

Application News

High Performance Liquid Chromatography

No.L391

Application of Evaporative Light Scattering Detector (Part 8) Analysis of Terpenoids in Ginkgo Biloba

Ginkgo biloba extract contains flavonoids and terpenoids that have been reported to be effective for improving poor blood circulation in the brain as well as poor peripheral blood vessel circulation. This ginkgo biloba extract is used as a health dietary supplement

in Japan and the United States.

Here we present an example of analysis of terpenoids in ginkgo biloba extract using the ELSD-LT II evaporative light scattering detector.

■ Analysis of Standard Solution

Terpenoids that are known to be present in large quantities in ginkgo biloba include bilobalide, ginkgolide A, ginkgolide B and ginkgolide C (Fig. 1). Because these compounds have no chromophores, use of the evaporative light scattering detector together with reversed-phase gradient elution is an effective means of analysis. Fig. 2 shows a chromatogram obtained from analysis of a standard solution of these four terpenoids (200 mg/L each, methanol), and Table 1 shows the analytical conditions used. (Column washing with 80 % methanol is included in the analytical conditions.)

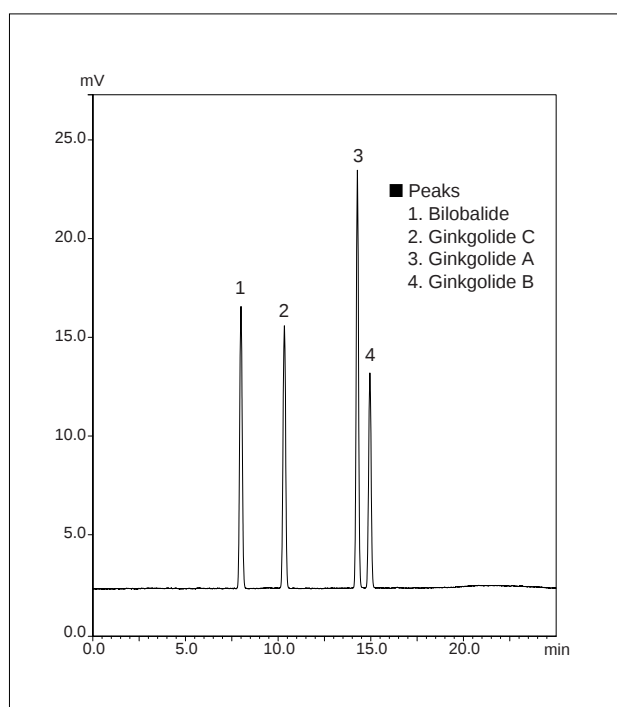


Fig. 2 Chromatogram of a Standard Mixture of 4 Terpenoids in Ginkgo Biloba (200 mg/L each, 10 μ L injected)

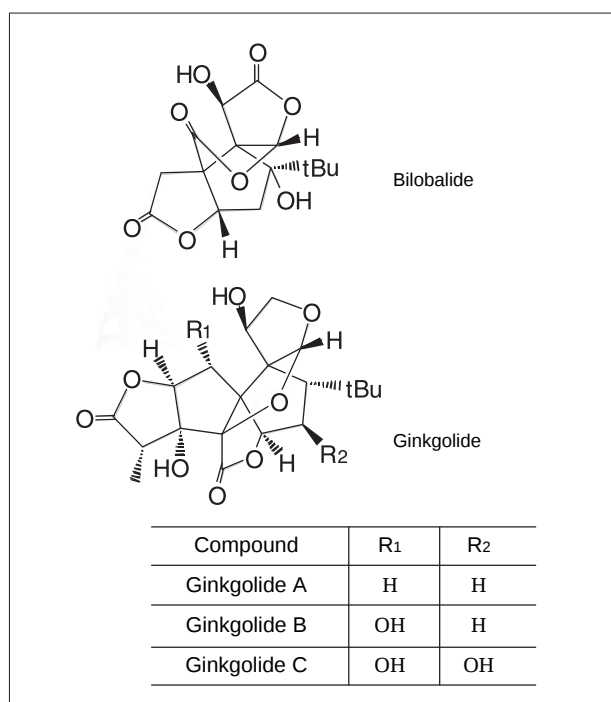


Fig. 1 Structures of 4 Terpenoids

Table 1 Analytical Conditions

Column	: Shim-pack FC-ODS (150 mm L. $\times$ 4.6 mm I.D.)
Mobile Phase	: A; Water B; Methanol B conc. 20 % (0 min) $\rightarrow$ 45 % (16 min) $\rightarrow$ 80 % (16.01-20 min) $\rightarrow$ 20 % (20.01-30 min)
Flow Rate	: 1.0 mL/min
Column Temp.	: 50 $^{\circ}$ C
Injection Volume	: 10 μ L
Detection	: ELSD-LT II Temperature : 40 $^{\circ}$ C Gain : 6 Nebulizer Gas : N ₂ Gas Pressure : 350 kPa

■ Calibration Curve and Repeatability

Fig. 3 shows the calibration curves generated using a concentration range of 20-1000 mg/L for each compound, and the peak area repeatability values (n = 60) based on a concentration of 200 mg/L. Because the response of the ELSD is exponential with

respect to the quantity of the substance injected, the calibration curves are plotted using logarithms of both the absorbance and the concentration, and are then used to conduct quantitation.

