Analysis of Catechins in Tea

Introduction

Catechins and epicatechins are polyphenolic phytochemicals that are found in high concentrations in foods such as grapes, apricots, red wine, and cocoa. Studies have found that increased catechin intake in humans has an overall positive effect on health and may help prevent and treat some chronic diseases. One of the most commonly consumed beverages around the world that is high in catechin content is green tea. Catechin content will vary depending on tea species, preparation, and other factors during its cultivation but can constitute as much as 25% dry mass of tea leaves.

Green tea is a variety of tea made from the leaves and buds of the *Camellia sinensis* plant. The preparation of green tea subjects the leaves to the least amount of oxidation; the oxidation of leaves is halted by a rapid application of heat to the leaves through either steam or dry roasting. In contrast, black and oolong teas have some of their moisture removed by leaving the leaves in the sun or in cool airy rooms in a process called withering. Green tea is extremely popular throughout China, Japan, and Korea.

Materials and Methods

Reagents

HPLC-grade methanol, HPLC grade acetonitrile, and HPLC-grade water were purchased from Sigma Aldrich. A Green Tea Catechin Mix (G-016-1ML) was purchased from Sigma Aldrich and used to create the calibration curve. Green tea leaves were sourced from local grocers.

Sample Preparation

A 1 g portion of green tea was weighed into an assembled Q-Cup containing a S1 Q-Disc® stack (C9+G1+C9 sandwich). The prepared Q-Cup was placed in an EDGE rack, along with 50 mL centrifuge vials. The rack was slid into position on the EDGE. The CEM-approved EDGE method for catechins was run. The extracts were diluted ten-fold and transferred into vials for analysis.

EDGE Method for Catechins

Q-Disc: S1 Q-Disc stack (C9+G1+C9 sandwich)

Cycle 1
Extraction Solvent: (80:20) Acetonitrile/Water
Top Add: 15 mL
Bottom Add: 0 mL
Rinse: 0 mL
Temperature: 80 ºC
Hold Time: 5:00 (mm:ss)

Wash 1
Wash Solvent: (80:20) Acetonitrile/Water
Wash Volume: 10 mL
Temperature: 80 ºC
Hold Time: 0:10 (mm:ss)

Analysis

HPLC Method

Table 1. HPLC Method

Parameter	Value
Mobile Phase	A: Water B: (3:1) Methanol/Acetonitrile
Column	Restek ARC-18 (5 µm, 150 x 4.6 mm) (PN 9314565)
Flow Rate	1.5 mL/min
Elution Conditions	0 to 10 minutes 10% B 10 to 17 minutes 10 to 30% B 17 to 19 minutes 30% B 19 to 19.5 minutes 30 to 10% B 19.5 to 20 minutes 10% B
Stop Time	20 minutes
Injection volume	10 µL
Needle wash	Methanol
Column temperature	40 °C
Detection	272 and 242 nm

Results

The standard was used to create calibration curves. **Figure 2** shows the calibration curves of the compounds; good linearities were obtained with a coefficient of determination (R^2) ≥ 0.99 for all compounds.
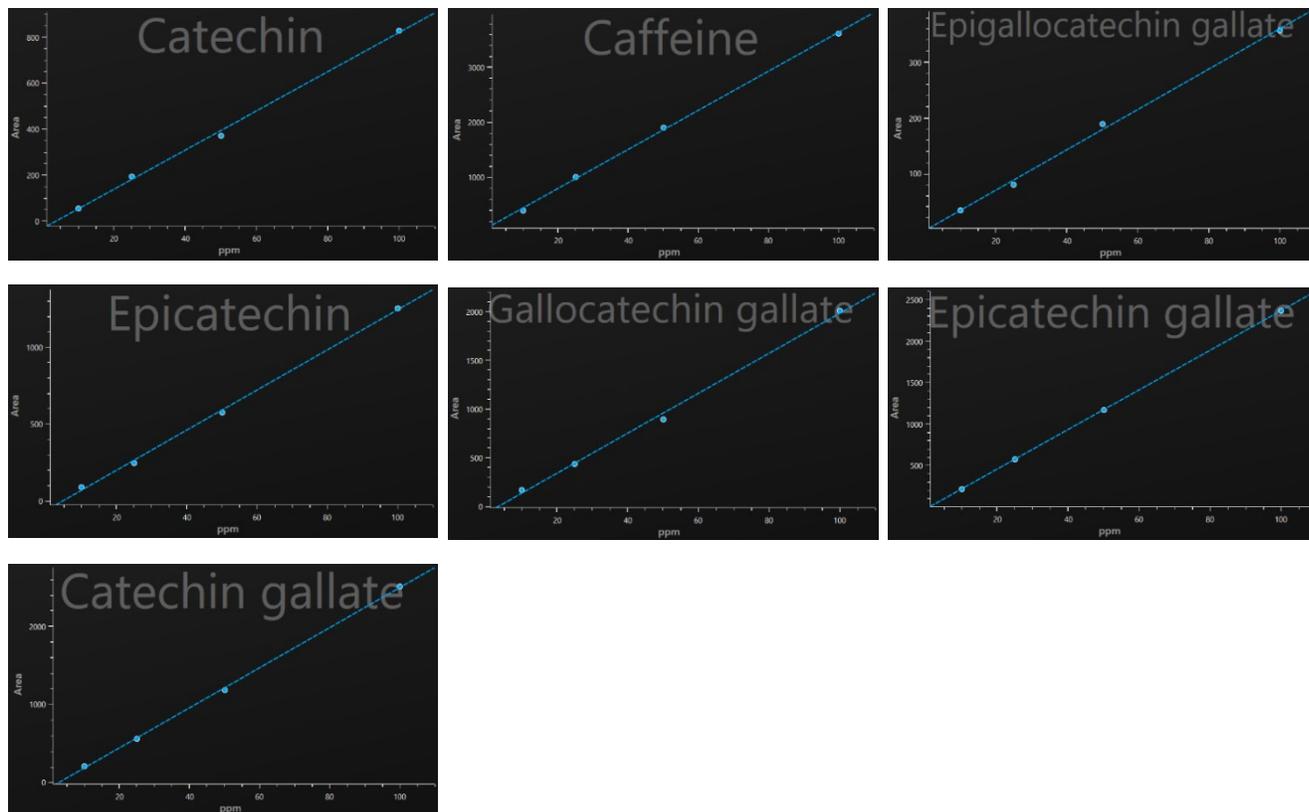

