Solution A: *Buffer* and acetonitrile (75:25)
Solution B: *Buffer* and acetonitrile (50:50)
Mobile phase: See *Table 1*

Table 1

Time (min)	Solution A (%)	Solution B (%)
0.00	100	0
5.00	100	0
36.25	0	100
38.25	100	0
48.00	100	0

[NOTE—Adjust the start time of the gradient step on the basis of the instrument's dwell volume.]
Diluent: Acetonitrile and *Buffer* (60:40)
System suitability stock solution: Transfer 6 mg of USP Raloxifene Hydrochloride RS to a 50-mL volumetric flask, and add 15.0 mL of acetonitrile, 3.0 mL of water, and 5.0 mL of 30% hydrogen peroxide (unstabilized). Mix, and dissolve the raloxifene hydrochloride. Shake the solution for approximately 30 min, followed by approximately 30 min of sonication. Let it stand at 30° for at least 6 h. Dilute with *Diluent* to 50.0 mL. [NOTE—Raloxifene hydrochloride is partly converted to raloxifene *N*-oxide under these conditions. The reaction time can be varied as necessary to achieve an appropriate level of raloxifene *N*-oxide.]
System suitability solution: Transfer 15 mg of USP Raloxifene Hydrochloride RS to a 50-mL volumetric flask, and add 5.0 mL of *System suitability stock solution* and 20 mL of *Diluent*. Dilute with *Solution A* to volume.
Standard stock solution: 0.06 mg/mL of USP Raloxifene Hydrochloride RS in *Diluent*
Standard solution: Mix 5 mL of the *Standard stock solution* and 45 mL of *Diluent*, and dilute with *Solution A* to 100.0 mL (0.003 mg/mL).
Sample solution: Transfer a sufficient quantity of Tablets to a volumetric flask of a suitable size to obtain a solution of raloxifene hydrochloride having a concentration of 6 mg/mL, based on the label claim. Add *Diluent*, and shake to disintegrate the Tablets. Sonicate, if necessary, and add *Diluent* to volume. Transfer 5 mL of this solution to a 10-mL volumetric flask, and dilute with *Solution A* to volume to obtain a solution having a concentration of 3 mg/mL of raloxifene hydrochloride, based on the label claim. Filter, and use the clear solution.
Chromatographic system
(See *Chromatography* ⟨621⟩*, System Suitability*.)
Mode: LC
Detector: UV 280 nm
Column: 4.6-mm × 25-cm; 5-µm base-deactivated packing L7
Column temperature: 35°
Flow rate: 1 mL/min
Injection size: 10 µL
System suitability
Sample: *System suitability solution*
Suitability requirements
Resolution: NLT 3.0 between raloxifene and raloxifene *N*-oxide
Tailing factor: NMT 2.0 for the raloxifene peak
Analysis
Samples: *Standard solution* and *Sample solution*
Record the chromatograms for NLT two times the retention time of the raloxifene peak, and measure all of the peak responses.
Calculate the percentage of each impurity in the portion of Tablets taken:

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

r_U = peak response of each impurity from the *Sample solution*
r_S = peak response of raloxifene from the *Standard solution*
C_S = concentration of USP Raloxifene Hydrochloride RS in the *Standard solution* (mg/mL)
C_U = nominal concentration of the *Sample solution* (mg/mL)
Acceptance criteria: See *Table 2*.

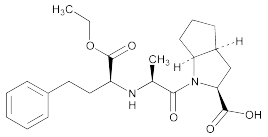
Table 2

Name	Relative Retention Time	Acceptance Criteria, NMT (%)
Raloxifene	1.00	—
Raloxifene *N*-oxide[a]	1.17	0.3
Any unspecified individual impurity	—	0.2
Total impurities	—	1.0

[a] Methanone, [6-hydroxy-2-(4-hydroxyphenyl) benzo[*b*]thien-3-yl][4-[2-(1-oxido-1-piperidinyl)ethoxy]phenyl].

ADDITIONAL REQUIREMENTS
- **PACKAGING AND STORAGE:** Preserve in tight containers, and store at controlled room temperature.
- **USP REFERENCE STANDARDS** ⟨11⟩
  USP Raloxifene Hydrochloride RS

Ramipril



$C_{23}H_{32}N_2O_5$ 416.51
Cyclopenta[*b*]pyrrole-2-carboxylic acid, 1-[2-[[1-(ethoxycarbonyl)-3-phenylpropyl]amino]-1-oxopropyl]octahydro-, [2*S*-[1[*R**(*R**)], 2α,3a*β*,6a*β*]]-.
(2*S*,3a*S*,6a*S*)-1-[(*S*)-*N*-[(*S*)-1-Carboxy-3-phenylpropyl]alanyl] octahydrocyclopenta[*b*]pyrrole-2-carboxylic acid, 1-ethyl ester [87333-19-5].

