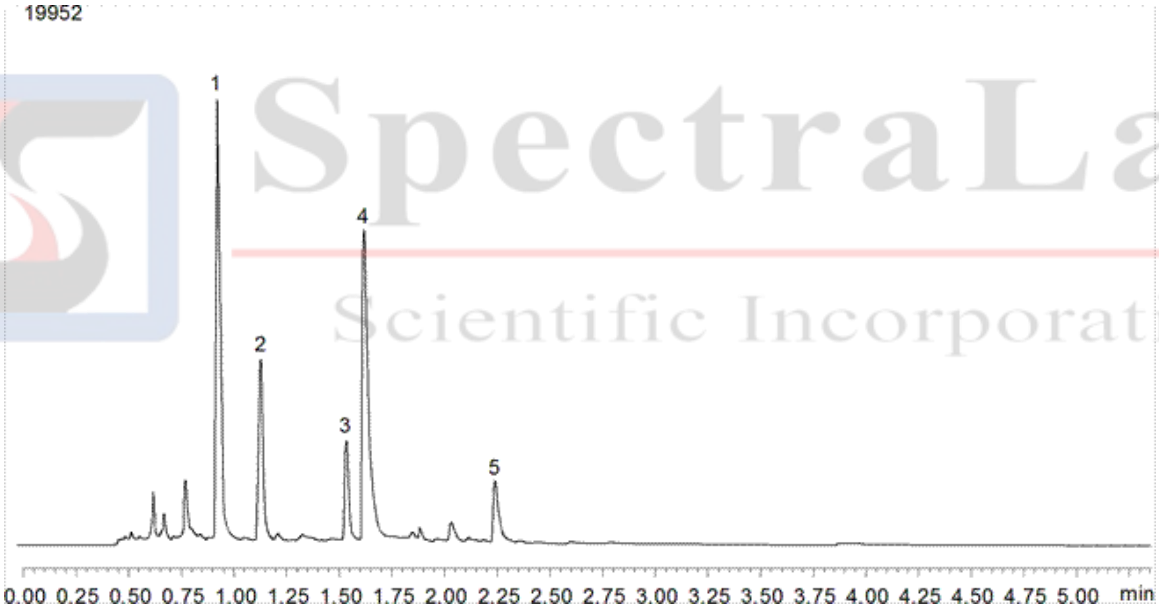
Analysis of Green Tea Extracts (Catechins) by HPLC-UV

Method 1:

Column: Kinetex™ 2.6 µm XB-C18 100 Å, LC Column 100 x 4.6 mm
Elution Type: Gradient
Elution A: 0.1% Phosphoric Acid in Water
Elution B: 0.1% Phosphoric Acid in Acetonitrile
Gradient Profile:

Step No.	Time (min)	Pct A	Pct B
1	0.01	85	15
2	0.45	85	15
3	2.68	65	35
4	3.34	10	90
5	3.35	85	15
6	5.34	85	15

Flow Rate: 1.8 mL/min
Col. Temp: Ambient
Detection: UV-Vis Abs. - Variable Wavelength (UV) @ 215 nm (22°C) (Ctrl + Click to follow link)



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1: Epigallocatechin
2: Catechin
3: Epicatechin
4: Epigallocatechin gallate
5: Epicatechin gallate

Method 2:

Column: Acclaim 120 C18, 2.2 µm (2.1 × 150 mm) or Waters Inertsil ODS-2 .5µm 4.6*150mm
Elution Type: Gradient
Elution A: 0.1% TFA, 5% acetonitrile
Elution B: 0.1% TFA in acetonitrile
Gradient Profile:

Step No.	Time (min)	Pct A	Pct B
1	0.0	100	0
2	1.2	100	0
3	15.5	71.5	28.5
4	17.0	71.5	28.5
5	17.0	100	0
6	25.0	100	0

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Flow Rate: 0.45 mL/min
Inj. Volume: 1.0 µL
Col. Temp: 25 ºC
Tray Temp.: 4 ºC
System Backpressure: ~6025–6200 psi during the gradient
Detection: **UV-Vis Abs. - Variable Wavelength (UV)** @ 280 nm **(Ctrl + Click to follow link)**

The retention times of gallic acid, GC, EGC, caffeine, catechin, EC, EGCG, GCG, and ECG are 2.15, 4.60, 7.00, 7.25, 7.40, 8.70, 8.90, 9.30, and 10.7 min, respectively. All catechins are well resolved and the total analysis time is less than 20 min.

Method 3:

Column: Symmetry C18, 5 µm (4.6 × 250 mm)
Elution Type: Isocratic
Elution Mobile Phase: 0.1% acetic acid and methanol (70:30 v/v)
Flow Rate: 1 mL/min
Inj. Volume: 10 µL
Col. Temp: Ambient
Detection: **UV-Vis Abs. - Variable Wavelength (UV)** @ 230 nm **(Ctrl + Click to follow link)**

The retention times of EGC, catechin, EGCG, caffeine, EC and ECG are 3.7, 4.1, 4.9, 5.6, 6.6, and 10.5 min., respectively. In addition catechin gallate (CG), and two other xanthines, theobromine and theophylline were 13.4, 3.1, and 4.4 min., respectively. All catechins are well resolved and the total analysis time is less than 15 min.

References: http://www.phenomenex.com/Application/Detail/19952?returnURL=/Application/Search
http://www.dionex.com/en-us/webdocs/110783-AN275-HPLC-Catechins-Tea-19May2011
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