

**Connecticut Department of Public Safety
Division of Scientific Services
Forensic Laboratory**

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Page 1 of 2

Document Title: Test for Creatinine (Jaffe)

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Date: _____

A. PURPOSE:

To determine the presence of creatinine in a Forensic sample, which indicates the presence of urine.

B. RESPONSIBILITY:

Forensic Science Examiners from the Connecticut State Forensic Science Laboratory who have been trained in the discipline of testing for creatinine according to SOP-FB-31 (Training Manual).

C. SAFETY:

Use appropriate measures for the proper handling of picric acid solution, sodium hydroxide and glacial acetic acid according to SOP-GL-2 (Safety Manual).

D. PROCEDURE:

This test will be performed at the discretion of the examiner based on the submitting agency requests, case information and the condition of the evidence.

1. Materials:

- a. Picric Acid Solution (saturated)
- b. 10% NaOH
- c. Distilled water (dH₂O)
- c. Glacial Acetic Acid
- d. Controls: known urine stain and blank filter paper (include substrate control when needed)
- e. Disposable pipet or micropipet and tips
- f. Test tubes

2. Procedure:

- a. Test a positive and negative control with the following procedure (steps 2.b. – 2.g.).
 - aa. The controls may be run concurrently with the question samples.
 - bb. If limited question sample is available, run the controls prior to testing the question sample. If controls yield the appropriate results then test the questioned sample.
 - cc. If controls do not yield the appropriate results, review the procedure and retest the controls prior to the question samples.

- D. 2. b. Extract a portion of the questioned sample or stain in a test tube or spot plate with enough dH₂O to cover the sample. Let stand for a minimum of 15 minutes or longer as necessary.
- c. Remove substrate from test tube.
- d. Add equal amounts of picric acid solution and 10% NaOH to an equal amount of the extract.
- e. Allow to stand for 15 minutes and observe the color of the extract.
- f. Add glacial acetic acid in excess (double the total volume of extract, picric acid and 10% NaOH) and observe the color of the extract.
- g. It should be noted that the detection of a urine or ammonia-type odor may also be used in the interpretation of the results (sample may need to be heated at 37°).
3. Results:
- a. *Positive*: A positive result is indicated by a color change from yellow to orange/reddish-orange and develops fully within 15 minutes. A color change back to yellow after the addition of glacial acetic acid, confirms the presence of creatinine.
- b. *Negative*: A negative result is indicated by the bright yellow color of the picric acid.
- c. *Inconclusive*: A sample that remains unchanged after the addition of glacial acetic acid is considered inconclusive.
- d. It is important to compare results against the positive and negative controls.
- e. Record the results of the controls and samples on the appropriate Quality Record Worksheet.
- f. A 2nd examiner will observe and confirm results and initial the appropriate Quality Record Worksheet.

E. REFERENCES:

1. Jaffe, M. "Uebeden Niederschlag, Welchen Pikrinsäure in normalen Harn erzeugt und über eine neue Reaktion des Kreatinins." Z. Physical Chem, 10: 391-400. 1886.
2. Metropolitan Police Forensic Science Laboratory. Biology Methods Manual. 1978, pp. 4-4 to 4-5.
3. SOP-GL-2 (Safety Manual).