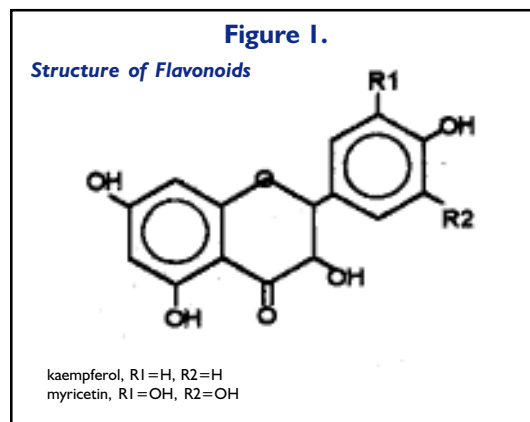


Analysis of Flavonoids in Green Tea

Flavonoids occur naturally in plant foods and are a common component of our diet. Flavonoids have demonstrated a wide range of biochemical and pharmacological effects, including antiinflammatory and antiallergic effects. For example, food-derived flavonoids such as myricetin and kaempferol (Figure 1) inhibited carcinogen-induced tumors in rats and mice¹. Antioxidant flavonoids such as quercetin also inhibited oxidation and cytotoxicity of low density lipoproteins in vitro², which may decrease their atherogenicity and subsequent risk of coronary heart disease.

Important dietary sources of flavonoids are vegetables, fruits, beverages, the latter accounts for a least 25-30% of the total daily flavonoids intake³. Kuhnau³ estimated that the intake of all flavonoids in the United States is approximately 1g/day. However, this estimation was based mainly on food analyses using techniques of doubtful accuracy. Evaluation of flavonoid content in food is not only useful in establishing accurate intake level of these compounds but also crucial for quality assurance. Flavonoids in green tea are of particular interest because of the recent reports on their cancer-protective effects⁴.

We report here a reversed-phase HPLC method for the determination of five major flavonoids and/or ingredients in green tea*: epigallocatechin, caffeine, epicatechin, epigallocatechin gallate, and epicatechin gallate. The flavonoids are prepared by pouring 100mL of boiling water onto 5g of loose green tea leaves. After 10 minutes, the infusion was passed through a sieve and allowed to cool prior to analysis. The influence of brewing time on flavonoids can also be studied by varying the time before the infusion is passed through the sieve. Figure 2 shows the chromatogram obtained on a typical sample. The ability to resolve these closely related organic compounds is very important for accurate quantitation.



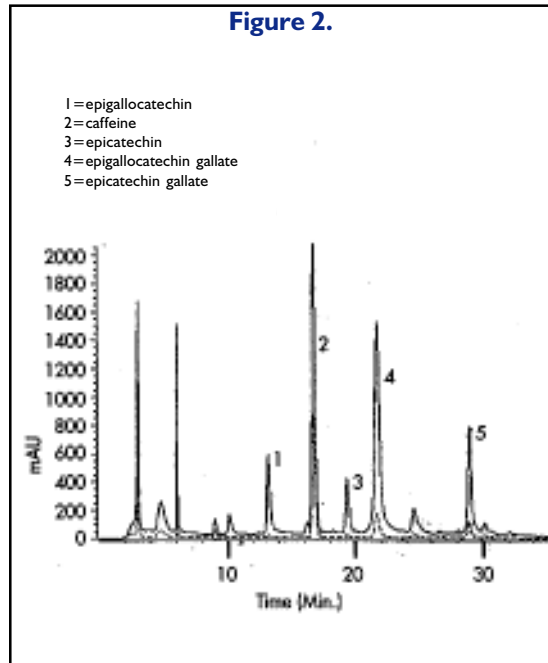
Chromatographic Conditions (Figure 2)

Column: SMT **0D-5-100 (250x4.6mm)**, **0D-5-100G**
Mobile Phase: 0.5 min, 12:88 ACN:Acetate buffer
18 min, 21:79 ACN:Acetate buffer
30 min, 61:39 ACN:Acetate buffer
Loop

Flow: 0.7mL/min
Detector: UV, 210 and 270nm

Column Specifications:

Particle: Spherical silica, 5 μ m
Pore Size: 100Å
Surface Area: 340 m²/g
% Carbon: 22%
pH range: 1-12



*SMT wishes to thank Dr. William Bronner of USDA, Beltsville, MD for the method development.

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2. De Whalley et. al. Biochem. Pharmacol. 1990, 39, 1743
3. Kuhnau, J. World Rev. Nutr. Diet. 1976, 24, 117
4. Conney et. al. Prev. Med. 1992, 21, 361



**SEPARATION
METHODS
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