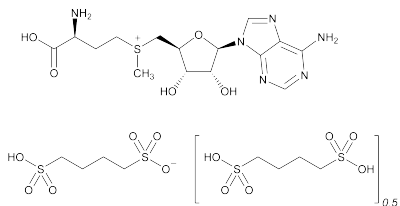


Add the following:

**▲ S-Adenosyl-L-methionine
1,4-Butanedisulfonate**



$C_{15}H_{23}N_6O_5S^+ \cdot C_4H_9O_6S_2^- \cdot 0.5 C_4H_{10}O_6S_2$ 725.80

S-(Adenosyl)-L-methionine 1,4-butanedisulfonate;
Adenosine, 5'-[[[(3S)-3-amino-3-carboxypropyl]
methylsulfonio]-5'-deoxy-, 1,4-butanedisulfonate (salt),
1,4-butanedisulfonate (salt) (2:1:2) CAS RN®: 101020-79-
5.

Adenosine, 5'-[[[(3S)-3-amino-3-carboxypropyl]
methylsulfonio]-5'-deoxy-, 1,4-hydrogen butanedisulfonate
(salt), 1,4-butanedisulfonic acid (2:2:1).

DEFINITION

S-Adenosyl-L-methionine 1,4-Butanedisulfonate is the salt of a mixture of diastereomers of the S-adenosyl-L-methionine ion with 1,4-butanedisulfonate. It may contain additional 1,4-butanedisulfonic acid ($C_4H_{10}O_6S_2$). It contains NLT 50.0% and NMT 56.0% of S-adenosyl-L-methionine ($C_{15}H_{23}N_6O_5S^+$), and NLT 44.0% and NMT 49.0% of 1,4-butanedisulfonic acid ($C_4H_{10}O_6S_2$), calculated on the anhydrous basis. It contains NLT 97.0% and NMT 101.0% of S-adenosyl-L-methionine 1,4-butanedisulfonate, calculated on the anhydrous basis.

IDENTIFICATION

- **A. SPECTROSCOPIC IDENTIFICATION TESTS** (197), *Infrared Spectroscopy*: 197K
- **B.** The retention time of the major peak of the *Sample solution* corresponds to that of S-adenosyl-L-methionine in the *System suitability solution*, as obtained in the test for *Content of S-Adenosyl-L-methionine*.

COMPOSITION

• **CONTENT OF S-ADENOSYL-L-METHIONINE**

[NOTE—Prepare the test solutions fresh and analyze within 1 h.]

Buffer solution: Dissolve 1.0 g of sodium 1-octanesulfonate and 12.6 g of ammonium formate in 1000 mL of water and adjust with formic acid to a pH of 2.8.

Mobile phase: Acetonitrile and *Buffer solution* (10:90)

Standard solution: 0.8 mg/mL of USP

S-Adenosyl-L-methionine 1,4-Butanedisulfonate RS in *Mobile phase*

System suitability solution: 4.0 µg/mL each of USP

S-Adenosyl-L-homocysteine RS and USP

S-Adenosyl-L-methionine 1,4-Butanedisulfonate RS in *Mobile phase*

Sample solution: 0.8 mg/mL of S-Adenosyl-L-methionine 1,4-Butanedisulfonate in *Mobile phase*

Chromatographic system

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

Mode: LC

Detector: UV 260 nm

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

Temperatures

Autosampler: 2°–8°

Column: 30°

Flow rate: 1.5 mL/min

Injection volume: 20 µL

Run time: NLT 5 times the retention time of S-adenosyl-L-methionine

System suitability

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

[NOTE—The relative retention times of S-adenosyl-L-homocysteine and S-adenosyl-L-methionine are about 0.8 and 1.0, respectively.]

