







E-mail: codex@fao.org - www.codexalimentarius.org

STANDARD FOR GINSENG PRODUCTS CODEX STAN 321-2015

Adopted in 2015.

This Standard supersedes the regional standard CODEX STAN 295R-2009.

## 1. SCOPE

This Standard applies to the ginseng products as defined in Section 2 below and offered for direct consumption, including for catering purposes or for repacking, if required. This Standard applies to ginseng products used as a food or food ingredient and does not apply to products used for medicinal purposes<sup>1</sup>.

## 2. DESCRIPTION

# 2.1 Product definition

Ginseng product is the product:

- (a) prepared from all part of fresh and sound ginseng roots, derived from *Panax ginseng* C.A.Meyer or *P. quinquefolius* L., cultivated for commercial purposes and used for foods;
- (b) packaged in such a manner as to safeguard the safety and nutritional and quality characteristics of the products;
- (c) processed in an appropriate manner, undergoing operations such as drying, steaming, cutting, powdering, extraction and concentration in conformity with Section 2.2.

## 2.2 Types of Ginseng Products

Ginseng products covered by this Standard may be as follows:

#### 2.2.1 Dried Ginseng

*Dried Ginseng* is manufactured when ginseng roots defined in Section 2.1 (a) are dried in an appropriate manner such as sun drying, hot air drying or other recognized drying methods. The product may be classified into one of such product types that have the main root and/or lateral roots or that are powdered or sliced.

## 2.2.2 Dried Steamed Ginseng

*Dried Steamed Ginseng* is manufactured when ginseng roots defined in Section 2.1 (a) are prepared using the steaming method and the drying method stated in Section 2.2.1. The product may be classified into one of such product types that have the main root and/or lateral roots or that are powdered or sliced.

## 2.2.3 Ginseng Extract

*Ginseng Extract* is manufactured when soluble components of ginseng roots defined in Section 2.1 (a) or *Dried Ginseng* defined in Section 2.2.1. are extracted by using water, ethanol or their mixture, filtered and concentrated. This product has a dark brown colour and a high viscosity. The product may be also presented as a powdered type through spray- or freeze-drying.

## 2.2.4 Steamed Ginseng Extract

Steamed Ginseng Extract is manufactured when soluble components of Dried Steamed Ginseng defined in Section 2.2.2 are extracted by using water, ethanol or their mixture, filtered and concentrated. This product has a dark brown colour and a high viscosity. The product may be also presented as a powdered type through spray- or freeze-drying.

#### 2.3 Styles

Styles should be permitted provided that the product meets all relevant requirements of the Standard and is adequately described on the label to avoid confusing or misleading the consumer.

# 3. ESSENTIAL COMPOSITION AND QUALITY FACTORS

# 3.1 Composition

# 3.1.1 Basic Ingredients

Ginseng roots as defined in Section 2.1 (a).

<sup>&</sup>lt;sup>1</sup> Some countries also consider ginseng as a medicine.

#### 3.2 Quality Criteria

## 3.2.1 Flavour, Colour, and Ginsenoside Pattern

Ginseng products shall have normal flavour, colour, taste and a ginsenoside pattern<sup>2</sup> unique to specific species of ginseng as well as be free from foreign matter.

# 3.2.2 Chemical and Physical Characteristics

- **3.2.2.1** Dried Ginseng and Dried Steamed Ginseng
  - (a) Moisture: no more than 14.0% (Powdered type: no more than 9.0%).
  - (b) Ash: no more than 6.0%.
  - (c) Water-saturated n-butanol extracts: no less than 20 mg/g3.
  - (d) Ginsenoside Rb1: qualitatively detected.

In addition, in the case of product is manufactured from *P. ginseng* C.A. Meyer, ginsenoside Rf should be also be qualitatively detected.

#### 3.2.2.2 Ginseng Extract and Steamed Ginseng Extract

3.2.2.2.1 Ginseng Extract (liquid form)

- (a) Solids: no less than 60.0%.
- (b) Water-insoluble solids: no more than 3.0%.
- (c) Water-saturated n -butanol extracts: no less than  $40 \text{ mg/g}^3$ .
- (d) Ginsenoside Rb1: qualitatively detected.

