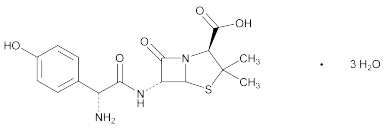
Amoxicillin



$C_{16}H_{19}N_3O_5S \cdot 3H_2O$ 419.45
4-Thia-1-azabicyclo[3.2.0]heptane-2-carboxylic acid, 6-[[amino(4-hydroxyphenyl)acetyl]amino]-3,3-dimethyl-7-oxo-, trihydrate [2*S*-[2α,5α,6β(*S**)]]-;
(2*S*,5*R*,6*R*)-6-[(*R*)-(–)-2-Amino-2-(*p*-hydroxyphenyl)-acetamido]-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxylic acid trihydrate [61336-70-7].
Anhydrous 365.41
[26787-78-0].

DEFINITION

Amoxicillin contains NLT 900 µg and NMT 1050 µg of $C_{16}H_{19}N_3O_5S$ per mg, calculated on the anhydrous basis.

IDENTIFICATION

• **INFRARED ABSORPTION** ⟨197K⟩

ASSAY

• **PROCEDURE**

Diluent: 6.8 g/L of monobasic potassium phosphate in water. Adjust with a 45% (w/w) solution of potassium hydroxide to a pH of 5.0 ± 0.1.
Mobile phase: Acetonitrile and *Diluent* (1:24)
Standard solution: 1.2 mg/mL of USP Amoxicillin RS in *Diluent*. [NOTE—Use this solution within 6 h.]
Sample solution: 1.2 mg/mL of Amoxicillin in *Diluent*. [NOTE—Use this solution within 6 h.]
Chromatographic system
(See *Chromatography* ⟨621⟩, *System Suitability*.)
Mode: LC
Detector: UV 230 nm
Column: 4-mm × 25-cm; packing L1
Flow rate: 1.5 mL/min
Injection size: 10 µL
System suitability
Sample: *Standard solution*
Suitability requirements
Tailing factor: NMT 2.5
Relative standard deviation: NMT 2.0%
Analysis
Samples: *Standard solution* and *Sample solution*
Calculate the quantity, in µg/mg, of $C_{16}H_{19}N_3O_5S$ in the portion of Amoxicillin taken:

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

r_U = peak response from the *Sample solution*
r_S = peak response from the *Standard solution*
C_S = concentration of USP Amoxicillin RS in the *Standard solution* (mg/mL)
C_U = concentration of *Sample solution* (mg/mL)
P = potency of amoxicillin in USP Amoxicillin RS (µg/mg)

Acceptance criteria: 900–1050 µg of $C_{16}H_{19}N_3O_5S$ per mg on the anhydrous basis

IMPURITIES

Organic Impurities

• **PROCEDURE**

Solution A: 2.72 g/L of monobasic potassium phosphate. Adjust with 1 N potassium hydroxide or 20% phosphoric acid to a pH of 5.0 ± 0.1.
Solution B: Methanol
Mobile phase: See the gradient table below.

Time (min)	Solution A (%)	Solution B (%)
0	97	3
10	97	3
22	75	25
26	97	3

Standard solution: 12.5 µg/mL of USP Amoxicillin RS in *Solution A*
System suitability solution: 12.5 µg/mL each of USP Amoxicillin Related Compound A RS and USP Amoxicillin Related Compound D RS in *Solution A*
Sample solution: 1.25 mg/mL of Amoxicillin in *Solution A*. [NOTE—Store this solution at 4° and use within 4 h.]
Chromatographic system
(See *Chromatography* ⟨621⟩, *System Suitability*.)
Mode: LC
Detector: UV 210 nm
Column: 4.6-mm × 10-cm; 5-µm packing L1
Column temperature: 40°
Flow rate: 1.5 mL/min
Injection size: 10 µL
Autosampler temperature: 4°
System suitability
Samples: *Standard solution* and *System suitability solution*
Suitability requirements
[NOTE— Identify peaks by the relative retention times in *Impurity Table 1*.]
Resolution: NLT 1.5 between amoxicillin related compound A and the second peak for amoxicillin related compound D, *System suitability solution*
Relative standard deviation: NMT 10%, *Standard solution*
Analysis
Samples: *Standard solution* and *Sample solution*
Calculate the percentage of each impurity in the portion of Amoxicillin taken:

