Ampelopsin (AMP) Solubilization by Inclusion Complexes

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INTRODUCTION

Ampelopsin (AMP) or better known under the name of Dihydromyricetin is a flavonoid extract with many pharmacological properties such as; anti-inflammatory, antimicrobial, antioxidant, hepatoprotective and anti-carcinogenic. Its development into a formulation for oral administration is limited due to its low water solubility and bioavailability beside light sensitivity. The objective of this project was to evaluate the ability of native and modified β-cyclodextrins to enhance AMP solubility and stability.

OBJECTIVES:

- To evaluate the phase solubility curve profile, stability constant ($K_{1:1}$) and the complexation efficiency (CE) of AMP in native β -Cyclodextrin (β-CD, KLEPTOSE®) and Hydroxypropyl β -Cyclodextrin (HP-β-CD, KLEPTOSE®HPB).
- To evaluate the complex formation and stability of complexes in liquid formulations.

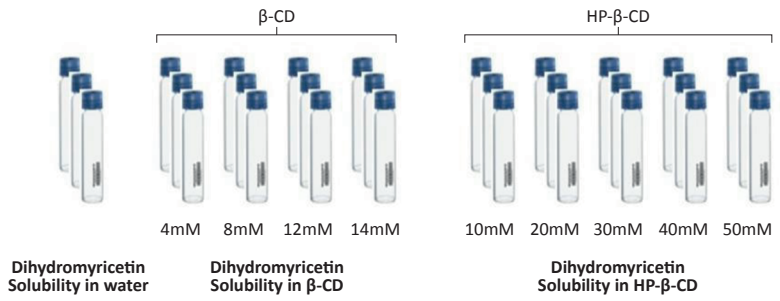
MATERIALS & METHODS

- The phase solubility profile , $K_{1:1}$ and CE of AMP were evaluated by adding excess amount of the API to different concentrations of β-CD and HP-β-CD (**Table 1**) in deionized water (DI). Samples were evaluated at day 1, 3 (data not shown) and 7 for saturation solubility in order to determine the necessary mixing time at 25°C. At equilibrium, samples were filtered using Millipore (0.45µm) syringe filter (**Fig.1**). The filtrates were analyzed using HPLC method for dihydromyricetin after appropriate dilution.
- Stability studies were also carried out for a period of 60 days at 25°C and 40°C.

Table 1. CDs relevant information.

Cyclodextrin	Molecular Weight	Concentration (mM)
β-CD	1135	4,8,12,14
HP-β-CD	1400	10,20,30,40,50

Figure 1. Phase solubility experiment design.



RESULTS & DISCUSSION

Stability Constants ($K_{1:1}$) and Complexation Efficiency (CE)

$$K_{1:1} = \frac{m}{S_0(1-m)} \qquad CE = \frac{m}{(1-m)}$$

- Where m is the slope of the experimental phase solubility curve of the AMP in β-CD and HP-β-CD at different concentrations determined by linear regression and S_0 is the drug solubility in DI water as determined after 7 days of mixing.

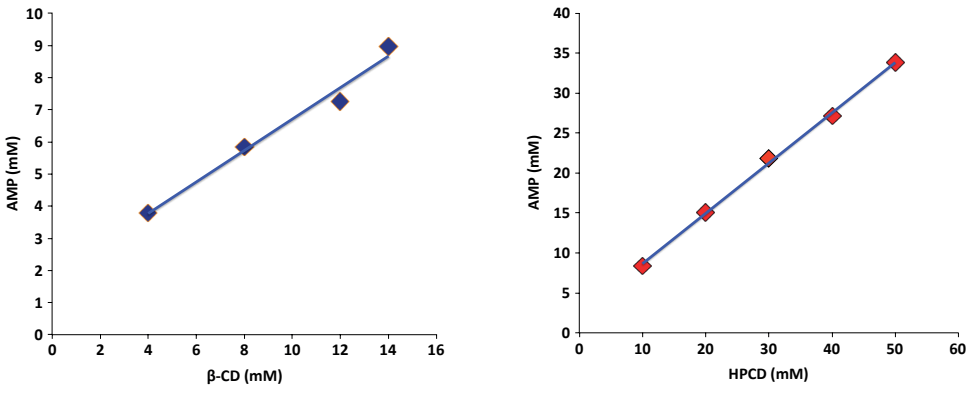


Figure 2. Phase-solubility profiles of AMP in β-CD and HP-β-CD.