In addition, in case of the product manufactured from *P. ginseng* C.A. Meyer, ginsenoside Rf should be also be qualitatively detected.

## **3.2.2.2.2** Ginseng Extract (powdered form)

- (a) Moisture: no more than 8.0%.
- (b) Water-insoluble solids: no more than 3.0%.
- (c) Water-saturated n -butanol extracts: no less than  $60 \text{ mg/g}^3$ .
- (d) Ginsenoside Rb1: qualitatively detected.

In addition, in case of the product manufactured from *P. ginseng* C.A. Meyer, ginsenoside Rf should be also be qualitatively detected.

#### 3.3 Definition of Defects

The following defects shall be applied to the dried ginseng and dried steamed ginseng.

- (a) Insect-damaged ginseng: Ginseng that is visibly damaged by insects or contains dead insects.
- (b) Mouldy ginseng: Ginseng that is visibly affected by mould.

#### 3.4 Classification of "Defectives"

A container that fails to meet one or more of the applicable quality requirements, set out in Sections 3.2 and 3.3, should be considered a "defective".

### 3.5 Lot Acceptance

A lot should be considered as meeting the applicable quality requirements referred to in Sections 3.2 and 3.3, when the number of "defectives", as defined in Section 3.4, does not exceed the acceptance number (c) of the appropriate sampling plan with an AQL of 6.5.

# 4. FOOD ADDITIVES

No additives are permitted in the products covered by this Standard.

<sup>&</sup>lt;sup>2</sup> The unique constituents of ginseng are found to be a complex mixture of saponins often referred to as ginsenosides, and more than 30 ginsenosides are known. Ginsenoside Rb1 or ginsenoside Rf is one of the major ginsenosides. Ginsenoside Rb1 is identified in all ginseng species in quantities, while ginsenoside Rf is identified mainly in *Panax ginseng* C.A. Meyer.

<sup>&</sup>lt;sup>3</sup> Indicating the content of crude saponin

#### 5. CONTAMINANTS

- **5.1** The products covered by this Standard shall comply with the maximum levels of the *General Standard for Contaminants and Toxins in Food and Feed* (CODEX/STAN 193-1995).
- **5.2** The products covered by this Standard shall comply with the maximum residue limits for pesticides established by the Codex Alimentarius Commission.

## 6. HYGIENE

- **6.1** It is recommended that the products covered by the provisions of this Standard be prepared and handled in accordance with the appropriate sections of the General Principles of Food Hygiene (CAC/RCP 1-1969), and other relevant Codex texts, such as codes of hygienic practice and codes of practice.
- **6.2** The products should comply with any microbiological criteria established in accordance with the Principles and Guidelines for the Establishment and Application of Microbiological Criteria related to Foods (CAC/GL 21-1997).

## 7. LABELLING

The products covered by this Standard shall be labelled in accordance with the *General Standard for the Labelling of Pre-packaged Foods* (CODEX STAN 1-1985). Any health claims should comply with the *Guidelines for Use of Nutrition and Health Claims* (CAC/GL 23-1997), if necessary.

In addition, the following specific provisions apply:

#### 7.1 Name of the Product

- 7.1.1 The name of the products defined in Sections 2.2.1, 2.2.2, 2.2.3 and 2.2.4 shall be *Dried Ginseng, Dried Steamed Ginseng, Ginseng Extract* and *Steamed Ginseng Extract*, respectively. In this case, the products manufactured with *P. ginseng* C.A. Meyer can be named *White Ginseng, Red Ginseng, White Ginseng Extract* and *Red Ginseng Extract*.
- **7.1.2** The style shall appear on the label in conjunction with, or in close proximity to the name of the product, to avoid misleading or confusing the consumer.

## 7.2 Name of the Ginseng Species

All ginseng products shall be labelled with the scientific or common name of the ginseng that is used as raw material. The common names of the ginseng species shall be declared in accordance with the law and custom of the country where the products is consumed, in a manner not to mislead the consumer.

#### 7.3 Country of Origin

The country of origin of the product and/or raw material shall be declared if its omission is likely to mislead or deceive the consumer.

