# Chapter 13

Part 2

Plasma Enzymes

# Enzymes Clinical Diagnosis

Plasma enzymes can be **classified into two major groups.**

- **First**, enzymes are secreted into the blood .. Example, enzymes involved in blood coagulation.
- **Second**, enzyme are released from cells during normal cell turnover. These enzymes almost always function intracellularly, and have no physiologic use in the plasma.

**Increased** plasma levels of these enzyme may indicate tissue damage so can be use for diagnostics and prognosis for the patient. (Figure 5.20).

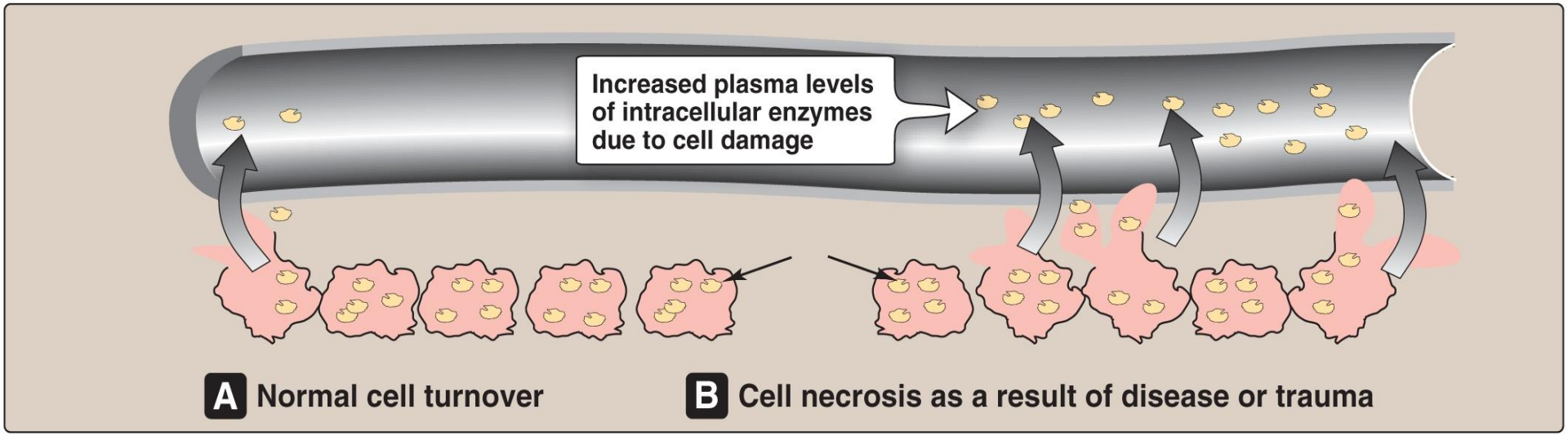



**Figure 5.20**
Release of enzymes from normal and diseased or traumatized cells.

# Possible causes for increase in plasma enzyme concentrations(activity)

- Increased cell turnover
- Cellular proliferation (e.g neoplasia/cancer)
- Increase enzyme synthesis (enzyme induction)
- Obstruction to secretion
- Decreased clearance

# Enzyme Activity

- **International unit – One IU is defined as the activity of the enzyme which transforms one micro mole of substrate into products per minute per liter of sample under optimal conditions and at defined temperature . It is expressed as IU/L**

# Disadvantages of enzyme assays

1. Lack of specificity for a particular tissue or cell type.
2. Many enzymes are common to more than one tissue.

This problem obviated to some extent in two ways:

1. Different tissues may contain two or more enzymes in different proportions.
2. Some enzymes exist in different forms (isoenzymes). Individual isoforms related to particular tissue.

# Alkaline phosphatase (ALP)

- Responsible for removing phosphate groups from many types of molecules, including nucleotides, proteins, and alkaloids.


Phosphate monoester  Alcohol  Phosphate

- **ALP** is the common name for a group of enzymes **(5 isoenzymes)**
- **ALP** is produced and present in high concentration in liver, bone, placenta and intestinal…etc.
- Every tissue produce one characteristic form so, **electrophoresis** is used to determine the type of ALP to know the origin tissue.

skeletal).