Suitability requirements

Resolution: NLT 3.0 between

S-adenosyl-L-homocysteine and

S-adenosyl-L-methionine, *System suitability solution*

Relative standard deviation: NMT 2.0% for S-adenosyl-L-methionine, *Standard solution*

Analysis

Samples: *Standard solution* and *Sample solution*

Calculate the percentage of S-adenosyl-L-methionine ($C_{15}H_{23}N_6O_5S^+$) in the portion of S-Adenosyl-L-methionine 1,4-Butanedisulfonate taken:

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

r_U = peak area of S-adenosyl-L-methionine from the *Sample solution*

r_S = peak area of S-adenosyl-L-methionine from the *Standard solution*

C_S = concentration of USP S-Adenosyl-L-methionine 1,4-Butanedisulfonate RS in the *Standard solution* (mg/mL)

C_U = concentration of S-Adenosyl-L-methionine 1,4-Butanedisulfonate in the *Sample solution* (mg/mL)

P = assigned value for the content of S-adenosyl-L-methionine in USP S-Adenosyl-L-methionine 1,4-Butanedisulfonate RS

Acceptance criteria: 50.0%–56.0% on the anhydrous basis

• **CONTENT OF 1,4-BUTANEDISULFONIC ACID**

Mobile phase: 35 mM potassium hydroxide in water

Standard solution: 0.08 mg/mL of USP

1,4-Butanedisulfonic Acid RS in water

Sample solution: 0.2 mg/mL of S-Adenosyl-L-methionine

1,4-Butanedisulfonate in water

Chromatographic system

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

Mode: Ion chromatography

Detector: Conductivity with self-regenerating anion suppressor current at 87 mA (or according to manufacturer's recommendations for optimal signal-to-noise ratio)

Column: 4.0-mm × 25-cm; 7.5-µm packing L113

Column temperature: 30°

Flow rate: 1.0 mL/min

Injection volume: 25 µL

Run time: NLT 3 times the retention time of 1,4-butanedisulfonate

System suitability

Sample: *Standard solution*

Suitability requirements

Column efficiency: NLT 4000

Relative standard deviation: NMT 2.0%

Analysis

Samples: *Standard solution* and *Sample solution*

Calculate the percentage of 1,4-butanedisulfonic acid ($C_4H_{10}O_6S_2$) in the portion of S-Adenosyl-L-methionine 1,4-Butanedisulfonate taken:

Result = (r_U/r_S) × (C_S/C_U) × 100

- r_U = peak area of 1,4-butanedisulfonate from the *Sample solution*
- r_S = peak area of 1,4-butanedisulfonate from the *Standard solution*
- C_S = concentration of USP 1,4-Butanedisulfonic Acid RS in the *Standard solution* (mg/mL)
- C_U = concentration of S-Adenosyl-L-methionine 1,4-Butanedisulfonate in the *Sample solution* (mg/mL)

Acceptance criteria: 44.0%–49.0% on the anhydrous basis

• **CONTENT OF S-ADENOSYL-L-METHIONINE 1,4-BUTANEDISULFONATE**

Analysis: Calculate the total content of S-adenosyl-L-methionine 1,4-Butanedisulfonate by adding the individual percentages, as obtained in the tests for *Content of S-Adenosyl-L-methionine* and *Content of 1,4-Butanedisulfonic Acid*, on the anhydrous basis.

Acceptance criteria: 97.0%–101.0% on the anhydrous basis

IMPURITIES

• **RELATED COMPOUNDS**

Mobile phase, Sample solution, and Chromatographic system: Proceed as directed in the *Content of S-Adenosyl-L-methionine*.

Standard solution: 4.0 µg/mL each of USP Adenine RS, USP S-Adenosyl-L-homocysteine RS, and USP S-Adenosyl-L-methionine 1,4-Butanedisulfonate RS in *Mobile Phase*

System suitability

Sample: *Standard solution*

[NOTE—See *Table 1* for relative retention times.]

Suitability requirements

Resolution: NLT 3.0 between S-adenosyl-L-homocysteine and S-adenosyl-L-methionine; NLT 2.0 between each impurity

Relative standard deviation: NMT 10.0% for S-adenosyl-L-homocysteine and S-adenosyl-L-methionine

Analysis

Samples: *Standard solution* and *Sample solution*
Separately calculate the percentages of adenine and S-adenosyl-L-homocysteine in the portion of S-Adenosyl-L-methionine 1,4-Butanedisulfonate taken:

Result = (r_U/r_S) × (C_S/C_U) × 100

- r_U = peak area of adenine or S-adenosyl-L-homocysteine from the *Sample solution*
- r_S = peak area of adenine or S-adenosyl-L-homocysteine from the *Standard solution*
- C_S = concentration of USP Adenine RS or USP S-Adenosyl-L-homocysteine RS in the *Standard solution* (mg/mL)
- C_U = concentration of S-Adenosyl-L-methionine 1,4-Butanedisulfonate in the *Sample solution* (mg/mL)

