MARROW ED8

Biochemistry

Comprehensive Question Bank

Contents

Chemistry of Carbohydrates, Amino sugars and Mucopolysaccharides

Question 1:

Which of the following is an aldose sugar?

a) Erythrulose

b) Ribulose

c) Fructose

d) Erythrose

Question 2:

Which of the following two monosaccharides are constituents of lactose?

a) Glucose and Fructose

b) Glucose and Mannose

c) Glucose and Galactose

d) Glucose and Glucose

Question 3:

Which of the following are enantiomers?

a) D-galactose and L-fructose

b) Galactose and glucose

c) D-mannose and L-mannose

d) D-glucose and D-mannose

Question 4:

Which among the following is an example of epimers?

a) D-glucose and L-glucose

b) D-glucose and D- mannose

c) D-glucose and D-fructose

d) D-fructose and D-talose

Question 5:

Which type of bond is there at the branching points in the structure of glycogen?

a) α1 ° 4 glucosidic bond

b) α1 ° 6 glucosidic bond

c) α1 ° 1 glucosidic bond

d) α1 ° 2 glucosidic bond

Question 6:

Which of the following has the highest glycemic index?

a) Glucose

b) Fructose

c) Sucrose

d) Sugar alcohols

Question 7:

You want to assess glucose in the urine of a patient with diabetes. Which of the following tests/methods cannot be used to detect the same?

a) Nelson Somogyi method

b) Ferric chloride test

c) O-Toluidine method

d) Folin and Wu method

Question 8:

Which of the following compounds in a patient's urine will not give a positive reaction to■ Benedict's test?

a) Ascorbic acid

b) Homogentisic acid

c) Sucrose

d) Lactose

Question 9:

What would be the respiratory quotient if someone is on an exclusive carbohydrate diet?

a) 0.7

b) 0.8

c) 1

d) 1.2

Question 10:

Which of the following tests can be used to estimate the liver function in a patient with chronic liver disease?

a) Glucose Tolerance Test

b) Fructose Tolerance Test

c) Galactose Tolerance Test

d) Lactose Tolerance Test

Question 11:

Which of the following is the most common mucopolysaccharidosis?

a) Hurler's disease

b) Hunter's disease

c) Sanfilippo disease

d) Sly syndrome

Question 12:

A patient with deafness and mental retardation is found to have an excess accumulation of glycosaminoglycans in his body. After a detailed evaluation, he is diagnosed with Hunter disease. What is the site of synthesis of these accumulated substances?

a) Mitochondria and Golgi apparatus

b) Endoplasmic reticulum and Golgi apparatus

c) Lysosomes and endoplasmic reticulum

d) Endoplasmic reticulum and mitochondria

Question 13:

What are the constituents of chondroitin sulfate?

a) N-acetyl glucosamine and glucuronic acid

b) N-acetyl galactosamine and glucuronic acid

c) N-acetyl glucosamine and galactose

d) N-sulfated glucosamine plus iduronic acid

Question 14:

Which of the following mucopolysaccharides does not contain uronic acid?

a) Chondroitin sulfate

b) Hyaluronic acid

c) Keratan sulfate

d) Dermatan sulfate

Question 15:

In a patient presenting with corneal opacity, deficiency of which of the following could be a likely cause?

a) Keratan sulfate

b) Hyaluronic acid

c) Chondroitin sulfate

d) Heparan sulfate

Question 16:

Which of the following glycosaminoglycan plays an important role in cellular migration?

a) Dermatan sulfate

b) Heparan sulfate

c) Chondroitin sulfate

d) Hyaluronic acid

Question 17:

Which of the following GAG is responsible for the charge selectiveness of the renal glomerular basement membrane?

a) Heparan sulfate

b) Heparin

c) Dermatan sulfate

d) Hyaluronic acid

Question 18:

All mucopolysaccharidoses are inherited in an autosomal recessive manner, except:

a) Hurler's disease

b) Hunter's disease

c) Maroteaux - lamy disease

d) Sanfilippo A disease

Question 19:

A 6-year-old mentally retarded girl presents with a protuberant abdomen, short stature, coarse facial features, and cloudy corneas. Skeletal malformations include dysostosis multiplex and bullet-shaped middle phalanx. Which of the following enzyme deficiencies is most likely in this patient?

a) Iduronate sulfatase

b) β-Galactosidase

c) α-L-Iduronidase

d) β-Glucuronidase

Question 20:

A 4-year-old boy presents with mental retardation, dysostosis multiplex, coarse facial features, and clear cornea. What is the most likely diagnosis?

a) Hurler's disease

b) Hunter's disease

c) Maroteaux-Lamy disease

d) Sly disease

Question 21:

A 7-year-old boy presented with dwarfism and skeletal abnormalities. Upon examination, the abdomen was distended with no hepatosplenomegaly. The child was assessed and was found to be of normal intelligence. Which of the following mucopolysaccharidoses could be a likely cause?

a) Hurler's disease

b) Hunter's disease

c) Sanfilippo disease

d) Morquio's disease

Question 22:

Naglazyme is used for treating patients with which of the following diseases?

a) Maroteaux-Lamy syndrome

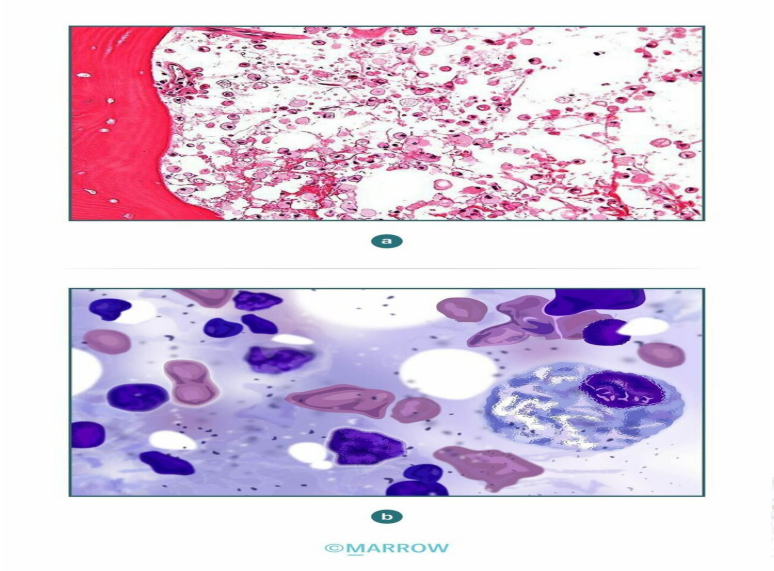
b) Morquio's disease

c) Von Gierkes disease

d) Niemann-Picks disease

Question 23:

A 5-year-old boy presented with easy fatigability, irritability and inability to concentrate. Labs revealed the following findings a and b (light and oil immersion respectively) on the bone marrow aspiration. Which of the following is the most likely enzyme deficient in this condition?



a) Hexosaminidase

b) Glucocerebrosidase

c) Sphingomyelinase

d) N-acetylglucosaminidase

Answer Key

Question No.	Correct Option
1	d
2	c
3	c
4	b
5	b
6	a
7	b
8	c
9	c
10	c
11	c
12	b
13	b
14	c

15	a
16	d
17	a
18	b
19	c
20	b
21	d
22	a
23	b

Detailed Explanations

Solution to Question 1:

Erythrose is not a ketose sugar but an aldose sugar as it has an aldehyde group in it.

Erythrulose, ribulose, and fructose are ketose sugars as they have a ketone group in them.

Solution to Question 2:

Lactose is a disaccharide that consists of galactose and glucose linked by a β-1,4-glycosidic linkage.

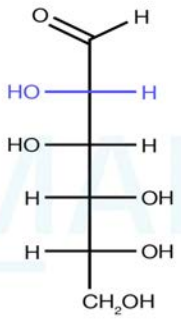
Solution to Question 3:

D-mannose and L-mannose are enantiomers. Enantiomers are mirror images of each other.

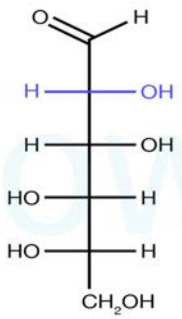
The designation of a sugar isomer as the D- or L- form is determined by its spatial relationship to the parent compound of the carbohydrates, that is, the three-carbon sugar glyceraldehyde. The orientation of the —H and —OH groups around the penultimate carbon atom determines whether the sugar belongs to the D- or L- series. When the —OH group on the penultimate carbon atom of the sugar is on the right side, then the sugar is designated as a D- isomer. When the –OH group is on the left, it is an L-isomer.

Naturally occurring monosaccharides are D-sugars and naturally occurring amino acids are L-isomers. Racemase enzyme interconverts between D and L isomers.

Examples of enantiomers include D-glucose and L-glucose, D-fructose and L-fructose and D-mannose and L-mannose



D-Mannose



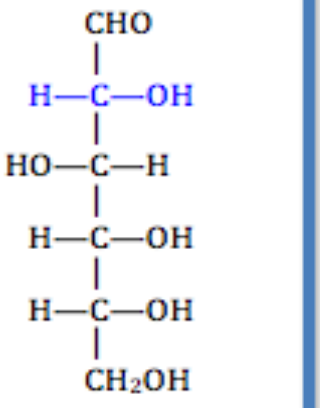
L-Mannose

Solution to Question 4:

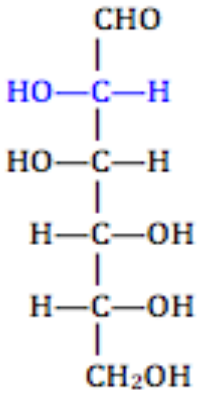
D-glucose and D-mannose are examples of epimers.

Isomers that differ in configuration around only one specific carbon atom (with the exception of the carbonyl carbon) are defined as epimers of each other.

Examples for epimers of glucose include mannose (epimerized at carbon 2) and galactose (epimerized at carbon 4)



D-Glucose



D-Mannose

Solution to Question 5:

Branching points in the glycogen structure have α1 ° 6 glucosidic bonds.

Glycogen is the storage polysaccharide in animals. It consists of α-D glucopyranose residues in α1 ° 4 glucosidic linkage in straight chains and branching by means of α1 ° 6 glucosidic bonds.

Solution to Question 6:

The glycemic index is highest for glucose.

The increase in blood glucose after a test dose of carbohydrate compared with that after an equivalent amount of glucose (as glucose or from a reference starchy food) is known as the glycemic index.

The glycemic Index of glucose and galactose is 1 while that of dietary fibres is 0.

Low glycemic index sugars include fructose, sucrose, starch and sugar alcohol.

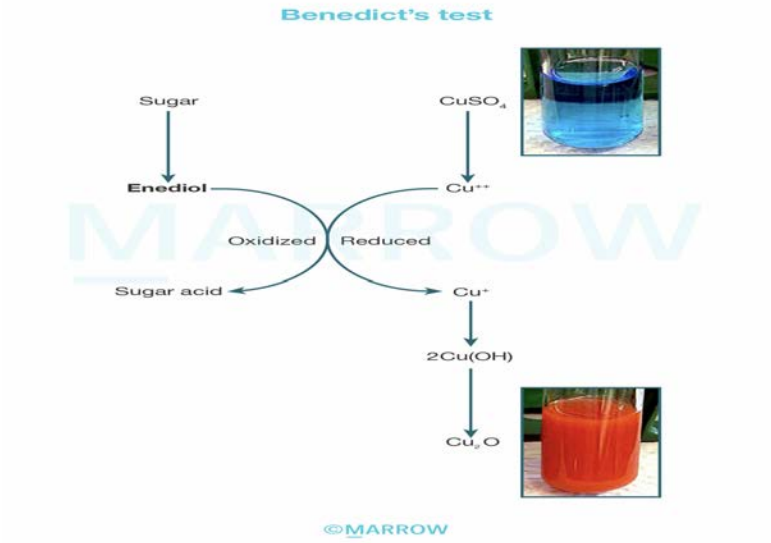
Solution to Question 7:

A Ferric chloride test is not used to detect glucose. It is used to detect alkaptonuria and phenylketonuria.

Solution to Question 8:

Benedict's test is a test for reducing sugars. Sucrose does not give a positive reaction to Benedict's test as it is a non-reducing sugar.

Principle: All monosaccharides and reducing disaccharides contain a free functional aldehyde or ketone group. When heated in the presence of an alkali, they are converted to enediols, which reduce the cupric compounds (Cu2+) present in Benedict's reagent to an insoluble precipitate of red cuprous oxide (Cu2O).



Composition of Benedict's reagent:

• Copper sulfate: Provides the source of cupric (Cu^{2+}) ions

• Sodium citrate: Complexes with cupric (Cu^{2+}) ions to prevent its deterioration to cuprous ions spontaneously

• Sodium carbonate: Provides an alkaline pH

Substances giving a positive reaction on Benedict's test:

• All monosaccharides such as glucose, galactose, fructose, pentoses

• Reducing disaccharides such as lactose and maltose

• Glucuronic acid, homogentisic acid, and ascorbic acid

Clinical correlate: Benedict's test is used to detect the presence of glucose in urine.

The color of the precipitate gives a rough idea of the amount of sugar present in urine. Hence, it is a semi-quantitative test.

Colour of the precipitate	Amount of reducing sugar
Green	0.5%
Yellow	1%
Orange	1.5%
Red	2%

Solution to Question 9:

The respiratory quotient, if someone is on an exclusive carbohydrate diet, is 1.

The respiratory quotient is the amount of CO_2 produced /O_2 consumed.

The respiratory quotient for various substances:

Substance	RQ
Carbohydrate	1.00
Protein	0.81
Fat	0.71
Alcohol	0.66

Solution to Question 10:

The galactose tolerance test is used to estimate liver function.

Galactose is metabolized to glucose almost exclusively by the liver. Hence, the galactose tolerance test is done to assess the functional capacity of the liver.

In liver dysfunction, galactose is excreted in the urine in increased amounts. The liver function is measured as the rate of excretion of galactose in urine following the
ingestion or intravenous injection of a known amount of galactose.
Normally, less than 3 g appear in the urine within 5 hours after the ingestion of 40 g of galactose.

Solution to Question 11:

The most common mucopolysaccharidosis (MPS) is Sanfilippo disease (MPS III), followed by Hurler's disease and Hunter's disease.

Sanfilippo disease makes up a genetically heterogeneous but clinically similar group of 4 recognized types. It is due to the deficiency of enzymes involved in the degradation of heparan sulfate. The onset of symptoms occurs between 2 and 6 years of age.

It is characterized by severe CNS involvement with a mild somatic involvement. Disproportionate involvement of CNS is a unique feature of MPS III. Other features include sleep disturbances, aggressive behavior problems, and hirsutism.

Solution to Question 12:

The sites of synthesis of glycosaminoglycans (GAG) are the endoplasmic reticulum and Golgi apparatus.

Synthesis of the core proteins occurs in the endoplasmic reticulum, whereas, biosynthesis of glycosaminoglycan chains and their subsequent modifications occurs in the Golgi apparatus

Glycosaminoglycans or mucopolysaccharides are complex carbohydrates made up of amino sugars and uronic acids. They may be attached to a protein molecule to form a proteoglycan.
Proteoglycans are components of the ground substance of connective tissue. Proteoglycans have a bottle brush appearance.

Note: Hyaluronic acid is the only GAG that is not synthesized by the Golgi apparatus. It is formed at the plasma membrane. It participates in cellular migration among other things.

Solution to Question 13:

Chondroitin sulfate is composed of N-acetyl galactosamine with sulfate and glucuronic acid.

Chondroitin sulfate is the most abundant GAG in the body. It is located at sites of calcification in endochondral bone, cartilage, tendons, ligaments, aorta, and CNS. In cartilages, it binds collagen and holds fibers in a tight, strong network.

Solution to Question 14:

Keratan sulfate is a mucopolysaccharide that does not contain uronic acid. It is constituted by repeating disaccharide units of N-acetyl glucosamine and galactose.

Two types of keratan sulfate are seen:

• Keratan sulfate I - in the cornea

• Keratan sulfate II - in the cartilage

It is present between the collagen fibrils in the cornea and plays a major role in corneal transparency. Hence, its deficiency can lead to corneal opacity.

Solution to Question 15:

Keratan sulfate I is present between the collagen fibrils in the cornea and plays a major role in corneal transparency. Hence, its deficiency can lead to corneal opacity.

Keratan sulfate is made of repeating units of N-acetyl glucosamine and galactose. Two types of keratan sulfate are seen:

• Keratan sulfate I - in the cornea

• Keratan sulfate II - in the cartilage

Dermatan sulfate (DS) also plays a role in corneal transparency. However, DS is widely distributed in the body, mainly in the skin.

Solution to Question 16:

Hyaluronic acid plays an important role in cellular migration.

It is composed of repeating disaccharide units of N-acetyl glucosamine and glucuronic acid. It has no sulfate group. Hyaluronic acid is the only GAG that is not covalently attached to core protein. It is located in Synovial fluid, vitreous humor, the umbilical cord, cartilage.

It serves as a lubricant and shock absorber, especially in joints. It also plays an important role in permitting cell migration during morphogenesis, wound repair, and tumor cell metastasis.

Solution to Question 17:

Heparan sulfate is the GAG involved in the charge selectiveness of the renal glomerular basement membrane.

Heparan sulfate is a GAG made up of repeating units of glucosamine and glucuronic acid.

Functions of heparan sulfate:

• Mediation of cell growth and cell-cell communication by acting as receptors on the cell membrane

• Charge selectiveness of glomerular filtration: Since heparan sulfate contains a lot of negative charges, its ability to repel proteins is important in glomerular filtration

• As an anchor for lipoprotein lipase

Note: The major GAG synthesized by arterial smooth muscle cells is dermatan sulfate, which binds LDL and plays a role in the formation of atherosclerotic plaque.

Solution to Question 18:

All mucopolysaccharidoses are inherited in an autosomal recessive manner, except Hunter's disease which is inherited in an X-linked recessive manner.

In Hunter's disease, the enzyme iduronate 2-sulphatase is deficient. It is an X-linked condition, affecting males only.

Clinical features of the condition include:

• Grouped skin papules

• Extensive Mongolian spots may be an early marker

• No corneal clouding

• Communicating hydrocephalus and spastic paraplegia may be seen due to the thickening of meninges

• Severe form: extensive and progressive CNS deterioration, followed by death by 10–15 years of age

Solution to Question 19:

The clinical features point to the diagnosis of Hurler's disease (MPS 1). Deficiency of the enzyme α-L-iduronidase that degrades glycosaminoglycans results in Hurler's disease.

Hurler's disease was also known previously as gargoylism (coarse facial features).

Clinical features of the disease include coarse facial features, corneal clouding, large tongue, and inguinal hernias. They also present with hepatosplenomegaly, short stature, skeletal dysplasia, and can present with hydrocephalus or valvular heart disease. These patients also have Reilly body inclusions in their leukocytes.

The gene encoding α-L-iduronidase is present on chromosome 4. Heparan sulfate and dermatan sulfate are accumulated. Enzyme replacement therapy is available in the form of aldurazyme.

Mucopolysaccharidoses are a group of metabolic disorders caused by the absence or malfunctioning of lysosomal enzymes needed to break down molecules called glycosaminoglycans.

• Most common MPS: Sanfilippo > Hunter's and Hurler's

• MPS with no mental retardation: Scheie's disease, Morquio syndrome, and Maroteaux–Lamy syndrome.

• MPS with no corneal clouding: Hunter's disease and Sanfilippo syndrome

• MPS with XLR inheritance: Hunter's disease

• MPS with no visceromegaly: Morquio's disease

• Skeletal deformity associated with all MPS: Dysostosis multiplex

• MPS with no leucocyte inclusions: Morquio's disease

Solution to Question 20:

The clinical features point to a diagnosis of Hunter's disease. Corneal clouding is absent in Hunter's disease.

In this condition, the enzyme iduronate 2-sulphatase is deficient. It is an X-linked condition, affecting males only.

Clinical features of the condition include:

• Grouped skin papules

• Extensive Mongolian spots may be an early marker

• No corneal clouding

• Communicating hydrocephalus and spastic paraplegia may be seen due to the thickening of meninges

• Severe form: extensive and progressive CNS deterioration, followed by death by 10–15 years of age

Solution to Question 21:

The key features in the above scenario are the absence of hepatosplenomegaly and the preservation of intelligence. Both of these features make the diagnosis of Morquio's disease most likely. All the other conditions above are associated with hepatosplenomegaly.

Morquio disease (MPS-IV) is caused by a deficiency of N-acetylgalactosamine-6-sulfatase (MPS-IVA) or of β-galactosidase (MPS IVB). Both lead to defective degradation of keratan sulfate.

Early symptoms:

• Genu valgus

• Kyphosis

• Growth retardation with short trunk and neck

• Waddling gait

• Preservation of intelligence

• Instability of the odontoid process and ligamentous laxity is regularly present and can result in life-threatening atlantoaxial instability and dislocation.

It is of two types:

Note:

• MPS with no mental retardation: Scheie's disease, Morquio syndrome, and Maroteaux–Lamy syndrome.

• MPS with no visceromegaly: Morquio's disease

Type	Enzyme involved	Chromosome
Morquio syndrome A	Galactosamine 6-sulfatase	Chromosome 16
Morquio syndrome B	β-Galactosidase	Chromosome 4

Solution to Question 22:

Naglazyme is used in the treatment of Maroteaux–Lamy syndrome.

Maroteaux–Lamy syndrome/MPS VI is caused by the deficiency of arylsulfatase B enzyme whose gene locus is on chromosome 5. Preservation of intelligence is the feature of this syndrome. It is characterized by macrocephaly, hydrocephalus, macroglossia, short stature, and contractures.

Naglazyme is used as an enzyme replacement therapy.

Solution to Question 23:

The given clinical scenario of a child with easy fatiguability, irritability, and inability to concentrate with the images showing a crumpled tissue paper appearance(Gaucher's cell) is suggestive of Gaucher's disease. The deficient enzyme is glucocerebrosidase (also known as beta-glucosidase). I

Gaucher's disease is an inborn error of metabolism caused due to a deficiency of beta glucocerebrosidase. It is the most common lysosomal storage disorder and the mode of inheritance is autosomal recessive. There is an accumulation of lipid-laden macrophages in the spleen, liver, and other tissues.

There are three clinical subtypes - type I (chronic non-neuronopathic), type II (acute neuronopathic), and type III (intermediate). Clinical features include anemia, thrombocytopenia, hepatosplenomegaly, and neurological features depending on the subtype.

Gaucher's cells - These are formed as a result of the accumulation of massive amounts of glucocerebrosides within the phagocytes. The cells have a fibrillary cytoplasm and give a crumpled tissue paper appearance.

Option A: Deficiency of beta-hexosaminidase A is seen in Tay-Sachs disease. The deficiency of beta-hexosaminidase A and B causes Sandhoff's disease. Clinical features are seizures, macrocephaly, and hyperacusis.

Option C: Deficiency of sphingomyelinase leads to Niemann-Pick disease. It is characterized by the presence of foam cells in the bone marrow. There may be pulmonary infiltrates leading to lung failure.

Option D: Deficiency of N-acetylglucosaminidase leads to Sanfillipo's disease. It is a type of mucopolysaccharidosis (MPS - III). There is an accumulation of heparan sulfate leading to severe cognitive degeneration.

Glycolysis and gluconeogenesis

Question 1:

Which of the following glucose transporter in the myocytes is under the influence of insulin?

a) GLUT 1

b) GLUT 2

c) GLUT 3

d) GLUT 4

Question 2:

Which of the following glucose transporter is specific to the transport of fructose from the intestine to enterocytes?

a) GLUT 1

b) GLUT 3

c) GLUT 5

d) GLUT 7

Question 3:

Insulin was administered in a patient with severe hyperglycemia. This drug will stimulate glucose uptake in all the following except:

a) Heart

b) Skeletal muscle

c) Liver

d) Adipose tissue

Question 4:

Which glucose transporter is present in erythrocytes?

a) GLUT 1

b) GLUT 2

c) GLUT 3

d) GLUT 4

Question 5:

In a patient after overnight fasting, which of the following will have reduced levels of glucose transporters?

a) Brain cells

b) Adipocytes

c) Hepatocytes

d) RBCs

Question 6:

Which of the following is false about glycolysis?

a) Occurs in the cytoplasm

b) Conversion of glucose to 3-carbon units

c) Complete breakdown of glucose

d) 2 ATPs are utilized in the pathway

Question 7:

The step involving which of the following enzyme is reversible in glycolysis?

a) Enolase

b) Phosphofructokinase

c) Pyruvate kinase

d) Hexokinase

Question 8:

Which of the following substances inhibits the rate-limiting enzyme in glycolysis?

a) AMP

b) Glucose-6-PO4

c) Citrate

d) Insulin

Question 9:

All of the following statements about glucokinase are true in comparison to other hexokinases, except:

a) Higher Km value

b) Phosphorylates multiple hexose substrates

c) An inducible enzyme

d) Higher Vmax value

Question 10:

Which of the following is the first committed step of glycolysis?

a) Glucose to glucose 6 PO4

b) Fructose 6 phosphate to Fructose 1, 6 bisphosphate

c) 1,3 bisphosphoglycerate to 3-phosphoglycerate

d) Phosphoenolpyruvate to pyruvate

Question 11:

Which of the following is a false statement regarding anaerobic glycolysis?

a) End product is lactic acid

b) Net production of ATP is 2

c) Pyruvate dehydrogenase enzyme is needed

d) Occurs in places like the eye lens and RBCs

Question 12:

Which of the following glycolytic enzymes catalyze the step where an inorganic phosphate is utilized?

a) Phosphoglyceromutase

b) Phosphohexose isomerase

c) Glyceraldehyde-3-phosphate dehydrogenase

d) Phosphofructokinase

Question 13:

Which of the following enzymes takes part in substrate-level phosphorylation in glycolysis?

a) Pyruvate kinase

b) PFK-1

c) Hexokinase

d) Pyruvate dehydrogenase

Question 14:

How many net ATPs are generated in aerobic glycolysis?

a) 2

b) 7

c) 10

d) 9

Question 15:

You are collecting a blood sample from a patient for blood glucose estimation. You put the sample in a vacutainer containing sodium fluoride. Which of the following enzyme is inhibited by this compound?

a) Glyceraldehyde 3 PO4 dehydrogenase

b) Enolase

c) Pyruvate kinase

d) 1, 3 bisphosphoglycerate kinase

Question 16:

What is fructose 2,6 bisphosphate?

a) Intermediate metabolite of glycolysis

b) Positive allosteric regulator of PFK1

c) Negative allosteric regulator of PFK2

d) Positive allosteric regulator of PFK2

Question 17:

A patient with anemia and hepatosplenomegaly is diagnosed with hemolytic anemia. Deficiency of which of the following glycolytic enzymes is most likely associated with this presentation?

a) Enolase

b) Pyruvate kinase

c) Aconitase

d) Phosphofructokinase-1

Question 18:

Where does the Rapaport Leubering cycle occur?

a) Liver

b) RBCs

c) Kidney

d) Brain

Question 19:

Which of the following pathways act as a major energy source for cancer cells?

a) Glycolysis

b) Oxidative phosphorylation

c) Krebs cycle

d) β-oxidation of fatty acids

Question 20:

In which of the following tissues does gluconeogenesis take place?

a) Mature RBC's

b) Kidney

c) Adipocytes

d) Myocyte

Question 21:

Which of the following is not a substrate for gluconeogenesis?

a) Glucogenic amino acids

b) Propionate

c) Glycerol

d) Acetyl CoA

Question 22:

What is the process of reconversion of lactate formed in the muscles and RBC's, back into glucose inside the liver called?

a) Cahill cycle

b) Cori cycle

c) Kreb's cycle

d) Dicken-Horecker cycle

Question 23:

Which of the following organ pairs is involved in the Cahill cycle?

a) Liver and Muscle

b) Liver and Kidney

c) Muscle and Kidney

d) Brain and Liver

Question 24:

Malate shuttle is important in which of the following processes?

a) Glycolysis only

b) Gluconeogenesis and glycolysis

c) Glycogenolysis

d) Glycogen synthesis

Question 25:

Which of the following is a repressor of the enzyme pyruvate carboxylase?

a) Cortisol

b) Glucagon

c) Insulin

d) Adrenaline

Question 26:

All of the following enzymes are involved in gluconeogenesis except:

a) Phosphoglucomutase

b) Fructose 1,6 bisphosphatase

c) Pyruvate carboxylase

d) Phosphoglycerate kinase

Question 27:

Which of the following is a step in the gluconeogenic pathway?

a) Pyruvate to Lactate

b) Glucose-6-PO4 to Fructose-6-PO4

c) Pyruvate to Acetyl CoA

d) Oxaloacetate to Phosphoenolpyruvate

Question 28:

Which of the following is not seen in low insulin-glucagon ratio?

a) Glycogen breakdown

b) Ketogenesis

c) Glycogen storage

d) Gluconeogenesis

Question 29:

Which of the following enzyme activity decreases in fasting?

a) Hormone-sensitive lipase

b) Glycogen phosphorylase

c) Pyruvate carboxylase

d) Phosphofructokinase 1

Question 30:

Enzyme activated by a decrease in insulin: glucagon ratio is:

a) Glucokinase

b) Hexokinase

c) Phosphofructokinase

d) Glucose 6-phosphatase

Answer Key

Question No.	Correct Option
1	d
2	c
3	c
4	a
5	b
6	c
7	a
8	c
9	b
10	b

11	c
12	c
13	a
14	b
15	b
16	b
17	b
18	b
19	a
20	b
21	d
22	b
23	a
24	b
25	c
26	a
27	d
28	c
29	d
30	d

Detailed Explanations

Solution to Question 1:

GLUT 4 is the glucose transporter in the myocytes under the influence of insulin.

Glucose transport in our body mainly occurs through sodium-independent glucose transporters, which include GLUT transporters and to a small extent through sodium-dependent glucose transporters, which include SGLT-1 and SGLT-2. Insulin induces the movement of intracellular GLUT 4 molecules to the cell surface and thus increases glucose uptake. In type 2 diabetes mellitus, membrane GLUT 4 is reduced, leading to insulin resistance in muscle and fat cells.

Solution to Question 2:

Glucose transporter specific to the transport of fructose from the intestine to enterocytes is GLUT 5. It is present in small intestine, testis, and sperm, and it primarily transports fructose.

Solution to Question 3:

In the liver, the transport of glucose is through GLUT 2, which is insulin-independent.

Insulin-stimulated glucose uptake takes place in the heart, skeletal muscle, and adipose tissue through GLUT 4.

Solution to Question 4:

GLUT 1 is the glucose transporter present in erythrocytes.

It mediates basal glucose uptake. GLUT 1 is also the major glucose transporter in the brain and placenta and is widely distributed.

Solution to Question 5:

Adipose tissue will have reduced levels of glucose transporters after overnight fasting.

The passive uptake of glucose by muscle and adipose tissue is catalyzed by the GLUT-4 transporter. Levels of insulin-sensitive receptor GLUT-4 decrease after overnight fasting.

In the absence of insulin, most GLUT 4 molecules are sequestered in membrane vesicles within the cell, but when blood glucose rises, the release of insulin triggers GLUT 4 movement to the plasma membrane.

Solution to Question 6:

The complete breakdown of glucose involves the Krebs cycle (TCA cycle), which occurs in the mitochondria, and not glycolysis.

Glycolysis takes place in the cytoplasm of all cells. 6 carbon-containing glucose is converted to 3 carbon-containing pyruvate in aerobic and lactate in an anaerobic environment. RBC lacks mitochondria and hence completely relies on glucose for energy needs.

Phosphofructokinase-1 (PFK-1) is the rate-limiting enzyme in glycolysis and is involved in the conversion of fructose-6-PO_4 to fructose 1,6 bisphosphate. This is the slowest reaction in glycolysis and is, therefore, the rate-limiting step. Hexokinase, PFK-1, and pyruvate kinase are involved in 3 irreversible steps of glycolysis.

Net ATP from aerobic glycolysis is 7 ATP. Net ATP from anaerobic glycolysis is 2 ATP. 2 ATP molecules are utilized in glycolysis. Steps releasing ATP in glycolysis are as follows:

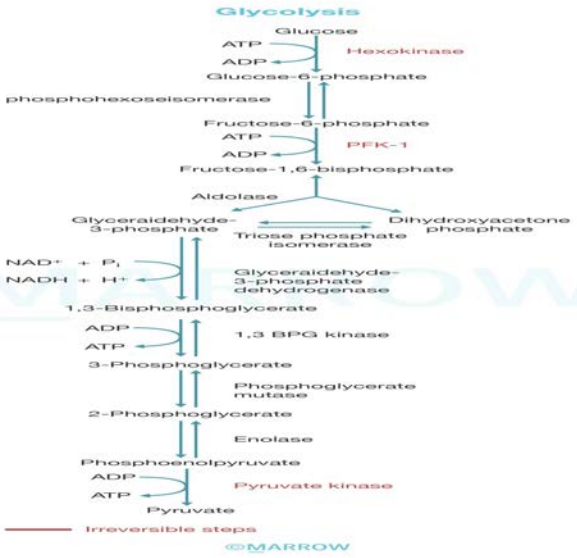
- 1,3 bisphosphoglycerate to 3 phosphoglycerate
- Phosphoenolpyruvate to pyruvate

Inhibitors of glycolytic enzymes:

- Iodoacetate inhibits glyceraldehyde 3 PO_4 dehydrogenase

• Fluoride inhibits enolase

• Arsenate inhibits 1, 3 bisphosphoglycerate kinase step. Arsenic causes glycolysis to produce zero net ATP.



Solution to Question 7:

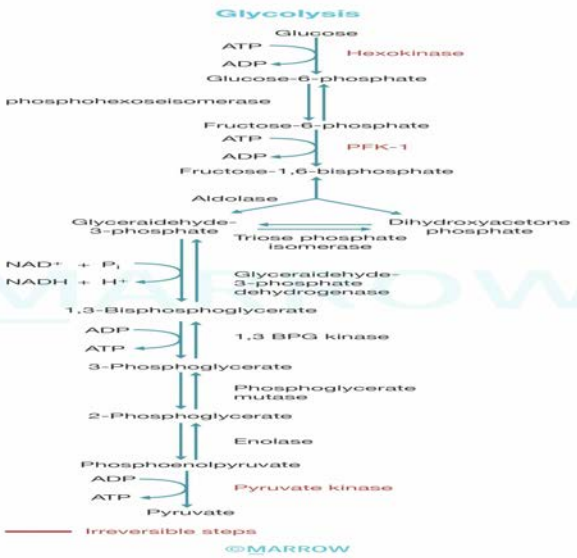
Enolase catalyzes the conversion of 2-phosphoglycerate to phosphoenolpyruvate, which is a reversible step of glycolysis.

The irreversible steps of glycolysis are catalyzed by the following enzymes:

• Hexokinase

• Phosphofructokinase

• Pyruvate kinase



Solution to Question 8:

Phosphofructokinase (PFK-1) is the rate-limiting enzyme in glycolysis. Citrate inhibits the PFK-1.

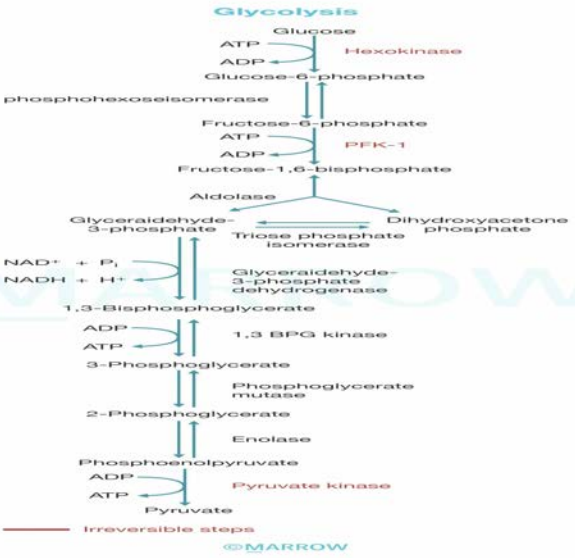
Mg2+ serves as the co-factor for the enzyme. This step is the committed step of glycolysis.

In general, end products of a reaction on accumulation can inhibit the forward direction progress of any reaction. Substrates and cofactors of an enzyme/reaction will stimulate a reaction/to activate the enzyme, and hence push the reaction in the forward direction.

Inhibitors of PFK-1: ATP, Citrate

Allosteric activators of PFK-1: AMP, ADP, Fructose 2,6 bisphosphate (Most potent activator)

Hormonal regulation: Inducer — Insulin, Repressor— Glucagon



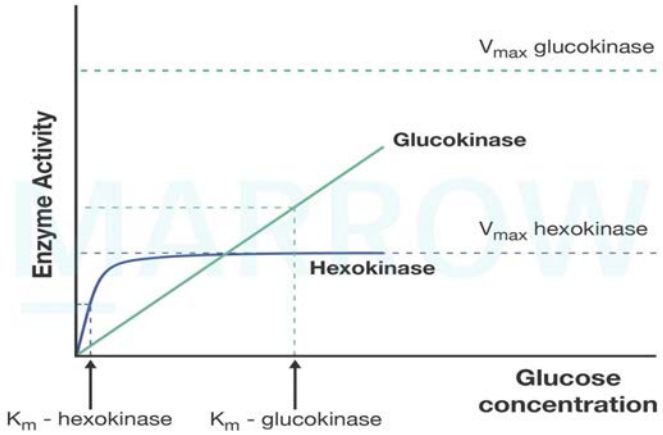
Solution to Question 9:

Glucose is the only cellular substrate that can be phosphorylated by glucokinase.

Glucokinase has a higher Vmax and a higher Km value than hexokinase. This means it requires a higher concentration of glucose for half saturation. Hence, glucokinase functions only when the intracellular concentration of glucose is elevated, as seen immediately after a carbohydrate-rich meal.

Characteristics	Hexokinase	Glucokinase
Tissue distribution	Most tissues	Liver and beta cells of the pa ncreas
Km	Low	High

Characteristics	Hexokinase	Glucokinase
Vmax	Low	High
Inhibition by G6P	Yes	No
Inducible	No	Inducible
Clinical significance	Deficiency causes hemolytic a nemia	Less activity in diabetes melli tus
Biological significance	Involved in maintaining intra cellular glucose concentratio n	Involved in maintaining bloo d glucose concentration

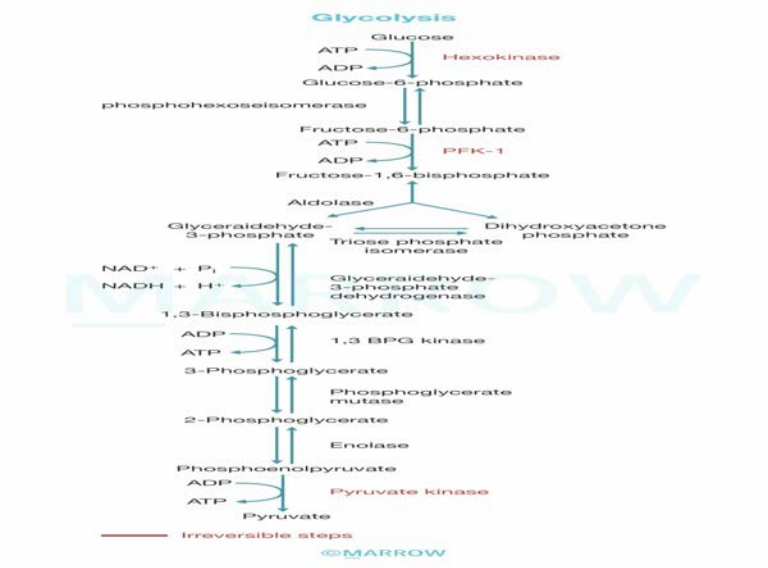


Note: Glucokinase acts on other hexoses such as fructose, mannose too. However, in vivo, the Km for this reaction is so high that at the intracellular concentrations available, the activity is not significant. Hence, the best option has been chosen as the answer in this MCQ.

Solution to Question 10:

Fructose 6 phosphate to fructose 1,6 bisphosphate catalyzed by PFK-1 is the first commited step of glycolysis. As fructose 1,6 bisphosphate once formed, it cannot revert back and go into any pathway other than glycolysis.

Note: Glucose to glucose-6-PO4 is also an irreversible step catalyzed by hexokinase, but as this glucose-6-PO4 can still enter the HMP shunt, it is not a step committed to glycolysis.



Solution to Question 11:

Lactate dehydrogenase enzyme is needed in anaerobic glycolysis, not pyruvate dehydrogenase.

Under anaerobic conditions, pyruvate is reduced to lactate. This reaction is catalyzed by lactate dehydrogenase.

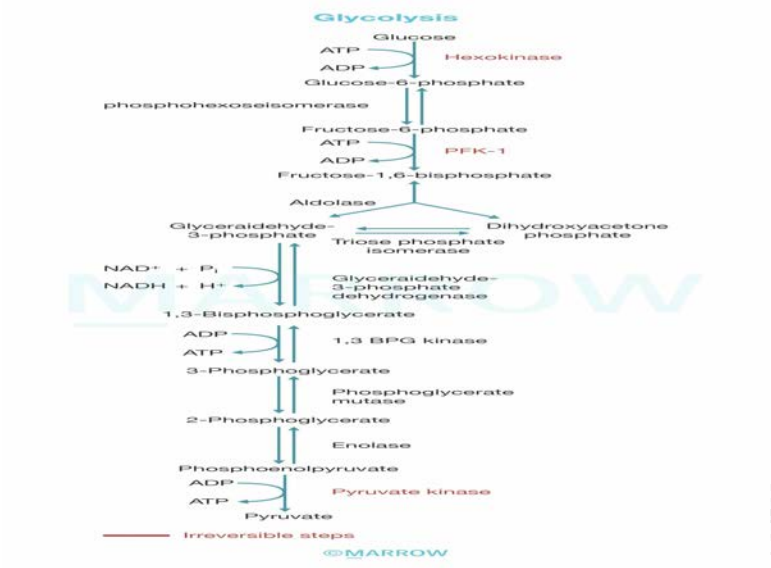
Since RBC's lack mitochondria, the further reactions of pyruvate in the TCA cycle cannot take place. Hence, glycolysis in erythrocytes always ends in the formation of lactate. Also, anaerobic glycolysis is the major fate for pyruvate in the lens and cornea of the eye, kidney medulla, leukocytes, because these are all poorly vascularized structures.

Net ATP produced per molecule of glucose under anaerobic conditions is 2. The NADH does not contribute to the ATP production because it gets used up in the reduction of pyruvate to lactate, which need NADH.

Solution to Question 12:

Conversion of glyceraldehyde-3-phosphate to 1,3 biphosphoglycerate catalyzed by glyceraldehyde 3-phosphate dehydrogenase involves the addition of inorganic phosphate.

Note: The step catalyzed by phosphofructokinase utilizes ATP as a phosphate donor and not inorganic phosphate.



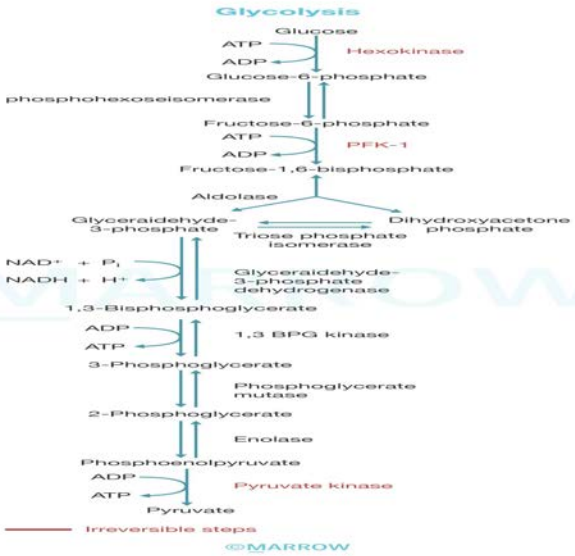
Solution to Question 13:

The enzyme involved in substrate-level phosphorylation is pyruvate kinase.

In substrate-level phosphorylation, there is a direct generation of ATP. In glycolysis, there are two such reactions, which are examples of substrate-level phosphorylation. They are as follows:

• 1,3 bisphosphoglycerate to 3-phosphoglycerate, catalyzed by the enzyme phosphoglycerate kinase

• Phosphoenol pyruvate to pyruvate, catalyzed by pyruvate kinase



Solution to Question 14:

Net generation of ATP up to pyruvate in aerobic glycolysis is 7.

Steps of ATP production in glycolysis:

But, remember that 2 molecules of ATP get utilized in the previous steps of glycolysis, catalyzed by the enzymes hexokinase and PFK-1.

Hence, net ATP generation in aerobic glycolysis is 9-2 = 7 ATP.

Reaction	Enzyme	Method of ATP produced	Net ATP produced
Glyceraldehyde-3 phosphate to 1,3 bisphosphoglycerate	Glyceraldehyde 3 phosphate dehydrogenase	From 2 molecules of NADH via respiratory chain oxidation	5 ATP (2*2.5 per NADH)
1,3 bisphosphoglycerate to 3 phosphoglycerate	Phosphoglycerate kinase	Substrate-level phosphorylation	2 ATP
Phosphoenolpyruvate to pyruvate	Pyruvate kinase	substrate-level phosphorylation	2 ATP
			Total = 9 ATP

Solution to Question 15:

Sodium fluoride and potassium oxalate are added to the blood sample to estimate glucose. Fluoride is an inhibitor of enolase and hence halts the process of glycolysis. By doing so, it prevents blood glucose levels from being falsely estimated as low.

Inhibitors of glycolytic enzymes:

- Fluoride inhibits enolase
- Iodoacetate inhibits glyceraldehyde 3 PO_4 dehydrogenase
- Arsenate inhibits phosphoglycerate kinase

Solution to Question 16:

Fructose 2,6-bisphosphate is the most potent positive allosteric activator of phosphofructokinase-1 and inhibitor of fructose 1, 6-bisphosphatase. It is not an intermediate of glycolysis.

Fructose 2, 6-bisphosphate is formed by phosphorylation of fructose-6-phosphate by phosphofructokinase-2. It acts as an intracellular signal, indicating that glucose is abundant and hence causes an increase in the rate of glycolysis during the well-fed state.

Solution to Question 17:

Deficiency of pyruvate kinase leads to hemolytic anemia.

Erythrocytes lack mitochondria and hence generate ATP exclusively via glycolysis. In a glycolytic enzyme deficiency, there is a reduced rate of glycolysis, leading to decreased ATP production. This results in a few alterations in the RBC membrane, thereby leading to changes in cell shape. Such deformed RBCs are later phagocytosed by cells of the reticuloendothelial system, and this results in a state of hemolytic anemia.

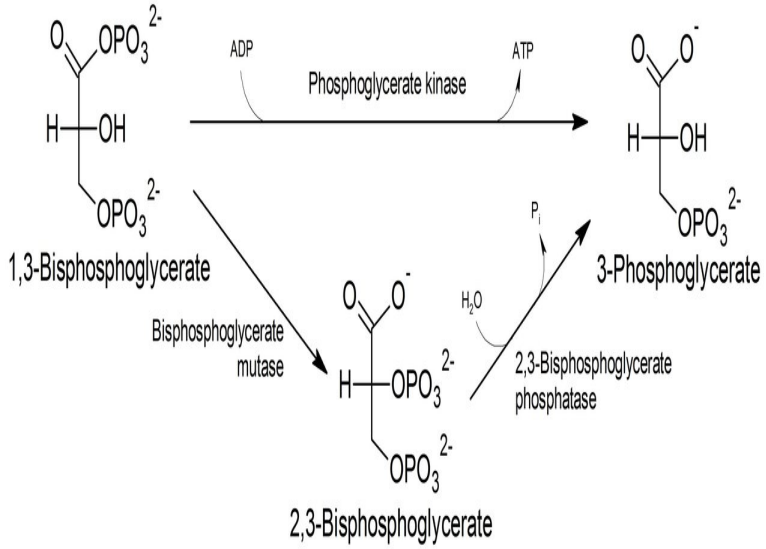
The most common enzyme deficiency causing hemolytic anemia is glucose-6-phosphate dehydrogenase deficiency (pentose phosphate pathway).

The second most common enzyme deficiency causing hemolytic anemia is the pyruvate kinase.

Note: Though hemolysis is seen in PFK-1 deficiency, it does not always manifest as anemia.

Solution to Question 18:

Rapaport Leubering cycle (2,3 BPG shunt) occurs in RBCs.



Rapaport Leubering cycle:

• 1, 3-Bisphosphoglycerate is converted to 2, 3-bisphosphoglycerate by the action of the enzyme bisphosphoglycerate mutase. 2, 3-BPG is present at high concentration in RBCs and serves to increase O2 delivery. 2, 3-BPG decreases the O2 affinity of hemoglobin by binding to deoxyhemoglobin and shifts the O2-Hb dissociation curve to the right. Hence, there is more oxygen release in conditions like hypoxia.

• 2,3-bisphosphoglycerate is hydrolyzed to 3-phosphoglycerate and Pi by 2,3-bisphosphoglycerate phosphatase and no ATP is generated.

Since the reaction catalysed by 1, 3- Bisphosphoglycerate kinase is bypassed, no ATPs are produced in that step. Only 2 ATPs are generated by the pyruvate kinase step and 2 ATPs are utilized in the steps catalyzed by hexokinase and PFK-1. Anaerobic glycolysis takes place due to the absence of mitochondria. So this pathway yields no net generation of ATP from glycolysis.

Solution to Question 19:

Cancer cells derive energy mainly from glycolysis.

Cancer cells utilize large amounts of glucose and metabolize it to lactic acid, even in the presence of oxygen. This effect was termed the Warburg effect.

The increased rate of glycolysis is due to certain defects in the mitochondrial electron transport chain. Besides, this enhanced glycolysis enables the cancer cells to proliferate in the reduced oxygen tension conditions, which is seen in tumors. This switch from aerobic to anaerobic glucose metabolism also acts as an impetus to tumorigenesis.

The increased glucose consumption is used as a carbon source for anabolic processes needed to support cell proliferation such as the de novo generation of nucleotides, lipids, and proteins. Also, the major isoenzyme of pyruvate kinase in cancer cells is PK-2 (in contrast to PK-1 in normal cells), which appears to play a role in the Warburg effect.

Note: This effect is the basis for detecting cancers by FDG-PET scanning.

Solution to Question 20:

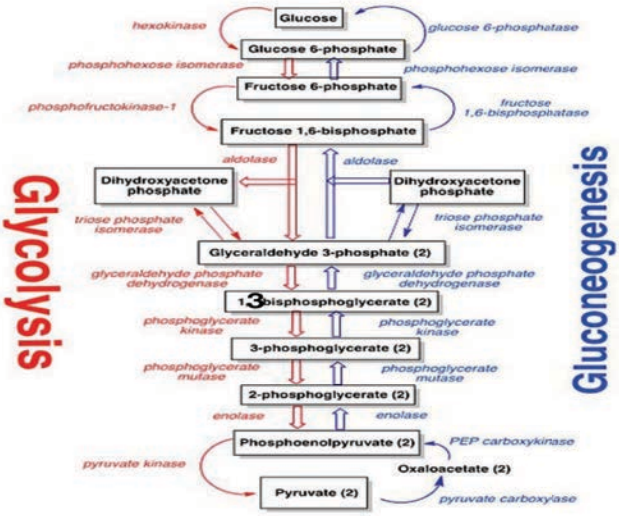
The liver and kidney are the two major locations where gluconeogenesis takes place.

During short periods of fasting, most gluconeogenesis takes place in the liver, while the remaining takes place in the kidneys. However, during periods of prolonged fasting, the kidneys become a major site of gluconeogenesis.

Gluconeogenesis is the process of synthesizing glucose or glycogen from non-carbohydrate precursors. It occurs in cytosol and mitochondria of the liver and kidney. Significant substrates are glucogenic amino acids, lactate, glycerol, and propionate.

Lactate generated in various tissues is transported to the liver and is converted back to pyruvate and enters gluconeogenesis. This pathway is referred to as Cori's cycle. The Glucose-alanine cycle is referred to as the Cahill cycle.

Muscle cannot participate in gluconeogenesis since it lacks glucose-6-phosphatase.



Solution to Question 21:

Acetyl CoA is not a substrate for gluconeogenesis. Instead, it enters the Krebs cycle for oxidative phosphorylation.

The following serve as substrates for gluconeogenesis:

• Glucogenic amino acids, alanine being the major contributor (alanine is the source of pyruvate). Glucogenic amino acids are converted into pyruvate, oxaloacetate, succinyl CoA, and alpha-ketoglutarate to enter gluconeogenesis and the Krebs cycle at various points.

• Lactate

• Pyruvate

• Glycerol

• Propionate (ruminants)

Solution to Question 22:

The process of reconversion of lactate formed in the muscles and RBCs, back into glucose inside the liver is called Cori's cycle.

Lactic acid is a metabolic byproduct formed after glycolysis in tissues such as the skeletal muscles and RBCs. The lactate so formed is transported into the liver and kidney. Here, it gets reconverted back into glucose. This entire process is called Cori's cycle or the glucose–lactate cycle.

Cahill cycle is also called as glucose–alanine cycle. In the fasting state, there is a significant output of alanine from skeletal muscle obtained from glycolysis of muscle glycogen. Alanine is derived from the transamination of pyruvate obtained from glycolysis. Alanine is then exported to the liver where it undergoes transamination again back to pyruvate and enters gluconeogenesis.

Note: Pentose phosphate pathway is otherwise known as Warburg-Dickens-Horecker Cycle.

Solution to Question 23:

The liver and muscle are involved in the Cahill cycle.

During periods of fasting, there is the production of alanine in the skeletal muscles, which is formed by transamination reactions from pyruvate. This alanine is shunted out into the circulation, and later on into the liver. Here in the liver, another round of transamination takes place, and the alanine is reconverted back into pyruvate. This pyruvate serves as a substrate for gluconeogenesis. This entire pathway has been called as the Cahill's cycle or the glucose–alanine pathway.

Solution to Question 24:

Malate shuttle is important in both, gluconeogenesis and glycolysis.

Malate shuttle is a biochemical system for translocating impermeable electrons (NADH) or substrates (oxaloacetate), in the form of a permeable substance, malate. The semipermeable inner mitochondrial membrane is permeable to malate.

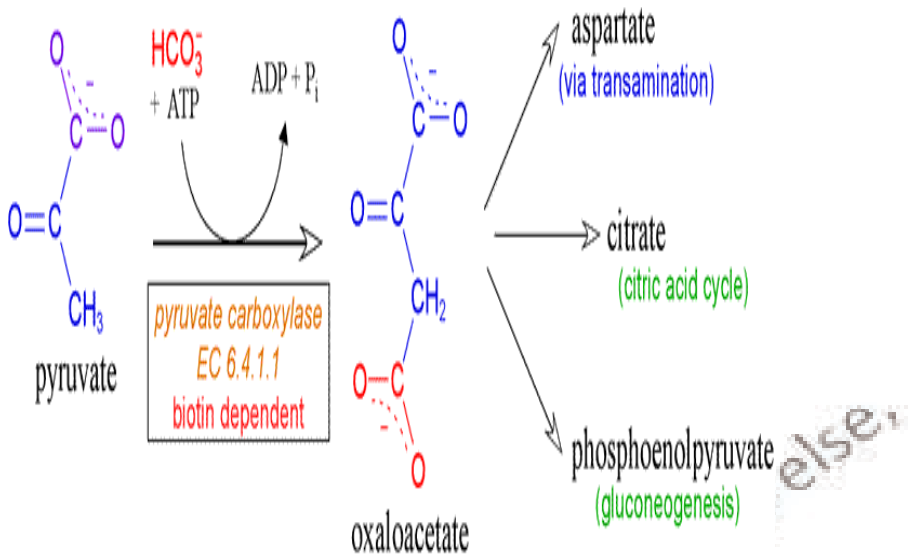
In glycolysis, the impermeable NADH produced in the cytosol (of liver and cardiac cells) are taken up into mitochondria in the form of malate for oxidation via malate shuttle.