# Alkaline phosphatase (ALP)

**Clinical significance:**

**ALP Physiologically increase in**

- Pregnancy
- Childhood
- Fatty meals (intestinal ALP).

diagnosis of two groups of conditions;

**ALP increase** in;

**1. hepatobiliary disease**

- ✓(obstructive jaundice, cirrhosis, viral hepatitis and metastatic).

**2. bone disease**

- ✓**Osteomyelitis**
- ✓**Paget's disease**
- ✓Primary hyperparathyroidism with bone involvement.

**To determine the damage tissue with high ALP** **(vvi)**

- **ALP** and **γ-glutamyl transferase** that is found in **liver** but not in bone **indicate liver disease.**
- Increase **ALP** with **hypercalcemia** (increase level of **calcium** in plasma) multiple myeloma or **leukemia, bone disease**, osteomyelities and Paget's disease.

Figure 13.13 Causes of an increased plasma alkaline phosphatase activity. ULN, upper limit of normal.

**Causes of an increased plasma alkaline phosphatase activity**

***Physiological***

pregnancy (last trimester)
childhood

***Pathological***

often >5 × ULN
- Paget's disease of bone
- osteomalacia, rickets
- cholestasis (intra- and extrahepatic)
- cirrhosis

usually <5 × ULN
- bone tumours (primary and secondary)
- renal bone disease
- primary hyperparathyroidism with bone involvement
- healing fractures
- osteomyelitis
- hepatic space-occupying lesions (tumour, abscess)
- infiltrative hepatic disease
- hepatitis
- inflammatory bowel disease

# Alkaline phosphatase (ALP)

**ALP Physiologically increase in**

- Pregnancy
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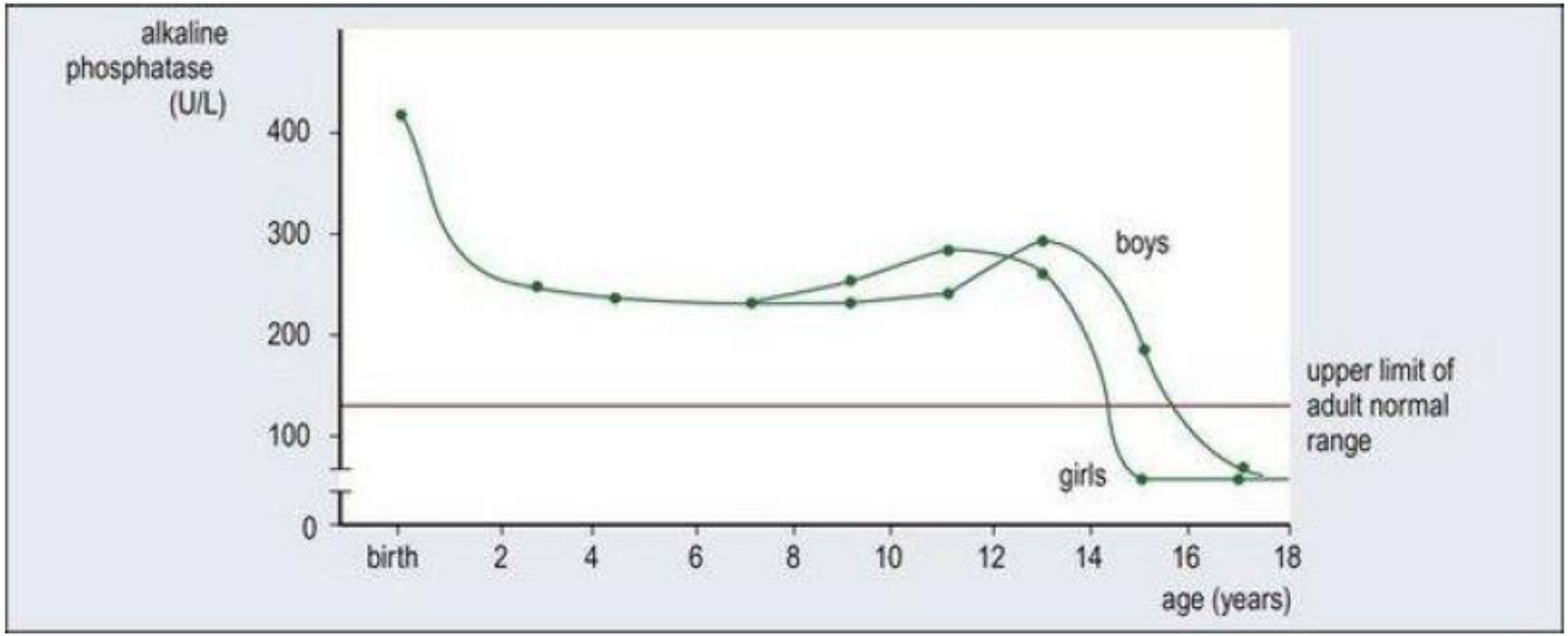
- Fatty meals (intestinal ALP).