- Both the CDs in the phase solubility diagram (**Fig. 2**) are displaying a linear solubility increase as a function of molarity increase indicating an AL type complexation. The affinity (stability) constants ($K_{1:1}$) and complexation efficiencies (CE) of AMP in each CD were calculated based on the parameters of the phase solubility graphs. The values are shown in **Table 2**. Increase in the solubility of AMP in the presence of both the cyclodextrin was calculated and the values are shown in **Table 3**.

Table 2. AMP: cyclodextrin affinity constants and complexation efficiencies.

Summary of Cyclodextrin Stability Constants and Complexation Efficiency		
	β-CD	HP-β-CD
S_0 (mole/L)	0.0018	0.0018
m (Slope)	0.490	0.630
1-m	0.510	0.371
S_0 (1-m)	0.001	0.001
$m/S_0(1-m)$	533.34	943.92
$K_{1:1}$	**533.34**	**943.92**
CE	**0.960**	**1.699**

Table 3. AMP solubility enhancement.

β-CD (mM)	Solubility enhancement ratio (β-CD)	HP-β-CD (mM)	Solubility enhancement ratio (HP-β-CD)
0	1	0	1
4	2.155	10	4.768
8	3.330	20	8.574
12	4.135	30	12.432
14	5.101	40	15.429
		50	19.252

Stability Studies

Figure 3. AMP: Cyclodextrin complex stability evaluation at 25°C and 40°C after 0, 60 and 180 Days.

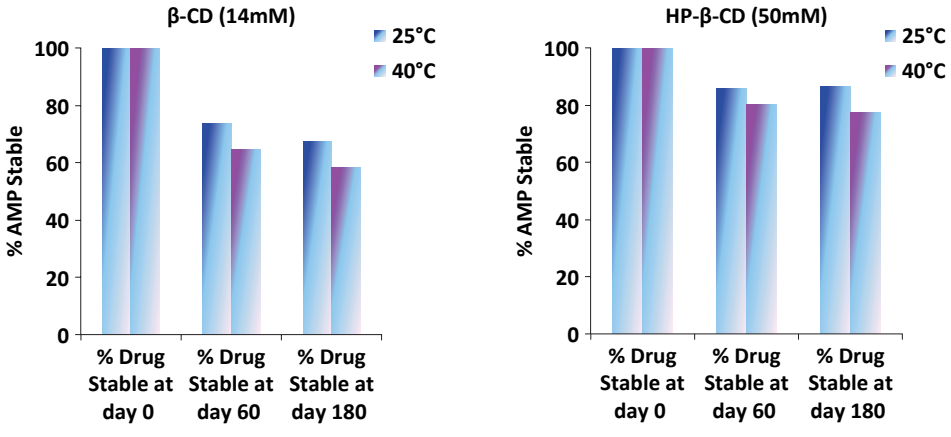


Figure 4. AMP: Cyclodextrin complex stability in water at 25°C and 40°C after 0, 60 and 180 Days.

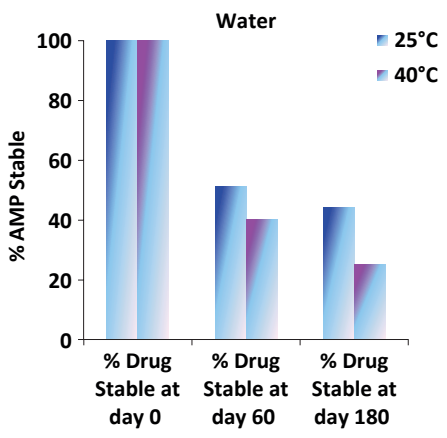


Table 4. Enhancement in the stability of AMP after complexation.

	Temp. (◦C)	% Enhancement in AMP stability after complexation after 180 days
β-CD (14mM)	25	23.03
	40	33.30
HP-β-CD (50mM)	25	42.32
	40	52.33

CONCLUSION

- An AL type phase solubility was observed with both cyclodextrins.
- A high complexation efficiency and stability constants were obtained for HP-β-CD.
- AMP solubility increased by ~5 % and ~19 % in the presence of β-CD and HP-β-CD respectively, compared to its solubility in water.
- The stability of the complexes formed in presence of HP-β-CD > β-CD following 60 days at 25°C and 40°C.
- Both β-CD and HP-β-CD would be ideal candidates for AMP solubilization, but HPBCD might be more preferred taking into account the fact that there are many existing drugs on the market formulated with it.

REFERENCES

P Manda, C Popescu, A Juluri, L Zhou, M A Repka and S N Murthy. Are Cyclodextrins a Viable Tool for Zotepine Solubilization? AAPS 2013.