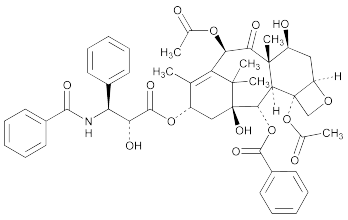
Paclitaxel



$C_{47}H_{51}NO_{14}$ 853.91

Benzenepropanoic acid, β-(benzoylamino)-α-hydroxy-, 6,12b-bis(acetyloxy)-12-(benzoyloxy)-2a,3,4,4a,5,6,9,10,11,12,12a,12b-dodecahydro-4,11-dihydroxy-4a,8,13,13-tetramethyl-5-oxo-7,11-methano-1*H*-cyclodeca[3,4]benz[1,2-b]oxet-9-yl ester, [2a*R*-[2aα,4β,4aβ,6β,9α(α*R**,β*S**),11α,12α,12aα,12bα]]-.

(2a*R*,4*S*,4a*S*,6*R*,9*S*,11*S*,12*S*,12a*R*,12b*S*)-1,2a,3,4,4a,6,9,10,11,12,12a,12b-Dodecahydro-4,6,9,11,12,12b-hexahydroxy-4a,8,13,13-tetramethyl-7,11-methano-5*H*-cyclodeca[3,4]-benz[1,2-*b*]oxet-5-one 6,12b-diacetate, 12-benzoate, 9-ester with (2*R*,3*S*)-*N*-benzoyl-3-phenylisoserine [33069-62-4].

» Paclitaxel contains not less than 97.0 percent and not more than 102.0 percent of $C_{47}H_{51}NO_{14}$, calculated on the anhydrous, solvent-free basis.

Caution—Paclitaxel is cytotoxic. Great care should be taken to prevent inhaling particles of Paclitaxel and exposing the skin to it.

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

Labeling—The labeling indicates the type of process used to produce the material and the *Related compounds* test with which the material complies.

USP Reference standards ⟨11⟩**—**
USP Endotoxin RS
USP Paclitaxel RS
USP Paclitaxel Related Compound A RS
Cephalomannine.
USP Paclitaxel Related Compound B RS
10-Deacetyl-7-epipaclitaxel.
USP Paclitaxel Impurity Mixture RS
Mixture of paclitaxel and the following related compounds: propyl analog, cephalomannine, *sec*-butyl analog, *n*-butyl analog, benzyl analog, baccatin VI, pentyl analog, and 7-epipaclitaxel.

Identification—

A: *Infrared Absorption* ⟨197K⟩.

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.*

Specific rotation ⟨781S⟩**:** between –49.0° and –55.0° at 20°, calculated on the anhydrous, solvent-free basis.

Test solution: 10 mg per mL, in methanol.

Microbial enumeration tests ⟨61⟩ **and Tests for specified microorganisms** ⟨62⟩**—**The total aerobic microbial count does not exceed 100 cfu per g. It meets the requirements of the tests for the absence of *Staphylococcus aureus, Pseudomonas aeruginosa, Salmonella* species, and *Escherichia coli.*

Bacterial endotoxins ⟨85⟩**—**It contains not more than 0.4 USP Endotoxin Unit per mg of paclitaxel.

Water, *Method Ic* ⟨921⟩**:** not more than 4.0%.

Residue on ignition ⟨281⟩**:** not more than 0.2%.

Heavy metals, *Method II* ⟨231⟩**:** 0.002%.

Related compounds—

TEST 1 (FOR MATERIAL LABELED AS ISOLATED FROM NATURAL SOURCES)—If the material complies with this test, the labeling indicates that it meets USP *Related compounds Test 1.*

Diluent—Prepare as directed in the *Assay.*

Solution A—Prepare filtered and degassed acetonitrile.

Solution B—Prepare filtered and degassed water.

Mobile phase—Use variable mixtures of *Solution A* and *Solution B* as directed for *Chromatographic system.* Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

System suitability solution—Dissolve accurately weighed quantities of USP Paclitaxel Related Compound A RS and USP Paclitaxel Related Compound B RS in methanol to obtain a solution having known concentrations of about 10 µg of each per mL. Transfer 5.0 mL of this solution to a 50-mL volumetric flask, dilute with *Diluent* to volume, and mix.

Standard solution—Dissolve, with the aid of sonication, an accurately weighed quantity of USP Paclitaxel RS in *Diluent,* and dilute quantitatively, and stepwise if necessary, with *Diluent* to obtain a solution having a known concentration of about 5 µg per mL.

Test solution—Use the *Assay preparation.*

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 227-nm detector and a 4.6-mm × 25-cm column that contains 5-µm packing L43. The flow rate is about 2.6 mL per minute. The column temperature is maintained at 30°. The chromatograph is programmed as follows.