» Ramipril contains not less than 98.0 percent and not more than 102.0 percent of $C_{23}H_{32}N_2O_5$, calculated on the dried basis.

Packaging and storage—Preserve in tight containers.

USP Reference standards ⟨11⟩**—**
USP Ramipril RS
USP Ramipril Related Compound A RS
(2*S*,3a*S*,6a*S*)-1-[(*S*)2-[[(*S*)1-(Methoxycarbonyl)-3-phenylpropyl]amino]-1-oxopropyl]-octahydrocyclopenta[*b*]pyrrole-2-carboxylic acid.
$C_{22}H_{30}N_2O_5$ 402.48
USP Ramipril Related Compound B RS
(2*S*,3a*S*,6a*S*)-1-[(*S*)2-[[(*S*)1-(Methylethoxy)carbonyl-3-phenylpropyl]amino]-1-oxopropyl]-octahydrocyclopenta[*b*]pyrrole-2-carboxylic acid.
$C_{24}H_{34}N_2O_5$ 430.54
USP Ramipril Related Compound C RS
(2*S*,3a*S*,6a*S*)-1-[(*S*)2-[[(*S*)1-Ethoxycarbonyl-3-cyclohexyl propyl]amino]-1-oxopropyl]-octahydrocyclopenta[*b*]pyrrole-2-carboxylic acid.
$C_{23}H_{38}N_2O_5$ 422.56

USP Ramipril Related Compound D RS
Ethyl (2*S*)2-[(3*S*,5a*S*,8a*S*, 9*aS*)-3-methyl-1,4-dioxodecahydro-1*H*-cyclopenta[*e*]pyrrolo[1,2-*a*]pyrazin-2-yl]-4-phenyl-butanoate.
$C_{23}H_{30}N_2O_4$ 398.50

Identification, *Infrared Absorption* ⟨197K⟩.

Melting range ⟨741⟩: between 105° and 112°.

Specific rotation ⟨781S⟩: between +32.0° and +38.0°, determined at 20°.

Test solution: 10 mg per mL, in 0.1 M methanolic hydrochloric acid.

Loss on drying ⟨731⟩—Dry it in vacuum at a pressure not exceeding 5 mm of mercury at 60° for 6 hours: it loses not more than 0.2% of its weight.

Residue on ignition ⟨281⟩: not more than 0.1%.

Limit of palladium—

Diluent—Prepare a mixture of water and nitric acid (997:3).

Standard stock solution—Transfer about 50 mg of palladium metal, accurately weighed, to a 100-mL volumetric flask, dissolve in 9 mL of hydrochloric acid, and dilute with water to volume.

Standard solutions—Dilute the *Standard stock solution* quantitatively, and stepwise if necessary, with *Diluent* to obtain solutions containing 0.02, 0.03, and 0.05 µg of palladium per mL.

Test solution—Transfer about 200 mg of Ramipril, accurately weighed, to a 100-mL volumetric flask, and dissolve in and dilute with *Diluent* to volume.

Blank solution—Transfer about 150 mg of magnesium nitrate to a 100-mL volumetric flask, and dissolve in and dilute with *Diluent* to volume.

Procedure—Concomitantly determine the absorbances of equal volumes of the *Standard solutions* and the *Test solution* (about 20 µL), at the palladium emission line at 247.6 nm, with a suitable atomic absorption spectrophotometer (see *Spectrophotometry and Light-Scattering* ⟨851⟩) equipped with a palladium hollow-cathode lamp, using a 10-µL injection of *Blank solution* as the blank. Plot the absorbances of the *Standard solutions* versus concentration, in µg per mL, of palladium, and draw the straight line best fitting the three plotted points. From the graph so obtained, determine the concentration, C_P, in µg per mL, of palladium in the *Test solution.* Calculate the percentage of palladium in the portion of Ramipril taken by the formula:

$$0.1C_P / C_R,$$

in which C_R is the concentration, in mg per mL, of Ramipril taken to prepare the *Test solution.* The limit is 0.002%.

Related compounds—

Solution A—Dissolve 2.0 g of sodium perchlorate in a mixture of 800 mL of water and 0.5 mL of triethylamine, adjust with phosphoric acid to a pH of about 3.6 ± 0.1, add 200 mL of acetonitrile, and mix.

Solution B—Dissolve 2.0 g of sodium perchlorate in a mixture of 300 mL of water and 0.5 mL of triethylamine, adjust with phosphoric acid to a pH of about 2.6 ± 0.1, add 700 mL of acetonitrile, and mix.