# Acid Phosphatase (ACP)

- **ACP** is present in prostate, liver, bone, spleen, kidney, erythrocyte and platelets.

Orthophosphoric monoester + $H_2O$ → alcohol + $H_3PO_4$ **(remove phosphate group)**

## Clinical significance:

**Elevated levels** of serum Acid Phosphatase (ACP) in:

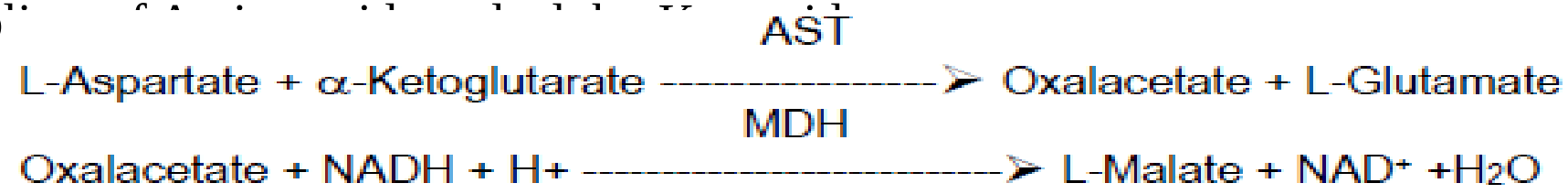
- Confirming and evaluating a diagnosis of **prostatic carcinoma**. (tumor marker).
- Paget's disease,
- hyperparathyroidism with skeletal involvement.
- Cancers which have invaded the bones.

# Aminotrasferases

Two used in diagnosis; aspartate aminotrasferases **(AST)** alanine aminotrasferases **(ALT).**

**1. aspartate aminotrasferases (AST) or glutamic-oxaloacetic transaminase (GOT). vvi**

**Functions**: The enzyme catalyses the transfer of amino groups during the metabo




**Tissue source**

high level of AST in cardiac, liver & SK muscle.

low level of AST decrease in kidney, pancreas &erythrocyte.

**Clinical significance:** AST levels are elevated in:

**1. myocardial infarction (MI)** **2. liver disease (hepatocellular damage).**

☹***heart attack*** **(myocardial infarction (MI))**

☹primary muscle disease

✓recent surgery and severe burns

# Aminotrasferases

**Clinical significance:** AST or (SGOT) levels are elevated in:

**Maximum elevations (> 20 times normal) (Fig. 13.15)**

- ✓ ***Acute viral hepatitis** **vvi**
- ✓ Sever tissue hypoxaemia

**High level (10-20 times normal)**

- ✓ ***myocardial infarction (MI)** **vvi**

**High level (5-10 times normal)**

- ✓ ***Chronic hepatites**
- ✓ **Cholestasis** (bile cannot flow from the liver to the duodenum)

**High level (2-5 times normal)**

- ✓ Metastatic hepatic tumors
- ✓ Acute pancreatitis
- ✓ Hemolytic anemia
- ✓ Hemolysis (ex; statins)

