

Firearms/Toolmarks Unit

Standard Operating Procedure for

The Modified Griess Test for Nitrite Residues

1 Scope

This procedure is designed to provide a specific technique for detecting and preserving patterns of nitrite residues around a suspected bullet hole as a basis for estimating muzzle-to-target distances. Such patterns may be on evidence such as clothing items, furniture, bedding, and wallboard. This procedure applies to Firearms/Toolmarks Discipline (FTD) personnel conducting forensic examinations in the following category of testing:

- Firearms

2 Equipment/Materials/Reagents

- Scissors
- Zip-lock bags
- Cheesecloth
- Graduated cylinder (500 ml)
- Blotters/brown wrapping paper
- Processing tray
- Photographic paper or similar media
- Filter paper
- Beaker (1000 ml)
- Sodium nitrite (reagent grade or better)
- Sulfanilic acid (reagent grade or better)
- Glacial Acetic acid (reagent grade or better)
- Personal protective equipment
- Laboratory coat
- Tweezers
- Flat Iron
- Micro-spatula
- Exhaust hood
- Polyethylene bottles
- Distilled and/or deionized water
- Cotton swabs
- Alpha-naphthol (reagent grade or better)
- Methanol (reagent grade or better)

3 Standards and Controls

3.1 Standards are not applicable.

3.2 Positive and negative controls are prepared for the Modified Griess test to detect for the contamination of nitrites.

3.2.1 Preparation of Nitrite Test Swabs

3.2.1.1 Prepare a solution of 0.6 grams of sodium nitrite in 100 milliliters of distilled water.

3.2.1.2 Divide a package of cotton swabs into equal amounts. Soak half of the cotton-tipped ends in the nitrite solution. Dispose of any remaining solution.

3.2.1.3 Set the swabs aside to dry, and store in a labeled zip-lock bag. These will serve as the positive control. There is no known limit to the shelf life of these swabs.

3.2.1.4 Retain the other half of the non-treated swabs for the negative control, and store in a labeled zip-lock bag.

3.3 If a chemical reagent must be prepared before an examination, the following information must be recorded on the appropriate Firearms/Toolmarks Unit (FTU) Chemistry Log which can be found in the Firearms/Toolmarks Unit, *Comprehensive Gunshot Residue Examinations in Muzzle-to-Target Distance Determination* procedure: FTU lot number, preparer, date, parent chemical (lot number), FBI Laboratory serial number and the result of the performance check.

3.3.1 The FTU lot number for reagents used during examinations will be recorded in the examination notes.

4 Sampling or Sample Selection

Not applicable.

5 Procedures

5.1 Preparation of Reagents and Test Media

The following instructions apply to the preparation of the reagents and test media for use in the Modified Griess Test for nitrite residues.

5.1.1 Processing of Photographic Paper

5.1.1.1 Prepare a solution of 0.5 grams of sulfanilic acid in 100 milliliters of distilled water.

5.1.1.2 Prepare a solution of 0.28 grams of alpha-naphthol in 100 milliliters of methanol.

5.1.1.3 Combine the above solutions.

5.1.1.4 Pour the combined solution into a non-reactive photo processing tray and briefly dip pre-cut sheets of the photographic paper into the tray. Submerge the sheets completely and remove them. Note: as a substitute for photographic paper, ordinary laboratory filter paper or similar media may be processed in the same manner. (See the suggested method in the specific

test procedure which follows.)

5.1.1.5 Set the sheets aside to dry on a clean surface (a table covered with brown wrapping paper). Dispose of any remaining solution.

5.1.2 Preparation of a 15% Acetic Acid Solution

5.1.2.1 Combine 150 milliliters of glacial acetic acid with 850 milliliters of distilled water. Gently pour the acid into the water to preclude the potential spattering of undiluted acid.

5.1.2.2 Store the solution in a properly sealed and labeled container. There is no known limit to the shelf life of this reagent.

5.2 Procedure for a Standard Modified Griess Test

5.2.1 To ensure the chemically treated photographic paper or similar paper media is functioning properly, test the four corners of the emulsion-coated side with a positive control. This is accomplished by saturating a nitrite test swab (positive control) in a small amount of 15% acetic acid solution and dabbing the four corners of the paper. An orange color should appear at each corner, confirming sensitivity. The results will be recorded in the examination records.

5.2.2 For a negative control, repeat step 5.3.1 above, but use clean, non-nitrite treated acetic acid-saturated test swabs. Ensure that this test follows the positive control test, and that there is sufficient physical separation between test marks to preclude bleeding from one mark to the other. The results will be recorded in the examination records.

5.2.3 Place the evidence or known-distance test (see the Firearms and Toolmarks Unit, *Shot Pattern Examinations in Muzzle-to-Target Distance Determinations* procedure for conducting known-distance tests) questioned side down on the emulsion-coated side of the treated photographic paper. Indicate on the paper or similar media, using a pencil, such objects as seams, buttons, button holes, rips, pockets, suspected bullet holes, tears, cuts, etc., for possible future reference by marking with a pencil.

5.2.3.1 Do not use ink at this point because it may transfer back onto the tested item.

5.2.4 Soak a piece of cheesecloth in the 15% acetic acid solution (in a large beaker) and wring it out. Place the cheesecloth on the questioned item or known-distance test as the third layer of the "sandwich". Press the "sandwich" with a hot iron. On many irons, the setting for "cotton" is appropriate.

5.2.5 Discard the cheesecloth and separate the questioned item or known-distance test-firing from the photographic paper.