Mobile phase—Use variable filtered and degassed mixtures of *Solution A* and *Solution B* as directed for *Chromatographic system.* Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Test solution—Transfer about 25 mg of Ramipril, accurately weighed, to a 25-mL volumetric flask, dissolve in and dilute with *Solution A* to volume, and mix. [NOTE—Keep the *Test solution* cold until injected.]

Resolution solution—Dissolve a quantity of USP Ramipril RS, USP Ramipril Related Compound A RS, USP Ramipril Related Compound B RS, USP Ramipril Related Compound C RS, and USP Ramipril Related Compound D RS in *Solution B* to obtain a solution with a concentration of about 0.5 mg of each per mL.

Standard solution—Dissolve an accurately weighed quantity of USP Ramipril RS in *Solution B,* and dilute quantitatively, and stepwise if necessary, with *Solution B* to obtain a solution having a known concentration of about 0.005 mg per mL.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 210-nm detector and a 4.0-mm × 25-cm column that contains 3-µm packing L1, and is maintained at a temperature of 65°. The flow rate is about 1 mL per minute. The chromatograph is programmed as follows.

Time (minutes)	*Solution A* (%)	*Solution B* (%)	Elution
0–6	90	10	isocratic
6–7	90→75	10→25	linear gradient
7–20	75→65	25→35	linear gradient
20–30	65→25	35→75	linear gradient
30–40	25	75	isocratic
40–45	25→90	75→10	linear gradient
45–55	90	10	isocratic

NOTE—Make adjustments at the 75:25 ratio stage, if necessary, to achieve elution of ramipril between 16 and 19 minutes after injection of the *Standard solution.* Chromatograph the *Resolution solution,* and record the peak responses as directed for *Procedure:* the resolution, *R,* between ramipril related compound A and ramipril is not less than 3.0. Similarly chromatograph the *Test solution,* and record the peak responses as directed for *Procedure:* the retention time for ramipril is between 16 and 19 minutes; and the tailing factor for the ramipril peak is between 0.8 and 2.0. Chromatograph the *Standard solution,* and record the peak responses as directed for *Procedure:* the relative standard deviation for replicate injections is not more than 5.0%. [NOTE—The relative retention times are about 0.8 for ramipril related compound A, 1.0 for ramipril, 1.3 for ramipril related compound B, 1.5 for ramipril related compound C, and 1.6 for ramipril related compound D.]

Procedure—Separately inject equal volumes (about 10 µL) of the *Test solution* and the *Standard solution* into the chromatograph, record the chromatograms, and measure the peak response for ramipril obtained from the *Standard solution* and the responses of all the peaks, other than the ramipril peak, obtained from the *Test solution.* Calculate the percentage of each related compound and unknown impurity in the portion of Ramipril taken by the formula:

$$100F(C_S / C_T)(r_i / r_S)$$

in which F is the relative response factor for the related compound, which is 2.4 for ramipril related compound C, and 1.0 for all other individual impurities; C_S is the concentration, in mg per mL, of USP Ramipril RS in the *Standard solution;* C_T is the concentration, in mg per mL, of ramipril in the *Test solution;* r_i is the peak response for each individual peak obtained from the *Test solution;* and r_S is the ramipril peak response obtained from the *Standard solution:* not more than 0.5% of ramipril related compound A, ramipril related compound B, ramipril related compound C, or ramipril related compound D is found; not more than 0.1% of any other individual impurity is found; and not more than 1.0% of total impurities is found.

Assay—

Sodium dodecyl sulfate solution—Prepare a 0.1% solution of sodium dodecyl sulfate. Adjust with phosphoric acid to a pH of 2.4 ± 0.1, filter, and degas.

Mobile phase—Prepare a mixture of *Sodium dodecyl sulfate solution* and acetonitrile (55:45). Adjust with phosphoric acid to a pH of 2.75 ± 0.1, filter, and degas. Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

System suitability preparation—Dissolve accurately weighed quantities of USP Ramipril RS and USP Ramipril Related Compound A RS in *Mobile phase* to obtain a solution having known concentrations of about 0.2 mg per mL and 0.01 mg per mL, respectively.

Standard preparation—Dissolve an accurately weighed quantity of USP Ramipril RS in *Mobile phase* to obtain a solution having a known concentration of about 0.2 mg per mL.

