

## High Speed Separation of Catechins by Extreme Liquid Chromatography ( $\lambda$ -LC<sup>®</sup>) and its Application to Analysis of Green Tea

### Introduction

The main components of catechins in green tea include epicatechin (EC), epigallocatechin (EGC, hydroxy derivative of EC), epicatechin gallate (ECg, gallic acid ester of EC), and epigallocatechin gallate (EGCg, gallic acid ester of EGC).

We examined the applicability of  $\lambda$ -LC for ultra-high speed analysis of 7 components consisting of the above 4 catechins,

catechin(C), catechin gallate(Cg), and Caffeine.

### Experimental

The system utilized in this experiment was a JASCO  $\lambda$ -LC system consisting of a 3185PU pump (2 sets), a 3080DG degasser, a 3180MX mixing unit, a 3067CO column oven, a 3070UV UV/Vis detector, a 3059AS autosampler, and ChromNAV chromatography data system.

A commercially-available tea beverage was filtered with 0.2  $\mu$ m membrane filter. A portion of the filtrate was injected.

### Results and Discussion

Figure 1 shows an  $\lambda$ -LC chromatogram of a standard mixture of 7 catechins. These components were clearly separated within 5 minutes by using a gradient elution method. Figure 2 shows an  $\lambda$ -LC chromatogram of the tea sample. Each component is well separated from unknown peaks.

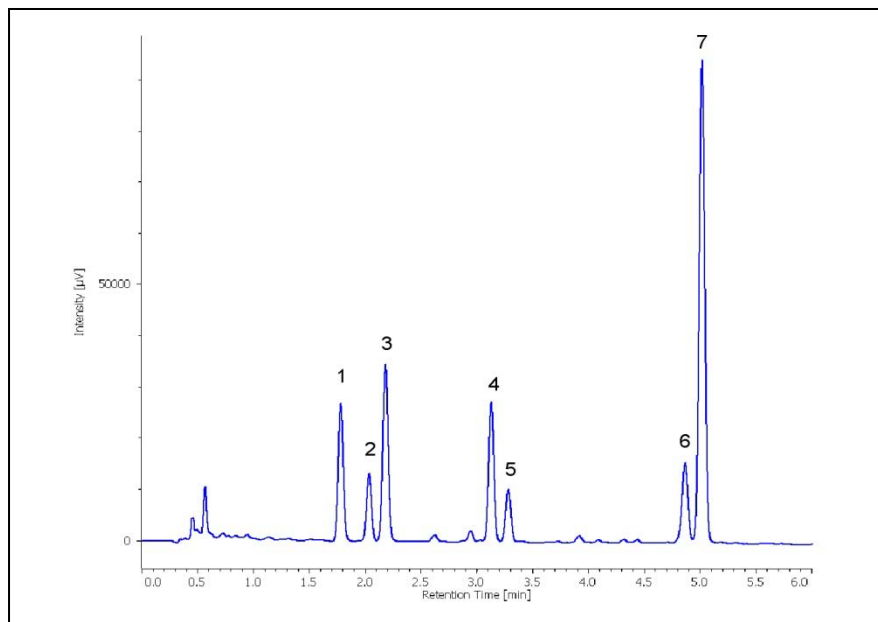


Figure 1  $\lambda$ -LC chromatogram of standard mixture

Peaks: 1=Epigallocatechin (500  $\mu$ g/mL), 2=Caffeine (10  $\mu$ g/mL), 3=Catechin (100  $\mu$ g/mL), 4=Epicatechin (100  $\mu$ g/mL), 5=Epigallocatechin gallate (25  $\mu$ g/mL), 6=Epicatechin gallate (20  $\mu$ g/mL), 7=Catechin gallate (100  $\mu$ g/mL)

Conditions: Column = X-PressPak AQ-C18W (2 mmID x 50 mmL, 2  $\mu$ m), Column temperature = 30 $^{\circ}$  C, Mobile phase: A = 0.2% phosphoric acid / acetonitrile (90/10), B = 0.2% phosphoric acid /acetonitrile (50/50), 0 min = A/B(100/0) — 0.5 min = A/B(100/0) — 4.0 min = A/B(70/30) — 5.0 min = A/B(70/30) — 5.05 min = A/B(100/0), Flow rate = 0.7 mL/min, Detection wavelength = 280 nm, Injection volume = 1  $\mu$ L

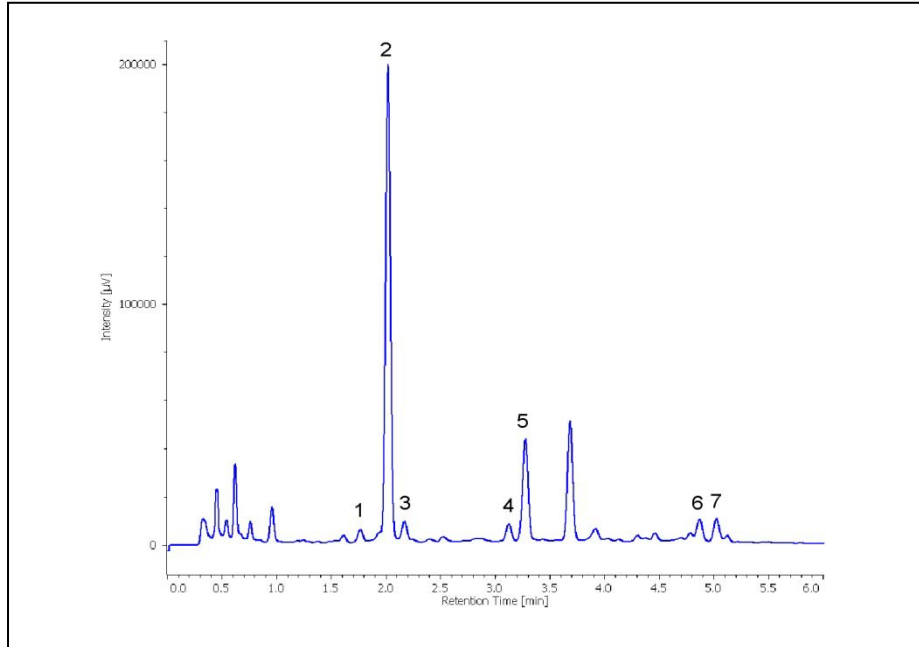


Figure 2 HPLC chromatogram of commercially-available tea beverage  
Conditions: the same as in the Figure 1