In gluconeogenesis, pyruvate in the mitochondria generates impermeable oxaloacetate, which is converted to aspartate and translocated to the cytosol via malate shuttle for the synthesis of glucose.

Solution to Question 25:

Insulin is a repressor of the enzyme pyruvate carboxylase. Since pyruvate carboxylase is a gluconeogenic enzyme, its activity is increased during fasting and decreased following carbohydrate feeds.

Factors influencing pyruvate carboxylase	
Inducers	Glucocorticoids Glucagon Epi nephrine
Repressor	Insulin
Activator	Acetyl CoA
Inhibitor	ADP



Solution to Question 26:

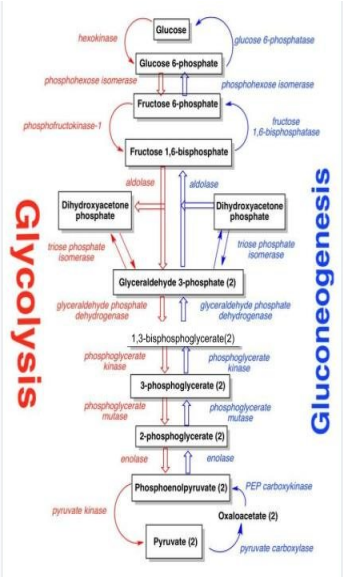
Phosphoglucomutase enzyme is not involved in gluconeogenesis.

Phosphoglucomutase catalyzes the conversion of glucose-6-phosphate to glucose-1-phosphate, which is important for the synthesis of UDP glucose. This is essential for glycogen synthesis, glycogenolysis, and the uronic acid pathway.

Option B: Fructose-1,6-bisphosphatase is an irreversible enzyme in gluconeogenesis and mediates the conversion of fructose-1,6-bisphosphate ° fructose-6-phosphate (cytosol)

Option C: Pyruvate carboxylase is an irreversible enzyme in gluconeogenesis and mediates the conversion of pyruvate ° oxaloacetate in mitochondria.

Option D: Phosphoglycerate kinase is a reversible enzyme in gluconeogenesis and mediates the conversion of 3-phosphoglycerate ° 1,3 biphosphoglycerate.



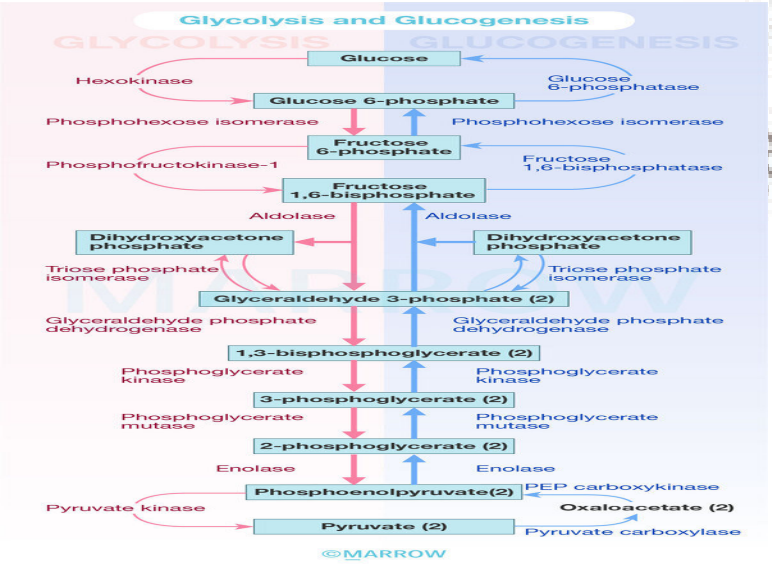
Solution to Question 27:

Oxaloacetate to phosphoenolpyruvate is one of the important irreversible steps in gluconeogenesis.

It is important to note that GTP is utilized in this reaction and CO_2 is liberated.

This reaction occurs in the cytoplasm. Oxaloacetate to phosphoenolpyruvate (PEP) is a step in gluconeogenesis, which is catalyzed by PEP carboxykinase.

The image shows the process of glycolysis and glucogenesis.



Option A: Pyruvate to lactate is a step in anaerobic glycolysis.

Option B: Glucose 6 phosphate to fructose 6 phosphate is a step in glycolysis.

Option C: Pyruvate to acetyl CoA is an anaerobic step in the oxidation of glucose.

Solution to Question 28:

A low insulin/glucagon ratio indicates low insulin and high glucagon levels, which are observed in the fasting state. In the fasting state, all of the above processes (i.e., glycogen breakdown, ketogenesis, and gluconeogenesis) occur, but not glycogen storage.

Metabolic process	Fasting state/ low insulin-glucagon ratio	Fed state/ high insulin-glucagon ratio
Gluconeogenesis	Increased	Decreased
Glycogenolysis	Increased	Decreased

Metabolic process	Fasting state/ low insulin-glucagon ratio	Fed state/ high insulin-glucagon ratio
Ketogenesis	Increased	Decreased
Glycogen storage	Decreased	Increased

Solution to Question 29:

During fasting, there is low insulin, and hence, the activity of phosphofructokinase-1 gets inhibited.

Phosphofructokinase 1 is an enzyme of glycolysis. It is induced by insulin and a well-fed state.

Other options:

Option A: Hormone-sensitive lipase mobilizes stored fats in a fasting state.

Option B: Glycogen phosphorylase is active in fasting state so as to produce glucose from glycogen.

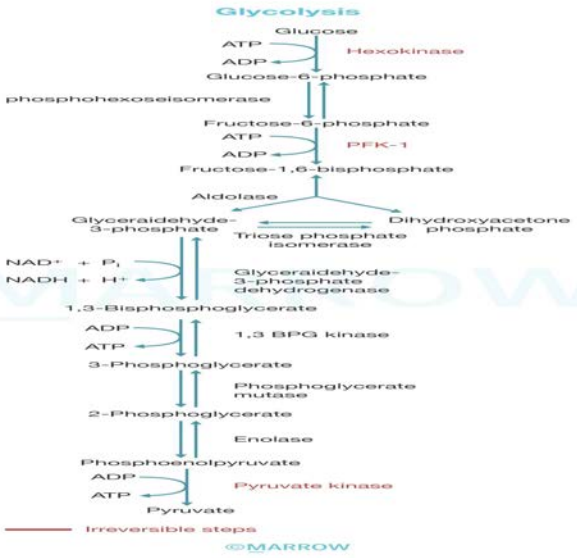
Option C: Pyruvate carboxylase is an enzyme of gluconeogenesis. During the fasting state, the liver is flooded with fatty acids mobilized from adipose tissue. The resulting elevated hepatic acetyl CoA activates pyruvate carboxylase. Other enzymes of gluconeogenesis that are elevated in fasting include Phosphoenolpyruvate carboxykinase, Fructose-1,6-bisphosphatase, and Glucose-6-phosphatase.

Solution to Question 30:

Enzyme activated by a decrease in insulin: glucagon ratio is glucose 6-phosphatase.

A decrease in insulin: glucagon ratio is seen in fasting state, in which there is the activation of gluconeogenesis and inhibition of glycolysis.

Among the given options, only glucose 6-phosphatase is an enzyme of gluconeogenesis and therefore is activated. The enzymes mentioned as other options are irreversible enzymes of glycolysis and are inhibited in the fasting state.



Glycogen metabolism and glycogen storage disorders

Question 1:

Which of the following pairs correctly represents the primary glycosidic bond linkage and the branch-point bond linkage in glycogen, respectively?

a) α(1° 4) linkage; α(1° 6) linkage

b) α (1° 6) linkage; α (1° 4) linkage

c) α (1° 4) linkage; α (1° 4) linkage

d) α (1° 6) linkage; α(1° 6) linkage

Question 2:

In which of the following organs does glycogen synthesis takes place?

a) Liver and kidney

b) Liver and muscle

c) Muscle and RBC

d) Muscle and kidney

Question 3:

Where does the process of glycogen synthesis take place?

a) Mitochondria

b) Golgi apparatus

c) Cytosol

d) Endoplasmic reticulum

Question 4:

Which of the following is a primer acting as an acceptor of glucose residues in the process of glycogen synthesis?

a) Protein

b) Carbohydrate

c) Lipid

d) Nucleic acid

Question 5:

Glucose attached to which of the following serves as the donor source of glucose in the process of glycogen synthesis?

a) UDP

b) ITP

c) ATP

d) GTP

Question 6:

Which of the following depicts the correct sequence of enzymes involved in glycogenolysis in the liver?

a) Phosphorylase, Glucan transferase, Glucose-6-phosphatase

b) Glycogen synthase, Phosphorylase, Glucose-6-phosphatase

c) Glucose-6-phosphatase, Phosphorylase, Debranching enzyme

d) Phosphorylase, Glucose-6-phosphatase, Glucan transferase

Question 7:

Which of the following enzymes catalyze the rate-limiting step of glycogenolysis?

a) Glycogen synthase

b) Glycogen phosphorylase

c) Glucose 6 Phosphatase

d) Phosphoglucomutase

Question 8:

Muscle cannot release glucose from glycogen because of the deficiency of which of the following enzymes?

a) Glucokinase

b) Phosphoglucomutase

c) Glucose-6-phosphatase

d) Muscle phosphorylase

Question 9:

Which of the following vitamins serves as the cofactor of the enzyme glycogen phosphorylase?

a) Vitamin B1

b) Vitamin B6

c) Vitamin B2

d) VItamin B7

Question 10:

Which of the following is an allosteric activator of muscle glycogen phosphorylase?

a) ATP

b) Insulin

c) Glucose

d) 5'AMP

Question 11:

Which of the following do not take part in regulating the enzyme glycogen phosphorylase?

a) cAMP

b) Calmodulin

c) Protein phosphatase-1

d) Glycogenin

Question 12:

Which of the following is the debranching enzyme?

a) Phosphogluconomutase

b) Glucose 6-phosphatase

c) Amylo 1,6 glucosidase

d) Amylo 1,4- 1,6 transglucosidase

Question 13:

Under anaerobic metabolism which of the following substrates yields 3 molecules of ATP?

a) Glycogen

b) Amino acid

c) Glucose

d) Galactose

Question 14:

Which of the following is not a glycogen-storage disorder?

a) Her's Disease

b) Tarui's Disease

c) Scheie's Disease

d) Andersen's Disease

Question 15:

Which of the following glycogen storage diseases is also a lysosomal storage disorder?

a) Von Gierke's disease

b) Pompe's disease

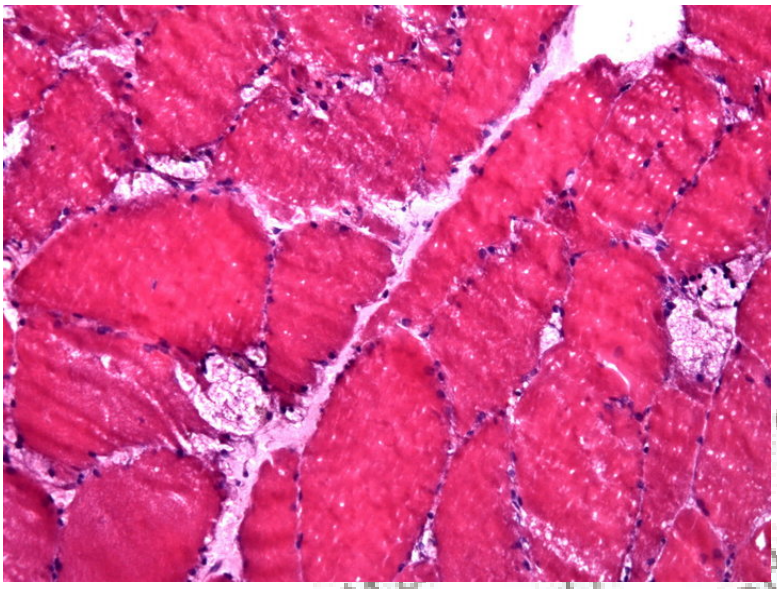
c) McArdle's disease

d) Cori's disease

Question 16:

A 6-month-old baby was brought to the clinic with a history of failure to thrive. Further examination shows generalized hypotonia and hepatomegaly. Chest X-ray image shows massive cardiomegaly. Microscopy of a muscle biopsy is shown in the given image. Which of

the following is the treatment for this condition?



a) Naglazyme

b) Alglucosidase alfa

c) Aldurazyme

d) Elaprase

Question 17:

A 10-year-old boy rapidly develops hypoglycemia after moderate activity. Blood examination reveals raised levels of ketone bodies, lactic acid, and triglycerides. On examination, the liver and kidneys are found to be enlarged. Histopathology of the liver shows deposits of glycogen in an excess amount. What is the diagnosis?

a) von Gierke's disease

b) Pompe's disease

c) McArdle's disease

d) Cori's disease

Question 18:

After evaluation, a child with severe hypoglycemia, lactic acidosis, and hyperuricemia is diagnosed with von Gierke's disease. What is the inheritance pattern of this disease?

a) Autosomal dominant

b) Autosomal recessive

c) X-linked recessive

d) X-linked dominant

Question 19:

A 3-year-old child presents with growth retardation and a history of recurrent attacks of hypoglycemia. On examination, you notice doll-like facies with relatively thin extremities and massive hepatomegaly. Blood examination reveals increased lactic acid, triglyceride, and uric acid levels. All of the following complications can be expected in this child except:

a) Hepatic adenoma

b) Pancreatitis

c) Cardiomyopathy

d) Renal failure

Question 20:

An adolescent male patient presented with pain in the calf muscles on exercise. On biopsy, an excessive amount of glycogen was found to be present in the muscle. What is the most likely enzyme deficiency?

a) Muscle-debranching enzyme

b) Phosphofructokinase 1

c) Glucose 6 phosphatase

d) Muscle glycogen phosphorylase

Answer Key

Question No.	Correct Option
1	a
2	b
3	c
4	a
5	a

6	a
7	b
8	c
9	b
10	d
11	d
12	c
13	a
14	c
15	b
16	b
17	a
18	b
19	c
20	d

Detailed Explanations

Solution to Question 1:

In glycogen, the primary glycosidic bond is an α (1° 4) linkage, whereas the branch point contains an α (1° 6) linkage.

Glycogen is a branched-chain homopolysaccharide with α-D-glucose being the monosaccharide, its major constituent.

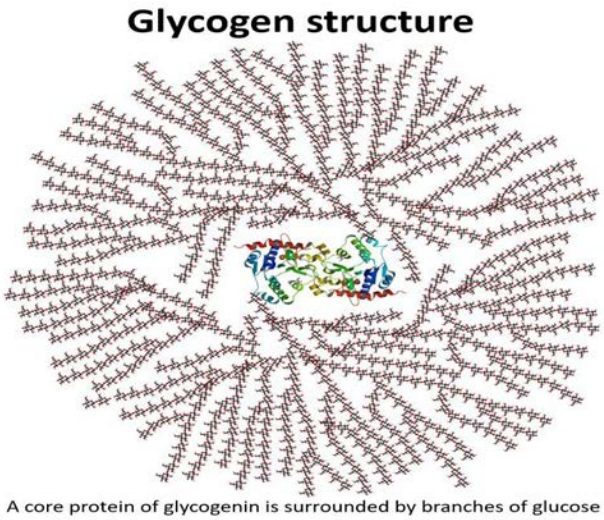
Solution to Question 2:

Glycogenesis takes place majorly in the skeletal muscle and the liver.

Glycogen functions as the secondary long-term energy storage, with the primary energy stores being fats held in adipose tissue.

Glucose 6-phosphate derived from glycogen breakdown can

- Be used as a fuel for glycolysis in muscles
- Be converted into free glucose in the liver and subsequently released into the blood
- Enter into the pentose phosphate pathway to generate NADPH or ribose in a variety of tissues



A core protein of glycogenin is surrounded by branches of glucose units.
The entire globular complex may contain approximately 30,000 glucose units.

Solution to Question 3:

The process of glycogen synthesis takes place within the cytosol of skeletal muscle and liver, where it is stored in the hydrated form with three or four parts of water.

Solution to Question 4:

A protein named glycogenin is the primer acting as an acceptor of glucose residues in the process of glycogen synthesis.

The ensuing entity is glycosylated further by UDP-glucose. Glycogenin further catalyzes the transfer of seven glucose residues from UDP-Glucose, in 1 ° 4 linkages, to form a glycogen primer. This in turn serves as the substrate for the enzyme, glycogen synthase, which is the rate-limiting enzyme of glycogen synthesis.

Solution to Question 5:

α-D-Glucose attached to uridine diphosphate (UDP) serves as the source of glucose, which is added to the glycogen molecule.

UDP-glucose is, in turn, synthesized from glucose 1-phosphate and UTP by the enzyme UDP-glucose pyrophosphorylase.

Solution to Question 6:

The sequence of enzymes involved in steps of glycogenolysis is phosphorylase, glucan transferase, and glucose-6-phosphatase.

Solution to Question 7:

The rate-limiting step in glycogenolysis is catalyzed by the enzyme, glycogen phosphorylase.

The process of phosphorolytic cleavage of the α1 ° 4 linkages of glycogen to yield single units of glucose 1-phosphate, is catalyzed by the enzyme glycogen phosphorylase.

Glycogen phosphorylase requires pyridoxal phosphate as the coenzyme The phosphate group is catalytically active. There are different isoenzymes of glycogen phosphorylase in the liver, muscle, and brain.

Solution to Question 8:

Muscles cannot release glucose into the blood due to the absence of the enzyme glucose-6-phosphatase.

Glucose-6-phosphatase is present in the lumen of the smooth endoplasmic reticulum of hepatocytes. Hence, hepatocytes can convert glucose-6-phosphate into glucose and release glycogen-derived glucose into the blood in order to maintain blood glucose levels stable, until gluconeogenesis kicks in.

However, myocytes lack the enzyme glucose 6-phosphatase. As a result, glucose-6-phosphate cannot be dephosphorylated and released into the blood. Muscle glycogen provides a readily available source of glucose-1-phosphate for glycolysis within the muscle itself.

Defects in the enzyme or glucose-6-phosphate transporter (GLUT 7) can cause type I glycogen storage disease.

Solution to Question 9:

Glycogen phosphorylase requires pyridoxal phosphate, i.e., vitamin B6, as its coenzyme.

The process of phosphorolytic cleavage of the 1 ° 4 linkages of glycogen to yield single units of glucose 1-phosphate, is catalyzed by the enzyme glycogen phosphorylase.

Solution to Question 10:

5'AMP serves as the allosteric activator of muscle glycogen phosphorylase.

Glycogen phosphorylase catalyzes the rate-limiting step in glycogenolysis. It is regulated by allosteric mechanisms and covalent modification by reversible phosphorylation and dephosphorylation in response to hormone action.

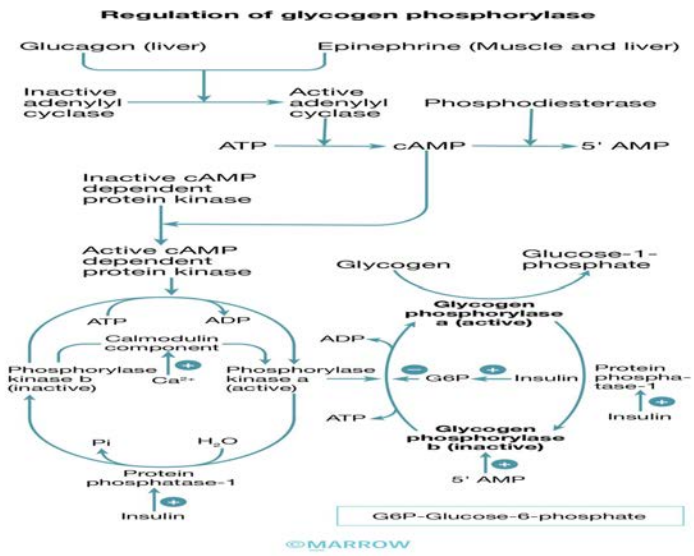
In both liver and muscle, the glycogen phosphorylase is:

• Activated by phosphorylation catalyzed by phosphorylase kinase (to yield phosphorylase a)

• Inactivated by dephosphorylation catalyzed by protein phosphatase-1 (to yield phosphorylase b)

Muscle phosphorylase differs from the liver isoenzyme in having a binding site for 5′ AMP. It acts as an allosteric activator of the (inactive) dephosphorylated b-form of the enzyme.

	Allosteric Activator	Allosteric Inhibitor
Liver		ATPGlucose-6-phosphateFree glucose
Muscle	5' AMP	ATPGlucose-6-phosphate



Solution to Question 11:

Glycogenin cannot regulate glycogen phosphorylase enzyme. It is a polypeptide serving as a primer in the initiation of glycogen synthesis.

Regulation of glycogen metabolism at the level of glycogen phosphorylase:

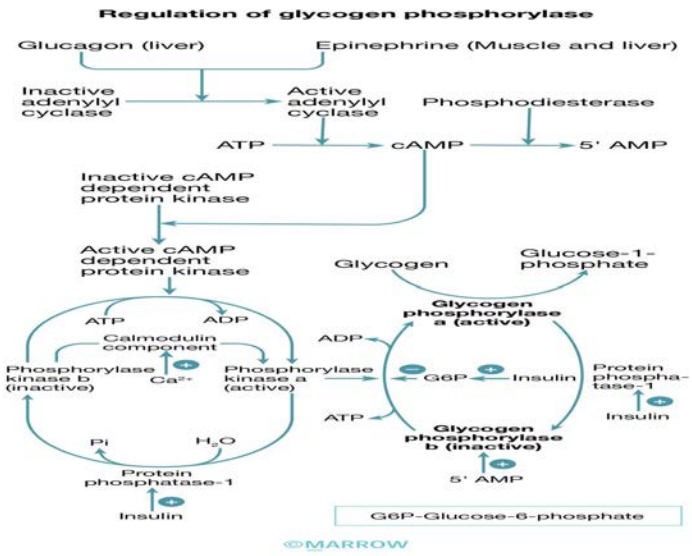
• cAMP activates phosphorylase kinase (by a cAMP-dependent protein kinase), which in turn activates glycogen phosphorylase.

• Calcium activates the calmodulin component of phosphorylase kinase directly, which in turn activates glycogen phosphorylase.

• 5' AMP acts allosterically to activate glycogen phosphorylase.

• Protein phosphatase-1 inactivates glycogen phosphorylase.

The regulation of glycogen phosphorylase is shown in the image below.



Solution to Question 12:

Amylo 1,6 glucosidase + α(1,4)->(1,4) glucan transferase forms the debranching enzyme.

When four glucose residues remain on either side of the branching point, α(1,4)->(1,4) glucan transferase transfers a trisaccharide unit from one branch of glycogen to the other following which, amylo 1,6-glucosidase catalyzes the hydrolysis of the α(1,6) glycosidic bond to liberate free glucose.

Solution to Question 13:

Breakdown of glycogen under anaerobic conditions yields 3 ATPs.

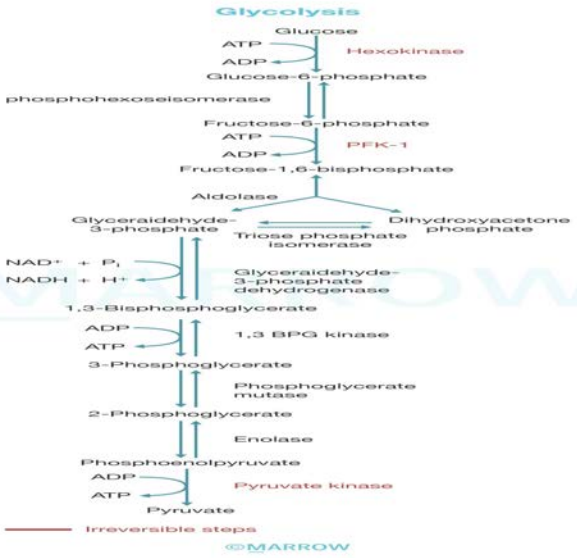
Breakdown of glycogen yields glucose-6-phosphate which can enter the glycolysis pathway. Since it bypasses the first step, one ATP is saved and anaerobic glycolysis yields 3 ATPs.

The first step in glycolysis involves the conversion of glucose to glucose-6-phosphate which requires one ATP. Glycogen breakdown leads to formation of glucose-6-phosphate which can enter glycolysis hence bypassing the first step and saving one ATP.

One ATP is used for conversion of fructose-6-phosphate to fructose 1,6 biphosphate hence net ATP formed from anaerobic glycolysis of glycogen is 3ATP

REACTION	ENZYME	METHOD OF ATP FORMAT ION	NET ATP PRO DUCED
1,3-bisphosphoglycerate to 3 phosphoglycerate	Phosphoglycerate kina se	Substrate level phosphorylati on	2 ATP
Phosphoenol pyruvate to Pyr uvate	Pyruvate kinase	Substrate level phosphorylati on	2 ATP

REACTION	ENZYME	METHOD OF ATP FORMAT ION	NET ATP PRO DUCED
		TOTAL PRODUCED	4 ATP



Solution to Question 14:

Scheie's disease is not a glycogen-storage disease, it is a type of mucopolysaccharidosis.

Solution to Question 15:

Pompe's disease is the glycogen storage disease (GSD), which is also a lysosomal storage disorder.

A small amount of glycogen is degraded by the lysosomal enzyme, α (1° 4)-glucosidase also known as acid maltase. A deficiency of this enzyme causes the accumulation of glycogen in vacuoles in the lysosomes. This is called type II GSD or Pompe's disease.

Glycogen storage disorders that are associated with the following findings:

• Liver cirrhosis: Types III, IV, IX GSD

• Renal dysfunction: Type I GSD

• Neurologic involvement: Type II GSD

Note: Another GSD that is also a lysosomal storage disorder is Danon disease (or glycogen storage disease Type IIb)

Solution to Question 16:

The clinical symptoms of hypotonia, hepatomegaly, and cardiomegaly, along with muscle biopsy showing vacuoles are suggestive of Pompe's disease.

Pompe's disease (type II glycogen storage disorder) occurs due to a mutation in alpha-1,4-glucosidase (also known as acid maltase), a lysosomal enzyme. A defect in this enzyme leads to the accumulation of glycogen in the lysosomes of peripheral, voluntary muscles and progresses to involve the cardiac muscle and diaphragm. Clinical features include hypotonia (floppy baby syndrome), cardiomyopathy, hepatomegaly, and failure to thrive. Myozyme

The treatment for Pompe disease is recombinant enzyme alglucosidase alfa (Myozyme), which can prevent worsening or reverse cardiac and skeletal abnormalities.

Solution to Question 17:

The given features are consistent with von Gierke's disease. It is the most common glycogen storage disease.

Enzyme deficient in this condition is glucose-6-phosphatase (common to both glycogenolysis and gluconeogenesis). Inheritance is autosomal recessive.

Von Gierke disease (type I glycogen storage disorder) occurs due to the deficiency of glucose-6-phosphatase enzyme in the liver, kidney, and intestinal mucosa. Patients present commonly by 3-4 months of age with hypoglycemia (leading to seizures), lactic acidosis, and hepatomegaly. Affected children often have a doll-like face with fat cheeks, relatively thin extremities, short stature, and a protuberant abdomen due to massive hepatomegaly. The kidneys are also enlarged, whereas the spleen and heart are not involved. Laboratory findings include hypoglycemia, lactic acidosis, hyperuricemia, and hyperlipidemia.

A definitive diagnosis is made using a liver biopsy. Fructose and galactose restriction in the diet is advised.

Solution to Question 18:

Von Gierke's disease is inherited as an autosomal recessive trait.

Von Gierke disease (type I glycogen storage disorder) occurs due to the deficiency of glucose-6-phosphatase enzyme in the liver, kidney, and intestinal mucosa. Patients present commonly by 3-4 months of age with hypoglycemia (leading to seizures), lactic acidosis, and hepatomegaly.

Affected children often have a doll-like face with fat cheeks, relatively thin extremities, short stature, and a protuberant abdomen due to massive hepatomegaly. The kidneys are also enlarged, whereas the spleen and heart are not involved. Laboratory findings include hypoglycemia, lactic acidosis, hyperuricemia, and hyperlipidemia.

Solution to Question 19:

The most likely diagnosis in this clinical scenario is von Gierke's disease. Cardiomyopathy is not a complication of von Gierke's disease. On the contrary, cardiomyopathy is associated with Pompe's disease.

Enzyme deficient in von Gierke's disease is glucose-6-phosphatase (common to both glycogenolysis and gluconeogenesis). Inheritance is autosomal recessive.

Glycogen accumulation occurs in the liver and renal tubular cells. Clinical features include:

• Severe hypoglycemia

• Ketosis

• Lactic acidosis

• Hypertriglyceridemia

• Hyperuricemia

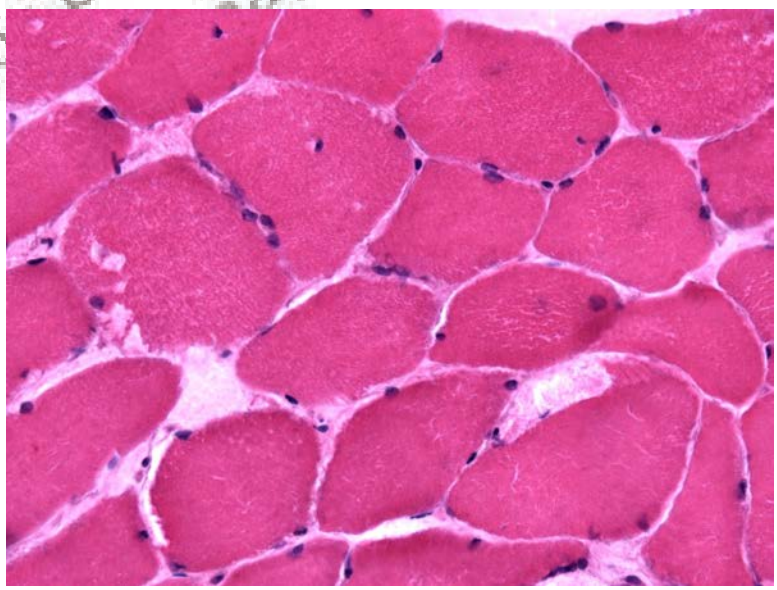
A definitive diagnosis is made using a liver biopsy. Fructose and galactose restriction in the diet is advised.

Complications of von Gierke's disease includes polycystic ovaries, osteopenia, hepatic adenoma, pulmonary hypertension, pancreatitis and renal failure.

Solution to Question 20:

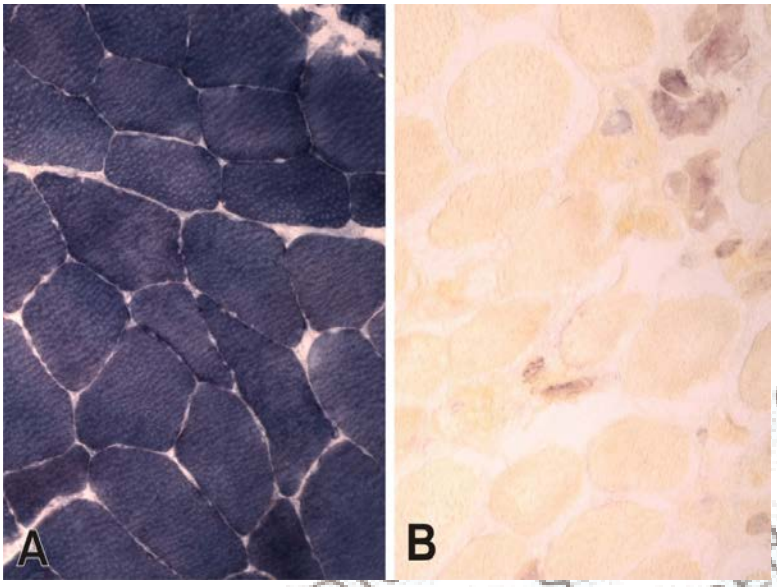
The given clinical features are seen in McArdle's disease. This patient has muscle glycogen phosphorylase deficiency.

The lack of muscle glycogen phosphorylase results in excess glycogen build-up in muscles. The image given below shows the vacuolar myopathy seen in McArdle's disease.



Clinical features include poor exercise tolerance, abnormally high muscle glycogen (2.5-4%), and a very low blood lactate after exercise

The images given below show myophosphorylase enzyme histochemistry showing (A) Normal muscle and (B) McArdle's disease.



HMP shunt pathway, Fructose , Galactose metabolism

Question 1:

Which of the following is false about HMP shunt pathway?

a) No ATP formation

b) Formation of NADPH occurs

c) Synthesis of ribose-5-phosphate occurs

d) Non-oxidative phase is irreversible

Question 2:

In which of the following sites does HMP shunt occur?

a) Golgi apparatus

b) Mitochondria

c) Endoplasmic reticulum

d) Cytosol

Question 3:

HMP shunt can occur at all of the following sites except:

a) Liver

b) Adipose tissue

c) Adrenal cortex

d) Muscles

Question 4:

NADPH is used in _____.

a) Fatty acid synthesis

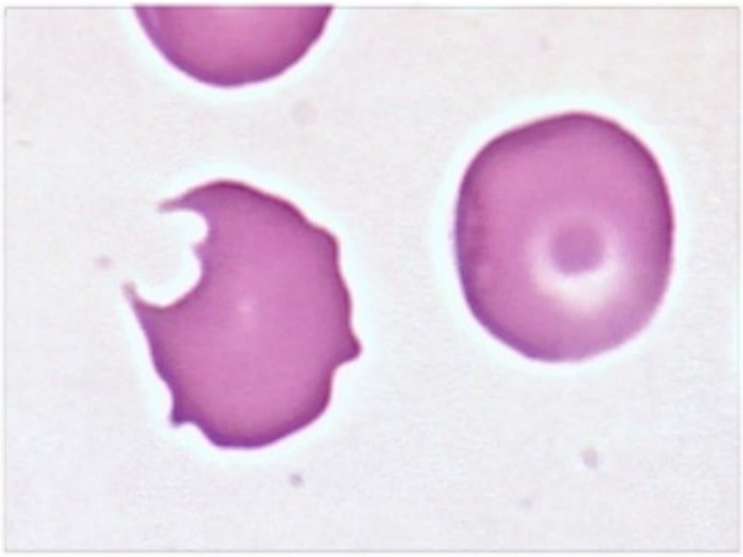
b) Ketone synthesis

c) Gluconeogenesis

d) Glycolysis

Question 5:

A young man with chronic bacterial prostatitis was treated with cotrimoxazole. During treatment, he developed malaise, jaundice, abdominal pain, and dark urine. His blood picture is given below. What is the most likely diagnosis?



a) Hemolytic uremic syndrome

b) Autoimmune hemolytic anemia

c) Paroxysmal nocturnal hemoglobinuria

d) Glucose 6 phosphate dehydrogenase deficiency

Question 6:

All of the following are the products obtained in the uronic acid pathway in humans, except:

a) Vitamin C

b) Glucuronic acid

c) Pentose sugars

d) Proteoglycans

Question 7:

A 28-year-old woman was found to have excessive urinary glucose levels. However, her blood glucose levels were normal. Further investigations revealed high levels of only L-xylulose in the urine. A genetic defect in which of the following pathways is most likely?

a) Krebs cycle

b) Uronic acid pathway

c) Ketone synthesis

d) HMP Shunt

Question 8:

Which of the following is the first enzyme to act on fructose metabolism?

a) Aldolase B

b) Fructokinase

c) Glucokinase

d) Hexokinase

Question 9:

A 10-month-old male baby is brought with vomiting, lethargy, and severe jaundice when weaning was started with fruit juice. Which of the following enzymes is most likely defective?

a) Aldolase A

b) Aldolase B

c) Fructokinase

d) Hexokinase

Question 10:

A patient was incidentally found to have essential fructosuria. Which of the following enzymes is deficient in this condition?

a) Aldolase A

b) Aldolase B

c) Fructokinase

d) Hexokinase

Question 11:

A newborn baby refuses breast milk since the 2nd day of birth but accepts glucose water. He develops vomiting and severe jaundice by the 5th day. Benedict's test was positive for urine and blood glucose was low. The most likely cause is due to the deficiency of

a) Aldose reductase

b) Galactokinase

c) Galactose-1-phosphate uridyl transferase

d) UDP galactose-4-epimerase

Answer Key

Question No.	Correct Option
1	d
2	d
3	d
4	a
5	d
6	a
7	b
8	b
9	b
10	c
11	c

Detailed Explanations

Solution to Question 1:

The non-oxidative phase of the HMP pathway is reversible.

In the HMP shunt, no ATP is formed, but the formation of NADPH and ribose takes place. The pentose phosphate pathway is an alternative route for the metabolism of glucose.

It does not lead to the formation of ATP, but it has two major functions:

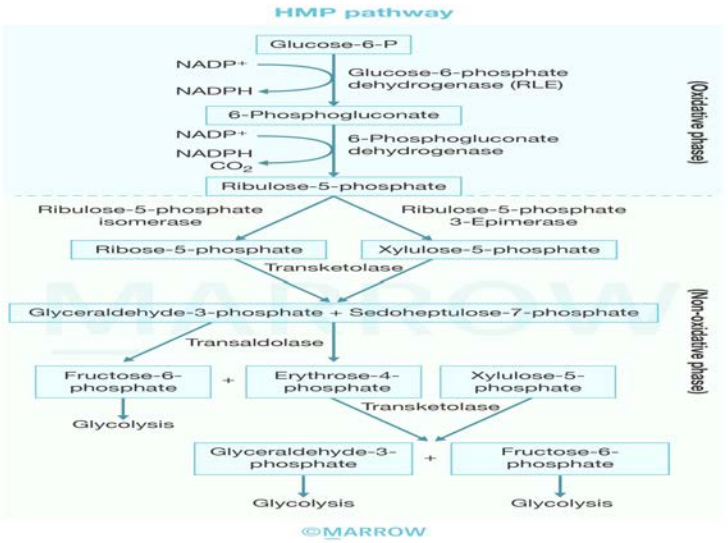
• Main source of NADPH: important for reductive biosynthesis of fatty acids and steroids, and maintaining reduced glutathione for antioxidant activity

• Synthesis of ribose-5-phosphate

There are 2 phases in the HMP pathway:

• Oxidative: irreversible

• Non-oxidative: reversible



Solution to Question 2:

HMP shunt takes place in the cytosol as enzymes needed for the HMP shunt (pentose phosphate) pathway are located in the cytoplasm.

Solution to Question 3:

HMP shunt pathway can not occur in muscles because muscle tissue is deficient in glucose 6-phosphate dehydrogenase, the rate-limiting enzyme in the oxidative phase of the pathway.

HMP shunt pathway takes place in the following:

• Liver

• Adipose tissue

• Adrenal cortex

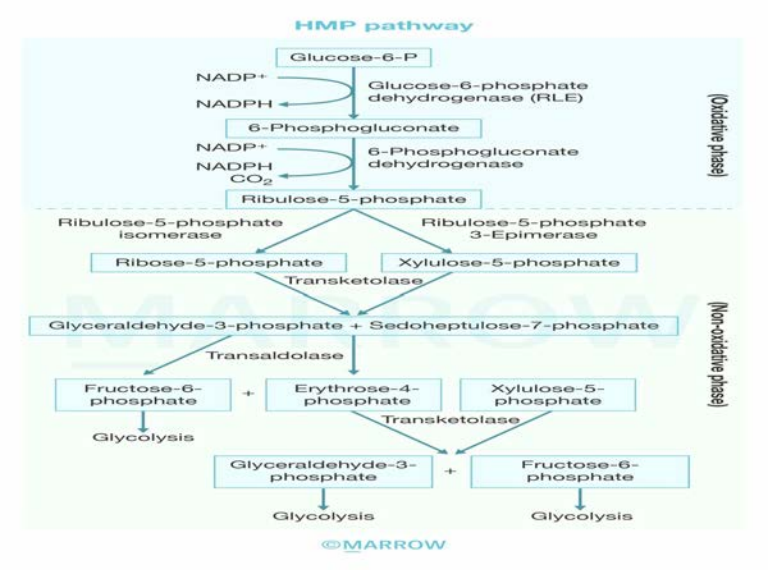
• Thyroid

• Erythrocytes

• Testis

• Lactating mammary glands

Given below is the HMP shunt pathway.



Solution to Question 4:

NADPH is the source of reducing equivalent for fatty acid synthesis.

This NADPH is derived from 3 sources:

- HMP shunt pathway (main source)
- Extramitochondrial isocitrate dehydrogenase (minor source)
- Malic enzyme

Solution to Question 5:

The most likely diagnosis in the above case scenario is glucose-6-phosphate dehydrogenase (G6PD) deficiency.

The clinical features are suggestive of an acute hemolytic reaction following the administration of cotrimoxazole. A hemolytic attack starts with abrupt onset of malaise, weakness, and abdominal or lumbar pain. After an interval of several hours to 2–3 days, the patient develops jaundice and often dark urine, due to hemoglobinuria.

The peripheral smear shows characteristic "bite cells" or degmacytes or blister cells. In G6PD deficiency uncontrolled oxidative stress causes hemoglobin to denature and form Heinz bodies. Bite cells result from the removal of Heinz bodies by macrophages in the spleen.

Cotrimoxazole contains sulfamethoxazole which triggers a hemolytic reaction in persons with G6PD deficiency.

Drugs that carry a risk of clinical hemolysis in persons with G6PD Deficiency:

Drugs	
Antimalarials	PrimaquineChloroquineDapsone/chlorproguanil
Sulphonamides/sulphones	SulfamethoxazoleSulfasalazineSulfisoxazole
Antibacterial/antibiotics	CotrimoxazoleCiprofloxacinChloramphenicolNalidixic acid NorfloxacinNitrofurantoin
Others	NaphthaleneMethylene blue Rasburicase

Solution to Question 6:

Vitamin C is not a product of the uronic acid pathway in humans.

Glucuronic acid, pentose sugars, proteoglycans are all the products obtained in the uronic acid pathway in humans.

In the liver, the uronic acid pathway is the source of glucuronic acid for conjugation of many endogenous and exogenous substances before excretion as glucuronides in urine and bile.

The lack of one enzyme of the uronic acid pathway called L-gulonolactone oxidase in primates and some other animals including human beings explains why ascorbic acid (vitamin C) cannot be endogenously synthesized. Hence, it is a dietary requirement for human beings.

Solution to Question 7:

High levels of L-xylulose in urine point to a diagnosis of essential pentosuria. It is a benign condition caused due to a defect in the uronic acid pathway.

Essential pentosuria has no clinical consequences. However, it can give false-positive results when urinary glucose is measured using alkaline copper reagents (Benedict's test).

The lack of enzyme xylulose reductase of the uronic acid pathway leads to essential Pentosuria. Affected individuals excrete large amounts of L-xylulose in urine. Xylulose being a reducing sugar gives Benedict's test positive. Bial's test is positive as xylulose is a pentose.

Essential pentosuria is a part of Garrod's tetrad which includes :

• Pentosuria

• Albinism

• Cystinuria

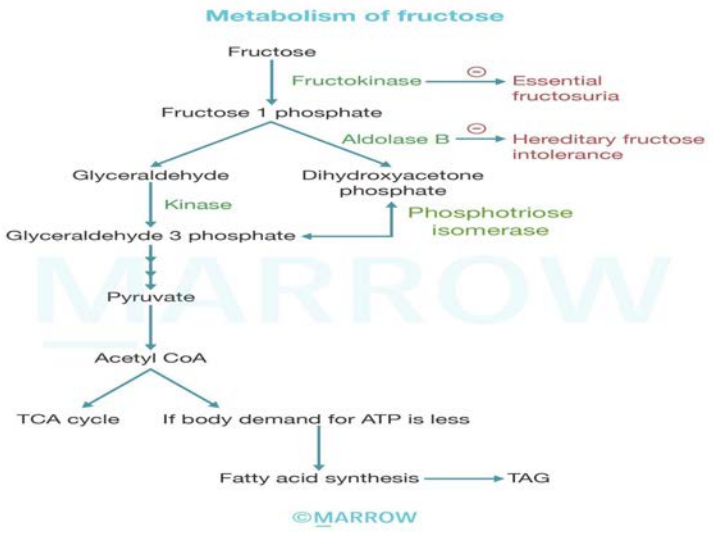
• Alkaptonuria

Solution to Question 8:

Fructokinase is the first enzyme to act on fructose metabolism.

Fructokinase in the liver, kidneys and intestine catalyzes the phosphorylation of fructose to fructose-1-phosphate. This enzyme does not act on glucose and, unlike glucokinase, its activity is not affected by fasting or by insulin, which may explain why fructose is cleared from the blood of diabetic patients at a normal rate.

Deficiency of fructokinase results in essential fructosuria.



Solution to Question 9:

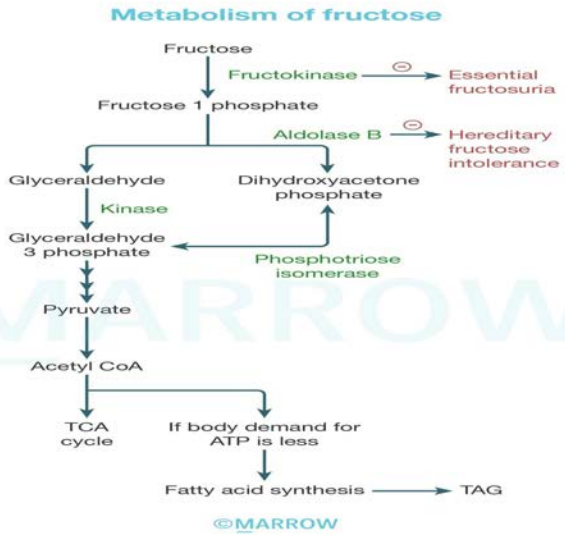
The given clinical scenario is suggestive of hereditary fructose intolerance (HFI), which is due to deficiency of the enzyme aldolase B.

Individuals affected with HFI are asymptomatic until they ingest fructose or sucrose (usually in the form of fruit, fruit juice, or sweetened cereal). Symptoms include:

- Hepatomegaly
- Jaundice
- Vomiting
- Lethargy
- Convulsions
- Hypoglycemia

Urine-reducing sugar (Benedict's test) and tests for ketoses (Rapid Furfural and Seliwanoff's test) are positive. Treatment is the removal of fructose and sucrose from the diet.

The flowchart given below depicts fructose metabolism.



Other options:

Option A: A deficiency of aldolase A leads to myopathy and hemolytic anemia.

Option C: Fructokinase deficiency leads to essential fructosuria, which is a benign condition and does not have significant clinical manifestations.

Option D: Hexokinase deficiency is a rare mutation that can present with hemolytic anemia and does not present with the symptoms in this patient.

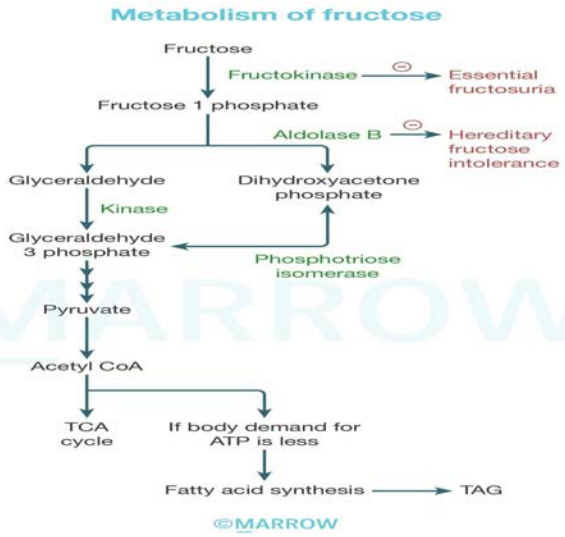
Solution to Question 10:

Essential fructosuria is due to the deficiency of the enzyme hepatic fructokinase. It is inherited in an autosomal recessive pattern.

This defective degradation does not cause any clinical symptoms. Fructose is either excreted unchanged in the urine or metabolized to fructose-6-phosphate by alternate pathways in the body, most commonly by hexokinase in adipose tissue and muscle.

No treatment is indicated for essential fructosuria.

The flowchart given below depicts fructose metabolism.



Solution to Question 11:

The above clinical features on ingestion of breastmilk at birth suggest classical galactosemia, a disorder of galactose metabolism. Galactose-1-phosphate uridyl transferase deficiency leads to classical galactosemia.

It is an autosomal recessive disorder. Clinical features include:

• Accumulation of galactose-1-phosphate in the liver causes liver damage & leads to hepatomegaly, vomiting, convulsions.

• Accumulation of galactose-1-phosphate depletes the liver of inorganic phosphate leading to diminished ATP synthesis which affects gluconeogenesis resulting in hypoglycemia.

• Accumulation of galactose-1-phosphate in the nerves causes intellectual disability.

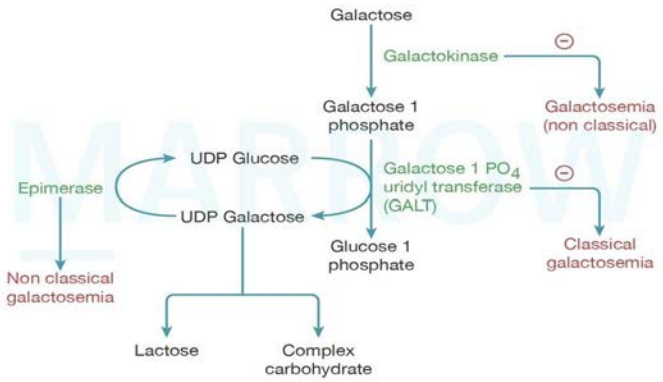
• Accumulation of galactitol in the lens causes an oil-drop cataract.

Urinary reducing sugar (Benedict's test) and the Mucic acid test are positive.

Treatment is a galactose-free diet.

The flowchart given below depicts galactose metabolism.

Metabolism of galactose



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ETC and bioenergetics

Question 1:

Where are the components of the electron transport chain located?

a) Inner mitochondrial membrane

b) Outer mitochondrial membrane

c) Mitochondrial matrix

d) Intermembranous space

Question 2:

Which of the following enzymes is not present in the mitochondrial matrix?

a) Citric acid cycle enzymes

b) β-oxidation enzymes

c) Pyruvate dehydrogenase

d) ATP synthase

Question 3:

Which of the following forms complex I of the electron transport chain?

a) Q-cytochrome c oxidoreductase

b) Succinate-Q reductase

c) NADH-Q oxidoreductase

d) Cytochrome c oxidase

Question 4:

Succinate-Q reductase belongs to which complex of the ETC?

a) Complex I

b) Complex II

c) Complex III

d) Complex IV

Question 5:

Which of the following is a component of cytochrome oxidase?

a) Ca^{2+}

b) Cu^{2+}

c) Mn^{2+}

d) Zn^{2+}

Question 6:

Which of the following ETC complexes does not pump hydrogen ions?

a) Complex I

b) Complex II

c) Complex III

d) Complex IV

Question 7:

Which of the following is known as complex V of the ETC?

a) Q-cytochrome c oxidoreductase

b) ATP synthase complex

c) NADH-Q oxidoreductase

d) Cytochrome c oxidase

Question 8:

Which of the following is an inhibitor of the Fo-F1 domain of ATP synthase?

a) Atractyloside

b) Oligomycin

c) BAL

d) Cyanide

Question 9:

Which of the following substances is an inhibitor of the ETC complex I?

a) Carboxin

b) Hydrogen sulphide

c) Amobarbital

d) Malonate

Question 10:

Which of the following substances inhibits oxidative phosphorylation by blocking the transportation of ADP and ATP?

a) Oligomycin

b) Atractyloside

c) Amobarbital

d) Rotenone

Question 11:

Which of the following is an example of a physiological uncoupler?

a) 2,4 Dinitrophenol

b) Oligomycin

c) Atractyloside

d) Thermogenin

Question 12:

All of the following are high energy compounds except?

a) Phosphoenolpyruvate

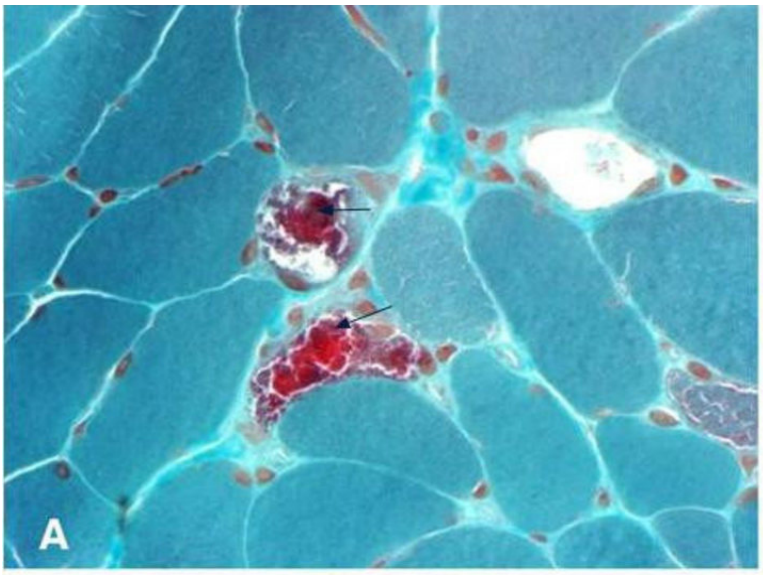
b) Carbamoyl phosphate

c) Creatine phosphate

d) Fumarate

Question 13:

A 3-year-old child presents with muscle weakness, vomiting, and seizures accompanied by focal neurological deficits. Her developmental milestones were normal during the previous visits. Laboratory tests show lactic acidosis and microscopic findings of muscle biopsy are shown below. What is the likely diagnosis?



a) MELAS

b) Duchenne muscular dystrophy

c) Becker muscular dystrophy

d) Myotonic dystrophy

Question 14:

Which of the following is not the source of phosphate?

a) Oxidative phosphorylation

b) Glycolysis

c) Citric acid cycle

d) Gluconeogenesis

Question 15:

In which of the following sites is creatine phosphate not present in?

a) Skeletal muscle

b) Heart

c) Spermatozoa

d) Nephron

Question 16:

Which of the following is a hemoprotein?

a) Cytochrome oxidase

b) Xanthine oxidase

c) L-amino acid oxidase

d) Succinate dehydrogenase

Question 17:

Cytochrome oxidase is inhibited in all of the following poisonings except:

a) Hydrogen sulphide

b) Carbon monoxide

c) Methane

d) Cyanide

Question 18:

A TPN bag consists of 100g glucose, 30g amino acids, and 40g lipids. What is the amount of calories delivered?

a) 840 KCal

b) 880 KCal

c) 640 KCal

d) 680 KCal

Answer Key

Question No.	Correct Option
1	a
2	d
3	c
4	b
5	b
6	b
7	b
8	b
9	c
10	b
11	d
12	d
13	a
14	d
15	d
16	a
17	c
18	b

Detailed Explanations

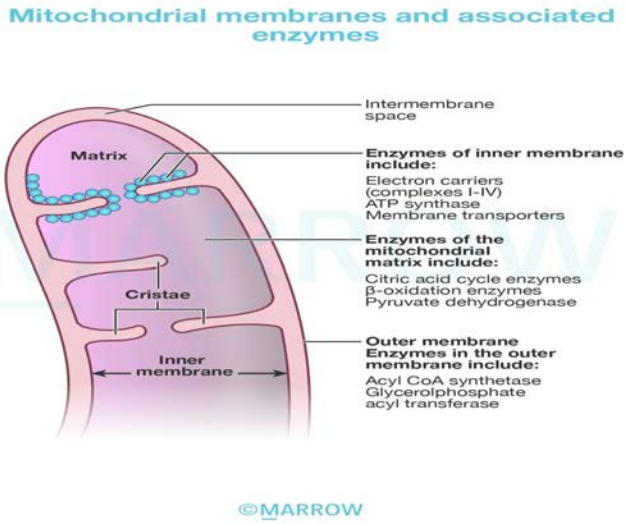
Solution to Question 1:

ETC (electron transport chain) carriers are located on the inner mitochondrial membrane.

