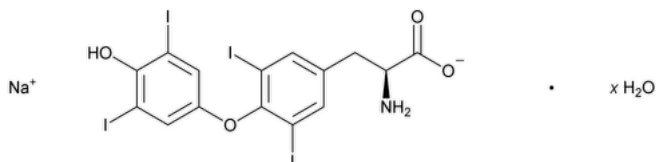


Status: Currently Official on 15-Feb-2025
 Official Date: Official as of 01-Nov-2020
 Document Type: USP Monographs
 DocId: GUID-FA5D4042-0D16-4851-ABF4-D7D047FE3B32_7_en-US
 DOI: https://doi.org/10.31003/USPNF_M45000_07_01
 DOI Ref: 1sf0t

© 2025 USPC
 Do not distribute

Levothyroxine Sodium



$C_{15}H_{10}I_4NNaO_4 \cdot xH_2O$ (anhydrous) 798.85
 L-Tyrosine, O-(4-hydroxy-3,5-diiodophenyl)-3,5-diiodo-, monosodium salt, hydrate;
 Monosodium L-thyroxine hydrate CAS RN®: 25416-65-3.
 Anhydrous CAS RN®: 55-03-8; UNII: 054I36CPMN.

DEFINITION

Levothyroxine Sodium is the sodium salt of L-3,3',5,5'-tetraiodothyronine. It contains NLT 97.0% and NMT 103.0% of levothyroxine sodium ($C_{15}H_{10}I_4NNaO_4$), calculated on the anhydrous basis.

IDENTIFICATION

- **A. SPECTROSCOPIC IDENTIFICATION TESTS (197), Infrared Spectroscopy:** (197A) or (197K)
- **B.** The retention time of the major peak of the *Sample solution* corresponds to that of the *Standard solution*, as obtained in the Assay.
- **C. IDENTIFICATION TESTS—GENERAL (191), Chemical Identification Tests, Sodium**

Sample solution: To 200 mg add 2 mL of [2 N sulfuric acid](#). Heat on a water bath and then carefully heat over an open flame, increasing the temperature gradually up to about 600°. [NOTE—Alternative procedures for igniting the material could also be used.] Continue the ignition until most of the particles have disappeared. Dissolve the residue in 2 mL of [water](#).

Acceptance criteria: The *Sample solution* meets the requirements of test A.

ASSAY

Change to read:

• PROCEDURE

Mobile phase: [Acetonitrile](#) and [water](#) (4:6) that contains 0.5 mL of [phosphoric acid](#) in each 1000 mL

Solution A: 400 mg of [sodium hydroxide](#) in 500 mL of [water](#). Cool and add 500 mL of [methanol](#).

Levothyroxine stock solution: 0.4 mg/mL of [USP Levotyroxine RS](#) in *Solution A*

Liothyronine stock solution: 0.4 mg/mL of ▲ (USP 1-Aug-2020) [USP Liothyronine RS](#) in *Solution A*. ▲ Further dilute with *Mobile phase* to 0.004 mg/mL. ▲ (USP 1-Aug-2020)

Standard solution: 10 µg/mL of ▲ [USP Levotyroxine RS](#) ▲ (USP 1-Aug-2020) from *Levothyroxine stock solution* and 0.2 µg/mL of ▲ [USP Liothyronine RS](#) ▲ (USP 1-Aug-2020) from *Liothyronine stock solution* in *Mobile phase*

Sample solution: 10 µg/mL of Levotyroxine Sodium in *Mobile phase*. [NOTE—A small amount of [0.01 M sodium hydroxide](#) ▲ in [methanol](#) ▲ (USP 1-Aug-2020) can be used to facilitate the dissolution of the sample.]

Chromatographic system

(See [Chromatography \(621\), System Suitability](#).)

Mode: LC

Detector: UV 225 nm

Column: 4.6-mm × 25-cm; ▲ 5-µm ▲ (USP 1-Aug-2020) packing [L10](#)

Flow rate: 1.5 mL/min

Injection volume: 100 µL

System suitability

Sample: *Standard solution*

Suitability requirements

Resolution: NLT 5.0 between liothyronine and levothyroxine

Relative standard deviation: NMT 2.0% for levothyroxine

Analysis

Samples: *Standard solution* and *Sample solution*

Calculate the percentage of levothyroxine sodium ($C_{15}H_{10}I_4NNaO_4$) in the portion of Levothyroxine Sodium taken:

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

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

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

C_S = concentration of [USP Levothyroxine RS](#) in the *Standard solution* (µg/mL)

C_U = concentration of Levothyroxine Sodium in the *Sample solution* (µg/mL)

M_{r1} = molecular weight of levothyroxine sodium, 798.85

M_{r2} = molecular weight of levothyroxine, 776.87

Acceptance criteria: 97.0%–103.0% on the anhydrous basis

IMPURITIES

[NOTE—On the basis of the synthetic route, perform either *Organic Impurities, Procedure 1* or *Organic Impurities, Procedure 2*. Procedure 2 is recommended when related compounds listed in [Table 3](#) may be present.]

