

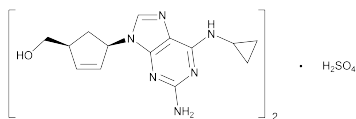
BRIEFING

Abacavir Sulfate. This USP Pending Monograph was published on the USP Website and authorized as Version 1 on January 28, 2008. It is proposed to use a flexible monograph approach to indicate that manufacturers could perform either *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- μ m packing L7. The typical retention time for abacavir in the *Assay* procedure is 4.6 min. The HPLC procedure in *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 *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 *Organic Impurities, Procedure 2* is 13 min. The HPLC procedure in *Limit of the abacavir enantiomer* is based on analysis performed using the Chiralpak-AD brand of L51 column. It is proposed to revise the system suitability requirement in *Organic Impurities, Procedure 1* to replace column efficiency with resolution between abacavir and abacavir related compound C. It is also proposed to update the names of impurities in *Impurity Table 1* for consistency with the impurities in *Impurity Table 2*.

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

Abacavir Sulfate

Draft 2



$(C_{14}H_{18}N_6O)_2 \cdot H_2SO_4$ 670.74
2-Cyclopentene-1-methanol, 4-[2-amino-6-(cyclopropylamino)-9H-purin-9-yl]-, (1S,4R)-, sulfate (2:1) (salt)
(1S,4R)-4-[2-Amino-6-(cyclopropylamino)-9H-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 $(C_{14}H_{18}N_6O)_2 \cdot H_2SO_4$, 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 per 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 \times 15-cm; 5- μ m packing L7
- Flow rate:** 1 mL/min
- Injection size:** 20 μ 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 $(C_{14}H_{18}N_6O)_2 \cdot H_2SO_4$ 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

Change to read:

Organic Impurities ▶[NOTE—If Abacavir Related Compound A or Abacavir Related Compound B are known process impurities, *Procedure 2* is recommended.]◀(1-Mar-2010)

• **PROCEDURE** ▶1◀(1-Mar-2010)

- 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 \times 25-cm; 5- μ m packing L7
- Flow rate:** 1.5 mL/min
- Injection size:** 20 μ L

System suitability

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

Suitability requirements

- Column efficiency:** NLT 25,000 theoretical plates
- ▶**Resolution:** NLT 2 between abacavir and abacavir related compound C◀(1-Mar-2010)
- Tailing factor:** NMT 2.0, *System suitability solution*
- Relative standard deviation:** NMT 5.0%, *Standard solution*

2 / Abacavir Sulfate

Analysis

Samples: *Standard solution* and *Sample solution*
[NOTE—Relative retention times are listed in *Impurity Table 1*.]

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*)

Impurity Table 1

Name	Relative Retention Time	Relative Response Factor	Acceptance Criteria, NMT (%)
Descyclopropyl abacavir ^a	0.64	1.6	0.1
Abacavir	1.0	—	—
Dihydro abacavir ^b	1.12	0.87	0.1
Abacavir penulti- mate ^c Abacavir related compound C ^c (1-Mar-2010) ^c	1.16	1.5	0.1
O-Pyrimidinyl abacavir ^d Abacavir O-pyrimidine derivative ^d (1-Mar-2010) ^d	1.43	0.93	0.15
Unspecified impurity	—	1.0	0.1

^a (1S,4R)-4-[2,6-Diamino-9H-purin-9-yl]-2-cyclopentene-1-methanol.

^b (1S,4R)-4-[2-Amino-6-(cyclopropylamino)-9H-purin-9-yl]cyclopentane-1-methanol.

^c (1S,4R)-4-[2-Amino-6-chloro-9H-purin-9-yl]-2-cyclopentene-1-methanol hydrochloride^c [(1S, 4R)-4-(2-Amino-6-chloro-9H-purin-9-yl)cyclopent-2-enyl]methanol. (1-Mar-2010)

^d (1S,4R)-4-[2-Amino-6-(cyclopropylamino)-9H-purin-9-yl]-1-(2,5-diamino-6-chloro-4-pyrimidinyl-2-cyclopentene-1-yl)methyl-2-cyclopentene-1-methanol^d N⁶-Cyclopropyl-9-[(1R,4S)-4-[[2,5-diamino-6-chloropyrimidin-4-yloxy)methyl]cyclopent-2-enyl]-9H-purine-2,6-diamine. (1-Mar-2010)

Acceptance criteria

Total impurities: NMT 0.5%

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: *System suitability solution*, *Standard stock solution*, and *Standard 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—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*)

Impurity Table 2

Name	Relative Retention Time	Relative Response Factor	Acceptance Criteria, NMT (%)
Abacavir related compound A ^a	0.35	1.5	0.15
Abacavir related compound B ^b	0.65	0.92	0.15
Abacavir related compound C ^c	0.73	0.79	0.15
Abacavir	1.00	—	—
Unspecified impurity	—	1.0	0.10

^a 4-(2,6-Diamino-1H-purin-9-yl)cyclopent-2-ene-yl-methanol.

^b [4-(2,5-Diamino-6-chloropyrimidin-4-ylamino)cyclopent-2-enyl]methanol.

^c [(1S, 4R)-4-(2-Amino-6-chloro-9H-purin-9-yl)cyclopent-2-enyl]methanol.

Acceptance criteria

Total impurities: NMT 1.0% (1-Mar-2010)

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: *System suitability solution* and *Standard 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.

Add the following:

- ▶ **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. ◀ (1-Mar-2010)

Change to read:

- **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 ◀ (1-Mar-2010)