Status: Currently Official on 14-Feb-2025
Official Date: Official as of 01-Aug-2022
Document Type: USP Monographs
DocId: GUID-7EC222B5-6160-436A-9492-5256ED93F317_7_en-US
DOI: https://doi.org/10.31003/USPNF_M11318_07_01
DOI Ref: 39912

Calcipotriene Ointment

DEFINITION

To view the Notice from the Expert Committee that posted in conjunction with this accelerated revision, please click *www.uspnf.com/rb-calcipotriene-ointment-20220429*.

Calcipotriene Ointment contains NLT 90.0% and NMT 110.0% of the labeled amount of calcipotriene ($C_{27}H_{40}O_3$), in a suitable ointment base.

IDENTIFICATION

• **A.** The retention time of the major peak of the *Sample solution* corresponds to that of the *Standard solution*, as obtained in the *Assay*.

Change to read:

• **B.** ▲The UV spectrum of the major peak of the *Sample solution* corresponds to that of the *Standard solution*, as obtained in the *Assay*.▲ (USP 1-Aug-2022)

ASSAY

Change to read:

• **Procedure**

▲Protect solutions containing calcipotriene from light and air. Prepare the solutions containing calcipotriene NMT 1 h before use.▲ (USP 1-Aug-2022)

Mobile phase: Methanol and water (70:30)

Buffer: 132 g/L of monobasic ammonium phosphate in water

Diluent: Methanol, *Buffer*, and water (700:3:297)

Standard stock solution: 0.1 mg/mL of USP Calcipotriene RS in *Diluent*. Sonicate if necessary.

Standard solution: 2 µg/mL of USP Calcipotriene RS prepared as follows. Transfer 5 mL of *Standard stock solution* into a 250-mL volumetric flask, add 50 mL of tetrahydrofuran, and dilute with *Diluent* to volume.

Sample stock solution: Nominally equivalent to 0.01 mg/mL of calcipotriene in tetrahydrofuran prepared as follows. Transfer Ointment equivalent to 0.25 mg of calcipotriene into a 25-mL volumetric flask. Add 15 mL of tetrahydrofuran and sonicate, with intermittent shaking, for 20 min in a cold water bath. Dilute with tetrahydrofuran to volume.

Sample solution: Nominally equivalent to 2 µg/mL of calcipotriene prepared as follows. Transfer 5 mL of the *Sample stock solution* into a suitable container. Add 20 mL of *Diluent*, mix, and sonicate for 10 min. Pass through a suitable filter of 0.45-µm pore size. Inject immediately after preparation.

Chromatographic system

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

Mode: LC

Detector: UV 264 nm. ▲For *Identification B*, use a diode array detector in the range of 190–400 nm.▲ (USP 1-Aug-2022)

Column: 4.6-mm × 15-cm; 3-µm packing L1

Flow rate: 1.0 mL/min

Injection volume: 50 µL

System suitability

Sample: *Standard solution*

Suitability requirements

Tailing factor: NMT 2.0

Relative standard deviation: NMT 2.0%

Analysis

Samples: *Standard solution* and *Sample solution*

Calculate the percentage of the labeled amount of calcipotriene ($C_{27}H_{40}O_3$) in the portion of Ointment taken:

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

r_U = peak response of calcipotriene from the *Sample solution*

r_S = peak response of calcipotriene from the *Standard solution*

C_S = concentration of USP Calcipotriene RS in the *Standard solution* (µg/mL)

C_U = nominal concentration of calcipotriene in the *Sample solution* (µg/mL)

Acceptance criteria: 90.0%–110.0%

PERFORMANCE TESTS

• **Minimum Fill ⟨755⟩:** Meets the requirements

IMPURITIES

Change to read:

• **Organic Impurities**

▲Protect solutions containing calcipotriene from light and air. Prepare the *Standard solution* and the *Sample solution* NMT 1 h before use. Prepare the *System suitability solution* daily.▲ (USP 1-Aug-2022)

Mobile phase, Buffer, and **Diluent:** Prepare as directed in the *Assay*.

System suitability solution: 10.0 µg/mL of USP Calcipotriene RS and 0.1 µg/mL of USP Calcipotriene Related Compound C RS in *Diluent*

Standard stock solution: 1.0 µg/mL of USP Calcipotriene RS in *Diluent*

Standard solution: 0.1 µg/mL of USP Calcipotriene RS prepared as follows. Transfer 1.0 mL of the *Standard stock solution* into a 10-mL volumetric flask, add 1.0 mL tetrahydrofuran, and dilute with *Diluent* to volume.

