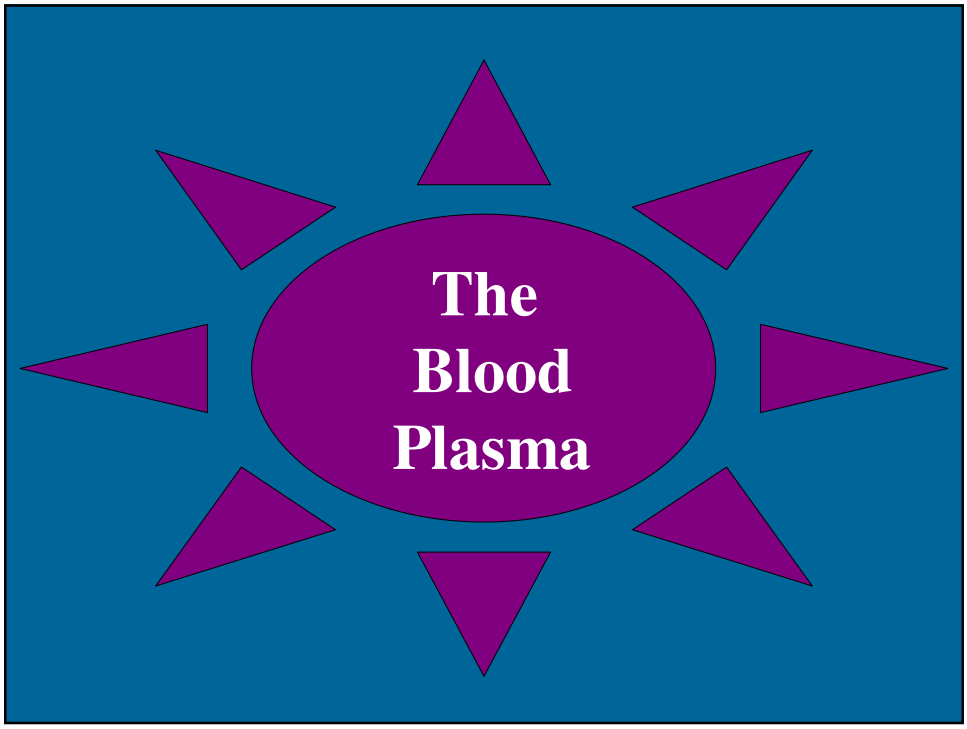


The Blood Plasma

-Contains 91-95% water.
- Solutes in plasma range from 5-9%
- Proteins are the major solute in the plasma and their level ranges from 6-8 gm %.

Principal Inorganic Constituents of Human Blood Plasma

Anions	Concentration meq/Liter	Cations	Concentration meq/Liter
Total	142-150	Total	142-158
Bicarbonate	24-30	Calcium	4.5-5.6
Chloride	100-110	Magnesium	1.6-2.2
Phosphate	1.6-2.7	Potassium	3.8-5.4
Sulfate	0.7-1.5	Sodium	132-150
Iodine,total	8-15*	Iron	50-180*
Protein bound	6-8*	Copper	80-160*

*These concentrations are in micrograms per 100 ml.

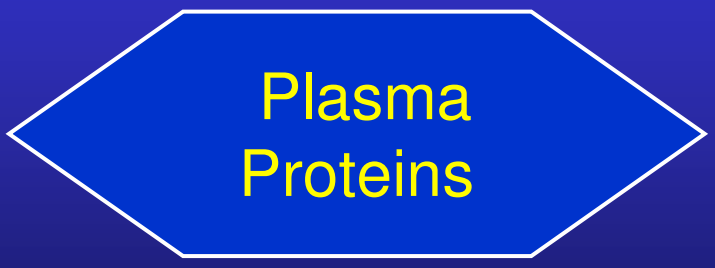
Principal Non-Protein Organic Constituents of Human Blood Plasma

Constituents	Normal Range
Non-Protein N :	25 - 40
Amino acid N	4 - 8
Amino acids	36 - 65
Bilirubin	0.2 - 1.4
Creatine	0.2 - 0.9
Creatinine	1 - 2
Uric acid	2 - 6
Carbohydrates:	
Glucose	65 - 90
Fructose	6 - 8

Contd...

Principal Non-Protein Organic Constituents of Human Blood Plasma

Constituents	Normal Range
Organic acids:	
Citric acid	1.4 - 3.0
α-ketoglutaric acid	0.2 - 1.0
Lactic acid	8 - 17
Lipids:	
Total lipids	285 - 675
Neutral fat	80 - 240
Cholesterol, total	130 - 260
Phosphoglyceride:	
Total	150 - 250



TOTAL PLASMA PROTEINS

- **The normal serum protein level is 63-83 g/dL.**
- **Plasma proteins include:**
  a. **Albumin**
  b. **Globulins**
     α– globulin: α1 & α2–globulins
     β– globulin: β1 & β2 globulins
     γ– globulins
  c. **Fibrinogen**
- **Under different pathological conditions the protein levels depart from the normal range.**

Functions of Plasma proteins

- **Transport**: e.g.
  - Transferrin transports iron.
  - Ceruloplasmin transports copper.
  - Albumin transports fatty acids, bilirubin calcium, many drugs etc.
  - Transcortin transports cortisol and corticosterone
  - Retinol binding protein transports retinol.
  - Lipoproteins transport lipids.
  - Haptoglobin transports free haemoglobin.
  - Thyroxin binding globulin transports thyroxin.

