each mL of saline TS. Calculate the quantity, in µg, of $C_{11}H_{16}N_2O_2$ in each solution taken by the formula:

$$(208.26 / 244.72)(A_U / A_S)27C$$

in which 208.26 and 244.72 are the molecular weights of pilocarpine and pilocarpine hydrochloride, respectively; A_U and A_S are the absorbances of the test solution and the Standard solution, respectively; and *C* is the concentration, in µg per mL, of USP Pilocarpine Hydrochloride RS in the Standard solution. Calculate the amount of pilocarpine released in 168 hours by adding the pilocarpine content of each set of tubes collected over 168 hours.

Tolerances—The amount of $C_{11}H_{16}N_2O_2$ from each Ocular System released during the total 0 to 168 hours tested conforms to *Acceptance Table 1* under *Drug Release* ⟨724⟩. The drug release range for this time period is not less than 80.0% and not more than 120.0% of the labeled release pattern.

Assay—

Buffer solution, Mobile phase, Standard preparation, System suitability preparation, and *Chromatographic system*—Proceed as directed in the *Assay* under *Pilocarpine*.

Assay preparation—Select not fewer than 10 Ocular Systems. Cut each System into 4 pieces, transfer quantitatively to a 500-mL volumetric flask, and rinse all cutting utensils with 20 to 30 mL of methanol into the flask. Make additional rinses of the utensils with about 250 mL of *Mobile phase*, and collect all the rinses in the flasks. Allow the flasks to stand for 30 minutes, sonicate for about 15 minutes, dilute with water to volume, and mix. Transfer an aliquot of the supernatant, equivalent to 6 mg of pilocarpine to a 200-mL volumetric flask, dilute with water to volume, mix, and filter.

Procedure—Proceed as directed for *Procedure* in the *Assay* under *Pilocarpine*. Calculate the quantity, in mg, of pilocarpine in each Ocular System taken by the formula:

$$(208.26 / 271.27)(10 / V)(C / N)(r_U / r_S)$$

in which 208.26 and 271.27 are the molecular weights of pilocarpine and pilocarpine nitrate, respectively; *V* is the volume, in mL, of the supernatant taken (see *Assay preparation*); *C* is the concentration, in µg per mL, of USP Pilocarpine Nitrate RS in the *Standard preparation; N* is the number of Ocular Systems taken; and r_U and r_S are the peak responses for pilocarpine obtained from the *Assay preparation* and the *Standard preparation,* respectively.

Pilocarpine Hydrochloride

$C_{11}H_{16}N_2O_2 \cdot HCl$ 244.72

2(3*H*)-Furanone, 3-ethyldihydro-4-[(1-methyl-1*H*-imidazol-5-yl)methyl]-, monohydrochloride, (3*S*-*cis*)-;
Pilocarpine monohydrochloride [54-71-7].

DEFINITION

Pilocarpine Hydrochloride contains NLT 98.0% and NMT 102.0% of $C_{11}H_{16}N_2O_2 \cdot HCl$, calculated on the dried basis.

IDENTIFICATION

- **A. Infrared Absorption** ⟨**197M**⟩
- **B. Identification Tests—General,** *Chloride* ⟨**191**⟩**:** Meets the requirements

Sample solution: 50 mg/mL

ASSAY

- **Procedure**

Buffer: 4.4 g/L of dibasic potassium phosphate in water. Adjust with phosphoric acid to a pH of 6.5 ± 0.1.

Mobile phase: Acetonitrile, methanol, and *Buffer* (2:35:63)

Standard solution: 0.5 mg/mL of USP Pilocarpine Hydrochloride RS in water. [Note—Sonicate if necessary.]

System suitability solution: Transfer a known quantity of USP Pilocarpine Hydrochloride RS in a suitable volumetric flask, and add water, equivalent to 10% of the volume of the flask, to dissolve. [Note—Sonicate as needed.] Add 0.1 N sodium hydroxide, equivalent to 10% of the volume of the flask, quench immediately with the same volume of 0.1 N hydrochloric acid, and mix. Dilute with water to volume. [Note—The initial concentration of USP Pilocarpine Hydrochloride RS is 0.5 mg/mL. Isopilocarpine is formed in the *System suitability solution* preparation.]

