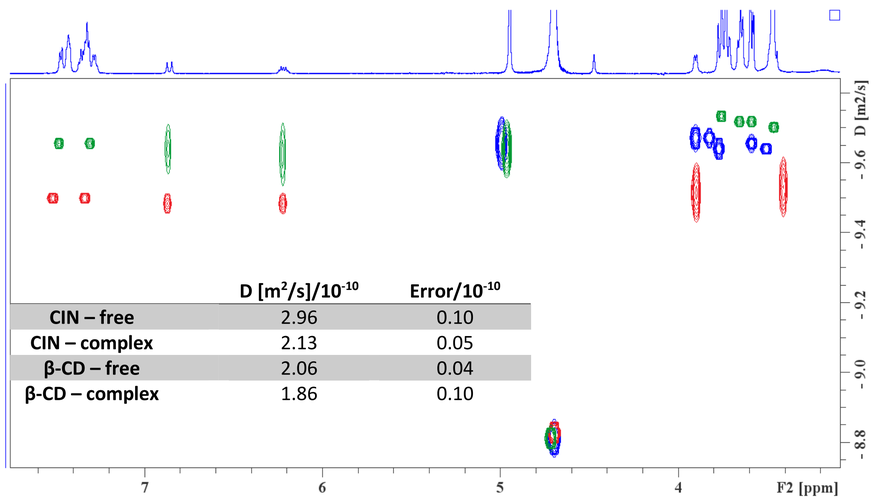


Fig. 3. Shematic inclusion of CIN in β-CD

Diffusion-Ordered Spectroscopy, DOSY



	D [m²/s]/10⁻¹⁰	Error/10⁻¹⁰
CIN – free	2.96	0.10
CIN – complex	2.13	0.05
β-CD – free	2.06	0.04
β-CD – complex	1.86	0.10

Figure 5. Overlay of the DOSY spectra: CIN (red), β-CD (blue), and CIN:β-CD complex (green)

DOSY measurements were used to evaluate the diffusion behaviour of free and complexed species and to estimate the percentage of complexation.

For β-CD, the diffusion coefficient remained nearly unchanged, while CIN showed a reduced value, consistent with fast exchange between free and bound states.

From these results, the percentage of complexation was estimated at ~91%.