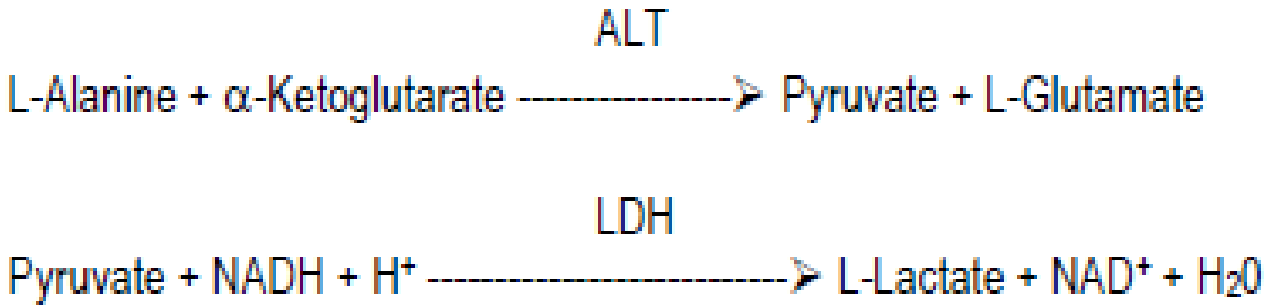
# Aminotrasferases

- AST (SGOT) increased 4-8 hours following a **myocardial infarction (MI)**, reaching its' peak in 2-3 days and declining on the fifth and sixth days.
- AST is not a specific or sensitive enough marker for the diagnosis of mycocardial infraction so cardiac troponins is much used.

# Aminotrasferases

2. alanine aminotrasferases **(ALT)** or Glutamic-Pyruvic Transaminase **(SGPT).**

Functions: ALT catalyzes the transfer of the amino group from L-alanine to α-ketoglutara ate.




**Tissue source**

**high level in liver**

**low level in cardiac, kidney & skeletal muscle.**

**ALT is considered more liver-specific than AST.**

# Aminotrasferases

**Clinical significance:**

**ALT** levels are **elevated** in:

- ✓**Acute or chronic hepatitis (cellular damage)**
- ✓ cirrhosis or scarring of the liver with loss of function.
- ✓Viral hepatitis.
- ✓cholestasis or congestion of the bile ducts
- ✓metastatic carcinoma.
- ✓Extensive liver damage from toxins or drugs.

**ALT is considered more liver-specific than AST.** **vvi**

- ❖**ALP** used to **detect and evaluate treatment** of acute hepatitis disease.
- ❖**ALP** **distinguish** between **MI** and **hepatic damage** (used with AST).
- ❖**ALP** used to **assess the hepatotoxicity of some drugs**.

# γ-Glutamyl transferase (GGT)

- GGT present in high concentration in liver, kidny and pancreas.
- Sensitive for hepatobiliary disease.

**Function:** GGT catalyzes the transfer of the γ-glutamyl group from Glutathione to amino.

**Clinical significance:** GGT levels are elevated in( fig 13.16)

- **High level (>10 times normal)**
  - ✓**Cholestasis**
  - ✓**alcoholic liver disease**
- **High level (5-10 times normal)**
  - ✓**Acute and chronic Hepatitis**
  - ✓**Cirrhosis (without cholestasis)**
  - ✓**Pancreatitis**
- **High level (< 5 times normal)**
  - ✓**Excessive alcohol ingestion**
  - ✓**Enzyme-inducing drugs**
  - ✓**Congestive cardiac failure**

# γ-Glutamyl transferase (GGT)

- GGT elevated in patients with liver diseases taking **alcohol**, **phenytoin**, **pheonobarbital** and rifampicin and can remain elevated for up to **3-4 weeks.**

  - increase both **ALP** and **GGT** indicate liver disease specially **cholestasis**. **VVI**
  - Measuring isoenzyme of **ALP** and **GGT** that is found in liver but not in bone so, **identify the origin tissue of ALP**. **VVI**

    - Measuring isoenzyme of ALP and γ-glutamyl transferase that is found in liver but not in bone so, **identify the origin tissue of ALP**.
    - **See figure 13.16 236**