ETC is the final pathway by which all the electrons derived from oxidation of carbohydrates, fatty acids, and amino acids are directed towards the final reaction with oxygen to form water and ATP.

Solution to Question 2:

ATP synthase is located on the inner mitochondrial membrane, not mitochondrial matrix.



Note: In contrast to the other enzymes of citric acid cycle, succinate dehydrogenase is bound to the inner surface of inner mitochondrial membrane. Succinate dehydrogenase functions as complex II of electron transport chain.

Solution to Question 3:

Complex I of the electron transport chain (ETC) is NADH-Q oxidoreductase.

The ETC constitutes 4 enzyme complexes:

• Complex I: NADH-Q oxidoreductase

• Complex II: Succinate-Q reductase

• Complex III: Q-cytochrome c oxidoreductase

• Complex IV: cytochrome c oxidase

Components of complex I are FMN and Fe-S complex. NADH-Q oxidoreductase catalyzes the transfer of electrons, from NADH to coenzyme Q, along with the transfer of four H+ across the membrane.

But, this occurs by means of the electrons being transferred from NADH to FMN initially, and from then on, through a series of Fe-S centers, to finally reach coenzyme Q, then to complex III.

Solution to Question 4:

Succinate Q reductase belongs to complex II of ETC.

Components include FAD and Fe-S. It receives electrons from succinate and donates it to a mobile carrier, coenzyme Q, thereby, reducing it and then to complex III.

There is a catalyzed oxidation of succinate to fumarate.

Solution to Question 5:

Cu^{2+} is a component of cytochrome oxidase. Cytochrome C oxidase is complex IV of the ETC.

Components include two heme groups a and a3, and copper (Cu). It is an irreversible complex.

It receives electrons from complex III (Q-cytochrome C oxidoreductase). Components are Cyt b, Cyt c1.

The final electron acceptor from complex IV is oxygen and it is reduced to water.

Solution to Question 6:

Complex II does not pump any proton.

ETC complexes:

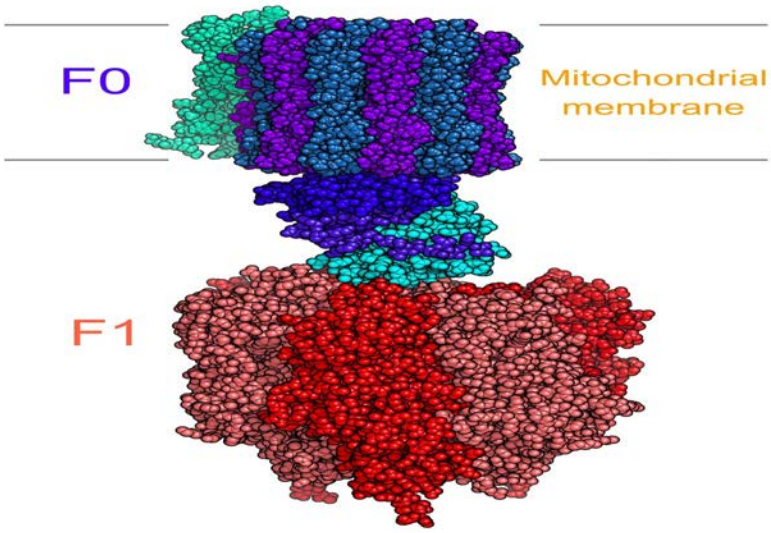
- Complex I pumps 4 protons
- Complex II doesn't pump any proton
- Complex III pumps 4 protons
- Complex IV pumps 2 protons

Solution to Question 7:

ATP synthase is a multi-subunit enzyme complex, also referred to as complex V of the ETC. It synthesizes ATP by utilizing the energy of the proton gradient that is generated by the ETC.

It contains two domains:

- Domain (F_o): spans the inner mitochondrial membrane
- Domain (F_1): extra membranous, which also protrudes into the mitochondrial matrix



The chemiosmotic theory states that the two processes (respiratory chain and synthesis of ATP) are coupled by a proton gradient across the inner mitochondrial membrane. The proton motive force caused by the electrochemical potential difference drives the mechanism of ATP synthesis.

Solution to Question 8:

Oligomycin is a competitive inhibitor of the Fo complex and blocks oxidation and phosphorylation by stopping the flow of protons through the ATP synthase enzyme complex.

Binds to the Fo domain of ATP synthase

↓

Closes the proton channel

↓

Prevents the reentry of protons into the matrix

↓

Stops the phosphorylation of ADP to ATP

Solution to Question 9:

Amobarbital acts as an inhibitor of the ETC complex I by blocking the transfer from Fe-S to Q.

Inhibitors of complex I of the ETC:

- Barbiturates such as amobarbital
- Piercidin A
- Rotenone

Solution to Question 10:

Atractyloside is a drug, which inhibits the process of oxidative phosphorylation by preventing the transportation of ADP into, and ATP out of, the mitochondria.

Solution to Question 11:

Thermogenin is a physiological uncoupler.

Uncouplers mediate their effects by increasing the permeability of the mitochondrial membrane to ions, thereby leading to a collapse in the proton gradient established by allowing the H+ to pass across the membrane without going through the ATP synthase enzyme complex.

Examples:

• Synthetic: 2, 4 dinitrophenol

• Natural: Thermogenin which is found naturally in the brown adipose tissue, whose function is heat generation, especially in a newborn, and thyroxine

Solution to Question 12:

Except for fumarate, all others are high-energy compounds.

The high-energy compounds (in decreasing order of energy):

• Phosphoenolpyruvate (-61.9 KJ/mol)

• Carbamoyl phosphate

• 1,3 Bisphosphoglycerate

• Creatine phosphate

• ATP to AMP

• ATP to ADP

• PPi

• Fructose 6 Phosphate

• Glucose 6 Phosphate

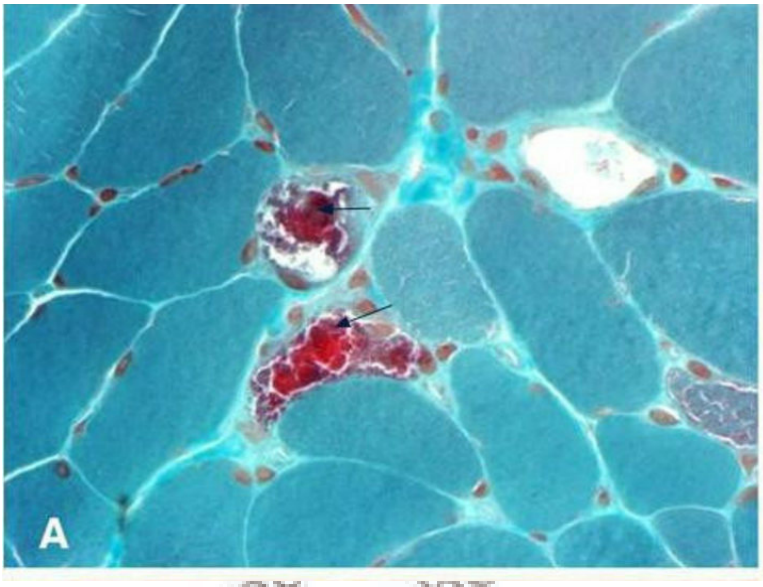
• Glycerol 3 Phosphate (-9.2 KJ/mol)

Solution to Question 13:

The given clinical scenario with microscopy showing ragged red fibres is suggestive of MELAS (mitochondrial encephalopathy, lactic acidosis, and stroke).

It is a mitochondrial inherited condition due to complex I or complex IV deficiency. Patients have normal development previously and then present with symptoms of lactic acidosis and stroke (seizures with focal neurological deficits).

The microscopy of muscle biopsy with modified Gomori trichrome staining shows ragged red fibers (arrowhead) as shown below.



Other mitochondrial inherited diseases:

- Leigh's disease
- Leber hereditary optic neuropathy
- Kearns–Sayre syndrome
- Chronic progressive external ophthalmoplegia
- Pearson syndrome

Solution to Question 14:

Gluconeogenesis is not a source of phosphate that takes part in energy conservation or energy capture.

There are three major sources of phosphate are as follows:

- Oxidative phosphorylation
- Glycolysis
- Citric acid cycle

Solution to Question 15:

Creatine phosphate is not present in nephrons.

Creatine phosphate occurs in vertebrate skeletal muscle, heart, spermatozoa, and brain. Phosphagens act as storage forms of high-energy phosphate.

Solution to Question 16:

Cytochrome oxidase is a hemoprotein widely distributed in many tissues.

Hemoproteins are metalloproteins that have a typical heme prosthetic group. They are present in myoglobin, hemoglobin, and other cytochromes.

Cytochrome oxidase is the terminal component of the chain of respiratory carriers found in mitochondria and transfers electrons resulting from the oxidation of substrate molecules by dehydrogenases to their final acceptor, oxygen. It contains two molecules of heme, each having one Fe atom that oscillates between Fe^{3+} and Fe^{2+} during oxidation and reduction. It also contains two atoms of copper, each associated with a heme unit.

The action of the enzyme is blocked by carbon monoxide, cyanide, and hydrogen sulfide, and this causes poisoning by preventing cellular respiration.

Solution to Question 17:

Cytochrome oxidase is a hemoprotein, which is the terminal component of the respiratory chain carriers (complex IV) in mitochondria. The action of cytochrome oxidase is inhibited in carbon monoxide, cyanide, and hydrogen sulfide poisoning, resulting in prevention of cellular respiration.

Hemoproteins are metalloproteins that have a typical heme prosthetic group. They are present in myoglobin, hemoglobin, and other cytochromes. Cytochrome oxidase contains two molecules of heme, each having one Fe atom that oscillates between Fe^{3+} and Fe^{2+} during oxidation and reduction. It also contains two atoms of copper, each associated with a heme unit.

Complexes of ETC :

Complex I: NADH-CoQ oxidoreductase

Contains - FeS, FMN, Coenzyme Q

Inhibitors – Rotenone

Complex II: Succinate-CoQ reductase

Contains - FeS, Coenzyme Q

Inhibitor - Carboxin

Complex III: $CoQH_2$-cytochrome c oxidoreductase

Contains - cytochrome c, b, C_1, and FeS

Inhibitor - Antimycin A

Complex IV: cytochrome oxidase

Contains - Cytochrome a, a3, and Copper

Inhibitor - Cyanide, Azide, Carbon monoxide

Complex V (informally): ATP Synthase

Inhibitor - Oligomycin

Note: MELAS (mitochondrial encephalopathy, lactic acidosis, and stroke) is an inherited condition due to NADH-Q oxidoreductase (Complex I) or cytochrome oxidase (Complex IV) deficiency.

Solution to Question 18:

The amount of calories delivered by the given TPN (Total Parenteral Nutrition) bag is 880 KCal.

The caloric value of different food components are:

• Carbohydrate (glucose) - 4KCal/g

• Proteins (amino acids) - 4.2KCal/g = ~4KCal/g

• Fat (lipids) - 9Kcal/g

Calculation of the number of calories:

• 100 gm of glucose = 100 x 4KCal=400 KCal

• 30 gm of amino acids = 30 x 4 KCal= 120 KCal

• 40 gm of lipids = 40 x 9 KCal= 360 KCal

Total amount of calories delivered= 400+120+360=880 Kcal

Note: The caloric value of alcohol is 7KCal/g.

Krebs Cycle

Question 1:

What is the site of the TCA cycle?

a) Cytosol

b) Endoplasmic reticulum

c) Mitochondria

d) Lysosomes

Question 2:

Which of the following is true about Krebs cycle?

a) It is the final common pathway for the oxidation of carbohydrates, lipids and proteins

b) Pyruvate condenses with oxaloacetate to form citrate

c) Oxidative phosphorylation occurs in the cytoplasm

d) Alpha keto glutarate is a 4 carbon compound

Question 3:

Which of the following is not an intermediate of the TCA cycle?

a) Citrate

b) Acetyl CoA

c) Succinyl CoA

d) Fumarate

Question 4:

Hyperammonemia inhibits the TCA cycle, by depleting levels of which of the following substances?

a) Oxaloacetate

b) Alpha ketoglutarate

c) Succinyl CoA

d) Fumarate

Question 5:

Aconitase contains which of the following?

a) Ca

b) Mg

c) Fe

d) Mn

Question 6:

Which of the following substances inhibits aconitase enzyme?

a) Arsenite

b) Malonate

c) Fluoroacetate

d) Iodoacetate

Question 7:

At which level does valine enter the TCA cycle?

a) Alpha ketoglutarate

b) Fumarate

c) Succinyl CoA

d) Acetyl CoA

Question 8:

In the Krebs cycle, Which of the following catalyses the step in which the first CO_2 is released?

a) Aconitase

b) Isocitrate dehydrogenase

c) Succinate thiokinase

d) Succinate dehydrogenase

Question 9:

What is the co-factor needed for the isocitrate dehydrogenase enzyme for decarboxylation?

a) Cu^{2+}

b) Mn^{2+}

c) Fe^{2+}

d) Ca^{2+}

Question 10:

Substrate level phosphorylation occurs in which of the following steps of the TCA cycle?

a) Isocitrate to alpha keto glutarate

b) Alpha keto glutarate to succinyl CoA

c) Succinyl CoA to Succinate

d) Succinate to fumarate

Question 11:

How many ATPs are generated per turn of the TCA cycle?

a) 10

b) 20

c) 25

d) 30

Question 12:

All of the following serve as cofactors for the enzymes of the TCA cycle, except:

a) Biotin

b) Pantothenic acid

c) Riboflavin

d) Niacin

Question 13:

Why is the TCA cycle called as an amphibolic pathway?

a) Both oxidative and synthetic reactions take place

b) Metabolites are utilized in other pathways

c) It can proceed both in forward and backward direction

d) Same enzymes can be used in reverse direction

Question 14:

Which of the following substances inhibit the enzyme alpha-ketoglutarate dehydrogenase in the TCA cycle?

a) Iodoacetate

b) Fluoroacetate

c) Arsenite

d) Malonate

Question 15:

Which of the following is not a cofactor of the enzyme alpha-ketoglutarate dehydrogenase?

a) Lipoate

b) Thiamine pyrophosphate

c) FAD

d) Pyridoxal phosphate

Question 16:

All of the following enzymes are components of the pyruvate dehydrogenase enzyme complex, except

a) Pyruvate dehydrogenase

b) Dihydrolipoyl transacetylase

c) Pyruvate kinase

d) Dihydrolipoyl dehydrogenase

Question 17:

Which of the following is inhibited in thiamine deficiency?

a) Isocitrate to alpha-ketoglutarate

b) Succinate to fumarate

c) Pyruvate to acetyl-CoA

d) Fumarate to malate

Question 18:

Which of the following is an allosteric inhibitor of pyruvate dehydrogenase?

a) AMP

b) ADP

c) Acetyl CoA

d) Citrate

Question 19:

Which of the following enzyme deficiencies is likely to cause congenital lactic acidosis in a patient?

a) Pyruvate dehydrogenase enzyme complex

b) Pyruvate kinase

c) Pyruvate Decarboxylase

d) Transketolase

Question 20:

Identify the anaplerotic reaction among the following?

a) Conversion of pyruvate to Acetyl CoA

b) Conversion of pyruvate to Oxaloacetate

c) Conversion of pyruvate to Acetaldehyde

d) Conversion of pyruvate to Lactic acid

Answer Key

Question No.	Correct Option
1	c
2	a
3	b
4	b
5	c
6	c
7	c
8	b
9	b
10	c
11	a
12	a
13	a
14	c
15	d
16	c
17	c
18	c
19	a
20	b

Detailed Explanations

Solution to Question 1:

The TCA cycle (Krebs cycle) takes place in the mitochondria. It occurs in all organs of the body.

Solution to Question 2:

The Krebs cycle is the final common pathway for the oxidation of carbohydrates, lipids, and proteins as glucose, fatty acids, and most amino acids are metabolized to acetyl-CoA or intermediates of the cycle.

Other options:

Option B: Acetyl CoA condenses with oxaloacetate to form citrate.

Option C: Oxidative phosphorylation occurs in mitochondria by ETC (electron transport chain).

Option D: Alpha-ketoglutarate is a 5-carbon compound.

Solution to Question 3:

Acetyl CoA is not an intermediate formed in the TCA cycle, but rather it is a starting substrate.

There are three sources of acetyl-CoA:

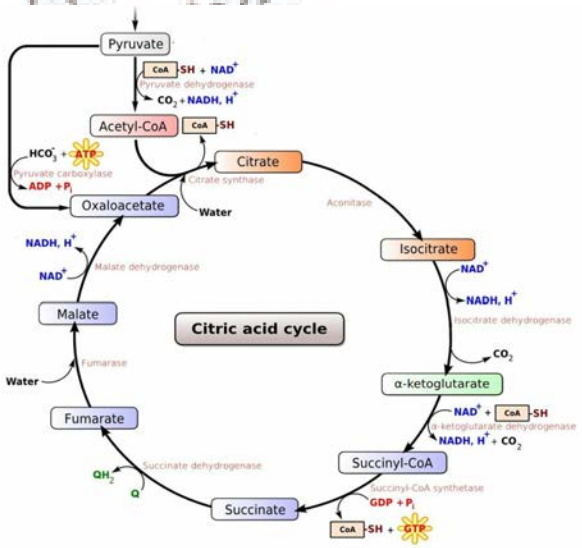
• From glucose ° Pyruvate ° Acetyl-CoA

• Fatty acid oxidation ° Acetyl-CoA

• Ketogenic amino acid oxidation° acetyl-CoA

The starting materials of Kreb's cycle are Acetyl CoA and oxaloacetic acid. All the other options are intermediates in the TCA cycle and oxaloacetic acid is released in the end.

The image given below depicts the Krebs cycle.



Solution to Question 4:

Hyperammonemia results in the depletion of alpha-ketoglutarate.

This happens in two steps:

• The excess free ammonia binds to glutamate and forms glutamine.

• To compensate for the reduced glutamate, alpha-ketoglutarate is aminated to glutamate.

Thus, hyperammonemia eventually depletes alpha-ketoglutarate, leading to a stoppage of the TCA cycle.

Solution to Question 5:

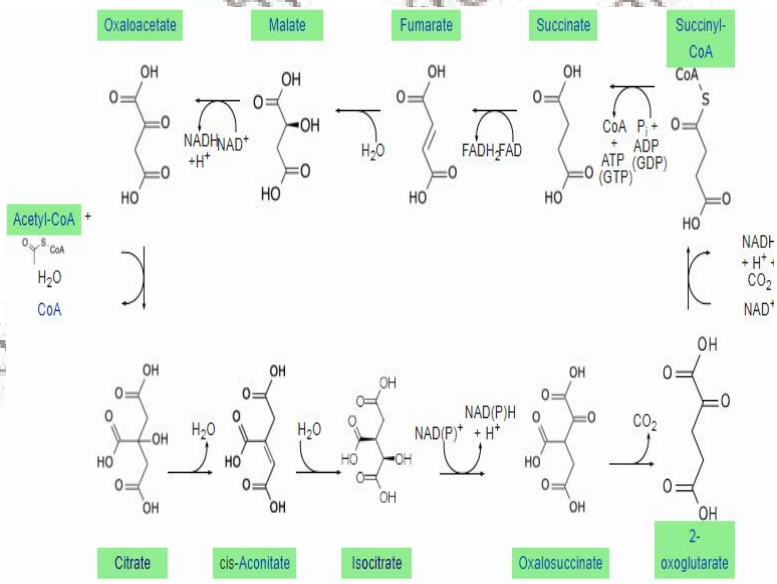
Aconitase is a Fe-containing enzyme. It is involved in the conversion of citrate to isocitrate.

The process occurs as a two-step reaction catalyzed by the enzyme, aconitase.

• There is an initial dehydration reaction with the removal of a molecule of water, from Cis-Aconitate.

• This is followed up by a rehydration reaction, where a molecule of water is added to Cis-Aconitate, thereby forming Isocitrate.

Note: Aconitase is involved in iron homeostasis.



Solution to Question 6:

Flouroacetate is an inhibitor of the aconitase enzyme.

Fluoroacetate combines with Co-A to form fluoroacetyl-CoA. Fluoroacetyl-CoA then condenses with oxaloacetate to form Fluorocitrate. Fluorocitrate then inhibits the enzyme aconitase.

Solution to Question 7:

Valine enters the tricarboxylic acid cycle (TCA) cycle at the succinyl CoA level.

Entry-level in TCA cycle	Amino Acids
Alpha keto-glutarate	ArginineHistidineGlutamine ProlineGlutamate
Succinyl CoA	ValineIsoleucineMethionine
Fumarate	PhenylalanineTyrosine
Oxaloacetate	AspartateAsparagine
Pyruvate	SerineAlanineCysteineGlycineHydroxyproline
Acetyl CoA	LeucineIsoleucineLysineTryptophan

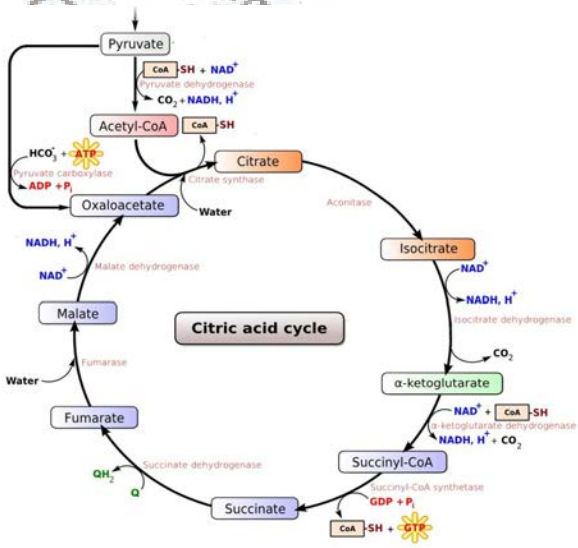
Solution to Question 8:

Isocitrate dehydrogenase catalyses the step in which the first CO_2 is released.

CO_2 is released and NAD+ is reduced to NADH in this reaction.

There are two oxidative decarboxylation steps in TCA cycle:

- Isocitrate to alpha ketoglutarate catalysed by isocitrate dehydrogenase.
- Alpha ketoglutarate to succinyl CoA catalysed by alpha ketoglutarate dehydrogenase.



Solution to Question 9:

Isocitrate dehydrogenase catalyzes the decarboxylation of isocitrate, to form α-ketoglutarate. This decarboxylation reaction, requires Mg2+ or Mn2+ ions.

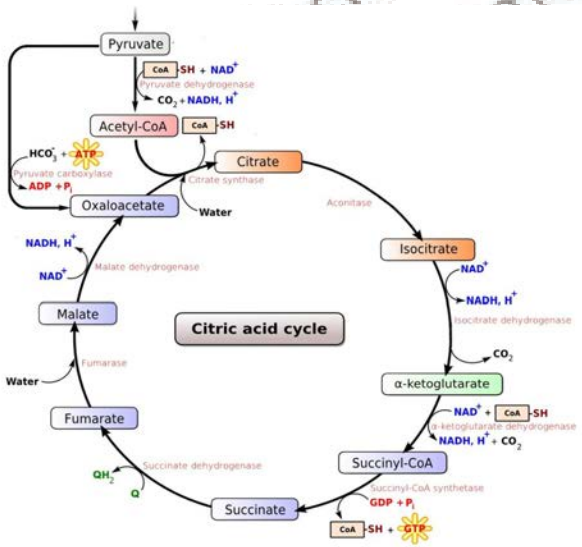
Solution to Question 10:

Substrate level phosphorylation occurs when succinyl CoA gets converted to succinate in the TCA cycle.

Succinyl-CoA is converted to succinate by the enzyme succinate thiokinase or also called succinyl-CoA synthetase.

GTP formation takes place in this step, and hence this is the one and only example of substrate-level phosphorylation in the TCA cycle.

The GTP so formed is used for the decarboxylation of oxaloacetate to phosphoenolpyruvate catalysed by PEPCK in gluconeogenesis.



Solution to Question 11:

Total ATPs generated per turn of the TCA cycle is 10.

The following steps of the TCA yield ATP:

• Isocitrate to alpha-ketoglutarate – Yields 1 NADH per turn

• Alpha-ketoglutarate to Succinyl CoA – yields 1 NADH per turn

• Succinyl Co A to Succinate – yields 1 GTP (Substrate level phosphorylation)

• Succinate to fumarate – Yields 1 FADH2

• Malate to Oxaloacetate – Yields 1 NADH

So total ATP formed will be= 10

3×NADH=7.5 ATP

1× FADH2=1.5 ATP

1× GTP= 1 ATP

TOTAL= 10 ATP PER TURN

Solution to Question 12:

Biotin (B7) is not a cofactor for TCA cycle enzymes.

The following vitamins serve as cofactors for various enzymes of the TCA cycle:

• Riboflavin (B2)

• Active form: FAD

• Cofactor for: Succinate dehydrogenase

• Niacin (B3)

• Active form: NAD+

• Cofactor for: Isocitrate dehydrogenase, α-ketoglutarate dehydrogenase, and Malate dehydrogenase complex

• Thiamine (B1)

• Active form: Thiamin diphosphate

• Cofactor for: decarboxylation step of alpha-ketoglutarate dehydrogenase complex

• Pantothenic acid (B5)

• Active form: As a part of Coenzyme A

• Cofactor for: esterified to the carboxylic acid residues, i.e. acetyl-CoA and succinyl-CoA.

Solution to Question 13:

In the TCA cycle, both oxidative and synthetic reactions take place.

The TCA cycle is not only a pathway for oxidation but also that of interconversion of metabolites arising from the transamination and deamination of amino acids. It provides the substrates for amino acid synthesis by transamination as well as for gluconeogenesis and fatty acid synthesis.

Solution to Question 14:

Arsenite inhibits alpha-ketoglutarate dehydrogenase.

Inhibitors of TCA cycle:

Enzyme	Inhibitor
Aconitase	Fluoroacetate
Alpha-ketoglutarate dehydrogenase	Arsenite
Succinate dehydrogenase	Malonate

Solution to Question 15:

Pyridoxal phosphate is not a cofactor of the enzyme alpha-ketoglutarate dehydrogenase.

Alpha-ketoglutarate dehydrogenase enzyme complex has 5 coenzymes, They are :

• Lipoate

• Thiamine pyrophosphate

• NAD+

• FAD

• Coenzyme A

All the enzymes carrying out oxidative decarboxylation require these 5 coenzymes, namely:

• Pyruvate dehydrogenase, catalyzing the reaction Pyruvate to acetyl CoA

• Alpha-ketoglutarate dehydrogenase

• Branched-chain keto acid dehydrogenase

Solution to Question 16:

Pyruvate kinase is not a component of the Pyruvate dehydrogenase (PDH) enzyme complex.

Pyruvate dehydrogenase complex is a multienzyme complex that is associated with the inner mitochondrial membrane which catalyzes the oxidative decarboxylation of pyruvate to Acetyl-CoA.

The three enzyme components are:

• Pyruvate dehydrogenase

• Dihydrolipoyl transacetylase

• Dihydrolipoyl dehydrogenase

The Cofactors are:

• Thiamine pyrophosphate (TPP)

• Lipoic acid

• Coenzyme A

• Flavin adenine dinucleotide (FAD)

• Nicotinamide adenine dinucleotide (NAD+)

The overall reaction is : Pyruvate + NAD+ + CoA ° Acetyl-CoA + NADH + H+ + CO2

Clinical aspects:

• Arsenite and mercuric ions react with the -SH groups of lipoic acid and inhibit pyruvate dehydrogenase.

• Dietary deficiency of thiamin inhibits PDH eg. Chronic alcoholics may develop potentially fatal pyruvic and lactic acidosis.

• Patients with inherited pyruvate dehydrogenase deficiency present with lactic acidosis, particularly after a glucose load and neurological disturbances.

Note : Pyruvate kinase catalyses the last step of glycolysis ie- transfering phosphate group from phosphoenol pyruvate to ADP forming pyruvate and ATP respectively.

Solution to Question 17:

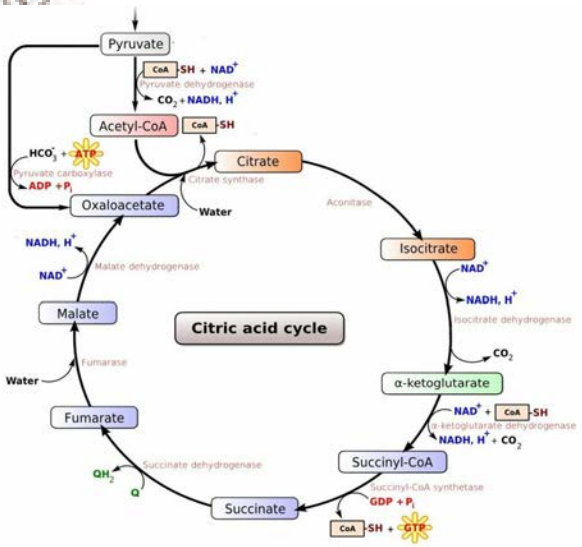
In thiamine deficiency, conversion of pyruvate to acetyl CoA is inhibited.

Thiamine is a cofactor of the pyruvate dehydrogenase component of the PDH complex. In thiamine deficiency, pyruvate dehydrogenase is inhibited. Hence pyruvate is not converted to acetyl CoA.

Other options:

Option A: The enzyme involved in converting isocitrate to alpha-ketoglutarate is isocitrate dehydrogenase which requires Mn2+or Mg2+ as a co-factor.

Option B: Riboflavin (FAD) is the co-factor for succinate dehydrogenase enzyme, which is responsible for converting succinate to fumarate.



Solution to Question 18:

Allosteric inhibitors of the PDH enzyme complex are NADH, acetyl CoA, and ATP.

Solution to Question 19:

Inherited pyruvate dehydrogenase deficiency, can be due to a defect in any one/ more of the components of the PDH enzyme complex. This presents with lactic acidosis.

Causes of lactic acidosis:

- Tissue hypoxia
- Mitochondrial disorders
- Deficiency of:
- PDH Enzyme
- TCA cycle enzymes
- Vit B1
- Poisoning:
- Cyanide
- Mercury
- Arsenite

Solution to Question 20:

The conversion of pyruvate to oxaloacetate is an anaplerotic reaction. It is catalyzed by pyruvate carboxylase.

Anaplerotic reactions are chemical reactions that form intermediates of a metabolic pathway. The above reaction is important in maintaining an adequate concentration of oxaloacetate for the condensation reaction with acetyl-CoA. If acetyl-CoA accumulates, it acts as an activator of pyruvate carboxylase and an inhibitor of pyruvate dehydrogenase, thereby ensuring an adequate supply of oxaloacetate.

Other anaplerotic reactions are:

- Compounds that are metabolized to yield propionyl CoA, which can be then carboxylated and isomerized to succinyl CoA.
- Glutamate and glutamine are important anaplerotic substrates because they yield α-ketoglutarate as a result of the reactions catalyzed by glutaminase and glutamate dehydrogenase.

• Transamination of aspartate leads directly to the formation of oxaloacetate.

Amino acids: Basics

Question 1:

How many amino acids are encoded by the human genetic code?

a) 20

b) 25

c) 17

d) 15

Question 2:

A neonate presents with feeding difficulty and failure to thrive. Her urine has a peculiar burnt sugar odor due to an excess of branched-chain amino acids. Which of the following is unlikely to be elevated?

a) Valine

b) Isoleucine

c) Leucine

d) Lysine

Question 3:

Which of the following is not an aromatic amino acid?

a) Phenylalanine

b) Tyrosine

c) Tryptophan

d) Arginine

Question 4:

Which of the following amino acids has an imino ring?

a) Proline

b) Tyrosine

c) Tryptophan

d) Histidine

Question 5:

A 45-year-old woman presents with loose stools, memory loss, and rashes over sun-exposed areas. On examination, a Cassal's necklace lesion was identified. What group does the amino acid she is deficient in contain?

a) Indole group

b) Imidazole group

c) Thioalcohol

d) Thioester linkage

Question 6:

Which of the following contain a phenol group?

a) Arginine

b) Phenylalanine

c) Tyrosine

d) Proline

Question 7:

Which special group is present in arginine?

a) Guanidine

b) Benzene

c) Imidazole

d) Indole

Question 8:

Which of the following is not a basic amino acid?

a) Histidine

b) Arginine

c) Lysine

d) Glycine

Question 9:

Which of the following amino acids is polar in nature?

a) Aspartic acid

b) Alanine

c) Proline

d) Methionine

Question 10:

A medical student was admitted to the hospital following a hunger strike. Which of the following amino acids cannot be used for gluconeogenesis by his body?

a) Tyrosine

b) Leucine

c) Phenylalanine

d) Tryptophan

Question 11:

Which of the following is a purely glucogenic amino acid?

a) Leucine

b) Tyrosine

c) Tryptophan

d) Alanine

Question 12:

Out of the following, _____ is a semi-essential amino acid.

a) Methionine

b) Tryptophan

c) Leucine

d) Arginine

Question 13:

Which of the following is a non-essential amino acid?

a) Methionine

b) Valine

c) Lysine

d) Aspartate

Question 14:

Beta-alanine is seen in____.

a) Thiamine

b) Niacin

c) Pantothenic acid

d) Biotin

Question 15:

Which of the following amino acids is not formed after post-translational modification?

a) Selenocysteine

b) Triiodothyronine

c) Hydroxy proline

d) Hydroxylysine

Question 16:

Amino acids absorb UV light due to _____.

a) Peptide bond

b) Aromatic ring

c) Disulphide bond

d) Imino group

Question 17:

Which of the following amino acids is the most sterically flexible?

a) Glycine

b) Lysine

c) Leucine

d) Aspartate

Question 18:

Which of the following is not a property of amino acids at isoelectric pH?

a) Maximum precipitability

b) Maximum buffering action

c) Minimum solubility

d) No mobility in electric field

Question 19:

At physiological pH, buffering action in blood is caused by_____.

a) Histidine

b) Methionine

c) Arginine

d) Glycine

Question 20:

A mixture of insulin is being tested in a lab to determine its amino acid sequence. Which of the following reagents can be used?

a) Sanger's reagent

b) Benedict's reagent

c) Seliwanoff's reagent

d) None of the above

Question 21:

A forensic team performed a ninhydrin test on some samples obtained from a crime scene. This test can detect which of the following?

a) Bile salts

b) Amino acids

c) Nucleic acids

d) Lipids

Question 22:

Sakaguchi test is answered by ____.

a) Amino acid

b) Bile acid

c) Lipids

d) Nucleic acid

Question 23:

A neonate was incidentally diagnosed with a benign inborn error of metabolism. A positive Pauly's test would indicate the accumulation of which of the following amino acids in his urine?

a) Tryptophan

b) Cysteine

c) Histidine

d) Arginine

Question 24:

Which of the following is responsible for the negative charge in fibrinopeptide A?

a) Glutamate and Valine

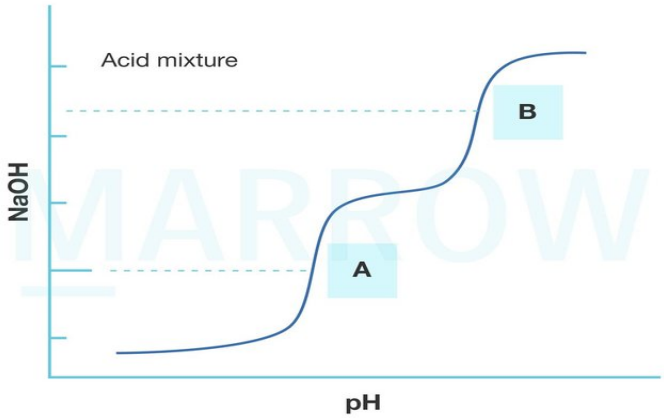
b) Histidine and Lysine

c) Aspartate and glutamate

d) Serine and Threonine

Question 25:

The graph shown below is the titration curve of a biochemical compound. Which of the following statement is true?



a) The maximum buffering capacity of the compound is represented by points A and B

b) The points A and B represent the range of maximum ionization of the amine and carboxyl group

c) The compound has three ionisable side chains

d) The compound has one ionisable group

Question 26:

Replacing alanine by which amino acid, will increase UV absorbance of protein at 280nm wavelength?

a) Leucine

b) Proline

c) Arginine

d) Tryptophan

Question 27:

Nitric oxide is synthesized from:

a) L - arginine

b) L - citrulline

c) Glycine

d) Lysine

Answer Key

Question No.	Correct Option
1	a
2	d
3	d
4	a
5	a
6	c
7	a
8	d
9	a
10	b
11	d
12	d
13	d
14	c
15	a
16	b
17	a
18	b
19	a
20	a
21	b
22	a
23	c
24	c

25	a
26	d
27	a

Detailed Explanations

Solution to Question 1:

The total number of standard aminoacids encoded by triplet codons in the human genetic code is 20.

Selenocysteine (Sec) in eukaryotes and pyrrolysine (Pyl) in prokaryotes are cotranslationally inserted into proteins and are known as the 21st and 22nd amino acids respectively.

Selenocysteine can be found in various human selenoproteins such as thioredoxin reductase, glutathione peroxidase, and deiodinase.

In this question, 21 (20 standard AA + selenocysteine) is not provided as an option, so the correct answer would be 20.

Solution to Question 2:

The given clinical scenario is suggestive of maple syrup urine disease (MSUD). Lysine is not a branched-chain amino acid and hence is unlikely to be elevated.

Branched-chain amino acids are:

• Valine (V)

• Isoleucine (I)

• Leucine (L)

MSUD is due to the deficiency of branched-chain alpha-keto acid dehydrogenase complex (BCKDC), leading to the formation of branched-chain amino acids and their toxic by-products (ketoacids) in the blood and urine. There is a peculiar odor of burnt sugar found in urine, sweat, and cerumen.

Solution to Question 3:

Arginine is not an aromatic amino acid.

The four aromatic amino acids, which include an aromatic ring, are as follows:

• Phenylalanine

• Tyrosine

• Tryptophan

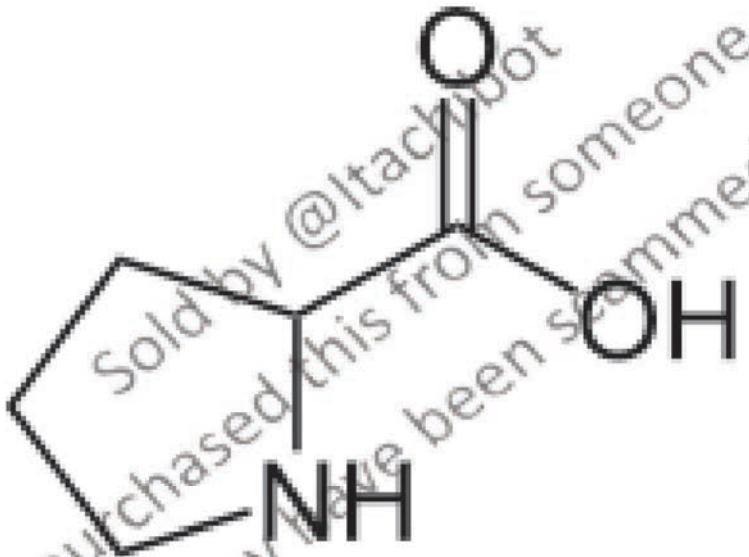
• Histidine

Proline is a heterocyclic amino acid.

Solution to Question 4:

Proline is an amino acid with an imino ring.

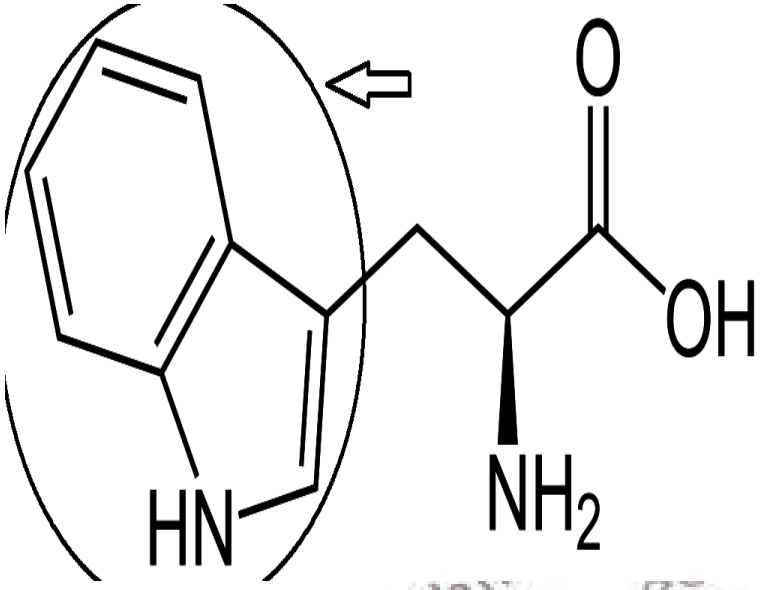
The distinctive cyclic structure of the side chain of proline gives it exceptional conformational rigidity as compared to other amino acids. Hence, proline plays a major role in collagen stability.



Solution to Question 5:

The given clinical scenario is suggestive of pellagra which can be seen in the deficiency of the amino acid tryptophan. Tryptophan contains an indole group.

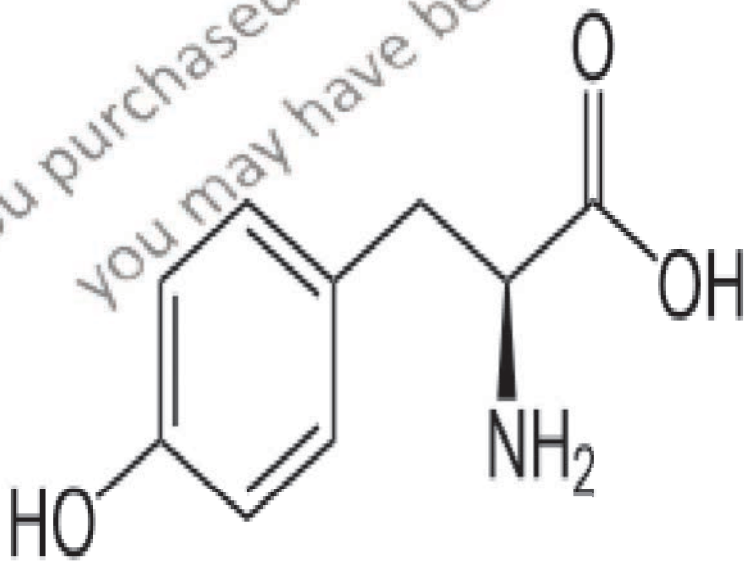
Tryptophan with the marked indole group:



Solution to Question 6:

The amino acid containing a phenol group is tyrosine.

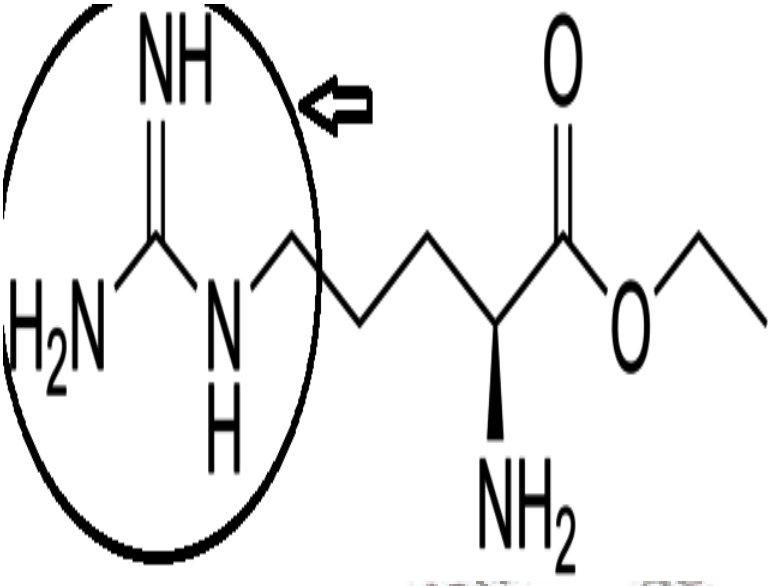
Structure of tyrosine:



Solution to Question 7:

Guanidine is present in arginine.

Image: Structure of arginine indicating the guanidine group



Solution to Question 8:

Glycine is not a basic amino acid.

Mnemonic for basic amino acids-HAL:

• Histidine

• Arginine (most basic)

• Lysine

The acidic amino acids are:

• Aspartic acid (aspartate): most acidic

• Glutamic acid (glutamate)

Solution to Question 9:

Aspartic acid is polar in nature.

Solution to Question 10:

Leucine cannot be used for gluconeogenesis.

Leucine and lysine are the only purely ketogenic amino acids. These two amino acids yield only acetyl-CoA on oxidation (ketogenesis), and hence, they cannot be used for gluconeogenesis.

Four other amino acids give rise to both acetyl-CoA and intermediates. Therefore, they can be used for both ketogenesis and gluconeogenesis. They are:

• Phenylalanine

• Isoleucine

• Tyrosine

• Tryptophan

Solution to Question 11:

Alanine is a purely glucogenic amino acid.

Solution to Question 12:

Arginine is a semi-essential amino acid.

• Essential or indispensable amino acids are those that cannot be synthesized by the body and need to be supplied through diet.

• Semi-essential amino acids are those that are synthesized adequately by adults but synthesized at rates inadequate to support growth in children.

• 8 amino acids are absolutely essential, while 2 are semi-essential.

Essential Amino Acids	Semi-Essential Amino Acids	Non-Essential Amino Acids
IsoleucineLeucineLysineMethioninePhenylalanineThreonineTryptophanValine	ArginineHistidine	AlanineAsparagineAspartateCysteineGlutamateGlutamineGlycineHydroxyprolineHydroxylysineProlineSerineTyrosine

Solution to Question 13:

Aspartate is a non-essential amino acid.

Solution to Question 14:

Beta-alanine is seen in:

• Pantothenic acid (vitamin B5)

• Coenzyme A

• Carnosine

• Anserine

• Acyl carrier protein

Solution to Question 15:

Selenocysteine, the 21st amino acid, is formed by co-translational modification. It is coded by the UGA codon, which is normally a stop codon. The protein synthetic apparatus can identify a selenocysteine-encoding UGA by the presence of the selenocysteine insertion sequence (SECIS) in the untranslated region of mRNA.

Selenocysteine is an analog of cysteine and is synthesized from serine.

Selenocysteine is present at the active site of several human enzymes that catalyze redox reactions such as thioredoxin reductase, glutathione peroxidase, and deiodinase. Since Glutathione peroxidase contains selenocysteine and hence it is an antioxidant.

Impairments in human selenoproteins have been implicated in tumorigenesis and atherosclerosis and are associated with selenium deficiency cardiomyopathy (Keshan disease).

Solution to Question 16:

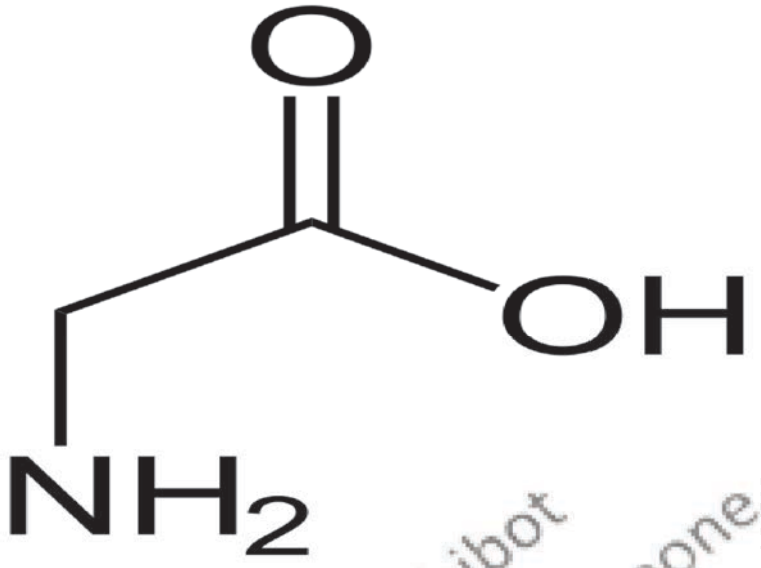
Amino acids absorb UV light due to aromatic ring. Aromatic amino acids such as tryptophan, tyrosine, and phenylalanine absorb 250 to 290 nm UV light. The maximum absorption of UV light is by tryptophan.

Compound	Wavelength absorbed
Aromatic Amino acids	250-290 nm (UV)
Nucleotides (pH=7)	260 nm (UV)
Porphyrins	400 nm (Soret band, visible)

Solution to Question 17:

Glycine is the smallest amino acid with R group as H atom.

So, as it has no R group, it increases the flexibility of the protein. Proline and glycine are frequently found in beta turns.



Solution to Question 18:

The properties of an amino acid at isoelectric pH are as follows:

• Maximum precipitability

• Minimum buffering action (note: maximum buffering action is when the pH=pKa, and not at isoelectric pH)

• Minimum solubility

• No mobility in an electric field

Solution to Question 19:

At physiological pH, buffering action in blood is caused by histidine.

The imidazole group of histidine has a pKa value of 6.5-7.4, which is close to physiological pH. Hence, histidine has the maximum buffering capacity at physiological pH.

Solution to Question 20:

Sanger's reagent is used to determine the amino acid sequence.

Peptides are isolated and hydrolyzed into a mixture of smaller peptides and treated with 1-fluoro-2, 4-dinitrobenzene (Sanger's reagent) which reacts with the alpha amino groups of the amino terminal residues. The amino acid content of each peptide is then determined, and the amino-terminal amino acid is identified.

Solution to Question 21:

Ninhydrin test is used to detect amino acids.

A purple-colored complex (Ruhemann's purple) indicates a positive reaction. Proline and hydroxyproline form a yellow adduct with ninhydrin. Glutamine and asparagine produce a brown color.

This test is used effectively used at crime scenes to develop and identify latent fingerprints.

Solution to Question 22:

Sakaguchi test is answered by amino acid arginine, which contains guanidium group.

Solution to Question 23:

Pauly's test is used for detecting the presence of histidine in proteins.

When proteins containing histidine react with diazotized sulfanilic acid under alkaline conditions, a red color is formed by a coupling reaction. The same reagent will give an orange colored product with the phenol group of tyrosine.

Solution to Question 24:

Aspartate and glutamate are responsible for the negative charge in fibrinopeptide A and B of fibrinogen.

The negative charges contribute to the solubility of fibrinogen in plasma and importantly also serve to prevent aggregation by causing electrostatic repulsion between fibrinogen molecules.

Solution to Question 25:

The maximum buffering capacity of the compound is represented by points A and B, which corresponds to the pK1 and pK2 of the compound.

Other options:

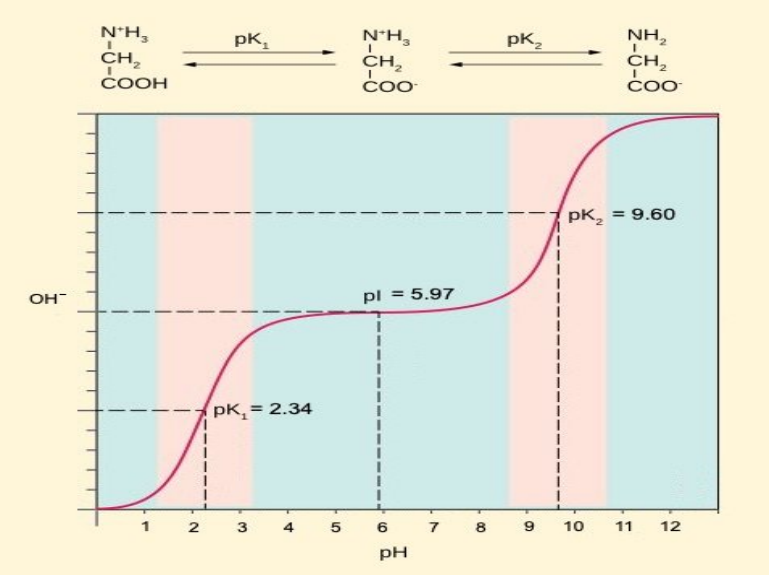
• The points A and B represent the pKa of carboxyl and amino groups respectively.

• The amino acid has no ionizable side chains as the graph shows only two pKas.

• The amino acid has two ionizable groups: an amino group and a carboxyl group.

The above graph resembles the titration curve of the amino acid glycine.

Titration curve of Glycine:



The titration is completed at a pH of 12, where the glycine predominantly exists in the form of H2N - CH2 - COO-.

pK1 and pK2 correspond to the point where the buffering capacity is maximum.

Characteristics of the titration curve:

• Buffering capacity is referred to as the ability of the aminoacid to resist the change in pH due to the addition of acid (H+) or base (OH- equivalents), by accepting or losing the protons respectively.

• An ionizable group refers to the group which can either lose or accept a proton and becomes negatively charged or positively charged ion respectively.

• During the titration, the buffering capacity of the compound reaches its maximum, at points where the pKa of the ionizable groups is reached.

• Each of these pKa are labeled as pK1, pK2, pK3, etc.

• The presence of two pKas indicates that the compound has two ionizable groups. Ex. Glycine.

• The presence of three or more pKas indicates the presence of side chains on the amino acid, with additional ionizable groups.

• Ex. histidine has three ionizable groups, an amino group, an imidazole group (on the side chain), and a carboxyl group. The pKas of these groups respectively correspond to the pK1, pK2, pK3 of the titration curve of histidine.

Solution to Question 26:

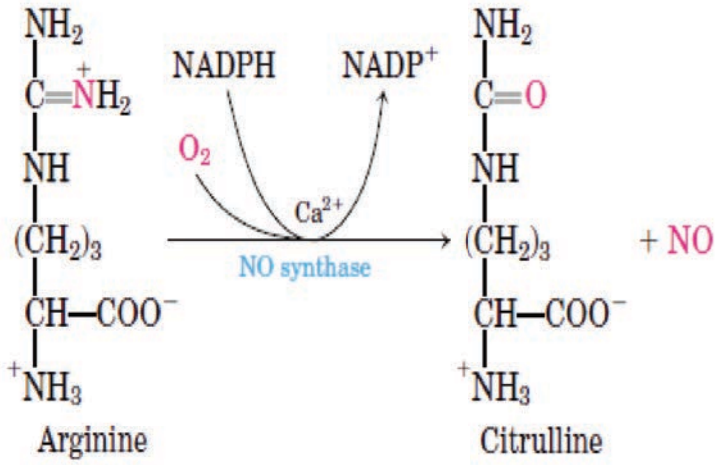
Replacing alanine with tryptophan will increase UV absorbance of protein at 280nm wavelength

The amino acids which absorb 250-290nm (maximum at 280nm) UV light are Tryptophan (Maximum Absorption), Tyrosine, Phenylalanine.

Solution to Question 27:

Nitric oxide is synthesized from arginine by a calcium-dependent reaction catalyzed by NO synthase.

The co-enzymes of NO synthase include FMN, FAD, Heme, and Tetrahydrobiopterin (BH4).