5.2.6 When dry, the photographic paper will be marked appropriately (Note: photographic

paper is considered secondary evidence and will be marked with the item identifier associated with the primary evidence item, preceded by an “*f*” indicating *from*, for example: *f* Item 5.) Photographic paper will be properly marked in ink for future identification and returned to the contributor as secondary evidence.

5.3 An Alternative Procedure for a Modified Griess Test Using Treated Filter Paper

5.3.1 Treat the filter paper in the same solutions used for treating the photographic paper. Allow it to dry. See section 5.1.1.

5.3.2 Test for nitrite sensitivity using the positive and negative controls.

5.3.3 Place the filter paper on the questioned surface.

5.3.4 Process the filter paper using one of the following methods:

- a. Saturate a piece of cheesecloth in the 15% acetic acid solution and wring it out. Place the cheesecloth over the filter paper and apply a hot iron.
- b. Spray the filter paper with the 15% acetic solution until very damp. Cover with two or three additional layers of filter paper and iron until dry.

5.3.5 Separate the test media and check the results.

5.3.6 When dry, mark and preserve results for retention as in 5.2.6 above.

5.4 Procedure for a Reverse Modified Griess Test.

This procedure is reserved for items that are thick or otherwise non-porous materials. Typically, these materials don't allow for the Acetic Acid solution to penetrate through the item.

5.4.1 Similar to Section 5.2.1, test for nitrite sensitivity using the positive and negative controls, confirming sensitivity. The results will be recorded in the examination records.

5.4.2 Place the photographic paper or similar media emulsion/treated side down on the questioned surface. Use a pencil to indicate on the paper or similar media such objects as seams, buttons, suspected bullet holes, pockets, rips, tears, and cuts for future reference.

5.4.3 Wipe the emulsion/treated side of the photographic paper with a piece of cheesecloth saturated with a 15% acetic acid solution. Lightly apply the solution to the entire surface. (Too much solution will cause indistinct or hazy results due to pigment migration.)

5.4.4 Immediately place the photographic paper or similar media emulsion/treated side down on the questioned surface.

5.4.4.1 Prior to applying a hot iron, attach a piece of material (e.g., filter paper, cheesecloth,

clean twill jean, clean cotton material) to the back of a piece of treated photographic paper or similar media. Failure to attach a piece of material will likely result in the paper or similar media sticking to the iron.

5.4.4.2 Apply a hot iron to the back of the photographic paper or similar media.

5.4.5 Separate the photographic paper and the questioned area. Any orange indications on the photographic paper are the result of a chromophoric reaction specific for the presence of nitrite residues.

5.4.6 When dry, mark and preserve results for retention as in 5.3.6 above.

6 Calculations

Not applicable.

7 Measurement Uncertainty

Not applicable.

8 Limitations

8.1 Based on the chemistry outlined in this procedure, the Modified Griess Test is not suitable for the detection of purely nitrate compounds, such as unburned smokeless powder. It should be noted that unburned powder particles (nitrates) are commonly coated with burned powder residues (nitrites) and positive reactions take place.

8.2 The Modified Griess Test yields results for nitrite residues regardless of whether these are in fact gunshot residues.

9 Safety

Ensure that proper ventilation is provided during both the preparation of the reagents and the procedure itself. When handling clothing that is potentially contaminated with biological hazards or preparing reagents for chemical tests, protective latex or vinyl gloves, safety glasses, and laboratory coat will be worn at all times.

For disposal of the chemicals/hazardous waste used for this procedure, refer to Section 5, titled Hazardous Waste Disposal, of the *FBI Laboratory Safety Manual*.

10 References

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Dillon, J.H., "A Protocol for Gunshot Residue Examinations in Muzzle-to-Target Distance Determinations," AFTE Journal, Vol. 22, No. 3, 1990, pp. 257-274.

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Gamboa, Frances A. and Kusumi, Raymond, "Evaluation of Photographic Paper Alternatives for the Modified Griess Test," AFTE Journal, Vol. 38, No. 4, 2006, pp. 339-347.

Malikowski, Shawn G., "Alternative Modified Griess Test Paper," AFTE Journal, Vol. 35, No. 2, 2003, pp. 243.

Watson, D.J., "Nitrites Examination in Propellant Powder," AFTE Journal, Vol. 11, No. 1, 1979, p. 32.

FBI Laboratory Quality Assurance Manual

FBI Laboratory Operations Manual

FBI Laboratory Safety Manual

FBI Laboratory, FTU Quality Assurance Manual

Rev. #	Issue Date	History
3	08/21/12	Section 3 added cite where Chemistry Logs can be found.
4	04/17/19	Header and document title were updated to conform to the document control requirements. Updated Section 1 to specify category of testing. Updated the formatting of the listed equipment, materials and reagents in Section 2. Included the recording of the FTU lot number as part of Section 3 and included nitrite controls. Removed Section 4 Calibration and renumbered. Renumbered the beginning of Section 5 and completed grammatical updates to Sections 5.2.1 through 5.2.3.1. Removed the listing of a specific type of photographic paper to allow for other media to be used in Section 5.2, 5.2.1, 5.3.1 and 5.5.1. Updated Section 5.3.6 with current terminology for evidence generated in Forensic Advantage. Split 5.3.2 into two separate steps and renumbered remaining section. Updated title for Section 5.4 and renumbered and reordered the remaining requirement. Relocated disposal statement from Section 5.1 to Section 9. Clarified limitations statement in Section 8.1. Additional references were added to Section 10 to include articles relating to the use of various photographic papers that are used in the Modified Griess Test.

Approval

Redacted - Signatures on File

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