P = percentage of procyanidines in the USP Maritime Pine Extract RS
Acceptance criteria: 65%–75% on the dried basis

CONTAMINANTS

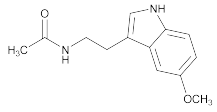
Delete the following:

•• **BOTANICAL EXTRACTS,** *Heavy Metals* ⟨**565**⟩**:** Meets the requirements• (Official 1-Jan-2018)
• **ARTICLES OF BOTANICAL ORIGIN,** *Pesticide Residue* ⟨**561**⟩**:** Meets the requirements
• **MICROBIAL ENUMERATION TESTS** ⟨**2021**⟩**:** The total aerobic microbial count does not exceed 10^4 cfu/g, and the total combined molds and yeasts count does not exceed 10^3 cfu/g.
• **ABSENCE OF SPECIFIED MICROORGANISMS** ⟨**2022**⟩**:** It meets the requirements of the tests for absence of *Salmonella* species and *Escherichia coli*.

SPECIFIC TESTS
• **LOSS ON DRYING** ⟨**731**⟩**:** Dry 1.0 g of Extract for 3 h at 110°: it loses NMT 8.0% of its weight.
• **ARTICLES OF BOTANICAL ORIGIN,** *Total Ash* ⟨**561**⟩**:** NMT 0.7%
• **LIMIT OF WATER-INSOLUBLE SUBSTANCES**
Analysis: Weigh 0.50 g of Extract, and stir in 50 mL of water at 20° for 15 min. Pass through a fine sintered glass filter, previously weighed. Dry the filter at 110° for 3 h, cool to room temperature, and weigh the filter. Calculate the amount of water-insoluble material.
Acceptance criteria: NMT 10% of the amount of Extract taken

ADDITIONAL REQUIREMENTS
• **PACKAGING AND STORAGE:** Preserve in tight containers, and store at 25°, excursion permitted between 15° and 30°. Protect from light.
• **LABELING:** The label states the Latin binomial and, following the official name of the article, the part of the plant from which the article was prepared, in addition to the information required for *Botanical Extracts* ⟨565⟩, *Labeling*.
• **USP REFERENCE STANDARDS** ⟨**11**⟩
USP Maritime Pine Extract RS

Melatonin



$C_{13}H_{16}N_2O_2$ 232.28
N-Acetyl-5-methoxytryptamine;
N-(2-(5-Methoxy-1*H*-indol-3-yl)ethyl) acetamide [73-31-4].

DEFINITION
Melatonin contains NLT 98.5% and NMT 101.5% of melatonin ($C_{13}H_{16}N_2O_2$), calculated on the dried basis.

IDENTIFICATION
• **A. INFRARED ABSORPTION** ⟨**197K**⟩
• **B. ULTRAVIOLET ABSORPTION** ⟨**197U**⟩
Analytical wavelength: 277 nm
Sample solution: 10 µg/mL of Melatonin in isopropyl alcohol
Acceptance criteria: Meets the requirements. Absorptivities, calculated on the dried basis, do not differ by more than 3.0%.
• **C. HPLC IDENTIFICATION TEST**
Analysis: Proceed as directed in the *Assay*.
Acceptance criteria: The retention time of the major peak of the *Sample solution* corresponds to that of the *Standard solution*.

ASSAY
• **PROCEDURE**
Buffer: 0.5 g/L of monobasic potassium phosphate in water. Adjust with phosphoric acid to a pH of 3.5, and filter.
Mobile phase: Acetonitrile and *Buffer* (25:75)
System suitability solution: 0.1 mg/mL of USP Melatonin RS and 0.02 mg/mL USP Melatonin Related Compound A RS in *Mobile phase*
Standard solution: 0.1 mg/mL of USP Melatonin RS in *Mobile phase*
Sample solution: 0.1 mg/mL of Melatonin in *Mobile phase*
Chromatographic system
(See *Chromatography* ⟨621⟩, *System Suitability*.)
Mode: LC
Detector: UV 222 nm
Column: 4.6-mm × 15-cm; 5-µm packing L1
Flow rate: 1.0 mL/min
Injection size: 10 µL
System suitability
Samples: *System suitability solution* and *Standard solution*
[NOTE—The relative retention times for melatonin related compound A and melatonin are 0.4 and 1.0, respectively.]
Suitability requirements
Resolution: NLT 4 between melatonin and melatonin related compound A, *System suitability solution*
Relative standard deviation: NMT 2.0%, *Standard solution*
Analysis
Samples: *Standard solution* and *Sample solution*
Calculate the percentage of melatonin in the portion of Melatonin taken:

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

r_U = peak response from the *Sample solution*
r_S = peak response from the *Standard solution*
C_S = concentration of USP Melatonin RS in the *Standard solution* (mg/mL)
C_U = concentration of Melatonin in the *Sample solution* (mg/mL)
Acceptance criteria: 98.5–101.5% on the dried basis

IMPURITIES
• **RESIDUE ON IGNITION** ⟨**281**⟩**:** NMT 0.1%
• **CHLORIDE AND SULFATE,** *Chloride* ⟨**221**⟩
Standard: 0.10 mL of 0.020 N hydrochloric acid
Sample: 0.36 g of Melatonin
Acceptance criteria: NMT 0.02%

Delete the following:

•• **HEAVY METALS** ⟨**231**⟩**:** NMT 20 µg/g• (Official 1-Jan-2018)
• **RELATED COMPOUNDS**
Solution A: Acetonitrile
Solution B: Use *Buffer*, prepared as directed in the *Assay*.
Mobile phase: See *Table 1*.

