

### 0045 - Feverfew for Parthenolide by HPLC

**Botanical Name:** *Tanacetum parthenium* (L.) Sch. Bip [syn. *Chrysanthemum parthenium* (L.) Bernh.]

**Common Names:** Bachelor's buttons, feverfew.

**Parts of Plant Used:** Flowering tops, aerial parts or leaves; usually collected when the plant is flowering.

**Uses:** Treatment of migraine headaches.

#### Modes of Action:

A few clinical trials support the antimigraine activity of feverfew, but the mechanism is still not very clear. Extracts of feverfew were found to inhibit in vitro platelet aggregation, serotonin release, and smooth muscle cell function, which may contribute to its antimigraine activity.<sup>1</sup>

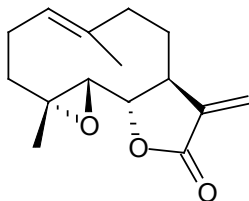
#### Chemical Markers:

Sesquiterpenoid lactones (parthenolide, epoxyartemomin, artemcanin, canin, santamarin) are believed to be the major chemical components in feverfew.<sup>2,3</sup>

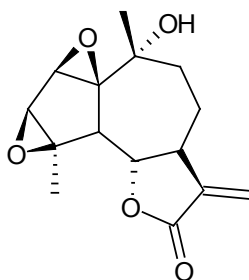
The occurrence of different sesquiterpene lactones was found to vary by the habitat of the plant. Flavonoids (6-hydroxykaempferol 3,6,4'-trimethyl ether, 6-hydroxykaempferol 3,6-dimethyl ether, quercetagenin 3,6-dimethyl ether, quercetagenin 3,6,4'-trimethyl ether and quercetagenin 3,6,3'-trimethyl ether, luteolin, apigenin, luteolin 7-glucuronides), melatonin, and tannins also were detected in feverfew.<sup>4-6</sup> The compounds identified in feverfew essential oil include camphor, chrysanthemyl acetate, camphene, and p-cymene.<sup>7</sup> For most varieties of feverfew, parthenolide is found to be the main sesquiterpene lactone, with the flowering tops and leaves containing the highest amounts (the stems and roots are not a good source for parthenolide). Therefore, parthenolide was selected as a chemical marker for quality control of feverfew. In Canada, it is recommended that feverfew powder must contain at least 0.2% parthenolide.



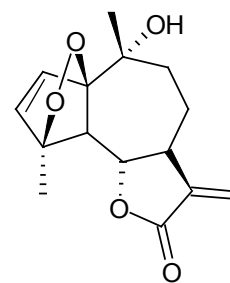
#### Representative Sesquiterpene Lactones Found in Feverfew.



Parthenolide



Artemcanin



Tanaparthin-beta-peroxide

## Methods of Analysis

Most published analytical methods for the analysis of parthenolide in feverfew use reversed-phase HPLC with isocratic elution and low-wavelength detection.<sup>1,8-10</sup> Since parthenolide is the only main chemical in feverfew, the analysis of feverfew is not very challenging and most of the published HPLC methods are good enough for parthenolide analysis. A GC method is an alternative method for parthenolide analysis.<sup>11</sup>

Extraction is key for accurate analysis of parthenolide in feverfew. Since parthenolide is a low polar compound, pure organic solvents such as methanol, ethanol, acetonitrile, and acetone are good extraction solvents. A small portion of water (10%, 20%, or 30%) in these solvents helps dissolve the hydrophilic components and helps the organic solvent catch parthenolide.<sup>10</sup> Based on the authors' experience, sonication of 250 mg of feverfew powder with 50 mL of methanol for 30 minutes is sufficient to extract parthenolide totally from feverfew.

### Method 1:

The method of Zhou et al. was used.<sup>10</sup>

#### *Sample Preparation:*

Various extraction solvents and extraction methods were tried; 90% acetonitrile aqueous solution was selected. The sample was prepared by bottle-stirring 1.5 g of feverfew with 100 mL of extraction solvent for 30 minutes.

#### *Chromatography:*

Column: Nacalai Tesque Cosmosil C18-AR column, 5  $\mu$ m, 150  $\times$  4.6 mm, 120 Å.

Mobile phase: Acetonitrile–water (55:45) isocratic.

Flow rate: 1.5 mL/minute

Injection volume: 5  $\mu$ L

Detection wavelength: 210 nm

#### *Validation Data:*

Linearity: 0.16 to 850 mcg/mL with a correlation coefficient of 0.9999.

Accuracy: In the spiked recovery tests, an average of 99.3% of 7.50 $\pm$ 0.30 mg of parthenolide standard spiked into a 0.75-g feverfew sample in 100 mL of extraction solvent was recovered with an RSD of 1.6%.