Functions of Plasma proteins

(contd)

- **Osmotic regulation**:
  - Plasma proteins are colloidal and non-diffusable and exert a colloidal osmotic pressure which helps to maintain a normal blood volume and a normal water content in the interstitial fluid and the tissues.
  - Albumin content is most important in regulation of colloidal osmotic or oncotic pressure.
  - Decrease in albumin level results in loss of water from blood and its entry into interstitial fluids causing oedema.
- Catalytic function (enzymes):
  - e.g lipases for removal of lipids from the blood.

Functions of Plasma proteins

(contd)

- **Protective function**:
  - Immunoglobulins combine with foreign antigens and remove them.
  - -Complement system removes cellular antigens.
  - Enzyme inhibitors remove enzymes by forming complexes with them. e.g. $\alpha 1$ antitrypsin combines with elastase, trypsin and protects the hydrolytic damage of tissues such as lungs.
  - Some proteins increase during acute phase and protect the body. E.g. $\alpha 1$ antitrypsin, $\alpha 2$ macroglobulins

Functions of Plasma proteins (contd)

- Blood clotting:

  - Many factors are involved in clotting mechanism and prevent loss of excessive amount of blood. e.g. clotting factors IX, VIII, thrombin, fibrinogen etc.

  - An excess of deficiency leads to a disease. e.g hemophilia, thrombus formation.

- Anticoagulant activity (thrombolysis):

  - Plasmin breaks down thrombin and dissolves the clot

- Buffering capacity:

  - Proteins in plasma help to maintain acid-base balance.

Main Functions/Concentration of proteins

PROTEIN	PLASMA CONC. (g/L)	FUNCTION
Pre-albumin	0.3	Binds T3 & T4
Albumin	40.0	Transport, colloid oncotic pressure
α1- globulin : α1- antitrypsin	3.0	Anti proteinase
α2- globulins		
ceruloplasmin	0.4	Copper transport
haptoglobulin	1.2	Binds haemoglobin
α2-macroglobulin	3.0	Transport, anti-proteinase

Contd.....

Main Functions/ Concentration of proteins

PROTEIN	PLASMA CONC. (g/L)	FUNCTION
β- Globulins		
Transferrin	2.5	- Iron - transport
Hemopexin	1.0	- Binds haem
Plasminogen	0.7	- Fibrinolysis
Fibrinogen	4.0	- Haemostasis
γ - Globulin		
IgA	0.9-4.5	-Ig in external secretions
IgM	0.7-2.8	- First Ab synthesised
IgG	8-18.0	-Main classes of antibody
IgE		- Involved in allergy
IgD		

MEASUREMENT OF PROTEIN FRACTIONS

- The protein fraction in plasma can be separated and estimated using the following methods:
  - Zone electrophoresis
  - Immunochemical methods
  - Chemical methods
  - Ultracentrifugation

CHARACTERIZATION, MEASUREMENT AND ISOLATION OF PLASMA PROTEINS

- Physical Techniques
  1. Ultracentrifugation (analytical or Sedimentation velocity ultracentrifuge) at 60,000 per.min. (Refractive index the boundary between the solvent and the protein is visualized by an optical system - called Sehlieren System).

  Advantage

  Most useful for the determination of the mol. wt of proteins

  Disadvantage

  High cost of each analysis and poor resolving capacity (when applied to whole serum or plasma)

CHARACTERIZATION, MEASUREMENT AND ISOLATION OF PLASMA PROTEINS

- 2. Biochemical Methods:

  i. Electrophoresis

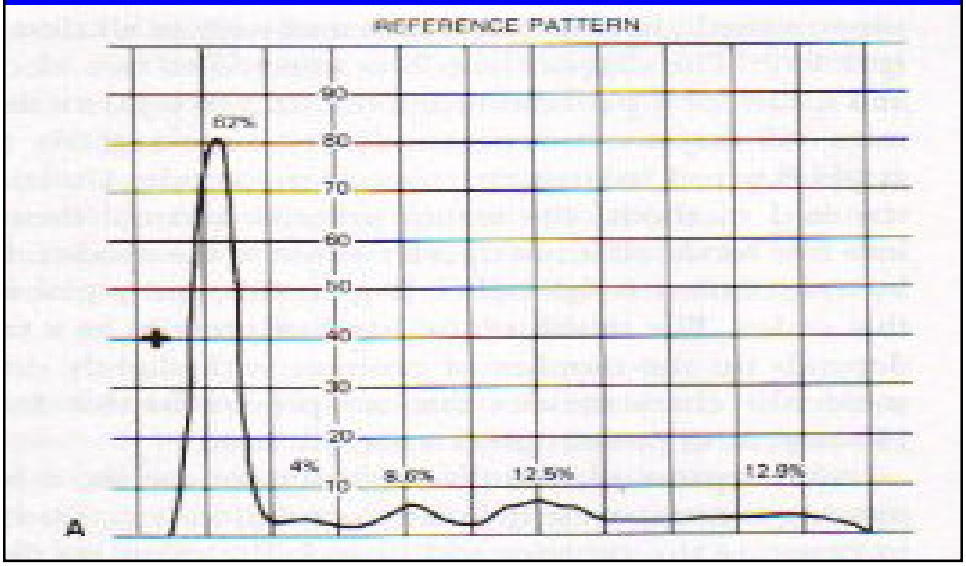
  Protein in aqueous solution are charged groups (e.g. carboxylic (Asp. Glu), amino groups (Lys, Arg), they can be separated under an electric field using various stabilizing media.

