

PLASMA PROTEINS & ENZYMES

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LEARNING OUTCOMES

- Discuss the clinical significance of measuring total plasma protein and different fractions
- Define the acute phase response and discuss the clinical significance of acute phase proteins
- List the common enzymes used in clinical diagnostics and explain the pathological basis of their alterations
- Describe sample collection requirements for proteins & enzymes

WHAT ARE THE CLINICALLY SIGNIFICANT PLASMA PROTEINS?

- Albumin
- Haptoglobin
- Ceruloplasmin
- Transferrin
- Ferritin
- Gamma globulins

WHAT ARE THE FACTORS AFFECTING PLASMA PROTEIN CONCENTRATION?

- Rate of protein synthesis
 - Paraproteinaemia
 - Liver disease
- Rate of removal
 - Increased excretion
 - Increased capillary permeability
- Volume of distribution
 - Dehydration/overhydration
- Artefactual

WHAT IS ACUTE PHASE RESPONSE?

- Physiological or systemic changes accompanying inflammation
- Occur distant to the site/sites of inflammation
- Seen in both acute and chronic inflammation
- Alterations in concentrations of many plasma proteins

WHAT ARE ACUTE PHASE PROTEINS?

- Definition – a plasma protein of which the concentration changes by 25% (at least) during inflammation
- Produced by the liver
- Mechanism – monocytes and macrophages at sites of inflammation



release cytokines



induce acute phase response

ACUTE PHASE PROTEINS

Positive

- C reactive protein
- Haptoglobin
- Ferritin
- Fibrinogen
- Ceruloplasmin
- Alpha1-Antitrypsin

Negative

Albumin
Transferrin

C-REACTIVE PROTEIN (CRP)

- An important marker of inflammation
- Normal <5mg/L
- Increased in
 - Rheumatoid arthritis
 - Crohn disease
 - Bacterial induced inflammation
- Unchanged in
 - Viral infections
 - SLE

HIGH SENSITIVITY C-REACTIVE PROTEIN (hsCRP)

- High sensitivity C (hsCRP)
- Rises of CRP in the range of 0.1 – 5mg/L
- Marker of cardiovascular risk
- Other markers of sepsis/inflammation
 - Procalcitonin
 - Lactate

OTHER MARKERS OF INFLAMMATION

- Procalcitonin
- Serum Lactate
- Systemic Inflammatory Response Syndrome

ALBUMIN

- Most abundant plasma protein
- Half life ($t_{1/2}$) –20 days
- Transport many substances
 - Calcium, fatty acids, bilirubin
- How does a low serum albumin cause oedema?
- Is albumin the only determinant of oncotic pressure?

WHAT ARE THE CAUSES OF HYPOALBUMINAEMIA?

- Reduced synthesis
 - –Malnutrition
 - –Malabsorption
 - –Chronic liver disease
- •Increased excretion or degradation
 - –Nephrotic syndrome
 - –Burns
 - –Protein-losing enteropathies
- •Increased volume of distribution
 - –Overhydration
 - –Increased capillary permeability

ASSAY OF SERUM ALBUMIN: INDICATIONS ?

- Patients with oedema
- To assess liver function
- Multiple myeloma and other plasma cell disorders
- Serum calcium assessment
- Nutritional assessment

OTHER PROTEINS

- **Haptoglobin**

- Binds free Hb in intravascular haemolysis
- The complex is removed by RES
- Concentration reduced in haemolysis

- **Ceruloplasmin**

- Copper transport protein
- Reduced in Wilson disease
- Increased in inflammation

TRANSFERRIN & FERRITIN

- Important in assessing iron status
- Fasting transferrin saturation >45% suggestive of haemochromatosis
- Ferritin –sensitive marker for iron deficiency
- Both affected by acute phase response

GAMMA GLOBULINS

- Mono clonal increase
 - –Multiple myeloma
 - –Waldenstrom macroglobulinaemia
 - –Chronic lymphocytic leukaemia and B cell lymphoma
 - –Benign paraproteinaemia
- Para protein –Immunoglobulin produced by a single clone of cells

QUESTIONS

- What is the tube used to collect blood for protein electrophoresis?
- What are the biochemical investigations requested in a patient suspected with multiple myeloma?

- **PLASMA ENZYMES**
- **CLINICAL SIGNIFICANCE**

PLASMA ENZYMES -OBJECTIVES

- What are the clinically significant plasma enzymes?
- What are the limitations of enzyme assays?
- Why do enzyme activities increase in plasma?
- Explain the pathological basis for alterations, in disease conditions

PLASMA ENZYMES OF CLINICAL SIGNIFICANCE

- Amylase
- Creatine kinase and Creatine kinase-MB
- Lactate dehydrogenase
- Aspartate aminotransferase
- Alanine aminotransferase
- Alkaline phosphatase
- Gamma glutamyl transferase

DISADVANTAGES OF ENZYME ASSAYS

- Lack of specificity
- Many are common to more than one tissue
- E.g. AST -liver, heart, red cells
- How to overcome?
- assay two or more enzymes of the same tissue
- Isoform/isoenzyme analysis
- Clinical correlation
- Serial enzyme assays

INCREASED PLASMA ENZYME ACTIVITY- CAUSES

Cell damage

–E.g.ALT in acute viral hepatitis

–Lactate dehydrogenase in haemolysis

•Increased enzyme synthesis

–E.g.GGT by Alcohol, drugs

•Reduced clearance from plasma

–E.g.Serum amylase in renal failure

SERUM AMYLASE

- Sources: exocrine pancreas, salivary glands
- Excretion : 25% via the kidney
- Indication : acute pancreatitis
- Persistent elevation : pancreatic pseudo cyst

- Other causes of increased activity?

CREATINE KINASE

- CK & CK-MB
- Total CK increased in
 - Physical exercise
 - Rhabdomyolysis/myositis
 - Statin therapy
 - Hypothyroidism
- CK-MB –useful in the diagnosis of re-infarction of the myocardium

ACUTE CORONARY SYNDROMES

- ST-elevation myocardial infarction
- Non-ST elevation myocardial infarction
- Unstable angina

BIOCHEMICAL MARKERS FOR ACUTE MYOCARDIAL INFARCTION

- Conventional
 - –Cardiac troponin T
 - –Cardiac troponin I
- High sensitivity cardiac troponin
 - –hs Troponin T
 - –hs Troponin I

HIGH SENSITIVITY CARDIAC TROPONIN (hs TROPONIN)

- Able to detect lower levels of Troponin earlier compared to conventional assays
- When to check?
 - On admission
 - 3 hours after
- If there is no elevation in the 3 hour sample, It is unlikely to be an MI.

CARDIAC TROPONINS

- Structural proteins within myocytes
- Bound to actin and myosin
- **Troponin I more specific for myocytes**
- Troponin T expressed in skeletal muscle
- Levels begin to rise soon after myocyte injury
- Conventional assays show elevated levels 4 hours after the onset of chest pain.

ELEVATED CARDIAC TROPONIN –NON CORONARY CAUSES

- Renal failure
- Heart failure
- Extreme tachyarrhythmias/bradyarrhythmias
- Pulmonary embolism
- Myocarditis

LACTATE DEHYDROGENASE

- No longer a useful marker for cardiac injury.
- Important marker of haemolysis
 - Megaloblastic anaemia
 - Haemolytic anaemia
- Prognostic marker in
 - Lymphoma
 - Testicular cancer

REFERENCES

- Marshall WJ, Bangert SK and Lapsley M. Plasma proteins and enzymes. In: Clinical Chemistry.9th ed.2017.p.285-301.