

BRIEFING

Abacavir Oral Solution. This monograph has been posted on the USP Pending Standards Web page for review and public comment for more than 90 days. The MD-AA Expert Committee reviewed the submitted comments and has approved the monograph as an Authorized USP Pending Standard. The following is a summary of the comments and the Expert Committee's decisions:

Comment 1: Commenter suggested to clarify the definition to reflect that the Oral Solution is labeled in terms of the content of abacavir (base), and to add coefficients to the calculation in the test for *Related compounds* and in the *Assay*.

Response: Comment was incorporated.

Comment 2: Commenter suggested to modify the *Identification* test to specify the concentration of the *Standard solution* and to reformat the *Procedure* for clarity.

Response: Comment was incorporated.

Comment 3: Commenter suggested to indicate the autosampler temperature is to be maintained at 4° in the test for *Related compounds*.

Response: Comment was incorporated.

Comment 4: Commenter suggested to indicate the autosampler temperature is to be maintained at 4° in the *Assay*.

Response: Comment was incorporated.

Comment 5: Commenter suggested to multiply rather than divide the response factor in the formula for *Related compounds*.

Response: Relative response factor values in *Table 1* were revised to reciprocal values and used as a denominator in the *Related compounds* formula.

Comment 6: Commenter suggested to use 2.0 mg per mL of abacavir in the *Identification* test by TLC.

Response: Comment was incorporated.

The HPLC procedures used in the test for *Related compounds* and in the *Assay* are based on analyses performed with the Inertsil ODS 3V brand of L1 column. The typical retention times for abacavir sulfate and the three related compounds, namely, abacavir related compound A, abacavir related compound B, and abacavir related compound C are 11.5, 4.1, 7.9, and 8.9 minutes, respectively.

(MD-AA: L. Santos; B. Davani) RTS—C58872

Add the following:

■ **Abacavir Oral Solution**

v.1 Authorized January 28, 2008

» Abacavir Oral Solution contains an amount of abacavir sulfate equivalent to not less than 90.0 percent and not more than 110.0 percent of the labeled amount of abacavir (C₁₄H₁₈N₆O).

Packaging and storage—Preserve in tight, light-resistant containers, and store at room temperature.

USP Reference standards ⟨11⟩—*USP Abacavir Sulfate RS*, *USP Abacavir Related Compound A RS*, *USP Abacavir Related Compound B RS*, *USP Abacavir Related Compound C RS*.

Identification—

A: *Thin-Layer Chromatographic Identification Test* ⟨201⟩—

Test solution—Transfer the amount of solution, equivalent to 100 mg of abacavir into a 50-mL volumetric flask, dilute to volume with methanol, and mix.

Standard solution—Dissolve an accurately weighed quantity of USP Abacavir Sulfate RS in methanol to obtain a solution having a known concentration equivalent to about 2.0 mg of abacavir per mL, using sonication if necessary.

Procedure—Separately apply 5 μL of the *Test solution* and 5 μL of the *Standard solution* of USP Abacavir Sulfate RS in methanol to a thin-layer chromatographic plate (see *Chromatography* ⟨621⟩) coated with a 0.25-mm layer of chromatographic silica gel mixture containing a fluorescent indicator having an optimal intensity at 254 nm. Allow the applications to dry, and develop the chromatogram in a solvent system consisting of a mixture of ethyl acetate and methanol (4 : 1) until the solvent front has moved about three-fourths of the length of the plate. Remove the plate from the developing chamber, mark the solvent front, and allow the solvent to evaporate. Observe the plate under short-wavelength UV light: the *R_F* value of the principal spot obtained from the *Test solution* corresponds to that of the principal spot obtained from the *Standard solution*.

B: The retention time of the major peak in the chromatogram of the *Assay preparation* corresponds to that in the chromatogram of the *Standard preparation*, as obtained in the *Assay*.

Microbial limits ⟨61⟩—The total aerobic microbial count does not exceed 100 cfu per mL, and the total combined yeasts and molds count does not exceed 10 cfu per mL. It meets the requirements of the tests for absence of *Escherichia coli*.

2 / Abacavir Oral Solution

Uniformity of dosage units (905)—

FOR ORAL SOLUTION PACKAGED IN SINGLE-UNIT CONTAINERS: meets the requirements.

Deliverable volume (698)—

FOR ORAL SOLUTION PACKAGED IN MULTIPLE-UNIT CONTAINERS: meets the requirements.

pH (791): between 4.60 and 5.00.

Related compounds—

[NOTE—The *Test solution* and the *Standard solution* are to be refrigerated at or below 4° immediately after preparation and during analysis using a refrigerated autosampler. The solutions are stable at or below 4° for about 24 hours.]

Buffer solution and *Mobile phase*—Proceed as directed in the *Assay*.

Impurity stock solutions—Dissolve in separate containers an accurately weighed quantity of USP Abacavir Related Compound A RS, USP Abacavir Related Compound B RS, and USP Abacavir Related Compound C RS in *Mobile phase*. Dilute quantitatively, and stepwise if necessary, with *Mobile phase* to obtain a solution having a known concentration of 0.15 mg per mL of each of abacavir related compound A, abacavir related compound B, and abacavir related compound C.

System suitability solution—Weigh accurately about 120 mg of USP Abacavir Sulfate RS, transfer to a 200-mL volumetric flask, and add 1 mL of each *Impurity stock solution*. Dissolve in and dilute with *Mobile phase* to volume, and mix.