Time (minutes)	*Solution A* (%)	*Solution B* (%)	Elution
0–35	35	65	isocratic
35–60	35→80	65→20	linear gradient
60–70	80→35	20→65	linear gradient
70–80	35	65	isocratic

Chromatograph the *System suitability solution,* and record the peak responses as directed for *Procedure:* the relative retention times are about 0.78 for paclitaxel related compound A and 0.86 for paclitaxel related compound B (relative to the retention time for paclitaxel obtained from the *Test solution*); and the resolution, *R,* between paclitaxel related compound A and paclitaxel related compound B is not less than 1.0. Chromatograph the *Standard solution,* and record the peak responses as directed for *Procedure:* the relative standard deviation for replicate injections is not more than 2.0%.

Procedure—Inject a volume (about 15 µL) of the *Test solution* into the chromatograph, record the chromatogram, and measure the areas for the major peaks. Calculate the percentage of each impurity in the portion of Paclitaxel taken by the formula:

$$100(Fr_i / r_U)$$

in which F is the relative response factor for each impurity peak (see *Table 1* for values); r_i is the peak area for each individual impurity; and r_U is the peak area for paclitaxel.

Table 1

Relative Retention Time	Relative Response Factor (*F*)	Name	Limit (%)
0.24	1.29	Baccatin III	0.2
0.53	1.00	10-Deacetylpaclitaxel	0.5
0.57	1.00	7-Xylosylpaclitaxel	0.2
0.78	1.26	Cephalomannine (paclitaxel related compound A)	a_1[1]
0.78	1.26	2″,3″-Dihydrocephalomannine	a_2[1]
0.86	1.00	10-Deacetyl-7-epipaclitaxel (paclitaxel related compound B)	0.5
1.10	1.00	Benzyl analog[3]	b_1[2]
1.10	1.00	3″,4″-Dehydropaclitaxel C	b_2[2]
1.40	1.00	7-Epicephalomannine	0.3
1.85	1.00	7-Epipaclitaxel	0.5

[1] Resolution may be incomplete for these peaks, depending upon the relative amounts present; the sum of a_1 and a_2 is not more than 0.5%.
[2] Resolution may be incomplete for these peaks, depending upon the relative amounts present; the sum of b_1 and b_2 is not more than 0.5%.
[3] The following chemical name is assigned to the related compound, benzyl analog: Baccatin III 13-ester with (2*R*,3*S*)-2-hydroxy-3-phenyl-3-(2-phenylacetylamino)propanoic acid.

In addition to not exceeding the limits for paclitaxel related impurities in *Table 1,* not more than 0.1% of any other single impurity is found; and not more than 2.0% of total impurities is found.

TEST 2 (FOR MATERIAL LABELED AS PRODUCED BY A SEMISYNTHETIC PROCESS)—If the material complies with this test, the labeling indicates that it meets USP *Related compounds Test 2.*

Diluent—Use acetonitrile.

Solution A—Use a filtered and degassed mixture of water and acetonitrile (3:2).

Solution B—Use filtered and degassed acetonitrile.

Mobile phase—Use variable mixtures of *Solution A* and *Solution B* as directed for *Chromatographic system.* Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

System suitability solution—Dissolve accurately weighed quantities of USP Paclitaxel RS and USP Paclitaxel Related Compound B RS in *Diluent,* shaking and sonicating if necessary, to obtain a solution having known concentrations of about 0.96 mg and 0.008 mg per mL, respectively.

Test solution—Transfer about 10 mg of Paclitaxel, accurately weighed, to a 10-mL volumetric flask, dissolve in and dilute with *Diluent* to volume, shaking and sonicating if necessary, and mix.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 227-nm detector and a 4.6-mm × 15-cm column that contains 3-µm packing L1. The flow rate is about 1.2 mL per minute. The column temperature is maintained at 35°. The chromatograph is programmed as follows.

Time (minutes)	*Solution A* (%)	*Solution B* (%)	Elution
0–20	100	0	isocratic
20–60	100→10	0→90	linear gradient
60–62	10→100	90→0	linear gradient
62–70	100	0	isocratic

Chromatograph the *System suitability solution,* and record the peak responses as directed for *Procedure:* the relative retention times are about 0.94 for paclitaxel related compound B and 1.0 for paclitaxel; the resolution, *R,* between paclitaxel related compound B and paclitaxel is not less than 1.2; and the relative standard deviation for replicate injections is not more than 2.0%.