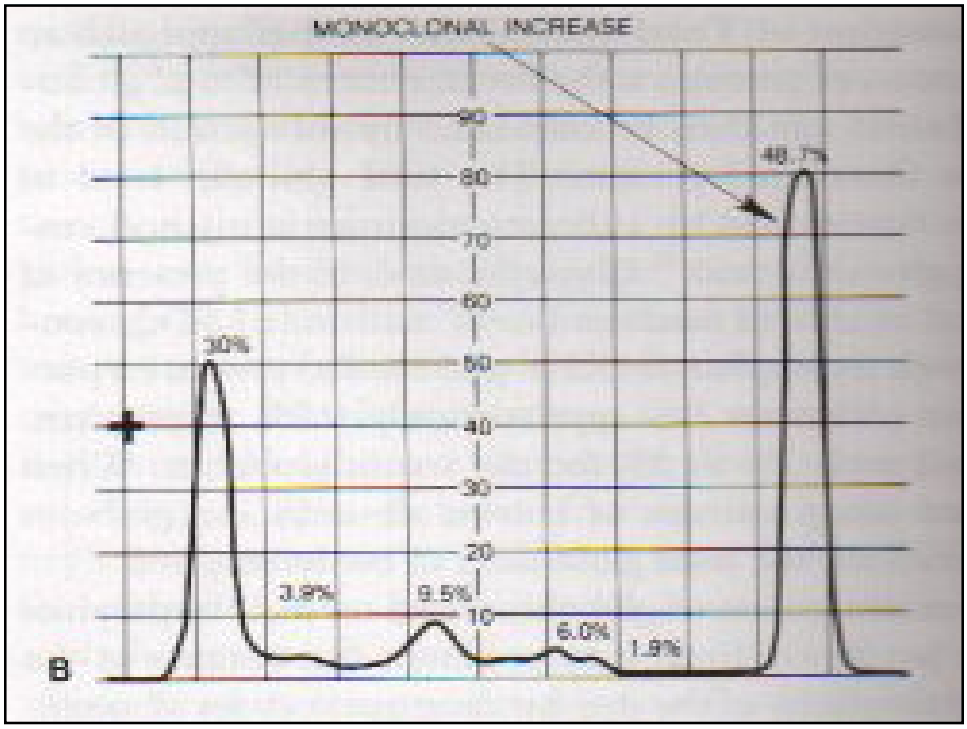
  N.B. Amino groups undergo ionic dissociation at alkaline pH and carboxylic undergo dissociation at acid pH. Most proteins are -ve at pH 8.6. The pH at which +ve charges equal to -ve charges is characteristic for a protein and is called isoelectric point PI).

