

Sir H N Reliance Foundation Hospital And Research Centre Mumbai  
Department of Biochemistry  
Mentor-Dr Nitin Chavan  
Report by Samika Jain Date: 2nd August 2024  
Purpose of Report: Lab Observership

## **OBSERVATION OF LAB TESTS :**

### **I. Aim: Quantitative estimation of Albumin**

**Location:** Found in human serum and plasma  
pH: 4.1

AT 4.1, Albumin has cationic properties. It can bind with BCG(bromocresolgreen), an anionic dye, to form a blue-green complex. The color intensity of the complex is proportional to the albumin concentration. Albumin is stable at pH between 4 and 9 at normal temperatures.

#### **Sample:**

- Albumin should be free of bubbles
- It should be free of RBC's
- It should be free of fibrin.

#### **Result:**

Hyperalbuminemia is a condition where there is an increased concentration of albumin in the blood. The common cause of Hyperalbuminemia is dehydration.

#### **Causes:**

Hyperalbuminemia is caused by liver disease, catabolism due to tissue damage, malabsorption of amino acids( Crohn's disease), protein loss via stools(neoplastic disease)

### **II Aim: Quantitative estimation of the amount of Calcium**

**Location** Human serum, plasma, and urine

**Principle:** The NM-BAPTA method is a quantitative method for measuring calcium in human serum. Calcium ions react with NM-BAPTA under alkaline conditions to form a complex. This complex reacts with EDTA to form a colored product whose intensity is directly proportional to calcium ions concentration in the serum. It is measured photometrically at 340nm.

**Biology:** Calcium levels are controlled by parathyroid hormone(PTH), Calcitriol and Vitamin D.

**III Aim:** Quantitative estimation of ALP (Alkaline Phosphatase) in human serum and plasma

**Location:** Human Serum and Plasma

**Principle Cofactors:** When alkaline phosphatase is present in human serum along with zinc and magnesium ions, it catalyzes the hydrolysis of p-nitrophenyl phosphate into p-nitrophenol and phosphate. The amount of p-nitrophenol produced is directly proportional to Alkaline phosphatase levels.

**Result:**

- Higher levels of ALP can lead to Cholestasis, a condition where the flow of bile from the liver is blocked or reduced.
- Obstructive Jaundice
- Skeletal System disease, a chronic disease that affects bones and the breast
- Osteomalacia and rickets including bone tumors
- Hyperparathyroidism

**Causes:**

Increased Osteoblast activity

Accelerated bone growth

Vitamin D deficiency can cause increased levels of ALP.

**IV Aim:** Quantitative estimation of ALT (alanine aminotransferase)

**Location:** Human serum and plasma

**Principle:** ALT catalyzes the reaction between two substances L-alanine and  $\alpha$ -ketoglutarate to form L-glutamate and pyruvate. The subsequent reaction by NADH in the presence of lactate dehydrogenase (LDH) FORMS L-lactate and  $\text{NAD}^+$

L-alanine +  $\alpha$ -ketoglutarate  $\xrightarrow{\text{ALT}}$  pyruvate + L-glutamate

Pyruvate +  $\text{NADH} + \text{H}^+$   $\xrightarrow{\text{LDH}}$  L-lactate +  $\text{NAD}^+$

The rate of NADH oxidation is directly proportional to ALT activity.

A decrease in absorbance is directly proportional to ALT activity. This is due to oxidation of NADH to  $\text{NAD}^+$ , which is measured at a wavelength of 340 nanometers.

**Result:** High levels of ALT can be a sign of liver damage from several conditions including Hepatitis, Cirrhosis, Carcinoma of the liver, and obstructive jaundice.

**Note:** They are elevated in patients with myocardial infarctions.

**V Aim:** Quantitative estimation of total cholesterol

**Location:** Human Serum and Plasma

**Principle:** Cholesterol esters are enzymatically hydrolyzed by the enzyme cholesterol esterase to cholesterol and free fatty acids. Free cholesterol, including that originally present, is then oxidized by cholesterol oxidase to cholest-4-en-3-one and hydrogen peroxide. The hydrogen peroxide combines with 4-amino antipyrine to form a chromophore (quinone imine dye) which may be quantitated at 505 nm.

**Biology:**

Quantitative estimation of total cholesterol is performed to screen for atherosclerotic risk and lipoprotein metabolic disorders.