Amino acid: Metabolism

Question 1:

Which of the following enzymes is involved in the transfer of an amino group from an amino acid to a keto acid?

a) Transaminases

b) Aminases

c) Transketolases

d) Deaminases

Question 2:

Which of the following amino acids does not undergo transamination?

a) Methionine

b) Threonine

c) Leucine

d) Glycine

Question 3:

Tetrahydrobiopterin is a coenzyme for all of the following except:

a) Phenylalanine hydroxylase

b) Tyrosine hydroxylase

c) Tryptophan hydroxylase

d) Homogentisate oxidase

Question 4:

Phenylalanine is the precursor of all of the following except?

a) Tyrosine

b) Epinephrine

c) Melatonin

d) Thyroxine

Question 5:

Which of the following amino acid is used for the conversion of norepinephrine to epinephrine?

a) Glycine

b) Alanine

c) Methionine

d) Serine

Question 6:

Which of the following is a metabolic derivative of tryptophan?

a) Melatonin

b) Thyroxine

c) Epinephrine

d) Nor-epinephrine

Question 7:

A child is brought to the OPD with a history of impaired vision and severe photophobia. He has achieved all the developmental milestones at the appropriate age. Examination shows fair hair, light skin color, and pale blue eyes. Which enzyme is most likely deficient in this case?

a) Tyrosinase

b) Tyrosine hydroxylase

c) Tyrosine transaminase

d) Phenylalanine hydroxylase

Question 8:

Vitamin B3 deficiencies in patients can be minimized by the administration of which of the following amino acids?

a) Arginine

b) Tyrosine

c) Tryptophan

d) Histidine

Question 9:

A patient presented with intermittent upper abdominal pain for 4 months, episodic hot flushes of the face for 2 months, and 5-6 episodes of watery stools per day for 1 month. His urinalysis was positive for 5-OH indole acetic acid. Which of the following is responsible for these symptoms in him?

a) 5 hydroxytryptamine

b) 5 hydroxytryptophan

c) 5 carboxytryptamine

d) 5 carboxytryptophan

Question 10:

Conversion of serine to glycine requires which of the following vitamin as a co-factor?

a) Folic acid

b) Thiamine

c) Vitamin C

d) Cyanocobalamin

Question 11:

Which of the following is not obtained from glycine?

a) Creatine

b) Carbon atoms of purines

c) Heme

d) Nitric oxide

Question 12:

Ethanolamine is synthesized from which of the following amino acids?

a) Serine

b) Tryptophan

c) Lysine

d) Histidine

Question 13:

Which of the following vitamins is not involved in sulphur containing amino acid metabolism?

a) Vitamin B6

b) Vitamin B9

c) Vitamin B1

d) Vitamin B12

Question 14:

Taurine is bio-synthesized from which of the following amino acid?

a) Leucine

b) Arginine

c) Cysteine

d) Valine

Question 15:

Which of the following is not a derivative of histidine?

a) Carnosine

b) Ergothioneine

c) Anserine

d) Homocysteine

Question 16:

FIGLU is an intermediate in the metabolism of which of the following?

a) Valine

b) Histidine

c) Methionine

d) Arginine

Question 17:

Gamma-aminobutyric acid (GABA) is synthesized from____.

a) Fumarate

b) Glutamate

c) Histidine

d) Glycine

Question 18:

Creatine is synthesized from all except?

a) Glycine

b) Arginine

c) Methionine

d) Alanine

Question 19:

Glutathione is composed of____.

a) Cysteine and glycine

b) Glutamic acid, cysteine, and glycine

c) Glutamic acid, glycine, and cystine

d) Lysine and methionine

Question 20:

Common precursor amino acid for urea, creatine and nitric oxide is____.

a) Glycine

b) Arginine

c) Aspartate

d) Alanine

Question 21:

In a patient with hyperammonemia, which of the following will also be elevated?

a) Urea

b) Glutamine

c) GABA

d) Uric acid

Question 22:

Which of the following is the major transporter of ammonia from muscles to liver?

a) Alanine

b) Glutamine

c) Glutamate

d) Aspartate

Question 23:

Which of the following enzymes is involved in the process of oxidative deamination?

a) Glutamate synthetase

b) Glutaminase

c) Glutamate dehydrogenase

d) Glutamate aminotransferase

Answer Key

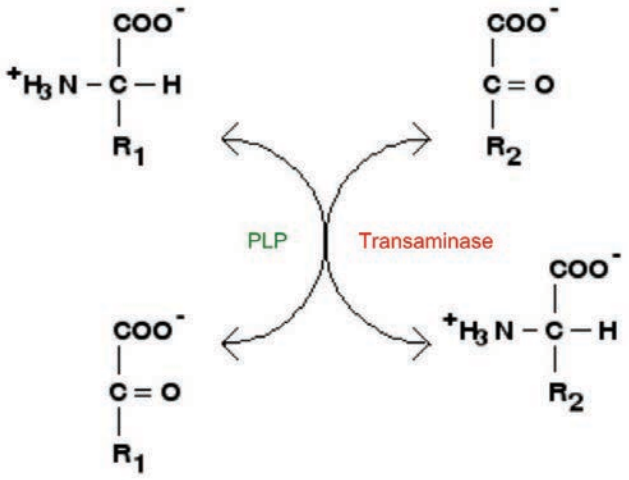
Question No.	Correct Option
1	a
2	b
3	d
4	c
5	c
6	a
7	a
8	c
9	a
10	a
11	d
12	a
13	c
14	c
15	d
16	b
17	b
18	d
19	b
20	b
21	b
22	a
23	c

Detailed Explanations

Solution to Question 1:

In transamination, there is a transfer of an amino group from an amino acid to a keto acid to form another pair of amino acid and keto acid, and this reaction is catalyzed by transaminase.

Transaminases are also known as aminotransferases and belong to the class of transferases. They require pyridoxal pyrophosphate or vitamin B6 as their coenzyme. AST (aspartate transaminase) and ALT (alanine transaminase) are examples of clinically important transaminases. These are elevated in virtually all liver diseases and serve as liver function tests.



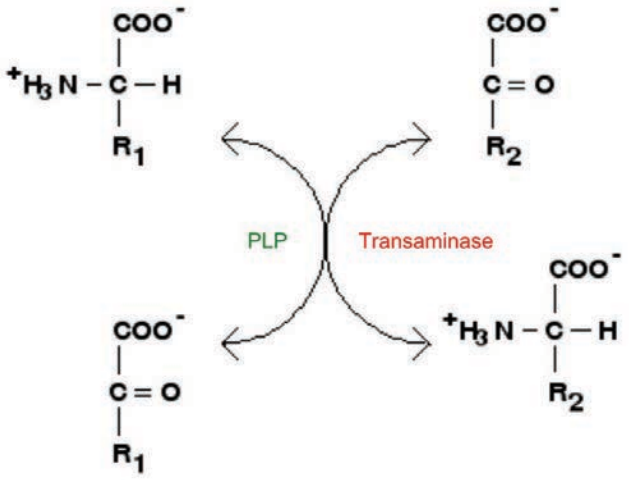
Solution to Question 2:

Threonine does not undergo transamination.

All amino acids undergo transamination except:

- Proline
- Lysine
- Threonine
- Hydroxyproline

In transamination, there is a transfer of an amino group from an amino acid to a keto acid to form another pair of amino acids and keto acid, and this reaction is catalyzed by transaminase. Transaminases are also known as aminotransferases and belong to the class of transferases. They require pyridoxal pyrophosphate or vitamin B6 as their coenzyme. It is important in the biosynthesis of non-essential amino acids.



Solution to Question 3:

Tetrahydrobiopterin (BH4) is a coenzyme for the following enzymes:

- Phenylalanine hydroxylase
- Tyrosine hydroxylase
- Tryptophan hydroxylase
- Nitric oxide synthase

Tetrahydrobiopterin (BH4) resembles folic acid, but it is not a vitamin. It is derived from GTP.

Solution to Question 4:

Phenylalanine is not a precursor of melatonin.

Serotonin is the precursor of melatonin and is derived from tryptophan.

The derivatives of phenylalanine are:

- Tyrosine ° Thyroxine
- Dopa ° Melanin
- Dopamine
- Nor-epinephrine
- Epinephrine

Solution to Question 5:

Methionine in the form of S-adenosyl methionine is used during the conversion of norepinephrine to epinephrine.

Function of S-Adenosyl methionine:

1. Transmethylation reactions:

• Norepinephrine to Epinephrine

• Ethanolamine to Choline

• Acetyl serotonin to Melatonin

• Guanidinoacetate to Creatine

2. DNA methylation

3. Polyamine synthesis:

• Cadaverine

• Putrescine

• Spermidine

• Spermine

Solution to Question 6:

Melatonin is a metabolic derivative of tryptophan.

Tryptophan is hydroxylated by tryptophan hydroxylase to form 5-hydroxy tryptophan which undergoes decarboxylation to form serotonin (5-hydroxy tryptamine). Serotonin is further converted to melatonin in the pineal gland via acetylation and methylation in which S-adenosyl methionine (SAM) acts as the methyl donor.

Melatonin is involved in the regulation of the sleep-wake cycle.

Solution to Question 7:

The clinical scenario is suggestive of albinism which occurs due to a deficiency of copper-containing enzyme tyrosinase causing a defect in melanin production.

Oculocutaneous albinism (OCA) is a group of inherited disorders characterized by a reduction or complete lack of melanin pigment in the skin, hair, and eyes. They have an increased risk for skin cancer.

Its symptoms are albinism with vision problems, which include:

• Strabismus

• Photophobia

• Nystagmus

• Impaired vision or blindness

• Astigmatism

Neuropsychiatric manifestations are not seen.

Solution to Question 8:

Vitamin B3 deficiencies in patients can be minimized by tryptophan as it can be converted into niacin (vitamin B3).

Approximately 60 mg of tryptophan is equivalent to 1 mg of dietary niacin (NADH & NAD+). Tryptophan also produces serotonin and melatonin.

Clinical correlation: Deficiency of the enzyme kynureninase or Vit B6 results in niacin deficiency and pellagra-like symptoms and xanthurenic aciduria.

Solution to Question 9:

The given scenario of a patient with facial flushing, episodic abdominal pain, and watery diarrhea is suggestive of carcinoid syndrome. Excessive levels of serotonin, also known as 5 hydroxy-tryptamine is responsible for it.

Serotonin is a monoamine neurotransmitter. It is produced by hydroxylation and subsequent decarboxylation of tryptophan. It is a potent vasoconstrictor and stimulator of smooth muscle contraction.

In carcinoid syndrome, excessive levels of serotonin is produced which causes flushing, diarrhea, wheezing, damage to the tricuspid valve, etc. The levels of 5-OH indole acetic acid (serotonin metabolite) are elevated in the urine.

Solution to Question 10:

Vitamins required for the conversion of serine to glycine are pyridoxal phosphate (B6) and folic acid.

Serine is also required for the synthesis of :

• Phosphatidylserine: plays a role in apoptosis and a component of cell membrane phospholipids.

• Ethanolamine and choline: a component of lecithin and cephalin.

• Selenocysteine: Serine serves as a precursor for peptidyl selenocysteine.

• Sphingosine: Serine and palmitoyl CoA are the starting material. It is a part of glycolipids.

Serine is also one of the most common sites of phosphorylation.

Solution to Question 11:

Nitric oxide is not a special product obtained from glycine.

Products obtained from glycine:

• Creatine (from glycine, arginine, and methionine)

• Heme (from succinyl CoA and glycine)

• Glutathione (from glutamic acid, cysteine, and glycine)

• C4, C5, and N7 of the purine ring structure

Note: Nitric oxide is a special product obtained from arginine.

Solution to Question 12:

Ethanolamine is synthesized from serine by decarboxylation. Amino acids undergo decarboxylation to form their corresponding amines.

Ethanolamine is involved in the synthesis of cephalin (phosphatidylethanolamine) which is a component of cell membrane phospholipids.

Choline (trimethylethanolamine) is synthesized from ethanolamine in a reaction where S-adenosyl methionine (SAM) is the methyl donor.

Solution to Question 13:

Vitamin B1 (Thiamine) is not involved in sulfur-containing amino acid metabolism.

Vitamins B6, B9, and B12 are involved in sulfur-containing amino acid metabolism.

• Vitamin B6 (Pyridoxine) is the co-enzyme of cystathionine β synthase.

• Vitamin B9 (Folic acid) and B12 (Cobalamin) are involved in the conversion of homocysteine to methionine by methionine synthase.

Clinical correlate: A defect in the enzyme cystathionine β synthase leads to classic homocystinuria.

• Homocystinuria can also be due to a deficiency of vitamin B6, B9 or B12.

Solution to Question 14:

Taurine is biosynthesized from cysteine. The reaction is catalyzed by the enzyme cysteine dioxygenase.

Taurine is involved in the conjugation of primary bile acids (cholic acid and chenodeoxycholic acid) to form the bile salts taurocholic acid and taurochenodeoxycholic acid. Bile salts are important in the emulsification of lipids for their absorption.

Solution to Question 15:

Homocysteine is not a derivative of histidine. It is a derivative of methionine.

Histidine derivatives include the following:

• Carnosine: a dipeptide made up of beta-alanine and histidine, that is found in excitable tissues like the brain and muscles.

• Ergothioneine: a diet-derived amino acid with anti-oxidant properties.

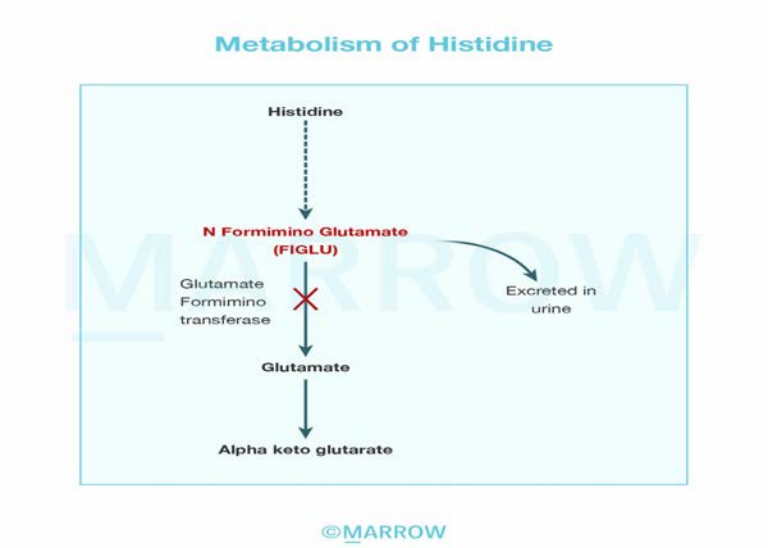
• Anserine: a dipeptide made up of beta-alanine and 1-methylhistidine and is a marker of meat consumption.

Solution to Question 16:

N-Formimino glutamate (FIGLU) is an intermediate in the metabolism of histidine.

The formimino group is transferred to tetrahydrofolate to form glutamate, then alpha-ketoglutarate. In folic acid deficiency, the transfer of the formimino group is impaired, and FIGLU is excreted.

Importance of FIGLU: Individuals deficient in folic acid excrete increased amounts of FIGLU in the urine, particularly after ingestion of a large dose of histidine. The FIGLU excretion test has been used in the diagnosis of folic acid deficiency.



Solution to Question 17:

Gamma-aminobutyric acid (GABA) is formed by the decarboxylation of glutamate by the enzyme glutamate decarboxylase.

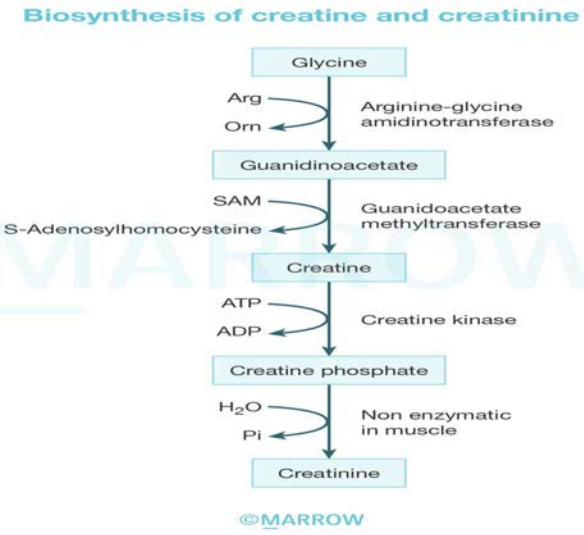
GABA is an inhibitory neurotransmitter in the brain.

Amino acid/ Derivative	Function
GABA	Inhibitory neurotransmitter in the brain and spinal cord
Glycine	Inhibitory neurotransmitter in the spinal cord
Glutamate, Aspartate	Excitatory neurotransmitters in the CNS

Solution to Question 18:

Creatine is synthesized from glycine, arginine, and methionine.

The 1st, 2nd and 3rd steps take place in kidney, liver, and muscle, respectively.



Creatine phosphate donates a phosphate group to ADP to form ATP to be used during the first few minutes of intense muscular contraction. Creatinine is formed in the muscle by non-enzymatic dehydration of creatine and also from creatine phosphate. It is then excreted in the urine. The amount of creatinine excreted is proportional to the muscle mass.

Clinical correlates:

1. Creatine kinase has 3 isoenzymes, which are elevated in the following diseases:

• CK-MM: Muscular dystrophy, acute kidney injury, autoimmune myositis, hypothyroidism, malignant hyperthermia, and drug-induced myopathy

• CK-BB: Stroke, brain injury, and meningitis

• CK-MB: Myocardial infarction, and myocarditis

2. Normally, creatinine is rapidly removed from the blood and excreted in the urine. Hence, creatinine clearance is a sensitive indicator of kidney function.

Solution to Question 19:

Glutathione is a tripeptide composed of glutamate, cysteine and glycine

Glutathione is an antioxidant that helps maintain the integrity of the RBC membrane. It is also used in phase 2 xenobiotic reactions (conjugation).

Note:

• Cystine: Two molecules of cysteine linked by a disulfide bond.

• Cystine is reduced to cysteine by cystine reductase or cysteine can be oxidized to its disulfide derivative cystine.

Solution to Question 20:

Arginine is the common precursor for urea, creatine and nitric oxide.

Solution to Question 21:

In a patient with hyperammonemia, glutamine will also be elevated as ammonia is detoxified in the brain to form glutamine.

Glutamine synthetase combines ammonia with glutamate to form glutamine. It requires the hydrolysis of ATP.

Solution to Question 22:

Alanine is the major transporter of ammonia from muscles to liver. This is by a process called the Cahill cycle.

Alanine is extracted by the liver and undergoes transamination with alpha-ketoglutarate by alanine transaminase (ALT) to form pyruvate and glutamate. Pyruvate is in turn, converted to glucose by gluconeogenesis and glucose is released into the blood.

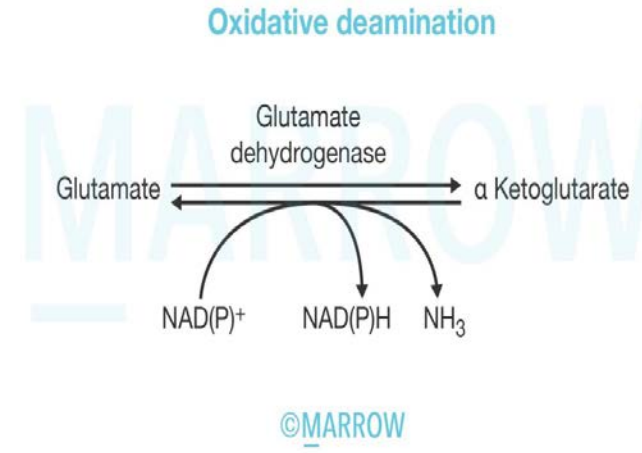
Glutamate undergoes oxidative deamination and release ammonia which further enters urea cycle.

Solution to Question 23:

Glutamate dehydrogenase is involved in the process of oxidative deamination.

In contrast to transamination (transfer amino groups), oxidative deamination by glutamate dehydrogenase results in the liberation of the amino group as free ammonia. Glutamate dehydrogenase can use NADH or NADPH during oxidative deamination.

The image below shows oxidative deamination by glutamate dehydrogenase:



Amino acid: Metabolic disorder

Question 1:

An infant is brought with a history of vomiting and poor feeding. A musty odor is noted in the baby. Guthrie test was done and it was found to be positive. All are true regarding this disease except

a) Phenylalanine hydroxylase enzyme defect

b) White patch of hair due to tryptophan deficiency

c) Phenylacetate positive in urine

d) Mental retardation is present

Question 2:

A 3-month-old infant born to consanguineous parents presented with global developmental delay and seizures. Plasma amino acid analysis revealed elevated phenylalanine levels. A urine $FeCl_3$ test done was positive. What is detected in this test?

a) Phenylacetate

b) Phenyllactate

c) Phenylpyruvate

d) Phenylalanine

Question 3:

Defective fumarylacetoacetate hydrolase enzyme is associated with _____.

a) Type 1 Tyrosinemia

b) Type 2 Tyrosinemia

c) Type 3 Tyrosinemia

d) Type 4 Tyrosinemia

Question 4:

An infant is brought with abdominal distention and increased irritability for 10 days. Physical examination showed jaundice, hepatomegaly, and a boiled cabbage odor. Investigations showed elevated transaminases and succinylacetone. What is the treatment of choice for this condition?

a) Dietary restriction of phenylalanine

b) Dietary restriction of tyrosine

c) Nitisinone (NTBC)

d) All of the above

Question 5:

Richner-Hanhart syndrome is associated with a defect in which of the following?

a) Fumarylacetoacetate hydrolase

b) Tyrosine aminotransferase

c) p-Hydroxyphenylpyruvate hydroxylase

d) Homogentisate oxidase

Question 6:

Neonatal tyrosinemia is due to the deficiency of which of the following enzymes?

a) Fumaryl acetoacetate hydrolase

b) Tyrosine transaminase

c) Tyrosinase

d) 4-hydroxyphenylpyruvate dioxygenase

Question 7:

Which of the following enzyme deficiency most commonly causes the disease shown in the image below:

a) Tyrosinase

b) Tyrosine hydroxylase

c) Phenylalanine hydroxylase

d) Tyrosine transaminase

Question 8:

A man complains of bluish-black discoloration of the sclera and pinna for the last 6 months. Since childhood, his urine has turned black on standing, as shown in the image below. What is the most probable diagnosis?

a) Phenylketonuria

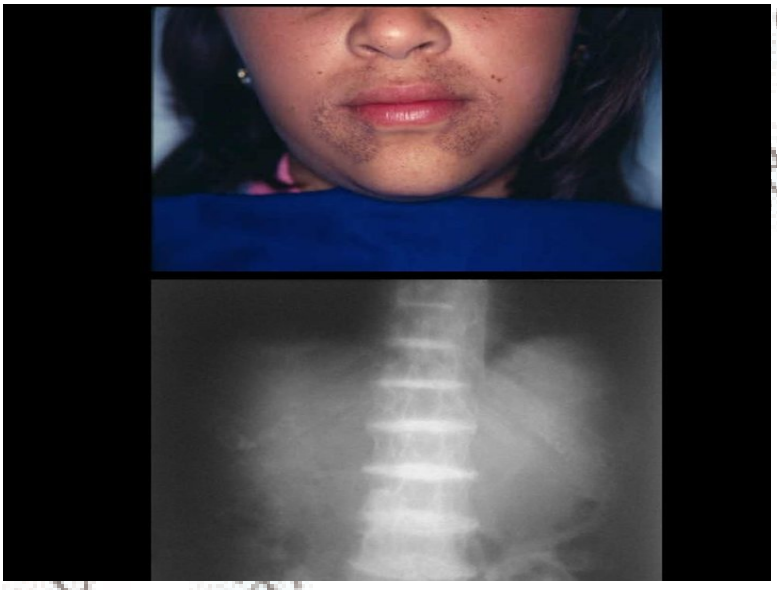
b) Alkaptonuria

c) Tyrosinemia

d) Maple syrup urine disease

Question 9:

The patient presents with pigmentation of the face as shown in the image below and gradually increasing backache and stiffness. The X-ray image of the spine is shown. What is the most probable diagnosis?



a) Phenylketonuria

b) Alkaptonuria

c) Maple syrup urine disease

d) Tyrosinemia

Question 10:

A child is brought with complaints of diarrhea, skin eruptions, and delayed mental development. She is found to have neutral aminoaciduria and a positive Obermeyer test. The impaired renal and intestinal transport of which amino acid must be suspected in this child?

a) Tyrosine

b) Tryptophan

c) Phenylalanine

d) Threonine

Question 11:

Xanthurenic aciduria is seen in which of the following vitamin deficiency?

a) Vitamin B2

b) Vitamin B3

c) Vitamin B5

d) Vitamin B6

Question 12:

In which condition is excessive urinary excretion of 5- hydroxy indole acetic acid seen?

a) Alkaptonuria

b) Albinism

c) Carcinoid syndrome

d) Phenylketonuria

Question 13:

A patient presents with recurrent kidney stones. Microscopic examination finding of the urine specimen is shown below. Which of the following is not seen in the urine of this patient?

a) Ornithine

b) Lysine

c) Arginine

d) Histidine

Question 14:

Defect in the metabolism of which of the following is associated with increased cardiovascular risk?

a) Branched amino acids

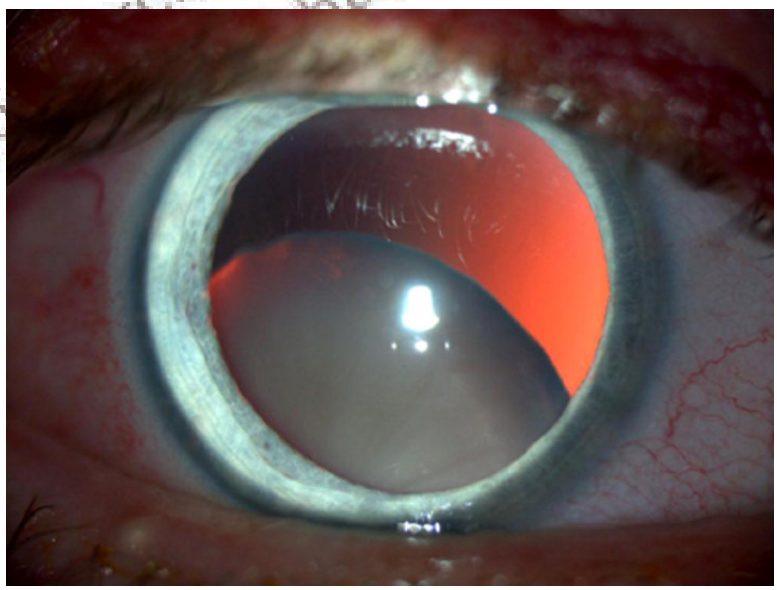
b) Aromatic amino acids

c) Sulfur-containing amino acids

d) Imino acids

Question 15:

A 4-year-old child developed a sudden onset weakness of the left side of the body with facial deviation to the right side. On examination, he is tall and thin with elongated limbs. Ocular findings are shown in the image below. What is the most likely inborn error of metabolism in this child?



a) Cystinuria

b) Homocystinuria

c) Cystinosis

d) Alkaptonuria

Question 16:

Garrod's tetrad includes all of the following, except ____.

a) Alkaptonuria

b) Cystinuria

c) Pentosuria

d) Phenylketonuria

Question 17:

Primary hyperoxaluria is due to the defect in the metabolism of _____.

a) Threonine

b) Serine

c) Glycine

d) Alanine

Question 18:

The histidine load test is done to identify the deficiency of which of the following?

a) Iron

b) Folic acid

c) Pyridoxine

d) Vitamin B12

Question 19:

Deficiency of aspartoacylase leads to:

a) Canavan disease

b) Oasthouse syndrome

c) Hawkinsinuria

d) Hartnup disease

Question 20:

Maple syrup urine disease type I is due to a defect in _____.

a) Decarboxylation

b) Oxidation

c) Carboxylation

d) Deamination

Question 21:

Type II MSUD results from the mutation in the genes encoding _____.

a) α-Ketoacid decarboxylase

b) Dihydrolipoyl transacylase

c) Dihydrolipoamide dehydrogenase

d) None of the above

Question 22:

A neonate with poor feeding, vomiting, and lethargy was found to have increased isovaleric acid in urine. This is due to a defect in the metabolism of _____.

a) Isoleucine

b) Valine

c) Leucine

d) Lysine

Question 23:

A 16-year-old boy with progressive myopia and night blindness was found to have gyrate atrophy of the retina and choroid. Which of the following will be elevated in plasma?

a) Cystine

b) Ornithine

c) Lysine

d) Alanine

Question 24:

Segawa syndrome is due to the deficiency of which of the following?

a) Tetrahydrobiopterin

b) Pantothenic acid

c) Biotin

d) Tetrahydrofolic acid

Question 25:

During the routine follow-up of a 9-month-old child, you notice a peculiar boiled cabbage odor. What is the most likely diagnosis?

a) Hypermethioninemia

b) Isovaleric aciduria

c) Multiple carboxylase deficiency

d) Trimethylaminuria

Question 26:

Which of the following should be avoided by a patient of fish odour syndrome?

a) Biotin

b) Choline

c) Niacin

d) Pantothenic acid

Question 27:

In a patient with Wilson's disease which of the following is decreased in urine?

a) Tyrosine

b) Phosphorous

c) Serine

d) 3-methylhistidine

Question 28:

A patient came to the hospital with complaint of abdominal pain. Routine investigations were unremarkable. On observation, the urine turned black on standing. Which of the following enzymes is likely to be defective in this patient?

a) Homogentisate oxidase

b) Xanthine oxidase

c) Phenylalanine hydroxylase

d) Dihydro oratate dehydrogenase

Answer Key

Question No.	Correct Option
1	b
2	c
3	a
4	d
5	b
6	d
7	a
8	b
9	b
10	b
11	d
12	c
13	d
14	c
15	b
16	d
17	c
18	b
19	a
20	a
21	b
22	c

23	b
24	a
25	a
26	b
27	d
28	a

Detailed Explanations

Solution to Question 1:

The given scenario of an infant with a history of vomiting and poor feeding with a musty odour is suggestive of phenylketonuria (PKU). Tryptophan deficiency is not a feature of this disorder. The deficiency of the enzyme phenylalanine hydroxylase causes PKU.

Characteristic clinical findings in Phenylketonuria:

- Mental retardation
- Failure to walk or talk
- Seizures
- Hyperactivity
- Tremor
- Microcephaly
- Failure to grow
- Deficiency of pigmentation (fair hair, light skin color, and blue eyes)

The Guthrie bacterial inhibition test is a screening test for phenylketonuria. This test uses a phenylalanine requiring strain of Bacillus subtilis. The ferric chloride test is also used as a screening test for PKU. Tandem mass spectrometry is the gold standard investigation.

Solution to Question 2:

The infant with developmental delays and seizures with elevated phenylalanine levels is most likely suffering from phenylketonuria. The FeCl3 test is used to detect urinary phenylpyruvate.

In this test, a test tube with 1 ml of fresh urine, 3-4 drops of 10gm% ferric chloride is added. A positive test reveals a transient green color due to the formation of a ferric phenol complex. Phenylpyruvate, phenyl lactate, phenylacetate and phenyl acetyl glutamate are products of alternative pathways of phenylalanine catabolism.

Specificity of FeCl3 test:

Reacts with FeCl3	Does not react with FeCl 3
Phenylpyruvate	Phenylacetate
	Phenyllactate
	Phenyl acetyl glutamate
	Phenylalanine
	Pyruvic acid
	Tyrosine

Solution to Question 3:

Defective fumarylacetoacetate hydrolase enzyme is associated with type I tyrosinemia (tyrosinosis). It is the most common type of tyrosinemia.

There are three types of tyrosinemias:

• Type 1 (Hepatorenal tyrosinemia): Fumaryl acetoacetate hydrolase deficiency

• Type 2 (Oculocutaneous tyrosinemia/Richner-Hanhart syndrome): Tyrosine transaminase deficiency

• Type 3 (Neonatal tyrosinemia)

Solution to Question 4:

The given clinical scenario of a child with abdominal distension, jaundice, hepatomegaly, and a boiled cabbage odor is suggestive of type 1 tyrosinemia (tyrosinosis). It is treated by a combination of nitisinone and dietary restriction of phenylalanine and tyrosine.

Type 1 tyrosinemia is the most common form of tyrosinemia caused by defective fumarylacetoacetate hydrolase enzyme. They usually present between 2-6 months of age with acute liver failure. They may also present with peripheral neuropathy resembling acute porphyria. Renal involvement is manifested as a Fanconi-like syndrome with hyperphosphaturia, hypophosphatemia, and vitamin D-resistant rickets.

An odor resembling boiled cabbage resulting from increased methionine metabolites can be noted. Elevated levels of succinylacetone in blood or urine is diagnostic of type 1 tyrosinemia.

Note: A boiled cabbage odor can also occur in hypermethioninemia.

Solution to Question 5:

Richner- Hanhart syndrome (Tyrosinemia type II) is associated with a defect in tyrosine aminotransferase. It is also called oculocutaneous tyrosinemia. Alternate metabolites of tyrosine

are excreted in type II Tyrosinemia.

It is an autosomal recessive condition with onset between ages 2 and 4 years when painful circumscribed calluses develop on the pressure points of the palms and soles.

Solution to Question 6:

Neonatal tyrosinemia is due to the deficiency of 4-hydroxyphenylpyruvate dioxygenase

There are three types of tyrosinemias:

• Type 1 (Hepatorenal tyrosinemia): Fumaryl acetoacetate hydrolase deficiency

• Type 2 (oculocutaneous tyrosinemia/Richner-hanhart syndrome): Tyrosine transaminase deficiency

• Type 3 (Neonatal tyrosinemia)

Solution to Question 7:

The image shows a child with albinism and is due to a deficiency of copper-containing enzyme tyrosinase causing a defect in melanin production. The affected child has milky white hair and milky white skin.

In untreated phenylketonuria, the baby looks normal at birth, with a skin and hair color lighter than other members of the family. Later, the child has blonde hair, skin with a light complexion, and blue irides. Manifestations are not so severe as in OCA. The image shows an infant with white hair, so albinism is the best choice.

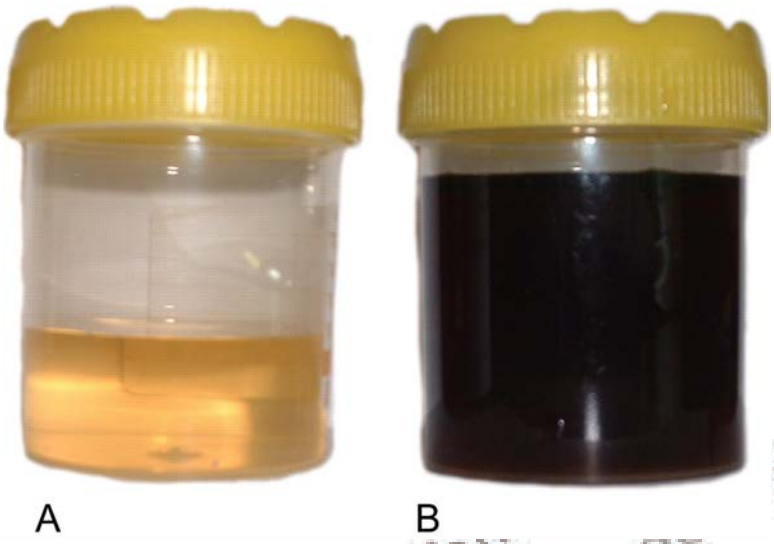
Note - Tyrosine hydroxylase is the rate-limiting enzyme in the biosynthesis of catecholamines. Tyrosine is first hydroxylated to form 3,4-dihydroxyphenylalanine (DOPA) by the enzyme tyrosine hydroxylase. This enzyme is abundant in the central nervous system, the sympathetic ganglia, and the adrenal medulla.

Solution to Question 8:

The given scenario of a patient with urine that turns black on standing and blackish discoloration of soft tissues (oochronosis) is characteristic of alkaptonuria. It occurs due to the excretion of homogentisic acid in the urine.

Alkaptonuria is an autosomal recessive condition due to the deficiency of homogentisate oxidase (also called homogentisate 1,2-dioxygenase). The homogentisic acid in these patients is oxidized to benzoquinone acetate and polymerized to form black-colored alkapton bodies, which deposit in soft tissues like cartilage, and this phenomenon is called ochronosis.

In the following images, A shows normal urine and B shows urine from patients with alkaptonuria



Solution to Question 9:

The given clinical images show the accumulation of blackish discoloration on the face and the X-Ray shows calcification of the intervertebral discs. These findings are associated with alkaptonuria.

Alkaptonuria is a condition caused due to the deficiency of homogentisic acid oxidase. This results in the accumulation of homogentisic acid.

This condition is associated with the following symptoms:

• Black ochronotic pigmentation (Alkapton bodies) of cartilage and collagenous tissue

• Homogentisic aciduria (patient's urine contains elevated levels of homogentisic acid, which is oxidized to a dark pigment on standing)

• Large joint arthritis

• Calcification of intervertebral discs known as as ochronotic spondyloarthropathy

Solution to Question 10:

This child presents with diarrhea, dermatitis, and delayed mental development. This combined with neutral aminoaciduria and a positive Obermeyer test should raise suspicions of Hartnup's disease. This is due to impaired renal and intestinal absorption of tryptophan and other neutral amino acids.

Hartnup's disease is characterized by a varied clinical presentation including ataxia (lack of muscle control), delayed mental development, diarrhea, and dermatitis. Patients can also develop symptoms similar to pellagra as tryptophan is a precursor of niacin.

Diagnosis is based on aminoaciduria and increased excretion of indole compounds detected by Obermeyer test.

Solution to Question 11:

Xanthurenic aciduria is seen in deficiency of vitamin B6 (pyridoxal phosphate), a coenzyme required by kynureninase enzyme to convert 3-OH kynurenine to 3-OH anthranilic acid during tryptophan metabolism.

Solution to Question 12:

Excessive urinary excretion of 5-hydroxy indole acetic acid (5-HIAA) is seen in carcinoid syndrome.

Carcinoid syndrome is marked by symptoms of bronchospasm, cutaneous flushing, abdominal cramps, and diarrhea. It occurs due to excessive production of serotonin, a specialized product of tryptophan metabolism by tumors commonly in the small intestine, liver and lungs. It is then metabolized and excreted in urine as 5-HIAA.

Solution to Question 13:

The image shows hexagonal, translucent and white crystals suggestive of cystinuria. Histidine is not seen in the urine of this patient.

Cystinuria is an autosomal-recessive disease due to a defect in renal PCT & intestinal amino acid transporter that is essential for reabsorptive transport of Cystine and the dibasic amino acids Ornithine, Lysine, and Arginine (COLA).

Diagnosis of cystinuria is by the cyanide–nitroprusside test. Initial treatment is with adequate hydration, alkalization of the urine with citrate supplementation or acetazolamide, and dietary modification to reduce salt and protein intake. If this fails then patients are usually started on chelation therapy with penicillamine.

Solution to Question 14:

Defect in the metabolism of sulfur-containing amino acids (homocysteine) due to the deficiency of cystathionine-β-synthase leads to homocystinuria and increases cardiovascular risk.

Homocysteine is highly cytotoxic and elevated levels within the bloodstream are believed to damage the endothelial lining of arterial vessels, leading to intravascular coagulation.

Solution to Question 15:

This child presented with an acute stroke. This in addition to elongated limbs and a tall body habitus with ocular examination showing downward subluxation of the lens is suggestive of homocystinuria.

Homocystinuria is an important cause of pediatric stroke. It is most commonly caused by a defect in cystathionine β synthase enzyme leading to homocysteine accumulation in the urine and blood. They are generally normal at birth; the first sign usually is subluxation of the lens (ectopia lentis) seen after 3 years of age.

Manifestations of homocystinuria:

• Ectopia lentis.

• Intellectual disability and psychiatric and behavioral disorders.

• Thromboembolic episodes.

• Skeletal abnormalities resembling those of Marfan syndrome: They are tall and thin with elongated limbs and arachnodactyly.

Elevations of both methionine and homocysteine in body fluids are the diagnostic lab findings.

Note: Marfan syndrome is the other cause of heritable ectopia lentis. However, in Marfan's, the lens is dislocated in an upward and temporal direction whereas in homocystinuria the lens is dislocated in a downward and nasal direction.

Solution to Question 16:

Phenylketonuria is not a component of Garrod's tetrad.

Garrod's tetrad includes the following:

• Alkaptonuria: Due to homogentisate oxidase deficiency

• Albinism: Due to tyrosinase deficiency

• Cystinuria: A type of aminoaciduria that prevents proper reabsorption of basic, or positively charged, amino acids that includes cystine, ornithine, lysine and arginine(COLA)

• Pentosuria: Due to the deficiency of L-xylulose reductase or xylitol dehydrogenase, which are necessary for xylitol metabolism

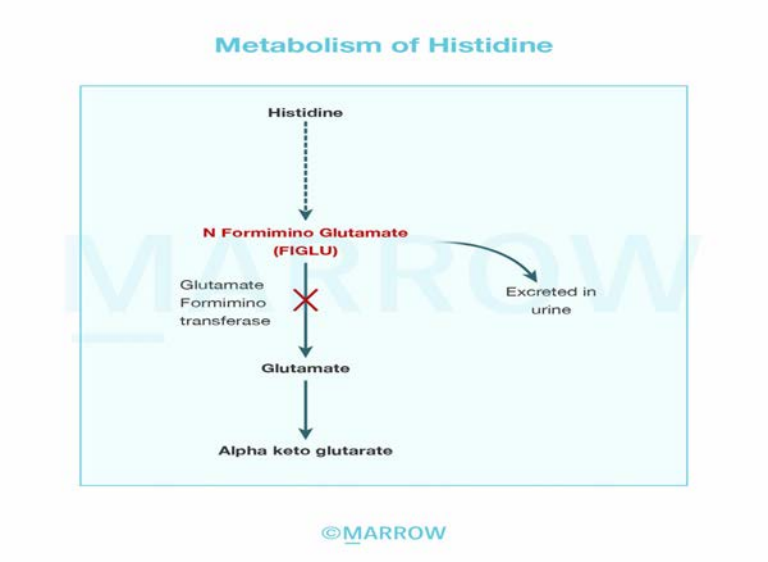
Solution to Question 17:

Primary hyperoxaluria is a defect in the metabolism of glycine.

The main defect in primary hyperoxaluria type I is the deficiency of the alanine glyoxalate aminotransferase in liver peroxisomes causing failure to catabolize glyoxylate formed by the deamination of glycine. This leads to a gross overproduction of oxalate, leading to the formation of oxalate stones and subsequent kidney damage.

Solution to Question 18:

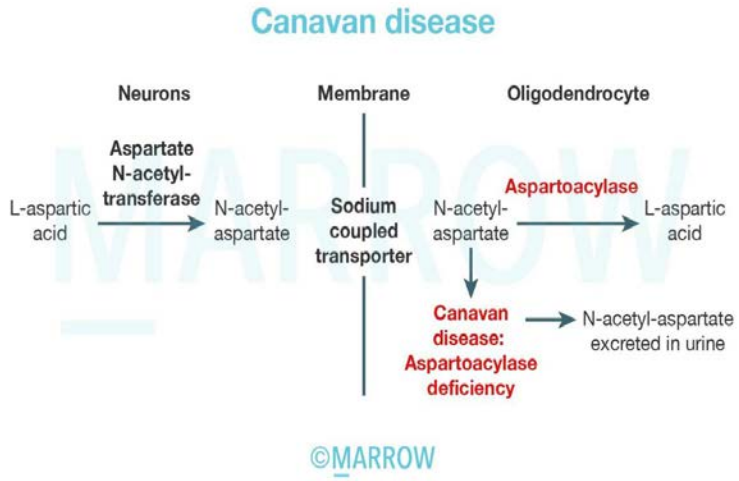
The histidine load test is done to identify folic acid deficiency. Formiminoglutamic acid (FIGLU) is excreted in urine, which is measured by the histidine load test.



Solution to Question 19:

Deficiency of aspartoacylase leads to Canavan disease. It is an autosomal-recessive disorder, which leads to excessive excretion of N-acetylaspartic acid in the urine.

It is characterized by leukodystrophy.



Solution to Question 20:

Maple syrup urine disease (MSUD) type I is due to impaired alpha-Ketoacid decarboxylase component of BCKAD (branched chain alpha keto acid dehydrogenase) complex, leading to defective decarboxylation reaction and the formation of branched-chain amino acids (leucine, isoleucine, and valine) and their toxic byproducts (keto-acids) in blood and urine.

A peculiar odor (burnt sugar) is found in urine, sweat, and cerumen.

Note: Amino-acids and disorders of their metabolism are invariably being asked in NEET as well as in AIIMS examinations.

Solution to Question 21:

Type II maple syrup urine disease (MSUD) results from a mutation in the genes encoding dihydrolipoyl transacylase.

There are 4 known types of Maple Syrup Urine Disease:

• Type IA and IB MSUD arise from mutations in the E1α and E1b genes, which encode the enzyme α-Ketoacid decarboxylase.

• Type II arises from mutations in the E2 gene, which encodes dihydrolipoyl transacylase.

• Type III arises from mutations in the E3 gene, which encodes Dihydrolipoamide dehydrogenase.

Solution to Question 22:

This infant with poor feeding and increased isovaleric acid in the urine likely has isovaleric aciduria. This condition is caused due to a defect in the metabolism of leucine.

Isovaleric aciduria is a condition characterized by vomiting, refusal to eat, listlessness, and a characteristic sweaty foot odor. The enzyme defect in this condition is isovaleryl Co-A dehydrogenase.

Solution to Question 23:

Plasma levels of ornithine are elevated in gyrate atrophy of the retina and choroid.

The initial reactions in arginine catabolism are the conversion of arginine to ornithine followed by the transamination of ornithine. Any mutations in ornithine aminotransferase lead to elevated plasma and urinary concentrations of ornithine.

This is associated with a condition known as gyrate atrophy of the choroid and retina. It presents with myopia and night blindness in adolescence which progresses to legal blindness by 50 years. The condition is treated with dietary restriction of arginine.

Solution to Question 24:

Segawa syndrome is a hereditary progressive dystonia due to the deficiency of tetrahydrobiopterin(BH4).

Segawa syndrome, also known as dopamine-responsive dystonia occurs due to a defect in the GTP cyclohydrolase I enzyme required for BH4 synthesis.

This condition is characterised by increased muscle tone and Parkinsonian features, typically absent in the morning or after rest but worsening during the day and with exertion. Treatment for this condition is with Levodopa.

Solution to Question 25:

Boiled cabbage odor in urine, sweat, and breath occurs in hypermethioninemia. Dimethylsulphide, a metabolite of methionine, causes the characteristic boiled cabbage smell.

People with a genetic deficiency of α/β-methionine adenosyltransferase present with hypermethioninemia and neural demyelination. However, they're usually asymptomatic until early adulthood.

Methionine adenosyltransferase converts methionine to S-adenosyl Methionine (SAM) - a principal methyl donor. Hence the enzyme deficincy causes excess accumulation of methionine and its broken down to metabolites.

Solution to Question 26:

A fishy odor is seen in trimethylaminuria. Consumption of food products with choline should be avoided in fish odor syndrome.

Fish odor syndrome (trimethylaminuria) is an autosomal recessive disease where TMA(trimethylamine) is not converted to the non-odorous TMAO (TMA-N-oxide) in the liver. This is due to the deficiency of flavin-containing mono oxygenase 3 (FMO3). TMA then builds up in the body and is released in sweat, urine, and breath, giving off a strong odor of rotting fish from the body.

Choline, lecithin, and TMAO are precursors of trimethylamine which are found in food products. These food products are eggs, liver, peanuts, meat, poultry, fish, dairy foods, pasta, and rice.

Solution to Question 27:

Urinary levels of 3-methylhistidine are decreased in patients with Wilson disease.

Amino acids in urine exist either in conjugated form (most) or in free form. In Wilson's disease the amount of conjugated amino acids far exceeds the amount of free amino acids. Methyl Histidine largely exists in free form and does not increase in amount on hydrolysis of acids in urine. Hence low amounts is seen in Wilson's disease.

Wilson disease is an autosomal-recessive disorder caused by mutation of the ATP7B gene located on chromosome 13. The ATP7B gene, encodes a transmembrane copper-transporting ATPase, expressed on the hepatocyte canalicular membrane.

Deficiency in the ATP7B protein causes a decrease in copper excretion into the bile, impairs its incorporation into ceruloplasmin, and inhibits ceruloplasmin secretion into the blood, leading to the accumulation of toxic levels of copper in the liver, brain, and eye.

Solution to Question 28:

Urine turning black on exposure to air is suggestive of alkaptonuria which occurs due to the defect in homogentisate oxidase enzyme.

Alkaptonuria is a disorder of tyrosine metabolism, that develops due to the defect in homogentisate oxidase, which oxidizes homogentisate to maleylacetoacetate. The defect in this enzyme leads to the accumulation of homogentisate, which is then excreted in the urine. The excreted homogentisate gets oxidized when exposed to the air and causes the urine to turn black.

In some cases, the excessive homogentisate may get oxidized to benzoquinone acetate, which binds to the connective tissue. This leads to arthritis and pigmentation of the connective tissue, a condition called ochronosis.

Other options:

Option B: Xanthine oxidase deficiency results in hypouricemia, xanthinuria, and xanthine lithiasis.

Option C: Defects in phenylalanine hydroxylase can lead to phenylketonuria characterised by hyperreflexia, seizures, hypopigmentation, low IQ, hyperactivity, and tremors.

Option D: Defects in dihydroorotate dehydrogenase are rarely seen in clinical practice. Miller syndrome is associated with this defect.

Protein structure and function

Question 1:

The primary structure of a protein refers to _____.

a) Linear structure and order of the amino acids present

b) Regular conformational forms of a protein

c) Complete three dimensional structure of the poly peptide units of a given protein

d) Subunit structure of the protein

Question 2:

Alpha helix and beta pleated sheet are examples of the _____ structure of a protein.

a) Primary

b) Secondary

c) Tertiary

d) Quarternary

Question 3:

Which of the following is false regarding alpha helix?

a) Stability of an alpha helix arises primarily from the peptide bond.

b) A complete turn of an alpha helix contains 3.6 amino acyl residues.

c) Pitch of the helix is 0.54 nm.

d) Large numbers of charged amino acids disrupt the helix.

Question 4:

What is the most common and stable conformation for a polypeptide chain?

a) Alpha helix

b) Beta pleated sheet

c) Turns and bends

d) Motifs

Question 5:

Which of the following amino acids do not participate in the formation of an alpha helix?

a) Leucine

b) Methionine

c) Proline

d) Lysine

Question 6:

All of the following types of interactions cooperate in stabilizing the tertiary structures of globular proteins except _____.

a) Disulphide bonds

b) Hydrogen bonds

c) Ionic interactions

d) Peptide bonds

Question 7:

Proteins are linear polymers of amino acids folded into compact structures. Sometimes, these folded structures associate to form homo- or hetero- dimers. Which of the following terms refers to this associated form?

a) Primary structure

b) Secondary structure

c) Tertiary structure

d) Quaternary structure

Question 8:

Which among the following is true about denaturation of proteins?

a) Biological property of the protein is retained.

b) Primary structure is lost.

c) It is always irreversible.

d) It mostly renders the protein insoluble.

Question 9:

What do chaperones assist in?

a) Ubiquitinization of proteins

b) Protein modification

c) Protein folding

d) Protein cleavage

Question 10:

All of the following are storage proteins except _____.

a) Myoglobin

b) Ovalbumin

c) Ferritin

d) Glutelin

Question 11:

Which of the following is a protein separation technique based on molecular/ ionic charge?

a) Affinity chromatography

b) SDS-PAGE

c) Absorption chromatography

d) Ion exchange chromatography

Question 12:

Which of the following protein separation techniques is based on the molecular size?

a) Agarose gel electrophoresis

b) Size exclusion chromatography

c) Ion exchange chromatography

d) Affinity chromatography

Question 13:

All of the following are methods used to detect the three dimensional structure of proteins except _____.

a) X-ray crystallography

b) NMR spectroscopy

c) Electrophoresis

d) Infra-red spectroscopy

Question 14:

Rossman fold-associated NADH domain is found in _____.

a) Pyruvate dehydrogenase

b) Lactate dehydrogenase

c) Acetyl CoA dehydrogenase

d) Isocitrate dehydrogenase

Question 15:

All of the following can determine protein structure except _____.

a) Mass spectrometry

b) X-ray crystallography

c) High-performance liquid chromatography

d) NMR spectroscopy

Question 16:

Where is quarter-staggered arrangement seen?

a) Immunoglobulin

b) Hemoglobin

c) Keratin

d) Collagen

Question 17:

Which of the following is not a protein-misfolding disease?

a) Prion disease

b) Alzheimer's disease

c) Beta thalassemia

d) Ehlers–Danlos syndrome

Question 18:

Which of the following is not a conjugated protein?

a) Albumin and globulin

b) Casein and vitellin

c) Hemoglobin

d) Carbonic anhydrase

Question 19:

Which among the following is not a transport protein?

a) Insulin

b) Myoglobin

c) Hemoglobin

d) Albumin

Question 20:

All are true about collagen except that _____

a) It is the most abundant protein in the body.

b) Type IV collagen is found in basement membrane

c) Every third amino acid is alanine

d) Procollagen is formed inside the fibroblast

Question 21:

An infant is brought to the pediatric OPD with clinical findings as shown in the image. Further examination shows that the child has a loss of developmental milestones, truncal hypotonia, and failure to thrive. Which of the following enzymes is defective in this child?

a) Branched chain keto-acid decarboxylase complex

b) Prolyl hydroxylase

c) Lysyl oxidase

d) Cystathionine synthase

Question 22:

Which among the following will be reduced in a patient with chronic inflammation?

a) C-reactive protein

b) Fibrinogen

c) Transferrin

d) Ceruloplasmin

Question 23:

Which of the following is present in glycoproteins?

a) Monosaccharide

b) Oligosaccharide

c) Polysaccharide

d) Disaccharide

Question 24:

A postmenopausal woman presents to her gynaecologist for her annual check-up. An ovarian cyst was noted on ultrasound. Histopathology revealed a mucinous cystadenoma. Which test can be used on the sample to detect the presence of mucin?

a) Periodic acid–Schiff reaction

b) Sepharose-lectin column chromatography

c) Mass spectrometry

d) Masson trichrome stain

Question 25:

What is the site of synthesis of O–linked glycoproteins?

a) Endoplasmic reticulum

b) Mitochondria

c) Golgi apparatus

d) Cytoplasm

Question 26:

What type of glycoprotein is collagen?

a) O-linkage

b) N-linkage

c) GPI linkage

d) None

Question 27:

Which of the following types of collagens is involved in wound healing?

a) Type II

b) Type III

c) Type IV

d) Type V

Question 28:

Which of the following is caused by the lack of a specific lysosomal hydrolase for glycoproteins?

a) Oligosaccharidoses

b) I cell disease

c) Scurvy

d) Ehlers-Danlos syndrome

Question 29:

Which of the following disease is associated with defective glycosyl-phosphatidylinositol (GPI)?

a) Paroxysmal nocturnal hemoglobinuria

b) Oligosaccharidoses

c) Ehlers–Danlos syndrome

d) I-cell disease

Question 30:

Which of the following features will not be seen in a patient with I-cell disease?

a) Coarse facial features

b) Corneal clouding

c) Failure to thrive

d) Buphthalmos

Question 31:

Which of the following would have no effect on the function of the protein product?

a) Glutamine replaced by asparagine

b) Glutamine replaced by alanine

c) Glutamine replaced by glutamate

d) Glutamine replaced by arginine

Question 32:

What is the function of the proteasome?

a) Protein folding

b) Post-translational modification

c) Protein degradation

d) Protein sorting

Question 33:

Which of the following is true about the structural organization of proteins?

a) Tertiary structure is three dimensional

b) Primary, secondary and tertiary structures are destroyed by denaturation

c) Secondary structure is stabilised by disulphide bonds

d) Secondary and tertiary structures depend upon amino acid sequence

Answer Key

Question No.	Correct Option
1	a
2	b
3	a
4	a
5	c
6	d
7	d
8	d
9	c

10	a
11	d
12	b
13	c
14	b
15	c
16	d
17	d
18	a
19	a
20	c
21	c
22	c
23	b
24	a
25	c
26	a
27	b
28	a
29	a
30	d
31	a
32	c
33	a

Detailed Explanations

Solution to Question 1:

The primary structure of a protein refers to the linear structure and order of the amino acids present.