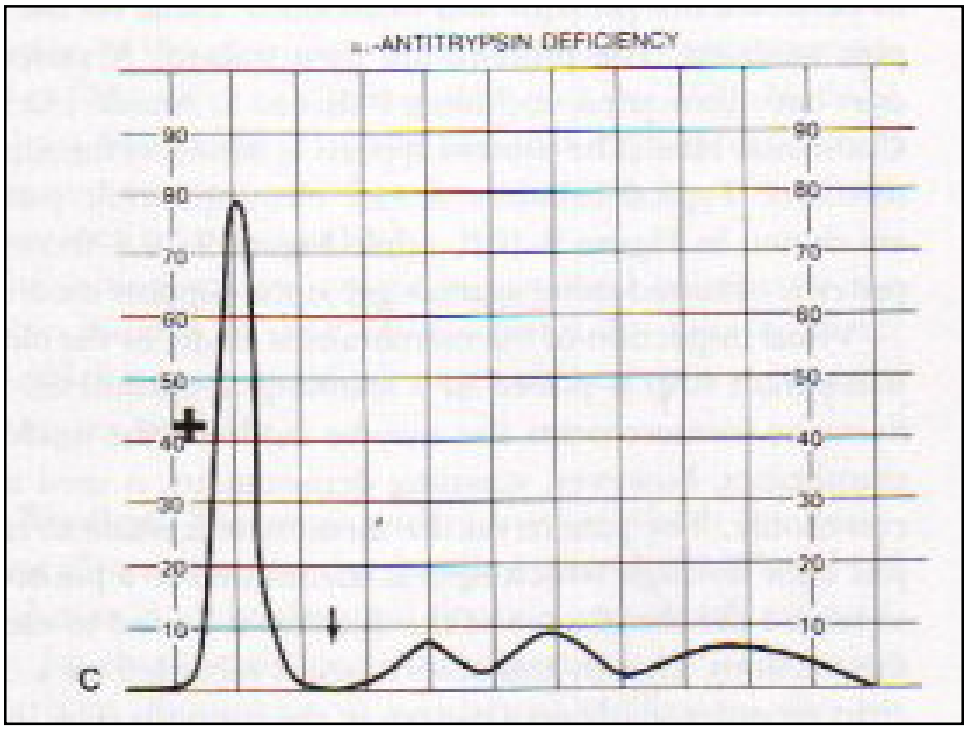
Electrophoresis

- Separates proteins on the basis of their charge.
- Types:
  - Free boundary: separation under an electric field in a fluid media. Separates plasma proteins five bands: albumin(54-58%), α1 globulins (6-7%), α2 globulins(8-9%), β globulins (13-14%), γ globulins (11-12%).
  - Zone electrophoresis: Separation under an electric field in a solid media e.g. paper, starch, cellulose, Acrylamide etc. Separates plasma proteins into: Albumin, α1 globulins, α2 globulins, β globulins, γ globulins and fibrinogen.

NORMAL HUMAN SERUM PROTEIN ELECTROPHORESIS




MONOCLONAL INCREASE
90
80
70
60
50
40
30
20
10
48.7%
30%
3.8%
9.5%
6.0%
1.9%
B


α1-ANTITRYPSIN DEFICIENCY
90
80
70
60
50
40
30
20
10
C

SERUM PROTEIN DEFECTS

- Normal serum protein levels:
  Total serum protein level: 63-83 g/dL.
- Hyperproteinaemia:
  Total serum protein level: > 90 g/dL.
- Hypoproteinaemia:
  Total serum protein level: < 63 g/dL.

INDIVIDUAL PROTEIN FRACTION

ALBUMIN

- A low molecular weight protein (M.Wt= 65,000).
- Functions include:
  - **Transport**
  - **Osmotic pressure regulation**
- Synthesized in the liver.
- Deficiency: in liver disease and kidney disease.

GLOBULINS

- Heterogenous group
- Can be separated into different fractions on the basis of their electrophoretic mobility and sedimentation coefficient:

α1-Globulin	-	α1- Fetoprotein
		α1- Antitrypsin
α2- Globulin	-	α2- Fetoprotein
		Haptoglobin
β- Globulin	-	Transferrin
		Ceruloplasmin
γ- Globulin	-	Antibodies (immunoglobulins)

FIBRINOGEN

- A globulin of very high mol. wt.
- Can be precipitated easily.
- Can be converted to fibrin which causes the blood clot formation.
- Synthesized exclusively in the liver.

BIOCHEMICAL ABNORMALITIES OF PROTEINS

- Total protein abnormalities.
- Abnormalities of individual protein fraction:
  - Serum albumin.
  - Carrier proteins.
  - Protease inhibitors.
  - Immunoglobulins.
  - Embryonic and fetal protein abnormalities. associated with human neoplasia.

TOTAL SERUM PROTEIN ABNORMALITIES

Hypoproteinaemia may result from:

1. Water access caused as a result of:
   a. Overhydration.
   b. Artifactual cause - blood taken from the "drip" arm.
2. Excessive loss of protein (mainly albumin):
   a. Through the kidney in nephrotic syndrome
   b. From the skin after burns
   c. Through the skin in protein losing enteropathy.
3. Decreased synthesis of proteins
   a*. Severe dietary protein deficiency e.g. in Kwashiokar
   b*. Severe liver disease (mainly albumin).
   c. Severe malabsorption.

* There may be no fall in total protein if γ-globulin is raised

HYPOLBUMINAEMIA

- Normal albumin level = 32-52 g/L.
- Hypoalbuminaemia: the level of albumin <32 g/L.
- Frequently encountered.
- Consequence:
  - Oedema
  - Hypocalcaemia
  - Alteration in the levels of protein-bound substance due to loss of carrier protein.

CAUSES OF HYPOALBUMINAEMIA

- Decrease albumin synthesis:
  a. Liver disease (specially chronic diseases).
  b. Malnutrition.
  c. Alcoholism
- Increased albumin loss:
  a. Renal disease (nephrotic syndrome).
     - Loss of albumin in urine (proteinuria).
  b. Extensive burns:
     - Loss of albumin through skin - transdution.

CAUSES OF HYPOALBUMINAEMIA

.....Contd

- Defective intake:
  a. Malabsorption due to gastro-intestinal disease
- Protein-losing enteropathy (rare)
  - Excessive loss of protein from the body into the gut.
  - Occurs in a variety of conditions such as :
    a. Ulceration of the bowel.
    b. Lymphatic obstruction.
    c. Intestinal lymphaangiectasis.

