The United States Pharmacopeia (USP) has recently published new Dietary Supplement monographs for Guarana Seed, Guarana Seed Powder and Guarana Seed Dry Extract in the Pharmacopeial Forum (PF) 44(2)-March-April, 2018. USP seeks the input and comments from stakeholders for the tests, specifications and acceptance criteria proposed in these monographs.

### Identification

#### A. HPTLC for Articles of Botanical Origin (203)

**Standard solution A:** 1.0 mg/mL of USP Guarana Seed RS in methanol

**Standard solution B:** 0.5 mg/mL each of USP (+)-Epicatechin RS and USP Procyanidin B1 RS in methanol

**Sample solution:** 1 g of Guarana Seed (finely powdered), Guarana Seed Powder or Guarana Seed Dry Extract in 10 mL of methanol. Sonicate for 10 min, centrifuge, and use the supernatant.

**Chromatographic system**

- Adsorbent: Silica gel, 25 µm; and 60 µm as a support in a single chromatographic run.
- Development reagent: 5% of anisaldehyde in 10% acetic acid.
- Solvent: Watermethanol (9:1).

**Sample Application:** Silica gel, 25 µm, and 60 µm as a support in a single chromatographic run.

**Analysis:** Proceed as directed in the test for Content of Caffeine and Flavonoids. For each sample, apply theStandard solution A, Standard solution B, and Sample solution on the HPTLC plate, develop, and examine under shortwave ultraviolet light and register the chromatogram between 300 and 380 nm.

**Results:** Content of caffeine in the portion of Guarana Seed taken:

- For Guarana Seed Dry Extract, the content ratio of catechin to epicatechin is about 2:1. The content ratio of caffeine to total flavonoids is NMT 3%.

### Composition

#### CONTENT OF CAFFEINE AND FLAVONOIDS

**Solution A:** 1.36 g/L of monobasic potassium phosphate; adjusted with 3.0% phosphoric acid to a pH of 3.1

**Solution B:** Acetic acid and methanol (70:30)

**Mobile phase:** See Table 1.

<table>
<thead>
<tr>
<th>Time (min)</th>
<th>Solution A (%)</th>
<th>Solution B (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>15</td>
<td>90</td>
<td>10</td>
</tr>
<tr>
<td>20</td>
<td>95</td>
<td>5</td>
</tr>
<tr>
<td>30</td>
<td>95</td>
<td>5</td>
</tr>
<tr>
<td>40</td>
<td>70</td>
<td>30</td>
</tr>
<tr>
<td>50</td>
<td>60</td>
<td>40</td>
</tr>
<tr>
<td>60</td>
<td>60</td>
<td>40</td>
</tr>
</tbody>
</table>

**HPLC Chromatogram at 275 nm of Caffeine and Flavonoids in Guarana Seed Powder**

---

**Development of USP Guarana Seed Monograph Family**

Maria Monagas*; Padmanabharaju P.; Chris Okunji*; Satish J.*; Eike Reich*; Gabriel Giancaspro*

*United States Pharmacopeia Convention (USP). 12601 Twinbrook Parkway, Rockville, MD 20852. CAMAG. Sonnennmatstrasse 11, 4132 Muttenz, Switzerland

---

**USP**

usa.org