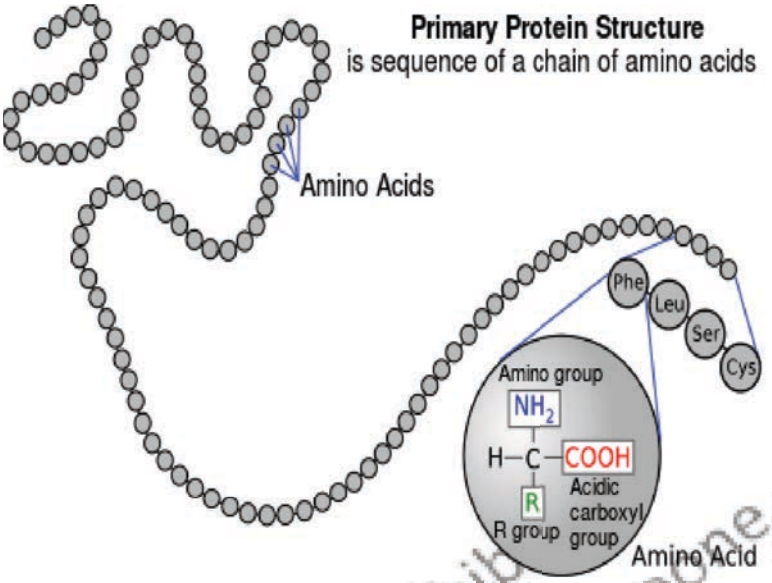
There are four known levels of protein structure:

• Primary: A linear sequence of amino acids in the polypeptide chain

• Secondary: Folded and geometrically ordered units of short segments of polypeptides. Eg: Alpha helix, beta-pleated sheets, loops, and bends

• Tertiary: Secondary structures arranged into larger functional units

• Quaternary: Different types of polypeptides arranged in a spatial manner



Solution to Question 2:

Alpha helix and beta-pleated sheets are examples of the secondary structure of a protein.

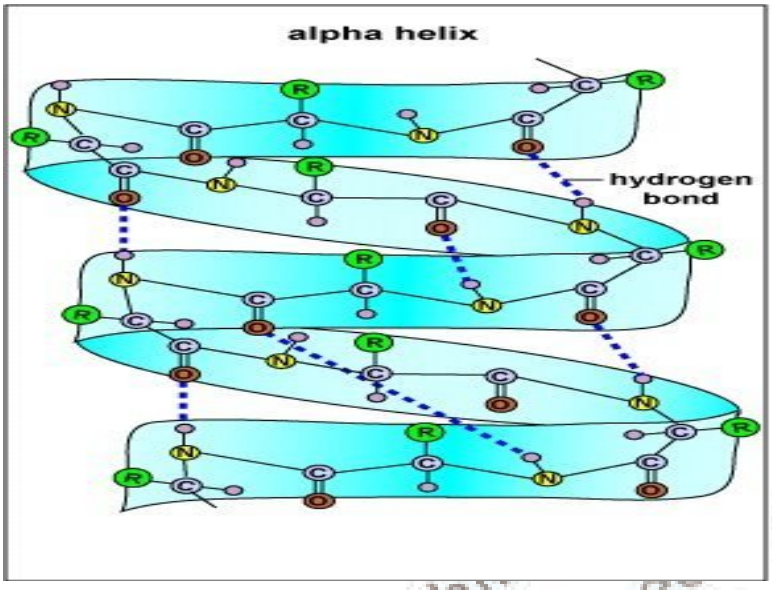
The secondary structure of a protein refers to the folded and geometrically ordered units of short segments of a polypeptide chain. Examples include alpha helix, beta sheets, turns and bends.

Solution to Question 3:

Alpha helix is stabilized primarily by intra-chain hydrogen bonds.

Alpha helix is the most common and stable secondary structure of protein. It is a rigid, right-handed spiral. A complete turn of the helix contains an average of 3.6 aminoacyl residues. The rise per turn of the helix (pitch) is 0.54nm.

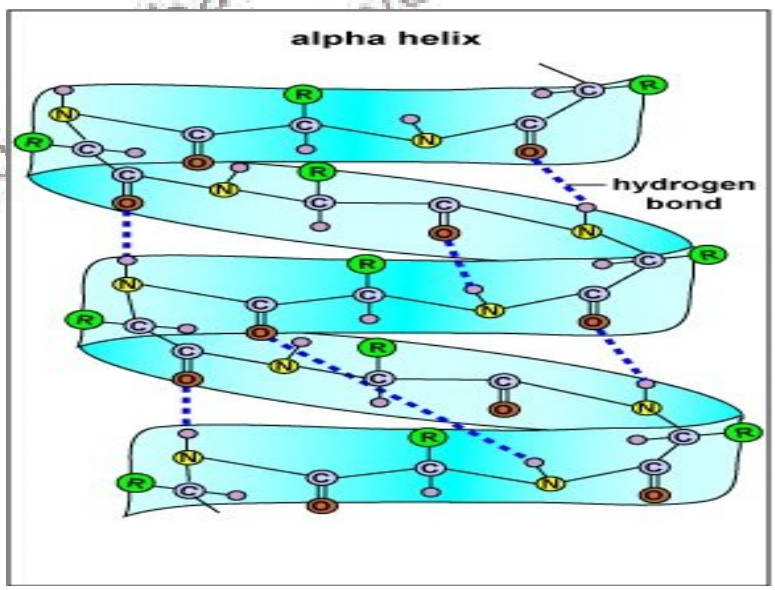
Large numbers of charged amino acids (glutamate, aspartate, histidine, lysine, and arginine) disrupt the helix by forming ionic bonds or by electrostatically repelling each other.



Solution to Question 4:

The most common and stable conformation for a polypeptide chain is alpha helix.

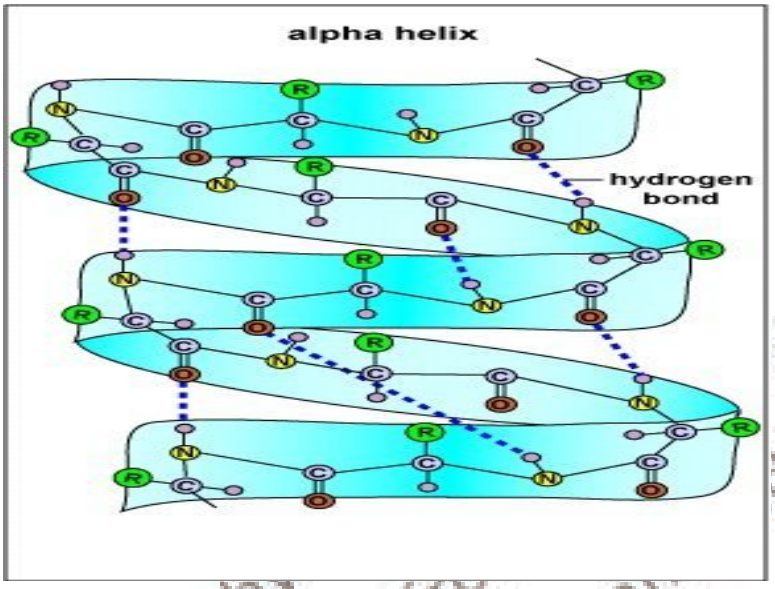
The alpha helix has high stability due to the presence of a large number of hydrogen bonds and van der Waals interactions between the amino acids. Hemoglobin and myoglobin are proteins whose major secondary structure is the alpha helix.



Solution to Question 5:

Proline does not participate in the formation of an alpha helix.

The stability of an alpha helix arises primarily from hydrogen bonds formed between the oxygen of the peptide bond carbonyl and the hydrogen atom of the peptide bond nitrogen of the fourth residue down the polypeptide chain. Since the peptide bond nitrogen of proline lacks a hydrogen atom, it is incapable of forming a hydrogen bond with carbonyl oxygen.



Solution to Question 6:

Peptide bonds are not involved in stabilizing tertiary structure of proteins. It forms the primary structure of proteins.

Interactions stabilizing tertiary structure of proteins are as follows:

• Hydrophobic interactions

• Hydrogen bonds

• Ionic interactions

• Disulphide bonds

Note:

Hydrogen bonds and Van der Waal interactions are involved in the formation of α helix (secondary structure).

Solution to Question 7:

Quaternary structure refers to the arrangement of multiple folded polypeptides that aggregate into homo- or hetero-dimers.

Homo-dimers (e.g., D-amino acid aminotransferase) contain two copies of the same polypeptide chain, while hetero-dimers (e.g., Rab geranylgeranly transferase) contain different polypeptides.

Solution to Question 8:

Denaturation mostly renders the protein insoluble.

Protein denaturation results in the unfolding and disorganization of the secondary and tertiary structures of protein. Thus, the biological property of the protein is lost. However, there is no hydrolysis of peptide bonds, which form the primary structure.

Denatured proteins are often insoluble and precipitate from the solution. This process may be reversible under ideal conditions, such that the protein refolds into its original structure when the denaturing agent is removed.

Solution to Question 9:

Chaperones assist in protein folding.

Chaperones are heat shock proteins. They bind to the short sequences of hydrophobic amino acids of an extended polypeptide. Thus, they shield the new polypeptide being synthesized from aggregating. This provides the polypeptide an opportunity to form an appropriate secondary structure.

When a protein is thermodynamically trapped in a misfolded dead-end form, chaperones can rescue the protein by unfolding their hydrophobic ends. Thus, they provide the protein a second chance to fold productively.

Solution to Question 10:

Myoglobin binds oxygen in muscles and is not a storage protein.

Proteins that act as a storehouse of amino acids and metal ions are called storage proteins. They are as follows:

- Ferritin
- Ovalbumin of egg white
- Glutelin of wheat
- Gliadin of wheat
- Vitellin of egg yolk
- Oryzenin of rice
- Milk casein

Solution to Question 11:

Ion exchange chromatography is a protein separation technique based on molecular/ ionic charge.

Other techniques based on molecular charge are:

- Agarose gel electrophoresis
- Cellulose acetate electrophoresis
- Isoelectric focusing
- High-performance liquid chromatography (HPLC)

Solution to Question 12:

Size-exclusion chromatography is a separation technique that separates solutes on the basis of the molecular size.

Solution to Question 13:

Electrophoresis is not used to detect the three-dimensional structure of proteins.

The techniques used to determine the three-dimensional structures of proteins are as follows:

- X-Ray crystallography
- NMR spectroscopy
- Infra-red spectroscopy
- Polychromatic X-ray crystallography
- UV light spectroscopy
- Optical rotatory dispersion
- Circular dichromism
- Computer-based modelling
- Mass spectrometry

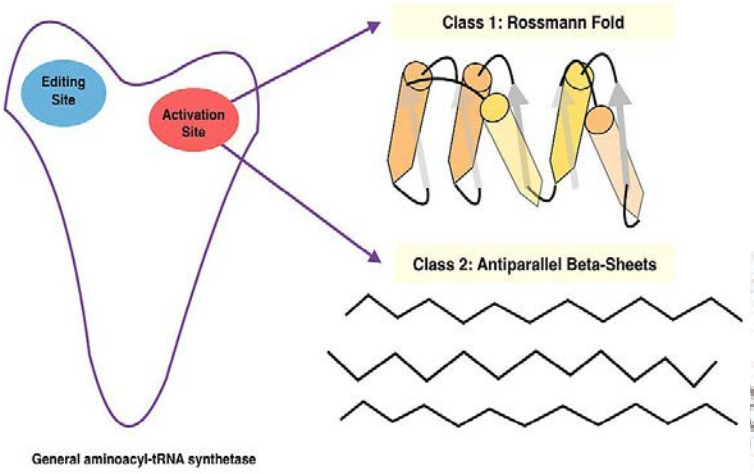
Solution to Question 14:

Rossmann fold-associated NADH domain is found in lactate dehydrogenase.

Rossmann fold is an N-terminal NAD(P)+ -binding domain. It is seen in:

- Lactate dehydrogenase
- Alcohol dehydrogenase
- Glyceraldehyde-3-phosphate dehydrogenase
- Malate dehydrogenase

- D-glycerate dehydrogenase
- 6-phosphogluconate dehydrogenase
- Formate dehydrogenase



Solution to Question 15:

High-performance liquid chromatography cannot determine protein structure. It is used for separation, purification, and identification of proteins.

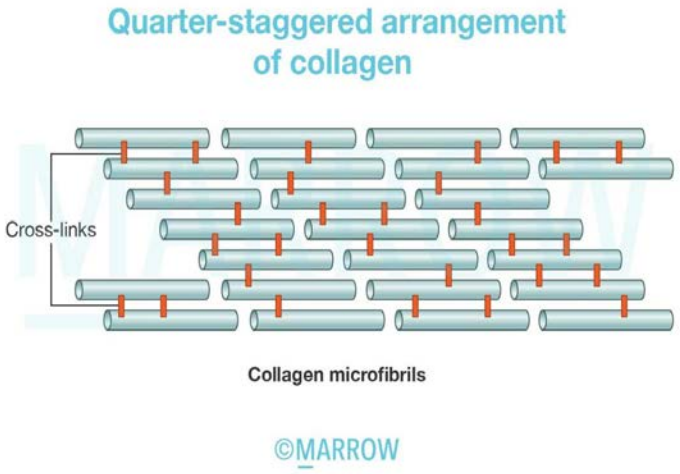
Given below are the various methods to determine protein structure:

- X-Ray crystallography
- NMR spectroscopy
- Mass spectrometry
- Infra-red spectroscopy
- Polychromatic X-ray crystallography
- UV light spectroscopy
- Optical rotatory dispersion
- Circular dichromism
- Computer-based modeling

Solution to Question 16:

The collagen fibers have a triple-stranded, quarter-staggered arrangement.

In a collagen fibre, tropocollagen molecules are arranged side-by-side. Each fibril is displaced longitudinally from its neighbour by less than one-quarter of its length. This is known as a "quarter-staggered array". This arrangement helps in mineralization.



Solution to Question 17:

Ehlers-Danlos syndrome is not a protein misfolding disease.

Ehlers–Danlos syndrome is a group of connective tissue disorders resulting in mobile joints and skin abnormalities. It is due to mutation in the genes encoding α collagen 1, procollagen N-peptidase and lysyl hydroxylase.

The following are examples of protein misfolding diseases:

• Prion disease: Creutzfeldt–Jakob disease, bovine spongiform encephalopathy, scrapie disease of sheep.

• Alzheimer's disease

• Beta thalassemia

Solution to Question 18:

Albumin and globulin are not conjugated proteins

Based on composition, proteins can be classified into:

1. Simple proteins: They contain only amino acids.

• Globular: Albumin, Globulin, histones

• Fibrous: Collagen, Elastin, Fibrin

2. Conjugated proteins: These contain a non-protein part (prosthetic group).

• Phosphoproteins: Casein, Vitellin

• Metalloproteins: Hemoglobin, Carbonic anhydrase

• Glycoproteins: Blood group antigens

• Nucleoproteins

• Lipoproteins

• Mucoproteins

3. Derived proteins: These are degradation products of native proteins.

• Primary: Coagulated proteins

• Secondary: Peptides

Solution to Question 19:

Insulin is not a transport protein.

Insulin is a regulatory protein (hormone) secreted by the beta cells of the pancreas. It promotes anabolism.

The following are examples of transport proteins:

• Hemoglobin transports oxygen from the lungs to the capillaries of the tissues. It is found exclusively in red blood cells

• Myoglobin functions both as a reservoir of oxygen and as an oxygen carrier. It increases the rate of transport of oxygen within the muscle cell. It is present in heart and skeletal muscle

• Albumin is a plasma protein that transports cations, fatty acids, bilirubin, etc. It also regulates the plasma oncotic pressure.

Solution to Question 20:

Every third amino acid in collagen is glycine.

Collagen is the most abundant protein in the body. The major collagen in the body is type I, in which every third amino acid is glycine. It is found in noncartilaginous connective tissues, including bone, tendon, skin. Type IV collagen is found in the basement membrane.

Collagen is synthesized intracelluarly by fibroblasts as a large precursor called procollagen. The procollagen is then cleaved extracellularly by specific peptidases to form tropocollagen.

Solution to Question 21:

The given image reveals an infant with sparse and kinky hair. This, along with loss of developmental milestones and truncal hypotonia, strongly suggests a diagnosis of Menkes disease,

which is caused by defective lysyl oxidase.

Menkes disease results from a deficiency of lysyl oxidase co-factor i.e copper. This is due to a defect in the ATP7A gene, which prevents the absorption of copper from the intestine.

Note :

• ATP7A mutation: Menkes disease - X-linked recessive

• ATP7B mutation: Wilson's disease - Autosomal recessive

Solution to Question 22:

Transferrin will be reduced in a patient with chronic inflammation as it is a negative acute phase reactant.

Acute-phase reactants are proteins that increase or decrease in levels in response to inflammatory and neoplastic conditions.

Positive acute-phase reactants	Negative acute-phase reactants
C-reactive proteinHaptoglobulinFibrinogenCeruloplasminAlpha 1 antiproteaseMannose-binding proteinComplement factorsFerritinSerum amyloid A	AlbuminTransthyretinRetinol-binding proteinTransferrinAntithrombinTranscortin

Solution to Question 23:

Oligosaccharides are present in glycoproteins.

Glycoproteins are proteins that contain oligosaccharide chains covalently bound to amino acids. The oligosaccharide (carbohydrate) is attached to the protein by cotranslational or posttranslational modification. This process is known as glycosylation. Some examples include collagens, mucin, antibodies and hormones like erythropoietin and follicle-stimulating hormone (FSH).

Solution to Question 24:

Periodic acid–Schiff (PAS) reagent helps to detect mucin which is a glycoprotein.

PAS is a staining method used to detect polysaccharides such as glycogen, and mucosubstances such as glycoproteins, glycolipids, and mucins in tissues.

PAS staining can be used to facilitate the diagnosis of several medical conditions:

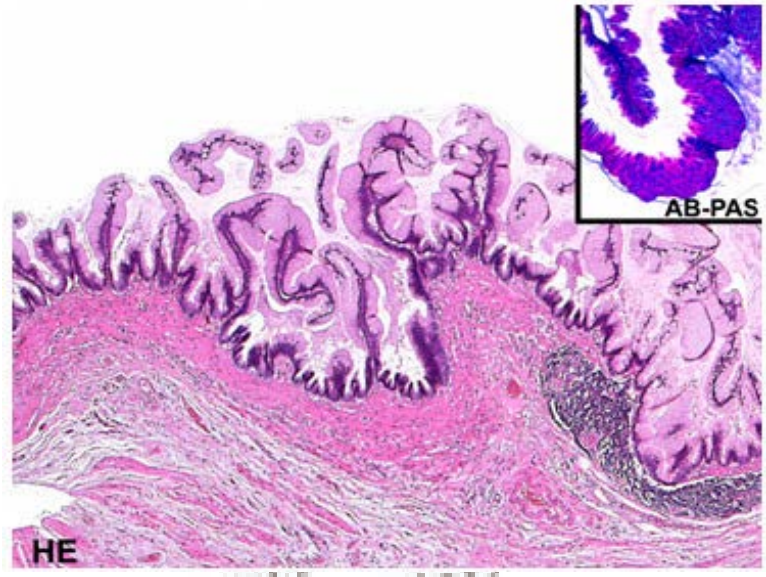
• Glycogen storage diseases (versus other storage disorders)

• Adenocarcinomas, which often secrete neutral mucins

• Staining of macrophages in Whipple's disease

• A1-antitrypsin deficiency if periportal liver hepatocytes stain positive

The image shows mucinous cystadenoma in HE (haematoxylin-eosin) stain and a positive PAS reaction.



Solution to Question 25:

O–linked glycoproteins are synthesized in the Golgi apparatus by the sequential transfer of sugars from their nucleotide carriers to the protein.

O-linked glycoproteins are proteins attached to a sugar (eg: N-acetylgalactosamine) through an O-glycosidic linkage between the hydroxyl side chain of serine/threonine and the sugar. Examples of O-linked glycoproteins include mucins, proteoglycans and collagens.

Solution to Question 26:

Collagen comes under glycoprotein sub-classes of O-glycosidic linkages.

Glycoproteins can be divided into three main groups:

• O-linked: They contain an O-glycosidic linkage. Examples include mucin, proteoglycans, collagens, nuclear and cytoplasmic proteins.

• N-linked: They contain an N-glycosidic linkage. The types of N-glycosidic linkage include complex, hybrid and high-mannose types.

• Glycosylphosphatidylinositol-anchored (GPI-anchored): Examples include acetylcholinesterase, alkaline phosphatase, decay-accelerating factor, 5′-Nucleotidase, thy-1 antigen and variable surface glycoprotein (Trypanosoma brucei).

Solution to Question 27:

Type III collagen is involved in wound healing.

Collagen types I and III predominate in the skin and aponeurotic layers. In the early stages of wound healing, type III collagen is synthesized. It is replaced by type I collagen as wound healing progresses.

The tensile strength of the healing wound is directly proportional to the amount and type of collagen present. During healing, as disorganized collagen is degraded and reformed, covalent cross-links are formed that enhance tensile strength. The maximum strength of the healed wound depends upon the interconnection of collagen subunits. Approximately 80 percent of the original strength of the tissue is obtained by six weeks after surgery. Wounds slowly continue to get stronger but may never achieve 100 percent of their previous strength.

Solution to Question 28:

Lack of a specific lysosomal hydrolase for glycoproteins cause a group of very rare, autosomal recessive genetic diseases called glycoprotein storage diseases (oligosaccharidoses).

Lysosomal acid hydrolases are specific for the removal of the oligosaccharide chains of glycoproteins. They are primarily exoenzymes that remove their respective groups in sequence in the reverse order of their incorporation (“last on, first off”). When one acid hydrolase is deficient, there is accumulation of partially degraded glycoproteins in the lysosomes. This results in oligosaccharidoses.

Solution to Question 29:

Paroxysmal nocturnal hemoglobinuria results from mutation of glycosylphosphatidylinositol (GPI)

GPI is necessary for the attachment of decay-accelerating factor (DAF) (CD55) and CD59 to the RBC membrane. Because of the absence of DAF when GPI is mutated, the complement system attacks the RBCs resulting in hemolysis. Patients present with symptoms of:

• Anemia

• Esophageal spasm

• Erectile dysfunction

• Abdominal pain

• Pulmonary hypertension

Treatment is with eculizumab, which is a terminal complement inhibitor.

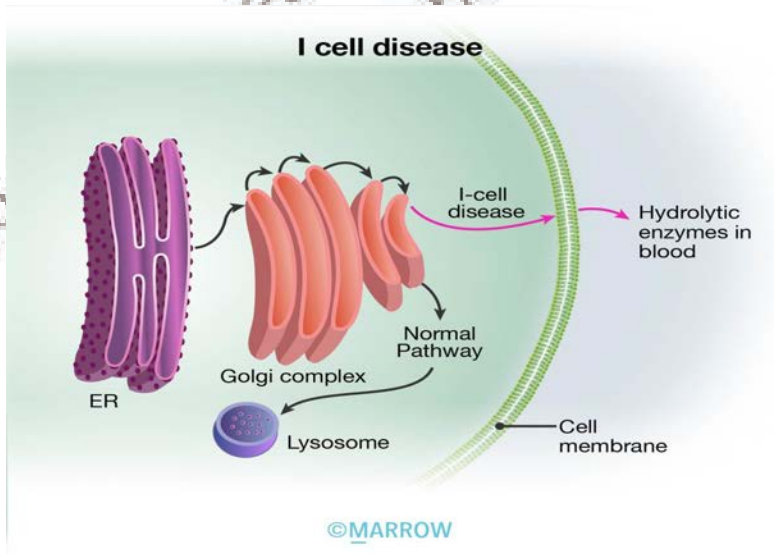
Solution to Question 30:

Buphthalmos is not a feature of I-cell disease.

I-cell disease is an inherited lysosomal storage disorder caused by a defect in the Golgi enzyme N-acetylglucosamine-1-phosphotransferase. This results in failure of the Golgi to phosphorylate mannose residues (↓mannose-6-phosphate) on glycoproteins. Hence, proteins are secreted extracellularly instead of being delivered to lysosomes. Lysosomes lack most normal enzymes and thus accumulate undegraded molecules, forming inclusion bodies.

It is characterized by the following:

- Coarse facial features
- Gingival hyperplasia
- Clouded corneas
- Restricted joint movements
- Claw hand deformities
- Kyphoscoliosis
- High plasma levels of lysosomal enzymes



Solution to Question 31:

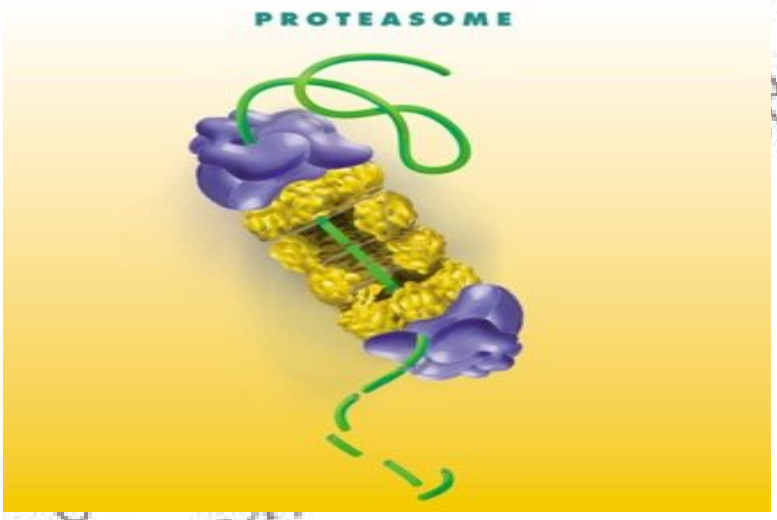
When glutamine is replaced by asparagine, there is no change in the function of the protein product.

This is due to the concept of conservative or homologous replacement of amino acids. Asparagine and glutamine are both uncharged polar (hydrophilic) amino acids. Thus, when glutamine is replaced with asparagine there is no change in the polarity or the charge of the peptide/protein.

Solution to Question 32:

The proteasome takes part in protein degradation.

The proteasome is a cylindrical structure in the cytosol that degrades polyubiquitinated proteins. It has a central core with proteolytic enzymes. Proteins that are tagged by ubiquitin to be degraded enter the core through regulatory rings on either side of the cylinder.



Clinical relevance: It plays an important role in antigen presentation to T lymphocytes as it degrades viruses and other molecules to small peptides.

Solution to Question 33:

The tertiary structure is three dimensional is the right answer among the given options.

Proteins are structurally organised into four levels, as follows:

• Primary: The linear sequence of amino acids in the polypeptide chain

• Secondary: Folded and geometrically ordered units of short segments of polypeptides. Eg: alpha helix, beta-pleated sheets, loops, and bends

• Tertiary: Secondary structures arranged into larger functional units called domains

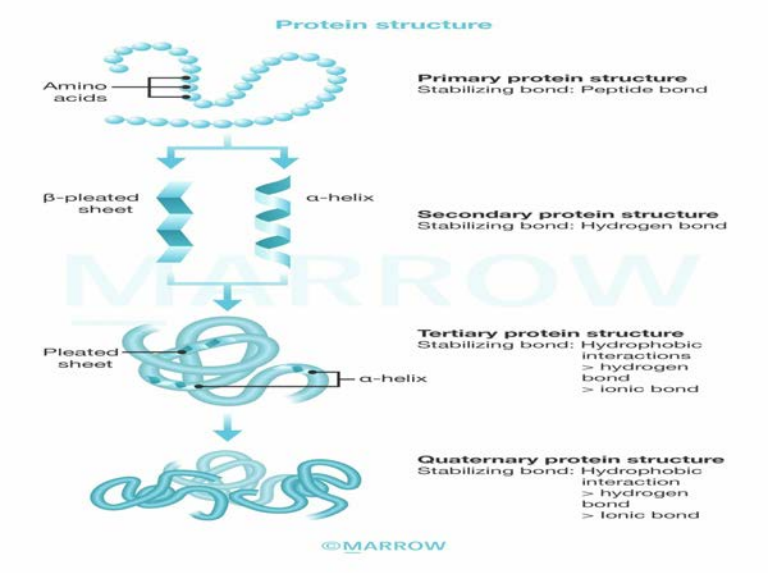
• Quaternary: Different types of polypeptides arranged in a spatial manner

Other options:

Option B: Protein denaturation results in the unfolding and disorganization of the secondary and tertiary structures without the hydrolysis of peptide bonds, which form the primary structure.

Option C: Hydrogen bond and Van der Waal interactions are involved in the stabilization of secondary structure(helix). Disulfide bonds stabilize the tertiary structure of proteins.

Option D: Primary structure depends upon the amino acid sequence, not secondary or tertiary.



Urea cycle and its disorders

Question 1:

Where does the ornithine cycle take place?

a) Cytoplasm

b) Mitochondria

c) Cytoplasm and mitochondria

d) Cytoplasm and nucleus

Question 2:

Which of the following allosteric activators are deficient in a patient with no activity of carbamoyl phosphate synthetase I?

a) Alanine

b) N-Acetyl glutamate

c) Ornithine

d) Citrulline

Question 3:

A 3-month-old child presented with vomiting, irritability, and lethargy. He was then diagnosed with a urea cycle disorder that is inherited as an X-linked recessive disorder. What is the likely diagnosis?

a) Citrullinemia

b) Hyperammonemia type I

c) Hyperammonemia type II

d) Argininosuccinic aciduria

Question 4:

An infant was admitted with failure to thrive. His reports show hypoglycemia, high glutamine, and uracil levels in urine. Gastric tube feeding was not tolerated and the child

became comatose. Parenteral dextrose was given after which the child recovered in 24 hours. What is the likely enzyme defect?

a) Carbamoyl phosphate synthase I

b) Ornithine transcarbamoylase

c) Argininosuccinate synthase

d) Argininosuccinate lyase

Question 5:

Deficiency of arginosuccinate synthetase results in _____.

a) Hyperargininemia

b) Argininosuccinic aciduria

c) Citrullinemia

d) Hyperammonemia type 1

Question 6:

The two nitrogen atoms in urea are derived from which of the following?

a) Ammonia and Glutamate

b) Ammonia and Aspartate

c) Glutamine and Arginine

d) Aspartate and Arginine

Question 7:

Urea cycle and Krebs cycle are linked at____.

a) Arginine

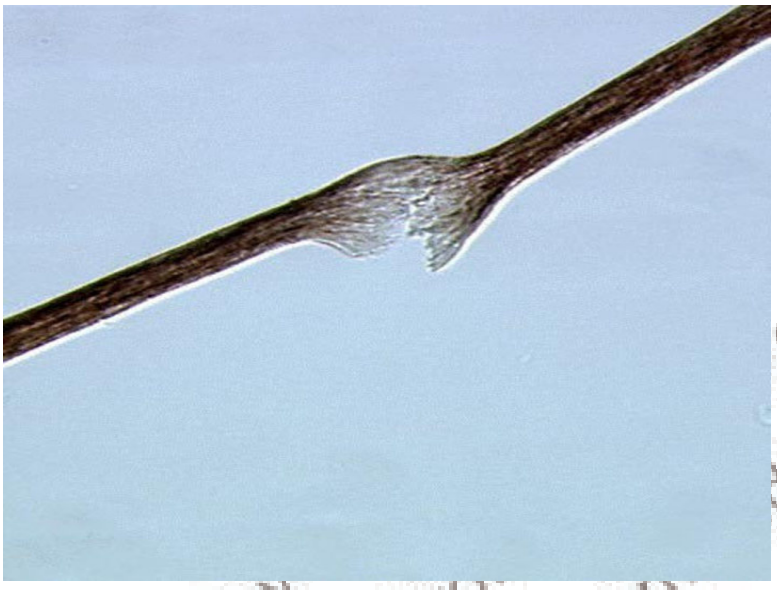
b) Ornithine

c) Fumarate

d) Oxaloacetate

Question 8:

A neonate was brought to the emergency room with lethargy and failure to thrive. A microscopic image of his hair strand is shown below. Which of the given enzymes may be deficient in this patient?



a) Arginosuccinate lyase

b) Arginase

c) Fumarase

d) Tyrosinase

Question 9:

A neonate was brought with tachypnoea and feeding difficulties 2 days after birth. Later he developed focal seizures that progressed to coma. The plasma ammonia level is 300 µg/dL. The argininosuccinate level is high but the arginine level is low. What is the enzyme deficiency?

a) Argininosuccinate synthetase

b) Argininosuccinate lyase

c) Arginase

d) Carbamoyl phosphate synthetase

Question 10:

At which step of the urea cycle does hydrolysis occur?

a) Cleavage of arginine

b) Formation of arginosuccinate

c) Formation of citrulline

d) Cleavage of arginosuccinate

Question 11:

A 3-day-old child developed lethargy, poor feeding, and hypotonia. An inborn error of metabolism (IEM) panel was sent. Reports reveal hyperammonemia, hyperornithinemia, and homocitrullinuria. What is the most likely defect?

a) Citrin defect

b) Ornithine transporter defect

c) Carnitine acyl transferase defect

d) Ornithine permease

Question 12:

In urea cycle disorders, which of the following can be used to reduce the levels of ammonia?

a) Glutamate

b) Phenylbutyrate

c) Isoleucine

d) L-carnitine

Question 13:

All are enzymes involved in the urea cycle except_____.

a) Carbamoyl phosphate synthase -ll

b) Ornithine transcarbamoylase

c) Argininosuccinate Lyase

d) Arginase

Question 14:

In which of the following biological processes is carbamoyl phosphate synthetase I used?

a) Urea cycle

b) Purine synthesis

c) Pyrimidine synthesis

d) Glucuronic pathway

Answer Key

Question No.	Correct Option
1	c
2	b
3	c
4	b
5	c
6	b
7	c
8	a
9	b
10	a
11	b
12	b
13	a
14	a

Detailed Explanations

Solution to Question 1:

Ornithine cycle/Krebs–Henseleit cycle/Urea cycle occurs in both the cytoplasm and mitochondria.

The reactions that take place both in the cytoplasm and mitochondria are:

- H: Heme synthesis
- U: Urea cycle
- G: Gluconeogenesis
- Pathway: Pyrimidine synthesis

Solution to Question 2:

In the given scenario, the patient is likely to be deficient in N-acetyl glutamate. N-Acetyl glutamate is the allosteric activator of carbamoyl phosphate synthetase I, the rate-limiting enzyme of the urea cycle.

Solution to Question 3:

This child with vomiting, lethargy, and irritability with an X-linked recessive urea cycle disorder most likely hyperammonemia type II

Hyperammonemia type II is due to the deficiency of the enzyme ornithine transcarbamoylase. It is the most frequent urea cycle defect. Biochemical findings include elevated levels of glutamine in the blood, urine, CSF, low citrulline, and high urinary orotic acid. This condition may be precipitated by a switch from formula to whole milk.

Urea cycle disorders are all characterized by hyperammonemia, encephalopathy, and respiratory alkalosis. There is an accumulation of the precursors of urea - ammonia, and glutamine. They include:

• Defect in carbamoyl phosphate synthetase I - Hyperammonemia type 1

• Defect in ornithine transcarbamoylase - Hyperammonemia type 2

• Defect in argininosuccinate synthetase - Citrullinemia

• Defect in argininosuccinate lyase- Argininosuccinic aciduria

• Defect in the gene for arginase- Hyperargininemia

Clinical features of urea cycle disorders are common to all and include vomiting, avoidance of high-protein foods, intermittent ataxia, irritability, lethargy, and severe mental retardation.

Note: Classically, Type 2 Hyperammonemia is an X- linked recessive disorder. But in certain heterozygous females (with one mutated gene), the disease manifests and they necessarily do not remain as carriers. In them, the disease follows an X-linked partially dominant pattern of inheritance.

Solution to Question 4:

This infant with failure to thrive, hypoglycemia, elevated levels of glutamine and uracil levels most likely has hyperammonemia type 2. This condition occurs due to the deficiency of the enzyme ornithine transcarbamoylase.

There is an accumulation of carbamoyl phosphate, which then enters into pyrimidine synthesis producing orotic acid and uracil and leading to orotic aciduria and high uracil in blood and urine. Since the ammonia from glutamine is not converted into urea, its level increases. Glutamate is not regenerated. Since glutamate is not available for transamination, amino acids

cannot be used for gluconeogenesis. Hence, the glucose level drops in the blood.

Treatment:

• Reducing intake of ammonia - Dietary protein, a metabolic source of ammonium, is restricted and caloric intake is provided by glucose and fat.

• Increasing excretion of ammonia - Addition of phenylbutyrate, which scavenges nitrogen by combining with amino acids to form a more water-soluble product. This can be easily excreted by the kidneys.

Solution to Question 5:

Deficiency of arginosuccinate synthetase results in citrullinemia.

Other options:

Option A: Hyperargininemia is due to a deficiency of arginase.

Option B: Argininosuccinic aciduria is due to a deficiency of argininosuccinate lyase.

Option D: Hyperammonemia type 1 due to a deficiency of carbamoyl phosphate synthetase I.

Solution to Question 6:

The two nitrogen atoms in urea are derived from ammonia and aspartate.

Solution to Question 7:

The urea cycle and Kreb's cycle are linked at fumarate.

In the urea cycle, cleavage of argininosuccinate to arginine releases the aspartate skeleton as fumarate. Fumarate in Kreb's cycle, by the addition of water, converts to L-malate.

Solution to Question 8:

The given microscopy image shows trichorrhexis nodosa and longitudinal breaks and is seen in argininosuccinic aciduria. This occurs due to a defect in the enzyme argininosuccinate lyase.

Trichorrhexis nodosa is a defect in the hair shaft characterized by thickening or weak points (nodes) that cause the hair to break off easily. It can also be caused by other disorders such as hypothyroidism, iron deficiency and Menkes' kinky hair syndrome

Solution to Question 9:

This infant with feeding difficulty, tachypnea and seizures, combined with an elevated arginosuccinate level and low arginine levels most likely has argininosuccinic aciduria. This is associated with an argininosuccinate lyase deficiency.

This condition is accompanied by high levels of argininosuccinate levels in blood, CSF, and urine. Like other classical urea cycle disorders, it presents 1-4 days after birth with lethargy and failure to thrive that progresses to coma and even death. It is also associated with friable, tufted hair known as trichorrhexis nodosa.

Diagnosis is by measurement of erythrocyte argininosuccinate lyase activity and can be performed on umbilical cord blood or amniotic fluid.

Solution to Question 10:

Hydrolysis occurs during the cleavage of arginine to form urea and ornithine in the urea cycle. It is catalyzed by the enzyme arginase.

Clinical correlation:

• Deficiency of the enzyme arginase results in hyperargininemia.

• Levels of arginine are elevated in both blood and cerebrospinal fluid (CSF).

Solution to Question 11:

This child with lethargy, poor feeding, and labs showing hyperammonemia, hyperornithinemia, and homocitrullinuria most likely has hyperammonemia, hyperornithinemia and homocitrullinuria syndrome (HHH syndrome). This condition is due to an ornithine transporter defect.

Solution to Question 12:

Phenylbutyrate can be used to reduce ammonia levels in patients with urea cycle disorders.

Phenylbutyrate diverts urea into an alternate pathway and reduces the amount of ammonia. Hence, acting as an ammonia scavenger. Other drugs that can be used to achieve the same outcome (ammonia scavengers) include phenylacetate and benzoate.

Note: Arginine is used as first-line treatment as it provides ornithine; a positive regulator of the urea cycle. Arginine also activates NAG synthase which increases NAG; a positive regulator of the urea cycle

Solution to Question 13:

Carbamoyl phosphate synthetase-ll is a cytoplasmic enzyme, which is involved in pyrimidine nucleotide synthesis.

Carbamoyl phosphate synthetase-l is a mitochondrial enzyme, which catalyzes the rate-limiting step of the urea cycle.

Solution to Question 14:

Carbamoyl phosphate synthetase I is a mitochondrial enzyme that catalyzes the rate-limiting step of the urea cycle.

Carbamoyl phosphate synthetase II is a cytoplasmic enzyme that catalyzes the synthesis of pyrimidine.

Lipids: Basics

Question 1:

Which of the following is a derived lipid?

a) Fats

b) Oils

c) Steroids

d) Waxes

Question 2:

Which of the following serve as the main storage form of lipids in our body?

a) Fatty acids

b) Triacylglycerols

c) Phospholipids

d) Lipoproteins

Question 3:

Which of the following is not a saturated fatty acid?

a) Linoleic acid

b) Caproic acid

c) Myristic acid

d) Palmitic acid

Question 4:

Which of the following is not an example of monounsaturated fatty acid?

a) Palmitoleic acid

b) Oleic acid

c) Elaidic acid

d) Linoleic acid

Question 5:

What is the total number of double bonds present in arachidonic acid?

a) 1

b) 2

c) 3

d) 4

Question 6:

Which of the following are known to possess anti-inflammatory activities?

a) Omega 3 fatty acids

b) Omega 6 fatty acids

c) Omega 9 fatty acids

d) Omega 7 fatty acids

Question 7:

Which of the following are essential fatty acids?

a) 1,2

b) 2,4

c) 1,3

d) 2,3,4

Question 8:

A boy presents with dry skin, scaly rash, hair loss and poor wound healing. He has cystic fibrosis and recently developed malabsorption syndrome secondary to pancreatic insufficiency. Deficiency of which of the following substances can explain these manifestations?

a) Arachidonic acid

b) Oleic acid

c) Linoleic acid

d) Elaidic acid

Question 9:

Which of the following is a by-product of hydrogenation of vegetable oils?

a) Eicosanoids

b) Glycerol

c) Cis fatty acids

d) Trans fatty acids

Question 10:

Prostaglandins are derived from which of the following fatty acids?

a) Palmitoleic acid

b) Oleic acid

c) Linoleic acid

d) Arachidonic acid

Question 11:

A 6-year-old boy presents with muscle aches, hypotonia and poor exercise tolerance. Further investigation reveals myoglobinuria and elevated creatine kinase enzyme level in blood. Which transporter deficiency is responsible for the pathophysiology of the disease?

a) Creatine

b) Creatinine

c) Carnitine

d) Acyl carrier protein

Question 12:

Which of the following proteins bind free fatty acids for transport in the blood?

a) Ceruloplasmin

b) Pre-albumin

c) Albumin

d) Transthyretin

Question 13:

What is the respiratory quotient of lipid?

a) 1

b) 0.8

c) 0.7

d) 0.6

Question 14:

A child presents with hypotonia and seizures. It was confirmed to be cerebrohepatorenal syndrome. Which of the following is accumulated in the brain in cerebrohepatorenal syndrome?

a) Very long chain fatty acid

b) Glucose

c) Lactic acid

d) Triglycerides

Answer Key

Question No.	Correct Option
1	c
2	b
3	a
4	d
5	d
6	a
7	c
8	c

9	d
10	d
11	c
12	c
13	c
14	a

Detailed Explanations

Solution to Question 1:

Steroids are derived lipids.

Lipids are classified as follows:

• Simple lipids: Esters of fatty acids with various alcohols. E.g. fats, oils, and waxes.

• Complex lipids: Esters of fatty acids with alcohols containing other groups like phosphate, nitrogenous base, carbohydrate, protein, etc. They can be divided into three groups:

• Phospholipids: Contain fatty acids, alcohol, phosphoric acid and a nitrogenous base.

• Glycolipids (glycosphingolipids): Contain fatty acids, sphingosine, and carbohydrate

• Others: Sulfolipids, amino lipids, and lipoproteins

• Derived lipids: These are the formed by the hydrolysis of simple and complex lipids. They possess the characteristics of lipids. Examples: Fatty acids, glycerol, steroids/sterols, lipid-soluble vitamins, ketone bodies etc.

Solution to Question 2:

Triacylglycerols serve as the major storage form of lipids in our body.

Triacylglycerols are esters derived from glycerol and three fatty acids. The fatty acids contained in lipids are mainly of two types:

• Saturated: Without any double bond in the chain

• Unsaturated: With one or more double bonds in the chain

Solution to Question 3:

Linoleic acid is not a saturated fatty acid.

Linoleic acid is a polyunsaturated fatty acid (PUFA) as it has 2 double bonds. Fatty acids can be classified into:

• Saturated fatty acids: No double bonds within the chain. Eg: Butyric acid, caproic acid, myristic acid, palmitic acid, stearic acid

• Unsaturated fatty acids:

• Monounsaturated fatty acids (MUFA): One double bond within the chain. Eg: Oleic acid

• Polyunsaturated fatty acids (PUFA): Two or more double bonds within the chain. Eg: Linoleic acid, Arachidonic acid

Solution to Question 4:

Linoleic acid is not an example of monounsaturated fatty acid. It is a polyunsaturated fatty acid (PUFA) with 2 double bonds.

Monounsaturated fatty acids (MUFA) contain a single double bond. Examples include palmitoleic acid (ω7), oleic acid (ω9) and elaidic acid (ω9).

Clinical correlate: When substituted for saturated fatty acids in the diet, MUFAs lower both total and LDL cholesterol and increase HDL. This may explain why the Mediterranean diet is associated with decreased serum total and LDL cholesterol compared to a typical Western diet high in saturated fats.

Solution to Question 5:

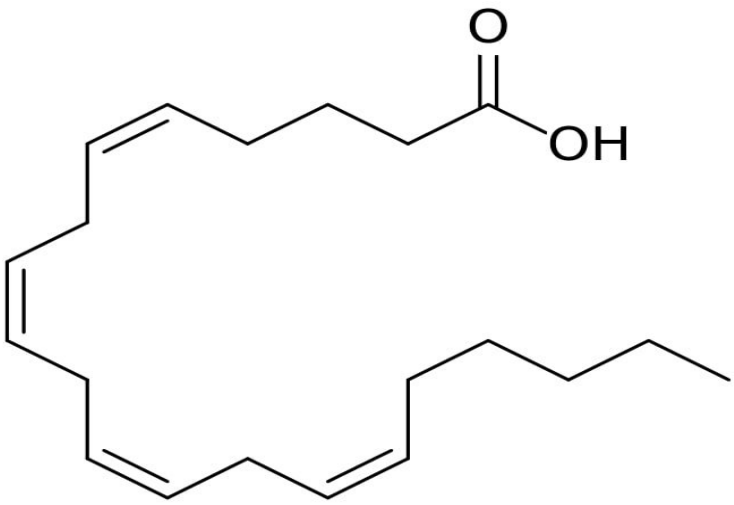
Arachidonic acid contains 4 double bonds in its structure.

Arachidonic acid is a polyunsaturated omega-6 fatty acid (PUFA). It has 4 double bonds present in a cis configuration. It is present in the phospholipids of cell membranes and is the precursor of prostanoids such as prostaglandins, thromboxanes, and leukotrienes.

Number of Double Bonds in Polyunsaturated Fatty Acids

Given below is an image of the structure of arachidonic acid.

Polyunsaturated fatty acids	No. of double bonds
Linoleic Acid	2
Linolenic Acid	3
Arachidonic Acid	4
Timnodonic Acid	5
Cervonic Acid	6



Solution to Question 6:

Omega 3 (ω3) fatty acids have anti-inflammatory effects.

ω3 fatty acids, such as linolenic, eicosapentaenoic and docosahexaenoic acids, result in synthesis of less inflammatory prostaglandins and leukotrienes. This contributes to their anti-inflammatory effect. They also have anti-thrombotic effects. This is because thromboxane A3 (formed from ω3 fatty acids) is less thrombogenic than thromboxane A2 (formed from ω6 fatty acids). ω3 fatty acids have also been shown to reduce serum triglyceride levels.

Solution to Question 7:

Linoleic acid and α-linolenic acid are essential fatty acids.

Essential fatty acids refer to polyunsaturated fatty acids(PUFAs) required for biological processes which are not synthesized by the body. Thus, they must be acquired from diet. The two essential fatty acids in humans are linoleic acid (Omega 6) - the precursor of arachidonic acid - and α-Linolenic acid (Omega 3) - the precursor of eicosapentaenoic acid and docosahexaenoic acid. Arachidonic acid and γ-Linolenic acid are semi-essential fatty acids since they can be derived from essential fatty acids.

Solution to Question 8:

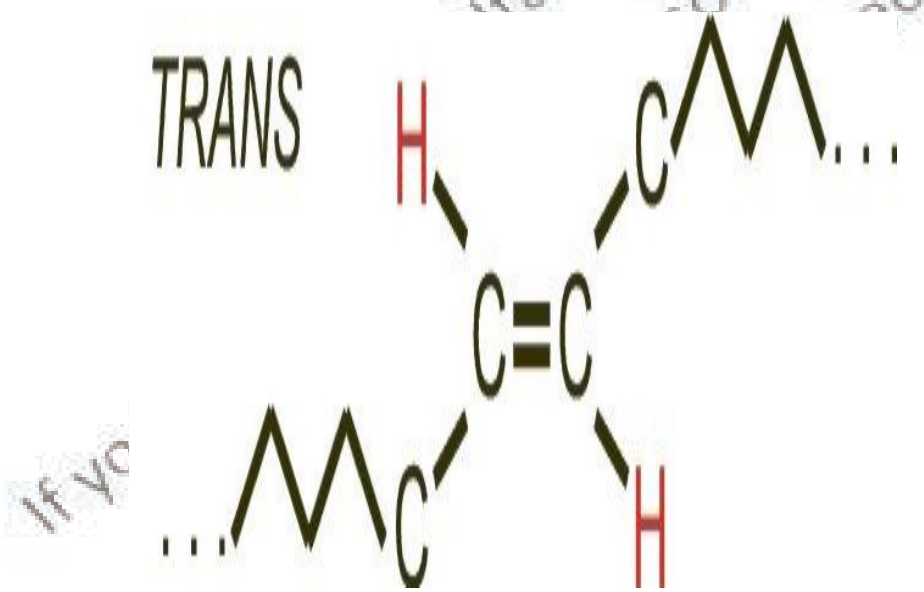
Dry skin, scaly rash, hair loss and poor wound healing with malabsorption syndrome are suggestive of essential fatty acid deficiency due to malabsorption. Linoleic acid is an essential fatty acid.

Essential fatty acids (Linoleic acid and α-Linolenic acid) are needed for many physiologic processes, including maintaining the integrity of skin and cell membranes. Deficiency of essential fatty acid is a rarely seen clinical entity in healthy adults and children who consume a varied diet. However, it can occur in individuals with risk factors affecting fatty acid absorption, like cystic fibrosis, pancreatic insufficiency, inflammatory bowel disease, massive bowel resection and prolonged parenteral nutrition. Patients with essential fatty acids deficiency have loss of structural integrity of the cell membranes, leading to like scaly dermatitis, hair loss and poor wound healing.

Solution to Question 9:

Trans fatty acids are a by-product of the hydrogenation of vegetable oils.

Trans fatty acids are isomers of cis fatty acids, the naturally occurring unsaturated fatty acids. The configuration of groups in a trans fatty acid is such that the acyl chains are on the opposite side of the double bond. They are formed during the hydrogenation of vegetable oils and in the production of margarine.



Trans fatty acids raise the level of triglycerides, total and LDL cholesterol and also lower the level of HDL. Hence their consumption is associated with an increased risk of cardiovascular disease. WHO recommends that the intake of trans fats should be less than 1% of the total energy intake.

Solution to Question 10:

Prostaglandins are derived from arachidonic acid.

Prostaglandins (PG) are a group of physiologically active lipid compounds. They act as local hormones. They belong to the chemical group known as prostanoids, which is a subclass of eicosanoids. They are synthesized from arachidonic acid through the cyclooxygenase (COX) pathway.

Solution to Question 11:

A young boy with muscle aches, hypotonia, poor exercise tolerance, myoglobinuria and elevated creatine kinase suggests a deficiency of carnitine.

Carnitine is a key transporter needed for beta-oxidation of fatty acids. It binds to acyl-CoAs to form acylcarnitine. This helps in the transfer of fatty acids across the impermeable inner mitochondrial membrane for beta-oxidation. Thus, carnitine deficiency results in impaired beta-oxidation of fatty acids. There will be inadequate ATP production in response to increased demands (e.g. exercise) with excess fatty acid accumulation in muscle cells. This results in muscle breakdown.

Patients with carnitine deficiency present with hypoglycemia, muscle aches, hypotonia and poor exercise tolerance (myopathy). They have high levels of serum creatine kinase. Treatment includes carnitine supplementation.

Solution to Question 12:

Free fatty acids are transported in the blood bound to albumin.

Solution to Question 13:

The respiratory quotient of lipid is 0.7

The respiratory quotient (RQ) is the molar ratio of carbon dioxide produced to the oxygen consumed when one mole of a substrate is metabolised.

Respiratory quotient for various substances

Substance	RQ (CO_2Produced/O_2Consumed)
Carbohydrate	1.00
Protein	0.81
Fat	0.71
Alcohol	0.66

Solution to Question 14:

Very long-chain fatty acids are accumulated in cerebrohepatorenal syndrome.

Cerebrohepatorenal syndrome, also known as Zellweger's syndrome, is a rare inherited disorder characterised by the absence of peroxisomes in all tissues. It is caused by mutations in the genes encoding peroxins, which take part in peroxisome biosynthesis. There is accumulation of C26-C38 polyenoic acids in brain tissues along with generalised loss of peroxisomal functions. Patients present with severe neurological symptoms and most die within a year. Other biochemical findings include abnormalities in the bile acid synthesis and a decreased plasmalogens.

Fatty acid oxidation and ketogenesis

Question 1:

Which of the following statements is false?

a) In beta oxidation, 2 carbon units are released as acetyl CoA per cycle.

b) In alpha oxidation, one carbon is lost in the form of CO_2 per cycle.

c) Alpha oxidation is an energetic process.

d) Beta oxidation is the major mechanism that occurs in the mitochondria matrix.

Question 2:

Which of the following statements is true?

a) Propionyl CoA is produced by the oxidation of odd-chain fatty acids.

b) Methylmalonyl CoA mutase is a biotin-dependent enzyme.

c) Propionyl CoA is not glucogenic.

d) Odd-chain fatty acids do not undergo oxidation.

