

Status: Currently Official on 17-Feb-2025
 Official Date: Official as of 01-Dec-2015
 Document Type: Dietary Supplement Monographs
 DocId: GUID-CC522008-2DAC-41EE-BC4A-27EA929E8165_1_en-US
 DOI: https://doi.org/10.31003/USPNF_M34981_01_01
 DOI Ref: z4461

© 2025 USPC
 Do not distribute

Ginkgo

DEFINITION

Ginkgo consists of the dried leaf of *Ginkgo biloba* L. (Fam. Ginkgoaceae). It contains NLT 0.5% of flavonoids, calculated as flavonol glycosides, and NLT 0.1% of terpene lactones, calculated as the sum of bilobalide ($C_{15}H_{18}O_8$), ginkgolide A ($C_{20}H_{24}O_9$), ginkgolide B ($C_{20}H_{24}O_{10}$), and ginkgolide C ($C_{20}H_{24}O_{11}$), both on the dried basis.

IDENTIFICATION

• A. [HPTLC FOR ARTICLES OF BOTANICAL ORIGIN \(203\)](#).

Test for flavonoids

Standard solution: A solution of 0.6 mg/mL of [USP Rutin RS](#), 0.2 mg/mL of [USP Chlorogenic Acid RS](#), and 0.2 mg/mL of [USP Quercetin RS](#) in methanol

Sample solution: Transfer 1.0 g of finely powdered Ginkgo to a 50-mL round-bottom flask fitted with a reflux condenser. Add 10 mL of methanol, reflux on a water bath for 10 min, allow to cool to room temperature, and filter. [NOTE—Reserve some of the *Sample solution* for use in the *Test for terpene lactones*.]

Adsorbent: Chromatographic silica gel with an average particle size of 5 µm (HPTLC plates)

Application volume: 5 µL

Developing solvent system: Ethyl acetate, water, anhydrous formic acid, and glacial acetic acid (100:26:11:11)

Derivatization reagent A: 5 mg/mL of 2-aminoethyl diphenylborinate in methanol

Derivatization reagent B: 50 mg/mL of polyethylene glycol 400 in alcohol

Analysis

Samples: *Standard solution* and *Sample solution*

Before development of the chromatograms, saturate the chamber for 20 min with *Developing solvent system*. If the relative humidity in the laboratory exceeds 50%, condition the plate to about 35% relative humidity using a suitable device. Apply the samples separately as bands, and allow to dry. Develop the plate over a path of 6 cm, remove from the chromatographic chamber, and dry in a circulating air oven at 105° for 5 min. Immediately treat the hot plate with *Derivatization reagent A*, then with *Derivatization reagent B*, dry, and examine under long-wave UV light (365 nm).

Acceptance criteria: The *Standard solution* shows in its lower part with increasing R_F values a yellowish-brown fluorescent zone due to rutin (R_F 0.28), a light blue fluorescent zone due to chlorogenic acid (R_F 0.36), and a yellow fluorescent zone due to quercetin (R_F 0.92). The *Sample solution* shows a yellowish-brown fluorescent zone, a light blue fluorescent zone, and a yellowish-brown fluorescent zone at R_F similar to those of rutin, chlorogenic acid, and quercetin, respectively, in the *Standard solution*. Additional yellowish to yellowish-green zones due to flavonoids detected in the *Sample solution* chromatogram include one zone below the rutin zone, two zones between the rutin and chlorogenic acid zones, and two zones above the chlorogenic acid zone. Other zones may be seen in the *Sample solution* chromatogram.

Test for terpene lactones

Standard solution: Dissolve an amount of [USP Ginkgo Terpene Lactones RS](#) in methanol to obtain a solution containing in each mL about 1.0, 0.9, 0.6, 0.7, and 0.2 mg of bilobalide, ginkgolide A, ginkgolide B, ginkgolide C, and ginkgolide J, respectively.

Sample solution: Use the *Sample solution* prepared in the *Test for flavonoids*.

Adsorbent: Chromatographic silica gel with an average particle size of 5 µm (HPTLC plates)

Application volume: 5 µL

Developing solvent system: Toluene, ethyl acetate, acetone, and methanol (20:10:10:1.2)

Derivatization reagent: Acetic anhydride

Analysis

Samples: *Standard solution* and *Sample solution*

Immerse the plate for 2 s in an 8 g/200-mL solution of sodium acetate in methanol. Allow the excess liquid to drip from the plate, dry in a forced-air oven at 70° for 30 min, and cool in a desiccator. Apply the samples separately as bands to the impregnated plate, and allow to air-dry. Before development of the chromatograms, saturate the chamber for 20 min with *Developing solvent system*. If the relative humidity in the laboratory exceeds 50%, condition the plate to about 35% relative humidity, using a suitable device. Develop the plate over a path of 6 cm, remove from the chromatographic chamber, and dry in cold air. Treat the plate with *Derivatization reagent*, heat at 180° for 10 min, cool, and examine under short-wave UV light (254 nm).

