

*Approved by Director: Dr. Guy Vallaro***A. PURPOSE:**

To determine the presence of amylase in a Forensic sample, which indicates the presence of saliva.

**B. RESPONSIBILITY:**

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

**C. DEFINITION:**

1. RSID: Rapid Stain Identification
2. sdH<sub>2</sub>O: Sterile distilled water

**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. Phadebas<sup>®</sup> powder
  - b. sdH<sub>2</sub>O
  - c. RSID<sup>™</sup>-Universal Buffer
  - d. Controls: positive (known saliva stain) and negative (blank filter paper), include substrate control as needed
  - e. Previously extracted samples
  - f. Disposable pipets
  - g. Test tubes
  - h. Filter paper
2. Procedure:
  - a. Test a positive and negative control with the following procedure (steps 2.b. – 2.i.).
    - aa. The controls may be run concurrently with the questioned samples and are made with the same extraction solution used for the questioned samples.
    - bb. If limited questioned sample is available, run the controls prior to testing the questioned sample. If controls yield the appropriate results then test the questioned sample.

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- D. 2. a. cc. If controls do not yield the appropriate results, review the procedure and retest the controls prior to the questioned samples.
- b. For a liquid sample, prepare a stain on filter paper and air dry.
- c. Place a portion of the questioned sample or stain in a labeled test tube.
- d. To each tube add approximately 0.025g of Phadebas<sup>®</sup> powder.
- e. Add approximately 4 drops (250µl) of sdH<sub>2</sub>O to each tube.
- f. For samples previously extracted in RSID<sup>™</sup>-Universal Buffer (as needed):
- aa. To each tube add approximately 0.025g of Phadebas<sup>®</sup> powder.
- bb. Add approximately 4 drops (250µl) of extract to each tube.
- g. Gently shake each test tube to mix contents.
- h. Incubate at 37°C for 15-20 minutes.
- i. Shake each tube again and centrifuge for one (1) minute.
- j. Observe the color of the supernatant of the samples.
3. Results
- a. *Positive.* In positive samples the supernatant will be blue, indicating the presence of amylase activity.
- b. *Negative.* In negative samples the supernatant will be clear, indicating the absence of amylase activity.
- c. *Inconclusive.* No distinguishable blue color of supernatant.
- 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.

Note: The reason a result is determined to be inconclusive must also be recorded.

**FB SOP-15 Test for Amylase (Phadebas)**

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- f. A 2<sup>nd</sup> examiner will observe and confirm results and initial the appropriate Quality Record Worksheet.
4. Record extraction solutions on the appropriate Quality Record Worksheet location and General Reagent Sheet (FBQR-09).

**E. REFERENCES:**

1. Willott, G.M. 1974. "An improved test for the detection of salivary amylase in stains". J. Forensic Sci. Soc., 14: 341-344.
2. Metropolitan Police Forensic Science Laboratory. Biology Methods Manual. 1978, pp. 3-10 to 3-11.