Table 1

Time (min)	Solution A (%)	Solution B (%)
0	25	75
7	25	75

DS Monographs

Table 1 *(Continued)*

Time (min)	Solution A (%)	Solution B (%)
15	80	20
18	25	75
25	25	75

Diluent: Mixture of *Solution A* and *Solution B* (25:75)
System suitability solution: 0.1 mg/mL of USP Melatonin RS and 0.02 mg/mL of USP Melatonin Related Compound A RS in *Diluent*
Standard solution: 5 µg/mL of USP Melatonin RS in *Diluent*
Sample solution: 1 mg/mL of Melatonin in *Diluent*
Chromatographic system
(See *Chromatography* ⟨621⟩, *System Suitability*.)
Mode: LC
Detector: UV 222 nm
Column: 4.6-mm × 15-cm; 5-µm packing L1
Flow rate: 1.0 mL/min
Injection size: 10 µL
System suitability
Sample: *System suitability solution*
[NOTE—The relative retention times for melatonin related compound A and melatonin are 0.4 and 1.0, respectively.]
Suitability requirements
Resolution: NLT 4.0 between melatonin and melatonin related compound A
Relative standard deviation: NMT 2.0% for the melatonin peak
Analysis
Samples: *Standard solution* and *Sample solution*
Calculate the percentage of each individual impurity in the portion of Melatonin taken:

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

r_U = peak response of each individual impurity from the *Sample solution*
r_S = peak response of melatonin from the *Standard solution*
C_S = concentration of USP Melatonin RS in the *Standard solution* (mg/mL)
C_U = concentration of Melatonin in the *Sample solution* (mg/mL)

Acceptance criteria
Individual impurities: NMT 0.1%
Total impurities: NMT 1.0%

SPECIFIC TESTS

- **LOSS ON DRYING** ⟨**731**⟩
  Analysis: Dry a sample at 80° in a vacuum for 3 h.
  Acceptance criteria: NMT 1.0%

ADDITIONAL REQUIREMENTS

- **PACKAGING AND STORAGE:** Preserve in tight containers, protected from light.
- **USP REFERENCE STANDARDS** ⟨**11**⟩
  USP Melatonin RS
  USP Melatonin Related Compound A RS
  2-(5-Methoxy-1*H*-indol-3-yl)ethanamine.
  $C_{11}H_{14}N_2O$ 190.24

Melatonin Tablets

DEFINITION

Melatonin Tablets contain NLT 90.0% and NMT 110.0% of the labeled amount of melatonin ($C_{13}H_{16}N_2O_2$).

IDENTIFICATION

- **A.** The retention time of the major peak of the *Sample solution* corresponds to that of the *Standard solution,* as obtained in *Strength.*

STRENGTH

- **PROCEDURE**
  Buffer: 0.5 g/L of monobasic potassium phosphate in water. Adjust with phosphoric acid to a pH of 3.5, and filter.
  Mobile phase: Acetonitrile and *Buffer* (25:75)
  System suitability solution: 0.1 mg/mL of USP Melatonin RS and 0.02 mg/mL of USP Melatonin Related Compound A RS in *Mobile phase*
  Standard solution: 0.1 mg/mL of USP Melatonin RS in *Mobile phase*
  Sample solution: Filtered portion of the solution in *Mobile phase*, equivalent to 0.1 mg/mL of Melatonin from NLT 20 finely powdered Tablets
  Chromatographic system
  (See *Chromatography* ⟨621⟩, *System Suitability*.)
  Mode: LC
  Detector: UV 222 nm
  Column: 4.6-mm × 15-cm; 5-µm packing L1
  Flow rate: 1.0 mL/min
  Injection size: 10 µL
  System suitability
  Samples: *System suitability solution* and *Standard solution*
  [NOTE—The relative retention times for melatonin related compound A and melatonin are 0.4 and 1.0, respectively.]
  Suitability requirements
  Resolution: NLT 4 between melatonin and melatonin related compound A, *System suitability solution*
  Relative standard deviation: NMT 2.0%, *Standard solution*
  Analysis
  Samples: *Standard solution* and *Sample solution*
  Calculate the percentage of the labeled amount of melatonin ($C_{13}H_{16}N_2O_2$) in the portion of Tablets taken:

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

r_U = peak response from the *Sample solution*
r_S = peak response from the *Standard solution*
C_S = concentration of USP Melatonin RS in the *Standard solution* (mg/mL)
C_U = nominal concentration of melatonin in the *Sample solution* (mg/mL)

Acceptance criteria: 90.0%–110.0%

PERFORMANCE TESTS

- **DISINTEGRATION AND DISSOLUTION OF DIETARY SUPPLEMENTS** ⟨**2040**⟩**:** Meet the requirements for *Dissolution*
  Medium: Water; 500 mL
  Apparatus 2: 50 rpm
  Time: 30 min
  Standard solution: Dissolve a suitable amount of USP Melatonin RS in water to obtain a concentration similar to that expected in the *Sample solution*.
  Sample solution: Filtered portion of the solution under test
  Analysis: Proceed as directed in *Strength*, making any necessary adjustments.
  Calculate the percentage of the labeled amount of melatonin ($C_{13}H_{16}N_2O_2$) dissolved:

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

r_U = peak area from the *Sample solution*
r_S = peak area from the *Standard solution*
C_S = concentration of USP Melatonin RS in the *Standard solution* (mg/mL)
V = volume of *Medium*, 500 mL
L = labeled amount of melatonin (mg/Tablet)

DS Monographs