0055 - Kava for Kavalactones by HPLC

Botanical Name:  
*Piper methysticum* G. Forst.

Common Names:  
Kava kava, kava pepper

Parts of Plant Used:  
Roots

Uses:  
Treatment of sleep disorders, anxiety, stress, and restlessness.

Modes of Action:

About 20 clinical trials have proven kava to be effective for treating anxiety and sleep disorders and for improving mental function. The active constituents in kava root consist of a group of structurally related lipophilic lactone derivatives, kavalactones, with an arylethylene-α-pyrene skeleton. Kava extract and individual kavalactones were found to show direct activity on central nervous system receptors and on neurotransmitters. Kavalactones also were found to interact with γ-aminobutyric acid (GABA)–benzodiazepine receptors, to inhibit noradrenaline uptake, and to modulate the voltage-dependent Na⁺ and Ca²⁺ channels.

Chemical Markers:

Kava roots contain kavalactones as the main chemical components, with the major ones being kavain, 5,5-dihydrokavain, methysticin, dihydromethysticin, yangonin, and desmethoxyyangonin. Currently, 18 kavalactones have been isolated from kava. Other types of compounds identified in kava include alkaloids, chalcones (flavokawains A, B, C), flavanones (pinostrobin, 5,7-dimethoxyflavanone), cinnamic acid derivatives (bornyl ester of 3,4-methylene dioxyxinnamic acid, cinnamic acid bornyl ester), long-chain fatty acids and alcohols, and sterols. Kavalactones (six main ones) are used as marker compounds for quality control of kava extract in the U.S. market.
Methods of Analysis
Several methods have been used to determine the total kavalactones in kava including colorimetry, TLC, GC, and HPLC. Currently, the HPLC method is generally used for quality control of kavalactones. Following are the two best HPLC methods for total separation of kavalactones in a short time.

Method 1:
The method found at www.nsfina.org was used.

Sample Preparation:
For kava herb powders, accurately weigh about 750±0.1 mg into a 50-mL volumetric flask, and add 40 mL of methanol–water (70:30). Sonicate for 60 minutes at room temperature. Cool to room temperature and fill to volume with methanol–water (70:30).

For kava paste (50% to 60% kavalactones), weigh about 100 mg into a 50-mL volumetric flask and add 40 mL of methanol. Sonicate for 10 minutes or until all of the solids are dissolved. Cool to room temperature and fill to volume.

Chromatography:
Mobile phase: 0.1% phosphoric acid–isopropyl alcohol–acetonitrile (64:16:20 vol/vol/vol) isocratic.
Column: YMCbasic, 5 μm, 4.6 × 250 mm.
Column temperature: 40°C
Flow rate: 0.6 mL/minute for YMCbasic; 1.0 mL/minute for J’Sphere.
Detection: 220 nm (246 nm alternatively to improve selectivity).
Injection volume: 5 μL
Run time: 30 minutes

Validation Data:
Not available

Method 2:
The method of Schmidt and Molnár was used.

Column: Phenomenex Luna C18, 5 μm, 4.6 × 250 mm.
Mobile phase: Solvent A = water, solvent B = isopropanol.
Gradient: Linear 20%B to 30%B in 16 minutes.
Column temperature: 60°C
Flow rate: 1.4 mL/minute
Detection wavelength: 240 nm
References:


