System suitability
Sample: *Standard solution*
Suitability requirements
Tailing factor: NMT 2.0 for the glucosamine peak
Column efficiency: NLT 1500 theoretical plates
Relative standard deviation: NMT 2.0%
Analysis
Samples: *Standard solution* and *Sample solution*
Calculate the percentage of the labeled amount of glucosamine ($C_6H_{13}NO_5$) in the portion of Tablets taken:

$$\text{Result} = (r_U/r_S) \times (C_S/C_U) \times (M_{r1}/M_{r2}) \times 100$$

r_U = peak response of glucosamine from the *Sample solution*
r_S = peak response of glucosamine from the *Standard solution*
C_S = concentration of USP Glucosamine Hydrochloride RS in the *Standard solution* (mg/mL)
C_U = nominal concentration of glucosamine in the *Sample solution* (mg/mL)
M_{r1} = molecular weight of glucosamine, 179.17
M_{r2} = molecular weight of glucosamine hydrochloride, 215.63

Acceptance criteria: 90.0%–110.0%

PERFORMANCE TESTS

- **DISINTEGRATION AND DISSOLUTION OF DIETARY SUPPLEMENTS ⟨2040⟩:** Meet the requirements for *Dissolution*
Medium: Water; 900 mL
Apparatus 2: 75 rpm
Time: 60 min
Standard solution: Dissolve a suitable amount of USP Glucosamine Hydrochloride RS in water to obtain a concentration similar to that expected in the *Sample solution*.
Sample solution: Filtered portion of the solution under test
Buffer: Mix 1.0 mL of phosphoric acid with 2 L of water, and adjust with potassium hydroxide to a pH of 3.0.
Mobile phase: Acetonitrile and *Buffer* (2:3)
Chromatographic system
(See *Chromatography* ⟨621⟩, *System Suitability*.)
Mode: LC
Detector: UV 195 nm
Column: 4.6-mm × 25-cm; packing L7
Flow rate: 0.6 mL/min
Injection size: 10 µL
System suitability
Sample: *Standard solution*
Suitability requirements
Tailing factor: NMT 2.0 for the glucosamine peak
Relative standard deviation: NMT 2.0%
Analysis
Samples: *Standard solution* and *Sample solution*
Calculate the percentage of the labeled amount of glucosamine ($C_6H_{13}NO_5$) dissolved:

$$\text{Result} = (r_U/r_S) \times (C_S \times V/L) \times (M_{r1}/M_{r2}) \times 100$$

r_U = peak area from the *Sample solution*
r_S = peak area from the *Standard solution*
C_S = concentration of USP Glucosamine Hydrochloride RS in the *Standard solution* (mg/mL)
V = volume of *Medium*, 900 mL
L = labeled amount of glucosamine (mg/Tablet)
M_{r1} = molecular weight of glucosamine, 179.17
M_{r2} = molecular weight of glucosamine hydrochloride, 215.63

Tolerances: NLT 75% of the labeled amount of glucosamine ($C_6H_{13}NO_5$) is dissolved.
- **WEIGHT VARIATION OF DIETARY SUPPLEMENTS ⟨2091⟩:** Meet the requirements

ADDITIONAL REQUIREMENTS

- **PACKAGING AND STORAGE:** Preserve in tight, light-resistant containers.
- **LABELING:** The label indicates the type of glucosamine salt contained in the article.
- **USP REFERENCE STANDARDS ⟨11⟩**
USP Glucosamine Hydrochloride RS

Glucosamine Sulfate Potassium Chloride

$(C_6H_{14}NO_5)_2SO_4 \cdot 2KCl$ 605.52
Bis(D-glucose, 2-amino-2-deoxy-), sulfate potassium chloride complex;
Bis(2-amino-2-deoxy-β-D-glucopyranose) sulfate potassium chloride complex (–,–) [38899-05-7].

DEFINITION

Glucosamine Sulfate Potassium Chloride contains NLT 98.0% and NMT 102.0% of glucosamine sulfate potassium chloride [$(C_6H_{14}NO_5)_2SO_4 \cdot 2KCl$], calculated on the dried basis.

