https://trungtamthuoc.com/

Chlordiazepoxide and Amitriptyline Hydrochloride Tablets

DEFINITION

Change to read:

Chlordiazepoxide and Amitriptyline Hydrochloride Tablets contain NLT 90.0% and NMT 110.0% of the labeled amount of chlordiazepoxide ($C_{16}H_{14}ClN_3O$) and an amount of amitriptyline hydrochloride equivalent to NLT 90.0% and NMT 110.0% of the labeled amount of amitriptyline ($C_{20}H_{23}N$).

▲Protect all solutions containing chlordiazepoxide from light.▲ (USP 1-Dec-2022)

IDENTIFICATION

• **A.** The retention times of the major peaks of the *Sample solution* correspond to those of the *Standard solution*, as obtained in the *Assay*.

Add the following:

▲• **B.** The UV spectra of the chlordiazepoxide and amitriptyline peaks of the *Sample solution* correspond to those of the *Standard solution*, as obtained in the *Assay*.▲ (USP 1-Dec-2022)

ASSAY

Change to read:

• PROCEDURE

▲**Solution A:** Transfer 7.0 mL of ammonium hydroxide to an appropriate 1-L flask containing about 500 mL of water. Dilute with water to volume.

Solution B: Acetonitrile

Mobile phase: See *Table 1*.

Table 1

Time (min)	Solution A (%)	Solution B (%)
0.0	65	35
1.2	65	35
25.0	35	65
25.1	65	35
30.0	65	35

Diluent: Acetonitrile and water (50:50)

Standard solution: 50 µg/mL of USP Chlordiazepoxide RS and 141 µg/mL of USP Amitriptyline Hydrochloride RS (equivalent to 125 µg/mL of amitriptyline) in *Diluent*

Sample stock solution: Nominally 0.1 mg/mL of chlordiazepoxide and nominally 0.25 mg/mL of amitriptyline from Tablets (NLT 20) prepared as follows. Finely powder Tablets (NLT 20) and transfer a portion of the powder equivalent to 5 mg of chlordiazepoxide and 12.5 mg of amitriptyline to a 50-mL volumetric flask. Add 80% of the flask volume of acetonitrile, sonicate for about 15 min, and then mechanically shake for about 1 h. Dilute with acetonitrile to volume. Centrifuge a portion of the resulting solution and use the supernatant. [NOTE—A centrifuge speed of 3250 rpm for 10 min may be suitable.]

Sample solution: Nominally 50 µg/mL of chlordiazepoxide and nominally 125 µg/mL of amitriptyline from *Sample stock solution* in water

Chromatographic system
(See *Chromatography ⟨621⟩, System Suitability*.)
Mode: LC
Detector: UV 254 nm. For *Identification B*, use a diode array detector in the range of 200–400 nm.
Column: 2.1-mm × 15-cm; 3.5-µm packing L1

Column temperature: 30°

Flow rate: 0.4 mL/min

Injection volume: 5 µL

System suitability

Sample: *Standard solution*

[Note—The relative retention times for chlordiazepoxide and amitriptyline are 1.0 and 5.5, respectively.]

Suitability requirements

Resolution: NLT 2.0 between chlordiazepoxide and amitriptyline

Tailing factor: NMT 1.5 for chlordiazepoxide; NMT 2.0 for amitriptyline

Relative standard deviation: NMT 1.0% each for chlordiazepoxide and amitriptyline▲ (USP 1-Dec-2022)

Analysis

Samples: *Standard solution* and *Sample solution*

Calculate the percentage of the labeled amount of chlordiazepoxide ($C_{16}H_{14}ClN_3O$) in the portion of Tablets taken:

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

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

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

C_S = concentration of [USP Chlordiazepoxide RS] in the *Standard solution* ▲(µg/mL)▲ (USP 1-Dec-2022)

C_U = nominal concentration of chlordiazepoxide in the *Sample solution* ▲(µg/mL)▲ (USP 1-Dec-2022)

Calculate the percentage of the labeled amount of amitriptyline ($C_{20}H_{23}N$) in the portion of Tablets taken:

