

**PREPARATION AND  
CHARACTERIZATION OF SPRAY  
DRIED INCLUSION COMPLEX  
BETWEEN ANDROGRAPHOLIDE  
AND CYCLODEXTRINS**

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**Preparation and Characterization of Spray  
Dried Inclusion Complex between  
Andrographolide and Cyclodextrins**  
**噴霧乾燥環糊精包合穿心蓮內酯  
的製備和特性研究**

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## **ABSTRACT**

Andrographolide (Andro) is the major bioactive component in *Andrographis paniculata*. Recent studies showed that Andro possesses promising anticancer effect by inducing apoptosis of various malignant cells in vitro and has significant hepatoprotective and immunostimulative functions. However, being a class of diterpene lactone, Andro is hardly soluble in water. It is also unstable under neutral and alkaline condition and may subject to the multi-drug efflux by P-glycoprotein (Pgp) in the GI tract. To address the shortcomings of Andro, the compound was molecularly encapsulated by cyclodextrins as inclusion complexes. Effects of mixing ratio between Andro and cyclodextrins (CDs) were studied. Good complexation was found in the mixture of Andro- $\beta$ CD and Andro- $\gamma$ CD at a molar ratio of 1:2 while unreacted Andro was still detectable in the mixture of Andro-HP $\beta$ CD at any molar ratios. Spray drying of Andro- $\beta$ CD and Andro- $\gamma$ CD at an inlet temperature of 150°C and feed rate of 5 ml/min produced particle size of the complex down to 948 and 837 nm. The physicochemical properties of the spray dried binary systems of Andro-CDs and ternary system of Andro-CDs-polymers were characterized by X-ray diffractometry, Fourier transform-infrared spectrometry, differential scanning calorimetry, scanning electron microscopy, moisture content analysis and particle size analysis. Addition of polymeric additives, namely HPMC

and PVP, had no improvement on the performances of Andro-CDs. When the interaction of Andro and CDs complexes was evaluated by the profiles of phase solubility in aqueous environment, it revealed that Andro-HP $\beta$ CD and Andro- $\gamma$ CD were the A<sub>L</sub>-type interaction and Andro- $\beta$ CD was the B<sub>S</sub>-type interaction. The stoichiometry of the complex between Andro and CD was 1:1 in the aqueous solution. Since the stability constant ( $K_c$ ) of Andro- $\gamma$ CD complexes was the largest based on the phase solubility study, it suggests that the highest extent of complex formation was between Andro and  $\gamma$ CD. The dissolution performance and stability of each type of complexes were determined at different pHs. Feeding rate of spray dryer was found to play an important role against hydrolytic degradation of Andro. Lower feed rate could enhance the stability of Andro in the inclusion complex. It was found that Andro- $\beta$ CD at 1:2 molar ratio spray dried at inlet temperature of 150°C and feed rate of 5 ml/min gave the best performance of dissolution; it was about 100% better than pure drug and the stabilizing effect on degradation of Andro was the best in comparison to the other two CDs.

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## **ABBREVIATIONS**

Andro:	andrographolide
$\alpha$ CD	$\alpha$ -cyclodextrin
$\beta$ CD	$\beta$ -cyclodextrin
Bax:	Bcl-2 associated X-factor
Bcl-2:	B cell lymphoma gene 2
Bcl-XL:	Bcl-2 related gene x, large splice isoform
CD:	cyclodextrin
CTL:	cytotoxic T lymphocyte
d (v, 0.5):	The volume diameters ( $\mu$ m) undersize of 50% of particles
DE <sub>120</sub> :	Dissolution efficiency at 120 min
DM- $\beta$ -CD:	heptakis (2,6-di-O-methyl)- $\beta$ -cyclodextrin
DP <sub>15</sub> :	Percentage of drug dissolved at 15 min
DR4:	death receptor 4
DSC:	differential scanning calorimetry
EAE	experimental autoimmune encephalomyelitis
FTIR:	fourier transform infrared spectroscopy
$\gamma$ CD	$\gamma$ -cyclodextrin
GCL:	gamma-glutamate cysteine ligase
GCLC:	gamma-glutamate cysteine ligase (GCL) catalytic subunit
GSH:	glutathione
HPMC:	hydroxypropyl methylcellulose
HP $\beta$ CD:	2-Hydroxypropyl- $\beta$ -cyclodextrin
HP $\gamma$ CD:	2-Hydroxypropyl- $\gamma$ -cyclodextrin
IFN:	Interferon
IL-2:	Interleukin-2
$K_c$	Apparent stability constant
$k_{obs}$ :	The observed rate constant of degradation of Andro
Pgp	P-glycoprotein
PVP:	polyvinylpyrrolidone
RM $\beta$ CD:	Randomly methylated $\beta$ -cyclodextrin
SBE4- $\beta$ -CyD:	sulphobutyl ether- $\beta$ -cyclodextrin
SEM:	scanning electron microscopy
VEFG:	vascular endothelial growth factor
XRD:	X-ray diffractometry

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