

	LPU-PM 3a.2 Amido Black	
	<i>Document #: 2591</i>	<i>Page 1 of 4</i>
	<i>Revision #: 2</i>	<i>Issued Date: 08/20/2020</i>
	<i>Document Manager: Tracee McIntosh</i>	<i>Approved By: Ryan Larrison</i>

3a.2 Amido Black

3a.2.1 Introduction

Amido black or naphthalene black 10B is a protein indicator that may be used to react with proteins present in suspected blood. Amido black is recommended for non-porous items but may be used on a porous item in certain circumstances. Amido black can be applied after cyanoacrylate fuming in many cases (see McCarthy and Grieve, 1989).

3a.2.2 Safety Considerations

3a.2.2.1

Application of methanol based amido black and the rinse solutions should be performed in a fume hood.

3a.2.2.2

Standard safety precautions from the SDS available at each worksite for the components in this protocol shall be followed.

3a.2.3 Storage

The methanol based solution shall be stored in a clear or dark glass stoppered bottle. The water base solution may be stored in a clear or dark glass or plastic stoppered bottle.

3a.2.4 Preparations

3a.2.4.1 Amido Black Working Solution (Methanol Base)

- Dissolve 2.0 grams of amido black 10B in 100 milliliters of acetic acid.
- Add 900 milliliters of methanol and thoroughly mix.
 - **Rinse #1**
Mix 100 milliliters of glacial acetic acid with 900 milliliters of methanol.
 - **Rinse #2**
Mix 50 milliliters of glacial acetic acid with 950 milliliters of distilled water.

3a.2.4.1.1

This methanol based Amido Black solution is available commercially and may be used as a purchased pre-mixed solution.

3a.2.4.2 Amido Black Working Solution (Water Base)

- Mix the following:

	LPU-PM 3a.2 Amido Black	
	<i>Document #: 2591</i>	<i>Page 2 of 4</i>
	<i>Revision #: 2</i>	<i>Issued Date: 08/20/2020</i>
	<i>Document Manager: Tracee McIntosh</i>	<i>Approved By: Ryan Larrison</i>

- 500 ml distilled water
- 20 g 5-Sulfosalicylic Acid
- 3 g Naphthol Blue-Black
- 3 g Sodium Carbonate
- 50 ml Formic Acid
- 50 ml Acetic Acid
- 12.5 ml Kodak Photo Flo 600 Solution
- Use Rinse #2 (above)

3a.2.4.2.1

This aqueous Amido Black solution is available commercially and may be used as a purchased, pre-mixed solution.

3a.2.5 Instrumentation

Photographic collection of the suitable developed latent print is required.

3a.2.6 Controls

3a.2.6.1

A swab of control blood shall be tested with Amido Black solution prior to use.

3a.2.6.2

Testing of a small area of the evidence or similar substrate that is not critical for latent print development may be necessary to ensure that the substrate will not be adversely affected by the methanol formula.

3a.2.7 Procedure

3a.2.7.1

Possible blood proteins should be fixed prior to amido black application. This can be accomplished by:

1. Baking the item at 100°C for 30 minutes. Heat-sensitive items may be baked at a lower temperature for a longer time or another fixing technique attempted.
2. Chemically fixing the possible blood with methanol. The Methanol fixing technique may only be applied to dried suspected blood stains.

3a.2.7.2

The working solution should be agitated before evidence application as well as during the immersion process.

3a.2.7.3

Amido black 10 B working solution is applied to the item by:

1. Immersion
2. Application with a wash bottle

	LPU-PM 3a.2 Amido Black	
	<i>Document #: 2591</i>	<i>Page 3 of 4</i>
	<i>Revision #: 2</i>	<i>Issued Date: 08/20/2020</i>
	<i>Document Manager: Tracee McIntosh</i>	<i>Approved By: Ryan Larrison</i>

3a.2.7.3

The item is covered with the working solution followed by the rinse solution(s) until optimum contrast has been observed.

3a.2.7.4

Dried impressions that lose contrast may be re-immersed in the second rinse solution for photographic collection.

3a.2.8 Interpretation of results

The reaction results in a dark blue coloration. The contrast of the suspected blood impression should be intensified and additional detail not previously visible may be revealed.

3a.2.9 Minimum Quality Standards

3a.2.9.1

Shelf life, expiration date, of the purchased reagent shall be used as the destruction date.

3a.2.9.2

Shelf life of the mixed reagent is approximately two years. Control testing shall dictate continued use throughout the two years.

3a.2.10 Other Related Procedures

Ninhydrin
LCV

3a.2.11 Limitations

3a.2.11.1

The staining is stable, however latent prints should be photographed at first opportunity.

3a.2.11.2

Amido Black reacts with proteins and is not intended as a presumptive or confirmatory test for the presence of blood.

3a.2.11.3

The background staining of Amido Black can be significant and limits the use of this reagent on nonporous items.

	LPU-PM 3a.2 Amido Black	
	<i>Document #: 2591</i>	<i>Page 4 of 4</i>
	<i>Revision #: 2</i>	<i>Issued Date: 08/20/2020</i>
	<i>Document Manager: Tracee McIntosh</i>	<i>Approved By: Ryan Larrison</i>

3a.2.11.4

Cyanoacrylate fuming may be detrimental to amido black, however if the item is not over-fumed then the amido black reaction tends to be as expected.

3a.2.11.5

Painted surfaces may be deteriorated by the methanol in the working and rinse solutions.

3a.2.11.6

Excessively blood-stained items and porous surfaces that strongly absorb the dye will yield little contrast to the developed detail.

3a.2.11.7

This process is detrimental to some biological examinations. The chemistry in the MSP Biology unit for DNA analysis appears to not be negatively impacted by prior application of amido black.

3a.2.11.8

Evidence can over-develop in the working solution causing deterioration of the quality of ridge structure.

3a.2.11.9

Blood shall be "dried" on the surface prior to applying this reagent.

3a.2.12 References

British Home Office. "Chemical Development and Intensification of Sweat and Blood Marks, Etc.," May 1981.

Lee, Henry C.; Gaensslen, R. E., eds. *Advances in Fingerprint Technology*; Elsevier Science Publishers: NY, 1991.

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

McCarthy, Mary M.; David L. Grieve. "Preprocessing with Cyanoacrylate Ester Fuming for Fingerprint Impressions in Blood"; *Journal of Forensic Identification*, 1989, 39, 1, 23-32.

<https://www.cbdiai.org/amido-black-methanol-base.html>