#### 7.4 Labelling of Non-retail Containers

Information about non-retail containers shall be given on the container or in accompanying documents, except that the name of the product, lot identification and the name and address of the manufacturer, packer or distributor, as well as storage instructions, shall appear on the container. However, lot identification, and the name and address of the manufacturer, packer or distributor may be replaced by an identification mark, provided that such a mark is clearly shown in the accompanying documents.

## 7.5 Optional Labelling

The products may have a clear marking to indicate that they are not intended for medicinal purposes, including other labeling requirements stipulated by the country where ginseng products are distributed.

## 8. METHODS OF ANALYSIS AND SAMPLING

#### 8.1 Preparation of Test Sample

Dried ginseng is pulverized using a grinder to make approximately 3 mm-sized particles for the analysis. Ginseng extract is used in the analysis as is.

# 8.2 Methods of Analysis

PROVISION	METHOD	PRINCIPLE	TYPE
Moisture	AOAC 925.45 B (Dried ginseng) Quantity of sample: 2 g AOAC 925.45 D (Ginseng extract) Quantity of sample: 1.5 g (mixing with 20 g of sea sand)	Gravimetry	IV
Solids	AOAC 925.45 B (Dried ginseng) - calculated by subtracting the content of moisture from 100% Quantity of sample: 2 g AOAC 925.45 D (Ginseng extract) - calculated by subtracting the content of moisture from 100% Quantity of sample: 1.5 g (mixing with 20 g of sea sand)	Calculation	IV
Ash	AOAC 923.03	Gravimetry	IV
Water-insoluble solids	As described in Annex I	Gravimetry	IV
Water-saturated n-butanol extracts	As described in Annex II	Gravimetry	IV
Identification of ginsenosides Rb1, and Rf	As described in Annex III	TLC or HPLC	IV

# References

1. Standard Operation Procedure (SOP) for Determination of Moisture (attached to the Standard)

2. Standard Operation Procedure (SOP) for Determination of Ash (attached to the Standard)

### ANNEX I

#### Determination of water-insoluble solid content

## 1. SCOPE OF APPLICATION

This method can be applied for the analysis of ginseng extract (liquid and powder form).

## 2. PRINCIPLES

Samples are dissolved in distilled water and centrifuged. The supernatant is removed, and the remaining solid is precipitated and dried. Its weight is determined to be the water-insoluble solid content.

## 3. EQUIPMENT & APPARATUS

- **3.1** Centrifuge (temperature controllable).
- 3.2 Centrifuge tubes for centrifugation.
- 3.3 Serum separation tube or micro-pipette.
- **3.4** Drying oven with a thermostat (±1°C temperature control).
- **3.5** Electronic balance (measurable down to 0.1 mg).
- 3.6 Desiccator (silica gel).
- 3.7 Tongs.

## 4. EXPERIMENTAL PROCEDURES

- **4.1** Dry a centrifuge tube in a drying oven at 105°C for 3 hours. After drying, place the centrifuge tube in a desiccator, let it stand at room temperature for 30 minutes, and then record its weight.
- **4.2** Repeat procedure step 4.1 until a constant weight is obtained for the centrifuge tube. Note, however, that the drying time should be 1-2 hours.
- **4.3** Precisely weigh out approximately 1 g of sample and place it in the centrifuge tube with known constant weight<sup>4</sup>.
- 4.4 Add 15 ml of distilled water to the centrifuge tube containing the sample to dissolve the sample.
- **4.5** Centrifuge the tube at room temperature at 1,000×g<sup>5</sup> for 15 minutes and then remove the supernatant immediately using a serum separation tube while trying not to touch the separated precipitate. The supernatant may not be able to be completely removed due to the necessity of leaving a small amount of the supernatant to prevent the loss of suspended solids.
- **4.6** Repeat procedural steps 4.4 and 4.5 two more times with the solid that remains in the centrifuge tube.
- **4.7** Dry the centrifuge tube with the remaining sample in a drying oven at 105°C for 5 hours.
- **4.8** After drying, place the centrifuge tube in a desiccator, let it stand at room temperature for 30 minutes, and then measure its weight.
- **4.9** Repeat procedures step 4.7 and 4.8 until a constant weight is obtained for the centrifuge tube containing the sample. Note, however, that the drying time should be 1-2 hours.
- **4.10** The water-insoluble solid content is calculated as follows:

Water-insoluble solid content (%) =  $\frac{W_1 - W_0}{S} \times 100$ 

 $W_0$ : Weight of the centrifuge tube (g)

 $W_1$ : Weight of the centrifuge tube with the solid residue after drying (g)

S: Weight of the sample (g)

<sup>&</sup>lt;sup>4</sup> The constant weight is the smaller value among weights measured successively when the weight difference between the current weight measurement and the previous weight measurement is less than 2 mg.