▲**Sensitivity solution:** 0.01 µg/mL of USP Calcipotriene RS prepared as follows. Transfer 1.0 mL of the *Standard solution* into a 10-mL volumetric flask, add 1.0 mL tetrahydrofuran, and dilute with *Diluent* to volume.▲ (USP 1-Aug-2022)

Sample solution: Nominally equivalent to 0.01 mg/mL of calcipotriene prepared as follows. Transfer Ointment equivalent to 0.1 mg of calcipotriene into a glass-stoppered test tube, and add 1 mL of tetrahydrofuran. Sonicate for 20 min with intermittent shaking. Add 9 mL of *Diluent*, and sonicate for 5 min. Shake the test tube vigorously, and then place it in a beaker containing ice cold water for 2–3 min. Pass the liquid portion through a nylon filter of 0.45-µm pore size, and discard the first few milliliters of the solution.

Chromatographic system

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

Mode: LC

Detector: UV 264 nm

Column: 4.6-mm × 15-cm; 3-µm packing L1

Flow rate: 1.0 mL/min

Injection volume: 100 µL

Run time: NLT 1.25 times of retention time of the calcipotriene peak

System suitability

Samples: *System suitability solution*, ▲▲ (USP 1-Aug-2022) *Standard solution*,▲and *Sensitivity solution*▲ (USP 1-Aug-2022)

Suitability requirements

Resolution: NLT 1.2 between calcipotriene related compound C and calcipotriene▲▲ (USP 1-Aug-2022) , *System suitability solution*

Relative standard deviation: NMT 5.0%, *Standard solution*

▲**Signal-to-noise ratio:** NLT 10, *Sensitivity solution*▲ (USP 1-Aug-2022)

Analysis

Samples: *Standard solution* and *Sample solution*

Calculate the percentage of any impurity in the portion of Ointment taken:

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

r_U = peak response of any impurity from the *Sample solution*

r_S = peak response of calcipotriene from the *Standard solution*

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

C_U = nominal concentration of calcipotriene in the *Sample solution* (mg/mL)

Acceptance criteria: See *Table 1*. ▲The reporting threshold is 0.1%.▲ (USP 1-Aug-2022)

Table 1

Name	Relative Retention Time	Acceptance Criteria, NMT (%)
Calcipotriene impurity B[a]	0.86	0.50

Name	Relative Retention Time	Acceptance Criteria, NMT (%)
Calcipotriene related compound C[b]	0.92	▲1.5▲ (USP 1-Aug-2022)
Calcipotriene	1.0	—
Calcipotriene impurity D[c]	1.31	▲3.0▲ (USP 1-Aug-2022)
▲▲ (USP 1-Aug-2022)	▲▲ (USP 1-Aug-2022)	▲▲ (USP 1-Aug-2022)
Any ▲▲ (USP 1-Aug-2022) unspecified impurity	—	▲0.7▲ (USP 1-Aug-2022)
Total impurities▲[d]▲ (USP 1-Aug-2022)	—	▲3.5▲ (USP 1-Aug-2022)

[a] (5*Z*,7*Z*,22*E*,24*S*)-24-Cyclopropyl-9,10-secochola-5,7,10(19),22-tetraene-1α,3β,24-triol.

[b] (5*E*,7*E*,22*E*,24*S*)-24-Cyclopropyl-9,10-secochola-5,7,10(19),22-tetraene-1α,3β,24-triol.

[c] (5*Z*,7*E*,22*E*,24*R*)-24-Cyclopropyl-9,10-secochola-5,7,10(19),22-tetraene-1α,3β,24-triol.

▲[d] Does not apply if propylene glycol ▲ethers▲ (RB 1-Aug-2022) of calcipotriene are present in the manufacturing process. Calculate the total impurities in the test for *Limit of Propylene Glycol ▲Ethers▲ (RB 1-Aug-2022) of Calcipotriene.*▲ (USP 1-Aug-2022)

Add the following:

▲• **Limit of Propylene Glycol Ethers of Calcipotriene** (if present)

Protect solutions containing calcipotriene from light and air. Prepare the solutions containing calcipotriene NMT 1 h before use.