IDENTIFICATION

- **A. INFRARED ABSORPTION ⟨197K⟩**
Sample: Transfer 50 mg of Glucosamine Sulfate Potassium Chloride to a centrifuge tube, and dissolve in 2 mL of water. Add 0.5 mL of barium chloride TS, and centrifuge. Collect the supernatant, and evaporate to dryness. Dry the residue at 105° for 2 h.
Acceptance criteria: The IR spectrum of the *Sample* matches that of a similar preparation of USP Glucosamine Hydrochloride RS, except that the addition of barium chloride TS is omitted.
- **B. IDENTIFICATION TESTS—GENERAL,** *Chloride* ⟨191⟩ and *Potassium* ⟨191⟩**:** Meets the requirements
- **C.** The retention time of the glucosamine peak of the *Sample solution* corresponds to that of the *Standard solution*, as obtained in the *Assay*.
- **D. SULFATE:** In the test for *Content of Sulfate*, after the addition of barium chloride TS a white precipitate is formed.

ASSAY

- **PROCEDURE**
Buffer: In a 1-L volumetric flask, dissolve 3.5 g of dibasic potassium phosphate in water, add 0.25 mL of ammonium hydroxide, dilute with water to volume, and mix. Adjust with phosphoric acid to a pH of 7.5.
Mobile phase: Acetonitrile and *Buffer* (75:25)
Diluent: Acetonitrile and water (50:50)
Standard solution: 3.8 mg/mL of USP Glucosamine Hydrochloride RS in *Diluent*. Shake for 5 min by mechanical means to completely dissolve.
Sample solution: Transfer 263 mg of Glucosamine Sulfate Potassium Chloride to a 50-mL volumetric flask. Dissolve in 30 mL of *Diluent*, and shake by mechanical means. Dilute with *Diluent* to volume.
Chromatographic system
(See *Chromatography* ⟨621⟩, *System Suitability*.)
Mode: LC
Detector: UV 195 nm
Column: 4.6-mm × 15-cm; 5-µm packing L8
Column temperature: 35°
Flow rate: 1.5 mL/min
Injection size: 10 µL
System suitability
Sample: *Standard solution*
[NOTE—The peak for the glucosamine moiety elutes at about 10 min. The chromatogram shows additional peaks near the void volume, due to the counter ions.]

Suitability requirements
Tailing factor: NMT 2.0 for the glucosamine peak
Efficiency: NLT 1500 theoretical plates
Relative standard deviation: NMT 2.0%.
Analysis
Samples: *Standard solution* and *Sample solution*
Calculate the percentage of glucosamine sulfate potassium chloride [$(C_6H_{14}NO_5)_2SO_4 \cdot 2KCl$] in the portion of Glucosamine Sulfate Potassium Chloride taken:

$$\text{Result} = (r_U/r_S) \times (C_S/C_U) \times (M_{r1}/M_{r2}) \times 100$$

r_U = peak response from the *Sample solution*
r_S = peak response from the *Standard solution*
C_S = concentration of USP Glucosamine Hydrochloride RS in the *Standard solution* (mg/mL)
C_U = concentration of Glucosamine Sulfate Potassium Chloride in the *Sample solution* (mg/mL)
M_{r1} = molecular weight of glucosamine sulfate potassium chloride, 605.52
M_{r2} = twice the molecular weight of glucosamine hydrochloride, 431.26
Acceptance criteria: 98.0%–102.0% on the dried basis

OTHER COMPONENTS

- **CONTENT OF SULFATE**
Sample: 1 g of Glucosamine Sulfate Potassium Chloride
Analysis: Transfer the *Sample* to a 250-mL beaker, and dissolve in 100 mL of water. Add 4 mL of 6 N hydrochloric acid. Heat the solution to boiling, and add, with constant stirring, sufficient boiling barium chloride TS to completely precipitate the sulfate. Add an additional 2 mL of barium chloride TS, and digest on a steam bath for 1 h. Pass the mixture through ashless filter paper. Transfer the residue quantitatively to a new filter, and wash the residue with hot water until no precipitate is obtained when 1 mL of silver nitrate TS is added to 5 mL of washing. Transfer the paper containing the residue to a tared crucible. Char the paper, without burning, and ignite the crucible and its contents to constant weight. Calculate the content of sulfate by multiplying the weight obtained by 0.4116.
Acceptance criteria: 15.5%–16.5%