Precision: 0.88% RSD

Selectivity: Peak identification was determined against standards.

Ruggedness: Not specified

Robustness: The method was repeated on two different types of column with comparable results.

LOD/LOQ: LOD = 0.10 ng; LOQ = 0.16 ng.

### Method 2:

The method of Abourashed and Khan<sup>9</sup> was used.

#### *Chromatography:*

Column: LiChrospher 5 C18, 5  $\mu$ m, 250  $\times$  4.6 mm, with a SecurityGuard cartridge system (Phenomenex).

Mobile phase: Solvent A = 50 mM NaH<sub>2</sub>PO<sub>4</sub> in water; solvent B = acetonitrile–methanol (90:10).

Gradient: 50%B to 85%B in 20 minutes.

Flow rate: 1.0 mL/minute

Injection volume: 20  $\mu$ L

Detection wavelength: 210 nm

**Validation Data:**

Linearity: 0.00 to 0.4 mg/mL with a correlation coefficient of 0.9999.

Accuracy: 103.1% recovery for parthenolide.

Precision: The RSDs with all measurements are between 0.00 and 3.24% (n = 3).

Selectivity: Peak identification was determined against standards.

Ruggedness: Not specified

Robustness: Not specified

LOD/LOQ: Not specified

**Method 3:**

The unpublished method of Mingfu Wang was used.

**Sample Preparation:**

Extract a standardized extract (0.7% parthenolide) or herb powders (about 250 mg) with about 35 mL of methanol in a 50-mL volumetric flask by sonication for 30 minutes. After cooling to room temperature, fill to volume with methanol.

**Chromatography:**

Column: Phenomenex Luna C18(2) 5  $\mu$ m, 4.60  $\times$  250 mm.

Mobile phase: Acetonitrile–water (0.2% phosphoric acid by volume) (50:50) isocratic.

Flow rate: 1.0 mL/minute.

Column temperature: Ambient

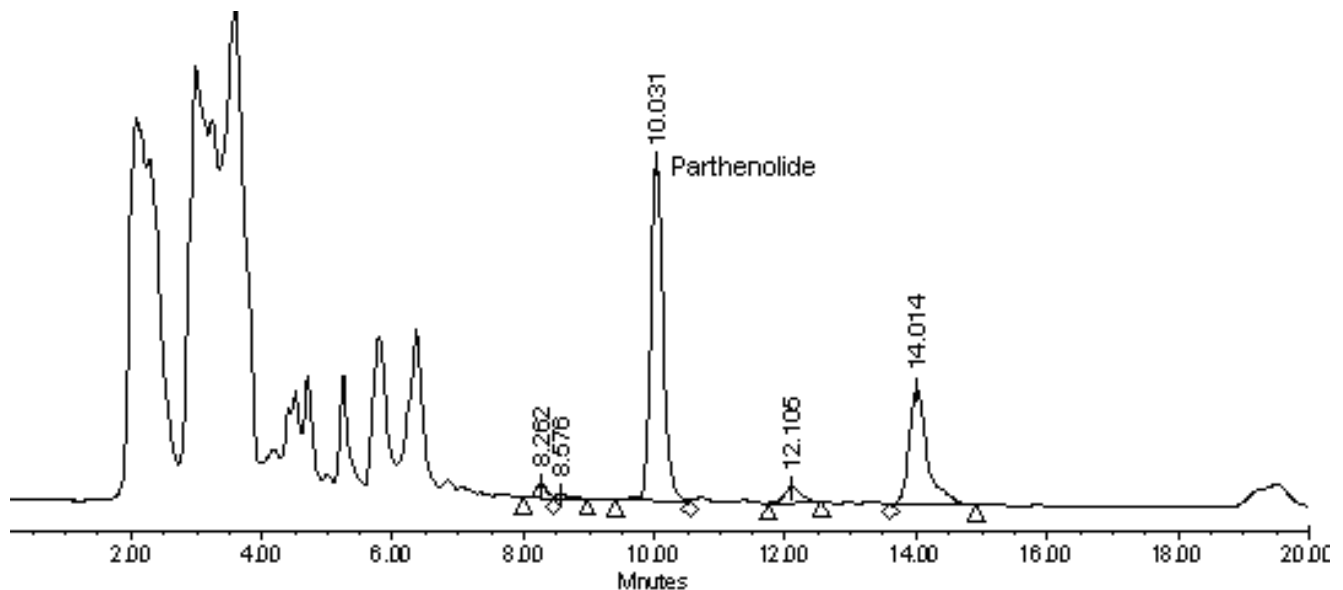
Injection volume: 20  $\mu$ L

Detection wavelength: 210 nm

**Validation Data:**

Not available.

### Representative HPLC Chromatogram of Feverfew Run by Method 3.



### References:

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