Solution A: Add 70 mL of phosphoric acid in 1000 mL of water

Solution B: 1.32 g/L of dibasic ammonium phosphate in water

Solution C: Adjust *Solution B* with *Solution A* to a pH of 6.0 ± 0.1.

Solvent for extraction: Acetonitrile, methanol, and *Solution B* (20:50:30)

Mobile phase: Acetonitrile, methanol, and *Solution C* (20:50:30)

System suitability stock solution: 0.05 mg/mL of USP Calcipotriene RS in *Solvent for extraction*

System suitability solution: 1.0 µg/mL of USP Calcipotriene RS prepared as follows. Transfer 1.0 mL of *System suitability stock solution* and 15 mL of water into a 50-mL volumetric flask and dilute with *Solvent for extraction* to volume.

Sample stock solution: Mix all contents of two tubes of Ointment. Weigh 2 g into a glass with a screw cap, and suspend the Ointment in 8 mL of *n*-heptane by gentle heating on a steam bath. Add 8 mL of *Solvent for extraction*, and shake vigorously for 15 min. Centrifuge to separate the two phases if needed.

Sample solution: Mix 3.5 mL of lower phase of the *Sample stock solution* with 1.5 mL of water. Pass the solution through glass fiber filter,[1] and use the filtrate for analysis.

Chromatographic system

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

Mode: LC

Detector: UV 265 nm

Column: 4-mm × 12.5-cm; 4-µm packing L1

Flow rate: 1.2 mL/min

Injection volume: 500 µL

Run time: NLT 3 times of retention time of the calcipotriene peak

System suitability

Sample: *System suitability solution*

Suitability requirements

Relative standard deviation: NMT 10.0%

Analysis

Sample: *Sample solution*

The peaks of propylene glycol ethers of calcipotriene are the peaks in the range of relative retention times between 1.4 and 3.0 measured with respect to the calcipotriene peak.

Calculate the percentage of the propylene glycol ethers of calcipotriene in the portion of Ointment taken:

$$\text{Result} = (r_U/r_T) \times 100$$

r_U = peak response of the propylene glycol ethers of calcipotriene from the *Sample solution*

r_T = sum of the peak responses from the *Sample solution*

Acceptance criteria

Propylene glycol ▲ethers▲ (RB 1-Aug-2022) of calcipotriene: NMT 6.0%

Total impurities: NMT 10.0%. The total impurities are the sum of all impurities in *Table 1* and propylene glycol ▲ethers▲ (RB 1-Aug-2022) of calcipotriene.▲ (USP 1-Aug-2022)

SPECIFIC TESTS

• **Microbial Enumeration Tests ⟨61⟩** and **Tests for Specified Microorganisms ⟨62⟩:** The total aerobic microbial count is NMT 10^2 cfu/g. The total yeasts and molds count is NMT 5×10^1 cfu/g. It meets the requirements of the tests for the absence of *Staphylococcus aureus* and *Pseudomonas aeruginosa* species.

Delete the following:

▲• **pH ⟨791⟩**▲ (USP 1-Aug-2022)

ADDITIONAL REQUIREMENTS

• **Packaging and Storage:** Preserve in well-closed containers, and store at controlled room temperature. Do not freeze.

Change to read:

• **USP Reference Standards ⟨11⟩**

USP Calcipotriene RS

USP Calcipotriene Related Compound C RS

(5*E*,7*E*,22*E*,24*S*)-24-Cyclopropyl-9,10-secochola-5,7,10(19),22-tetraene-1α,3β,24-triol.

$C_{27}H_{40}O_3$ ▲412.61▲ (USP 1-Aug-2022)

[1] A suitable filter is available as GD/A-grade glass fiber filter from Whatman, ▲https://www.cytivalifesciences.com/en/us/about-us/our-brands/whatman▲ (RB 1-Aug-2022) .

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

Topic/Question	Contact	Expert Committee
CALCIPOTRIENE OINTMENT	Documentary Standards Support	SM32020 Small Molecules 3

Chromatographic Database Information: Chromatographic Database

Most Recently Appeared In:
Pharmacopeial Forum: Volume No. 45(5)

Current DocID: GUID-7EC222B5-6160-436A-9492-5256ED93F317_7_en-US

DOI: https://doi.org/10.31003/USPNF_M11318_07_01

DOI ref: 39912