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

r_U = peak response of each impurity from the *Sample solution*
r_S = peak response of amoxicillin from the *Standard solution*
C_S = concentration of USP Amoxicillin RS in the *Standard solution* (µg/mL)
C_U = nominal concentration of Amoxicillin in the *Sample solution* (mg/mL)
F = unit conversion factor (0.001 mg/µg)

Acceptance criteria
[NOTE—The reporting limit is 0.03% of the amoxicillin peak from the *Standard solution*.]
Individual impurities: See *Impurity Table 1*.
Total impurities: NMT 5.0%

Impurity Table 1

Name	Relative Retention Time	Acceptance Criteria, NMT (%)
Amoxicillin related compound I[a] (D-hydroxyphenylglycine)	0.32	1.0
Amoxicillin related compound D[b,c] (amoxicillin open ring)	0.53	1.0
	0.68	1.0
Amoxicillin related compound A[d] (6-aminopenicillanic acid)	0.78	0.5
Amoxicillin related compound B[e,f] (L-amoxicillin)	0.87	—
Amoxicillin	1.0	—
Amoxicillin related compound G[g] (D-hydroxyphenylglycylamoxicillin)	2.9	1.0
Amoxicillin related compound E[h,i] (amoxicillin penilloic derivative)	4.5	1.0
Amoxicillin related compound M[j] (*N*-(penicillan-6-yl) open ring amoxicillinamide)	6.0	1.0
Amoxicillin related compound F[e,k] (phenylpyrazinediol)	6.3	—
Amoxicillin related compound C[l] (amoxicillin rearrangement product)	6.4	1.0
Amoxicillin related compound E[h,i] (amoxicillin penilloic derivative)	6.7	1.0
Amoxicillin related compound J[m] (amoxicillin open ring dimer)	8.8	1.0
Amoxicillin related compound L[n] (*N*-(penicillan-6-yl) amoxicillinamide)	9.0	1.0
Any unspecified individual impurity	—	1.0

[a] (*R*)-2-Amino-2-(4-hydroxyphenyl)acetic acid.

[b] The chromatographic system resolves two penicilloic acids from each other.

[c] (4*S*)-2-{[(*R*)-2-Amino-2-(4-hydroxyphenyl)acetamido](carboxy)methyl}-5,5-dimethylthiazolidine-4-carboxylic acid.

[d] (2*S*,5*R*,6*R*)-6-Amino-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]-heptane-2-carboxylic acid.

[e] These compounds are listed for information only and are not to be reported.

[f] (2*S*,5*R*,6*R*)-6-[(*S*)-2-Amino-2-(4-hydroxyphenyl)acetamido]-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxylic acid.

[g] (2*S*,5*R*,6*R*)-6-{(*R*)-2-[(*R*)-2-Amino-2-(4-hydroxyphenyl)acetamido]-2-(4-hydroxyphenyl)acetamido}-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]-heptane-2-carboxylic acid.

[h] The chromatographic system resolves two penilloic acids from each other.

[i] (4*S*)-2-{[(*R*)-2-Amino-2-(4-hydroxyphenyl)acetamido]methyl}-5,5-dimethylthiazolidine-4-carboxylic acid.

[j] (2*S*,5*R*,6*R*)-6-(2-[(*R*)-2-Amino-2-(4-hydroxyphenyl)acetamido]-2-((4*S*)-4-carboxy-5,5-dimethylthiazolidin-2-yl)acetamido)-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxylic acid.

[k] 3-(4-Hydroxyphenyl)pyrazin-2-ol.

[l] (4*S*)-2-[5-(4-Hydroxyphenyl)-3,6-dioxopiperazin-2-yl]-5,5-dimethylthiazolidine-4-carboxylic acid.