CAUSES OF HYPOALBUMINAEMIA
.....Contd

- Haemodilution
  a. Over hydration.
  b. Late stage of pregnancy.
- Artefactual
  a. Blood drawn from “drip” arm.
- Non-specific causes (common)
  - In many acute conditions including minor illnesses such as colds and boils.
  - Often in hospitalized patients.
  - Upright position when drawing blood.
  - Newborn babies.
- Increased degradation of albumin. In:
  - Idiopathic
  - Familial idiopathic hypercatabolic hypoproteinemia.
  - Wislcott-Aldrich syndrome

ABNORMALITIES OF CARRIER PROTEINS

α1-globulin

- The normal serum level of α1-globulin is 1-3g/L.
- α1-lipoprotein transport cholesterol.
  - In a rare genetic disorder, α1-lipoprotein deficiency (Tangiers disease), its level is reduced causing the accumulation of cholesterol esters in tissues resulting in:
    - Tonsillar enlargement.
    - Hepatomegaly.
    - Lymphadenopathy

α1-FETOPROTEIN (AFP)

- AFP is synthesized in fetus at 14-40 weeks of gestation.
- AFP levels decline rapidly after 2 weeks of age.
- In adults it is found primarily in:
  - association with hepatocellular cancer of liver and embryonic tumor of the ovary and testes.
  - Cases of gastric and prostatic carcinoma.
  - Viral hepatitis.
  - Cirrhosis.
- AFP detection is very useful in diagnosis of primary liver cancer.

α2-GLOBULIN

The normal α2–globulin level is 6-10 g/L of serum.

α2-Macroglobulin make up most of α2-globulin fraction.

- It is a large molecule
- In nephrotic syndrome, it is retained in serum and levels are found to increase.
- Haptoglobin : binds free haemoglobin. Low levels are found in hemolytic conditions since the haptoglobin/ haemoglobin complex is catabolised better than free haptoglobin.

β-GLOBULIN

Normal level of β-globulin in serum is 7-11 g/L.

- β-lipoprotein transport cholesterol in serum.
- Abetalipoproteinaemia is the complete absence of β-lipoprotein, pre β-lipoprotein and chylomicron. This causes:
  - . Inability to transport lipid from intestine or the liver.
  - . Plasma cholesterol deficiency.
  - It is clinically characterized by intestinal malabsorption under steatorrhea, progressive atasia, retinitis pigmentation and crenation of erythrocytes.
- High levels of β-globulin are found in pregnancy, biliary obstruction and nephrotic syndrome.

TRANSFERRIN

- Transferrin is a β-globulin.
- It binds free iron in serum.
- Normally it is about one third saturated with iron.
- Transferrin levels are decreased in:
  - *Liver disease (e.g. cirrhosis).*
  - *Chronic infections.*
  - *Nephrosis.*
  - *Congenital atransferrinaemia.*
- Increased serum transferrin levels occur during increased transferrin synthesis caused as a result of iron deficiency anaemia.

γ- Globulins

Mostly Immunoglobulins – Ig G,A,M, D & E

Cryoglobulins

- Pure monoclonal → IgG, IgM, IgA
- Mixed. Consist of complexes of immunoglobulins or altered immunoglobulins.
- Insoluble at 4°C. Aggregate at 30°C

BIOCHEMICAL INVESTIGATIONS IN THE DIAGNOSIS OF DISEASE STATES

TYPES OF BIOCHEMICAL TESTS

1. Discretionary tests.
2. Profile and screening investigations.
   a. On patients.
   b. On apparently healthy individuals.

EXAMPLES OF ORGAN-SPECIFIC PROFILES

	TESTS
- Electrolyte profile	Na+, K+, Cl^-, HCO^-_3
- Liver function tests	Bilirubin, alkaline phosphatase, alanine transaminase (SGPT), Plasma albumin.
- Bone Profile	Ca^{x2}, alkaline phosphatase, phosphate.
- Kidney function tests	Creatinine, urea.
- Acid-Base balance	pH, PCO_2, HCO^-_3
- Cardiac profile	Lactate dehydrogenase (LDH), Creatinine phosphokinase (CPK), Aspartate transaminase (SGOT)
- Endocrine profile	T3, T4, TSH, and Thyroid function

COMMONLY REQUESTED DISCRETIONARY BIOCHEMICAL TESTS

TEST	SUSPECTED DISEASE
- Bilirubin	Liver disorders.
- Glucose	Diabetes Mellitus
- Iron and Total Iron Binding capacity	Anaemias
- Urea	Renal function
- Creatinine	Renal function
- Uric acid	Gout
- Electrolytes	Water and electrolyte balance
- Plasma enzymes	Liver, cardiac, muscle, etc.
- Cholesterol/Lipids	Cardiac diseases
- Blood gases	Acid-Base balance

INHERITED ABNORMALITIES OF THE Plasma Proteins

DEFICIENCY	ASSOCIATED ABNORMALITY
$\alpha 1$-Antitrypsin	Obstructive pulmonary disease (Chronic or emphysema) liver disease.
Anti-thrombin	Thrombosis Pulmonary embolism
Immunoglobulin	Severe recurrent or chronic infection
Complement	Severe, recurrent infection.
C1 esterase inhibitor	Recurrent non-pruritic swelling of skin and mucus membrane (hereditary angioneurotic edema)

ALTERATION OF specific PLASMA PROTEIN CONCENTRATION

PROTEIN	INCREASED IN	DECREASED IN
Albumin	Dehydration	- Acute and chronic liver disease. - Malnutrition - Malabsorption - Cirrhosis of liver - Burns - Severe trauma - Nephrotic syndrome
Transferrin	- Iron deficiency - In woman taking oral contraceptives.	- Protein losing conditions - Infection; and - Neoplastic disease Contd.....