Standard solution—Dissolve an accurately weighed quantity of USP Abacavir Sulfate RS in *Mobile phase*. Dilute quantitatively, and stepwise if necessary, with *Mobile phase* to obtain a solution having a known concentration of 0.6 µg per mL.

Test solution—Transfer an accurately measured volume of Oral Solution, equivalent to about 100 mg of abacavir based on the label claim, to a 200-mL volumetric flask. Dissolve in and dilute with *Mobile phase* to volume, and mix.

Chromatographic system (see *Chromatography* (621))—

The liquid chromatograph is equipped with a 214-nm detector and a 4.6-mm × 25-cm column that contains packing L1. The flow rate is about 1.2 mL per minute. The column temperature is maintained at 25°. Chromatograph the *System suitability solution*, and record the peak responses as directed for *Procedure*: the resolution, *R*, between abacavir related compound B and abacavir related compound C is not less than 2.0; the column efficiency for the abacavir peak is not less than 3000 theoretical plates; the tailing factor for the abacavir peak is not more than 2.0; and the relative standard deviation for the peak area of the abacavir peak for six replicate injections is not more than 5.0%.

Procedure—Separately inject a volume (about 20 µL) of the *Standard solution* and the *Test solution* into the chromatograph. Identify the impurities based on relative retention times given in *Table 1*, and measure the peak responses. Calculate the percentage of each impurity in the portion of Oral Solution taken by the formula:

$$(100/F)(572.66/670.76)(C_s/C_v)(r_v/r_s)$$

in which *F* is the relative response factor for each impurity against abacavir as listed in *Table 1*; 572.66 and 670.76 are the molecular weights of abacavir and abacavir sulfate, respectively; *C_s* is the concentration, in mg per mL, of USP Abacavir Sulfate RS in the *Standard solution*; *C_v* is the concentration, in mg per mL, of abacavir calculated, based on the label claim, in the *Test solution*; *r_v* is the peak area for each impurity obtained from the *Test solution*; and *r_s* is the peak area of abacavir obtained from the *Standard solution*.

Assay—

[NOTE—The *Standard preparation* and *Assay preparation* are to be refrigerated at or below 4° immediately after preparation and during analysis using a refrigerated autosampler. The solutions are stable at or below 4° for about 24 hours.]

Table 1

Name	Approximate Relative Retention Time	Relative Response Factor (<i>F</i>)	Limit (%)
Abacavir related compound A impurity ¹	0.36	1.4	0.15
Abacavir	1.00	—	—
Abacavir related compound B impurity ²	0.69	0.9	0.15
Abacavir related compound C impurity ³	0.77	0.8	0.15
Unknown impurities	—	1.0	0.10
Total impurity	—	—	1.00

¹ [4-(2,6-Diamino-9*H*-purin-9-yl)cyclopent-2-enyl]methanol.

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

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

Buffer solution—Accurately weigh about 1.15 g of ammonium dihydrogen phosphate and 2 g of tetrabutylammonium hydrogen sulfate, and transfer into a 1000-mL volumetric flask. Dissolve in and dilute with water to volume, and mix. Adjust with triethylamine to a pH of 6.0 ± 0.05 . Pass through a membrane filter having a 0.45- μ m porosity.

Mobile phase—Prepare a filtered and degassed mixture of *Buffer solution* and acetonitrile (85 : 15). Make adjustments if necessary (see *System Suitability* under *Chromatography* (621)).

Standard preparation—Dissolve an accurately weighed quantity of USP Abacavir Sulfate RS in *Mobile phase*, and dilute quantitatively, and stepwise if necessary, with *Mobile phase* to obtain a solution having a known concentration of about 0.06 mg per mL.

Assay preparation—Transfer an accurately measured volume of Oral Solution, equivalent to about 100 mg of abacavir based on the label claim, to a 200-mL volumetric flask, dissolve in and dilute with *Mobile phase* to volume, and mix. Transfer 5.0 mL of this solution to a 50-mL volumetric flask, dilute with *Mobile phase* to volume, and mix.

Chromatographic system (see *Chromatography* (621))—The liquid chromatograph is equipped with a 214-nm detector and a 4.6-mm \times 15-cm column that contains packing L1. The flow rate is about 1.5 mL per minute. The column temperature is maintained at 40°. Chromatograph the

Standard preparation, and record the peak responses as directed for *Procedure*: the tailing factor is not more than 2.0; and the relative standard deviation for replicate injections is not more than 2.0%.

Procedure—Separately inject equal volumes (about 20 μ L) of the *Standard preparation* and the *Assay preparation* into the chromatograph, record the chromatograms, and measure the responses for the abacavir sulfate peak. Calculate the percentage of the labeled amount of abacavir (C₁₄H₁₈N₆O) in the portion of Oral Solution taken by the formula:

$$100(572.66/670.76)(C_s/C_U)(r_U/r_s)$$

in which 572.66 and 670.76 are the molecular weights of abacavir and abacavir sulfate, respectively; *C_s* is the concentration, in mg per mL, of USP Abacavir Sulfate RS in the *Standard preparation*; *C_U* is the concentration, in mg per mL, of abacavir calculated, based on label claim, in the *Assay preparation*; and *r_U* and *r_s* are the peak area responses of abacavir obtained from the *Assay preparation* and the *Standard preparation*, respectively. ■