 $g = G \frac{M}{R^2}$  (g: gravity acceleration, G: gravity constant, R: radius, M: mass)

#### ANNEX II Determination of water-saturated n-butanol extracts

## 1. SCOPE OF APPLICATION

This method can be applied for the analysis of dried ginseng and ginseng extracts (liquid and powder forms).

## 2. PRINCIPLES

Crude saponin is extracted from ginseng products using water-saturated n-butanol as the solvent after the removal of the nonpolar lipids and carbohydrates using diethyl ether and distilled water.

### 3. EQUIPMENT & APPARATUS

- 3.1 Separatory funnel (250 ml).
- 3.2 Round flat flask (200-300 ml).
- 3.3 Erlenmeyer flask (200-300 ml).
- 3.4 Standard sieve (No. 80).
- 3.5 Filter paper (No. 2).
- 3.6 Glass funnel.
- 3.7 Funnel Shaker.
- 3.8 Rotary evaporator.
- 3.9 Constant-temperature water bath.
- **3.10** Electronic balance (measurable down to 0.1 mg).
- **3.11** Drying oven with a thermostat (±1°C temperature control).
- 3.12 Desiccator (silica gel).
- 3.13 Grinder.
- 3.14 Tongs.

#### 4. REAGENTS

- 4.1 n-butanol (over EP grade).
- 4.2 Diethyl ether (over EP grade).
- 4.3 Distilled water.

# 5. PREPARATION OF THE WATER-SATURATED N-BUTANOL SOLUTION

- 5.1 Mix n-butanol and distilled water at a ratio of 70:30.
- **5.2** Shake the mixture sufficiently and let it stand so that the upper layer (water-saturated n-butanol layer) and the lower layer (water layer) separate completely.
- **5.3** After complete separation is achieved, the water-saturated n-butanol layer is stored in a container and capped until further use.

# 6. PRETREATMENT OF SAMPLES

Dried ginseng samples are pulverized using a grinder and sifted through an 80-mesh sieve for experimental use. The ginseng extract is used in the experiment as is.

# 7. EXPERIMENTAL PROCEDURES FOR DRIED GINSENG

- **7.1** Precisely weigh out approximately 5 g of sample and place it in a round flat flask (A). Then, add 50 ml of the water-saturated n-butanol solution. Perform reflux extraction in a constant-temperature water bath at 75-80°C for 1 hour and then let it stand for 30 minutes.
- 7.2 Transfer the solution obtained in step 7.1 into a separatory funnel after filtering it through filter paper.

- 7.3 Repeat procedures step 7.1 and 7.2 two more times for the solid remains in the round flat flask (A).
- **7.4** Add 50 ml of distilled water to the mixed solution obtained in step 7.2-7.3 and then shake the solution using a funnel shaker (approximately 15 minutes). Let it stand until the upper layer (water-saturated n-butanol layer) and the lower layer (water layer) are completely separated.
- **7.5** Transfer the upper layer (water-saturated n-butanol layer) into a previously weighed flat bottom flask (B) and vacuum-concentrate and dry (60°C) the sample until the liquid is completely removed.
- **7.6** Add 50 ml of diethyl ether to the round flat flask (B) containing the precipitates and reflux the sample again in a constant-temperature water bath at 46°C for 30 minutes.
- **7.7** Discard the diethyl ether in the flat bottom flask (B) by filtering the sample through filter paper and then collect the precipitates on the filter paper in a flat bottom flask (B) by dissolving them with methanol.
- 7.8 Concentrate the contents in the round flat flask (B) until the odors of diethyl ether and methanol disappear.
- **7.9** After drying the round flat flask (B) in a drying oven at 105°C for 1 hour, place it in a desiccator at room temperature, let it stand for 1 hour, and then measure its weight.
- 7.10 The water-saturated n-butanol content of dried ginseng is calculated as follows:

