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Department of Basic Science
Biochemistry Lab

Liver Function Tests
(AST, ALT and ALP)

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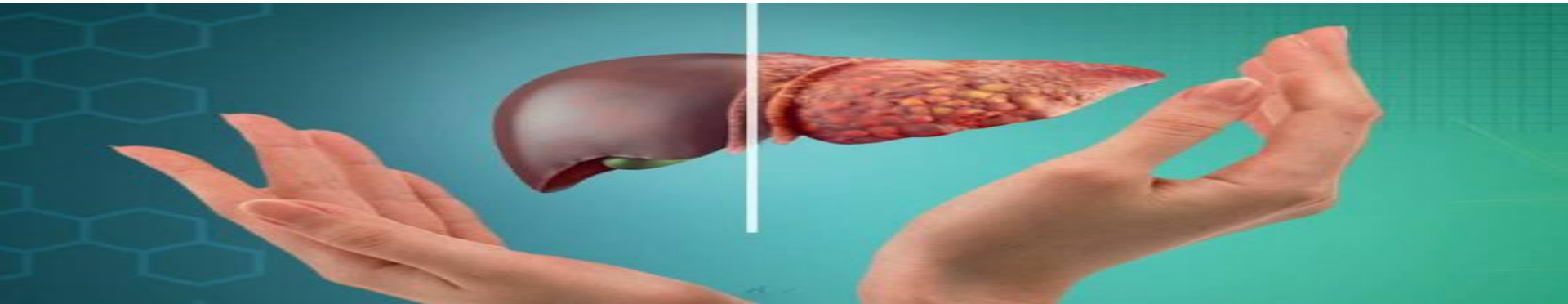
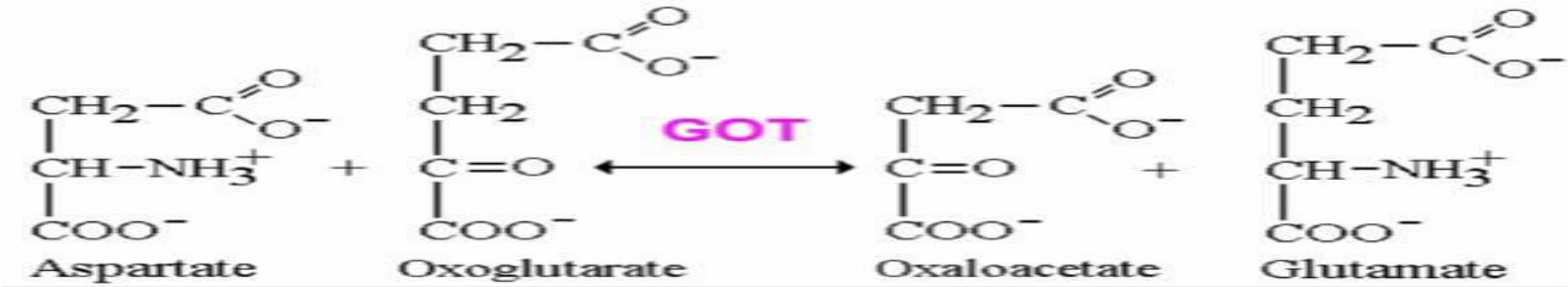