ALTERATION OF Specific PLASMA PROTEIN CONCENTRATION .....Contd

PROTEIN	INCREASED IN	DECREASED IN
Ceruloplasmin	- Chronic liver disease - Some infections.	Wilson disease
Haptoglobulin		Haemolytic anaemia
α_1-Antitrypsin		Pulmonary emphysema.
α_2-Macroglobulin	Nephrotic syndrome collagen disorder	Liver disease in children leading to cirrhosis.
α-Fetoprotein	Hepatocellular carcinoma	
Fibrinogen		- Congenital fibrinogen def. - Shock. - Complication of pregnancy. - Major surgery - Snake bites. - Disseminated carcinoma

INFLAMMATORY RESPONSE

- Assessment of the presence and degree of inflammation can be obtained from the levels of "acute phase protein"
  - Positive acute phase proteins: Increase during inflammation.
  - Negative acute phase proteins: decrease during inflammation.

ACUTE PHASE PROTEINS

- Indicators of inflammatory disease :
  - ESR
  - Leukocytosis
  - Fever
- Indicate active state of inflammation.
- Constitute: α1-antitrypsin
- Carrier proteins:
  - Haptoglobin.
  - Ceruloplasmin.
  - Fibrinogen.
  - C-reactive proteins
  - α1-acid glycoprotein

CLINICAL INDICATIONS FOR ASSESSMENT OF ACUTE PHASE PROTEINS

- Presence of inflammatory disease.
- Differential diagnosis of inflammatory disease.
- Estimation of the endpoint of therapy.
- Monitoring therapeutic effectiveness.
- Postsurgical follow-up in patients at risk of postoperative infections.
- Follow-up of patient with malignancy.

POSITIVE ACUTE PHASE PROTEINS

- $\alpha 1$-antitrypsin.
- $\alpha 1$-antichymotrypsin.
- $\alpha 1$-acid glycoprotein.
- Ceruloplasmin.
- Haptoglobin.
- Complement component C3 and C4.
- Antithrombin III.

SPECIFIC INDICATIONS FOR QUATIFICATION OF SOME ACUTE PHASE PROTEINS

PROTEIN	DISEASE
α1-antitrypsin	- Chronic obstructive pulmonary disease. - Neonatal hepatitis syndrome cytogenic cirrhosis.
Ceruloplasmin	Hepatitis or cirrhosis (unexplained)
Haptoglobin	In-vivo haemolysis. Ineffective erythropoiesis

NEGATIVE ACUTE PHASE PROTEINS

- Albumin.
- Transferrin.
- Pre-albumin.

EMBRYONIC AND FETAL PROTEIN ASSOCIATED WITH HUMAN NEOPLASIA

- Several fetal proteins and synthesized in human tumors.
- They are released in biological fluid.
- Useful in - diagnosis of malignancy
  - monitoring of therapy for cancer
  - evaluation of prognosis:
- The protein often found associated with tumors are:
  - – α1-fetoproteins
  - – α2-H fetoprotein
  - – β2-S fetoprotein
    - regain alkaline phosphatase
    - fetal sulphoglycoprotein antigen
  - – γ-fetoproteins
    - Carcinoembryonic antigen of the gastrointestinal tract.

INHERITED ABNORMALITIES OF PLASMA PROTEINS

DEFICIENCY	ASSOCIATED ABNORMALITY
α1-Antitrypsin	Obstructive pulmonary disease (Chronic bronchitus or emphysema), liver disease.
Anti-thrombin	Thrombosis. Pulmonary embolism.
Immunoglobulin	Severe recurrent or chronic infection
Complement	Severe, recurrent infection.
C1 esterase inhibitor	Recurrent non-pruritic swelling of skin and mucus membrane (Hereditary angioneurotic edema).

EMBRYONIC AND FETAL PROTEIN ASSOCIATED WITH HUMAN NEOPLASMA

- Several fetal proteins are synthesized in human tumors.
- They are released in biological fluid.
- Useful in:
  - diagnosis of malignancy.
  - monitoring of therapy for cancer.
  - evaluation of prognosis.
- The protein often found associated with tumors are:
  - α-feto proteins
  - α2–ferroprotein.
  - β-fetoprotein.
  - Alkaline phosphatase.
  - Fetal sulphoglycoprotein antigen.
  - γ-fetoproteins.
  - Carcinoembryonic antigen of the gastrointestinal tract.