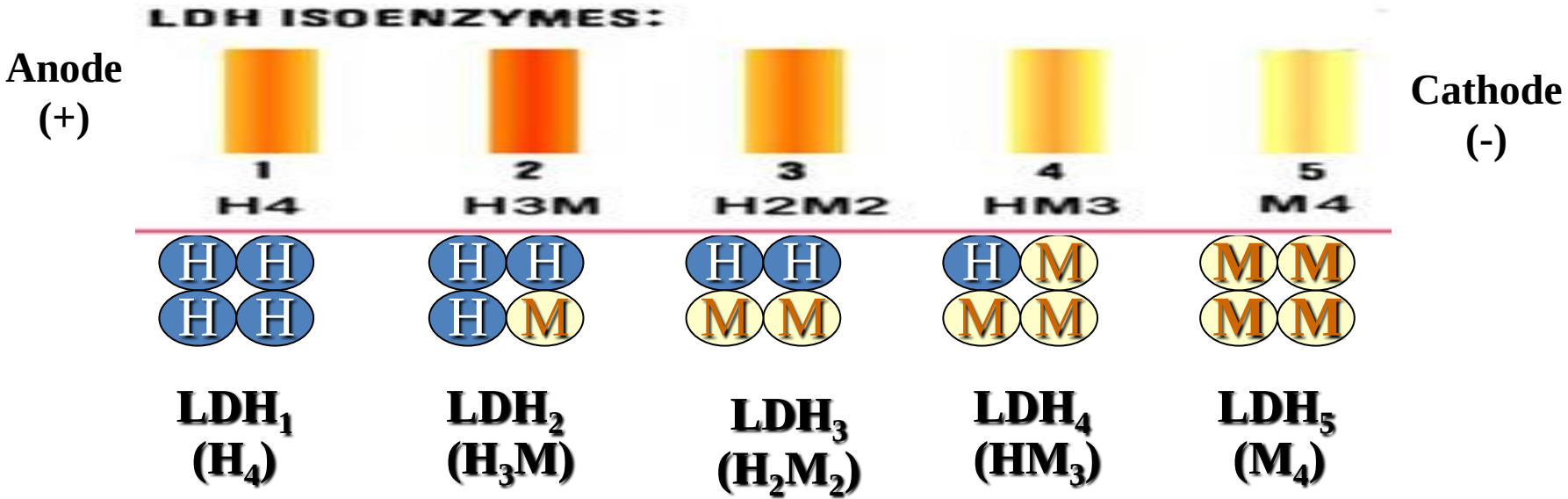
# Lactate dehydrogenase (LDH or LD)

- LDH is an enzyme found in nearly all living cells.
- LDH level are high in liver, skeletal muscle, kidney and erythrocytes.
- LDH exists as tetrameric composed of H and M subunits that form the five isoenzymes, LDH-1 (4H), LDH-2 (3H1M), LDH-4 (1H3M), LDH-5 (4M).

**Function:** Lactate dehydrogenase catalyzes the oxidation of lactate to pyruvate with simultaneous reduction of NAD to NADH.

$$\text{L-Lactate} + NAD^+ \xrightarrow{LD} \text{Pyruvate} + NADH + H^+$$

- LDH isoenzyme distinguished by electrophoresis due to different mobility.



# **Lactate dehydrogenase** (**LDH** or **LD**)

| LD | Tissue distribution | Clinical Significance |
|---|---|---|
| LD1 (H4) | Heart, RBC | MI/Hemolytic anemia megaloplastic anemia . Acute renal infraction |
| LD2 (H3M) | | Same as in LD1 |
| LD3 (H2M2) | mostly Lung Pancrease lymphocytes | Pulmonary embolism, pnemonia..etc |
| LD4 (HM3 | Mostly in liver | Hepatic inflamation and injury |
| LD5 (M4) | Mostly in Sk. muscles | Skeletal muscle injury |

- Both in RBC and heart muscle LD1 is dominant. It shows greater activity with substrate α– hydroybutarate rather than lactate. So it is also known as HBD/LD1.