Liver enzymes; transaminases

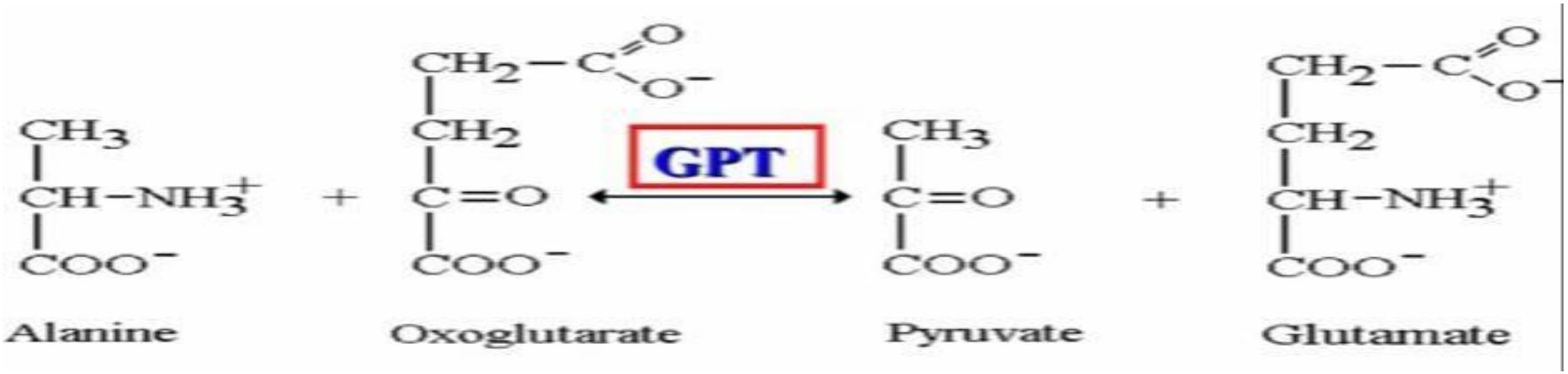
- Transaminases or an aminotransferases are enzymes that catalyze a type of reactions between an amino acid and an α-keto acid.

Most common transaminase enzymes are AST and ALT, which are used to test hepatocyte integrity and damage.

1. Aspartate transaminase (AST), also known as serum glutamate oxaloacetate transaminase (SGOT) which is catalyzes the reversible transfer of an α-amino group between aspartate and glutamate.



2. Alanine transaminase (ALT) or serum glutamate-pyruvate transaminase (SGPT) which is catalyzes the reversible transfer of an α-amino group between alanine and glutamate.

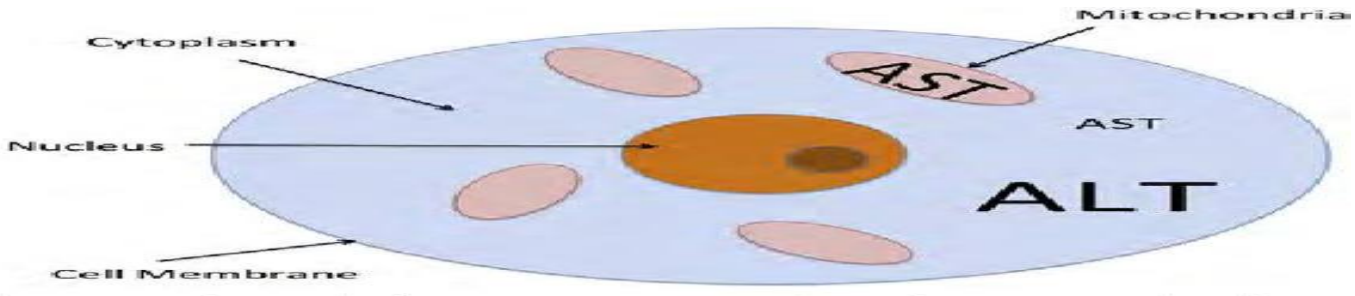


- AST is found normally in liver, heart, muscle, kidney and RBCs, (cytoplasm and mitochondria)
- ALT is predominantly found in liver (exclusively cytoplasm)

- AST and ALT are found in different distribution of hepatocytes, ALT exists in cytoplasm and if there is a slight liver cell damage, ALT firstly leak into the bloodstream, so that ALT increased.

- AST is mainly found in mitochondria of liver cells, the mitochondria are bubble in the liver cell cytoplasm, if there is a slight cell damage, AST don't leak in to the bloodstream, when AST significantly higher, mitochondria of liver cells are injuries.

- Increases ALT is more specific than AST for liver injury.