**VI Aim:** Quantitative estimation of glucose in human serum, plasma urine, and cerebrospinal fluid

**Principle:**

Hexokinase(HK) catalyzes the phosphorylation of glucose to produce glucose-6-phosphate and adenosine diphosphate(ADP). The hexokinase-catalyzed reactions produce NADH, which is analyzed using spectrophotometry to determine glucose concentration.

**Lab Interpretation:**

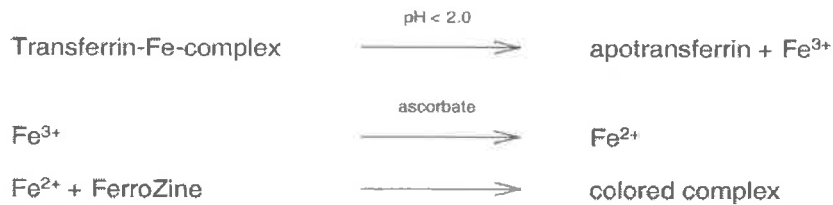
Glucose is the major carbohydrate present in the peripheral blood. Oxidation of glucose is the major source of cellular energy in the body. Glucose derived from dietary sources is converted to glycogen for storage in the liver or to fatty acids for storage in adipose tissue. The concentration of glucose in the blood is controlled within narrow limits by many hormones, the most important of which are produced by the pancreas.

The most frequent cause of hyperglycemia is diabetes mellitus resulting from a deficiency in insulin secretion or action. Several secondary factors also contribute to elevated blood glucose levels. These include pancreatitis, thyroid dysfunction, renal failure, and liver disease. Hypoglycemia is less frequently observed. A variety of conditions may cause low blood glucose levels such as insulinoma, hypopituitarism, or insulin-induced hypoglycemia. Glucose measurement in urine is used as a diabetes screening procedure and to aid in the evaluation of glycosuria, detection of renal tubular defects, and the management of diabetes mellitus. Glucose measurement in cerebrospinal fluid is used for the evaluation of meningitis, neoplastic involvement of meninges, and other neurological disorders.

## VII Aim: Quantitative estimation of iron

**Location:** Human serum and blood plasma

### **Principles:**



Under acidic conditions, iron is liberated from transferrin. Lipemic samples are clarified by the detergent. Ascorbate reduces the released  $\text{Fe}^{3+}$  ions to  $\text{Fe}^{2+}$  ions which then react with FerroZine to form a colored complex. This color intensity is directly proportional to iron concentration and can be measured photometrically.

### **Lab Interpretation:**

Iron (non-heme) measurements are used in the diagnosis and treatment of diseases such as **iron deficiency anemia, hemochromatosis** (a disease associated with widespread deposit in the tissue of the two iron-containing pigments, hemosiderin and hemofuscin, and characterized by pigmentation of the skin), **and chronic renal disease**. Iron determinations are performed for the diagnosis and monitoring of **microcytic anemia** (e.g. due to iron metabolism disorders and hemoglobinopathy), **macrocytic anemia** (e.g. due to vitamin B12 deficiency, folic acid deficiency and drug-induced metabolic disorders of unknown origin) as well as normocytic anemias such as renal anemia (erythropoietin deficiency), **hemolytic anemia, hemoglobinopathy, bone marrow disease, and toxic bone marrow damage**.

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## **IMMUNOLOGY**

Immunology is the study of the human immune system and how it protects the body from infection and diseases.

### **Type of Infectants**

#### **Thyroid peroxidase antibodies(TPOAb)**

The thyroid gland produces Thyroid Peroxidase enzyme(TPO)which helps the body produce hormones that control growth and metabolism.

When Thyroid Peroxidase antibodies (TPOAb) are detected in the blood it indicates thyroid dysfunction due to an immune system condition called Hashimoto's disease. It suggests that the thyroid gland is overstimulated.

#### **Anti-Thyroglobulin antibodies:**

Thyroglobulin(Tg) is a protein that is produced by the thyroid gland and is used to diagnose and monitor thyroid conditions:

Anti-thyroglobulin antibody test measures the amount of antibodies in our blood that are directed against thyroglobulin, a protein found in thyroid cells. Diseases like **thyroiditis** and **nodular goiter** can be detected.

#### **Alpha-fetoprotein(AFP)**

Alpha-fetoprotein is a protein that is produced by the liver and and yolk sac of a developing baby during pregnancy. The levels are elevated during pregnancy. It is a tumor marker for various cancers such as liver cancer, germ cell tumors, and brain cancer.