[m] (2*S*,5*R*,6*R*)-6-((2*R*)-2-{2-[(*R*)-2-Amino-2-(4-hydroxyphenyl)acetamido]-2-[(4*S*)-4-carboxy-5,5-dimethylthiazolidin-2-yl]acetamido}-2-(4-hydroxyphenyl)acetamido)-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]-heptane-2-carboxylic acid.

[n] (2*S*,5*R*,6*R*)-6-{(2*S*,5*R*,6*R*)-6-[(*R*)-2-Amino-2-(4-hydroxyphenyl)acetamido]-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxamido}-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxylic acid.

SPECIFIC TESTS

- **CRYSTALLINITY** ⟨**695**⟩**:** Meets the requirements
- **DIMETHYLANILINE** ⟨**223**⟩**:** Meets the requirement
- **pH** ⟨**791**⟩**:** 3.5–6.0
  Sample solution: 2 mg/mL
- **WATER DETERMINATION,** *Method I* ⟨**921**⟩**:** 11.5%–14.5%
- **STERILITY TESTS** ⟨**71**⟩**:** Where the label states that Amoxicillin is sterile, it meets the requirements when tested as directed in *Test for Sterility of the Product to Be Examined, Direct Inoculation of the Culture Medium,* except to use Fluid Thioglycollate Medium containing polysorbate 80 solution (5 mg/mL) and an amount of sterile penicillinase sufficient to inactivate the amoxicillin in each tube, to use Soybean–Casein Digest Medium containing polysorbate 80 solution (5 mg/mL) and an amount of sterile penicillinase sufficient to inactivate the amoxicillin in each tube, and to shake the tubes once daily.
- **BACTERIAL ENDOTOXINS TEST** ⟨**85**⟩**:** Where the label states that Amoxicillin is sterile or Amoxicillin must be subjected to further processing during the preparation of injectable dosage forms, it contains NMT 0.25 USP Endotoxin Unit/mg of amoxicillin.

ADDITIONAL REQUIREMENTS

- **PACKAGING AND STORAGE:** Preserve in tight containers, and store at controlled room temperature.
- **LABELING:** Where it is intended for use in preparing injectable dosage forms, the label states that it is intended for veterinary use only and that it is sterile or must be subjected to further processing during the preparation of injectable dosage forms. Label all other Amoxicillin to indicate that it is to be used in the manufacture of nonparenteral drugs only.
- **USP REFERENCE STANDARDS** ⟨**11**⟩
  USP Amoxicillin RS
  USP Amoxicillin Related Compound A RS
  (2*S*,5*R*,6*R*)-6-Amino-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxylic acid; 6-aminopenicillanic acid.
  $C_8H_{12}N_2O_3S$ 216.26
  USP Amoxicillin Related Compound D RS
  (4*S*)-2-{[(*R*)-2-Amino-2-(4-hydroxyphenyl)acetamido](carboxy)methyl}-5,5-dimethylthiazolidine-4-carboxylic acid; amoxicillin open ring.
  $C_{16}H_{21}N_3O_6S$ 383.42
  USP Endotoxin RS

Amoxicillin Boluses

» Amoxicillin Boluses contain not less than 90.0 percent and not more than 110.0 percent of the labeled amount of amoxicillin ($C_{16}H_{19}N_3O_5S$).

Packaging and storage—Preserve in tight containers, and store at controlled room temperature.

Labeling—Label Boluses to indicate that they are for veterinary use only.

USP Reference standards ⟨11⟩**—**
USP Amoxicillin RS

Identification—

Test solution—To a portion of powdered Boluses add 0.1 N hydrochloric acid to obtain a *Test solution* containing about 4 mg of amoxicillin per mL. Use within 10 minutes after preparation.

Application volume, Developing solvent system, Procedure—Proceed as directed for the *Identification* test under *Amoxicillin Tablets.*

Disintegration ⟨701⟩: 30 minutes, simulated gastric fluid being used instead of water.