- After severe damage, AST levels rise 10 to 20 times and greater than normal, whereas ALT can reach higher levels (up to 50 times greater than normal). On the other hand, the ratio of AST to ALT (AST/ALT) sometimes can help determine whether the liver has been damaged.

- The normal concentrations in the blood are (5-40) U/L for AST and (5-35) U/L for ALT.

Elevated levels of AST may indicate:

- Alcoholic hepatitis (AST>ALT)
- Acute heart failure (AST>ALT)
- Acute hemolytic anemia
- Acute pancreatitis or inflammation of the pancreas.
- Cirrhosis of the liver.
- Hepatitis
- Heart attack
- Primary muscle disease also muscle injury
- Recent surgery
- Severe burns

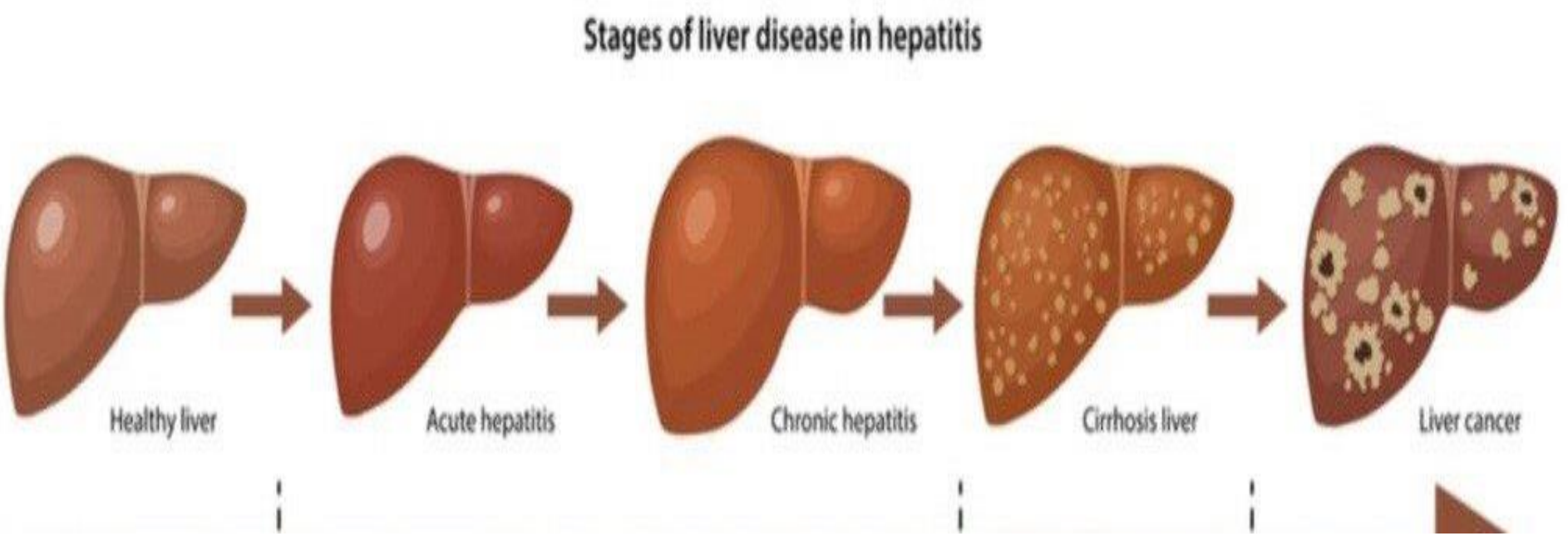
Elevated levels of ALT may indicate:

- Viral and chronic hepatitis
- Alcoholic liver disease
- Cancer of the liver
- Cholestasis or congestion of the bile ducts
- Cirrhosis or scarring of the liver with loss of function
- Death of liver tissue
- Noncancerous tumor of the liver
- Use of medicines or drugs toxic to the liver

What does the test result mean?