Titrations of CIN with β-CD, HP-β-CD and SBE-β-CD at different acidic conditions

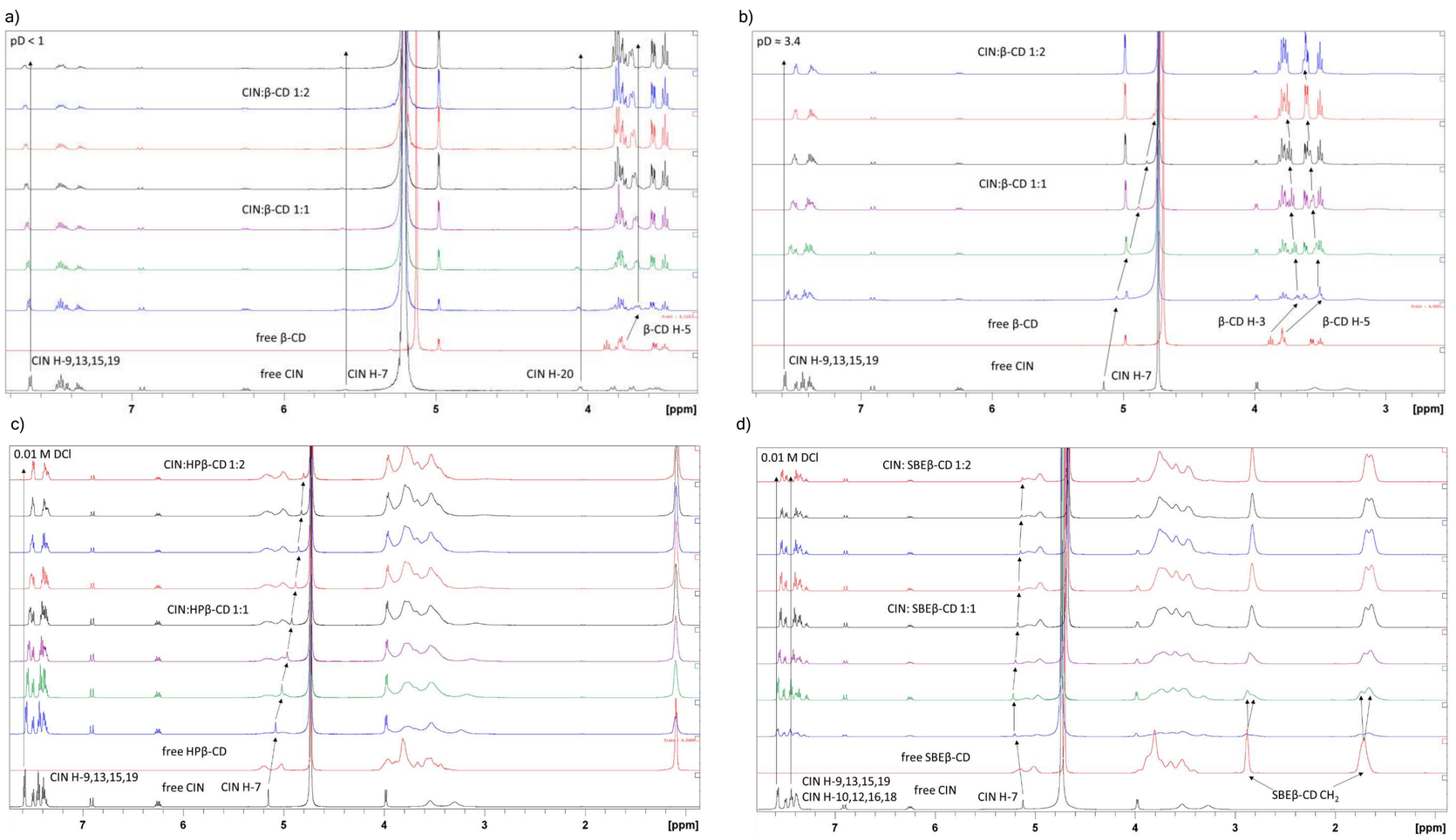


Fig. 6. Titrations of CIN with a) β-CD at pD<1, b) β-CD at pD 3.4, c) HP HPβ-CD at pD 3.4 and d) SBEβ-CD at pD 3.4

Conclusions

- DOSY experiments confirmed ~91% complexation of CIN with β-CD
- Titration with β-CD, HP-β-CD and SBE-β-CD at strongly acidic (pD < 1) conditions, where CIN is predominantly bi-protonated:
  - downfield shifts of CIN protons, with no strong evidence of inclusion, reflecting the low affinity of the fully bi-protonated form of CIN for the hydrophobic CD cavity.
- Titration with β-CD, HP-β-CD and SBE-β-CD at mildly acidic conditions (pD ≈ 3.4), where CIN exists as a mixture of mono- and bi-protonated forms:
  - The β-CD and HP-β-CD data supports the involvement of both the piperazine and diphenylmethyl moieties of CIN in the inclusion process, with encapsulation occurring through the wider rim of β-CD.
  - increasing the ratio to 1:2 did not shift β-CD H-3 and H-5 back toward the chemical shifts of free β-CD, suggesting the possibility of partial 1:2 complex formation.[5]
  - Titration of β-CD and HP-β-CD show similar CIN ^{1}H movements, while CIN titrated with SBE-β-CD behaves differently indicating slightly different mode of action

References

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Experimental

For DOSY, the samples were prepared in D_2O with 0.1 M DCl. For the β-CD titrations, the samples were prepared in 0.01 M and 1 M DCl. NMR spectra were recorded with standard pulse sequences on the Bruker Avance AV600 equipped with RT 5 mm diameter BBO probe with z-gradient accessory. Measurement of pH/pD was performed using the Mettler Toledo MP220 pH meter.

Acknowledgements

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