



<b>ATF-LS-14 Ninhydrin</b>	Published Online: <b>March 2018</b>
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I. **Scope:** Ninhydrin, or tri-keto-hydrindene hydrate, is an extremely sensitive indicator of alpha-amino acids, proteins, peptides and polypeptides. The reaction produces a violet to blue-violet coloring of these substances and is effective with older deposits with even minute amounts of amino acids. While ninhydrin can be used on any surface, normally processing is confined to porous items which have not subsequently become water-soaked or do not contain inherent animal proteins.

II. **References:**

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Cowger, James F. Friction Ridge Skin Comparison and Identification of Fingerprints; Boca Raton: CRC Press, 1993.

Lee, Henry C.; Gaensslen, R.E., eds. Advances in Fingerprint Technology 2<sup>nd</sup> Edition; CRC Press: Boca Raton, 2001.

Lennard, Christopher J.; Pierre A. Margot; "Sequencing of Reagents for the Improved visualization of Latent Fingerprints"; Journal of Forensic Identification, September/October 1988, 38, 5, 197-210.

Kent, Terry, ed. Fingerprint Development Techniques; Heanor Gate Publisher; Derbyshire, England, 1993.

Olson, Robert; Scott's Fingerprint Mechanics; Charles C. Thomas Publisher; Springfield, IL, 1978.

Pounds, C.A.; R.J. Jones; "Physicochemical Techniques in Development of Latent Fingerprints; Trends in Analytical Chemistry, 1983, 2, 8, 180-183.

Trozzi, Timothy et al; "FBI Processing Guide for Developing Latent Prints, 2000

III. **Apparatus/Reagents:**

- Fume Hood
- Preparations:
- Recommended preparation – 0.05%

**Petroleum Ether:**

1. Dissolve 5 grams of ninhydrin in 10 milliliters (ml) of ethanol or methanol
2. When crystals are completely dissolved slowly add 90 milliliters of ethyl acetate
3. Add 900 milliliters of petroleum ether or other carrier
4. Allow to stand for five to ten minutes. Two separate layers will form, a large pale yellow layer on top of a smaller darker yellow layer. The darker layer is discarded and the pale yellow layer is the working solution.

**Alternate formula:**

1. Dissolve 5 grams of ninhydrin in 30 ml of ethanol or methanol
2. Add 40 ml of isopropanol
3. Add 930 ml of petroleum ether or other carrier

**Acetone formula:**

1. Dissolve 6 grams ninhydrin in 1 liter acetone

Instrumentation: Environmental chambers should be used to control the heat and relative humidity that the item of evidence is exposed to after processing. The chambers should be calibrated periodically with a hygrometer and thermometer to insure that the chamber is maintaining the proper level of relative humidity and temperature.

**IV. Safety Precautions:** This procedure involves the use of hazardous materials. This procedure does not purport to address all of the safety problems associated with its use. It is the responsibility of the user of this procedure to establish appropriate safety and health practices and determine the applicability of regulatory limitations before use. Proper caution should be exercised and the use of personal protective equipment should be utilized to avoid exposure to dangerous chemicals. Consult the appropriate MSDS for each chemical before use. Preparation and evidence processing should be performed in a fume hood.

**V. Procedures:**

Dipping (preferred method of application)

1. In a tray large enough to accommodate the evidence, pour enough working solution to cover all of the items.
2. Completely immerse each item to be processed in the working solution until the item is completely saturated, usually five seconds or less. The item can be manipulated using tongs or forceps.
3. Remove and allow the item to dry completely.
4. Place the item in the heat/humidity chamber at no greater than 80 degrees centigrade and between 60% and 80% relative humidity.
5. Check the item periodically to monitor the impression development. Care should be taken not to saturate the item with water vapor.
6. Remove the item from the heat/humidity chamber and photograph any developed impressions using an appropriate photographic preservation technique.

Alternate application methods

Brushing, Spraying, or use of a squirt bottle

Larger Items that will not fit conveniently into processing trays should be painted with the ninhydrin solution using a soft bristle brush. Two inch to four-inch nylon paintbrushes are adequate. Care must be taken to apply an even and thorough amount to all surfaces. Applying ninhydrin via aerosolized spray cans or squirt bottles to items of evidence is also permissible.

Additional formulas are available for use (commercial and manual preparation) and are widely accepted.

**VI. Quality Assurance/Quality Control:** The standards and controls for the ninhydrin procedure consist of placing test impressions on porous items to make test strips. The test strips are then immersed in the working solution and subjected to the proper level of humidity. If impressions are visualized the working solution can be used to process evidence. Tests for each working solution shall be documented using the appropriate reagent log.