Acceptance criteria: The *Standard solution* chromatogram shows five distinct quenching zones corresponding to the different ginkgo terpene lactones: ginkgolide C, ginkgolide J, ginkgolide B, ginkgolide A, and bilobalide at R_f values of about 0.13, 0.18, 0.32, 0.38, and 0.45, respectively. The *Sample solution* chromatogram shows a strong quenching zone at the application position, a broad quenching zone near the solvent front, and five distinct quenching zones corresponding to different ginkgo terpene lactones at R_f similar to those detected in the *Standard solution* chromatogram.

[NOTE— R_f values may differ from one plate to another due to the impregnation step.]

COMPOSITION

• CONTENT OF FLAVONOL GLYCOSIDES

Extraction solvent: Alcohol, hydrochloric acid, and water (25:4:10)

Mobile phase: Methanol, water, and phosphoric acid (100:100:1)

Standard solution A: 0.02 mg/mL of [USP Quercetin RS](#) in methanol

Standard solution B: 0.02 mg/mL of [USP Kaempferol RS](#) in methanol

Standard solution C: 0.005 mg/mL of [USP Isorhamnetin RS](#) in methanol

Sample solution: Transfer about 1.0 g of Ginkgo, finely powdered, to a 250-mL flask fitted with a reflux condenser. Add 78 mL of *Extraction solvent*, and reflux on a water bath for 135 min. [NOTE—The solution will turn deep red. The color of the solution is not a definitive indication of reaction completeness.] Allow to cool to room temperature. Decant into a 100-mL volumetric flask. Add 20 mL of methanol to the 250-mL flask, and sonicate for 30 min. Filter, transfer the filtrate into the 100-mL volumetric flask, wash the residue on the filter with a small amount of methanol, transfer the rinsate into the same 100-mL volumetric flask, dilute with methanol to volume, and mix.

Chromatographic system

(See [Chromatography \(621\), System Suitability](#).)

Mode: LC

Detector: UV 370 nm

Column: 4.6-mm × 25-cm; packing L1

Flow rate: 1.5 mL/min

Injection volume: 20 µL

System suitability

Samples: *Standard solution A*, *Standard solution B*, and *Standard solution C*

[NOTE—The relative retention times for quercetin, kaempferol, and isorhamnetin are about 1.0, 1.8, and 2.0, respectively, *Standard solution A*, *Standard solution B*, and *Standard solution C*.]

Suitability requirements

Relative standard deviation: NMT 2.0% determined from the quercetin peak in repeated injections, *Standard solution A*

Analysis

Samples: *Standard solution A*, *Standard solution B*, *Standard solution C*, and *Sample solution*

Calculate the percentage of each flavonol glycoside in the portion of Ginkgo taken:

$$\text{Result} = (r_U/r_S) \times (C_S/W) \times F \times 10$$

r_U = peak area of the relevant analyte from the *Sample solution*

r_S = peak area of the relevant analyte from *Standard solution A*, *Standard solution B*, or *Standard solution C*

C_S = concentration of the relevant analyte in *Standard solution A*, *Standard solution B*, or *Standard solution C* (mg/mL)

W = weight of Ginkgo taken to prepare the *Sample solution* (g)

F = factor to convert each flavonol aglycone into its respective flavonol glycoside: 2.504 for quercetin, 2.437 for isorhamnetin, and 2.588 for kaempferol

Calculate the total percentage of flavonol glycosides by adding the individual percentages calculated.

Acceptance criteria: NLT 0.5% of flavonoids, as flavonol glycosides, on the dried basis

• CONTENT OF TERPENE LACTONES

Solvent: Methanol and water (9:1)

Buffer solution: Dissolve 1.19 g of dibasic sodium phosphate and 8.25 g of monobasic potassium phosphate in 1000 mL of water, and adjust to a pH of 5.8.

Diluent: Methanol and water (1:1)

Solution A: Water

Solution B: Methanol

Mobile phase: See [Table 1](#).

Table 1