

BRIEFING

**Abacavir Sulfate.** The revision of this monograph was posted on the USP Pending Monographs Web page as a draft USP Pending Monograph on August 28, 2009. The MD-AA Expert Committee reviewed the following comment and approved the monograph as an Authorized USP Pending Monograph.

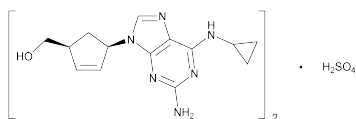
**Comment:** The chemical name of abacavir related compound A in *Impurity Table 2* was corrected from 4-(2,6-Diamino-1*H*-purin-9-yl)cyclopent-2-ene-yl-methanol to 4-(2,6-Diamino-9*H*-purin-9-yl)cyclopent-2-ene-yl-methanol.

This flexible monograph indicates that manufacturers could perform either the tests for *Organic Impurities, Procedure 1* or *Organic Impurities, Procedure 2* based on the impurity profile. The HPLC procedure in the *Assay* is based on analysis performed with the Symmetry C8 brand of column containing 5- $\mu$ m packing L7. The typical retention time for abacavir in the *Assay* procedure is 4.6 min. The HPLC procedure in the test for *Organic Impurities, Procedure 1* is based on analysis performed with the Hypersil BDS C8 brand of L7 column. The typical retention time for abacavir in *Organic Impurities, Procedure 1* is 17.7 min. The HPLC procedure in the test for *Organic Impurities, Procedure 2* is based on analysis performed using the Inertsil ODS 3V brand of L1 column. The typical retention time for abacavir in the test for *Organic Impurities, Procedure 2* is 13 min. The HPLC procedure in the test for *Limit of the Abacavir Enantiomer* is based on analysis performed using the Chiralpak-AD brand of L51 column.

(MD-AA: L. Santos, B. Davani.)      RTS—C46972;    C53869

**Abacavir Sulfate**

v.2 Authorized April 1, 2010



(C<sub>14</sub>H<sub>18</sub>N<sub>6</sub>O)<sub>2</sub> · H<sub>2</sub>SO<sub>4</sub>      670.74  
2-Cyclopentene-1-methanol, 4-[2-amino-6-(cyclopropylamino)-9*H*-purin-9-yl]-, (1*S*,4*R*)-, sulfate (2:1) (salt)  
(1*S*,4*R*)-4-[2-Amino-6-(cyclopropylamino)-9*H*-purin-9-yl]-2-cyclopentene-1-methanol sulfate (2:1) [188062-50-2].

**DEFINITION**

Abacavir Sulfate contains NLT 98.0% and NMT 102.0% of (C<sub>14</sub>H<sub>18</sub>N<sub>6</sub>O)<sub>2</sub> · H<sub>2</sub>SO<sub>4</sub>, calculated on the anhydrous basis.

**IDENTIFICATION**

- **A. INFRARED ABSORPTION (197K)**
- **B.** The retention time of the abacavir peak in the *Sample solution* corresponds to that in the *Standard solution*, as obtained in the *Assay*.
- **C. IDENTIFICATION TEST—GENERAL, Sulfate (191):** Meets the requirements

**ASSAY**

• **PROCEDURE**

**Buffer:** 4.0 g/L of sodium dihydrogen orthophosphate dihydrate in water. Adjust with orthophosphoric acid to a pH of 3.0.

**Mobile phase:** Acetonitrile and *Buffer* (1:9)

**Standard solution:** 0.05 mg/mL of USP Abacavir Sulfate RS in water

**Sample solution:** 0.05 mg/mL of Abacavir Sulfate in water

**Chromatographic system**

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

**Mode:** LC

**Detector:** UV 220 nm

**Column:** 4.6-mm × 15-cm; 5- $\mu$ m packing L7

**Flow rate:** 1 mL/min

**Injection size:** 20  $\mu$ L

**System suitability**

**Sample:** *Standard solution*

**Suitability requirements**

**Column efficiency:** NLT 3500 theoretical plates

**Tailing factor:** NMT 2.0

**Relative standard deviation:** NMT 1.0%

**Analysis**

**Samples:** *Standard solution* and *Sample solution*

Calculate the percentage of (C<sub>14</sub>H<sub>18</sub>N<sub>6</sub>O)<sub>2</sub> · H<sub>2</sub>SO<sub>4</sub> in the portion of Abacavir Sulfate taken:

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

$r_U$  = peak response of abacavir in the *Sample solution*

$r_S$  = peak response of abacavir in the *Standard solution*

$C_S$  = concentration of USP Abacavir Sulfate RS in the *Standard solution* (mg/mL)

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

**Acceptance criteria:** 98.0%–102.0% on the anhydrous basis

**IMPURITIES**

**Inorganic Impurities**

• **RESIDUE ON IGNITION (281):** NMT 0.3%, a 1.0-g test specimen being used

• **HEAVY METALS, Method II (231):** NMT 20 ppm

**Organic Impurities**

[NOTE—If abacavir related compound A or abacavir related compound B are known process impurities, *Procedure 2* is recommended.]