Water-saturated n-butanol extract (mg/g) =  $\frac{W_1 - W_0}{c}$ 

*W*<sub>0</sub>: Weight of the flask (mg)

 $W_1$ : Weight of the flask after concentration and drying (mg)

S: Weight of the sample (g)

# 8. EXPERIMENTAL PROCEDURES FOR GINSENG EXTRACTS

- **8.1** Precisely weigh out approximately 2 g of sample in an Erlenmeyer flask, add 60 ml of distilled water to dissolve the sample, and then transfer it to a separatory funnel (A).
- **8.2** Add 60 ml of diethyl ether, shake the funnel several times, and then remove the gas by opening the cork. Repeat the above procedure step 8.2, 2-3 times.
- **8.3** Shake the separatory funnel sufficiently in a funnel shaker (approximately 15 minutes) and then let it stand until the upper layer (diethyl ether layer) and the lower layer (water layer) are completely separated.
- **8.4** Transfer the lower portion (water layer) to a different separatory funnel (B), add 60 ml of the water-saturated n-butanol solution, shake the funnel under the same conditions as described in step 8.3, and let it stand until the layers are completely separated. The supernatant (water-saturated n-butanol layer) is collected (collected from above of the boundary surface) and transferred to another flask.

\* At this time, the lower layer (water layer) is considered the emulsion layer in the next two separation stages but not in the final separation stage.

- **8.5** Repeat procedure step 8.4 two more times on the lower layer (water layer) left in the separatory funnel (B). At the final separation stage, the supernatant including the emulsion is slowly removed, leaving only the upper layer, by opening the spout of the separatory funnel.
- **8.6** Collect the solution (supernatants from each separation stage) obtained from procedures step 8.4-8.6 into the separatory funnel (B), add 50 ml of distilled water, and shake the funnel under the same conditions as described in (c). Then, let it stand\_until the upper layer (n-butanol layer) and the lower layer (water layer) are completely separated.
- **8.7** Transfer the supernatant (n-butanol layer) into the previously weighed flat bottom flask and vacuum-concentrate (60°) it until the liquid is completely removed.
- **8.8** Dry the flat-bottomed flask in a drying oven at 105°C for 1 hour and then place in a desiccator at room temperature. Let it stand for 1 hour and then measure its weight.
- **8.9** Calculate the water-saturated n-butanol content in the ginseng extract using the same method as described in step 7.10.

#### ANNEX III

#### Identification of ginsenosides Rb<sub>1</sub>, and Rf

Ginsenosides in ginseng products can be identified by thin-layer chromatography (TLC) or high-performance liquid chromatography (HPLC).

#### 1. SAMPLE SOLUTION PREPARATION

The dried 1-butanol extract obtained according to the method for the measurement of the water-saturated n-butanol extract in Annex II is completely dissolved in 10 ml of methanol and then filtered through a 0.45-µm membrane filter.

# 2. STANDARD SOLUTION PREPARATION

Reference substances for ginsenoside  $Rb_1$  and ginsenoside Rf are dissolved in methanol to concentrations of 0.2%, and then the solutions are filtered through a 0.45-µm membrane filter.

#### 3. IDENTIFICATION

### 3.1 Thin-Layer Chromatography (TLC)

#### 3.1.1 Preparation of the developing solvent

- (a) Mix n-butanol: ethyl acetate:water at a ratio of 50:10:40 (A), or chloroform:methanol:water at a ratio of 65:35:10 (B) in a separatory funnel.
- (b) Shake the funnel sufficiently and let it stand until the solvent is completely separated.
- (c) Collect only the upper layer when using solvent (A) as the developing solvent and only the lower layer when using solvent (B) and store the layers for further use. Collect from above (A) or below (B) the boundary surface of the relevant solvent when each solvent is separated and stored to increase the purity of the developing solvent.