Change to read:

• ORGANIC IMPURITIES, PROCEDURE 1

Diluent: [Acetonitrile](#) and [water](#) (1:1)

Solution A: Dilute 5 mL of [phosphoric acid](#) with *Diluent* to 100.0 mL.

Mobile phase: ▲1.0 g/L of ▲(USP 1-Aug-2020) [sodium 1-heptanesulfonate](#)▲ prepared as follows. Transfer 1 g of [sodium 1-heptanesulfonate](#) to a 1-L volumetric flask and dissolve▲(USP 1-Aug-2020) in 200 mL of [water](#). Add 200 mL of [acetonitrile](#), 400 mL of [methanol](#), and 1.0 mL of [phosphoric acid](#). Dilute with [water](#) to ▲volume.

Levothyroxine stock solution: 0.25 mg/mL of [USP Levothyroxine RS](#) prepared as follows.▲(USP 1-Aug-2020) Transfer 25 mg of [USP Levothyroxine RS](#) to a 100-mL volumetric flask ▲containing▲(USP 1-Aug-2020) 50 mL of *Diluent* and 1 drop of [10 N sodium hydroxide](#), and sonicate until dissolved. Add 7 mL of *Solution A* and dilute with *Diluent* to volume.

▲**Liothyronine stock solution:** 0.25 mg/mL of [USP Liothyronine RS](#) prepared as follows.▲(USP 1-Aug-2020) Transfer 25 mg of [USP Liothyronine RS](#) to a 100-mL volumetric flask ▲containing▲(USP 1-Aug-2020) 50 mL of *Diluent* and 1 drop of [10 N sodium hydroxide](#), and sonicate until dissolved. Add 7 mL of *Solution A* and dilute with *Diluent* to volume.

System suitability solution: ▲0.0125 mg/mL each of [USP Levothyroxine RS](#) and [USP Liothyronine RS](#) prepared as follows.▲(USP 1-Aug-2020) Transfer 5.0 mL each of ▲*Levothyroxine stock solution*▲(USP 1-Aug-2020) and ▲*Liothyronine stock solution*▲(USP 1-Aug-2020) to a 100-mL volumetric flask. Add 7 mL of *Solution A*, and dilute with *Diluent* to volume.

Standard solution: ▲0.0005 mg/mL of [USP Levothyroxine RS](#) and [USP Liothyronine RS](#) prepared as follows.▲(USP 1-Aug-2020) Transfer 4.0 mL of *System suitability solution* into a 100-mL volumetric flask. Add 7 mL of *Solution A*, and dilute with *Diluent* to volume.

Sample solution: ▲0.25 mg/mL of Levothyroxine Sodium prepared as follows.▲(USP 1-Aug-2020) Transfer 25 mg of Levothyroxine Sodium to a 100-mL volumetric flask. Add 50 mL of *Diluent*, and sonicate until dissolved. Add 7 mL of *Solution A*, and dilute with *Diluent* to volume.

Blank solution: Transfer 7 mL of *Solution A* to a 100-mL volumetric flask, and dilute with *Diluent* to volume.

Chromatographic system

(See [Chromatography \(621\), System Suitability](#).)

Mode: LC

Detector: UV 225 nm

Column: 4.6-mm × 15-cm; 5-µm packing [L7](#)

Column temperature: 35°

Flow rate: 1.5 mL/min

Injection volume: 15 µL

System suitability

Samples: *System suitability solution* and *Standard solution*

Suitability requirements

Resolution: NLT 5.0 between levothyroxine and liothyronine, *System suitability solution*

Relative standard deviation: NMT 2.0% for the levothyroxine peak, *Standard solution*

Analysis

Samples: *Standard solution*, *Sample solution*, and *Blank solution*

[NOTE—Record the chromatograms for at least 6 times the retention time of the levothyroxine peak. Verify that no peaks elute in the *Blank solution* at the expected retention times for levothyroxine and related compounds.]