Calculate the percentage of each specified and unspecified impurity in the portion of S-Adenosyl-L-methionine 1,4-Butanedisulfonate taken:

Result = (r_U/r_S) × (C_S/C_U) × 100

- r_U = peak area of each specified or unspecified impurity from the *Sample solution*
- r_S = peak area of S-adenosyl-L-methionine 1,4-butanedisulfonate from the *Standard solution*
- C_S = concentration of USP S-Adenosyl-L-methionine 1,4-Butanedisulfonate RS in the *Standard solution* (mg/mL)
- C_U = concentration of S-Adenosyl-L-methionine 1,4-Butanedisulfonate in the *Sample solution* (mg/mL)

Acceptance criteria: See *Table 1*.

Table 1

Name	Relative Retention Time	Acceptance Criteria, NMT (%)
Adenine	0.63	0.5
S-Adenosyl-L-homocysteine	0.75	0.1
S-Adenosyl-L-methionine	1.00	—
Methylthioadenosine ^a	2.89	0.3
S-Adenosylmethionamine ^b	3.73	0.2
Any unspecified impurity	—	0.1
Total impurities	—	2.5

^a 5'-(Methylthio)-5'-deoxyadenosine.

^b 5'-[(3-Aminopropyl)methylsulfonio]-5'-deoxyadenosine.

• **RESIDUE ON IGNITION** (281): NMT 0.1%

SPECIFIC TESTS

• **ISOMERIC RATIO**

Buffer: Transfer 4.2 g of citric acid and 2.03 g of sodium dihydrogen phosphate dihydrate to a 1-L volumetric flask, and then dissolve in and dilute with water to volume.

Mobile phase: Transfer 4.0 g of sodium dodecyl sulfate to a 1-L volumetric flask, dissolve in 560 mL of *Buffer*, and add 440 mL of acetonitrile. Dilute with *Buffer* to volume.

Standard solution: 0.1 mg/mL of USP S-Adenosyl-L-methionine 1,4-Butanedisulfonate RS in water

Sample solution: 0.1 mg/mL of S-Adenosyl-L-methionine 1,4-Butanedisulfonate in water

Chromatographic system

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

Mode: LC

Detector: UV 254 nm

Column: 4.6-mm × 15-cm; 3-µm packing L1

Column temperature: 25°

Flow rate: 0.8 mL/min

Injection volume: 20 µL

Run time: NLT 3 times the retention time of S,S-isomer

System suitability

Samples: *Standard solution* and *Sample solution*

[NOTE—The relative retention times of R,S-isomer and S,S-isomer are 0.95 and 1.00, respectively.]

Suitability requirements

Resolution: NLT 1.5 between the R,S-isomer and S,S-isomer

Analysis

Samples: *Standard solution* and *Sample solution*
Identify the peaks due to the R,S- and S,S-isomers of S-adenosyl-L-methionine in the *Sample solution* by comparing with the *Standard solution*.
Calculate the percentage of the S,S-isomer:

$$\text{Result} = [r_{SS}/(r_{SS} + r_{RS})] \times 100$$

r_{SS} = peak area of the *S,S*-isomer in the *Sample solution*
 r_{RS} = peak area of the *R,S*-isomer in the *Sample solution*

Acceptance criteria: NLT 60.0% and NLT the labeled amount of the *S,S*-isomer

• **PH** <791>

Sample solution: 4.0 mg/mL

Acceptance criteria: 1.8–2.5

• **WATER DETERMINATION** <921>, *Method I*

Analysis: Dissolve a suitable amount of a sample in a mixture of formamide and methanol (1:1) and perform the test with the reagent without pyridine.

Acceptance criteria: NMT 3.0%

ADDITIONAL REQUIREMENTS

- **PACKAGING AND STORAGE:** Preserve in tight, light-resistant containers, and store in a refrigerator.
- **LABELING:** Label it to indicate the minimum content, as a percentage, of *S,S*-isomer.
- **USP REFERENCE STANDARDS** <11>
 - USP Adenine RS
 - USP S-Adenosyl-L-homocysteine RS
 - USP S-Adenosyl-L-methionine 1,4-Butanedisulfonate RS
 - USP 1,4-Butanedisulfonic Acid RS▲ (USP 1-Aug-2021)

Official