#### 3.1.2 Developing chamber

- (a) Use a developing chamber with a cover (the developing chamber is completely sealed by applying glycerin, etc.).
- (b) Attach filter paper to the sides and back of the inside of the developing chamber and soak them with the developing solvent.
- (c) Place the developing solvent slowly into the developing chamber (approximately halfway up to the starting line of the TLC plate).
- (d) Place the cover on and let it stand until the inside of the developing chamber is sufficiently saturated (30 minutes).

#### 3.1.3 TLC preparation

- (a) The TLC plate is cut into appropriate pieces over 10 cm in length and wide enough to accommodate the number of samples needed for identifying the ginsenosides.
- (b) Place the plate in a clean drying oven and dry it at 110°C for 10-15 minutes before use.
- (c) Draw a line (starting line) 1 cm from the bottom of the TLC plate and mark the spots for dropping the samples. Then, draw a line (ending line) at exactly 8 cm from the starting line.

#### 3.1.4 TLC identification

- (a) Five-microliter samples of the ginsenoside references and the sample solutions prepared as described above are dropped while drying using a dryer. Each 5-μl sample is dropped by dividing it into several drops carefully without scraping off the silica gel of the TLC plate and not by using one drop.
- (b) After the dropping is completed, dry the TLC plate with a dryer.
- (c) Place the TLC plate in the developing chamber with its starting line at the bottom and develop the samples.
- (d) When the developing solvent reaches the ending line, the TLC plate is taken out and dried with a dryer.
- (e) Spray a 10% sulfuric acid solution evenly on the TLC plate.

- (f) Place the plate in a dryer at 110°C for 5-10 minutes for the development of the colors.
- (g) Compare the R<sub>f</sub> values and colors of the substances separated from the sample with those of the ginsenoside references to identify the relevant ginsenosides in the ginseng products.

 $R_f = \frac{\textit{distance sample solution migrated}}{\textit{distance developing solvent migrated}}$ 

# 3.2 High-Performance Liquid Chromatography (HPLC)

The sample solution prepared according to the description above and the ginsenoside references are analyzed using HPLC under the conditions described below. Ginsenosides in the sample solutions can be identified by comparing their retention times with the peaks shown by the ginsenosides in the reference substances.

<Operating conditions>

- (a) Column: ODS column
- (b) Detector: UV (203 nm) or ELSD
- (c) Eluent
  - UV: acetonitrile:water (30:70, v/v)-
  - ELSD: acetonitrile:water:isopropanol (94.9:5.0:0.1, v/v/v)
- (d) Flow rate: 1.0 ml/min~2.0 ml/min
- % The analytical conditions can be adjusted depending on the laboratory conditions, but the peaks of Rb<sub>1</sub>, and Rf in the chromatogram should NOT be located in the first 5 minutes NOR in the last 5 minutes of the retention time.

# **Reference 1**

# Standard Operation Procedure for Determination of Moisture

# 1. SCOPE OF APPLICATION

This method can be applied for the analysis of dried ginseng and ginseng extract.

# 2. PRINCIPLES

It is assumed that the moisture is the only volatile component in food. When the pressure of the water vapor in food is increased by heating, that of the surroundings is reduced relative to that of the food. The moisture in a food sample can be completely evaporated during heating at 105°C without the occurrence of any chemical change.

# 3. EQUIPMENT & APPARATUS

- **3.1** Weighing bottle with a lid.
- **3.2** Glass rod (It should protrude at least 1.5 cm from the surface of the sea sand when inserted at a 45° angle into a weighing bottle containing 20 g of sea sand.).
- **3.3** Drying oven with a thermostat (±1°C temperature control).
- **3.4** Electronic balance (measurable down to 0.1 mg).
- 3.5 Sea sand (20-35 mesh).
- 3.6 Desiccator (silica gel).
- 3.7 Grinder.
- 3.8 Tongs.

# 4. PRE-TREATMENT OF SAMPLES

Dried ginseng samples are pulverized using a grinder to make approximately 3-mm-sized particles for the experiment. The ginseng extract is used in the experiment as is.

