Saliva and Its Importance in Forensic Science

Introduction: -

Saliva is a watery and frothy substance produce in the mouth of humans as well as animals by the salivary glands (**parotid, sub lingual, sub mandibular**). The measure gland for secretion is parotid (below ear), sub lingual (below tongue), Sub mandibular (below the floor of mouth).

Characteristics: -

Saliva is tasteless, odorless, clear, viscid, alkali. The amount secreted in 24 hours is estimated to be 1500 ml.

Composition: -

99.5 % water, Salts, Enzymes (amylase, lipase), Proteins (albumin, globulin), Antibacterial compounds (peroxide, lactoferin, thiocynates, secretory Ig A)

Functions: -

Digestive function, Disinfectant function, Non physiological function

Cases: -

Hanging, Poisoning, Rape, Kidnapping, Struggling, Murder, Bite mark

Evidence: -

Hanger cheap, Ligature, Gag, Cigarette buds, Cup, Glass, Bit marks, Rim of the bottle, Tooth pick, Bad sheet, Pillow, Food material

Collection of samples: -

To collect the sample of saliva for analysis the mouth should be thoroughly rinsed with water before collecting the sample. To stimulate flow of saliva the subject should chew a small swelled of paraffin wax.

Examination of saliva stains: -

1) Starch Iodine Test: -

Reagent preparation: -

0.5% soluble starch solution (50 mg of soluble starch in 10 ml water)

Lugol's Iodin solution (1 gram of iodine + 2 gram of potassium iodide + 200 ml of water)

Sample preparation: -

1 test tube: -

Place 5 mm into 5 mm piece of sample to be tested. (Presence or absence of amylase)

2 test tube: -

Place 5 mm into 5 mm piece of sample known saliva stain. (Presence of amylase)

3 test tube: -

Place 5 mm into 5 mm piece of unstained control any other. (Absence of amylase usually water)

Procedure: -

Place 3 test tubes in a rack. Add 3-4 drops of soluble starch solution to each test tube. Mix, cork and incubate the test tube for 1 hour at 37 degree Celsius. Add 2 drops of lugol's iodine solution and note the color formation.

Observation: -

Formation of blue color therefor absence of saliva.

Absence of blue color indicates the starch has been hydrolyzed by amylase and no longer available for complex with iodine. Therefor **lack of blue color** is a **positive result** for amylase activity and indicate **presence of saliva**.

2) **Radial diffusion test for amylase: -**

Reagent preparation: -

0.1 ml phosphate buffer pH 6.9 (for phosphate buffer)

Gel test plate (2% Agarose, 0.1% soluble starch)

Iodine development solution (KI 1.65, Iodine 2.54, distilled water 30 ml)

Sample preparation: -

Extract 3 mm square piece of stained material with 50 micro liter distilled water. Prepare and extract of unstained material in the same manner.

Procedure: -

Punch holes in the plate with a vacuum pipette or cork borer leaving 1.5 cm distance between the sample wells. Place sample to be tested in the sample wells using precision pipette each well holds 4 micro liter liquid. Cover the petri dish and place in incubator at 30 degree Celsius for 6 hours or overnight. Stain the plate by pouring a 1:50 dilution of saturated iodine solution onto the surface. Rinse with water.

Observation: -

Clear circles around the wells indicate areas of amylase activity. The diameter of the clear circle is proportional to the square root of the concentration of amylase. Radial diffusion test using for **concentration of amylase**.