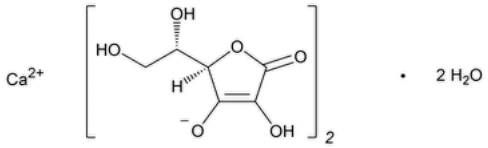
Status: Currently Official on 14-Feb-2025
Official Date: Official as of 01-Aug-2023
Document Type: USP Monographs
DocId: GUID-88C50D31-0D48-4A52-B187-266E8D0C8F8F_8_en-US
DOI: https://doi.org/10.31003/USPNF_M11410_08_01
DOI Ref: bnxxu

Calcium Ascorbate



$C_{12}H_{14}CaO_{12} \cdot 2H_2O$ 426.34

L-Ascorbic acid, calcium salt (2:1), dihydrate;

Calcium L-ascorbate (1:2), dihydrate [5743-28-2]; UNII:183E4W213W.

DEFINITION

Calcium Ascorbate contains NLT 98.0% and NMT 101.0% of calcium ascorbate dihydrate ($C_{12}H_{14}CaO_{12} \cdot 2H_2O$), calculated on the as-is basis.

IDENTIFICATION

Change to read:

• **A.** ▲Characteristic emission lines for calcium at 184.0, 315.9, and 317.9 nm from the *Sample solution* correspond to those from the *Standard solution*, as obtained in the ▲*Content of Calcium*.▲ (ERR 1-Aug-2023) ▲ (USP 1-Aug-2023)

Change to read:

• **B.** ▲The retention time of the ascorbic acid peak of the *Sample solution* corresponds to that of the *Standard solution*, as obtained in the *Assay*.▲ (USP 1-Aug-2023)

• **C.** **Spectroscopic Identification Tests** ⟨197⟩, *Infrared Spectroscopy*: 197M

ASSAY

Change to read:

• **Procedure**

▲**Mobile phase:** 50 mM monobasic sodium phosphate, adjusted with phosphoric acid to a pH of 2.5

Diluent: Dissolve 73 g of metaphosphoric acid in 1.0 L of water.

Standard stock solution: 2 mg/mL of USP Ascorbic Acid RS in *Diluent*

Standard solution: 0.2 mg/mL of USP Ascorbic Acid RS in *Mobile phase*, from the *Standard stock solution*

Sample stock solution: Transfer 220 mg of Calcium Ascorbate to a 100-mL volumetric flask. Dissolve and dilute with *Diluent* to volume.

Sample solution: 0.22 mg/mL of Calcium Ascorbate in *Mobile phase*, from the *Sample stock solution*

Blank: *Diluent*

Chromatographic system

(See *Chromatography* ⟨621⟩, *System Suitability*.)

Mode: LC

Detector: UV 245 nm

Column: 4.6-mm × 25-cm; 5-µm packing L96

Temperatures

Autosampler: 5°

Column: 10°

Flow rate: 0.8 mL/min

Injection volume: 10 µL

System suitability

Sample: *Standard solution*

Suitability requirements

Tailing factor: NMT 2.0

Relative standard deviation: NMT 2.0% for the ascorbate peak

Analysis

Samples: *Standard solution* and *Sample solution*

Calculate the percentage of calcium ascorbate dihydrate ($C_{12}H_{14}CaO_{12} \cdot 2H_2O$) in the portion of sample taken:

$$\text{Result} = (r_U/r_S) \times (C_S/C_U) \times (M_{r1}/2M_{r2}) \times 100$$

r_U = peak response of ascorbic acid from the *Sample solution*

r_S = peak response of ascorbic acid from the *Standard solution*

C_S = concentration of USP Ascorbic Acid RS in the *Standard solution* (mg/mL)

C_U = concentration of Calcium Ascorbate dihydrate in the *Sample solution* (mg/mL)

M_{r1} = molecular weight of calcium ascorbate dihydrate, ▲426.34▲ (ERR 1-Aug-2023)

M_{r2} = molecular weight of ascorbic acid, 176.12

Acceptance criteria: 98.0%–101.0% on the as-is basis▲ (USP 1-Aug-2023)

Add the following:

▲OTHER COMPONENTS

- **Content of Calcium**

Stock aqua regia solution: A mixture of hydrochloric acid and nitric acid (3:1), prepared as follows. Add the nitric acid to the hydrochloric acid. [Note—Periodically vent the solution in an appropriate fume hood.]