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

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

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

C_S = concentration of [USP Amitriptyline Hydrochloride RS] in the *Standard solution* ▲(µg/mL)▲ (USP 1-Dec-2022)

C_U = nominal concentration of amitriptyline in the *Sample solution* ▲(µg/mL)▲ (USP 1-Dec-2022)

M_{r1} = molecular weight of amitriptyline, 277.40

M_{r2} = molecular weight of amitriptyline hydrochloride, 313.87

Acceptance criteria: 90.0%–110.0% each of the labeled amounts of chlordiazepoxide ($C_{16}H_{14}ClN_3O$) and amitriptyline ($C_{20}H_{23}N$)

PERFORMANCE TESTS

Change to read:

• **DISSOLUTION ⟨711⟩**

Medium: [Simulated gastric fluid TS], prepared without [pepsin]; 900 mL

Apparatus 1: 100 rpm

Time: 30 min

Standard solution A: [USP Chlordiazepoxide RS] in *Medium*

Standard solution B: [USP Amitriptyline Hydrochloride RS] in *Medium*

Sample solution: Pass a portion of solution under test through a suitable filter. Dilute with *Medium,* if necessary.

Instrumental conditions

Mode: UV

Analytical wavelengths: 239 and 309 nm

Blank: *Medium*

Analysis

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

Calculate the percentage of the labeled amount of chlordiazepoxide ($C_{16}H_{14}ClN_3O$) in the portion of Tablets ▲dissolved:▲ (USP 1-Dec-2022)

$$\text{Result} = (A_U/A_S) \times C_{SA}▲\times V▲ \text{ (USP 1-Dec-2022)} \times D▲ \text{ (USP 1-Dec-2022)} \times (1/L) \times 100$$

A_U = absorbance from the *Sample solution*, 309 nm

A_S = absorbance from *Standard solution A*, 309 nm

C_{SA} = concentration of [USP Chlordiazepoxide RS](#) in *Standard solution A* (mg/mL)

▲V = volume of *Medium*, 900 mL▲ (USP 1-Dec-2022)

D = dilution factor of the *Sample solution*, if needed

▲▲ (USP 1-Dec-2022)

L = label claim of chlordiazepoxide (mg/Tablet)

Calculate the absorbance of amitriptyline in the *Sample solution* at 239 nm (A_x):

$$\text{Result} = A_{U239} - \{A_{U309} \times [(C_{S309} \times A_{S239})/(C_{S239} \times A_{S309})]\}$$

A_{U239} = absorbance from the *Sample solution*, 239 nm

A_{U309} = absorbance from the *Sample solution*, 309 nm

C_{S309} = concentration of chlordiazepoxide from *Standard solution A* ▲(mg/mL),▲ (USP 1-Dec-2022) 309 nm

A_{S239} = absorbance from *Standard solution A*, 239 nm

C_{S239} = concentration of chlordiazepoxide from *Standard solution A* ▲(mg/mL),▲ (USP 1-Dec-2022) 239 nm

A_{S309} = absorbance from *Standard solution A*, 309 nm

Calculate the percentage of the labeled amount of amitriptyline ($C_{20}H_{23}N$) in the portion of Tablets ▲dissolved:▲ (USP 1-Dec-2022)

$$\text{Result} = (A_X/A_S) \times C_S \blacktriangle \times V_{\blacktriangle \text{ (USP 1-Dec-2022)}} \times D \times (M_{r1}/M_{r2}) \blacktriangle_{\blacktriangle \text{ (USP 1-Dec-2022)}} \times (1/L) \times 100$$

A_X = absorbance from the *Sample solution*, as determined from the previous equation

A_S = absorbance of amitriptyline from *Standard solution B*, 239 nm

C_S = concentration of [USP Amitriptyline Hydrochloride RS](#) in *Standard solution B* (mg/mL)

▲V = volume of *Medium*, 900 mL▲ (USP 1-Dec-2022)

D = dilution factor of the *Sample solution*, if needed

M_{r1} = molecular weight of amitriptyline, 277.40

M_{r2} = molecular weight of amitriptyline hydrochloride, 313.87

▲▲ (USP 1-Dec-2022)

L = label claim of amitriptyline (mg/Tablet)

[Note—All of the chlordiazepoxide measurements may be made with either a single *Standard solution* or two separate *Standard solutions*.]

