Amoxicillin Tablets

Type of Posting	Revision Bulletin, Postponement
Posting Date	27–Apr–2018
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Expert Committee	Chemical Medicines Monographs 1
Reason for Revision	Compliance

In accordance with the Rules and Procedures of the 2015–2020 Council of Experts, the Chemical Medicines Monographs 1 Expert Committee has postponed the test for *Organic Impurities* in the Amoxicillin Tablets monograph. This postponement also necessitates postponing the additions of USP Amoxicillin Related Compound C RS and USP Amoxicillin Related Compound H RS to the *USP Reference Standards* section.

USP has received comments concerning the relative retention time of one of the impurities, and limits for several impurities are tighter than approved limits. Additional information will be required in order to resolve the concerns that have been raised. Interested parties are invited to contact USP for additional information on this topic and to get involved in the revision process. The process for and timing of the revision will be determined following additional considerations by the Expert Committee and USP staff.

The Amoxicillin Tablets Revision Bulletin supersedes the monograph becoming official in *USP 41–NF 36*.

Should you have any questions, please contact Ramanujam Prasad, Senior Scientific Liaison (301-816-8211 or rsp@usp.org).

Amoxicillin Tablets

DEFINITION

Amoxicillin Tablets contain NLT 90.0% and NMT 120.0% of the labeled amount of amoxicillin ($C_{16}H_{19}N_3O_5S$).

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

Change to read:

• **PROCEDURE**

Buffer: 6.8 g/L of monobasic potassium phosphate in water. Adjust with 45% potassium hydroxide TS to a pH of 5.0 ± 0.1.

Mobile phase: Acetonitrile and *Buffer* (1:24)

Standard solution: 1.2 mg/mL of USP Amoxicillin RS in *Buffer*. [NOTE—Use this solution within 6 h.]

Sample solution: Place NLT 5 Tablets in a high-speed glass blender jar containing *Buffer* sufficient to yield a concentration of 1 mg/mL of anhydrous amoxicillin. Blend for 4 ± 1 min, allow to stand for 5 min, and centrifuge a portion of the mixture. [NOTE—Where the volume of *Buffer* required would exceed 500 mL, place 5 Tablets in a volumetric flask of such capacity that when finally diluted to volume, a concentration of 1 mg of anhydrous amoxicillin per milliliter would be obtained. Add a volume of *Buffer* equivalent to three-fourths of the capacity of the volumetric flask, and sonicate for 5 min. Dilute with *Buffer* to volume, add a magnetic stirring bar, and stir for 30 min. Centrifuge a portion of this solution.]

Pass a portion of the clear supernatant through a suitable filter. [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; 10-µm packing L1

Flow rate: 1.5 mL/min

Injection volume: 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 percentage of ▲the labeled amount of▲ USP41 amoxicillin ($C_{16}H_{19}N_3O_5S$) in ▲the portion of Tablets▲ USP41 taken:

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

r_U = peak response of amoxicillin 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* (mg/mL)

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

P = potency of amoxicillin in USP Amoxicillin RS (µg/mg)

F = conversion factor, 0.001 mg/µg

Acceptance criteria: 90.0%–120.0%

PERFORMANCE TESTS

Change to read:

• **DISSOLUTION** ⟨**711**⟩

Medium: Water; 900 mL

Apparatus 2: 75 rpm

Time: 30 min

Determine the amount of amoxicillin ($C_{16}H_{19}N_3O_5S$) dissolved by using the following method.

Buffer: 27.2 g of monobasic potassium phosphate in 3 L of water. Adjust with 45% potassium hydroxide TS to a pH of 5.0 ± 0.1. Dilute with water to obtain 4 L of solution.

Mobile phase: Acetonitrile and *Buffer* (1:39)

Standard solution: 0.05 mg/mL of USP Amoxicillin RS in *Buffer*. [NOTE—Use this solution within 6 h.]

Sample solution: Pass a portion of the sample through a suitable filter of 0.5-µm pore size. Quantitatively dilute a volume of the filtrate with water to obtain an estimated concentration of 0.045 mg/mL of amoxicillin. Use this solution within 6 h.

Chromatographic system

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

Mode: LC

Detector: UV 230 nm

Columns

Guard: 2-mm × 2-cm; packing L2

Analytical: 3.9-mm × 30-cm; packing L1

Column temperature: 40 ± 1°

Flow rate: 0.7 mL/min

Injection volume: 10 µL

System suitability

Sample: *Standard solution*

Suitability requirements

▲▲ USP41

Tailing factor: NMT 2.5

Relative standard deviation: NMT 1.5%

Analysis

Samples: *Standard solution* and *Sample solution*

Calculate the percentage of ▲the labeled amount of▲ USP41 amoxicillin ($C_{16}H_{19}N_3O_5S$) dissolved:

$$\text{Result} = (r_U/r_S) \times (C_S/L) \times V \times D \times P \times F \times 100$$

r_U = peak response of amoxicillin 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* (mg/mL)

L = label claim (mg/Tablet)

V = volume of the dissolution medium, 900 mL

D = dilution factor for the *Sample solution*

P = potency of amoxicillin in USP Amoxicillin RS (µg/mg)

F = conversion factor, 0.001 mg/µg

Tolerances: NLT 75% (Q) of the labeled amount of amoxicillin ($C_{16}H_{19}N_3O_5S$) is dissolved.