Question 3:

ATP is hydrolyzed directly to AMP, with the release of PPi during a biochemical reaction catalyzed by which of the following enzymes?

a) Creatine kinase

b) Adenylate kinase

c) Myokinase

d) Acyl-CoA synthetase

Question 4:

For fatty acid oxidation which of the following intermediate is formed during the transport of acyl-CoA using a mitochondrial shuttle?

a) Acetyl-CoA

b) Acyl-coenzyme A

c) Acyl-carnitine

d) Aceto-acetyl-Coa

Question 5:

Which of the following enzymes is required for the only energy-requiring step of fatty acid oxidation?

a) Thiolase

b) Acyl CoA dehydrogenase

c) Thiokinase

d) Beta-hydroxy acyl Co A dehydrogenase

Question 6:

How many molecules of acetyl-CoA are produced in β- oxidation of palmitic acid?

a) 6

b) 7

c) 8

d) 9

Question 7:

Each cycle of β-oxidation produces _____.

a) 1 FADH2, 1 NAD+, and 1 acetyl-CoA

b) 1 FADH2, 1 NADH, and 1 acetyl-CoA

c) 1 FADH2, 1 NADH, and 2 CO2 molecules

d) 1 FAD, 1 NAD+, and 2 CO2 molecules

Question 8:

Which is the rate-limiting enzyme in beta-oxidation of fatty acids?

a) Acyl CoA dehydrogenase

b) Thiolase

c) Thiokinase

d) Carnitine palmitoyl transferase-1

Question 9:

What is the net ATP yield in the beta-oxidation of a straight-chain fatty acid having 2N carbon atoms?

a) 7N-6

b) 14N-6

c) 7N-3

d) 14N-3

Question 10:

A 28-year-old woman at 33 weeks of gestation presented with nausea and right upper quadrant abdominal pain. Her LFTs pointed towards a diagnosis of acute fatty liver of pregnancy. Which of the following enzyme deficiencies is most likely involved in the pathogenesis of this condition?

a) Fetal medium-chain acyl CoA dehydrogenase

b) Maternal long-chain 3-hydroxy acyl CoA dehydrogenase

c) Fetal long-chain 3-hydroxy acyl CoA dehydrogenase

d) Maternal medium-chain acyl CoA dehydrogenase

Question 11:

An 8-month-old infant was brought to the pediatric emergency department with recurrent seizures and hypoglycemia. His family history revealed the sudden death of his sibling at the age of 3 months. Labs showed increased dicarboxylic acid in blood and urine. Which of the following is the pathophysiology behind this clinical manifestation?

a) Impaired alpha oxidation of branched-chain fatty acids

b) Impaired beta-oxidation of medium-chain fatty acids

c) Impaired omega oxidation of medium-chain fatty acids

d) Impaired peroxisomal oxidation of very long-chain fatty acids

Question 12:

An infant is being assessed for hypotonia, seizures, and neurodevelopmental delay. Clinical examination reveals facial deformities, hyporeflexia, and hepatomegaly. A congenital disorder affecting extramitochondrial lipid metabolism is suspected. Which of the following processes is impaired in this patient?

a) Acetyl CoA formation

b) H_2O_2 mediated breakdown of fatty acids

c) Transport of fatty acyl CoA using shuttle mechanism

d) Peroxisomal alpha oxidation of fatty acids

Question 13:

A 3-year-old child is being evaluated for anosmia, deafness, and unstable gait. Further investigation reveals retinitis pigmentosa on fundoscopy and cardiac rhythm abnormalities on ECG. A rare congenital disease affecting lipid metabolism is being suspected. Deficiency of which of the following enzymes is expected to be found in this patient?

a) Phytanoyl CoA α-hydroxylase

b) Acyl CoA synthetase

c) Phytanic acid synthetase

d) None

Question 14:

Which of the following is a primary ketone body?

a) Acetone

b) Acetoacetate

c) Beta-hydroxybutyric acid

d) Acetyl CoA

Question 15:

A 13 year-old-boy with type 1 diabetes presents with abdominal pain, severe nausea, and vomiting. He has a history of increased urine output over the past 2 days. ABG analysis reveals high anion gap metabolic acidosis. The given clinical condition is due to the

over-activity of which of the following pathways?

a) HMP shunt

b) Gluconeogenesis

c) Lipolysis

d) Glycogenolysis

Question 16:

The rescue team has found a 33-year-old hiker who was stranded for 8 days. He was found in an extremely weak and dehydrated state. His breath has a fruity smell and urinalysis shows a positive ketostix test. The activity of which of the following enzymes is increased in him?

a) HMG CoA synthase

b) HMG CoA reductase

c) Acetyl CoA carboxylase

d) Pyruvate dehydrogenase

Question 17:

Which of the following cannot utilize ketone bodies?

a) Brain

b) Skeletal muscle

c) Liver

d) Kidney

Question 18:

A diabetic man presented with vomiting, abdominal pain, and polyuria. He gives a history of missing 2 doses of insulin since morning. He is hypotensive and tachycardic and his breath has a fruity odour. Blood glucose was 350 mg/dL. Which test, done here will give a positive result?

a) Hay's test

b) Fouchet's test

c) Ninhydrin test

d) Rothera's test

Question 19:

A 12-year-old girl suffers from an inadequately treated type 1 diabetes mellitus. She is currently hyperglycemic. Which of the following is most likely to occur?

a) Glycogenesis in muscle

b) Increased protein synthesis

c) Increased conversion of fatty acids to acetyl-CoA

d) Decreased cholesterol synthesis

Question 20:

Ketosis is common in type 1 DM due to:

a) 2,3 and 4

b) 1,2,3 and 4

c) 4 only

d) 1 and 2 only

Answer Key

Question No.	Correct Option
1	c
2	a
3	d
4	c
5	c
6	c
7	b
8	d
9	b
10	c
11	b
12	b

13	a
14	b
15	c
16	a
17	c
18	d
19	c
20	a

Detailed Explanations

Solution to Question 1:

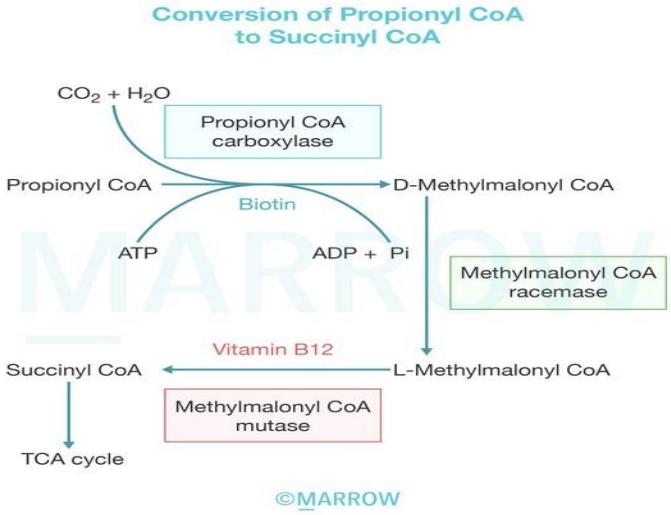
Alpha oxidation is not an energy yielding process.

Solution to Question 2:

'Propionyl CoA is produced by the oxidation of odd-chain fatty acids' is the correct statement.

In the oxidation of odd-chain fatty acids, the last cycle of oxidation yields a three-carbon-containing propionyl CoA. Propionyl CoA is carboxylated to methylmalonyl CoA by propionyl CoA carboxylase, a biotin-dependent enzyme that requires ATP hydrolysis.

Methylmalonyl CoA is further converted into succinyl CoA by racemase and mutase, (a vitamin B12 dependent enzyme) which enters into the TCA cycle.Thus, propionyl CoA can be converted into glucose by gluconeogenesis.



Note:

• Methylmalonyl CoA mutase is a vitamin B12-dependent enzyme.

• Propionyl residue from an odd-chain fatty acid is the only part of a fatty acid that is glucogenic.

Solution to Question 3:

ATP can also be hydrolyzed directly to AMP, with the release of PPi by the enzyme Acyl-CoA synthetase.

This occurs in the activation of long-chain fatty acids. This reaction is accompanied by the loss of free energy as heat, which ensures that the activation reaction will go to the right. It is further aided by the hydrolytic splitting of PPi, catalyzed by inorganic pyrophosphatase, a reaction that itself has a large $\Delta G0'$ of –19.2 kJ/mol.

Solution to Question 4:

In the fatty acid oxidation process, acyl-carnitine is formed as an intermediate during the transport of acyl-CoA using a mitochondrial shuttle.

Acyl CoA is converted to acyl-carnitine, which is the active form of fatty acid during transport from the cytoplasm to the inside of the mitochondria for oxidation. The enzyme CPT-1 (carnitine palmitoyl transferase-I), present on the outer mitochondrial membrane catalyzes the reaction.

Solution to Question 5:

The only energy-requiring step in fatty acid oxidation is catalyzed by the thiokinase enzyme.

In the presence of ATP and coenzyme A, the enzyme acyl-CoA synthetase or thiokinase catalyses the conversion of a fatty acid to acyl-CoA, the active form of fatty acid for oxidation.

Solution to Question 6:

In the β-oxidation of palmitic acid (C16), 8 molecules of acetyl-CoA are produced.

In the beta-oxidation pathway, two carbons at a time are cleaved from acyl-CoA molecules between alpha(2) and beta(3) carbons, starting at the carboxyl end. The two-carbon units formed are called acetyl-CoA.

Thus, in palmitic acid (C16), 7 cycles of β-oxidation take place, releasing 8 acetyl CoA molecules. In the first 6 cycles, 6 acetyl CoA molecules are released. In the 7th cycle, 2 acetyl CoA molecules are released.

Solution to Question 7:

One cycle of β-oxidation of fatty acids produces 1 $FADH_2$, 1 NADH, and 1 acetyl-CoA.

• $FADH_2$ is formed in the reaction catalyzed by acyl CoA dehydrogenase.

• NADH is formed in the reaction catalyzed by β-hydroxy acyl CoA dehydrogenase

• In each cycle of β-oxidation, the terminal two carbons are cleaved as acetyl CoA starting at carboxyl end.

Thus, the overall reaction of each cycle of β-oxidation can be summarized as follows:

(Cn)acyl CoA + FAD + NAD++ H_2O + CoA ° (Cn-2)acyl CoA + $FADH_2$ + NADH + Acetyl CoA +H+

Solution to Question 8:

Carnitine palmitoyl transferase-1 (CPT-1), is the rate-limiting enzyme in the beta-oxidation of fatty acids.

CPT-1 is responsible for transferring the acyl group from CoA to carnitine, forming acylcarnitine which can enter the outer mitochondrial membrane. Thus the acyl group enters the mitochondria from the cytoplasm.

Malonyl CoA is an inhibitor of CPT-I. Thus,

• In the fed state, with high insulin: glucagon ratio, acetyl CoA carboxylase is active and malonyl CoA is produced, which inhibits β-oxidation.

• In the fasting state, with low insulin: glucagon ratio, acetyl CoA carboxylase is inactive and malonyl CoA is not produced. Hence, β-oxidation takes place.

Solution to Question 9:

A straight-chain fatty acid comprising of 2N carbon atoms will undergo [(2N ÷ 2) -1] = (N-1) cycles of beta-oxidation and produce N number of acetyl CoA.

Each acetyl CoA will undergo TCA cycle, producing 10 ATP. On the other hand, each cycle of beta-oxidation also produces 1 $FADH_2$ and 1 NADH generating 1.5 ATP and 2.5 ATP respectively. Therefore (N-1) cycles of beta-oxidation will produce (N-1) $FADH_2$ and (N-1) NADH.

The energetics of β-oxidation of a straight-chain fatty acid comprising of 2N carbon atoms can be summarized as follows:

So, the net ATP yield after beta-oxidation of a straight-chain fatty acid having 2N carbon atoms will be (14N-4) - 2 = (14N- 6)We must keep in mind that during the initial activation of fatty acids (conversion of fatty acid to fatty acyl CoA), 2 ATP is utilized.

This formula can be used to calculate net ATP yield from a straight-chain fatty acid having an even number of carbon atoms. For example,

Net ATP yield from beta-oxidation of palmitic acid (16 carbon) will be 106.

Calculation: Here, 2N = 16 i.e, N = 8 . Therefore net ATP yield = (14x8 - 6) = 112-6 = 106

Intermediate produced	ATP yield per molecule	Total number produced	Total ATP
FADH2	1.5	N-1	1.5(N-1)
NADH	2.5	N-1	2.5(N-1)
Acetyl CoA	10	N	10N
Total ATP	1.5(N-1) +2.5(N-1) +10N =(14 N-4)		
ATP used in activation	- 2		
Net ATP yield	(14N-4) -2 = 14N-6		

Solution to Question 10:

A deficiency of fetal long-chain 3-hydroxy acyl CoA dehydrogenase (LCHAD) is involved in the pathogenesis of acute fatty liver of pregnancy.

Free fatty acids normally increase in pregnancy, particularly during late gestation to fuel fetoplacental growth and development. In fetuses homozygous for LCHAD deficiency, the fetoplacental unit is unable to undertake fatty acid metabolism adequately. This contributes to the accumulation of long-chain metabolites in maternal blood and hepatocytes, resulting in toxic effects.

Solution to Question 11:

The given clinical vignette is suggestive of medium-chain acyl-CoA dehydrogenase deficiency in which, there is impaired beta-oxidation of medium-chain fatty acids. This leads to the accumulation of dicarboxylic acids leading to dicarboxylic aciduria. It is characterized by:

• Excretion of C6-C10 ω-dicarboxylic acids (medium-chain) in the urine.

• Nonketotic hypoglycemia: due to the lack of ATP and acetyl CoA from β-oxidation (which is an activator of pyruvate carboxylase) to support gluconeogenesis.

Defects in medium-chain acyl-CoA dehydrogenase have been identified as a cause of sudden infant death syndrome (SIDS).

Omega-oxidation is an alternative way of fatty acid oxidation in the body when the beta-oxidation enzymes are either saturated or defective. Omega oxidation converts excess fatty acids to dicarboxylic acids that are soluble and hence can be excreted out via urine.

Other options:

Option A: Impaired Alpha oxidation leads to Refsum's disease. The manifestations of classic adult Refsum's disease include retinitis pigmentosa, peripheral neuropathy, ataxia, ichthyosis, anosmia,

deafness and cardiac arrhythmias.

Option C: Medium-chain acyl CoA dehydrogenase deficiency will lead to increased (not impaired) utilization of the omega oxidation pathway, resulting in the overproduction of dicarboxylic acids.

Option D: Impaired peroxisomal oxidation of very long-chain fatty acids (VLCFA) causes Zellweger (cerebrohepatorenal) syndrome. It is a rare inherited disorder characterized by the absence of peroxisomal enzymes in all tissues leading to the accumulation of C26—C38 polyenoic acids in brain tissue. The clinical manifestation of Zellweger syndrome includes:

• Facial deformities resembling Down syndrome (Mongoloid facies, Hypertelorism, Frontal bossing, Upslanting palpebral fissure)

• Brush field spots

• Hypotonia and hyporeflexia

• Seizures

• Neurodevelopmental delay

• Liver enlargement

Solution to Question 12:

The clinical vignette is suggestive of Zellweger (cerebrohepatorenal) syndrome in which H_2O_2-mediated peroxisomal beta-oxidation of very-long-chain fatty acids (VLCFA) is impaired.

Beta-oxidation in peroxisome is differentiated from that occurring in mitochondria by the formation of H_2O_2.

Beta-oxidation of fatty acid in peroxisomes is not coupled to ATP synthesis. The high-potential electrons are transferred to O_2, which yields H_2O_2 (instead of $FADH_2$). The enzyme catalase, found exclusively in peroxisomes, converts the hydrogen peroxide to end products water and oxygen.

Zellweger syndrome is a rare inherited disorder characterized by the absence of peroxisomal enzymes in all tissues leading to the accumulation of C26—C38 polyenoic acids in brain tissue. The clinical manifestation of Zellweger syndrome includes:

• Facial deformities resembling Down syndrome (Mongoloid facies, Hypertelorism, Frontal bossing, Upslanting palpebral fissure)

• Brush field spots

• Hypotonia and hyporeflexia

• Seizures

• Neurodevelopmental delay

• Liver enlargement

Solution to Question 13:

The given clinical vignette is suggestive of Refsum's disease, characterized by impaired alpha oxidation due to the deficiency of phytanoyl CoA α-hydroxylase (also known as phytanic acid alpha oxidase).

Refsum disease is a rare, autosomal recessive disorder caused by a deficiency of peroxisomal phytanoyl CoA α-hydroxylase, an enzyme involved in the alpha oxidation of phytanic acid (branched-chain fatty acid). This results in the accumulation of phytanic acid in the plasma and tissues.

The manifestation of classic adult Refsum disease includes

• Retinitis pigmentosa

• Peripheral neuropathy

• Ataxia

• Ichthyosis

• Anosmia

• Deafness

• Occasionally cardiac arrhythmias

Treatment: Restriction of dietary phytanic acid (found in dairy products and green leafy vegetables).

Solution to Question 14:

Acetoacetate is a primary ketone body as it is directly synthesized by the pathway of ketogenesis.

Secondary ketone bodies are those which are produced from the primary ketone body via spontaneous breakdown or by enzymatic reaction. Examples of secondary ketone bodies are: acetone and beta-hydroxybutyrate.

Ketone bodies are formed under conditions associated with a high rate of fatty acid oxidation such as:

• Fasting

• Carbohydrate restrictive diet

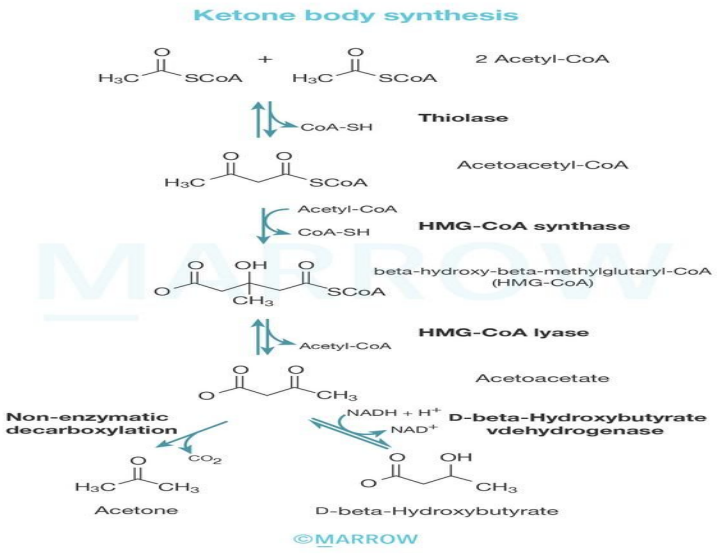
• Starvation

• Prolonged intense exercise

• Untreated (or inadequately treated) type 1 diabetes mellitus.

In such conditions, there is increased production of acetyl CoA from beta-oxidation of fatty acids which inhibits pyruvate dehydrogenase and activates pyruvate carboxylase. The oxaloacetate thus produced is used for gluconeogenesis rather than the TCA cycle. Hence, acetyl CoA is diverted into ketone body synthesis.

Acetyl CoA is converted into acetoacetate by the following reactions that occur in the liver mitochondria:



Thus, acetoacetate is the primary ketone body synthesized.

HMG CoA synthase is the rate-limiting step in ketone body synthesis.

Solution to Question 15:

The given clinical scenario is suggestive of diabetic ketoacidosis which is an insulin-deficient state leading to increased lipid breakdown (lipolysis) into fatty acids. This condition is also characterized by hyperglycemia and increased ketone body synthesis.

Type 1 diabetes mellitus is commonly found in the adolescent population. In response to certain precipitating factors, such patients may develop a severe insulin-deficient state. Since insulin is an inhibitor of hormone-sensitive lipase (HSL), an insulin-deficient state will cause HSL overactivity in adipocytes and the breakdown of lipid into fatty acids (lipolysis). Free fatty acids will undergo beta-oxidation and produce a large amount of acetyl CoA, which gets converted to ketone bodies (acetoacetate, acetone, beta-hydroxybutyrate, etc.) Such ketone bodies will accumulate in the blood and cause high anion gap metabolic acidosis.

Solution to Question 16:

A fruity smell (presence of acetone in breath) and positive ketostix test (ketonuria) on the background of prolonged starvation is suggestive of starvation ketosis. This is characterized by increased activity of HMG CoA synthase enzyme.

Prolonged starvation implies a catabolic state in which there is excessive β-oxidation leading to increased production of acetyl CoA. The excessive acetyl CoA is utilised by the hepatocytes to synthesise ketone bodies for extrahepatic utilisation.

HMG CoA synthase is the enzyme required for ketone body synthesis, it is also the rate-limiting enzyme.

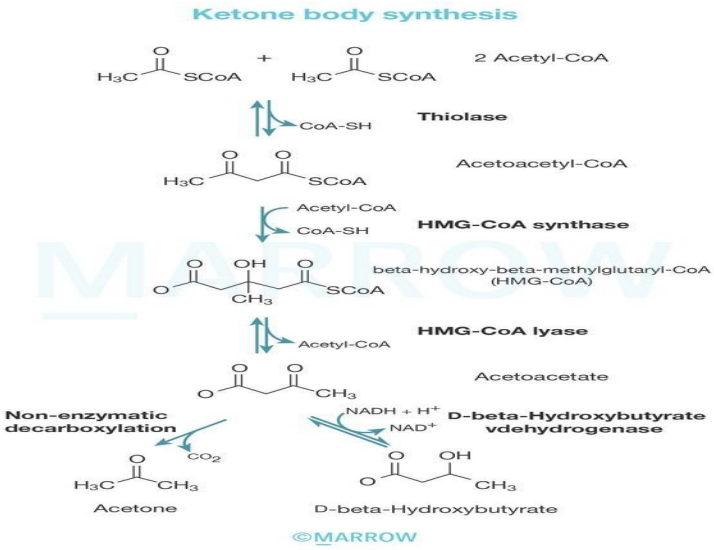
Other options:

• HMG CoA reductase is involved in the synthesis of cholesterol.

• Acetyl CoA carboxylase is involved in the fatty acid synthesis.

• Pyruvate dehydrogenase converts pyruvate into acetyl CoA.

All three enzymes are inactive in low insulin state (such as prolonged starvation).



Solution to Question 17:

The liver cannot utilize ketone bodies.

The ketone bodies synthesized in the liver mitochondria are transported in the blood to the peripheral tissues, where they are utilized by the following steps:

• β-hydroxybutyrate is oxidized to acetoacetate by β-hydroxybutyrate dehydrogenase producing NADH.

• Acetoacetate is then activated to acetoacetyl CoA by transferring a CoA molecule from succinyl CoA by succinyl CoA-acetoacetate CoA transferase (thiophorase).

• Acetoacetyl CoA is cleaved into two acetyl CoA molecules by thiolase and oxidized into CO_2 by the TCA cycle.

RBCs cannot utilize ketone bodies as they lack mitochondria.

The liver cannot utilize ketone bodies as it lacks the enzyme thiophorase.

Solution to Question 18:

The clinical picture is suggestive of diabetic ketoacidosis in which rothera's test will give positive results due to the presence of ketone bodies in urine.

Rothera's test by nitroprusside reagent is a specific test used to detect whether acetone or acetoacetate is present in the given sample of urine. A purple ring at the junction of two liquids indicates a positive test.

Note: Rothera's test does not detect β-hydroxybutyrate.

Other options:

Option A: Hay's test is used to test for bile salts.

Option B: Fouchet's test is used to test for bilirubin.

Option C: Ninhydrin test is used to test for alpha-amino acids.

Solution to Question 19:

With the background of inadequately treated type I diabetes mellitus, an insulin-deficient state in the body is expected which will lead to increased conversion of fatty acids to acetyl-CoA.

Type 1 diabetes mellitus is commonly found in the adolescent population. In response to certain precipitating factors, such patients may develop a severe insulin-deficient state. Since insulin is an inhibitor of hormone-sensitive lipase (HSL), an insulin-deficient state will cause HSL overactivity in adipocytes and the breakdown of lipid into fatty acids (lipolysis). Free fatty acids will undergo beta-oxidation and produce a large amount of acetyl CoA, which gets converted to ketone bodies (acetoacetate, acetone, beta-hydroxybutyrate, etc.)

Solution to Question 20:

2, 3 and 4 statements correctly explain the reason behind increased ketogenesis as seen in type1 DM.

Type 1 DM is characterized by a deficiency in insulin production. Since insulin is deficient, GLUT4 (insulin-dependent uptake) mediated glucose uptake is not possible in the heart or skeletal muscles. So adipose tissue mobilizes fat to be used as an energy source. These FFA (free fatty acids) are oxidized to acetoacetyl CoA which serves as a precursor for ketone body synthesis.

As the level of serum FFA is raised, proportionately more of the acetyl-CoA produced from their breakdown is converted to ketone bodies and less is oxidized via the citric acid cycle to CO_2. A fall in the concentration of oxaloacetate, particularly within the mitochondria, can impair the ability of the citric acid cycle to metabolize acetyl-CoA and divert fatty acid oxidation toward ketogenesis. Such a fall may occur because of an increase in the (NADH)/(NAD+) ratio caused when increased β-oxidation alters the equilibrium between oxaloacetate and malate so that the concentration of oxaloacetate is decreased.

Beta-hydroxy butyrate dehydrogenase is one of the enzymes used in ketone body synthesis. Increased rate of ketone body synthesis in insulin-deficient state implies an increased activity of beta-hydroxy butyrate dehydrogenase enzyme.

Biosynthesis of fatty acids and Eicosanoids

Question 1:

Which of the following biochemical reactions does not occur in mitochondria?

a) Kreb cycle

b) Urea cycle

c) Gluconeogenesis

d) Fatty acid synthesis

Question 2:

Which of the following pathways utilizes NADPH as the source of reducing equivalent?

a) Fatty acid synthesis

b) Ketone synthesis

c) Gluconeogenesis

d) Glycolysis

Question 3:

Which of the following pathway is the most important source of reducing equivalents for fatty acid synthesis in the liver?

a) Glycolysis

b) Tricarboxylic acid cycle

c) Uronic acid pathway

d) Hexose monophosphate shunt pathway

Question 4:

Which of the following is the rate-limiting enzyme of the fatty acid synthesis pathway?

a) Acetyl CoA carboxylase

b) Beta-hydroxy CoA dehydrogenase

c) Acyl CoA dehydrogenase

d) Ketoacyl synthase

Question 5:

Which of the following is an allosteric activator of acetyl CoA carboxylase?

a) Citrate

b) Acetate

c) Malate

d) Palmitate

Question 6:

Which is an important intermediate product of fatty acid biosynthesis ?

a) Cholesterol

b) Malonyl CoA

c) Acetyl CoA

d) Isovaleryl CoA

Question 7:

A child with severe acute malnutrition is undergoing nutritional rehabilitation. During follow-up visits, he shows significant improvement in appetite, an adequate gain of weight, and a gradual increase in subcutaneous body fat. Activity of which of the following enzymes is responsible for the restoration of fat storage in this patient?

a) Acetyl CoA carboxylase

b) Acyl-CoA dehydrogenase

c) Thiolase

d) Carnitine palmitoyl transferase - I

Question 8:

Which of the following is the most common end-product of fatty acid synthesis pathway?

a) Arachidonic acid

b) Oleic acid

c) Palmitic acid

d) Acetyl CoA

Question 9:

Which of the following statements about the mechanism of synthesis of fatty acids is correct?

a) Acetyl-CoA is the most active donor of two carbon atoms in fatty acid synthesis.

b) Malonyl-CoA is the most active donor of two carbon atoms in fatty acid synthesis.

c) Fatty acid synthesis is the reverse of oxidation of fatty acids.

d) Coenzyme A is the acyl group carrier of intermediates in fatty acid synthesis.

Question 10:

In which of the following steps of fatty acid synthesis is CO_2 produced?

a) Activation

b) Condensation

c) Elongation

d) Reduction

Question 11:

Which of the following is not a part of the fatty acid synthase complex?

a) Enoyl reductase

b) Ketoacyl synthase

c) Acetyl CoA carboxylase

d) Acetyl transacylase

Question 12:

The elongation of long chain fatty acids takes place in which if the following hepatocyte organelles?

a) 1,2

b) 3,5

c) 2,3,4

d) 1,3

Question 13:

Stearic acid is converted into which of the following fatty acids by the enzyme ■9 desaturase?

a) Linoleic acid

b) Oleic acid

c) Linolenic acid

d) Arachidonic acid

Question 14:

Which of the following is false regarding the role of insulin on lipid metabolism?

a) Increasing acetyl CoA carboxylase activity

b) Increasing the transport of glucose into the cells

c) Inhibition of pyruvate dehydrogenase

d) Decreasing intracellular cAMP level

Question 15:

Eicosanoids are derived from fatty acids with _____

a) 19C

b) 17C

c) 20C

d) 15C

Question 16:

What is the first step in synthesis of eicosanoids?

a) Activation of PGH2 synthetase

b) Activation of lipooxygenase

c) Activation of hydrolase

d) Activation of cyclooxygenase

Question 17:

Which substance is least likely to have its production decreased by aspirin?

a) Prostaglandins

b) Prostacyclin

c) Thromboxane

d) Leukotrienes

Question 18:

Which of the following are unstable intermediate products of the cyclooxygenase pathway?

a) 1,2

b) 3,4,5

c) 2,4

d) 1,3,5

Question 19:

Which of the following is not a primary prostaglandin?

a) PGD2

b) PGI2

c) PGE2

d) PGF2α

Question 20:

Which organ pair can synthesize the whole range of COX products?

a) Lung & spleen

b) Spleen & liver

c) Lung & liver

d) Blood vessel & spleen

Question 21:

Which enzyme is required in the synthesis of PGE2 from cyclic endoperoxides?

a) Isomerases

b) Cyclooxygenase

c) Hydrolase

d) Reductase

Question 22:

A 7-year-old girl presents with sudden onset of severe breathlessness with wheezing. Her peak expiratory flow rate is reduced significantly. Which of the following enzymes is important for the pathogenesis of the most likely diagnosis?

a) 7-Lipoxygenase

b) 12-Lipoxygenase

c) 15-Lipoxygenase

d) 5-Lipoxygenase

Question 23:

What is the first intermediate product of the LOX pathway?

a) HPETE

b) LTs

c) Lipoxins

d) HETE

Answer Key

Question No.	Correct Option
1	d

2	a
3	d
4	a
5	a
6	b
7	a
8	c
9	b
10	b
11	c
12	d
13	b
14	c
15	c
16	c
17	d
18	a
19	b
20	a
21	a
22	d
23	a

Detailed Explanations

Solution to Question 1:

Fatty acid synthesis does not occur in mitochondria.

Fatty acids are synthesized in the cytosol by a process called de novo fatty acid synthesis. All the steps, from acetyl-CoA to palmitate, are done by an extramitochondrial system. Fatty acid synthesis takes place in the liver, kidneys, brain, lungs, adipose tissue and lactating mammary gland.

Solution to Question 2:

NADPH is the source of reducing equivalent for fatty acid synthesis.

The NADPH utilised for fatty acid synthesis is derived from 3 sources:

- HMP shunt pathway (major source)
- Extramitochondrial isocitrate dehydrogenase (minor source)
- Malic enzyme

Solution to Question 3:

The reducing equivalent for fatty acid synthesis in the liver is NADPH. The most important source of NADPH is the hexose monophosphate shunt (HMP) pathway.

The HMP shunt pathway, or pentose phosphate pathway, is an alternative route for the metabolism of glucose. It generates NADPH and synthesizes ribose. NADP+ is hydrogenated to NADPH by glucose-6-phosphate dehydrogenase. Since the HMP shunt pathway and de novo fatty acid synthesis both occur in the cytosol of the cell, there are no membranes or permeability barriers against the transfer of NADPH. Thus, this forms the major source for NADPH for fatty acid synthesis.

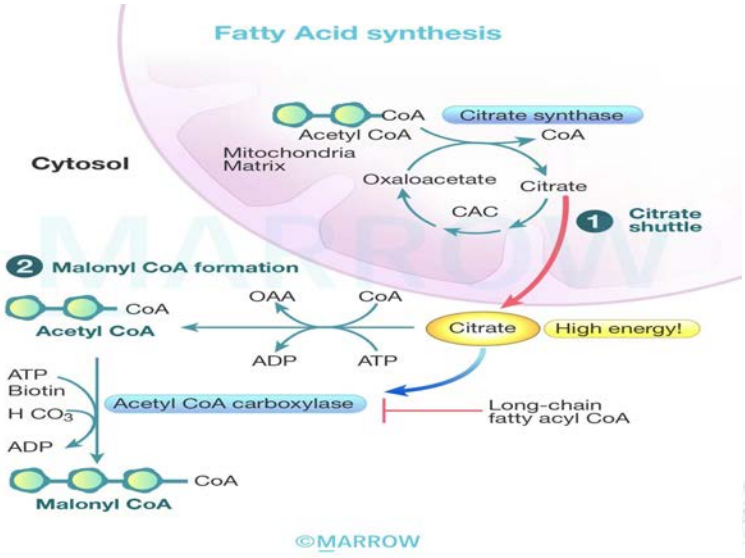
Other minor sources of NADPH are extramitochondrial isocitrate dehydrogenase and NADP malate dehydrogenase (malic enzyme).

Solution to Question 4:

Acetyl CoA carboxylase is the rate-limiting enzyme of the fatty acid synthesis pathway.

Acetyl CoA carboxylase converts acetyl CoA into malonyl CoA, the first step of de novo fatty acid synthesis. It is an allosteric enzyme activated by citrate in the well-fed state. Acetyl CoA carboxylase requires the following:

- NADPH as the reducing equivalent
- Biotin is a coenzyme needed by carboxylases
- ATP for energy
- Manganese (Mn^{2+})
- HCO_3^- as the carbon donor

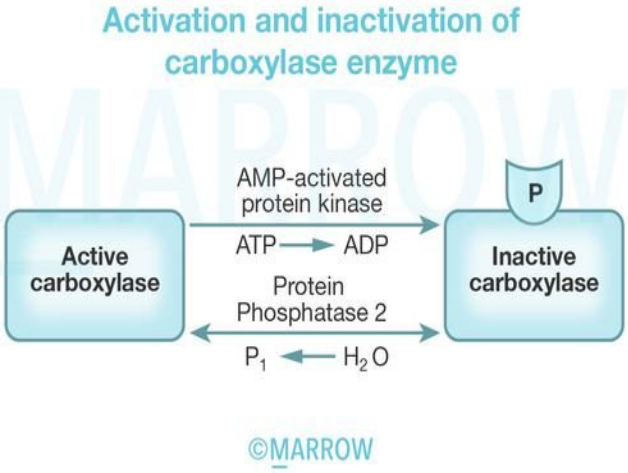


Solution to Question 5:

Citrate is an allosteric activator of acetyl-CoA carboxylase.

Acetyl-CoA carboxylase is the rate-limiting enzyme in de novo fatty acid synthesis. Citrate promotes the conversion of acetyl CoA carboxylase from an inactive dimer to an active polymeric form. Its concentration increases in the well-fed state. Insulin also activates the enzyme by dephosphorylating it.

Glucagon and epinephrine promote inactivation of acetyl-CoA-carboxylase by phosphorylation of the enzyme. Long-chain acyl-CoA molecules such as palmitoyl CoA also promotes inactivation of the enzyme.

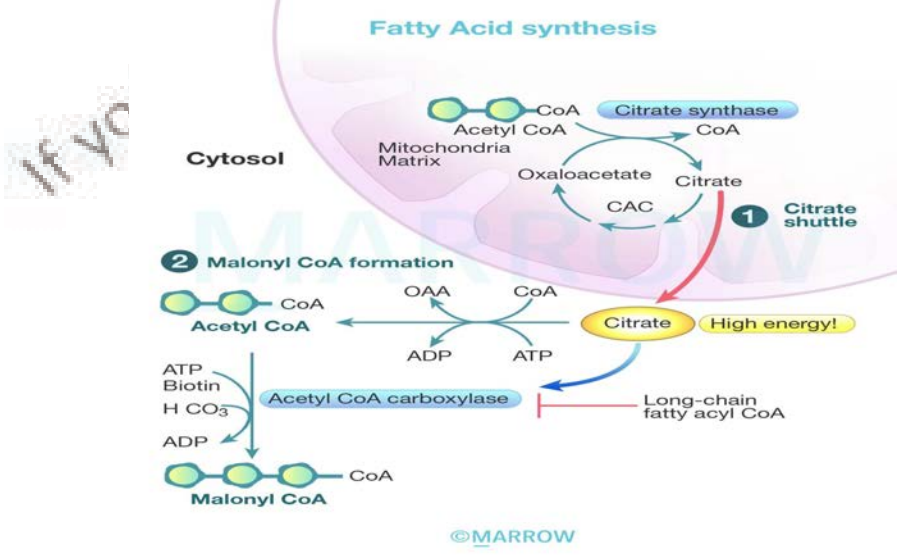


Examples of allosteric modifiers		
Enzyme	Allosteric inhibitor	Allosteric activator
1. ALA synthase	Heme	-
2. Aspartate transcarbamoylase	CTP	ATP
3. HMG CoA reductase	Cholesterol	-
4.Phosphofructokinase	Citrate, ATP	AMP, F2,6P
5. Acetyl CoA carboxylase	Acyl CoA	Citrate
6. Citrate synthase	ATP	-
7. Carbamoyl phosphate synthase-I	-	NAG
8. Carbamoyl phosphate synthase-II	-	ATP

Solution to Question 6:

Malonyl-CoA is an important intermediate product of fatty acid biosynthesis.

Production of malonyl-CoA from acetyl CoA is the initial and rate-limiting step in fatty acid synthesis. Bicarbonate is a source of CO_2 and is required in the initial reaction for the carboxylation of acetyl-CoA to malonyl-CoA in the presence of ATP and acetyl-CoA carboxylase.



Solution to Question 7:

The restoration of fat storage in a child undergoing nutritional rehabilitation, as evidenced by adequate weight gain and an increase in subcutaneous body fat, occurs with the help of acetyl CoA carboxylase.

With appropriate supplementation of nutrition, the child's body will gradually enter an anabolic phase. In the well fed state, there is an abundant supply of glucose, which is converted to pyruvate, which in turn is converted to acetyl CoA. Acetyl CoA is diverted into fatty acid synthesis, in which acetyl CoA carboxylase is the rate-limiting enzyme. There is also a high insulin : glucagon ratio in the well fed state. This favors dephosphorylation (activation) of acetyl CoA carboxylase, resulting in increased fatty acid synthesis. The fatty acids thus formed (most commonly palmitoyl CoA) are esterified into triacylglycerols. This process is called lipogenesis.

Note: The enzymes acyl-CoA dehydrogenase, thiolase, and carnitine palmitoyl transferase-I are involved in fatty acid oxidation and are inactive in the well-fed state.

Solution to Question 8:

The most common end product of fatty acid synthesis is palmitic acid.

Palmitic acid is a 16 carbon saturated long chain fatty acid (LCFA). The substrate for its synthesis is acetyl CoA. Acetyl CoA is also the end product of fatty acid beta-oxidation.

Solution to Question 9:

Malonyl-CoA is the most active donor of two carbon atoms in fatty acid synthesis.

Malonyl-CoA is a 3-carbon unit that donates 2 carbon units in fatty acid synthesis. It is formed from acetyl-CoA by acetyl-CoA carboxylase. During fatty acid synthesis, acetyl CoA primarily acts as the primer. The enzyme fatty acid synthase (FAS) adds two carbons from malonyl-CoA to the inital acetyl-CoA, forming a 4-carbon intermediate (butyryl-ACP). FAS then sequentially adds two carbons from malonyl-CoA to the ACP-intermediates (eg: butyryl-ACP) to elongate the fatty acid chain.

Thus, in the end product palmitic acid (C16), only carbon atoms 15 and 16 are from acetyl CoA. The remaining 14 carbon atoms are derived from malonyl-CoA. Thus, malonyl-CoA is the most active donor of two carbon atoms in fatty acid synthesis.

Note:

Option 3: Fatty acid synthesis is not a simple reversal of oxidation of fatty acids. These two processes involve completely different enzymes and occur in different compartments of the cell.

Option 4: Intermediates in fatty acid synthesis are covalently linked to the sulfhydryl group of an acyl carrier protein (ACP), not coenzyme A, of the enzyme fatty acid synthase. Intermediates in the fatty acid breakdown are covalently attached to the sulfhydryl group of coenzyme A.

Solution to Question 10:

In fatty acid synthesis, CO_2 is produced in the condensation step.

Solution to Question 11:

Acetyl CoA carboxylase is not a part of the fatty acid synthase complex.

The fatty acid synthase complex is a multienzyme polypeptide complex that synthesizes fatty acids from malonyl-CoA and acetyl-CoA. It is a dimer of two identical polypeptide monomers (homodimer). Each monomer contains six enzymes and the acyl carrier protein (ACP). ACP carries the intermediate products formed during fatty acid synthesis. The enzymes of the fatty acid synthase complex are as follows:

1. Condensing unit:

• Ketoacyl synthase

• Malonyl/acetyl transacylase

2. Reduction unit:

• Dehydratase

• Enoyl reductase

• Beta-ketoacyl reductase

3. Releasing unit:

• Thioesterase

Solution to Question 12:

The elongation of long chain fatty acids takes place in the 1. endoplasmic reticulum and 3. mitochondria of hepatocytes.

Fatty acid elongation occurs in three cellular compartments: the cytosol, mitochondria, and endoplasmic reticulum (microsomes). Both unsaturated and saturated fatty acids can be elongated, with malonyl-CoA acting as the 2 carbon donor and NADPH as the reducing agent.

Solution to Question 13:

Stearic acid is converted to oleic acid by the enzyme ■9 desaturase.

■9 desaturase is present in the endoplasmic reticulum. It catalyzes the synthesis of monounsaturated fatty acids (Eg: Oleic acid) from saturated fatty acids (Eg: Stearic acid). ■9 desaturase is a mixed-function oxidase, requiring NADPH and O_2. The first double bond is always introduced in the ■9 positions. Humans cannot introduce double bonds beyond ■9 positions (between C-10 and terminal methyl group). Hence, linoleic ($\Delta 9,12$) and linolenic acid ($\Delta 9,12,15$) are considered essential fatty acids and should be included in the diet.

Solution to Question 14:

Inhibition of pyruvate dehydrogenase is not an action of insulin. Insulin activates the pyruvate dehydrogenase enzyme.

Insulin stimulates lipogenesis by the following mechanisms:

• Decreasing the level of intracellular cAMP. This favours dephosphorylation of acetyl-CoA carboxylase, resulting in its activation.

• Increasing acetyl-CoA carboxylase activity. This accelerates the rate-limiting step of fatty acid synthesis.

• Increasing the transport of glucose into the cell and activation of pyruvate dehydrogenase in the adipose tissue. This yields more acetyl CoA for fatty acid synthesis.

Solution to Question 15:

Eicosanoids are derived from fatty acids with 20C.

Eicosanoids are formed from arachidonic acid and other C20 polyunsaturated fatty acids (PUFA). They are short-acting paracrine chemicals that bring about their action through a G-protein-coupled receptor.

Types of eicosanoids:

• Prostanoids

• Prostaglandins (E & F)

• Prostacyclins

• Thromboxanes

• Leukotrienes

• Lipoxins

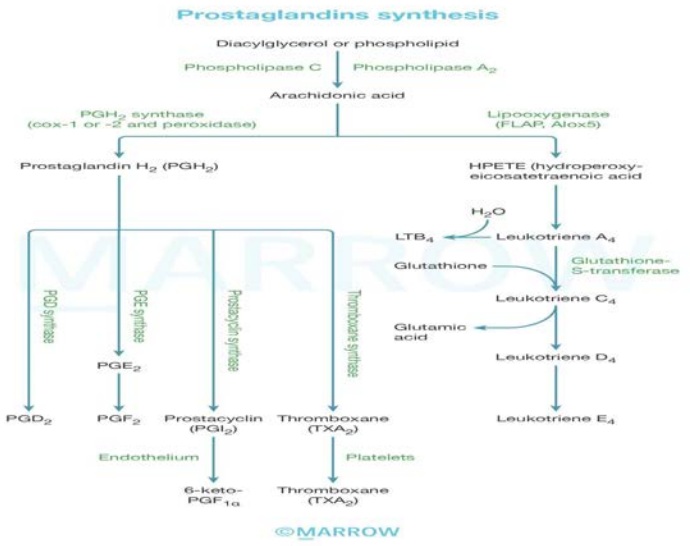
Solution to Question 16:

The first step in the synthesis of eicosanoids involves the activation of hydrolase.

Eicosanoid synthesis is governed by the rate of release of arachidonic acid from membrane lipids in response to appropriate stimuli (Eg: Increased calcium). These stimuli activate hydrolases, including phospholipase A2, which releases arachidonic acid from membrane phospholipids. The released arachidonic acid is then converted into:

• Prostaglandins and thromboxanes by the cyclooxygenase (COX) pathway. PGH2 synthase is the first enzyme of this pathway.

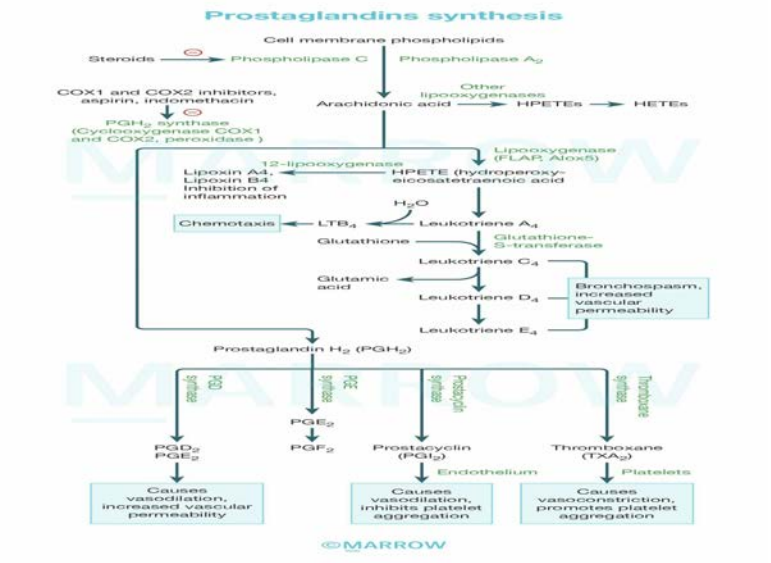
• Leukotrienes and lipoxins by the lipooxygenase (LOX) pathway



Solution to Question 17:

Aspirin is least likely to decrease the production of leukotrienes.

Aspirin is a non-steroidal anti-inflammatary drug (NSAID) that inhibits the cyclooxygenase (COX) pathway. The cyclooxygenase (COX) pathway generates eicosanoids with a ring structure - prostaglandins, thromboxane, prostacyclin. Leukotrienes, however, are produced by the lipoxygenase pathway (LOX), which is not inhibited by aspirin.



Solution to Question 18:

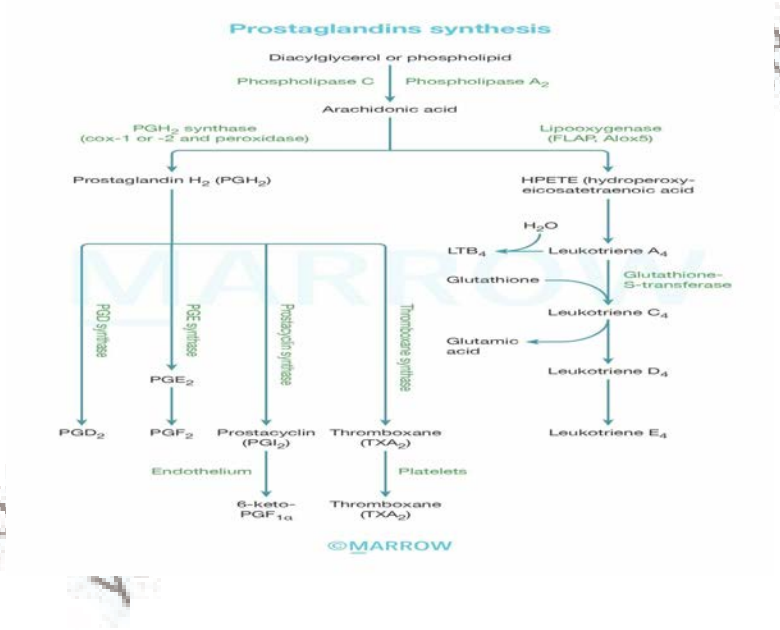
PGG2 and PGH2 are unstable intermediates of the cyclooxygenase (COX) pathway.

The cyclooxygenase enzyme of the COX pathway is a bifunctional enzyme. It first oxidizes arachidonic acid to PGG2 and then peroxidizes PGG2 to PGH2. These two intermediates (cyclic endoperoxides) are unstable. PGH2 is metabolized further by specific synthases and isomerases into a variety of eicosanoids including PGE2, PGD2, PGF2α, prostacyclin (PGI2), and thromboxane (TX) A2.

Solution to Question 19:

PGI2 is not a primary prostaglandin.

PGD2, PGE2, and PGF2α are called the primary prostaglandins based on their structure. Primary prostaglandins contain a single cyclopentane ring with a 15-hydroxyl group with a 13,14-trans double bond. Prostacyclin, or PGI2, structurally differs from the primary prostaglandins due to its double ring structure.



Solution to Question 20:

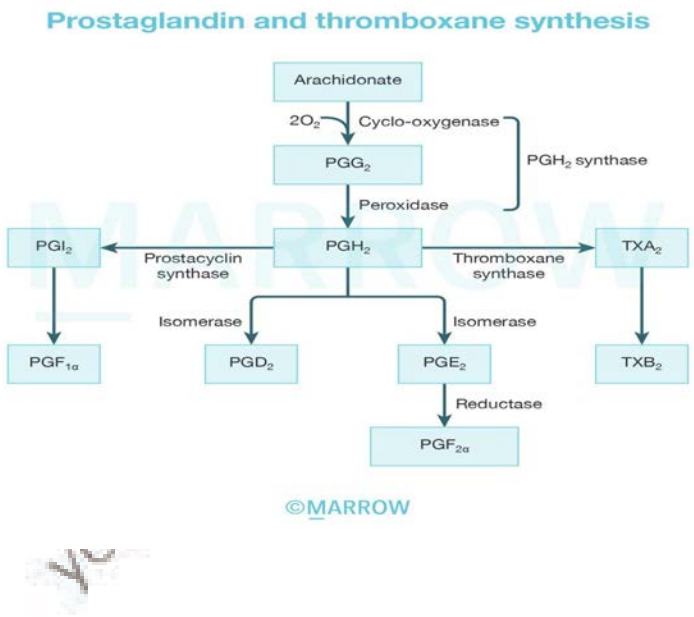
Lung and spleen can synthesize the whole range of COX products.

COX pathway products	Site of synthesis
Whole range of COX products	Lung & Spleen
Thromboxane	Primarily platelets
Prostacyclin	Endothelium of blood vessels

Solution to Question 21:

Isomerases are required in the synthesis of PGE2 from cyclic endoperoxides i.e., PGG2 and PGH2

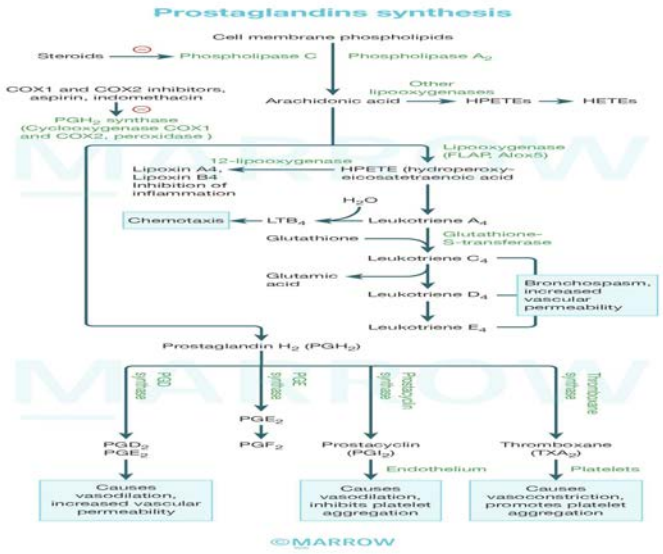
COX pathway products	Enzymes for synthesis
PGI2	Prostacyclin synthase
PGD2	Isomerase
PGE2	Isomerase
PGF2α	Reductase
TXA2	Thromboxane synthase



Solution to Question 22:

Sudden onset of severe breathlessness and wheezing with a reduced peak expiratory flow rate is suggestive of acute exacerbation of asthma. 5-lipoxygenase enzyme is important for the pathogenesis of this condition.

Acute exacerbation of asthma is characterized by bronchoconstriction due to chemicals mediators like leukotrienes (LTB4, LTC4, LTD4 etc.). Leukotrienes are produced from the breakdown of arachidonic acid by 5-lipoxygenase via the lipoxygenase pathway.

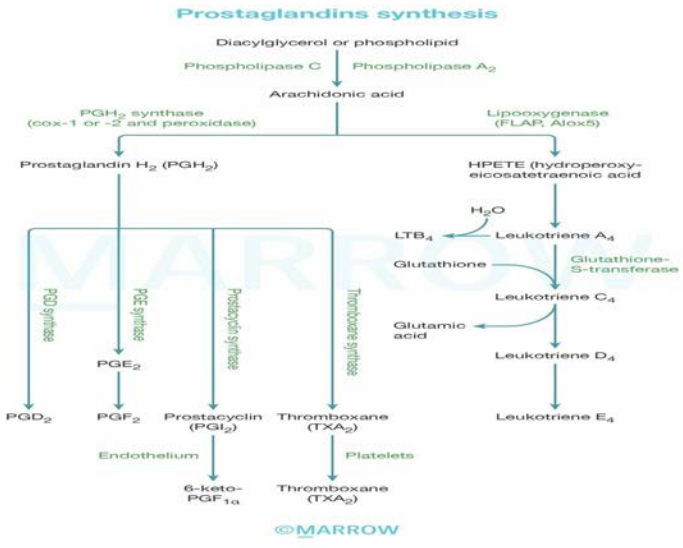


Leukotrienes are eicosanoic acid derivatives formed in leukocytes, mast cells, platelets, and macrophages. They are potent constrictors of the bronchial airway. Leukotrienes also cause increased vascular permeability and activation and attraction of leukocytes. Thus, they are involved in many inflammatory or immediate hypersensitivity reactions, such as asthma.

Solution to Question 23:

Hydroperoxyeicosatetraenoic acid (HPETE) is the first intermediate product produced in the LOX pathway.

5-lipoxygenase, the first enzyme of the LOX pathway, breaks down arachidonic acid into 5-hydroperoxyeicosatetraenoic acid (5-HPETE) which undergoes further chemical reactions to produce leukotrienes (LTs), hydroxyeicosatetraenoic acid (HETE) and lipoxins.