• **PROCEDURE 1**

**Buffer:** Proceed as directed in the *Assay*.

**Solution A:** *Buffer*

**Solution B:** Acetonitrile

**Mobile phase:** See the gradient table below.

Time (min)	Solution A (%)	Solution B (%)
0	98	2
20	88	12
30	40	60
35	40	60
36	98	2
45	98	2

**Standard solution:** 0.001 mg/mL of USP Abacavir Sulfate RS in water

**Sample solution:** 1 mg/mL of Abacavir Sulfate in water

**System suitability solution:** 0.1 mg/mL of USP Abacavir Sulfate RS in water

**Chromatographic system**

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

**Mode:** LC

**Detector:** UV 220 nm

**Column:** 4.6-mm × 25-cm; 5- $\mu$ m packing L7

**Flow rate:** 1.5 mL/min

**Injection size:** 20  $\mu$ L

**System suitability**

**Samples:** *Standard solution* and *System suitability solution*

**Suitability requirements**

**Resolution:** NLT 2 between abacavir and abacavir related compound C

**Tailing factor:** NMT 2.0, *System suitability solution*

**Relative standard deviation:** NMT 5.0%, *Standard solution*

**Analysis**

**Samples:** *Standard solution* and *Sample solution*

[NOTE—Relative retention times are listed in *Impurity Table 1.*]

2 / Abacavir Sulfate

Calculate the percentage of each impurity in the portion of Abacavir Sulfate taken:

$$\text{Result} = (r_u/r_s) \times (C_s/C_u) \times (1/F) \times 100$$

- $r_u$  = peak response of each impurity peak in the *Sample solution*
- $r_s$  = peak response of abacavir in the *Standard solution*
- $C_s$  = concentration of USP Abacavir Sulfate RS in the *Standard solution* (mg/mL)
- $C_u$  = nominal concentration of abacavir sulfate in the *Sample solution* (mg/mL)
- F = relative response factor (see *Impurity Table 1*)

**Acceptance criteria**  
**Total impurities:** NMT 0.5%

**Impurity Table 1**

Name	Relative Retention Time	Relative Response Factor	Acceptance Criteria, NMT (%)
Descyclopropyl abacavir <sup>a</sup>	0.64	1.6	0.1
Abacavir	1.0	—	—
Dihydro abacavir <sup>b</sup>	1.12	0.87	0.1
Abacavir related compound C <sup>c</sup>	1.16	1.5	0.1
Abacavir O-pyrimidine derivative <sup>d</sup>	1.43	0.93	0.15
Any other individual, unspecified impurity	—	1.0	0.1

- <sup>a</sup> (1*S*,4*R*)-4-[2,6-Diamino-9*H*-purin-9-yl]-2-cyclopentene-1-methanol.
- <sup>b</sup> (1*S*,4*R*)-4-[2-Amino-6-(cyclopropylamino)-9*H*-purin-9-yl]cyclopentane-1-methanol.
- <sup>c</sup> [(1*S*,4*R*)-4-(2-Amino-6-chloro-9*H*-purin-9-yl)cyclopent-2-enyl]methanol.
- <sup>d</sup> *N*<sup>6</sup>-Cyclopropyl-9-[(1*R*,4*S*)-4-[(2,5-diamino-6-chloropyrimidin-4-yl)oxy]methyl]cyclopent-2-enyl]-9*H*-purine-2,6-diamine.

**PROCEDURE 2**

**Buffer:** 1.15 g/L of ammonium dihydrogen phosphate and 2 mg/mL of tetrabutylammonium hydrogen sulfate in water. Adjust with triethylamine to a pH of 6.00 ± 0.05.

**Mobile phase:** Acetonitrile and *Buffer* (3:17)

**Standard solution:** 0.05 mg/mL of USP Abacavir Sulfate RS in *Mobile phase*

**Sample solution:** 0.5 mg/mL of Abacavir Sulfate in *Mobile phase*

**System suitability solution:** 0.75 µg/mL of USP Abacavir Related Compound A RS, 0.75 µg/mL of USP Abacavir Related Compound B RS, 0.75 µg/mL of USP Abacavir Related Compound C RS, and 0.5 mg/mL of USP Abacavir Sulfate RS in *Mobile phase*

**Chromatographic system**

(See *Chromatography* <621>, *System Suitability*.)