For products labeled as chewable Tablets: Proceed as directed above.

For chewable Tablets labeled to contain 200 or 400 mg

Time: 20 min

Tolerances: NLT 70% (Q) of the labeled amount of amoxicillin ($C_{16}H_{19}N_3O_5S$) is dissolved.

For chewable Tablets labeled to contain 125 or 250 mg
Time: 90 min
Tolerances: NLT 70% (*Q*) of the labeled amount of amoxicillin ($C_{16}H_{19}N_3O_5S$) is dissolved.
For veterinary products: Proceed as directed above, except use *Apparatus 2* at 100 rpm.

IMPURITIES

Change to read:

▲• **ORGANIC IMPURITIES**
Solution A: Dissolve 6.8 g/L of monobasic potassium phosphate in water. Adjust with a 20% (w/v) solution of sodium hydroxide to a pH of 5.0 ± 0.1.
Solution B: Acetonitrile
Mobile phase: See *Table 1*.

Table 1

Time (min)	Solution A (%)	Solution B (%)
0	100	0
5	100	0
25	94	6
40	84	16
50	84	16
51	100	0
60	100	0

Impurity stock solution: 0.15 mg/mL each of USP Amoxicillin Related Compound C RS and USP Amoxicillin Related Compound H RS in *Solution A* prepared as follows. Transfer a weighed amount of USP Amoxicillin Related Compound C RS and USP Amoxicillin Related Compound H RS to a suitable volumetric flask. Add acetonitrile to fill 10% of the flask volume and *Solution A* to fill 60% of the flask volume. Sonicate to dissolve and dilute with *Solution A* to volume.
System suitability solution: 1.5 mg/mL of USP Amoxicillin RS and 0.015 mg/mL each of USP Amoxicillin Related Compound C RS and USP Amoxicillin Related Compound H RS in *Solution A* prepared as follows. Transfer a weighed amount of USP Amoxicillin RS to a suitable volumetric flask. Add *Solution A* to fill 60% of the flask volume. Add an appropriate volume of *Impurity stock solution* to the volumetric flask. Sonicate to dissolve and dilute with *Solution A* to volume.
Standard solution: 0.017 mg/mL of USP Amoxicillin RS in *Solution A*. Sonicate if necessary to dissolve. Use this solution immediately after preparation.
Sample solution: Nominally 1.5 mg/mL of amoxicillin in *Solution A* from the powdered Tablets prepared as follows. Transfer powdered Tablets equivalent to 75 mg of amoxicillin into a 50-mL volumetric flask. Add *Solution A* to fill 60% of the final flask volume. Sonicate for 15 min and dilute with *Solution A* to volume. Pass through a suitable filter of 0.45-µm pore size. Use this solution immediately after preparation.
Chromatographic system
(See *Chromatography* ⟨621⟩, *System Suitability*.)
Mode: LC
Detector: UV 230 nm
Column: 4.6-mm × 15-cm; 5-µm packing L7
Column temperature: 40°
Flow rate: 2 mL/min
Injection volume: 20 µL
System suitability
Samples: *System suitability solution* and *Standard solution*
[NOTE—See *Table 2* for relative retention times.]
Suitability requirements
Resolution: NLT 1.5 between amoxicillin related compound C and amoxicillin related compound H, *System suitability solution*
Relative standard deviation: NMT 5.0%, *Standard solution*
Analysis
Samples: *Standard solution* and *Sample solution*
Calculate the percentage of each degradation product in the portion of Tablets taken:

$$\text{Result} = (r_U/r_S) \times (C_S/C_U) \times P \times (F_1/F_2) \times 100$$

r_U = peak response of each degradation product 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* (mg/mL)
C_U = nominal concentration of amoxicillin in the *Sample solution* (mg/mL)
P = potency of amoxicillin in USP Amoxicillin RS (µg/mg)
F_1 = conversion factor, 0.001 mg/µg
F_2 = relative response factor (see *Table 2*)

Acceptance criteria: See *Table 2*. Disregard any peak less than 0.05%.