Tolerances: NLT 85% (*Q*) of the labeled amount of chlordiazepoxide ($C_{16}H_{14}ClN_3O$), and an amount of amitriptyline hydrochloride equivalent to NLT 85% (*Q*) of the labeled amount of amitriptyline ($C_{20}H_{23}N$) are dissolved.

• **U**ɴɪꜰᴏʀᴍɪᴛʏ ᴏꜰ **D**ᴏꜱᴀɢᴇ **U**ɴɪᴛꜱ ⟨905⟩, *Content Uniformity*: Meet the requirements for both chlordiazepoxide and amitriptyline

IMPURITIES

• **O**ʀɢᴀɴɪᴄ **I**ᴍᴘᴜʀɪᴛɪᴇꜱ

Standard solution A: 1 mg/mL of [USP Chlordiazepoxide Related Compound A RS](#) in acetone

Standard solution B: 50 µg/mL of [USP 2-Amino-5-chlorobenzophenone RS](#) in acetone

Sample solution: Transfer a portion of finely powdered Tablets equivalent to 25 mg of chlordiazepoxide to a 10-mL conical flask, add 2.5 mL of acetone, and shake. Allow any undissolved particles to settle, and use the supernatant.

Chromatographic system

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

Mode: TLC

Adsorbent: 0.25-mm layer of chromatographic silica gel

Application volumes

Standard solution A: 20 µL

Standard solution B: 10 µL

Sample solution: 50 µL

Developing solvent system: Ethyl acetate

Spray reagent A: 2 N sulfuric acid

Spray reagent B: 1 mg/mL of sodium nitrite in water

Spray reagent C: 5 mg/mL of ammonium sulfamate in water

Spray reagent D: 1 mg/mL of *N*-(1-naphthyl)ethylenediamine dihydrochloride in water

Analysis

Samples: *Standard solution A, Standard solution B,* and *Sample solution*

Develop the chromatogram in a chromatographic chamber (not previously saturated with the *Developing solvent system*) using the *Developing solvent system* until the solvent front has moved about three-fourths of the length of the plate. Remove the plate from the developing chamber, mark the solvent front, and allow the solvent to evaporate. Locate the spots on the plate by lightly spraying with *Spray reagent A*, drying at 105° for 15 min, and then spraying in succession with *Spray reagent B, Spray reagent C,* and *Spray reagent D.*

Acceptance criteria: Any spots from the *Sample solution* are not greater in size or intensity than the spots at the respective R_F values produced by the *Standard solutions*, corresponding to NMT 4.0% of chlordiazepoxide related compound A, and NMT 0.1% of 2-amino-5-chlorobenzophenone.

ADDITIONAL REQUIREMENTS

Change to read:

• PACKAGING AND STORAGE**:** Preserve in tight, light-resistant containers. ▲Store at controlled room temperature.▲ (USP 1-Dec-2022)

Change to read:

• **USP REFERENCE STANDARDS** 〈11〉

USP 2-Amino-5-chlorobenzophenone RS $C_{13}H_{10}ClNO$ 231.68

USP Amitriptyline Hydrochloride RS

USP Chlordiazepoxide RS

USP Chlordiazepoxide Related Compound A RS

7-Chloro-1,3-dihydro-5-phenyl-2*H*-1,4-benzodiazepin-2-one 4-oxide.

$C_{15}H_{11}ClN_2O_2$ ▲286.71▲ (USP 1-Dec-2022)

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

Topic/Question	Contact	Expert Committee
CHLORDIAZEPOXIDE AND AMITRIPTYLINE HYDROCHLORIDE TABLETS	Documentary Standards Support	SM42020 Small Molecules 4

Chromatographic Database Information: Chromatographic Database

---