Procedure—Separately inject equal volumes (about 15 µL) of the *Diluent* and the *Test solution* into the chromatograph, record the chromatograms, and measure the areas for all the peaks. Disregard any peaks due to the *Diluent.* Calculate the percentage of each impurity in the portion of Paclitaxel taken by the formula:

$$100(Fr_i / r_s)$$

in which *F* is the relative response factor for each impurity (see *Table 2* for values); r_i is the peak area for each impurity obtained from the *Test solution;* and r_s is the sum of the areas of all the peaks obtained from the *Test solution.*

Table 2

Relative Retention Time	Relative Response Factor (*F*)	Name	Limit (%)
0.11	1.24	10-Deacetylbaccatin III	0.1
0.20	1.29	Baccatin III	0.2
0.42	1.39	Photodegradant[2]	0.1
0.47	1.00	10-Deacetylpaclitaxel	0.5
0.80	1.00	2-Debenzoylpaclitaxel-2-pentenoate	0.7
0.92[1]	1.00	Oxetane ring opened, acetyl and benzoyl migrated[2]	x_1
0.92[1]	1.00	10-Acetoacetylpaclitaxel	x_2
0.94[1]	1.00	10-Deacetyl-7-epipaclitaxel (paclitaxel related compound B)	x_3
1.37	1.00	7-Epipaclitaxel	0.4
1.45	1.00	10,13-Bissidechainpaclitaxel[2]	0.5
1.54	1.00	7-Acetylpaclitaxel	0.6
1.80	1.75	13-Tes-baccatin III	0.1
2.14	1.00	7-Tes-paclitaxel	0.3

[1] Resolution may be incomplete for these peaks, depending upon the relative amounts present; the sum of x_1, x_2, and x_3 is not more than 0.4%.
[2] The following chemical names are assigned to the related compounds Photodegradant; Oxetane ring opened, acetyl and benzoyl migrated; and 10,13-Bissidechainpaclitaxel:
Photodegradant
(1*R*,2*R*,4*S*,5*S*,7*R*,10*S*,11*R*,12*S*,13*S*,15*S*,16*S*)-2,10-diacetyloxy-5,13-dihydroxy-4,16,17,17-tetramethyl-8-oxa-3-oxo-12-phenylcarbonyloxypentacyclo[11.3.1.0^{1,11}.0^{4,11}.0^{7,10}]heptadec-15-yl (2*R*,3*S*)-2-hydroxy-3-phenyl-3-(phenylcarbonylamino)propanoate
Oxetane ring opened, acetyl and benzoyl migrated
(1*S*,2*S*,3*R*,4*S*,5*S*,7*S*,8*S*,10*R*,13*S*)-5,10-diacetyloxy-1,2,4,7-tetrahydroxy-8,12,15,15-tetramethyl-9-oxo-4-(phenylcarbonyloxymethyl)tricyclo[9.3.1.0^{3,8}]pentadec-11-en-13-yl (2*R*,3*S*)-2-hydroxy-3-phenyl-3-(phenylcarbonylamino)propanoate
10,13-Bissidechainpaclitaxel
Baccatin III 13-ester with (2*R*,3*S*)-2-hydroxy-3-phenyl-3-(phenylcarbonylamino)propanoic acid, 10-ester with (2*S*,3*S*)-2-hydroxy-3-phenyl-3-(phenylcarbonylamino)propanoic acid

In addition to not exceeding the limits for paclitaxel related impurities in *Table 2,* not more than 0.1% of any other single impurity is found; and not more than 2.0% of total impurities is found.

TEST 3 (FOR MATERIAL LABELED AS PRODUCED BY A PLANT CELL FERMENTATION PROCESS)—If the material complies with this test, the labeling indicates that it meets USP *Related compounds Test 3.*

Solution A—Prepare a filtered and degassed mixture of water and acetonitrile (3:2).

Solution B—Prepare filtered and degassed acetonitrile.

Mobile phase—Use variable mixtures of *Solution A* and *Solution B* as directed for *Chromatographic system.* Make adjust-

ments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

System suitability solution—Dissolve USP Paclitaxel Impurity Mixture RS in acetonitrile, sonicating if necessary, to obtain a solution having a known concentration of about 1 mg per mL.

Standard solution—Dissolve an accurately weighed quantity of USP Paclitaxel RS in acetonitrile, sonicating if necessary, to obtain a solution having a known concentration of about 1 mg per mL.

Test solution—Transfer about 10 mg of Paclitaxel, accurately weighed, to a 10-mL volumetric flask. Dissolve in and dilute with acetonitrile to volume, sonicating if necessary, and mix.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 227-nm detector and a 4.6-mm × 15-cm column that contains 3-µm packing L1. The flow rate is about 1.2 mL per minute. The chromatograph is programmed as follows.

Time (minutes)	*Solution A* (%)	*Solution B* (%)	Elution
0–28	100	0	isocratic
28–33	100→98	0→2	linear gradient
33–58	98→10	2→90	linear gradient
58–60	10	90	isocratic
60–63	10→100	90→0	linear gradient
63–70	100	0	isocratic

Chromatograph the *System suitability solution,* and record the peak responses as directed for *Procedure:* the resolution, *R,* between paclitaxel and benzyl analog is not less than 1.8. Chromatograph the *Standard solution,* and record the peak responses as directed for *Procedure:* the relative standard deviation for replicate injections is not more than 2.0%. [NOTE—For the purpose of peak identification, the approximate relative retention times are given in *Table 3*. The relative retention times are measured versus Paclitaxel.]