Metabolism of Acylglycerols and Sphingolipids

Question 1:

What type of lipid is triacylglycerol?

a) Glycerolipid

b) Glycerophospholipid

c) Glycolipid

d) Sterol Lipid

Question 2:

Which is the first step for catabolism of triacylglycerol?

a) Decarboxylation

b) Hydrolysis

c) Oxidation

d) Dehydrogenation

Question 3:

Which of the following organs can catabolize the glycerol component of triacylglycerol?

a) 2, 3, 4

b) 1, 2, 3, 5

c) 1, 3, 4, 5

d) 1, 2, 3, 4, 5

Question 4:

Which of the following represents the structure of a phospholipid?

a) Glycerol + phosphate + choline or ethanolamine

b) Monoglyceride + phosphate + choline or ethanolamine

c) Diglyceride + phosphate + choline or ethanolamine

d) Triglyceride + phosphate + choline or ethanolamine

Question 5:

Which of the following is not a phospholipid?

a) Plasmalogen

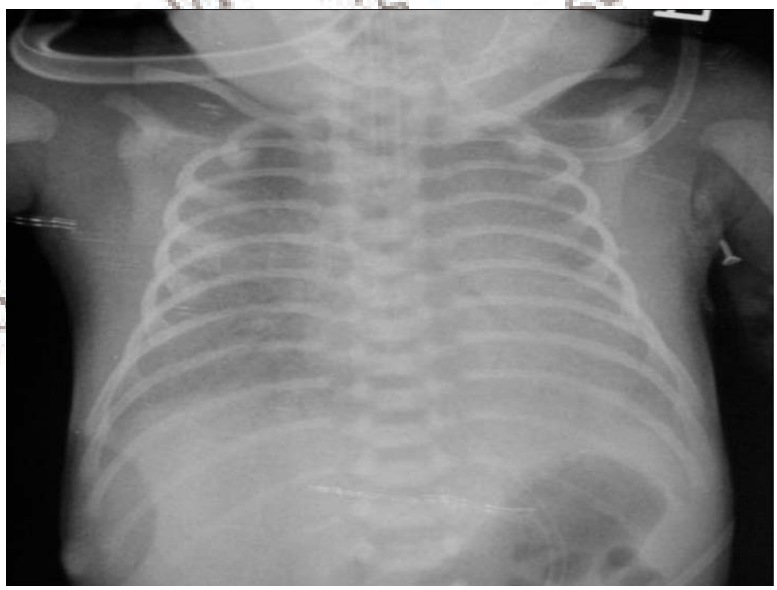
b) Dipalmitoyl lecithin

c) Ceramide

d) Cardiolipin

Question 6:

A 1.5 kg male baby was delivered vaginally at 32 weeks of gestation. The infant was cyanotic and required CPAP immediately after delivery. His chest radiograph is shown below. A deficiency of which of the following is the likely cause for the condition?



a) Phosphatidylethanolamine

b) Dipalmitoyl phosphatidylcholine

c) Diphosphatidyl glycerol

d) Phosphatidylinositol

Question 7:

What is the location of phospholipids synthesis?

a) Rough endoplasmic reticulum

b) Smooth endoplasmic reticulum

c) Golgi apparatus

d) Mitochondria

Question 8:

Which of the following is the common intermediate in the biosynthesis of triacylglycerols and phosphoglycerols?

a) Phosphatidate

b) Acyl CoA

c) Acetyl CoA

d) Plasmalogen

Question 9:

Which of the following is the composition of the phospholipid cephalin ?

a) Phosphatidylserine

b) Phosphatidylethanolamine

c) Phosphatidylcholine

d) Phosphatidylinositol

Question 10:

Which of the following phospholipids plays an important role in apoptosis?

a) Phosphatidylserine

b) Phosphatidylethanolamine

c) Phosphatidylcholine

d) Phosphatidylglycerol

Question 11:

Which of the following phospholipids is found only in the mitochondria and helps in the mitochondrial function?

a) Cephalin

b) Cardiolipin

c) Lecithin

d) Plasmalogen

Question 12:

Which of the following phospholipids serves as a reservoir for prostaglandin synthesis?

a) Phosphatidyl serine

b) Phosphatidyl inositol

c) Phosphatidyl choline

d) Phosphatidyl ethanolamine

Question 13:

Which of the following phospholipids has a role to play in signal transmission across membranes?

a) Phosphatidyl serine

b) Phosphatidyl inositol

c) Phosphatidyl choline

d) Phosphatidyl ethanolamine

Question 14:

A lady undergoing further evaluation after a miscarriage has a positive VDRL result. Gold standard investigation using FTA-ABS shows a negative result. Which of the following substances is responsible for this discrepancy in the results?

a) Cephalin

b) Lecithin

c) Cardiolipin

d) Plasmalogen

Question 15:

Sphingolipid is a type of _____

a) Glycerol esters saponifiable lipid

b) Non-glycerol esters saponifiable lipid

c) Non-saponifiable lipids

d) Derived lipid

Question 16:

Which of the following is not a component of glycosphingolipid ?

a) Carbohydrate

b) Glycerol

c) Sphingosine

d) Fatty acid

Question 17:

Which of the following is an example of a glycolipid?

a) Plasmalogen

b) Cerebroside

c) Sphingomyelin

d) Lecithin

Question 18:

Which of the following is not a role of glycosphingolipids?

a) Source of blood group antigens

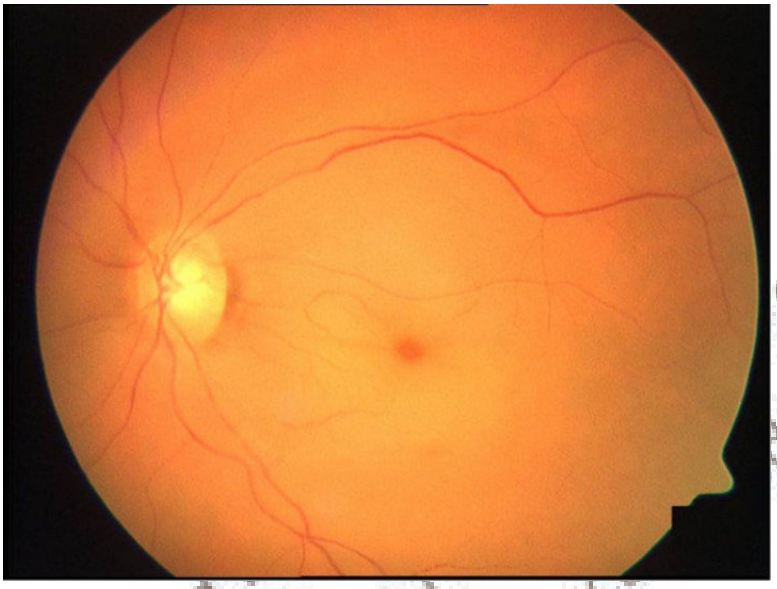
b) Cell surface receptors for cholera toxin

c) Cell surface receptors for tetanus toxin

d) Formation of lung surfactant

Question 19:

A 1-year old boy presented with loss of motor skills, increased startle response to noise and seizures. On fundoscopy, the following image was seen. Further evaluation reveals the diagnosis of a lysosomal storage disorder. Which of the following enzymes is most likely defective in this condition?



a) Hexosaminidase A

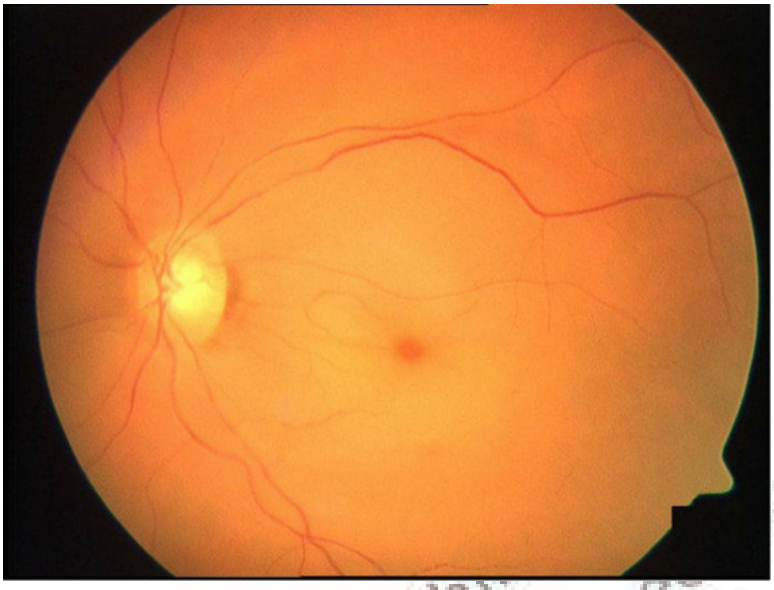
b) β-glucosidase

c) α-galactosidase

d) Glucose-6-phosphatase

Question 20:

A 6-month-old infant with progressive failure to thrive and milestone regression is being evaluated for an inborn error of metabolism. On examination, hypotonia, apathy, deafness, hepatosplenomegaly, and generalized lymphadenopathy are present. Fundus examination is shown below. A liver biopsy shows foamy hepatocytes loaded with sphingomyelin. Electron microscopy reveals zebra bodies. What is the likely diagnosis?



a) Niemann-Pick disease type A

b) Tay-Sach disease

c) Niemann-Pick disease type B

d) Sandhoff's disease

Question 21:

An 8-year-old boy presents with burning pain and numbness in his hands, heat intolerance and decreased visual acuity. Further evaluation reveals skin angiokeratomas and a decreased renal function. Which of the following is the mode of inheritance of the most likely diagnosis?

a) Autosomal recessive

b) X-linked recessive

c) Autosomal dominant

d) X-linked dominant

Question 22:

A 10-year old child presents chronic fatigue, growth retardation and epistaxis. On further evaluation, hepatosplenomegaly, thrombocytopenia and anemia was found. Bone marrow biopsy showed cells with wrinkled tissue paper appearance. What is the probable diagnosis?

a) Niemann Pick disease

b) Fabry's disease

c) Gaucher's disease

d) Tay Sach's disease

Question 23:

Which of the following enzymes is deficient in metachromatic leucodystrophy?

a) β-Galactosidase

b) Arylsulfatase

c) Sphingomyelinase

d) Ceramidase

Question 24:

Accumulation of which of the following substances is responsible for Farber's disease?

a) Lecithin

b) Ceramide

c) Sphingomyelin

d) Sulfatides

Answer Key

Question No.	Correct Option
1	a
2	b
3	b
4	c
5	c
6	b
7	b
8	a
9	b
10	a
11	b
12	b

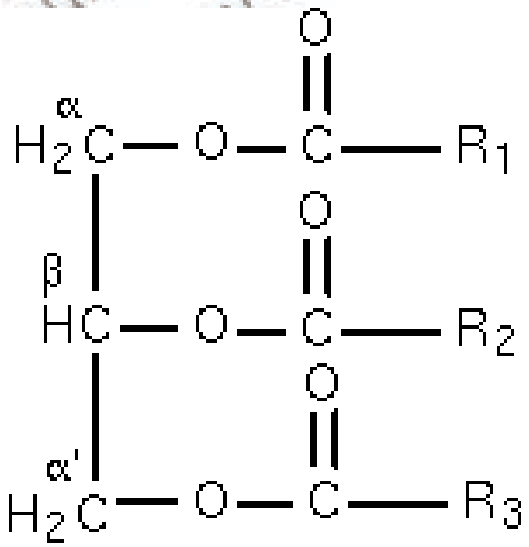
13	b
14	c
15	b
16	b
17	b
18	d
19	a
20	a
21	b
22	c
23	b
24	b

Detailed Explanations

Solution to Question 1:

Triacylglycerol is a glycerolipid.

Glycerolipids are esters of the trihydric alcohol glycerol and fatty acids. The types of glycerolipids include mono-, di- or tri-substituted glycerols. The best known glycerolipid is the fatty acid triester of glycerol known as triglycerides or triacylglycerols.



Solution to Question 2:

The first step for the catabolism of triacylglycerol is hydrolysis.

Triacylglycerols must be hydrolyzed by lipase to their constituent fatty acids and glycerol before further catabolism. Hydrolysis (lipolysis) occurs in adipose tissue with release of free fatty acids into the plasma. Free fatty acids combine with albumin to reach various tissues, where they are oxidized to obtain energy or re-esterified.

Solution to Question 3:

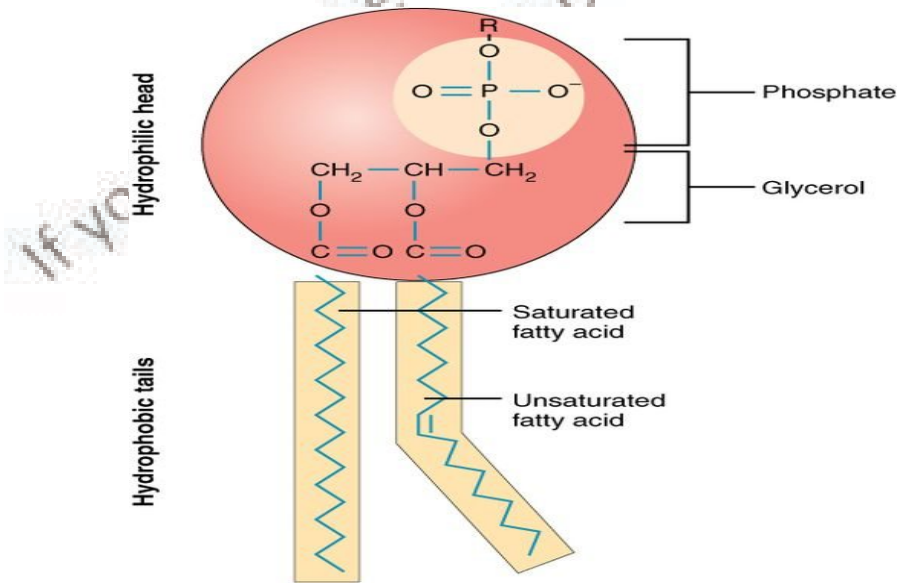
White adipose tissue cannot catabolize the glycerol component of triacylglycerol.

The utilization of glycerol depends upon whether the tissue has the enzyme glycerol kinase. It is found in the liver, kidney, intestine, brown adipose tissue and the lactating mammary gland. However, white adipose tissue lacks glycerol kinase.

Solution to Question 4:

Phospholipid consists of diglyceride + phosphate + choline or ethanolamine.

A phospholipid, such as lecithin or cephalin, is any lipid consisting of a diglyceride combined with a phosphate group. A simple organic molecule such as choline or ethanolamine is attached to the phosphate group. A diglyceride is glycerol molecule with 2 fatty acids attached to it.



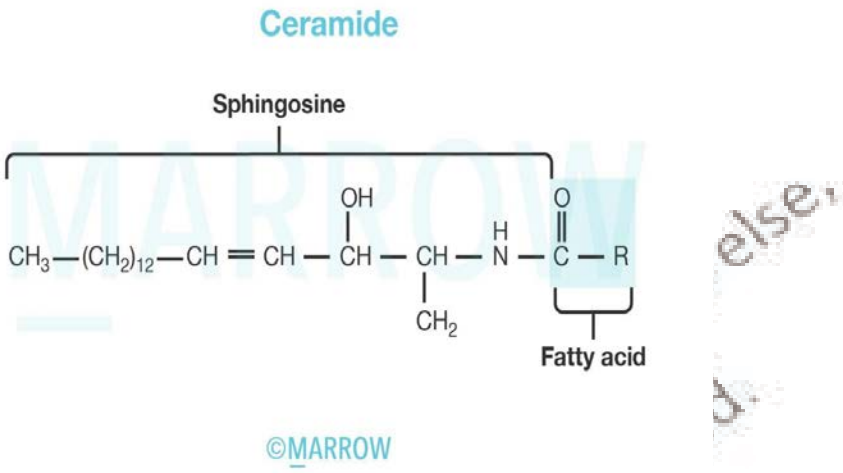
Solution to Question 5:

Ceramide is not a phospholipid.

Ceramide consists of sphingosine and a fatty acid (ceramide = sphingosine + fatty acid) . It can be a component of a phospholipid e.g sphingomyelin, which consists of ceramide, phosphoric acid

and choline. It is also a component of non-phosphorylated compound lipids like cerebrosides, globosides, and gangliosides. These are fatty acids esterified with alcohol and contain other groups.

Option A: Plasmalogens are a class of glycerophospholipids that contain vinyl-ether.



Solution to Question 6:

A preterm infant requiring resuscitation at birth with cyanosis and diffuse bilateral opacities on chest radiography is suggestive of infant respiratory distress syndrome. It is caused by a deficiency of dipalmitoyl phosphatidylcholine.

Dipalmitoyl lecithin or dipalmitoylphosphatidylcholine (DPPC) is a major component of lung surfactant. In DPPC, positions 1 & 2 on the glycerol are occupied by palmitate. Surfactant is secreted by the type 2 pneumocytes in the alveoli. It reduces surface tension at the air-liquid interface of the alveolus and increases compliance.

Mature levels of surfactant are present after 35 weeks of gestation. Thus, preterm babies are at risk of surfactant deficiency, leading to infant respiratory distress syndrome. Lung maturity of the fetus can be assessed by determining the ratio of lecithin to sphingomyelin (L:S ratio) in the amniotic fluid. A ratio of >2 indicates maturity.

Other options -

Option 1: Phosphatidylethanolamine, also called cephalin, is found in the cell membranes of nervous tissue, particularly in the brain.

Option 3: Diphosphatidyl glycerol, also called cardiolipin, is found in the inner membrane of mitochondria and plays a key role in apoptosis.

Option 4: Phosphatidylinositol present in the cell membrane acts as a precursor of hormone secondary messengers and platelet-activating factor.

Solution to Question 7:

The location of phospholipids synthesis is the smooth endoplasmic reticulum.

Once synthesized the phospholipids are transported into the Golgi apparatus, and further across cell membranes, to be extruded out of the cell by the process of exocytosis.

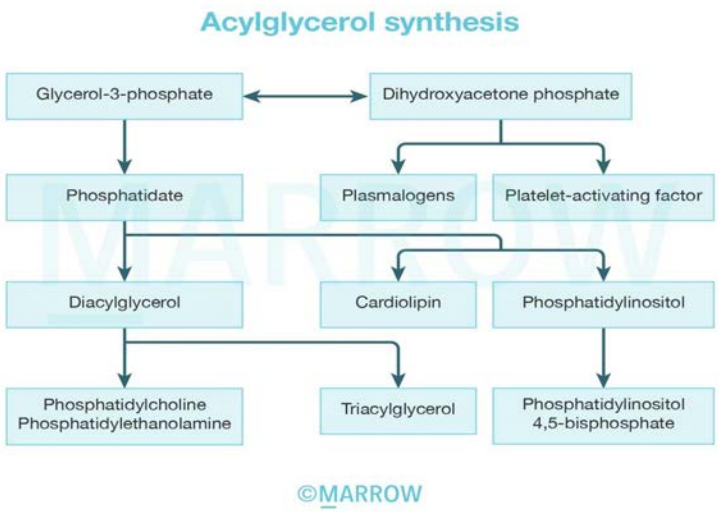
Solution to Question 8:

The common intermediate in the biosynthesis of triacylglycerols and phosphoglycerols is phosphatidate.

Acyl-CoA combines with glycerol-3-phosphate in two successive reactions to form phosphatidate or 1,2-diacylglycerol phosphate. 1,2- diacylglycerol phosphate contributes its phospahate for the formation of phosphatidyl inositol and becomes 1,2-diacylglycerol.

The 1,2-diacylglycerol is either converted to triacylgycerol by diacylglycerol acyl transferase, or reacts with CDP-choline or CDP-ethanolamine to form the phospholipids phosphatidylcholine or phosphatidylethanolamine.

(Note: Phosphoglycerol is the glycerol ester of phosphoric acid and is a component of phospholipids).



Solution to Question 9:

Phosphatidylethanolamine is the composition of the phospholipid cephalin.

Cephalin is a phospholipid composed of ethanolamine and phosphatidic acid. Thus, it is also known as phosphatidylethanolamine. It is found in cell membranes.

Solution to Question 10:

Phosphatidylserine plays an important role in apoptosis.

Phosphatidylserine, a phospholipid, is a component of the cell membrane. It is composed of serine and phosphatidic acid. It plays a key role in cell cycle signaling, coagulation and apoptosis (programmed cell death).

Solution to Question 11:

Cardiolipin is the phospholipid found only in the mitochondria.

Cardiolipin is an important component of the inner mitochondrial membrane. It is essential for the optimal function of numerous mitochondrial enzymes involved in mitochondrial energy metabolism and for the maintenance of the mitochondrial membrane structure.

Decreased cardiolipin or alteration in its metabolism results in mitochondrial dysfunction. This is seen in Barth syndrome (cardioskeletal myopathy) which is characterized by an enlarged and weakened heart (dilated cardiomyopathy), weakness in muscles used for movement (skeletal myopathy), recurrent infections due to small numbers of white blood cells (neutropenia), and short stature.

Solution to Question 12:

Phosphatidylinositol serves as a reservoir for prostaglandin synthesis.

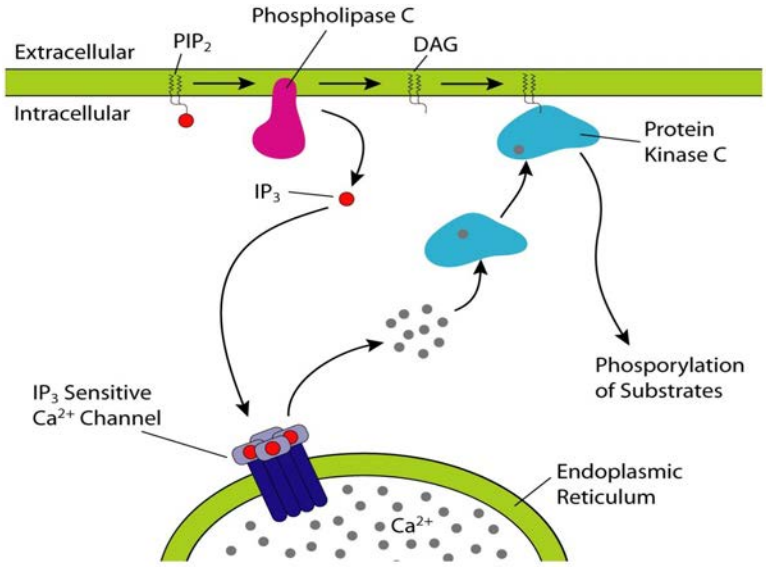
Phosphatidylinositol contains stearic acid and arachidonic acid attached to glycerol. Therefore, it serves as a reservoir of arachidonic acid in membranes and hence provides the necessary substrates for prostaglandin synthesis.

Solution to Question 13:

Phosphatidylinositol (PI) plays a role in signal transmission across membranes.

Membrane-bound phosphatidylinositols (PI) are phosphorylated to produce polyphosphoinositides such as phosphatidylinositol 4,5-bisphosphate (PIP2). When neurotransmitters, hormones, and growth factors to receptors on the cell membrane, degradation of PIP2 by phospholipase C.

The degradation products are inositol 1,4,5-trisphosphate (IP3), which mediates the mobilization of intracellular calcium, and diacylglycerol (DAG), which activates protein kinase C. Both of these act synergistically to evoke specific cellular responses thus accomplishing signal transmission.



Solution to Question 14:

Cardiolipin, a phospholipid with antigenic properties, is responsible for the discrepancy in results i.e positive VDRL test with negative FTA-ABS test.

A positive VDRL test with negative FTA-ABS test suggests a false-positive VDRL. The VDRL antigen consists of cardiolipin-cholesterol-lecithin mixtures for the detection of serum antibodies formed against Treponema pallidum.

The lady undergoing further evaluation for a miscarriage probably has an anti-cardiolipin antibody in her serum. This antibody has likely cross-reacted with the cardiolipin component of the VDRL reagent, resulting in a biological false-positive VDRL result. However, a confirmatory test i.e FTA-ABS test reveals that her anti-cardiolipin antibody is not from infection with Treponema pallidum but due to some other reasons.

Solution to Question 15:

Sphingolipid is a type of non-glycerol ester saponifiable lipid.

Sphingolipids (eg: sphingomyelin) are a class of lipids containing a backbone of sphingoid bases, a set of aliphatic amino alcohols e.g. sphingosine. They are known as non-glycerol esters. Sphingolipids are amphipathic molecules.

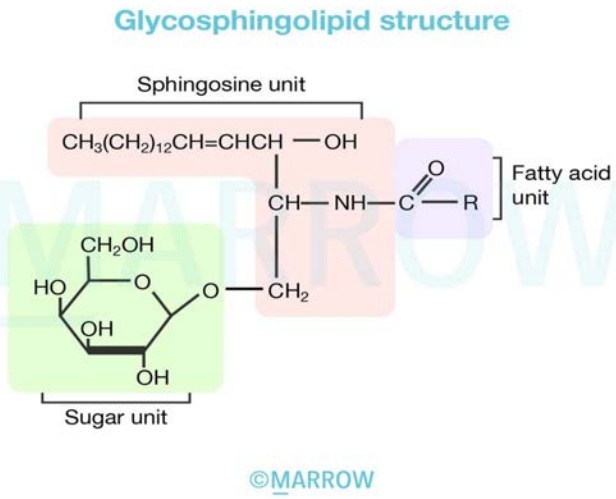
Sphingolipids are found on the outer layer of the plasma membrane and in the myelin sheath surrounding the nerves. They also play a role in cell signaling and apoptosis.

Solution to Question 16:

Glycerol is not a component of glycosphingolipids.

Glycolipids or glycosphingolipids are complex lipids that contain the alcohol sphingosine instead of glycerol. They also contain a fatty acid and a carbohydrate attached to sphingosine.

Glycosphingolipid = Sphingosine + fatty acid + carbohydrate



Solution to Question 17:

Cerebroside is an example of a glycolipid.

A glycolipid is a complex lipid containing a carbohydrate. Cerebrosides consist of sphingosine, a fatty acid and a single sugar linked to sphingosine. Thus, they are glycolipids. Cerebrosides are found predominantly in the brain and peripheral nervous tissue, with high concentrations in the myelin sheath.

Solution to Question 18:

Formation of lung surfactant is not a function of glycosphingolipids.

Lung surfactant is composed of the glycerophospholipid known as dipalmitoyl lecithin or dipalmitoylphosphatidylcholine.

Glycosphingolipids, like cerebrosides, are part of the outer leaflet of plasma membranes. They are important in cell adhesion, cell recognition, and signal transduction. Some function as blood group antigens, and as cell surface receptors for cholera (GM1 ganglioside) and tetanus toxins, and certain viruses and microbes. They also serve as various embryonic antigens specific for stages of fetal development. The carbohydrate portion of a glycolipid which serves as the antigenic determinant.

Solution to Question 19:

An infant with loss of motor skills, increased startle response to noise and seizures (neurological involvement) and a cherry-red spot in the macula suggests Tay-Sachs disease. This disease is caused by a deficiency of the enzyme hexosaminidase A.

Tay-Sachs disease is an autosomal recessive disorder resulting in the deficiency of hexosaminidase A. As a result, GM2 gangliosides are not degraded and they accumulate within lysosomes. Affected children present with loss of motor skills, increased startle response, seizures, hyperacusis, macrocephaly, neurodegeneration, blindness and muscular weakness. Cherry-red spots are seen in the macula of eye.

Note: The clinical features of Sandhoff's disease, due to deficiency of hexosaminidase A and B, are similar to Tay-Sachs. Affected children additionally have hepatosplenomegaly and cardiac involvement.

Solution to Question 20:

Progressive failure to thrive, milestone regression, hypotonia, apathy, deafness, hepatosplenomegaly, and generalized lymphadenopathy with the cherry-red spot on fundus examination suggests a diagnosis of Niemann-Pick disease type A.

Niemann-Pick disease is a lysosomal storage disorder due to an inherited deficiency of sphingomyelinase. This results in the accumulation of sphingomyelin in the monocyte-macrophage system. Type A disease is a severe infantile form. Infants present with extensive neurologic involvement (hypotonia, milestone regression, apathy), marked visceral accumulations of sphingomyelin (hepatosplenomegaly, foamy hepatocytes, zebra bodies on EM), progressive wasting and early death within the first 3 years of life.

In contrast, patients with Niemann-Pick disease type B have organomegaly but generally no CNS involvement.

Solution to Question 21:

Burning pain and numbness in hands, heat intolerance, decreased visual acuity with skin angiokeratomas and a decreased renal function is suggestive of Fabry's disease which is inherited as an X-linked recessive disorder.

Fabry's disease is a lipid storage disorder due to a deficiency of α-galactosidase A, resulting in the accumulation of globotriaosyl-ceramide in tissues. Young boys present with Fabry crises (burning pain in the hands, feet and proximal extremities), angiokeratomas (reddish blue telangiectatic skin lesions between the umbilicus and knees), corneal and lenticular opacities, renal failure and cardiomyopathy.

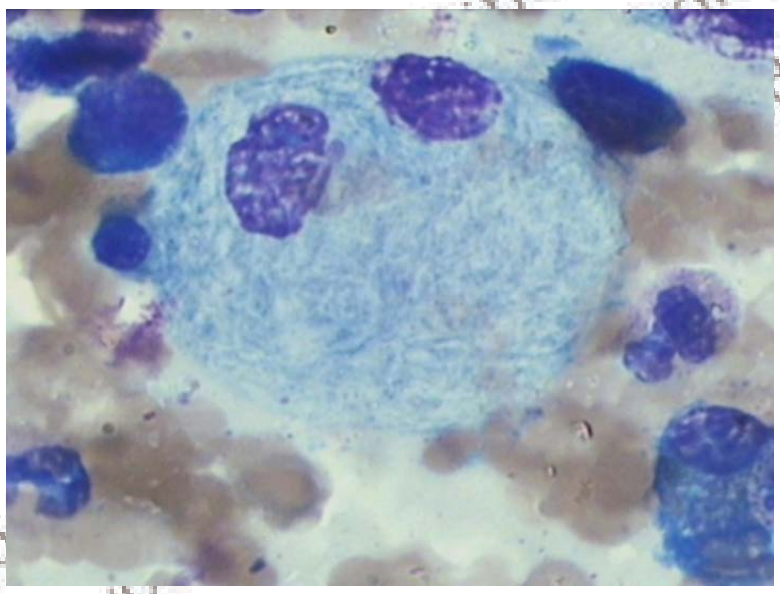
Note: All sphingolipidoses are inherited in an autosomal recessive manner, except Fabry's disease.

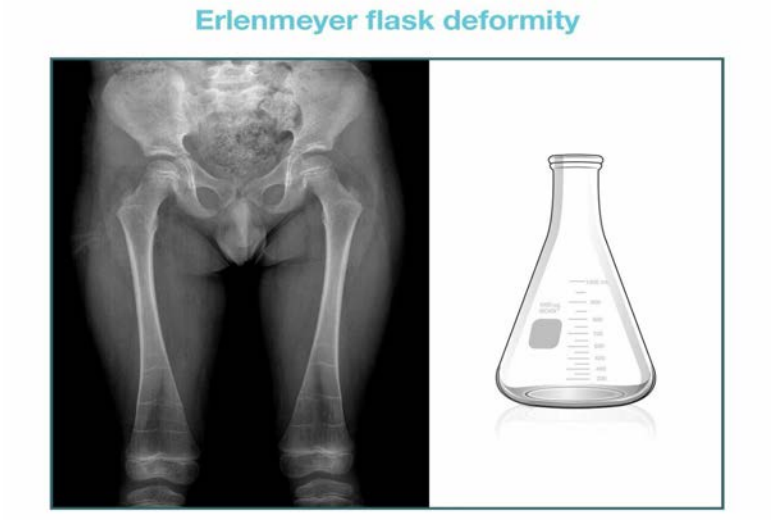
Solution to Question 22:

Chronic fatigue, growth retardation, epistaxis, hepatosplenomegaly, thrombocytopenia, anemia, Erlenmeyer flask deformity of femur with bone marrow biopsy showing cells with wrinkled tissue paper appearance suggest Gaucher's disease.

Gaucher's disease is a genetic disorder in which glucocerebroside accumulates in the reticuloendothelial system due to the deficiency of the enzyme glucocerebrosidase (acid β-glucosidase). It is the most common lysosomal storage disorder.

Affected children present with bleeding tendencies secondary to thrombocytopenia, chronic fatigue secondary to anemia, painless hepatomegaly and splenomegaly, and bone pain. They may have liver cirrhosis, osteoporosis and pathological fractures. Neurological symptoms such as developmental delay, hypertonia, strabismus, supranuclear gaze palsy are seen in type 3 Gaucher's disease. On bone marrow biopsy, Gaucher's cells with wrinkled tissue paper appearance are seen. Erlenmeyer flask deformity is seen on X-ray of femur.





Treatment is with enzyme replacement therapy in the form of human recombinant acid β-glucosidase (Imiglucerase), velaglucerase alfa or taliglucerase alfa.

Note: Neurological involvement is not seen in type I Gaucher's disease

Solution to Question 23:

Metachromatic leucodystrophy occurs due to the deficiency of arylsulfatase A enzyme.

Metachromatic leucodystrophy is an autosomal recessive disorder. Deficiency of arylsulfatase A enzyme leads to accumulation of sulfated glycosphingolipids in white matter with subsequent demyelination. Affected infants (infantile form) present with cognitive deterioration, muscle weakness, hypotonia, nystagmus, myoclonic seizures, optic atrophy and progressive paralysis.

Solution to Question 24:

Accumulation of ceramide is seen in Farber's disease.

Farber's disease occurs due to deficiency of ceramidase, which catalyses the breakdown of ceramide to fatty acid and sphingosine. Affected children present with painful joint swelling and nodules, progressive joint deformity, hoarse cry and breathing difficulty, mental retardation and failure to thrive.

Cholesterol Synthesis, Transport and Excretion

Question 1:

Where does cholesterol synthesis take place?

a) Cytosol and endoplasmic reticulum

b) Peroxisomes and mitochondria

c) Mitochondria and cytosol

d) Mitochondria and golgi body

Question 2:

What is the substrate for cholesterol synthesis?

a) 3-hydroxybutyrate

b) Acetoacetate

c) Acetyl CoA

d) Acyl CoA

Question 3:

Which of the following is not a primary bile acid?

a) Cholic acid

b) Chenodeoxycholic acid

c) Glycocholic acid

d) Deoxycholic acid

Question 4:

Which is the first intermediate in the conversion of cholesterol to bile acids?

a) 7-dehydrocholesterol

b) 7-hydroxycholesterol

c) Lanosterol

d) Ergosterol

Question 5:

A 47-year-old diabetic man comes to the OPD for a follow-up visit. In the most recent lab report, serum LDL level is high. He is therefore started on a lipid-lowering drug that acts on the rate-limiting step of cholesterol synthesis. Which of the following is the site of action of this drug?

a) HMG CoA synthase

b) HMG CoA reductase

c) Squalene epoxidase

d) HMG CoA lyase

Question 6:

Which is the first hormone to be produced in cholesterol synthesis?

a) Cholecystokinin

b) Epinephrin

c) Lanosterol

d) Secretin

Question 7:

What is the function of lipoprotein?

a) Transport majority of nonpolar lipids

b) Transport majority of polar lipids

c) Transport only cholesterol

d) Transport only fatty acids

Question 8:

Which of the following lipoproteins have the highest cholesterol:triglyceride ratio?

a) Chylomicron and VLDL

b) LDL and HDL

c) HDL and VLDL

d) IDL and HDL

Question 9:

In a patient with lipoprotein lipase deficiency, which of the following is increased following a fatty meal?

a) Chylomicron

b) LDL

c) HDL

d) Apo A

Question 10:

Which of the following lipoproteins are the major carriers of triacylglycerol?

a) Chylomicron and VLDL

b) LDL and HDL

c) HDL and VLDL

d) Chylomicron and HDL

Question 11:

Which is the largest human plasma lipoprotein?

a) VLDL

b) LDL

c) HDL

d) Chylomicron

Question 12:

Which of the following statements about HDL is false?

a) HDL act as repository for the apo C and apo E.

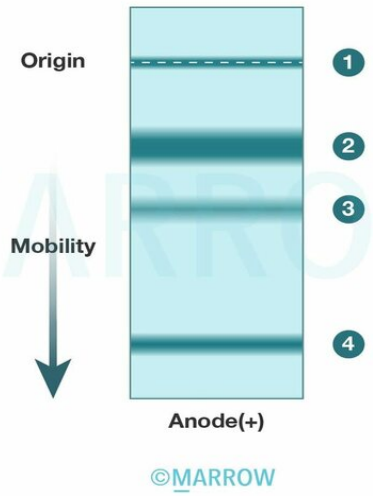
b) Class B scavenger receptor B1 (SRB-1) is HDL receptor.

c) HDL removes cholesterol from tissues.

d) HDL increases LDL oxidation.

Question 13:

An adolescent male with a history of recurrent abdominal pain and eruptive xanthoma is suspected to have a rare genetic condition affecting lipid metabolism. On serum lipid electrophoresis (diagram below) band 1 (origin) is found to be wider than normal. Activity of which of the following enzymes is reduced in this patient?



a) Lipoprotein Lipase

b) Hormone Sensitive Lipase

c) LCAT

d) HMG Co-A reductase

Question 14:

Which of the following contains apolipoprotein A?

a) 1,2,3

b) 1,2

c) 3,5

d) 2,4,5

Question 15:

Which of the following has the highest electrophoretic mobility and least lipid content?

a) Chylomicrons

b) HDL

c) LDL

d) VLDL

Question 16:

Which of the following is the main apolipoprotein present in LDL ?

a) Apo B-100

b) Apo E

c) Apo-48

d) Apo A-11

Question 17:

Which of the following statements about LDL is false?

a) It delivers cholesterol to cells.

b) It contains only one apoprotein.

c) It is the marker of cardiovascular disease.

d) It contains Apo B-48.

Question 18:

Which of the following is an activator of the key enzyme required in reverse cholesterol transport?

a) Apo B-100

b) Apo B-48

c) Apo A-IV

d) Apo A-I

Question 19:

Which apoprotein do chylomicrons require in order to enter the lymph?

a) Apo B-48

b) Apo B-100

c) Apo C

d) Apo E

Question 20:

Where does IDL become LDL?

a) Blood capillaries of the liver

b) Cytosol of liver

c) Heart

d) Mitochondria of liver

Question 21:

A 56-year-old man suffering from uncontrolled diabetes mellitus came for a routine checkup. His lipid profile revealed increased TG (triacylglycerol) and VLDL (very low-density lipoprotein). What is the cause of this abnormal lipid profile?

a) Increased activity of lipoprotein lipase and decreased activity of hormone sensitive lipase

b) Increased activity of hormone sensitive lipase and decreased lipoprotein lipase activity

c) Increase in peripheral function of LDL receptors

d) Increased in activity of hepatic lipase

Answer Key

Question No.	Correct Option
1	a
2	c
3	d
4	b

5	b
6	c
7	a
8	b
9	a
10	a
11	d
12	d
13	a
14	b
15	b
16	a
17	d
18	d
19	a
20	a
21	b

Detailed Explanations

Solution to Question 1:

Cholesterol synthesis takes place in the cytosol and endoplasmic reticulum.

Cytosolic acetyl-CoA forms acetoacetyl-CoA, which condenses with another acetyl-CoA to form HMG-CoA.

Cytosolic HMG-CoA, a key intermediate in cholesterol biosynthesis, is reduced in the endoplasmic reticulum to mevalonic acid by the regulatory enzyme HMG-CoA reductase. Mevalonic acid subsequently undergoes phosphorylation, decarboxylation , condensation and cyclicization reactions to produce cholesterol.

Summary of cholesterol synthesis pathway:

• Synthesis of mevalonate from acetyl-CoA

• Formation of isoprenoid units from mevalonate

• Condensation of six isoprenoid units form squalene

• Cyclization of squalene to give rise to the parent steroid, lanosterol

• Formation of cholesterol from lanosterol.

All tissues containing nucleated cells are capable of cholesterol synthesis, with the majority produced in the liver and intestine.

Solution to Question 2:

Cholesterol is synthesized from acetyl-CoA by a long pathway that may be divided into five steps

• Synthesis of mevalonate from acetyl-CoA

• Formation of isoprenoid units from mevalonate

• Condensation of six isoprenoid units form squalene

• Cyclization of squalene to give rise to the parent steroid, lanosterol

• Formation of cholesterol from lanosterol.

All tissues containing nucleated cells are capable of cholesterol synthesis, with the majority produced in the liver and intestine.

Solution to Question 3:

Deoxycholic acid is not a primary bile acid. It is a secondary bile acid.

The primary bile acids are cholic acid and chenodeoxycholic acid synthesized from cholesterol in the liver. They are next conjugated with glycine and taurine in the liver peroxisomes to form glycocholic, glycochenodeoxycholic, taurocholic, and taurochenodeoxycholic acids.

The secondary bile acids are formed in the intestine as the primary bile acids are deconjugated and dehydroxylated by the intestinal bacteria to form deoxycholic and lithocholic acid.

These are absorbed in the ileum and returned to the liver by enterohepatic circulation.

Solution to Question 4:

The first intermediate in the synthesis of bile acids is 7α-hydroxycholesterol

This reaction is catalyzed by the enzyme 7α-hydroxylase, a microsomal CYP450 enzyme, which requires NADPH, oxygen, and vitamin C as co-factors. It is the rate-limiting step in the synthesis of bile acids.

Regulation of bile acid synthesis occurs by feedback inhibition of 7α-hydroxylase via the Farnesoid X receptor. When the level of bile acids in the enterohepatic circulation increases, FXR is activated and transcription of the enzyme is suppressed.

Other options:

Option A: 7-dehydrocholesterol is an intermediate in the conversion of cholesterol to vitamin D.

Option C: Lanosterol is an intermediate in the synthesis of cholesterol.

Option D: Ergosterol is a sterol found in plants.

Solution to Question 5:

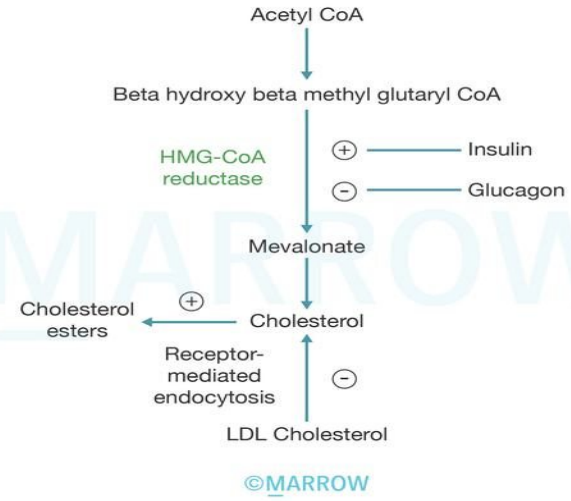
HMG-CoA reductase is the rate-limiting enzyme of cholesterol synthesis.

HMG-CoA reductase is inhibited by the statin group of drugs (e.g. Atorvastatin, Rosuvastatin, Pravastatin, etc.), which are widely used as lipid-lowering agents.

Regulation of HMG CoA reductase:

Glucagon and glucocorticoids repress the transcription of HMG CoA reductase via sterol regulatory element-binding protein (SREBP).

Activity increased by	Activity decreased by
Insulin	Glucagon, glucocorticoids
Thyroid hormone	Cholesterol, mevalonate
	Dietary cholesterol
	Statins



Solution to Question 6:

Lanosterol is the first steroid to be synthesized in cholesterol synthesis.

• Cyclization of squalene gives rise to the parent steroid, lanosterol.

• The formation of cholesterol from lanosterol involves changes in the steroid nucleus and the side chain.

Note: Though lanosterol is technically not a hormone, it is the best choice among the listed options. This is a recent NEET question.

Solution to Question 7:

Lipoproteins transport the majority of non-polar lipids to most tissues for oxidation and to adipose tissue for storage as follows:

• From the intestines as chylomicrons, it carries exogenous triglycerides.

• From the liver as very-low-density lipoproteins (VLDL), it carries endogenous triglycerides.

Solution to Question 8:

As cholesterol is the predominant lipid in LDL and HDL, they have a higher cholesterol: triglyceride ratio.

VLDL also has a significant amount of cholesterol but it predominantly comprises triglycerides (lower cholesterol: triglyceride ratio). Therefore LDL and HDL are considered the major carriers of cholesterol.

IDLs are formed by the removal of triglycerides from VLDL by muscle and adipose tissue and are hence enriched in cholesterol. LDLs are derived from VLDL and IDL particles and contain more cholesterol than IDLs.

Solution to Question 9:

After a fatty meal, chylomicrons are elevated in lipoprotein lipase deficiency.

Chylomicrons transport dietary lipids from the intestine to the peripheral tissues. They are synthesized in the intestine by assembling apo B-48 with lipids (mainly triacylglycerols). This is called a nascent chylomicron, which upon receiving apo C-II and apo E from HDL is remodelled into mature chylomicron.

Apo C-II activates lipoprotein lipase present on the endothelium. Lipoprotein lipase hydrolyzes the triacylglycerols into fatty acids and glycerol which are either used for storage or energy. Therefore, patients who are deficient in lipoprotein lipase or apo C-II (type I hyperlipoproteinemia) show increased levels of chylomicrons in the plasma after a fatty meal.

Solution to Question 10:

Triacylglycerol is predominantly carried by chylomicrons and VLDL.

Solution to Question 11:

Chylomicrons are the largest among plasma lipoproteins, which are especially rich in triacylglycerol.

Solution to Question 12:

HDL plays no role in LDL oxidation.

A major function of HDL is to act as a repository for the apo C and apo E required in the metabolism of chylomicrons and VLDL.

The class B scavenger receptor B1 (SRB-1) has been identified as an HDL receptor with a dual role in HDL metabolism. SRB-1 mediates the acceptance of cholesterol effluxed from the cells by HDL, which then transports it to the liver for excretion via the bile.

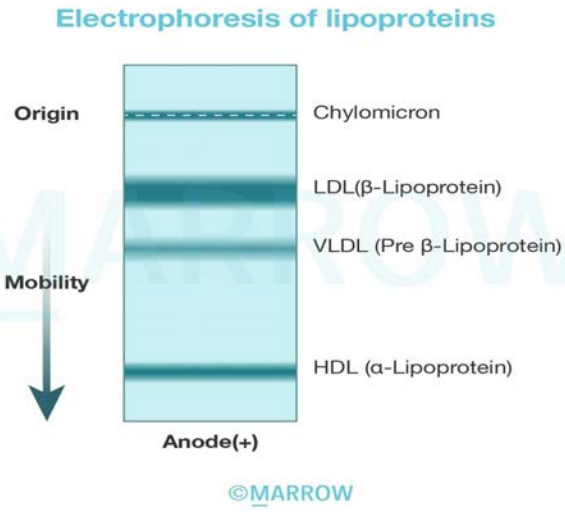
Solution to Question 13:

The given clinical vignette with a wide band at the origin on serum lipid electrophoresis is suggestive of familial hyperchylomicronemia (elevated chylomicron level in blood) which is associated with a deficiency of Lipoprotein Lipase. Familial Hyperchylomicronemia is also associated with hepatosplenomegaly, pancreatitis and eruptive/pruritic xanthoma. Serum biochemistry commonly reveals increased chylomicron, triglyceride and cholesterol levels in the blood.

Chylomicron has the lowest mobility on serum lipid electrophoresis and the chylomicron band appears at the origin location (initial location). On the other hand, HDL has the highest electrophoretic mobility and the least lipid content.

As a general rule, lipoproteins with higher protein content will move faster towards the anode and those with fewer proteins have minimal mobility.

	Chylomicron	VLDL	IDL	LDL	HDL
Electrophoretic mobility	origin	pre-beta	broad-beta	beta	alpha
Protein composition (%)	2	10	20	20	30-60



Note: Even though VLDL has a lesser percentage of proteins with respect to LDL, it moves ahead of LDL closer to the anode due to the presence of more net negative charge in VLDL

Solution to Question 14:

Apolipoprotein A is found in chylomicron and HDL.

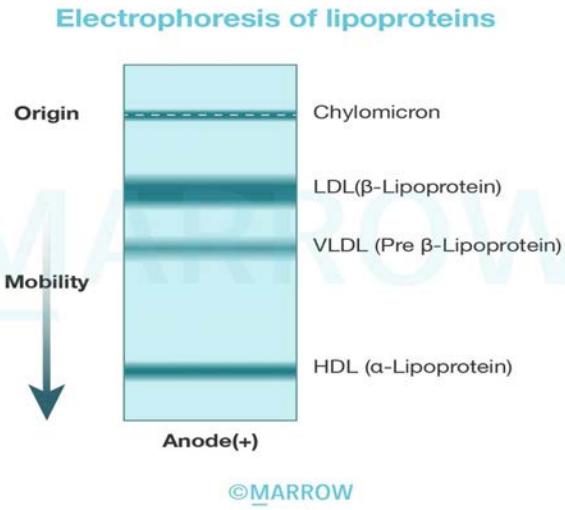
Lipoproteins	Corresponding Apolipoproteins			
Apo-	Apo-	Apo-	Apo-	
HDL	A	C	E	D
Chylomicron	A	C	E	B48
VLDL		C	E	B100
IDL			E	B100
LDL				B100

Solution to Question 15:

HDL has the highest electrophoretic mobility and the least lipid content. HDL particles are the densest.

On electrophoresis, HDL moves farthest on the electrophoretic plate towards the anode. On the other hand, the chylomicron does not move towards the charged end in electrophoresis, thus having the lowest mobility. As a general rule, lipoproteins with higher protein content will move faster towards the anode and those with fewer proteins have minimal mobility.

The density of various lipoproteins and their electrophoretic mobility are shown in the images below:



Solution to Question 16:

The main apolipoprotein of LDL (β-lipoprotein) is apo B(B-100), which is found also in VLDL.

Lipoproteins	Corresponding Apolipoproteins			
Apo-	Apo-	Apo-	Apo-	
HDL	A	C	E	D
Chylomicron	A	C	E	B48
VLDL		C	E	B100
IDL			E	B100
LDL				B100

Solution to Question 17:

Apo B-48 is not the ligand of LDL.

Apo B-48 is the apolipoprotein present on nascent chylomicrons. The main apolipoprotein of LDL (β-lipoprotein) is apo B(B-100), which is found also in VLDL.

Low-density lipoproteins (LDL) deliver cholesterol to the tissues, hence it is a marker of cardiovascular disease.

Lipoproteins	Corresponding Apolipoproteins			
Apo-	Apo-	Apo-	Apo-	
HDL	A	C	E	D
Chylomicron	A	C	E	B48
VLDL		C	E	B100
IDL			E	B100
LDL				B100

Solution to Question 18:

Apo A-I is the activator of Lecithin: cholesterol acyltransferase (LCAT), the enzyme present in HDL facilitating reverse cholesterol transport.

Lecithin-cholesterol acyltransferase (LCAT) is activated by apo A-I which then converts free cholesterol(amphipathic) in HDL to cholesterol ester(hydrophobic). As a result the initial discoidal shape now becomes spherical.

This facilitates reverse cholesterol transport.

Clinical correlation: Familial LCAT deficiency will lead to failure of reverse cholesterol transport resulting in cholesterol deposition in specific peripheral tissues - the cornea, kidneys, and erythrocytes. Major complications include premature atherosclerosis, renal failure, anemia and corneal opacities. HDL cholesterol level is decreased.

Solution to Question 19:

Each nascent chylomicron particle has one molecule of apolipoprotein(apo) B-48 with which chylomicrons are released from the cells into the lymphatic system and travel to the blood, where they receive apo C-II and apo E from HDLs.

Solution to Question 20:

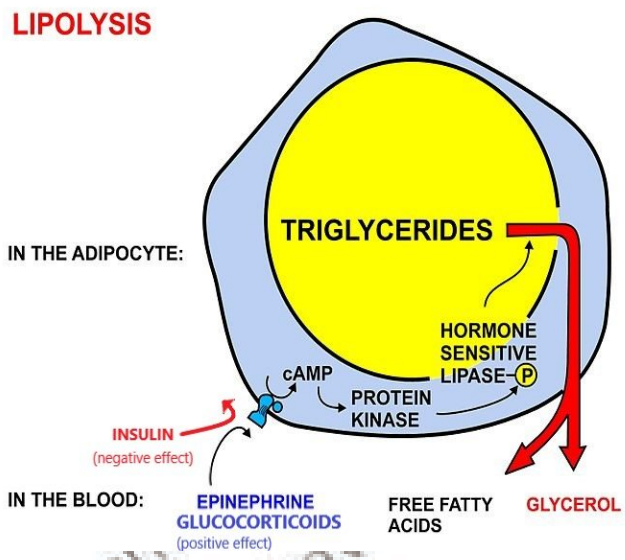
IDL (Intermediate-density lipoprotein) particles are rapidly taken up by the liver in circulation, where they undergo further triglyceride hydrolysis and are converted to LDL (Low density lipoprotein).

Solution to Question 21:

In chronic diabetes, the values of TG and VLDL are increased due to an increase in hormone-sensitive lipase and a decrease in lipoprotein lipase.

Insulin has an inhibitory effect on hormone-sensitive lipase (HSL). In diabetes, low insulin corresponds to an increase in HSL activity. This promotes the breakdown of lipids and the release of TG and free fatty acids from the adipose tissue.

Insulin induces the activity of lipoprotein lipase (LPL) which is present on the capillary walls. LPL promotes hydrolysis of TG and the degradation of the transport proteins, VLDL, and chylomicrons. In diabetes, low insulin leads to decreased LPL activity which corresponds to an increase in VLDL and TG.



Porphyrins and bile pigments

Question 1:

What is the porphyrin present in heme?

a) Protoporphyrin

b) Uroporphyrin

c) Coproporphyrin

d) Benzoporphyrin

Question 2:

Which of the following is not a site for heme synthesis?

a) Osteocyte

b) Liver

c) Erythroid cells of bone marrow

d) Mature erythrocyte

Question 3:

Which of the following TCA cycle intermediate is a part of heme metabolism?

a) Alpha ketoglutarate

b) Fumarate

c) Succinyl CoA

d) Malate

Question 4:

Which step of heme synthesis requires pyridoxal phosphate?

a) Succinyl CoA to α-amino-β-ketoadipate

b) α-Amino-β-ketoadipate to ALA

c) ALA to porphobilinogen

d) Porphobilinogen to hydroxymethylbilane

Question 5:

Which of the following is the rate-limiting enzyme in the hepatic biosynthesis of heme?

a) ALA Synthase

b) Uroporphyrinogen III synthase

c) ALA dehydratase

d) Ferrochelatase

Question 6:

A 44-year-old man with a past medical history of acute intermittent porphyria is planned for neurosurgery. Considering the risk of potential complications, the anaesthetist decides not to use barbiturates for induction. What is the biochemical basis behind the worsening of porphyria by barbiturates?

a) Repression of ALA synthase

b) De-repression of ALA synthase

c) Inhibiton of ferrochelatase

d) None of the above

Question 7:

A young patient came with complaints of acute abdominal pain on and off and tingling sensation of limbs and at times weakness of limbs. She had a history of eating paint from the wall of a newly built house. Which of the following enzyme defect will be the cause of her condition?

a) ALA synthase

b) ALA dehydratase

c) Heme synthase

d) Coproporphyrinogen oxidase

Question 8:

What is the by-product of the ■reaction catalyzed by cytosolic hydroxymethylbilane synthase in heme synthesis?

a) H_2O

b) NH_3

c) CO_2

d) CoA.SH

Question 9:

Which of the following enzymes catalyzes the cyclization of hydroxymethylbilane?

a) Uroporphyrinogen I synthase

b) Uroporphyrinogen II synthase

c) Uroporphyrinogen III synthase

d) Uroporphyrinogen decarboxylase

Question 10:

Which of the following porphyrias is not inherited in an autosomal dominant mode?

a) Erythropoietic protoporphyria

b) Variegate porphyria

c) Acute intermittent porphyria

d) Hereditary coproporphyria

Question 11:

A 50-year-old man presents with a history of fatigue, dark-coloured urine and photosensitivity. He consumes alcohol regularly. On examination, hyperpigmentation and scarring are present on the face and dorsum of the hand. Lab findings are given below. Which of the following is the most likely diagnosis?

a) Acute intermittent porphyria

b) Porphyria cutanea tarda

c) Hereditary coproporphyria

d) Variegate porphyria

Question 12:

A middle-aged female patient presented with intermittent abdominal pain, nausea, vomiting, constipation and bilateral lower limb numbness for 3 days. On examination, she is thin and pale. Her vitals and lab findings are given below. Which of the following is the most likely diagnosis?

a) Hereditary coproporphyria

b) Acute intermittent porphyria

c) Porphyria cutanea tarda

d) Variegate porphyria

Question 13:

Which of the following is not a hepatic porphyria?

a) Variegate porphyria

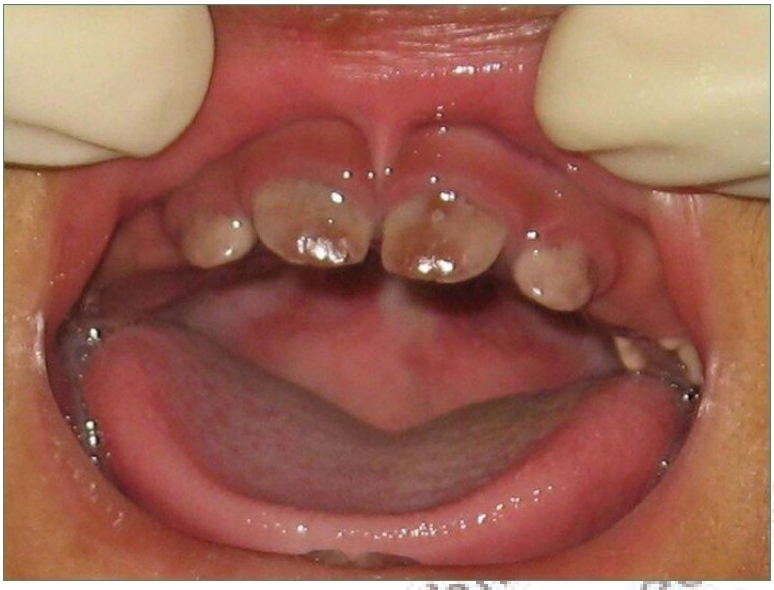
b) Acute intermittent porphyria

c) Porphyria cutanea tarda

d) Erythropoietic protoporphyria

Question 14:

An infant presented with delayed eruption of her teeth. An image of her teeth are shown below. She also has history of blisters over the face, hands and feet on exposure to sunlight since the first week of birth. What is the most likely diagnosis?



a) Congenital erythropoietic porphyria

b) Porphyria cutanea tarda

c) Erythropoietic protoporphyria

d) Acute intermittent porphyria

Question 15:

Which of the following porphyria is caused by a mutation of ALA synthase-2?

a) Acute intermittent porphyria

b) Congenital erythropoietic porphyria

c) X- linked protoporphyria

d) Erythropoietic protoporphyria

Question 16:

Which of the following enzyme is defective in variegate porphyria?

a) Coproporphyrinogen oxidase

b) Protoporphyrinogen oxidase

c) Uroporphyrinogen III synthase

d) Uroporphyrinogen decarboxylase

Question 17:

Carbon monoxide is released in which of the following reactions?

a) Conversion of biliverdin to bilirubin

b) Conversion of heme to biliverdin

c) Conversion of bilirubin to bilirubin monoglucuronide

d) Conversion of bilirubin monoglucuronide to bilirubin diglucuronide

Question 18:

Which of the following is the site of bilirubin conjugation within the cell?

a) Cytoplasm

b) Endoplasmic reticulum

c) Mitochondria

d) Golgi apparatus

Question 19:

Which of the following is the rate-limiting step for hepatic bilirubin metabolism?

a) Transport of bilirubin with albumin

b) Uptake of bilirubin by liver

c) Conjugation of bilirubin

d) Secretion of conjugated bilirubin into the bile

Question 20:

In a patient with jaundice which of the following tests can be used to determine serum bilirubin?

a) Ehrlich's test

b) Van den Bergh test

c) Hay's test

d) Benedict's test

Question 21:

A 65 year old man was admitted with generalised weakness, easy fatigability, itching and jaundice since 1 month. On examination, he was emaciated and had scratch marks all over the body. A mass was palpable in the upper abdomen. Which of the following can be seen on Van den Bergh reaction?

a) Direct positive

b) Indirect positive

c) Biphasic reaction

d) Negative

Question 22:

An adult male patient presents with yellow discolouration of the sclera. There was a history of recurrent mild jaundice since childhood. His lab values are mentioned below. A liver biopsy was performed which revealed normal liver architecture with coarse granular black pigmentation. The mutation of which protein is involved in the following condition?

a) OATP1B1

b) BSEP

c) MRP-3

d) MRP-2

Question 23:

A young adult presents with yellowish discolouration of the skin and sclerae for the last 3 days. He is otherwise asymptomatic. He had a viral fever one week ago which has now resolved. Investigations reveal unconjugated hyperbilirubinemia. Which of the following is true regarding this patient's condition?

a) BSEP protein is mutated

b) Active transport of bilirubin is impaired due to MRP-2 mutation

c) Promoter defect of UGT1A1 enzyme is the most common cause

d) Missense mutation of transferase enzyme is the most common cause

Question 24:

Which of the following is false regarding jaundice?

a) Urine bilirubin is absent in hemolytic jaundice

b) Urine urobilinogen is increased in obstructive jaundice

c) Criggler-Najjar syndrome presents with unconjugated hyperbilirubinemia

d) Hepatocellular jaundice can have normal or decreased urine urobilinogen

Answer Key

Question No.	Correct Option
1	a
2	d
3	c
4	a
5	a
6	b
7	b
8	b
9	c
10	a
11	b
12	b
13	d
14	a
15	c
16	b
17	b
18	b
19	d
20	b
21	a
22	d
23	c
24	b

Detailed Explanations

Solution to Question 1:

The porphyrin present in heme is protoporphyrin.

