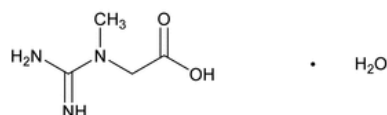


Status: Currently Official on 14-Feb-2025  
 Official Date: Official as of 01-Nov-2020  
 Document Type: Dietary Supplement Monographs  
 DocId: GUID-FB300FEA-1F49-49E9-AEF0-EDB7C7165AC9\_2\_en-US  
 DOI: [https://doi.org/10.31003/USPNF\\_M20457\\_02\\_01](https://doi.org/10.31003/USPNF_M20457_02_01)  
 DOI Ref: rml6h

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Add the following:

## ^Creatine



$C_4H_9N_3O_2 \cdot H_2O$  149.15

Glycine, *N*-(aminoiminomethyl)-*N*-methyl-, monohydrate;

*N*-Carbamimidoyl-*N*-methylglycine monohydrate CAS RN®: 6020-87-7.

$C_4H_9N_3O_2$  131.14

Anhydrous CAS RN®: 57-00-1.

### DEFINITION

Creatine contains one molecule of water of hydration. It contains NLT 98.0% and NMT 102.0% of creatine ( $C_4H_9N_3O_2$ ), calculated on the dried basis.

### IDENTIFICATION

- **A.** [SPECTROSCOPIC IDENTIFICATION TESTS \(197K\)](#)
- **B.** The retention time of the major peak of the *Sample solution* corresponds to that of the *Standard solution*, as obtained in the Assay.

### ASSAY

#### • PROCEDURE

**Mobile phase:** Dissolve 23 g of [monobasic ammonium phosphate](#) in 1 L of [water](#) and adjust with [phosphoric acid](#) to a pH of 4.0.

**Diluent:** [Water](#)

**System suitability solution:** 0.1 mg/mL of [USP Creatine RS](#), 0.1 mg/mL of [USP Creatinine RS](#), and 0.002 mg/mL of dicyandiamide<sup>1</sup> in *Diluent*

**Standard solution:** 2.5 mg/mL of [USP Creatine RS](#) in *Diluent*

**Sample solution:** 2.5 mg/mL of Creatine in *Diluent*

#### Chromatographic system

(See [Chromatography \(621\)](#), [System Suitability](#).)

**Mode:** LC

**Detector:** UV 225 nm

**Column:** 4.0-mm × 25-cm; 5-μm packing [L9](#)

**Column temperature:** 25°

**Flow rate:** 1.0 mL/min

**Injection volume:** 20 μL

#### System suitability

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

[NOTE—The relative retention times for dicyandiamide, creatine, and creatinine are about 0.8, 1.0, and 2.0, respectively, *System suitability solution*.]

#### Suitability requirements

**Resolution:** NLT 2.0 between dicyandiamide and creatine, *System suitability solution*

**Column efficiency:** NLT 1000 theoretical plates, *Standard solution*

**Relative standard deviation:** NMT 2.0%, *Standard solution*

#### Analysis

**Samples:** *Standard solution* and *Sample solution*

Calculate the percentage of creatine ( $C_4H_9N_3O_2$ ) in the portion of Creatine taken:

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

$r_U$  = peak response of creatine from the *Sample solution*

$r_S$  = peak response of creatine from the *Standard solution*

$C_S$  = concentration of [USP Creatine RS](#) in the *Standard solution* (mg/mL)

$C_U$  = concentration of Creatine in the *Sample solution* (mg/mL)

**Acceptance criteria:** 98.0%–102.0% on the dried basis

#### IMPURITIES

• [RESIDUE ON IGNITION \(281\)](#): NMT 0.1%

• [CHLORIDE AND SULFATE \(221\)](#), *Sulfate*

**Standard solution:** 0.10 mL of 0.020 M sulfuric acid

**Sample:** 200 mg of Creatine

**Acceptance criteria:** NMT 0.1%

#### PROCESS-RELATED IMPURITIES

[NOTE—On the basis of information regarding the manufacturing process, perform either *Procedure 1* or *Procedure 2*.]

##### • PROCEDURE 1

**Mobile phase, Diluent, System suitability solution, Chromatographic system, and System suitability:** Proceed as directed in the Assay.

**Standard solution:** 2.5 µg/mL of [USP Creatine RS](#) and 2.5 µg/mL of [USP Creatinine RS](#) in *Diluent*

**Sample solution:** 2.5 mg/mL of Creatine in *Diluent*

##### Analysis

**Samples:** *Standard solution* and *Sample solution*

Calculate the percentage of creatinine ( $C_4H_7N_3O$ ) in the portion of Creatine taken:

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

$r_U$  = peak response of creatinine from the *Sample solution*

$r_S$  = peak response of creatinine from the *Standard solution*

$C_S$  = concentration of [USP Creatinine RS](#) in the *Standard solution* (mg/mL)

$C_U$  = concentration of Creatine in the *Sample solution* (mg/mL)

Separately calculate the percentage of dicyandiamide and dihydrotriazine in the portion of Creatine taken:

$$\text{Result} = (r_U/r_S) \times (C_S/C_U) \times (1/F) \times 100$$

$r_U$  = peak response of dicyandiamide or dihydrotriazine from the *Sample solution*

$r_S$  = peak response of creatine from the *Standard solution*

$C_S$  = concentration of [USP Creatine RS](#) in the *Standard solution* (mg/mL)

$C_U$  = concentration of Creatine in the *Sample solution* (mg/mL)

$F$  = relative response factor (see [Table 1](#))

Calculate the percentage of any unspecified impurity in the portion of Creatine taken:

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

$r_U$  = peak response of any unspecified impurity from the *Sample solution*

$r_S$  = peak response of creatine from the *Standard solution*

$C_S$  = concentration of [USP Creatine RS](#) in the *Standard solution* (mg/mL)