A. PURPOSE:

To prepare reagents for blood screening tests and to perform quality control on prepared reagents.

B. RESPONSIBILITY:

Forensic Science Examiners 1 and 2 in the Forensic Biology Section. Ordering information is maintained in a log book in the Forensic Biology Section.

C. SAFETY:

Use appropriate measures for the proper handling of o-Tolidine, ethanol, glacial acetic acid and potassium hydroxide according to SOP-GL-2 (Safety Manual) and the Material Safety Data Sheets.

D. DEFINITIONS:

1. dH_2O: Distilled water
2. KM: Kastle-Meyer Test

E. PROCEDURE:

o-Tolidine Solution

1. Materials:
   a. o-Tolidine 1.6g
   b. Ethanol 40.0ml
   c. Glacial acetic acid 30.0ml
   d. dH_2O 30.0ml

2. Procedure:
   a. Mix all materials together and place into a brown dropper bottle.

   b. Test each new batch of reagent before use according to SOP-FB-07 (Screening Tests for Blood) and the o-Tolidine Reagent Log Sheet. Record the required information.

   c. If the appropriate results are not obtained, discard the reagent, review the procedure and make new reagent.

   d. If the reagent is suitable for use, record the solution, lot # (date of preparation), control date and examiner's initials on the dropper bottle and store in the refrigerator.

E. 2. e. Discard after one (1) year or sooner if the reagent changes from transparent beige to a dark brown color.

Phenolphthalin Stock Solution

1. Materials:
   a. Phenolphthalin 2g
   b. Potassium hydroxide 20g
   c. dH_2O 100ml
   d. Granular Zinc 20g

2. Procedure:
   a. Dissolve potassium hydroxide in dH_2O. Note: The solution will be warm from the reaction.

   b. Add granular zinc followed by phenolphthalin. Solution turns pink immediately.

   c. Swirl solution until it becomes colorless.

   d. Place stock solution in a brown bottle containing zinc.

   e. Discard after six (6) months or sooner if the reagent turns from colorless to pink.

Phenolphthalin Working Solution

1. Materials:
   a. Phenolphthalin stock solution 1 part
   b. Ethanol 4 parts
   c. Granular Zinc ~15g

2. Procedure:
   a. Dilute stock solution 1:5 in ethanol (1 part to 4 parts).

   b. Place working solution in brown dropper bottles containing zinc.

   c. Test each new batch of the working solution before use according to SOP-FB-07 (Screening Tests for Blood) and the KM Reagent Log Sheet. Record the required information.

   d. If the appropriate results are not obtained, discard the reagent, review the procedure and make new reagent.

e. If the reagent is suitable for use, record the solution, lot # (date of preparation), control date and examiner's initials on the stock and dropper bottles and store in the refrigerator.

f. Discard after six (6) months or sooner if the reagent turns from colorless to pink.

E. 3 % Hydrogen Peroxide

1. This chemical is purchased from an outside vendor and tested/used as received.

2. Test the new manufacturer's lot before use according to SOP-FB-07 (Screening Tests for Blood) and the KM and o-Tolidine Reagent Log Sheets. Record the required information.

3. If the appropriate results are not obtained, review the procedure, repeat the test and replace the chemical if necessary.

4. If the lot is suitable for use, record the date received, date opened and examiner's initials on the stock bottles and store in the refrigerator.

5. Record the chemical, lot #, manufacturer's expiration date, fill date and initials on the dropper bottles.

6. Discard according to the manufacturer's expiration date or sooner if a decrease in reaction activity is noted.

F. REFERENCES:

1. Metropolitan Police Forensic Science Laboratory. Biology Methods Manual. 1978, pp. 2-88 to 2-90.

2. SOP-GL-2 (Safety Manual).

3. Material Safety Data Sheets.