

3050 Spruce Street
Saint Louis, Missouri 63103 USA
Telephone 800-325-5832 • (314) 771-5765
Fax (314) 286-7828
email: techserv@sial.com
sigma-aldrich.com

ProductInformation

Ninhydrin Reagent solution

Product Number **N 7285** Storage Temperature 2-8 °C

Product Description

Product No. N 7285 is a replacement for Product No. N 1632, which is no longer available. This reagent is a solution of ninhydrin and hydrindantin in DMSO and lithium acetate buffer, pH 5.2.

It may be used for the determination of the concentration of most primary and secondary amines and amino acids in solution. A test for sensitivity to amino acids has been published. Methods for use of this reagent in both manual^{2,3} and automated⁴ systems have been published.

This product is not intended for use as a chromatography spray or dipping reagent.

Precautions and Disclaimer

For Laboratory Use Only. Not for drug, household or other uses.

This product is provided as a burgundy liquid. It is sensitive to air and will oxidize to a yellow color at which time it should not be used. Store protected from light and under nitrogen or argon gas.

Procedure

General procedure for the manual determination of amino acids using Product No. N 7285 Ninhydrin Reagent.

- 1. Prepare standard curve.
 - a. Make stock solution of an amino acid at a concentration of 0.05 μ moles/ml (50 μ M) in 0.05% glacial acetic acid.
 - b. Prepare five tubes as follows (volumes in ml):

Tube	1	2	3	4	5
Standard	0.0	0.5	1.0	1.5	2.0
Water	2.0	1.5	1.0	0.5	0.0
Ninhydrin	1.0	1.0	1.0	1.0	1.0
reagent					

2. Prepare samples, substituting 2.0 ml of sample in place of standard.

For both standard and samples:

- 3. Mix contents of standard and sample tubes gently.
- 4. Place into boiling water bath for 10 minutes exactly.
- 5. Cool to room temperature.
- 6. Add 5 ml of 95% ethanol to each tube.
- 7. Read absorbance at 570 nm.
 - a. For standard solution: Read the absorbance at 570 nm of tubes
 2 to 5 versus tube 1, as the reference for the standard curve. If A₅₇₀ exceeds 1.0, dilute samples with 95% ethanol.
 - For sample solutions: Read tubes versus tube 1, and record the absorbance at 570 nm. Amino acid concentration is determined by comparison of the A₅₇₀ readings to the standard curve.

Note: This procedure is offered for informational purposes only. This assay is not considered a GMP/GLP assay and should not be used as such.

References

- 1. Reagent Chemicals, 9th ed., Oxford University Press (New York, NY: 2000), pp. 438-439.
- Moore, S., et al., Chromatography of amino acids on sulfonated polystyrene resins. An improved system. Anal. Chem., 30, 1185-1190 (1958).
- Moore, S. Amino acid analysis: Aqueous dimethyl sulfoxide as solvent for the ninhydrin reaction. J. Biol. Chem., 243, 6281-6283 (1968).
- 4. Spackman, D. H., et al., Automatic recording apparatus for use in chromatography of amino acids. Anal. Chem., **30**, 1190-1206, (1958).

IRB/RXR 10/03