Table 3

Name	Relative Retention Time	Limit (%)
Propyl analog[1]	0.54	0.2
Cephalomannine (Paclitaxel related compound A)	0.76	0.5
sec-Butyl analog[2]	0.81	0.2
n-Butyl analog[3]	0.89	0.1
Benzyl analog	1.10	0.4
Baccatin VI	1.23	0.2
Pentyl analog[4]	1.31	0.2
7-Epipaclitaxel	1.51	0.4

[1] The following chemical name is assigned to the related compound Propyl analog: Baccatin III 13-ester with (2*R*,3*S*)-3-butanoylamino-2-hydroxy-3-phenylpropanoic acid.

[2] The following chemical name is assigned to the related compound *sec*-Butyl analog: Baccatin III 13-ester with (2*R*,3*S*)-2-hydroxy-3-(2-methylbutanoylamino)-3-phenylpropanoic acid.

[3] The following chemical name is assigned to the related compound *n*-Butyl analog: Baccatin III 13-ester with (2*S*,3*S*)-2-hydroxy-3-(pentanoylamino)-3-phenylpropanoic acid.

[4] The following chemical name is assigned to the related compound Pentyl analog: Baccatin III 13-ester with (2*R*,3*S*)-3-(hexanoylamino)-2-hydroxy-3-phenylpropanoic acid.

Procedure—Inject a volume (about 12 µL) of the *Test solution* into the chromatograph, record the chromatogram, and measure the areas for all the peaks. Calculate the percentage of each impurity in the portion of Paclitaxel taken by the formula:

$$100(r_i / r_U)$$

in which r_i is the response of each individual impurity; and r_U is the sum of the areas of all the peaks obtained from the *Test solution*. In addition to not exceeding the limits for paclitaxel related impurities in *Table 3,* not more than 0.1% of any other single impurity is found; and not more than 2.0% of total impurities is found.

Assay—

Diluent—Prepare a mixture of methanol and acetic acid (200:1).

Mobile phase—Prepare a filtered and degassed mixture of water and acetonitrile (11:9). Make adjustments if necessary (see *System Suitability* under *Chromatography* ⟨621⟩).

Standard preparation—Dissolve, using sonication if necessary, an accurately weighed quantity of USP Paclitaxel RS in *Diluent,* and dilute quantitatively, and stepwise if necessary, with *Diluent* to obtain a solution having a known concentration of about 1 mg per mL.

Assay preparation—Transfer about 10 mg of Paclitaxel, accurately weighed, to a 10-mL volumetric flask. Dissolve in *Diluent,* using sonication if necessary, dilute with *Diluent* to volume, and mix.

Chromatographic system (see *Chromatography* ⟨621⟩)—The liquid chromatograph is equipped with a 227-nm detector and a 4.6-mm × 25-cm column that contains 5-µm packing L43. The flow rate is about 1.5 mL per minute. Chromatograph the *Standard preparation,* and record the peak responses as directed for *Procedure:* the tailing factor is between 0.7 and 1.3; and the relative standard deviation for replicate injections is not more than 1.5%.

Procedure—Separately inject equal volumes (about 10 µL) of the *Standard preparation* and the *Assay preparation* into the chromatograph, record the chromatograms, and measure the areas for the major peaks. Calculate the quantity, in mg, of $C_{47}H_{51}NO_{14}$ in the portion of Paclitaxel taken by the formula:

$$10C(r_U / r_S)$$

in which *C* is the concentration, in mg per mL, of USP Paclitaxel RS in the *Standard preparation;* and r_U and r_S are the peak responses for paclitaxel obtained from the *Assay preparation* and the *Standard preparation,* respectively.

Paclitaxel Injection

» Paclitaxel Injection is a sterile, stabilized solution of Paclitaxel, suitable for dilution for intravenous administration. It contains not less than 90.0 percent and not more than 110.0 percent of the labeled amount of paclitaxel ($C_{47}H_{51}NO_{14}$).

Packaging and storage—Preserve in single-dose or multiple-dose containers, preferably of Type I glass, at controlled room temperature.

Labeling—Label it to indicate that it is to be diluted with a suitable parenteral vehicle prior to intravenous infusion.

USP Reference standards ⟨11⟩**—**
USP Endotoxin RS
USP Paclitaxel RS
USP Paclitaxel Related Compound B RS
10-Deacetyl-7-epipaclitaxel.

Identification—

A: The retention time of the major peak in the chromatogram of the *Test solution* corresponds to that in the chromato-