Sample solution: 0.5 mg/mL of Pilocarpine Hydrochloride in water. [Note—Sonicate if necessary.]

Chromatographic system
(See *Chromatography* ⟨621⟩*, System Suitability*.)
Mode: LC
Detector: UV 215 nm
Column: 4.6-mm × 15-cm; 3-µm packing L11
Column temperature: 35°
Flow rate: 1.0 mL/min
Injection size: 10 µL

System suitability
Samples: *Standard solution* and *System suitability solution*
Suitability requirements
Resolution: NLT 1.5 between isopilocarpine and pilocarpine, *System suitability solution*
Tailing factor: NMT 2.0, *Standard solution*
Relative standard deviation: NMT 2.0%, *Standard solution*

Analysis
Samples: *Standard solution* and *Sample solution*
Calculate the percentage of $C_{11}H_{16}N_2O_2 \cdot HCl$ in the portion of Pilocarpine Hydrochloride taken:

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

r_U = peak area from the *Sample solution*
r_S = peak area from the *Standard solution*
C_S = concentration of USP Pilocarpine Hydrochloride RS in the *Standard solution* (mg/mL)
C_U = concentration of Pilocarpine Hydrochloride in the *Sample solution* (mg/mL)

Acceptance criteria: 98.0%–102.0% on the dried basis

IMPURITIES

Organic Impurities

- **Procedure 1: Related Compounds**

Mobile phase, Standard solution, System suitability solution, and **Sample solution:** Proceed as directed in the *Assay*.
Sensitivity solution: 0.25 µg/mL of USP Pilocarpine Hydrochloride RS in water from the *Standard solution*
Chromatographic system
(See *Chromatography* ⟨621⟩*, System Suitability*.)
Mode: LC
Detector: UV 215 nm
Column: 4.6-mm × 15-cm; 3-µm packing L11
Column temperature: 35°
Flow rate: 1.0 mL/min
Run time: NLT 5 times the retention time of the pilocarpine peak
Injection size: 10 µL

System suitability
Samples: *Standard solution*, *System suitability solution*, and *Sensitivity solution*
Suitability requirements
Resolution: NLT 1.5 between isopilocarpine and pilocarpine, *System suitability solution*
Signal-to-noise ratio: NLT 10 for the pilocarpine peak, *Sensitivity solution*
Relative standard deviation: NMT 2.0% for the pilocarpine peak, *Standard solution*

Analysis
Samples: *Standard solution* and *Sample solution*
Calculate the percentage of each impurity in the portion of Pilocarpine Hydrochloride taken:

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

r_U = peak area of each individual impurity from the *Sample solution*
r_S = peak area of pilocarpine from the *Standard solution*
C_S = concentration of USP Pilocarpine Hydrochloride RS in the *Standard solution* (mg/mL)
C_U = concentration of Pilocarpine Hydrochloride in the *Sample solution* (mg/mL)

Acceptance criteria
Individual impurities: See *Impurity Table 1*. [NOTE—Disregard any unspecified impurity peaks less than 0.05%.]
Total impurities: NMT 1.0%

Impurity Table 1

Name	Relative Retention Time	Acceptance Criteria, NMT (%)
Isopilocarpine[a]	0.94	1.0
Pilocarpine	1.00	—
Pilocarpic acid[b]	1.15	0.5
Isopilocarpic acid[c]	1.19	0.1
Any unspecified impurity	—	0.1

[a] (3*R*,4*R*)-3-Ethyl-4-[(1-methyl-1*H*-imidazol-5-yl)methyl]dihydrofuran-2(3*H*)-one.
[b] (2*S*,3*R*)-2-Ethyl-4-hydroxy-3-[(1-methyl-1*H*-imidazol-5-yl)methyl]butanoic acid.
[c] (2*R*,3*R*)-2-Ethyl-4-hydroxy-3-[(1-methyl-1*H*-imidazol-5-yl)methyl]butanoic acid.