Assay preparation—Transfer about 100 mg of Ramipril, accurately weighed, to a 100-mL volumetric flask, dissolve in about 10 mL of acetonitrile, dilute with *Mobile phase* to volume, and mix. Pipet about 10 mL of this stock solution to a 50-mL volumetric flask, dilute with *Mobile phase* to volume, and mix.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 210-nm detector and a 4.6-mm × 15-cm column that contains packing L1. The flow rate is about 1.8 mL per minute. Chromatograph the *System suitability preparation,* and record the peak responses as directed for *Procedure:* the resolution, *R,* between ramipril and ramipril related compound A is not less than 2.0; the column efficiency determined from the ramipril peak is not less than 4000 theoretical plates; and the relative standard deviation for replicate injections determined from the ramipril peak is not more than 1.0%.

Procedure—Separately inject equal volumes (about 20 µL) of the *Standard preparation* and the *Assay preparation* into the chromatograph, record the chromatograms, and measure the responses for all of the peaks. Calculate the quantity, in mg, of $C_{23}H_{32}N_2O_5$ in the portion of Ramipril taken by the formula:

$$500C(r_U / r_S)$$

in which C is the concentration, in mg per mL, of USP Ramipril RS in the *Standard preparation;* and r_U and r_S are the peak responses obtained from the *Assay preparation* and the *Standard preparation,* respectively.

Ramipril Capsules

DEFINITION

Ramipril Capsules contain NLT 90.0% and NMT 110.0% of the labeled amount of $C_{23}H_{32}N_2O_5$.

IDENTIFICATION

- **A. Ultraviolet Absorption** ⟨197U⟩

  Phosphoric acid solution: 30 mL/L of phosphoric acid in water

  Diluent: Acetonitrile and *Phosphoric acid solution* (2:3)

  Standard solution: 0.2 mg/mL of USP Ramipril RS in *Diluent*. Sonicate for 1 min, if necessary, for complete dissolution.

  Sample solution: Use the *Sample solution* prepared as directed in the *Assay*.

  Wavelength range: 200–400 nm

  Path length: 0.1-cm cell

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

ASSAY

- **Procedure**

  Buffer: Dissolve 17 g of monobasic potassium phosphate and 11.2 g of sodium perchlorate in 750 mL of water in a 1-L flask. Dilute with water to volume. Adjust with phosphoric acid to a pH of 2.3.

  Solution A: Acetonitrile, *Buffer*, and water (1:2:2). [Note—Do not filter *Solution A.*]

  Solution B: Acetonitrile, *Buffer*, and water (9:10:6). [Note—Do not filter *Solution B.*]

  Phosphoric acid solution and **Diluent:** Prepare as directed in *Identification* test *A.*

  Mobile phase: Use the gradient table below.

Time (min)	Solution A (%)	Solution B (%)
0	100	0
5	100	0
50	0	100
51	0	100
51.1	100	0
60	100	0

  Standard solution: 0.2 mg/mL of USP Ramipril RS and 0.002 mg/mL of USP Ramipril Related Compound A RS in *Diluent*

  Sample stock solution: Transfer the contents of 8 Capsules into each of the flasks as described in *Table 1*. Add Capsule shells into the flasks. Add acetonitrile per *Table 1*, and swirl to agitate the contents. Sonicate for 15 min, and mechanically shake for 10 min. Dilute with acetonitrile to volume for Capsule strengths 5.0 and 10 mg only. For 1.25- and 2.5-mg Capsules, use the solution as is without further dilution. [Note—Extracts from the vial cap may result in extraneous peaks.]

Table 1

Strength of Capsule (mg)	Volumetric Flask Size (mL)	Acetonitrile (mL)
1.25	50	25
2.5	100	50
5.0	100	70
10	200	140

  Sample solution: Nominally 0.2 mg/mL of ramipril in *Phosphoric acid solution* from the *Sample stock solution*. Pass through a nylon filter of 0.20-µm pore size, and discard the first 2 mL of filtrate.

  Chromatographic system

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

  Mode: LC

  Detector: UV 215 nm

  Column: 4.6-mm × 15-cm; 5-µm packing L1 with a guard column, packing L1

  Temperature: 60°

  Flow rate: 1.5 mL/min

  Injection size: 50 µL

  System suitability

  Sample: *Standard solution*

  Suitability requirements

  Resolution: NLT 2.5 between ramipril and ramipril related compound A

  Tailing factor: NMT 2.5 for the ramipril peak

  Relative standard deviation: NMT 2.0% for the ramipril peak

  Analysis

  Samples: *Standard solution* and *Sample solution*

  Calculate the percentage of $C_{23}H_{32}N_2O_5$, based on the label claim, in the portion of Capsules taken:

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

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

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

  C_S = concentration of ramipril in the *Standard solution* (mg/mL)

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

  Acceptance criteria: 90.0%–110.0%