Diluent: A mixture of *Stock aqua regia solution* and deionized water (1:9), prepared as follows. Add 1 volume of *Stock aqua regia solution* to 2 volumes of deionized water. Dilute with additional deionized water to volume, and mix well.

Standard stock solution: Using a commercially available calcium standard solution in 5% (v/v) nitric acid solution, pipet an appropriate amount of calcium standard solution into a volumetric flask and dilute with 5% (v/v) nitric acid solution to obtain a 1000-mg/L calcium solution.

Standard solution A: Dilute the *Standard stock solution* with *Diluent* to obtain a concentration of 5.0 mg/L.

Standard solution B: Dilute the *Standard stock solution* with *Diluent* to obtain a concentration of 30.0 mg/L.

Standard solution C: Dilute the *Standard stock solution* with *Diluent* to obtain a concentration of 60.0 mg/L.

Standard solution D: Dilute the *Standard stock solution* with *Diluent* to obtain a concentration of 100.0 mg/L.

Standard solution E: Dilute the *Standard stock solution* with *Diluent* to obtain a concentration of 150.0 mg/L.

Standard solution F: Dilute the *Standard stock solution* with *Diluent* to obtain a concentration of 250.0 mg/L.

Sample solution: Transfer 266 mg of Calcium Ascorbate (equivalent to about 25 mg of calcium) to a 250-mL Erlenmeyer flask. Cautiously add 25 mL of *Stock aqua regia solution* in 5-mL increments and swirl after each addition. Once bubbling stops, bring to a boil on a hot plate set at low to medium heat. Continue gently boiling until fumes cease (for about an hour). Remove from the heat source and let the solution cool to room temperature. Transfer the solution quantitatively to a 250-mL volumetric flask, dilute with deionized water to volume, and mix well. Pass about 30 mL through a nylon syringe filter of 5-µm pore size into a polypropylene centrifuge tube.

Blank: *Diluent*

Instrumental conditions

(See *Plasma Spectrochemistry* ⟨730⟩.)

Mode: ICP-OES

Emission wavelength: About 315.9 nm or optimized wavelength for calcium. For *Identification A*, detect additional calcium emission lines at 184.0 and 317.9 nm. [Note—The operating conditions may be developed and optimized based on the manufacturer's recommendation. The wavelengths selected should be demonstrated experimentally to provide sufficient specificity, sensitivity, linearity, accuracy, and precision.]

System suitability

Sample: *Standard solutions*

Suitability requirements

Correlation coefficient: NLT 0.99, determined from the linear calibration constructed in the *Analysis*, *Standard solutions A–F*

Relative standard deviation: NMT 2.0% from five replicate analyses of *Standard solution D*

Analysis

Samples: *Diluent*, *Standard solutions*, and *Sample solution*

Construct a linear calibration curve using the intensity of the emission from the six *Standard solutions*. Determine the emission lines of calcium in each *Standard solution* and the *Sample solution*. Plot the emission values of calcium in the *Standard solutions* versus the concentration, in mg/L, of calcium, and draw the straight line best fitting the plotted points. From the graph, determine the concentration (C), in mg/L, of calcium in the *Sample solution*.

Calculate the percentage of calcium in the portion of Calcium Ascorbate taken:

$$\text{Result} = C \times (V/W) \times 100$$

C = concentration of calcium in the *Sample solution* (mg/L)

V = volume of the *Sample solution* (L)

W = sample weight (mg)

Acceptance criteria: 9.0%–10.0% on the as-is basis▲ (USP 1-Aug-2023)

IMPURITIES

• **Arsenic ⟨211⟩, *Procedures, Procedure 1*:** NMT 3 µg/g

• **Limit of Fluoride**

Prepare and store all solutions in plastic containers.

