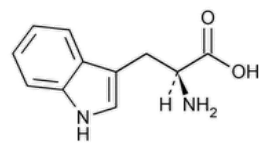


Status: Currently Official on 17-Feb-2025
Official Date: Official as of 01-Jun-2023
Document Type: USP Monographs
DocId: GUID-BE4CF742-3682-4138-802E-634E4AFD4B1D_6_en-US
DOI: https://doi.org/10.31003/USPNF_M86750_06_01
DOI Ref: r9pju

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Tryptophan



$C_{11}H_{12}N_2O_2$ 204.23
L-Tryptophan CAS RN®: 73-22-3; UNII: 8DUH1N11BX.

DEFINITION
Tryptophan contains NLT 98.5% and NMT 101.5% of $C_{11}H_{12}N_2O_2$, as L-tryptophan, calculated on the dried basis.

IDENTIFICATION
• [SPECTROSCOPIC IDENTIFICATION TESTS \(197\)](#), [Infrared Spectroscopy: 197K](#)

ASSAY
• **PROCEDURE**
Sample solution: Place 200 mg of Tryptophan in a 125-mL flask. Dissolve in a mixture of 3 mL of formic acid and 50 mL of glacial acetic acid.
Analysis: Titrate with 0.1 N perchloric acid VS, determining the endpoint potentiometrically. Perform a blank determination, and make any necessary correction (see [Titrimetry \(541\)](#)). Each mL of 0.1 N perchloric acid is equivalent to 20.42 mg of $C_{11}H_{12}N_2O_2$.
Acceptance criteria: 98.5%–101.5% on the dried basis

IMPURITIES
INORGANIC IMPURITIES
• [RESIDUE ON IGNITION \(281\)](#): NMT 0.1%
• [CHLORIDE AND SULFATE, Chloride\(221\)](#): A 0.73-g portion shows no more chloride than corresponds to 0.50 mL of 0.020 N hydrochloric acid (0.05%). [NOTE—Gently heat the sample preparation to dissolve, if necessary.]
• [CHLORIDE AND SULFATE, Sulfate\(221\)](#): A 0.33-g portion shows no more sulfate than corresponds to 0.10 mL of 0.020 N sulfuric acid (0.03%). [NOTE—Gently heat the sample preparation to dissolve, if necessary.]

Change to read:
• [▲IRON \(241\), Procedures, Procedure 1▲](#) (CN 1-JUN-2023) : NMT 30 ppm

ORGANIC IMPURITIES
• **PROCEDURE 1**
Solution A: Trifluoroacetic acid in water (1 mL/L)
Solution B: Trifluoroacetic acid in an acetonitrile and water solution (80:20) (1 mL/L trifluoroacetic acid solution)
Standard solution: 1.0 mg/L each of [USP Tryptophan Related Compound A RS](#) and [USP Tryptophan Related Compound B RS](#) in water
Sample solution: 10.0 mg/mL of tryptophan in water
System suitability solution: 1.0 mg/L of [USP Tryptophan Related Compound B RS](#) in water
Mobile phase: See the gradient table below.

Time (min)	Solution A (%)	Solution B (%)
0	95	

Time (min)	Solution A (%)	Solution B (%)
2	95	5
37	35	65
42	0	100
47	0	100
50	95	5
60	95	5

Chromatographic system(See [Chromatography \(621\), System Suitability.](#))**Mode:** LC**Detector:** UV 220 nm**Column:** 4.6-mm × 25-cm; 5-μm packing L1**Column temperature:** 30°**Flow rate:** 1 mL/min**Injection size:** 20 μL**System suitability****Sample:** System suitability solution**Suitability requirement****Relative standard deviation:** NMT 5.0%**Analysis****Samples:** Standard solution and Sample solution

Calculate the percentage of each unspecified impurity in the portion of Tryptophan taken:

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

 r_U = peak area of each unspecified impurity in the *Sample solution* r_S = peak area of tryptophan related compound B in the *Standard solution* C_S = concentration of [USP Tryptophan Related Compound B RS](#) in the *Standard solution* (μg/mL) C_U = concentration of Tryptophan in the *Sample solution* (μg/mL)**Acceptance criteria****Total impurities 1:** NMT 0.01% of the total impurities eluting prior to the tryptophan peak**Total impurities 2:** NMT 0.03% of the total impurities eluting after the tryptophan peak. [NOTE—Exclude the peak for tryptophan related compound B.]**Tryptophan related compound A:** If a peak for tryptophan related compound A is observed in the *Sample solution*, then perform the test for Procedure 2: Limit of Tryptophan Related Compound A, below.**• PROCEDURE 2: LIMIT OF TRYPTOPHAN RELATED COMPOUND A****Solution A:** 18 mM monobasic sodium phosphate, filtered and degassed (pH 2.5), and acetonitrile (9:1)**Solution B:** 10 mM monobasic sodium phosphate, filtered and degassed (pH 2.5), and acetonitrile (1:1)**Solution C:** Acetonitrile in water (7:3)**Standard solution:** 0.1 mg/L of [USP Tryptophan Related Compound A RS](#) in water**Sample solution:** 10.0 mg/mL of Tryptophan in water