# 5. EXPERIMENTAL PROCEDURES - DRIED GINSENG AND GINSENG EXTRACT (POWDER FORM)

- **5.1** Dry a weighing bottle and a lid separately in a drying oven at 105°C for 5 hours. Afterwards, place the weighing bottle capped tightly with the lid in a desiccator, let it stand at room temperature for 30 minutes, and then measure its weight.
- **5.2** Repeat procedure step 5.1 until a constant weight is obtained for the bottle and lid. Note, however, that the drying time should be 1-2 hours.
- **5.3** Precisely weigh out approximately 2 g of sample, and place it into the weighing bottle with known constant weight.
- **5.4** Dry the weighing bottle containing the sample in a drying oven at 105°C for 3 hours. The lid is placed slightly ajar to dry the sample in the weighing bottle.
- **5.5** Place the weighing bottle capped tightly with the lid in a desiccator, let it stand at room temperature for 30 minutes, and then measure its weight.
- **5.6** Repeat procedures 5.4 and 5.5 until a constant weight is obtained for the bottle containing the sample. Note, however, that the drying time should be 1-2 hours.
- 5.7 The moisture content is calculated as follows:

Moisture content in the sample (%) =  $\frac{S - (W_1 - W_0)}{S} \times 100$ 

 $W_0$ : Weight of the weighing bottle (g)

 $W_1$ : Weight of the weighing bottle with the sample after drying (g)

S: Weight of the sample (g)

# 6. EXPERIMENTAL PROCEDURES - GINSENG EXTRACT (LIQUID FORM)

6.1 Dry the weighing bottle containing 20 g of sea sand and a glass rod in a drying oven at 105°C for 5 hours.

- **6.2** After drying, place the weighing bottle in a desiccator, let it stand at room temperature for 30 minutes, and then measure its weight.
- **6.3** Repeat procedures 6.1 and 6.2 until a constant weight is obtained for the bottle containing the sea salt and the glass rod. Note, however, that the drying time should be 1-2 hours.
- **6.4** Precisely weigh out approximately 1.5 g of sample and place it into the weighing bottle with a known constant weight. Then, mix the sample well with the sea sand and evenly spread the mixture on the surfaces of the weighing bottle walls using the glass rod.
- 6.5 The remaining analytical steps and calculations are the same as for step 5.4 and 5.5 of Section 5 above.

## **Reference 2**

## Standard Operation Procedure for Determination of Ash

#### 1. SCOPE OF APPLICATION

This method can be applied for the analysis of dried ginseng samples.

## 2. PRINCIPLES

Samples are collected in a container (crucible) for ash analysis and burned at 525-600°C to remove the organic substances. The total mineral weight of the remaining sample is considered the ash content.

# 3. EQUIPMENT & APPARATUS

- **3.1** Porcelain crucible with a lid.
- 3.2 Electric heating plate.
- **3.3** Electric furnace with a thermostat (±1°C temperature control).
- 3.4 Electronic balance (measurable down to 0.1 mg).
- **3.5** Desiccator (silica gel).
- 3.6 Grinder.
- 3.7 Tongs.

## 4. PRETREATMENT OF SAMPLES

Dried ginseng samples are pulverized using a grinder to make approximately 3-mm-sized particles for the experiment.

# 5. EXPERIMENTAL PROCEDURES

- **5.1** Heat a clean porcelain crucible in an electric furnace at 550°C for 3 hours. Let it stand at room temperature for 1 hour, and then measure its weight.
- **5.2** Repeat procedure step 5.1 until a constant weight is obtained. Note, however, that the ashing time should be 1-2 hours.
- 5.3 Precisely weigh out approximately 3 g of sample in the porcelain crucible with known constant weight.
- **5.4** Place the porcelain crucible containing the sample in an electric furnace at 550°C and ash the sample by heating the crucible with the lid on it until white or bright grayish white ash is formed.
- **5.5** After ashing is complete, place the porcelain crucible containing the sample in a desiccator, let it stand at room temperature for 1 hour, and then measure its weight.
- **5.6** Repeat procedures step 5.4 to 5.5 until a constant weight is obtained for the crucible containing the sample. Note, however, that the ashing time should be 1-2 hours.
- 5.7 The ash content is calculated as follows:

Ash content in the sample (%) =  $\frac{(W_2 - W_1)}{s} \times 100$ 

 $W_1$ : Weight of the porcelain crucible before ashing (g)

 $W_2$ : Weight of the porcelain crucible after ashing (g)

S: Weight of the sample (g)