Buffer solution: 294 mg/mL of sodium citrate dihydrate in water

Standard stock solution: 1.1052 mg/mL of USP Sodium Fluoride RS in water

Standard solution: Transfer 20.0 mL of the *Standard stock solution* to a 100-mL volumetric flask containing 50.0 mL of *Buffer solution*, dilute with water to volume, and mix. Each milliliter of the *Standard solution* contains 100 µg of fluoride ion.

Sample solution: Transfer 2.0 g of Calcium Ascorbate to a beaker containing a plastic-coated stirring bar. Add 20 mL of water and 2.0 mL of hydrochloric acid, and stir until dissolved. Add 50.0 mL of *Buffer solution* and sufficient water to make 100 mL.

Electrode system: Use a fluoride-specific ion-indicating electrode and a silver–silver chloride reference electrode connected to a pH meter capable of measuring potentials with a minimum reproducibility of ± 0.2 mV (see *pH ⟨791⟩*).

Standard response line: Transfer 50.0 mL of *Buffer solution* and 2.0 mL of hydrochloric acid to a beaker, and add water to make 100 mL. Add a plastic-coated stirring bar, insert the electrodes into the solution, stir for 15 min, and read the potential, in mV. Continue stirring, and at 5-min intervals add 100, 100, 300, and 500 µL of the *Standard solution*, record the potential 5 min after each addition. Plot the logarithms of the cumulative fluoride ion concentrations (0.1, 0.2, 0.5, and 1.0 µg/mL) versus potential, in mV.

Analysis

Samples: *Standard solution* and *Sample solution*

Rinse and dry the electrodes, insert them into the *Sample solution*, stir for 5 min, and record the potential, in mV. From the potential and the *Standard response line*, determine the concentration (*C*), in µg/mL, of fluoride ion in the *Sample solution*.

Calculate the content, in ppm, of fluoride in the portion of Calcium Ascorbate taken:

$$\text{Result} = (C \times V)/W$$

C = concentration of fluoride ion in the *Sample solution* (µg/mL)

V = volume of the *Sample solution* (mL)

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

Acceptance criteria: NMT 10 ppm

SPECIFIC TESTS

• **Optical Rotation ⟨781S⟩, *Procedures, Specific Rotation***

Sample solution: 50 mg/mL in carbon dioxide-free water. [Note—Perform measurements immediately after preparation.]

Acceptance criteria: +95° to +97°

• **pH ⟨791⟩**

Sample solution: 100 mg/mL

Acceptance criteria: 6.8–7.4

• **Loss on Drying ⟨731⟩**

Sample: 3 g

Analysis: Dry the *Sample* at 105° for 2 h.

Acceptance criteria: NMT 0.1%

ADDITIONAL REQUIREMENTS

• **Packaging and Storage:** Preserve in tight, light-resistant containers.

Change to read:

• **USP Reference Standards ⟨11⟩**

▲USP Ascorbic Acid RS▲ (USP 1-Aug-2023)

USP Calcium Ascorbate RS

USP Sodium Fluoride RS

Auxiliary Information - Please check for your question in the FAQs before contacting USP.

Topic/Question	Contact	Expert Committee
CALCIUM ASCORBATE	Natalia Davydova Scientific Liaison	NBDS2020 Non-botanical Dietary Supplements

Chromatographic Database Information: Chromatographic Database

Most Recently Appeared In:
Pharmacopeial Forum: Volume No. 47(3)
Current DocID: GUID-88C50D31-0D48-4A52-B187-266E8D0C8F8F_8_en-US
DOI: https://doi.org/10.31003/USPNF_M11410_08_01
DOI ref: bnxxu