Figure 1. Calibration curves for the compounds found in the catechin standard.
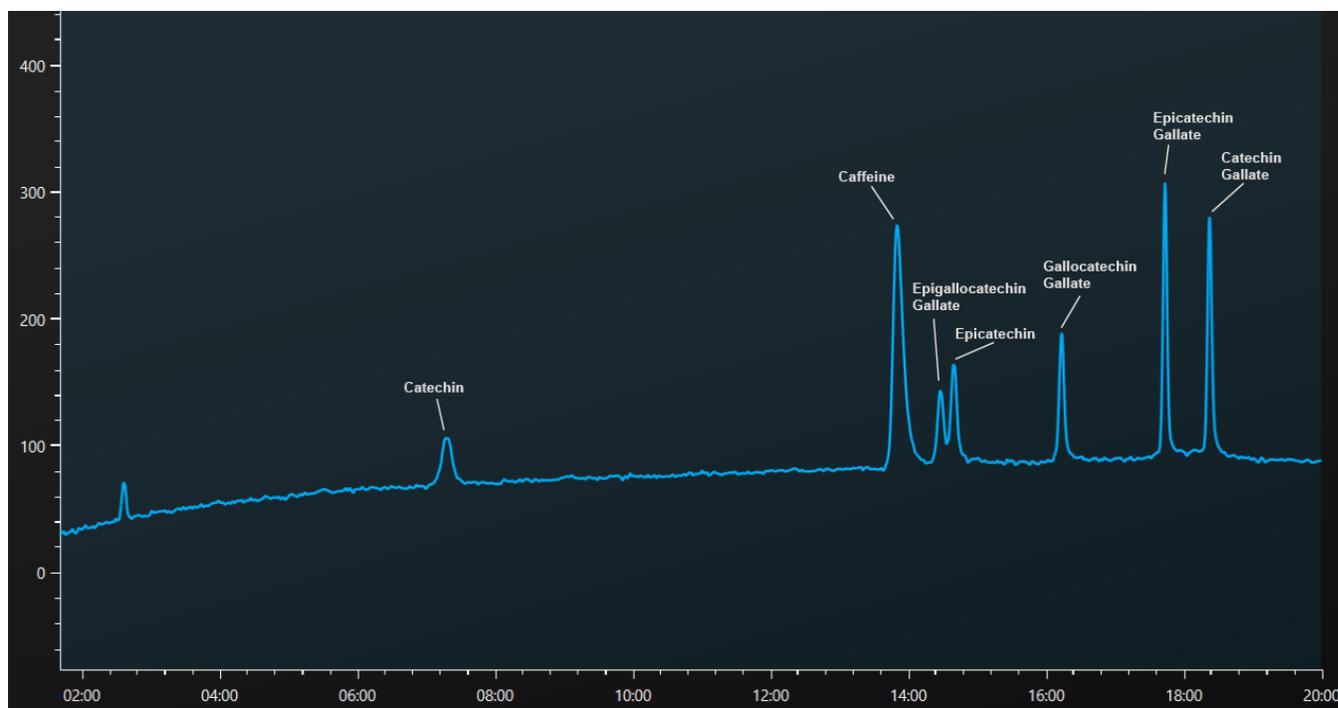
Figure 2. The chromatogram of catechin standard.

Table 2. Catechin concentrations in extracted tea (n=4)

Compound	Average concentration (µg/mL)	%RSD
Caffeine	83.77	2.54
Epicatechin	57.45	1.50
Epicatechin gallate	14.12	3.83

Conclusion

The Lucidity miniLC was able to generate calibration curves with good linearity for all compounds present in the standard. The CEM EDGE Automated Extraction system was able to extract green tea leaves, which was able to be analyzed on the miniLC.