**Mode:** LC

**Detector:** UV 214 nm

**Column:** 4.6-mm × 25-cm; 5-µm packing L1

**Flow rate:** 1.2 mL/min

**Injection size:** 20 µL

**Run time:** 45 min

**Sample temperature:** 4°

**System suitability**

**Samples:** *Standard solution* and *System suitability solution*

**Suitability requirements**

**Tailing factor:** NMT 2.0, *Standard solution*

**Resolution:** NLT 2.0 between abacavir related compound B and abacavir related compound C, *System suitability solution*

**Relative standard deviation:** NMT 5.0%, *Standard solution*

**Analysis**

**Samples:** *Standard solution* and *Sample solution*

[NOTE—The relative retention times are listed in *Impurity Table 2*.]

Calculate the percentage of each impurity in the portion of Abacavir Sulfate taken:

$$\text{Result} = (r_u/r_s) \times (C_s/C_u) \times (1/F) \times 100$$

- $r_u$  = peak response of each impurity peak in the *Sample solution*
- $r_s$  = peak response of abacavir in the *Standard solution*
- $C_s$  = concentration of USP Abacavir Sulfate RS in the *Standard solution* (mg/mL)
- $C_u$  = nominal concentration of Abacavir Sulfate in the *Sample solution* (mg/mL)
- F = relative response factor (see *Impurity Table 2*)

**Acceptance criteria**  
**Total impurities:** NMT 1.0%

**Impurity Table 2**

Name	Relative Retention Time	Relative Response Factor	Acceptance Criteria, NMT (%)
Abacavir related compound A <sup>a</sup>	0.35	1.5	0.15
Abacavir related compound B <sup>b</sup>	0.65	0.92	0.15
Abacavir related compound C <sup>c</sup>	0.73	0.79	0.15
Abacavir	1.00	—	—
Any other individual, unspecified impurity	—	1.0	0.10

- <sup>a</sup> 4-(2,6-Diamino-9*H*-purin-9-yl)cyclopent-2-ene-yl-methanol.
- <sup>b</sup> [4-(2,5-Diamino-6-chloropyrimidin-4-ylamino)cyclopent-2-enyl]methanol.
- <sup>c</sup> [(1*S*, 4*R*)-4-(2-Amino-6-chloro-9*H*-purin-9-yl)cyclopent-2-enyl]methanol.

**SPECIFIC TESTS**

**• LIMIT OF THE ABACAVIR ENANTIOMER**

**Mobile phase:** *n*-Hexane, dehydrated alcohol, and methanol (8:1:1)

**Standard stock solution:** 0.4 mg/mL of USP Abacavir Sulfate RS initially dissolved at 10% of final volume with methanol. Dilute with *Mobile phase* to final volume.

**Standard solution:** 1 µg/mL of USP Abacavir Sulfate RS in *Mobile phase*

**Sample solution:** 0.5 mg/mL of Abacavir Sulfate initially dissolved at 10% of final volume with methanol. Dilute with *Mobile phase* to final volume.

**System suitability solution:** 0.5 mg/mL of USP Abacavir Sulfate Racemic RS initially dissolved at 10% of final volume with methanol. Dilute with *Mobile phase* to final volume.

**Chromatographic system**

(See *Chromatography* <621>, *System Suitability*.)

**Mode:** LC

**Detector:** UV 220 nm

**Column:** 4.6-mm × 25-cm; packing L51

**Flow rate:** 1 mL/min

**Injection size:** 20 µL

**System suitability**

**Samples:** *Standard solution* and *System suitability solution*  
[NOTE—The relative retention times for the abacavir enantiomer and abacavir are about 0.8 and 1.0, respectively.]

**Suitability requirements**

**Resolution:** NLT 2.5 between abacavir enantiomer and abacavir, *System suitability solution*

**Relative standard deviation:** NMT 5.0%, *Standard solution*

**Analysis**

**Samples:** *Standard solution* and *Sample solution*

Calculate the percentage of the abacavir enantiomer in the portion of Abacavir Sulfate taken:

$$\text{Result} = (r_u/r_s) \times (C_s/C_u) \times 100$$

- $r_u$  = peak response of abacavir enantiomer in the *Sample solution*

- $r_s$  = peak response of abacavir in the *Standard solution*  
 $C_s$  = concentration of USP Abacavir Sulfate RS in the *Standard solution* (mg/mL)  
 $C_u$  = nominal concentration of Abacavir Sulfate in the *Sample solution* (mg/mL)

**Acceptance criteria**

- Abacavir enantiomer: NMT 0.2%  
• **WATER DETERMINATION, Method I (921):** NMT 1.0%

**ADDITIONAL REQUIREMENTS**

- **PACKAGING AND STORAGE:** Preserve in well-closed containers. Store at controlled room temperature.

- **LABELING:** If a test for *Organic Impurities* other than *Procedure 1* is used, then the labeling states with which *Organic Impurities* test the article complies.  
• **USP REFERENCE STANDARDS (11)**  
USP Abacavir Sulfate RS  
USP Abacavir Sulfate Racemic RS  
USP Abacavir Related Compound A RS  
USP Abacavir Related Compound B RS  
USP Abacavir Related Compound C RS