METHODS USED IN IDENTIFICATION AND QUANTITATION OF NORMAL AND ABNORMAL BLOOD PROTEINS

a. Plasma Proteins
b. Haemoglobin

METHODS FOR PLASMA PROTEIN ESTIMATION

- Quantitation:
  - *Total Protein*
  - *Albumin*
  - *Globulin*
- Manually - Biuret method. Colour development with Cu^{+2} reagent.
- Autoanalyser - SMAC
  - American monitor
- Method for specific protein:
  - *Immunodiffusion e.g. transferrin, immunoglobulins*
  - *Nephelometric method e.g. Albumin, α1-antitrypsin, immunoglobulins.*
  - *RIA method e.g. Ferritin, Immunoglobulin, Protein Hormones.*

IDENTIFICATION

- Electrophoresis:
  - Widely used method.
  - Simple.
  - Proteins are separated on the basis of the charges under an electric field.
  - Useful investigation of disease states e.g. liver, renal diseases, infections.

IMMUNODIFFUSION

- Used for specific protein identification.
- Simple procedure.
- Proteins are identified on the basis of precipitation reaction with respective antibodies.

IMMUNOELECTROPHORESIS

- Complex procedure.
- Accurate.
- Proteins are identified on the basis of their change and precipitation reaction with respective antibody.

METHODS USED FOR ESTIMATION OF HAEMOGLOBIN

- **Estimation of total haemoglobin:**

a. Manually: Cyanomethaemoglobin method
   - Not used commonly.
   - Not very accurate.

b. Autoanalyzer: Coulter Counter with haemoglobinometer attachment:
   - Widely used.
   - Very accurate.
   - Simple.
   - Estimates total RBC, WBC, MCV, MCH, MCHC.

IDENTIFICATION OF HAEMOGLOBIN TYPES

a. Electrophoresis at alkaline acid pH
   - Simple procedure
   - Accurate
   - Useful for identification of several Hb variants (not all).
   - Proteins can be quantitated by using a densitometer.

IDENTIFICATION OF HAEMOGLOBIN TYPES

b. Isoelectric Focussing:

- Separation on the basis of isoelectric pH of haemoglobin variants.
- Simple method.
- Does not separate all variants.

LABORATORY INVESTIGATIONS OF ANAEMIA

- Haemoglobin, RBC and PCV.
- Red cell indices.
- Red cell morphology.
- Iron and TIBC estimation.
- Hb A_2 and F estimation.
- Haemoglobin electrophoresis at acid and alkaline pH.

METHODS USED FOR INVESTIGATION OF HAEMOGLOBINOPATHIES

- Detection of haemoglobinopathies and thalassaemias - Haematological Tests.
  - Hb
  - RBC count
  - PCV
  - MCH
  - MCHC
  - Red cell morphology

Contd.....

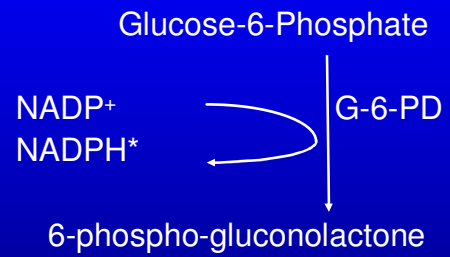
METHODS USED FOR INVESTIGATION OF HAEMOGLOBINOPATHIES ...Contd

- Differentiation and confirmation of haemoglobinopathies and thalassaemias:
  - Electrophoresis.
  - Hb A_2 quantitation.
  - Hb F quantitation.
  - Hb stability test.
  - Determination of α/non-α globin chain ratio.
  - Studies at gene level.

BIOCHEMICAL TEST IN THE INVESTIGATION OF G-6-PD PD DEFICIENCY

1. Estimation of red cell G-6-PD activity
   - Spot tests
   - Spectrophotometric method.
2. Phenotyping by electrophoresis.

ESTIMATION OF G-6-PD ACTIVITY REACTION



* NADPH is estimated by measuring
  a. Fluorescence (under UV Lamp)
  b. Absorbance at 340 nm

LIVER FUNCTION TESTS

The principal function of the liver include:

- Conjugation and excretion of bilirubin.
- Metabolism of carbohydrates, proteins and lipids.
- Detoxication of drugs, metabolite and hormone.
- Excretion of various natural and foreign substances into the biliary tract.
- Storage.

PATHOLOGICAL CHANGES IN LIVER DISEASE

a. Liver cell damage

(acute hepatitis, toxins, chronic hepatitis, prolonged biliary obstruction, cirrhosis, hepatic congestion).

b. Cholestasis

- Intrahepatic cholestasis:

(Viral hepatitis, biliary cirrhosis, infiltration of the liver).

- Extra hepatic cholestasis

(Gallstone in the common bile duct, fibrosis of the bile duct, carcinoma of head of pancreas, external presence of tumour).