- Very high levels of ALT (more than 10 times the highest normal level) are usually due to acute hepatitis,(ALT>AST) often due to a virus infection.
- In acute hepatitis, ALT levels usually stay high for about 1-2 months but can take as long as 3-6 months to return to normal.
- ALT levels are usually not as high in chronic hepatitis, AST is increased more than ALT (AST>ALT) and AST/ALT ratio more than 1.

- Other causes of moderate increases in ALT include obstruction of bile ducts, cirrhosis and with tumors in the liver.



Liver enzymes; ALP

- Alkaline phosphatase (ALP) is a hydrolase enzyme responsible for removing phosphate groups from many types of molecules, including nucleotides and proteins. ALP is more effective in an alkaline environment.
- in humans, ALP is present in all tissues throughout the entire body, but is particularly concentrated in liver, bone (>95%), bile duct, kidney and placenta.
- ALP is increase physiological and pathological such as bone disease, liver disease, complication of pregnancy and intestinal disease.

ALP Interpretation

- **Increase**
  - Physiological (Child, Pregnant, bone healing)
  - Bone disease:
    - Metastatic carcinoma of bone
    - Osteogenic sarcoma
    - Myeloma
    - Paget disease: > 10-20 times
    - Osteomalacia, Rickets
    - Osteomyelitis

ALP Interpretation

- Liver disease
- Obstructive biliary system (stone and carcinoma) also cholestasis.
- Hepatitis
- Toxemia of pregnancy.

Normal range of ALP is (40-125) U/L

H.W Q/ The biochemical investigation of the blood specimen showed Bilirubin- 4.5 mg/dl, ALT- 150 IU/L, AST- 200 IU/L, Albumin- 25 mg/dL.

What is the likely biochemical diagnosis?

(a) Chronic hepatic disease, (b) Acute hepatic disease, (c) Hemolytic Jaundice, (d) Obstructive liver disease.

AST and ALT Procedure

MATERIALS REQUIRED

- Photometer or spectrophotometer with a thermostatted cell compartment set at 30/37°C, capable of reading at 340 nm.
- Stopwatch, strip-chart recorder or printer.
- Cuvettes with 1-cm pathlength.
- Pipettes to measure reagent and samples.

PROCEDURE

1. Preincubate working reagent, samples and controls to reaction temperature.
2. Set the photometer to 0 absorbance with distilled water.
3. Pipette into a cuvette:

Reaction temperature	37°C	30°C
Working reagent	1.0 mL	1.0 mL
Sample	50 µL	100 µL

4. Mix gently by inversion. Insert cuvette into the cell holder and start stopwatch.
5. Incubate for 1 minute and record initial absorbance reading.
6. Repeat the absorbance readings exactly after 1, 2 and 3 minutes.
7. Calculate the difference between absorbances.
8. Calculate the mean of the results to obtain the average change in absorbance per minute (ΔA/min).

CALCULATIONS

U/L = ΔA/min x 3333 (37°C)
U/L = ΔA/min x 1746 (30°C)

ALP Procedure

MATERIALS REQUIRED

- Photometer or spectrophotometer with a thermostatted cell compartment set at 25/30/37°C, capable of reading at 405 nm.
- Stopwatch, strip-chart recorder or printer.
- Cuvettes with 1-cm pathlength.
- Pipettes to measure reagent and samples.

PROCEDURE

1. Preincubate working reagent, samples and controls to reaction temperature.
2. Set the photometer to 0 absorbance with distilled water.
3. Pipette into a cuvette:

Working reagent	1.0 mL
Sample or control	20 µL

4. Mix gently by inversion. Insert cuvette into the cell holder and start stopwatch.
5. Incubate for 1 minute and record initial absorbance reading.
6. Repeat the absorbance readings exactly after 1, 2 and 3 minutes.
7. Calculate the difference between absorbances.
8. Calculate the mean of the results to obtain the average change in absorbance per minute (ΔA/min).

CALCULATIONS

U/L = ΔA/min x 2764