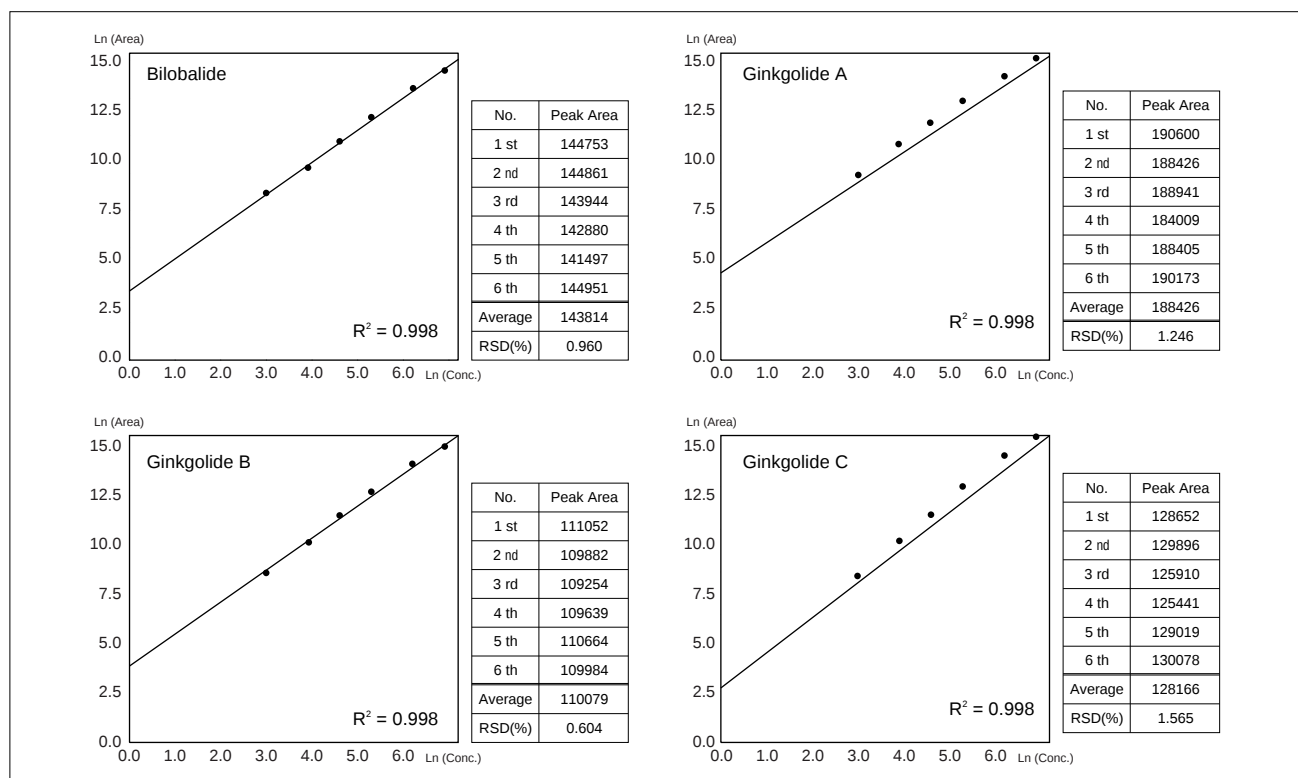


Fig. 3 Calibration Curves and Repeatability

■ Analysis of Dietary Ginkgo Biloba Supplement

Analysis was conducted after performing sample preparation of a commercially available dietary ginkgo biloba supplement according to the procedure shown in Fig. 4. Fig. 5 shows the chromatogram.

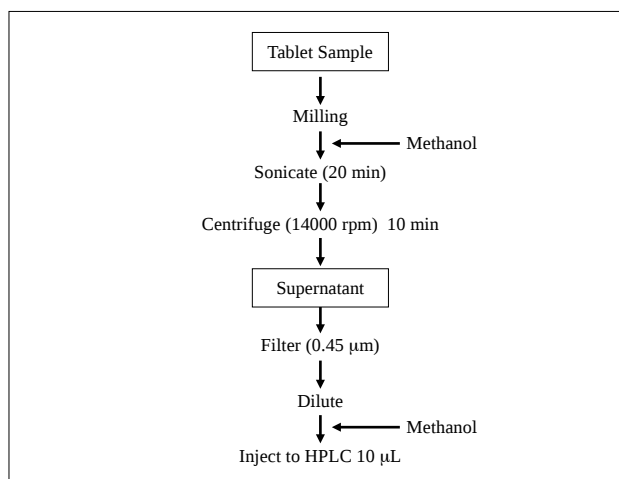


Fig. 4 Sample Preparation

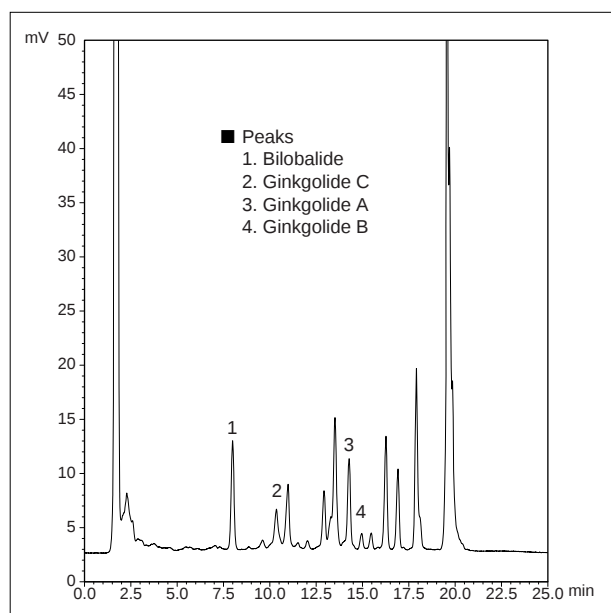


Fig. 5 Chromatogram of Dietary Ginkgo Biloba Supplement