PLASMA PROTEIN CHANGES IN LIVER DISEASES

Liver disease	HPT	A1b	C3	LDL	IgG	IgM	IgA	TRF	Pre-Alb	α1-AT
"Pure" Biliary Obstruction	↑↑		↑↑	↑↑						
Advanced Hepatic Cirrhosis	↓↓	↓	↓		↑↑	(↑↑)	↑↑	↓	↓↓	↑
Acute Viral Hepatitis	(↓)				(↑)	↑	(↑)			↑
Infection Mononucleosis	↓				(↑)	↑	(↑)			

↓ = Decrease
↑ = Increase
(↓) = May be decreased
(↑) = May be increased

CONSEQUENCES OF LIVER DISEASES

- ↓ Synthesis of plasma proteins.
- ↑ Release of hepatic proteins and enzymes.
- ↓ Excretion of some metabolites.

LIVER FUNCTION TESTS

- Total bilirubin.
- Transaminase (SGPT & SGOT)
- Alkaline phosphatase.
- Albumin
- Total protein

TESTS PERFORMED IN SUSPECTED CASES OF DIFFERENT LIVER DISEASES

Tests	Acute Hepatitis	Chronic Hepatitis	Cirrhosis	Choles-tosis	Hepatic Infiltration	Hepato-cellular Carcinoma
Plasma Bilirubin	√		√	√		
SGOT SGPT	√ √	√	√ √	√	√	√
Urinary Bilirubin Urobilirubin	√ √					
Hepatitis associated antigen		√				
Plasma protein electrophoresis		√	√			
Alkaline phosphatase			√	√	√	√
5' Nucleotidase			√	√	√	√
α-Fetoprotein						√
γ-Glutamyl Transfe-rase				√		

TYPES OF BILE PIGMENTS PRESENT IN PLASMA, URINE AND FEACEA IN DIFFERENT TYPES OF JAUNDICE

Disease	Plasma		Uric		Feaces
	Total Bilirubin	Excess Conju-gated Bilirubin (Direct Van der Brough Reaction)	Uro Bilirubin	Bilirubin	Urobilirogen
Normal	Present	-	Present	Absent	Present
Haemolytic Jaundice	+	-	Increased	Absent	⇈
Hepatic (Infective hepatitis)	⇈	+	Variable	+	Low
Post Hepatic	III	⇈	Absent	⇈	Absent

FUNCTIONS OF THE KIDNEY

- To excrete water and ions from the body.
- To maintain the composition of plasma normal by excreting or reabsorbing substances.
- To maintain acid-base balance.
- To excrete metabolic end products, hormones, drugs.
- To control blood pressure.

CHOICE OF RENAL FUNCTION TESTS

TESTS	CONDITION
- Examination of urine	Suspected renal damage.
- The water deprivation or vasopressin test	Most useful single test to confirm renal tubular impairment.
- Creatinine clearance	Quantitative test for glomerular impairment.
- Estimation of plasma urea and creatinine	Guide to progress and prognosis if there is severe renal damage or obstruction.

PLASMA PROTEIN CHANGES IN RENAL DISEASE

- ↓ Albumin
- ↑ α2-globulins
- ↓ γ-globulins (often)
- ↓ C3 and C4 in acute glomerulonephritis
- ↓ C3, normal C4 in membrane proliferative glomerulonephritis.

URINARY PROTEIN CHANGES IN RENAL DISEASES

Glomerulal Proteinura*	**Overflow Proteinura**	**Revert Tubular Disease Proteinuria****	**Nephrogenic Proteinuria**
↑ Albumin	↑ Bench flow protein	↑ α2-Globular	↑ IgG, IgM
↑ Transferrin	↑ Myoglobin ad	↑ β-Globular	IgE
↑ Acid Glycoprotein	Haemoglobin	Slightly ↑ albumin and transferrin	
↑ α1-Antitrypsin	↑ Acid glyco-protein	↑ β2-Microglobulin	
↑ IgG	↑ α1-Antitrypsin	↑ Lysozone	

* Ratio of albumin to low mol-wt. protein 20:1
** Ratio of albumin to low mol-wt. protein 1:1

BLOOD TESTS FOR BONE DISEASE

Blood Ca^{+2} level.

- Blood phosphate level.
- Alkaline phosphatase level.
- Parathyroid hormone level.

BLOOD TESTS FOR DIAGNOSIS OF DIABETES MELLITUS

- Random blood glucose estimation.
- Fasting blood glucose estimation.
- Testing for glucose in urine.
- Two-hour post-glucose blood glucose level.
- Glucose Tolerance Test (GTT).

MUSCLE DISEASE TESTS

Creatine phosphokinase (CPK).

- Creatine and Creatinine in Serum.
- Calcium.
- Na^{+} in Serum.
- K^{+} in Serum.
- Mg^{+2} in Serum

Myocardial infarction

Cardiac enzymes / Proteins

- Myoglobin- increase in serum within 2-6 hrs.
- CPK - first enzyme to respond.
  - Informative in recurrent infarction.
  - MB fraction more specific.
- Troponin T - Heart specific (as sesitive as CPK-MB in first 48 Hrs)
- Troponin I – Heart specific 6-8 hrs more sensitive than CPK (not increased by muscle injury)
- LDH – Non specific . LDH incearse in 10-12 hrs, peaks in 48-72 hrs, remain increase 10-14 days.