IMPURITIES

- **RESIDUE ON IGNITION ⟨281⟩:** 26.5%–31.0%
- **SODIUM:** A solution (1 in 10), tested on a platinum wire, does not impart a pronounced yellow color to a nonluminous flame.
- **ARSENIC,** *Method II* ⟨**211**⟩**:** NMT 3 µg/g
- **HEAVY METALS,** *Method II* ⟨**231**⟩**:** NMT 10 ppm

SPECIFIC TESTS

- **OPTICAL ROTATION,** *Specific Rotation* ⟨**781S**⟩
Sample solution: 35 mg/mL. Measure the specific rotation 3 h after preparation.
Acceptance criteria: +47.0° to +53.0°
- **pH** ⟨**791**⟩
Sample solution: 20 mg/mL
Acceptance criteria: 3.0–5.0
- **LOSS ON DRYING** ⟨**731**⟩**:** Dry a sample at 105° for 2 h: it loses NMT 1.0% of its weight.

ADDITIONAL REQUIREMENTS

- **PACKAGING AND STORAGE:** Preserve in tight, light-resistant containers.
- **USP REFERENCE STANDARDS** ⟨**11**⟩
USP Glucosamine Hydrochloride RS

Glucosamine Sulfate Sodium Chloride

$(C_6H_{14}NO_5)_2SO_4 \cdot 2NaCl$ 573.31
Bis(D-glucose, 2-amino-2-deoxy-), sulfate sodium chloride complex;
Bis(2-amino-2-deoxy-β-D-glucopyranose) sulfate sodium chloride complex (–,–) [38899-05-7].

DEFINITION

Glucosamine Sulfate Sodium Chloride contains NLT 98.0% and NMT 102.0% of glucosamine sulfate sodium chloride [$(C_6H_{14}NO_5)_2SO_4 \cdot 2NaCl$], calculated on the dried basis.

IDENTIFICATION

- **A. INFRARED ABSORPTION** ⟨**197K**⟩
Sample: Transfer 50 mg of Glucosamine Sulfate Sodium Chloride to a centrifuge tube, and dissolve in 2 mL of water. Add 0.5 mL of barium chloride TS, and centrifuge. Collect the supernatant, and evaporate to dryness. Dry the residue at 105° for 2 h.
Acceptance criteria: The IR spectrum of the *Sample* matches that of a similar preparation of USP Glucosamine Hydrochloride RS, except that the addition of barium chloride TS is omitted.
- **B. IDENTIFICATION TESTS—GENERAL,** *Chloride* ⟨**191**⟩ and *Sodium* ⟨**191**⟩**:** Meets the requirements
- **C.** The retention time of the glucosamine peak of the *Sample solution* corresponds to that of the *Standard solution*, as obtained in the *Assay*.
- **D. SULFATE:** In the test for *Content of Sulfate*, after the addition of barium chloride TS a white precipitate is formed.

ASSAY

- **PROCEDURE**
Buffer: In a 1-L volumetric flask, dissolve 3.5 g of dibasic potassium phosphate in water, add 0.25 mL of ammonium hydroxide, dilute with water to volume, and mix. Adjust with phosphoric acid to a pH of 7.5.
Mobile phase: Acetonitrile and *Buffer* (75:25)
Diluent: Acetonitrile and water (50:50)
Standard solution: 3.8 mg/mL of USP Glucosamine Hydrochloride RS in *Diluent*. Shake for 5 min by mechanical means to completely dissolve.
Sample solution: Transfer 250 mg of Glucosamine Sulfate Sodium Chloride to a 50-mL volumetric flask. Dissolve in 30 mL of *Diluent,* and shake by mechanical means. Dilute with *Diluent* to volume.
Chromatographic system
(See *Chromatography* ⟨621⟩*, System Suitability*.)
Mode: LC
Detector: UV 195 nm
Column: 4.6-mm × 15-cm; 5-µm packing L8
Column temperature: 35°
Flow rate: 1.5 mL/min
Injection size: 10 µL
System suitability
Sample: *Standard solution*
[NOTE—The peak for the glucosamine moiety elutes at about 10 min. The chromatogram shows additional peaks near the void volume, due to the counter ions.]