- **PROCEDURE 2: OTHER ALKALOIDS**
  Sample solution: 10 mg/mL in water
  Analysis: Divide the *Sample solution* into two portions. To one portion add a few drops of 6 N ammonium hydroxide, and to the other, add a few drops of potassium dichromate TS.
  Acceptance criteria: No turbidity is produced in either solution.

SPECIFIC TESTS
- **OPTICAL ROTATION,** *Specific Rotation* ⟨**781S**⟩**:** +88.5° to +91.5°
  Sample solution: 20 mg/mL, in water
- **LOSS ON DRYING** ⟨**731**⟩**:** Dry a sample at 105° for 2 h: it loses NMT 3.0% of its weight.
- **READILY CARBONIZABLE SUBSTANCES TEST** ⟨**271**⟩
  Sample solution: 50 mg/mL in sulfuric acid
  Acceptance criteria: The solution has no more color than *Matching Fluid B*.

ADDITIONAL REQUIREMENTS
- **PACKAGING AND STORAGE:** Preserve in tight, light-resistant containers. Store at room temperature.
- **USP REFERENCE STANDARDS** ⟨**11**⟩
  USP Pilocarpine Hydrochloride RS

Pilocarpine Hydrochloride Ophthalmic Solution

DEFINITION
Pilocarpine Hydrochloride Ophthalmic Solution is a sterile, buffered, aqueous solution of pilocarpine hydrochloride. It contains NLT 90.0% and NMT 110.0% of the labeled amount of pilocarpine hydrochloride ($C_{11}H_{16}N_2O_2 \cdot HCl$). It may contain suitable antimicrobial agents and stabilizers, and suitable additives to increase its viscosity.

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*.

ASSAY
- **PROCEDURE**
  Solution A: Ammonium hydroxide in isopropyl alcohol (1 in 50)
  Mobile phase: *n*-Hexane and *Solution A* (700:300)
  Standard solution: 1.6 mg/mL of USP Pilocarpine Hydrochloride RS in water
  Sample solution: Nominally equivalent to 1.6 mg/mL of pilocarpine hydrochloride from a volume of Ophthalmic Solution diluted with methanol
  Chromatographic system
  (See *Chromatography* ⟨621⟩, *System Suitability*.)
  Mode: LC
  Detector: UV 220 nm
  Column: 4.6-mm × 25-cm; packing L3
  Flow rate: 2 mL/min
  Injection size: 10 µL
  System suitability
  Sample: *Standard solution*
  [NOTE—The retention time for pilocarpine hydrochloride is 16 min.]
  Suitability requirements
  Relative standard deviation: NMT 2.0% for pilocarpine hydrochloride for three replicate injections
  Analysis
  Samples: *Standard solution* and *Sample solution*
  Calculate the percentage of the labeled amount of pilocarpine hydrochloride ($C_{11}H_{16}N_2O_2 \cdot HCl$) in each mL of Ophthalmic Solution taken:

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

r_U = peak response from the *Sample solution*
r_S = peak response from the *Standard solution*
C_S = concentration of USP Pilocarpine Hydrochloride RS in the *Standard solution* (mg/mL)
C_U = nominal concentration of the *Sample solution* (mg/mL)

Acceptance criteria: 90.0%–110.0%

SPECIFIC TESTS
- **STERILITY TESTS** ⟨**71**⟩**:** Meets the requirements
- **pH** ⟨**791**⟩**:** 3.5–5.5

ADDITIONAL REQUIREMENTS
- **PACKAGING AND STORAGE:** Preserve in tight containers.
- **USP REFERENCE STANDARDS** ⟨**11**⟩
  USP Pilocarpine Hydrochloride RS

Pilocarpine Hydrochloride Tablets

DEFINITION
Pilocarpine Hydrochloride Tablets contain NLT 90.0% and NMT 110.0% of the labeled amount of pilocarpine hydrochloride ($C_{11}H_{16}N_2O_2 \cdot HCl$).

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