#### **Vitamin B-12**

This is essential for Hematopoiesis, or the process of blood cell production.It carries out nervous functions smoothly.

Deficiency of Vitamin B-12 is due to lack of intrinsic Factor(IF)secretion due to pernicious anemia, intestinal malabsorption, and gastric mucosa. Symptoms include sore tongue, mouth ulcers, shortness of breath, muscle weakness, vision problems, and weight loss along with anemia.

## **hCG hormone**

Human chorionic gonadotropin(hCG) is a glycoprotein that is made of two subunits, an alpha chain and a beta chain linked together by disulfide bonds. This hormone is secreted by the placenta during pregnancy to maintain corpus luteum by the production of progesterone. Besides pregnancy, the hCG hormone is secreted by abnormal germ cells, tissues, and neoplasms.

## **Anti-cyclic citrullinated peptide antibodies**

An anti-cyclic citrullinated peptide (anti-CCP) antibody test is a blood test that looks for antibodies that attack healthy cells in the body. It's used to diagnose rheumatoid arthritis(RA).It also helps gauge connective tissue diseases that may be present with arthritis.

## **Carbohydrate antigen 19-9**

Carbohydrate antigen 19-9(CA 19-9) is a protein that's shed by some cancer cells and can be found in the blood. It's a tumor marker that's used to help diagnose and monitor certain types of cancer like pancreatic stomach and colon cancer.

## **Carbohydrate antigen 15-3(CA 15-3)**

It's a protein found in the blood that can be used as a tumor marker for many types of cancer, including breast cancer, lung, colon, endometrium, liver, and cervix. Pregnancy and lactation can also raise CA levels(15-3). Normal levels of CA 15-3 vary but are generally below 30 U/ml.

## **Estrogen Test**

Estrogens are a group of hormones that play a key role in female reproductive health, including puberty, menstruation, pregnancy, and menopause. Estrogens are also important for heart, bone, and brain health in males and females. But males need estrogen in much smaller amounts. There are many types of estrogen, but only three types are commonly tested:

- **Estrone**, also called E1, is the only estrogen that females continue to make after menopause. Menopause is the time after menstrual periods have stopped for a year. Males and females make estrone in the adrenal glands (glands that sit on top of each kidney), and in body fat. In females, the ovaries (the glands that contain eggs) also make estrone.
- **Estradiol**, also called E2, is the main estrogen in nonpregnant females of childbearing age. It's mostly made in the ovaries and is important for fertility (the ability to get pregnant). It also helps support brain and bone health. Males make small amounts of this estrogen in the testicles (the glands that make sperm).

- **Estriol**, also called E3, is an estrogen that increases during pregnancy. The placenta (the organ that grows in the uterus to provide nutrients and oxygen to the unborn baby) makes estriol. Measuring estriol levels can help monitor the health of a pregnancy and the unborn baby. Males and nonpregnant females have very low levels of this estrogen.

### **Progesterone**

Progesterone is a hormone that's made mainly by the ovaries, which are two glands in the female reproductive system that contain eggs. Progesterone makes the lining of your uterus grow thicker so that a fertilized egg can attach (implant) inside of the uterus and grow into a baby. It is useful to evaluate placental function in pregnancy and ascertain ovulation. Placental insufficiency has been noted with low levels while high levels are associated with Testicular adrenal rest tumors.

### **Thyroid Stimulating Hormone(TSH)**

A glycoprotein hormone is made up of two subunits alpha and beta linked non-covalently. A TSH test measures the amount of thyroid-stimulating hormone (TSH) in your blood. TSH is produced by the pituitary gland. TSH is used for diagnostic purposes and to stimulate iodine uptake in some patients with thyroid cancer. Other thyroid tests that may be done at the same time include:

- T3 test (free or total)
- T4 test (free or total)
- Anti-TPO (antibodies against thyroid peroxidase)

### **Testosterone**

The primary androgenic hormone is testosterone and is responsible for the development of the male external genitalia and secondary sexual characteristics. In females, its main role is as a precursor for estrogen. Low testosterone also known as hypogonadism is a condition when a body's sex glands produce little or no testosterone. Causes include primary secondary or tertiary testicular failure.

**My experience from Observership at Sir H N Reliance Hospital :**

During my Observership under Dr Nitin Chavan of Sir H N Reliance Hospital, I got an opportunity to understand concepts, theories, and skills related to various types of testing that are conducted in a lab. My mentor guided me and solved all my queries adequately. After this first-hand practical experience, I feel better prepared for my future endeavors.

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