- Normally LD2 is > LD1
- In MI LD1 will increase to a point at which LD1>LD2
- So it is called LD flipped pattern

# Creatine kinase (CK)

- Creatine Kinase exists as dimeric molecules composed of M and B subunits that form the isoenzymes CK-MM, CK-MB, and CK-BB.
- CK-MM are distributed primarily in the skeletal muscle.
- CK-MB are distributed primarily in the heart muscle.
- CK-BB is present mainly in the brain and in tissues composed of smooth muscle.

**Functions:** storage of energy in the form of phosphocreatine.

**Creatine** 

**Phosphocreatine**

**Clinical significance:**

- CK-MB increased in MI and rarely skeletal muscle damage.
- CK-MB detection is of importance in determining the degree of the injury and the efficacy of the treatment.
- **CK and LDH isoenzymes** provides a definitive diagnosis of acute **myocardial infarction (MI).**

Note: CK begins rise within 4–8 hours following onset of chest pain, reaches a peak of activity at 24 hours, and returns to baseline after 48–72 hours

Check Fig 13.17

# Amylase (serum 13-130U/L , urine 1-15U/Hr)

- Amylase is found in the salivary glands and exocrine pancreas.

**Functions:** that catalyses the hydrolysis of starch into sugars (act on α-1,4-glycosidic bonds).

## Clinical significance:

- In acute pancreatitis **α-amylase** starts to **rise** approximately 4 hours after the onset of pain, reaches a peak at 24 hours and remains elevated for 3-7 days.
- Amylase also increases in:

acute abdominal disorders appendictitis, intestinal obstruction

salivary gland disorders, mumps

Macroamylasamia (H.W.)???.

# Lipase

lipase catalyzes the hydrolysis of lipids to alcohol and fatty acids (RCOOH).

**Clinical significance:**

**Increase** Pancreatic lipase is important for diagnosis of **pancreatic diseases** and for associated **monitoring of therapeutic effects** (more specific).

- Persists for 5 days

# Cholinesterase (CHE)

- CHE enzyme secreted by the liver into the bloodstream.
- **Function**: cholinesterase break down an acetylcholine by preventing the accumulation of acetylcholine and the overstimulation of muscles and nerves.
- symptoms of overstimulation of muscle and nerve fibers cause difficulty in breathing or death.

**Clinical significance:**

**Low plasma activity of CHE in:**

- Physiologically during pregnancy
- chronic hepatic dysfunction.
- Liver disease
- Organophosphate (pesticides ) poisoning so ***Cholinesterase test helps doctors determine whether or not an individual is poisoned** **vvi**
- Cholinesterase hydrolysis a muscle-relaxant drug, used in anaesthesia (suxamethonium).
- Cholinesterase must be examined to avoid anesthesia in abnormal cases of cholinesterase activity .

# Tumor Markers
# Alpha-feto protein (AFP)

**Tumor markers** are substances, usually proteins, that are produced by the body in response to cancer growth.

- **AFP** is a major plasma protein produced by the yolk sac and the liver during **fetal development**.
- **AFP levels (>500ng/ml)** are increased in >90% of patients with **hepatocellular cancer.**
- AFP are useful in monitoring the response to therapy of hepatocellular cancer.
- **AFP** levels increase in **pregnancy (false positive)**. **vvi**

# References


- Clinical Biochemistry : Lecture notes, by Geoffery Beckett, Simon Walker, Peter Rae, Peter Ashby, Blackwell publishing, 7th edition, 2005, ISBN, 978-1-4051-2959-6
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- Clinical Biochemistry: an Illustrated color text, by Allan Gaw, Robert Cowan, Denis O'Reilly, and Michael Stewart Edinburgh: Churchill Livingstone, 3rd Edition, 2004,. ISBN 0-443-07269-8
- 
- Clinical Chemistry: Principles, Procedures, Correlations by Michael L. Bishop, Edward P. Fody, Larry E. Schoeff Publisher: Lippincott Williams & Wilkins; 5th edition (July 6, 2004) ISBN: 0781746116