Table 2

Name	Relative Retention Time	Relative Response Factor	Acceptance Criteria, NMT (%)
Amoxicillin related compound I[a, b] (D-hydroxyphenylglycine)	0.19	—	—
Amoxicillin related compound D[c, d] (amoxicillin open ring)	0.36, 0.47	0.8	2.0
Amoxicillin related compound A[a, e] (6-aminopenicillanic acid)	0.66	—	—
Amoxicillin related compound B[a, f] (L-amoxicillin)	0.82	—	—
Amoxicillin	1.0	1.0	—
Amoxicillin related compound E[d, g]	2.5, 3.32	1.0	5.0
Amoxicillin related compound G[a, h] (D-hydroxyphenylglycyla-moxicillin)	3.03	—	—
Amoxicillin related compound C[i] (amoxicillin rearrangement product)	3.63, 3.84	1.0	1.0
Amoxicillin related compound H[a, j] (*N*-pivaloyl pHPG)	4.03	—	—
Amoxicillin related compound F[k] (pyrazine-2-ol)	4.12	1.1	1.0

Table 2 *(continued)*

Name	Relative Retention Time	Relative Response Factor	Acceptance Criteria, NMT (%)
Amoxicillin related compound K[d, l] (amoxicilloic acid dimers 1 and 2)	4.39, 4.75	0.64	1.0
6-APA amoxicillin amide[a, m]	6.24	—	—
Amoxicilloic amoxilloic acid dimers 1, 2, 3, and 4[d]	6.18, 6.40, 6.56	0.46	1.0
Amoxicillin related compound J[n] (amoxicillin open ring dimer)	7.02	0.64	2.5
N-Pivaloyl amoxicillin	7.96	—	—
Any individual unspecified degradation product	—	—	1.0
Total impurities	—	—	10.0

[a] These are process impurities that are controlled in the drug substance. They are listed here for reference only and are not to be reported.
[b] (*R*)-2-Amino-2-(4-hydroxyphenyl)acetic acid.
[c] (4*S*)-2-{[(*R*)-2-Amino-2-(4-hydroxyphenyl)acetamido](carboxy)methyl}-5,5-dimethylthiazolidine-4-carboxylic acid.
[d] Some chromatographic systems may resolve the peaks from isomers, and the limit is for the sum of all the isomers.
[e] (2*S*,5*R*,6*R*)-6-Amino-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxylic acid.
[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] (4*S*)-2-{[(*R*)-2-Amino-2-(4-hydroxyphenyl)acetamido]methyl}-5,5-dimethylthiazolidine-4-carboxylic acid and (4*R*)-2-{[(*S*)-2-amino-2-(4-hydroxyphenyl)acetamido]methyl}-5,5-dimethylthiazolidine-4-carboxylic acid.
[h] (2*S*,5*R*,6*R*)-6-{(*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.
[i] (4*S*)-2-[5-(4-Hydroxyphenyl)-3,6-dioxopiperazin-2-yl]-5,5-dimethylthiazolidine-4-carboxylic acid.
[j] (*R*)-2-(4-Hydroxyphenyl)-2-pivalamidoacetic acid.
[k] 3-(4-Hydroxyphenyl) pyrazin-2-ol.
[l] Oligomers of penicilloic acids of amoxicillin.
[m] (2*S*,5*R*,6*R*)-6-[[(2*S*,5*R*,6*R*)-6-[(2*R*)-2-Amino-2-(4-hydroxyphenyl) acetamido]-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carbonyl] amino]-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxylic acid.
[n] Co-oligomers of amoxicillin and penicilloic acids of amoxicillin.▲ USP41

▲(Postponed on 1-May-2018)▲ (RB 1-May-2018)

SPECIFIC TESTS

- **MICROBIAL ENUMERATION TESTS ⟨61⟩** and **TESTS FOR SPECIFIED MICROORGANISMS ⟨62⟩:** The total aerobic microbial count does not exceed 10^3 cfu/g, and the total combined molds and yeasts count does not exceed 10^2 cfu/g.

ADDITIONAL REQUIREMENTS

- **PACKAGING AND STORAGE:** Preserve in tight containers, and store at controlled room temperature.
- **LABELING:** Label chewable Tablets to indicate that they are to be chewed before swallowing. Tablets intended solely for veterinary use are so labeled.

Change to read:

- **USP REFERENCE STANDARDS ⟨11⟩**

USP Amoxicillin RS

▲USP Amoxicillin Related Compound C RS
(4*S*)-2-[5-(4-Hydroxyphenyl)-3,6-dioxopiperazin-2-yl]-5,5-dimethylthiazolidine-4-carboxylic acid.
$C_{16}H_{19}N_3O_5S$ 365.40

USP Amoxicillin Related Compound H RS
(*R*)-2-(4-Hydroxyphenyl)-2-pivalamidoacetic acid.
$C_{13}H_{17}NO_4$ 251.28▲ USP41

▲(Postponed on 1-May-2018)▲ (RB 1-May-2018)