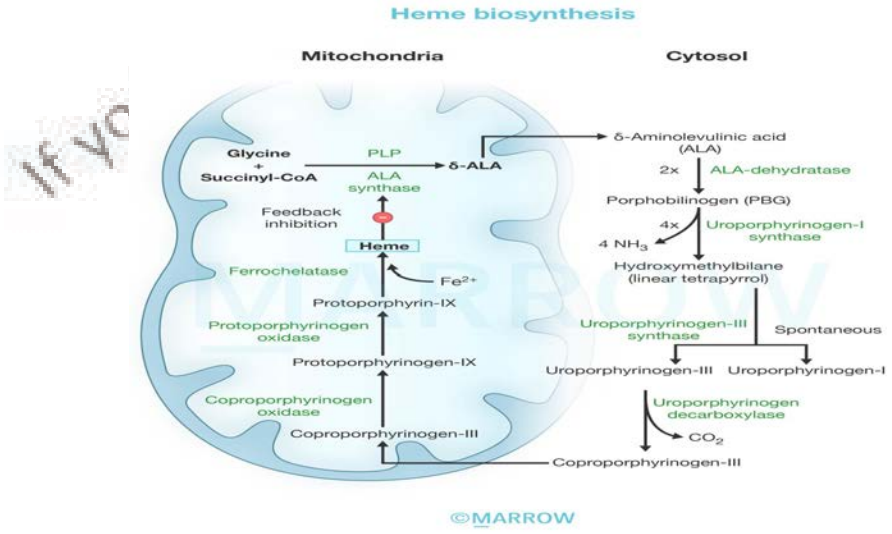
Heme is ferroprotoporphyrin. Iron is present in the ferrous (Fe^{2+}) state in heme. Porphyrins are cyclic coloured compounds formed by the linkage of four pyrrole rings through methyne or methenyl (=CH-) bridges. There are three types of porphyrins:

- Uroporphyrin (most water-soluble).
- Coproporphyrin (intermediate).
- Protoporphyrin (least water-soluble).

Solution to Question 2:

Mature erythrocyte is not a site for heme biosynthesis.

Approximately 85% of heme synthesis occurs in erythroid precursor cells in the bone marrow and the majority of the remainder in hepatocytes. Heme biosynthesis occurs both in mitochondria and cytoplasm. It is initiated by the condensation of succinyl–CoA and glycine in a reaction catalyzed by mitochondrial ALA synthase. This initial reaction, and the last three steps in the formation of heme occur in mitochondria, whereas the intermediate steps of the biosynthetic pathway occur in the cytosol.

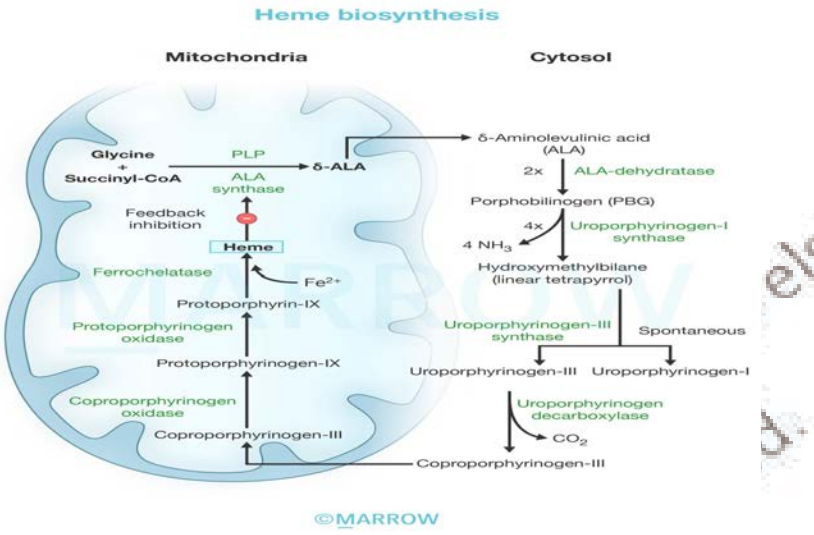


Solution to Question 3:

Succinyl Co-A, an essential part for heme biosynthesis, is an intermediate of the TCA cycle.

The synthesis of heme begins by condensation of succinyl Co-A and glycine to form α-amino-β-ketoadipate, which is then converted to δ-aminolevulinate. These steps are mediated by the enzyme ALA synthase in the mitochrondria.

Various steps involved in the synthesis of heme is illustrated in the image given below:



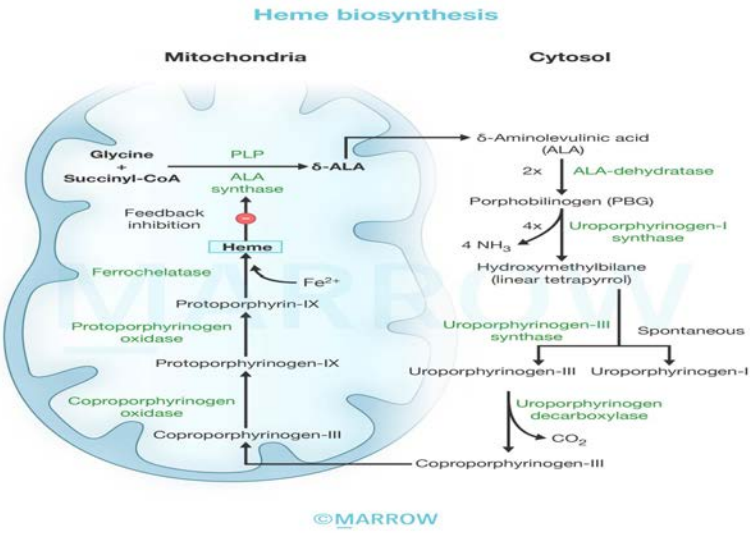
Solution to Question 4:

Conversion of succinyl CoA to α-amino-β-ketoadipate requires pyridoxal phosphate.

The first step of heme synthesis is the conversion of succinyl CoA to α-amino-β-ketoadipate. This reaction is dependent on pyridoxal phosphate. α-amino-β-ketoadipate is rapidly decarboxylated to produce δ -aminolevulinate. It is catalyzed by mitochondrial ALA synthase.

Succinyl-CoA + glycine ° α-amino-β-ketoadipate + CoA-SH ° δ-aminolevulinate + CO2

Various steps involved in the synthesis of heme is illustrated in the image given below:

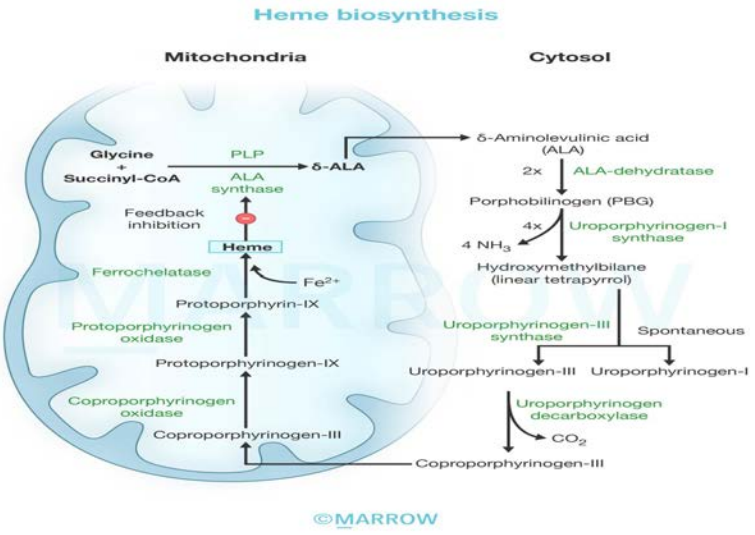


Solution to Question 5:

The rate-limiting enzyme in the hepatic biosynthesis of heme is mitochondrial ALA synthase.

ALA synthase (ALAS) is a mitochondrial enzyme catalysing the conversion of succinyl CoA and glycine to ALA. There are two isozymes of ALA synthase. ALAS-1 is expressed throughout the body. It is induced by drugs whose metabolism requires cytochromes (hemoprotein). Heme, probably acting through an aporepressor molecule, acts as a negative regulator of the synthesis of ALAS-1 i.e. ALAS-1 synthesis decreases in the presence of heme.

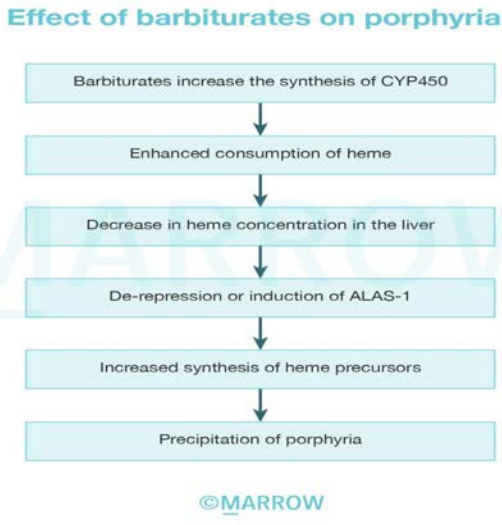
ALAS-2 is expressed in the erythrocyte precursor cells. It is neither induced by drugs nor is its feedback regulated by heme.



Solution to Question 6:

The biochemical basis of worsening of porphyria by barbiturates is due to de-repression of ALA synthase. De-repression is the removal of repression of an enzyme, or the induction of a blocked enzyme.

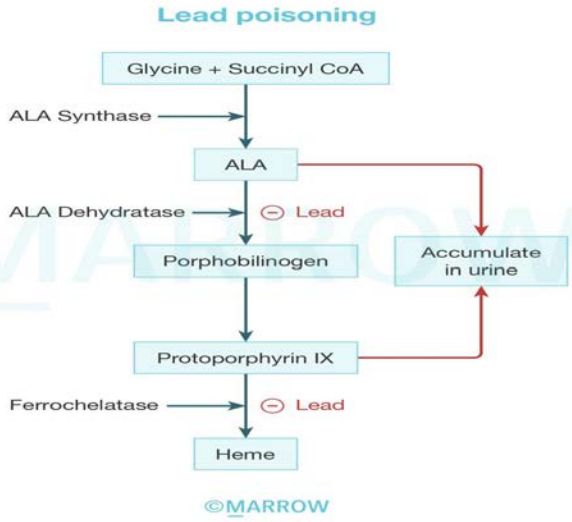
Barbiturates are metabolized by microsomal CYP450, a heme-protein oxidase system found in the liver. In patients with porphyria, barbiturates precipitate an attack of porphyria by the following mechanism:



Solution to Question 7:

A child with acute abdominal pain, tingling sensation and weakness of limbs with history of eating paint likely has lead poisoning. It is characterized by impaired ALA dehydratase enzyme activity.

Lead is a potent inhibitor of both ALA dehydratase and ferrochelatase/ heme synthase (option C). However, it has the greatest inhibitory action on ALA dehydratase. Lead poisonining thus leads to the buildup of protoporphyrin and ALA. Lead also substitutes for calcium in many fundamental cellular processes. It can cross red blood cell membranes as well as the blood-brain barrier and enter the neuroglial cells which support brain function. The fatal dose of lead is 20g of lead acetate and the fatal period is 1 to 2 days.



Other options:

Option A: ALA synthase 2 defect causes X-linked protoporphyria.

Option C: Heme synthase, or ferrochelatase, defect causes erythropoeitic protoporphyria.

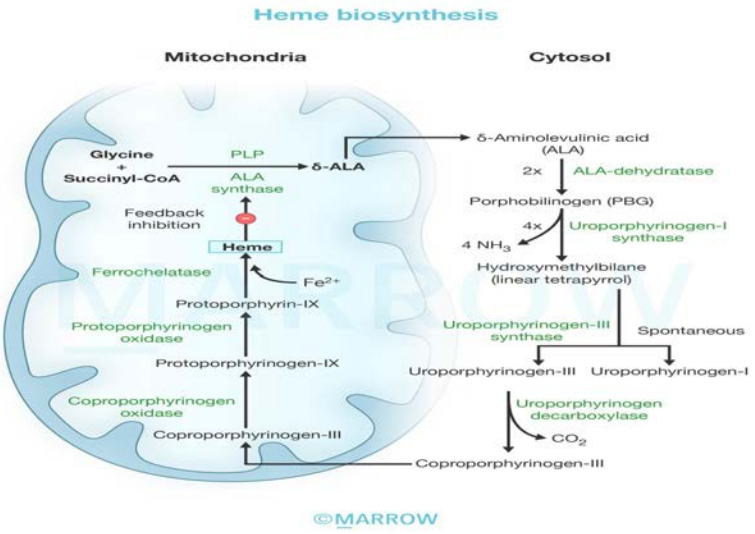
Option D: Coproporphyrinogen oxidase defect causes hereditary corproporphyria.

Solution to Question 8:

The by-product of the reaction catalyzed by cytosolic hydroxymethylbilane (HMB) synthase in heme synthesis is ammonia (NH_3).

HMB synthase is also known as porphobilinogen deaminase or uroporphyrinogen 1 synthase. It catalyses the condensation of 4 molecules of porphobilinogen to release hydroxymethylbilane and 4 molecules of NH_3.

4 Porphobilinogen + H_2O -------> Hydroxymethylbilane + 4 NH_3

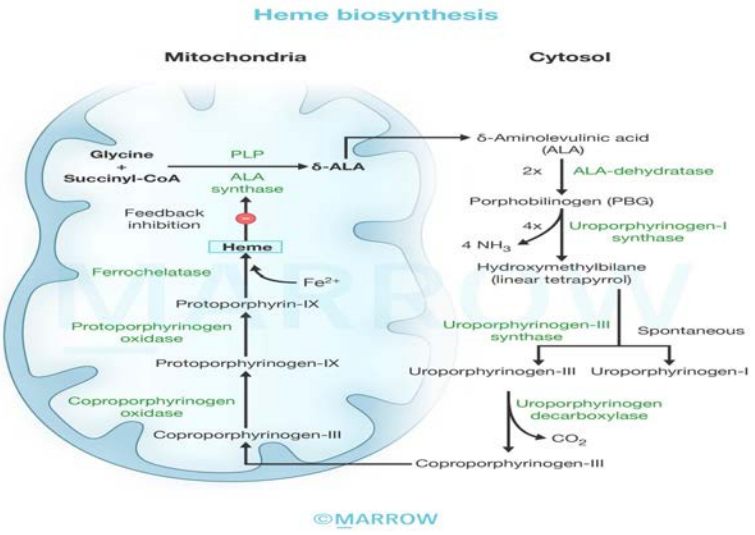


Solution to Question 9:

Cyclization of hydroxymethylbilane (HMB) is catalyzed by uroporphyrinogen III synthase

HMB is cyclised to uroporphyrinogen III by uroporphyrinogen III synthase. Uroporphyrinogen III is the first porphyrin precursor in heme synthesis.

Note: HMB can also cyclize spontaneously to form uroporphyrinogen I. But under normal circumstances, the porphyrinogen formed is predominantly uroporphyrinogen III.



Solution to Question 10:

Erythropoietic protoporphyria is not transmitted in an autosomal dominant manner..

Erythryopoietic protoporphyria is an autosomal recessive disease caused by a defect in the enzyme ferrochelatase (FECH). It is the most common porphyria in children, who present with non-blistering photosensitivity. An increase in the ratio of free protoporphyrin to zinc bound porphyrin in erythrocytes is the hallmark of this disease. The diagnosis is made by FECH mutation analysis.

Note: Most porphyrias are autosomal dominant except:

• ALA dehydratase deficiency porphyria - autosomal recessive

• Congenital erythropoietic porphyria - autosomal recessive

• Erythropoietic protoporphyria - autosomal recessive

• X-linked protoporphyria

Solution to Question 11:

A middle-aged adult presenting with features of hemolysis and photosensitivity with elevated urine uroporphyrin suggests a diagnosis of porphyria cutanea tarda.

Porphyria cutanea tarda (PCT) is the most common porphyria. It is caused by the deficiency of uroporphyrinogen decarboxylase, which converts uroporphyrinogen III to coproporphyrinogen III. It is characterized by photosensivity, which manifests as blistering skin lesions on the back of the hands with crusting and scarring. It is associated with hemochromatosis and patients may develop chronic liver disease. There is increased excretion of uroporphyrin in the urine.

Solution to Question 12:

An adult female with intermittent abdominal pain, nausea, vomiting, constipation, bilateral lower limb numbness with elevated urine ALA and porphobilinogen is most likely to have acute intermittent porphyria (AIP).

AIP is a type of hepatic porphyria characterized by neurovisceral symptoms. AIP is caused by a defect in hydroxymethylbilane synthase, which converts prophobilinogen to hydroxymethylbilane. Patients present with abdominal pain, tachycardia, hypertension and peripheral neuropathy. There is increased plasma and urinary ALA and prophobilinogen. Darkening of urine on exposure to air also occurs.

Other options:

Options A and D: Hereditary coproporphyria and variegate porphyria present with both photosensitivity and neurovisceral symptoms. The porphyrin intermediates appearing in urine help differentiate the two diseases.

Option C: Porphyria cutanea tarda presents with blistering photosensitivity with vesicles and bullae formation.

Solution to Question 13:

Erythropoietic protoporphyria is not a type of hepatic porphyria.

Porphyrias are classified into erythropoietic and hepatic porphyrias, based on the location of accumulation of heme pathway intermediates:

• Erythropoietic porphyrias:

• The overproduction of heme pathway intermediates occurs primarily in bone marrow erythroid cells.

• They are characterized by cutaneous symptoms and appear in early childhood.

• Examples: Congenital erythropoietic porphyria, erythropoietic protoporphyria, X-linked protoporphyria

• Hepatic porphyrias:

• The overproduction of heme pathway intermediates occurs primarily in the liver.

• Acute hepatic porphyrias are characterised by neurologic symptoms and present acutely. Examples: Acute intermittent porphyria, hereditary coproporphyria, variegate porphyria, ALA dehydratase deficient porphyria

• Chronic hepatic porphyrias present in adulthood with photosensitivity. Example: Porphyria cutanea tarda

Solution to Question 14:

An infant with delayed eruption and reddish discoloration of teeth, and blisters over sun-exposed areas suggests a diagnosis of congenital erythropoietic porphyria.

Congenital erythropoietic porphyria, also is known as Gunther's disease, is an autosomal recessive porphyria. There is uroporphyrinogen III synthase deficiency, leading to uroporphyrin I and coproporphyrin I accumulation in teeth, bone marrow, erythrocyte, plasma urine and feces.

Infants present with port wine urine or pink stained diapers, severe cutaenous hypersensitivity, and reddish-brown teeth known as erythrodontia. When illuminated with UV light, the teeth show reddish fluorescence. It may cause non-immune hydrops in utero or shortly after birth in severe cases.

Solution to Question 15:

X-linked protoporphyria is caused by a mutation of ALA synthase-2.

X-linked Protoporphyria is caused by a gain-of-function mutation, leading to the increased activity of ALA synthase-2. This results in increased ALA synthesis. Due to the saturation of iron, protoporphyrins are not converted to heme and thus protoporphyrins accumulate.

ALA synthase-2 mutation is also seen in X-linked sideroblastic anemia. However, this is not a porphyria. X-linked sideroblastic anemia is due to loss-of-function mutation, leading to decreased

activity of ALA synthase-2 and decreased protoporphyrin synthesis.

Solution to Question 16:

Protoporphyrinogen oxidase enzyme is defective in variegate porphyria.

Variegate porphyria is an autosominal dominant porphyria. Protoporphyrinogen IX accumulates in the urine. Patients present with acute attacks of abdominal pain and peripheral neuropathy. Cutaneous photosensitivity is also seen.

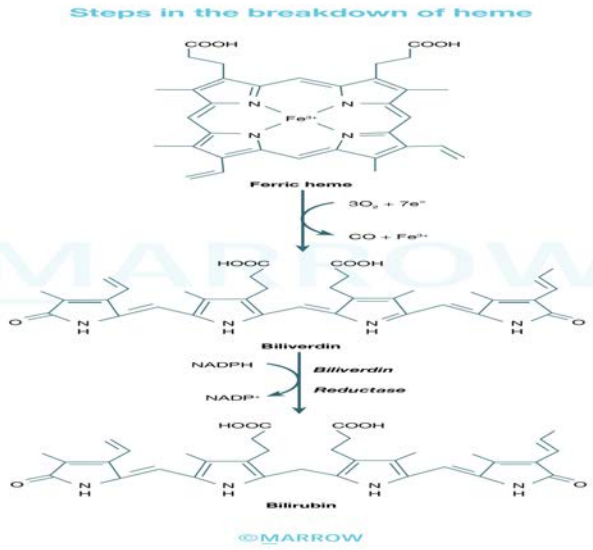
Solution to Question 17:

The conversion of heme to biliverdin produces carbon monoxide.

Heme oxygenase catabolises heme to biliverdin. Carbon monoxide and ferric iron are released in the process. This is the only source of endogenous carbon monoxide in the body. Biliverdin reductase further converts biliverdin to bilirubin in an NADPH-dependent reaction. Iron produced from this reaction is stored as ferritin.

The process of heme catabolism occurs in the microsomal fraction (endoplasmic reticulum) of reticuloendothelial cells.

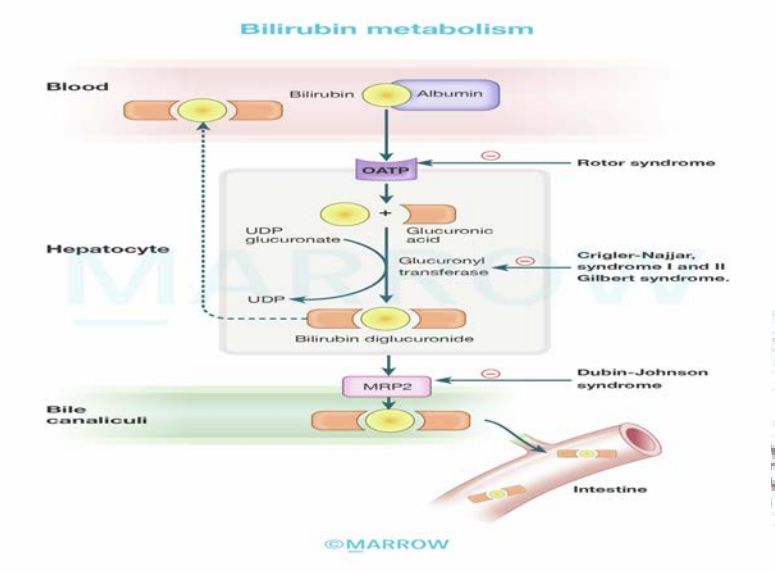
Note: 1 g of hemoglobin produces 35 mg of bilirubin. About 250-350 mg bilirubin is produced daily in the adults.



Solution to Question 18:

The endoplasmic reticulum is the site for the conjugation of bilirubin.

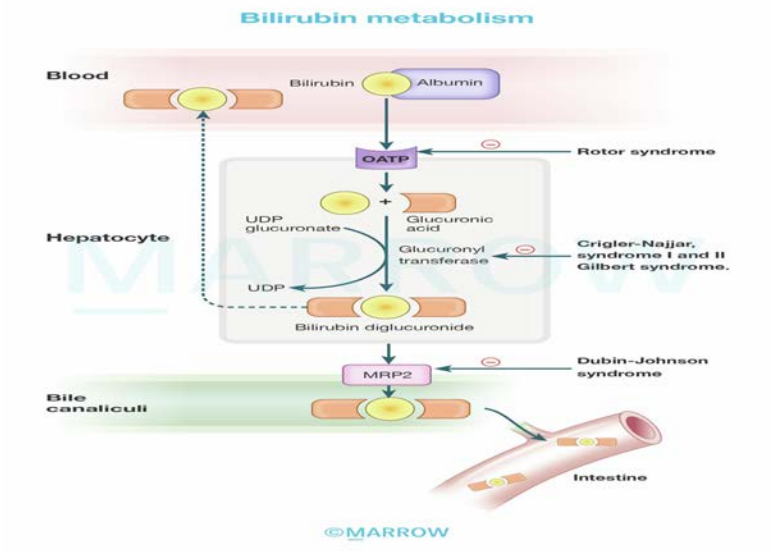
Hepatocytes convert bilirubin to a polar form by a process called conjugation. UDP-glucuronic acid acts as a glucuronyl donor in this reaction. It initially produces bilirubin monoglucuronide. This is subsequently converted to bilirubin diglucuronide which is excreted in bile. Conjugation is catalysed by the enzyme glucuronyl transferase, which is present in the endoplasmic reticulum.



Solution to Question 19:

The secretion of conjugated bilirubin into the bile is the rate-limiting step for hepatic bilirubin metabolism.

The secretion of conjugated bilirubin into the bile occurs by an active transport mechanism, which involves multidrug resistance-associated protein 2 (MRP-2), also called multispecific organic anion transporter (MOAT). It is located in the plasma membrane of the bile canaliculi.



Solution to Question 20:

Van den Bergh test can be used to determine the serum bilirubin level in a patient with jaundice.

Van den Bergh reaction is a colorimetric method to measure serum bilirubin. It is based on the formation of red-purple azodipyrroles when diazotized sulfanilic acid reacts with bilirubin.

Option A: Ehrlich's test is used to determine urinary and fecal urobilinogen.

Option C: Hay's test is used for bile salts in the urine.

Option D: Benedict's test is used to detect the presence of reducing sugars.

Clinical condition	Reaction with reagent	Interpretation
Normal serum	No reaction	Negative
HighconjugatedbilirubinEg: Obstructive jaundice	Reacts in the absence of met hanol	Directpositive
HighunconjugatedbilirubinE g: Hemolytic anemia	Reacts in the presence of met hanol	Indirectpositiv e
Highconjugatedandunconjug atedbilirubinEg: Hepatic jaun dice	Reacts strongly both with an d without methanol	Biphasicreacti on

Solution to Question 21:

Generalised weakness, easy fatigability, itching and jaundice with a palpable upper abdominal mass is suggestive of pancreatic malignancy leading to obstructive jaundice. This gives a direct positive result on Van den Bergh reaction.

In obstructive jaundice, obstruction of the biliary tree causes regurgitation of conjugated bilirubin into the blood. Patients present with itching, jaundice and pale coloured stools.

Van den Bergh reaction is a colorimetric method to measure the serum bilirubin level. It is based on the formation of red azodipyrroles when diazotized sulfanilic acid reacts with bilirubin.

Clinical condition	Reaction with reagent	Interpretation
Normal serum	No reaction	Negative
Highconjugatedbilirubineg:O bstructive jaundice	Reacts in the absence of met hanol	Directpositive
Highunconjugatedbilirubineg: Hemolytic anemia	Reacts in the presence of met hanol	Indirectpositiv e
Highconjugatedandunconjug atedbilirubineg: Hepatic jaun dice	Reacts strongly both with an d without methanol	Biphasicreacti on

Solution to Question 22:

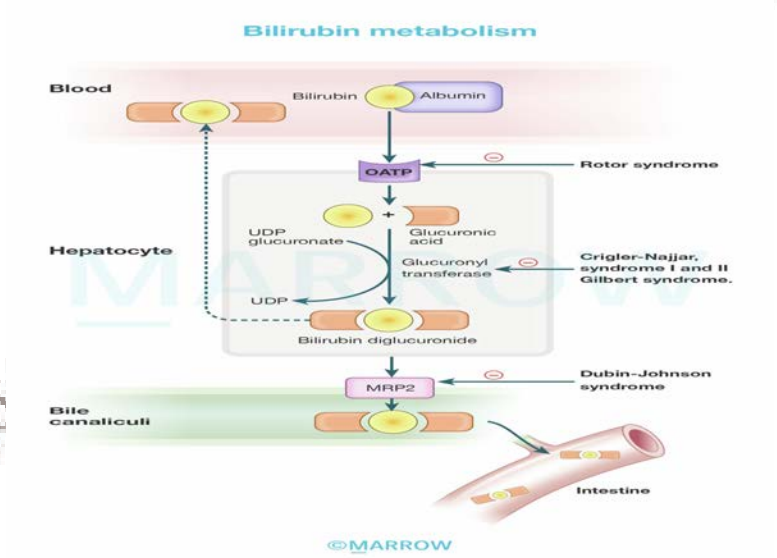
Yellow discolouration of the sclera with coarse granular black pigmentation of the liver is suggestive of Dubin–Johnson syndrome, characterized by a mutation in the MRP-2 protein.

Dubin–Johnson syndrome is an autosomal recessive disorder with predominantly conjugated hyperbilirubinemia. It is due to a defect in the active transport of bilirubin across the canalicular membrane, due to a mutation in the MRP-2 protein. It is also known as black liver jaundice due to the accumulation of a dark pigment, derived from epinephrine metabolites, in the lysosomes of centrilobular hepatocytes. Bromosulpthalein test on plasma shows 2 peaks.

Option A: OATP1B1 and OATP1B3 are mutated in Rotor syndrome. It is due to defective bilirubin excretion and causes conjugated hyperbilirubinemia.

Option B: BSEP protein is mutated in progressive familial intrahepatic cholestasis (PFIC type-2) and benign recurrent intrahepatic cholestasis (BRIC-2).

Option C: MRP-3 is mutated in PFIC type-3.

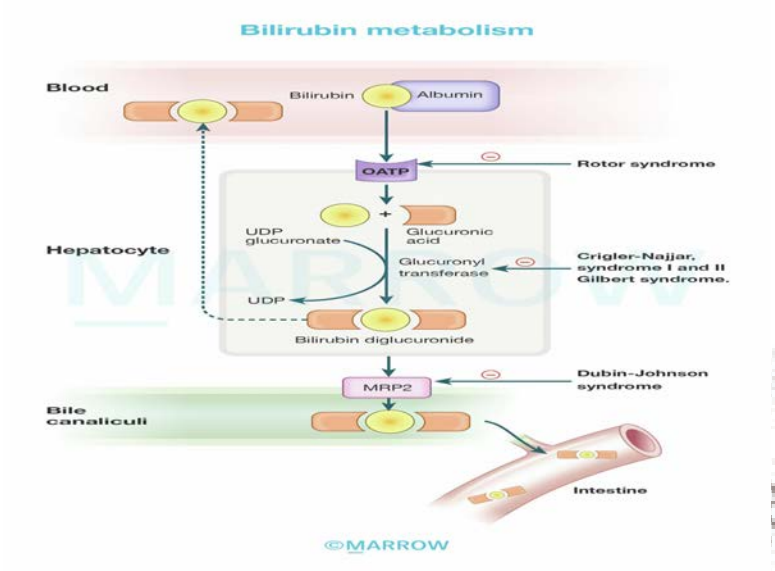


Solution to Question 23:

A young adult with a short history of jaundice following a period of stress (viral fever) with unconjugated hyperbilirubinemia likely has Gilbert syndrome. Gilbert syndrome most commonly occurs due to promoter defect of the UGT1A1 enzyme.

Gilbert syndrome is caused by a defect in the promoter region of the gene coding for the UDP-glucuronosyl transferase (UGT1A1) enzyme. This results in a variable rate of transcription of the normal enzyme. UGT1A1 activity is reduced to 10-33% of normal. It presents with periods of mild unconjugated hyperbilirubinemia with other liver function tests being normal. The disorder is precipitated by stress, alcohol and reduced caloric intake.

Liver histology shows increased lipofuscin pigment. It responds well to phenobarbitone. Gilbert's syndrome doesn't require treatment. The bilirubin levels may fluctuate over time, and patients may occasionally have jaundice, which usually resolves on its own with no ill effects.



Solution to Question 24:

Urine urobilinogen is not increased in obstructive jaundice.

Obstructive jaundice has the following features:

• Absent urinary urobilinogen: In complete obstruction of the biliary tree, conjugated bilirubin cannot enter the intestine. Thus it is not metabolized by the intestinal flora and urobilinogen is not formed.

• Increased serum direct bilirubin: Due to obstruction in the biliary tree, the bilirubin conjugates diffuse into the blood causing conjugated hyperbilirubinemia.

• Increased urine bilirubin: Conjugated bilirubin is water-soluble and thus appears in the urine in excess amounts.

Option A: Urine bilirubin is absent in hemolytic jaundice because unconjugated bilirubin is not water-soluble.

Option C: Criggler-Najjar syndrome, due to absent UGT1A1 activity, presents with raised unconjugated bilirubin.

Option D: Hepatocellular jaundice can have decreased urine urobilinogen if micro-obstruction occurs due to swollen, damaged hepatocytes.

Enzymes - Mechanism of Action & Clinical Importance

Question 1:

Which of the following is false regarding the properties of enzymes?

a) Enzymes are stereospecific catalysts

b) Enzymes are neither consumed nor permanently altered in a reaction

c) Most of them are proteins

d) They are heat-stable

Question 2:

Which of the following is not a mechanism of enzymes to enhance rate of reaction?

a) Catalysis by proximity

b) Acid-Base catalysis

c) Catalysis by strain

d) Catalysis by increasing the activation energy

Question 3:

Which of the following is the prosthetic group present in the enzyme dinitrogenase ?

a) Molybdenum

b) Zinc

c) Manganese

d) Copper

Question 4:

Which of the following is not a copper-containing enzyme?

a) Mitochondrial superoxide dismutase

b) Cytosolic superoxide dismutase

c) Tyrosinase

d) Lysyl oxidase

Question 5:

Which of the following is not a coenzyme?

a) Lipoic acid

b) ATP

c) Vitamin K

d) S-adenosyl methionine

Question 6:

Which of the following is false?

a) Cofactors have functions similar to those of prosthetic groups.

b) Cofactors bind in a transient, dissociable manner to enzymes.

c) The most common cofactors are metal ions.

d) Enzymes that use metal ion as a cofactor are termed as metalloenzymes.

Question 7:

Which of the following enzyme is not involved in oxidation-reduction reactions?

a) Dehydrogenase

b) Hydrolase

c) Peroxidase

d) Oxygenase

Question 8:

Which of the following is not a mono-oxygenase?

a) Tryptophan pyrrolase

b) Tryptophan hydroxylase

c) Tyrosine hydroxylase

d) Cytochrome p450

Question 9:

Which of the following is false about the function of oxygenases?

a) Can incorporate 2 atoms of oxygen in a substance

b) Can incorporate 1 atom of oxygen in a substance

c) Important in hydroxylation of steroids

d) Catalyze carboxylation of drugs

Question 10:

Which of the following is a lyase?

a) Aldolase B

b) Acetyl-CoA Carboxylase

c) Fatty Acyl- CoA Dehydrogenase

d) Acetyl-CoA Synthetase

Question 11:

The group of enzymes that catalyze the joining together of two molecules is called _____.

a) Transferases

b) Ligases

c) Oxidoreductases

d) Lyases

Question 12:

Which of the following is not a non functional enzyme?

a) Lipoprotein lipase

b) Alkaline phosphatase

c) Lactate Dehydrogenase

d) Gamma glutamyltranspeptidase

Question 13:

All of the following is true about isoenzymes except _____.

a) They catalyze the same reaction.

b) They have the same number of charged amino acids.

c) They can be separated by electrophoresis.

d) They differ in tissue localization.

Question 14:

A 20-year old woman presented with generalised weakness and lightheadedness for 2 weeks. Her peripheral smear showed RBC fragments and blood investigations were notable for decreased haemoglobin, increased indirect bilirubin and LDH. Which of the following isoenzymes of LDH is likely to be raised?

a) LDH 3

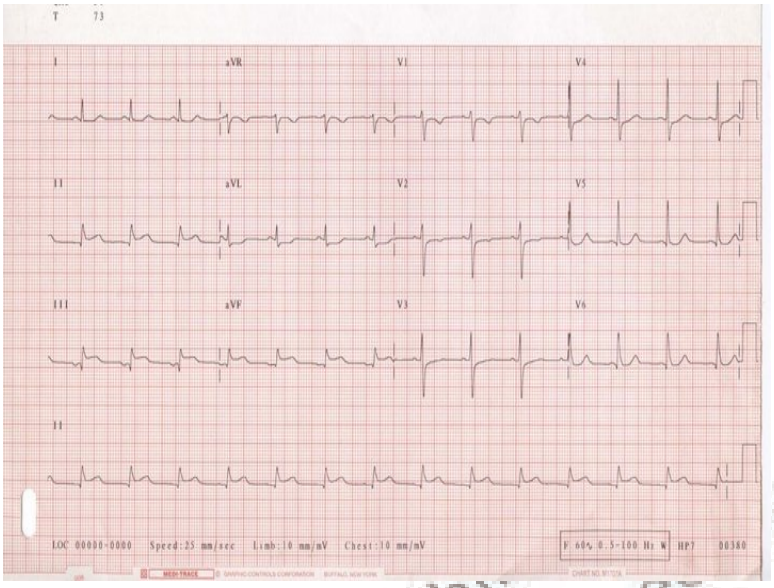
b) LDH 4

c) LDH-5

d) LDH-2

Question 15:

A 54-year-old man who is a known case of diabetes mellitus and hypertension experiences severe, sudden-onset chest pain radiating to the left arm and profuse sweating. He was admitted to the emergency ward. An ECG is taken which is as shown below. Which of the following is true regarding blood lactate dehydrogenase levels in this scenario?



a) LDH 1 > LDH 2

b) LDH2 > LDH1

c) LDH 2 > LDH 3

d) LDH 3 > LDH 2

Question 16:

A 40-year-old man was admitted with complaints of fever, anorexia, vomiting, abdominal pain and dark urine since 6 days. On investigation, he had elevated liver enzymes and serology was positive for IgM hepatitis A antibodies. Which of the following liver enzymes is most specific for his condition?

a) Alkaline phosphatase (ALP)

b) Aspartate aminotransferase (AST)

c) Alanine aminotransferase (ALT)

d) Gamma glutamyl transferase (GGT)

Question 17:

Which of the following is not a serine protease?

a) Chymotrypsinogen

b) Plasmin

c) Prostate specific antigen

d) Elastase

Question 18:

Elastase acts on the carboxy-terminal of which amino acid?

a) Arginine

b) Alanine

c) Tryptophan

d) Phenylalanine

Question 19:

Which of the following enzyme does not participate in a ping-pong reaction?

a) Serine proteases

b) Aminotransferases

c) Pyruvate carboxylase

d) Aldolase

Answer Key

Question No.	Correct Option
1	d
2	d
3	a
4	a
5	d
6	d
7	b
8	a
9	d
10	a
11	b
12	a

13	b
14	d
15	a
16	c
17	a
18	b
19	d

Detailed Explanations

Solution to Question 1:

Enzymes are not heat-stable.

Most enzymes are proteins and are thus susceptible to heat. Many enzymes also have temperature ranges at which they are most active, and they get deactivated at temperatures higher than the aforementioned range.

Coenzymes are heat-stable. Enzymes are stereospecific catalysts. They are neither consumed nor permanently altered in a reaction. Most of them are proteins except ribozymes. A ribozyme is RNA with catalytic activity. Some examples of ribozymes are:

- Sn Rna in the spliceosome.
- Ribonuclease P
- Peptidyl transferase
- RNAse P

Solution to Question 2:

Enzymes do not catalyze a reaction by increasing the activation energy, rather they do it by decreasing the activation energy.

Specific acid or base catalysis, catalysis by proximity, catalysis by strain all constitute mechanisms of enzyme to enhance the rate of reaction.

Enzymes use combinations of four general mechanisms to achieve enhancements of the rates of chemical reactions:

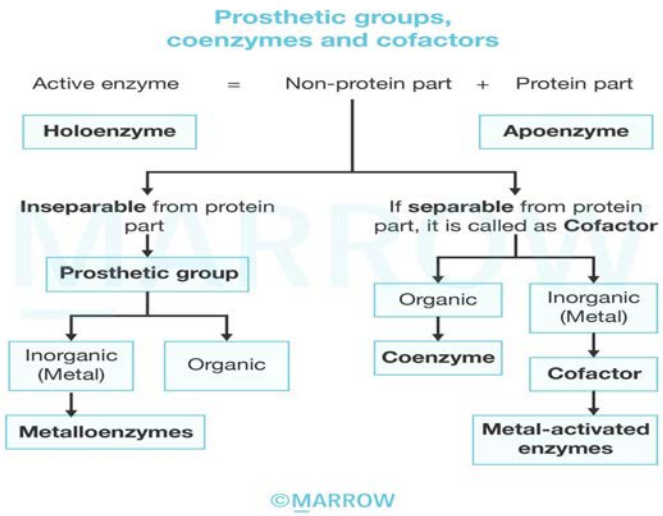
- Covalent catalysis
- Specific acid or base catalysis
- Catalysis by proximity
- Catalysis by strain

Solution to Question 3:

The prosthetic group present in dinitrogenase is Molybdenum.

Prosthetic groups are non-protein part of a holoenzyme which are tightly and irreversibly bound to the protein part by covalent or noncovalent forces. The prosthetic group may be organic (such as a vitamin, sugar, or lipid) or inorganic (such as a metal ion). Metal ions constitute the most common type of prosthetic group.

If an enzyme uses metal ion as a prosthetic group it is called a metalloenzyme. Metalloenzymes have metal ions as an integral part of their structure.



Solution to Question 4:

Mitochondrial superoxide dismutase is not a copper-containing enzyme. It contains manganese.

Solution to Question 5:

S-adenosyl methionine is not a coenzyme. Its a methyl group donor.

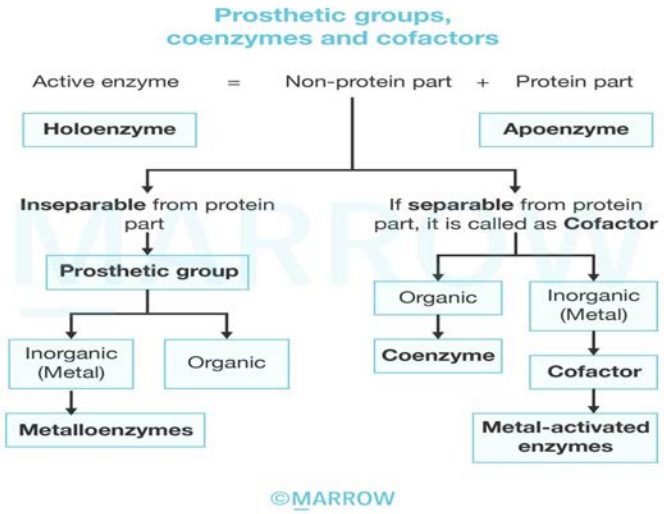
Coenzymes are cofactors in which organic compound is present. Coenzymes transiently and reversibly bind to the enzyme to catalyze the reaction. They play an important role as a two-way shuttle.

Some coenzymes:

• Coenzyme A

• ATP/GTP

- NAD/NADP
- FAD/FMN
- Lipoic acid
- B-complex vitamins
- Vitamin C
- Vitamin K

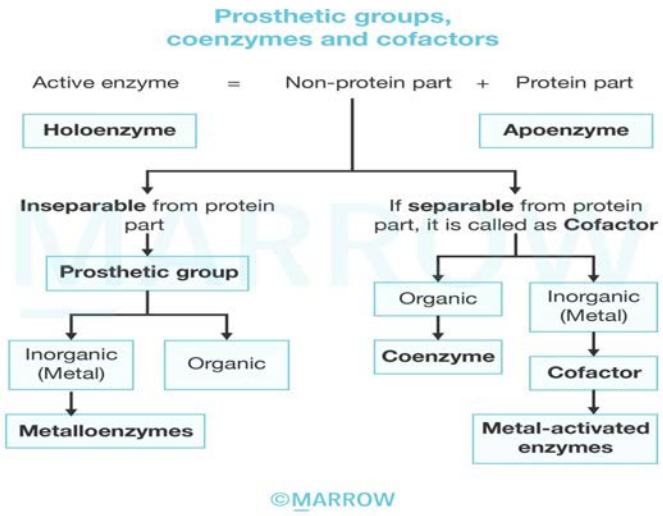


Solution to Question 6:

Enzymes that use metal-ion as a cofactor are termed as metal-activated enzymes (and not metalloenzymes).

Metalloenzymes are prosthetic groups in which the metal is tightly and irreversibly bound to the enzyme. The metal forms an integral part of its active structure. Metal-activated enzymes are those that have a cofactor containing metal (inorganic) which are loosely and reversibly bound to the enzyme. They show enhanced activity in the addition of metal.

A cofactor is a non-protein part of a holoenzyme that can be separated part from the protein part. Cofactors are mostly metal ions or inorganic. If an organic compound is present in a cofactor, it is called a coenzyme. Cofactors serve functions similar to those of prosthetic groups. Cofactors bind in a transient, dissociable manner to enzymes. Cofactors must be present in the medium surrounding the enzyme for catalysis to occur.



Solution to Question 7:

Hydrolases are not involved in oxidation-reduction reactions. Hydrolases are involved in the hydrolysis of the substrate using water.

Solution to Question 8:

Tryptophan pyrrolase is not a mono-oxygenase. Its a di-oxygenase.

Oxygenases catalyzes the incorporation of oxygen into a substrate. Oxygenases are of two types:

1) Mono-oxygenases (Hydroxylases/ Mixed-function oxidases): Incorporate one atom of oxygen into the substrate.

Examples:

- Tryptophan hydroxylase
- Tyrosine hydroxylase
- Cytochrome p450
- Phenylalanine hydroxylase
- 7-alpha hydroxylases

2) Di-oxygenases: Incorporate two atoms of oxygen into a substrate.

Examples:

- Tryptophan pyrrolase
- Homogentisate di-oxygenase (previously known as homogentisate oxidase)

Solution to Question 9:

Oxygenases do not catalyze the carboxylation of drugs. They catalyze the hydroxylation of drugs.

Cytochromes P450 are an important superfamily of heme-containing monooxygenases. They are located mainly in the endoplasmic reticulum in the liver and intestine but are also found in the mitochondria in some tissues. It is involved in a series of enzymatic reactions known as the hydroxylase cycle. Together cytochromes P450 and b5 are responsible for about 75% of the modification and degradation of drugs that occur in the body by hydroxylation and not decarboxylation.

Mitochondrial cytochrome P450 systems are found in steroidogenic tissues such as the adrenal cortex, testis, ovary, and placenta and are concerned with the biosynthesis of steroid hormones from cholesterol by hydroxylation. (Option C)

Depending on the number of atoms of oxygen being added, there are two types of oxygenase (Options A and B):

• Mono oxygenases: Incorporate one oxygen atom into a substrate.

• Dioxygenases: Incorporate two oxygen atoms into a substrate.

Solution to Question 10:

Aldolase B is a lyase.

Lyases are enzymes that catalyze the cleavage of C-C, C-O, C-N and other covalent bonds by atom elimination generating double bonds.

Some examples of lyases include:

• HMG-CoA lyase

• Argininosuccinate lyase

• ATP citrate lyase

• Aldolase

• Fumarase

Solution to Question 11:

The group of enzymes that catalyze the joining together of two molecules with the hydrolysis of ATP is called ligases.

Solution to Question 12:

Lipoprotein lipase is not a non-functional enzyme. It is a functional enzyme.

Functional enzymes have a specific function in the plasma such as coagulation factors, pseudocholinesterases, and lipoprotein lipases.

Nonfunctional enzymes have no specific function in the plasma.

• They are released from the cells as a result of normal wear and tear. Their level is very low in serum.

• They serve as biomarkers whose levels in the plasma can assist in the diagnosis and prognosis of diseases affecting specific tissues.

• Eg: Lactate dehydrogenase, Creatine kinase, alkaline phosphatase, gamma glutamyl transpeptidase.

Solution to Question 13:

Isoenzymes do not have the same number of charged amino acids.

Isoenzymes are physically distinct forms of the same enzyme. They catalyze the same reaction but differ in the following:

• Amino acid sequence and the number of charged amino acids

• Subunit composition: CK-1 contains BB while CK-2 contains MB subunits

• Electrophoretic mobility: CK-1 moves faster than CK-3

• Tissue localization: CK-1 is present in the brain, CK-2 in the heart

• Substrate affinity: Glucokinase has low affinity but hexokinase has high affinity.

Solution to Question 14:

The given clinical scenario of a woman with fragmented RBCs and decreased RBCs is suggestive of hemolytic anemia. LDH1 and LDH-2 isoenzymes are expressed in the RBC and particularly raised during hemolysis.

LDH is a tetrameric enzyme with 4 subunits. It has four subunits with two isoforms: H-isoform (Heart) and M-isoform (Muscle)

LDH Type	Composition	Location
LDH-1	HHHH	Heart and RBC
LDH-2	HHHM	RBC and kidney
LDH-3	HHMM	Lungs, Spleen, Brain, Lymphatic tissues, Platelets
LDH-4	HMMM	Liver and skeletal muscle

LDH Type	Composition	Location
LDH-5	MMMM	Liver and skeletal muscle

Solution to Question 15:

The given clinical scenario of sudden onset chest pain with profuse sweating and 12 Lead Electrocardiogram (ECG) with ST-segment elevation in Leads II, III, and aVF is suggestive of myocardial Infarction. Serum LDH 1 > LDH 2 levels are seen in this condition.

Under normal conditions, LDH 2 is predominantly present in the blood and LDH 1 is predominantly present in the heart. So, normally LDH 2 > LDH 1 in blood. When cardiac tissue is damaged LDH 1 is released into the bloodstream. This results in a flipped lactate dehydrogenase (LDH) ratio where LDH 1 > LDH 2.

LDH is a tetrameric enzyme with 4 subunits. It has four subunits with two isoforms: H isoform (heart) and M isoform (muscle)

LDH Type	Composition	Location
LDH-1	HHHH	Heart and RBC
LDH-2	HHHM	RBC and kidneys
LDH-3	HHMM	Lungs, brain, lymphatic tissues, and platelets
LDH-4	HMMM	Liver and skeletal muscle
LDH-5	MMMM	Liver and skeletal muscle

Solution to Question 16:

The enzyme most specific for his condition, acute viral hepatitis, is alanine aminotransferase (ALT).

Alanine aminotransferase (ALT) is predominantly found in the liver and therefore is a more specific marker for hepatocellular damage than AST.

Aspartate aminotransferase (AST) is found in various tissues. The AST/ALT ratio is very important.

• AST/ALT <1 is seen in hepatocellular damage like in NAFLD, toxic hepatitis, paracetamol toxicity. In cirrhosis, the ratio becomes more than 1.

• AST/ALT> 2 is suggestive while AST/ALT> 3 is highly suggestive of alcoholic liver disease.

Other options:

Option A: Alkaline phosphatase rise is seen in many liver diseases but more than fourfold rise indicates cholestasis.

Option D: GGT is used to identify occult alcohol use.

Solution to Question 17:

Chymotrypsinogen is a zymogen. Its active form chymotrypsin is a serine protease.

Serine proteases are proteolytic enzymes having serine, histidine and aspartate at the active site. Active site consists of two binding sites and a catalytic site. While serine is present in the catalytic site of serine proteases, histidine and aspartate are present on binding sites. Serine proteases differ in substrate specificity.

Examples of serine proteases :

• Chymotrypsin

• Trypsin

• Elastase

• Thrombin

• Plasmin

• Complements

• Factor X and XI

• Prostate-specific antigen.

Solution to Question 18:

Elastase is a serine protease that cleaves at the carboxy-terminal of neutral amino acids like alanine and glycine.

Serine proteases are proteolytic enzymes having Serine, Histidine and Aspartate at the active site. The active site consists of two binding sites and a catalytic site. While serine is present in the catalytic site of serine proteases, histidine and aspartate are present on binding sites.

Serine proteases differ in substrate specificity.

• Trypsin cleaves the carboxy-terminal of basic amino acids like arginine and lysine.

• Chymotrypsin cleaves at the carboxy-terminal of bulky hydrophobic amino acids like tryptophan, phenylalanine, and tyrosine.

• Elastase cleaves at the carboxy-terminal of neutral amino acids like alanine and glycine.

Solution to Question 19:

Aldolase does not participate in a ping-pong reaction. Ping pong reactions are a type of Bi-Bi reaction.

Bi-Bi reaction is a two substrate two product reaction. Most of the Bi-Bi reactions follow Michaelis-Menten kinetics. There are three types of Bi-Bi reactions:

• Ordered Bi-Bi reaction: Example- NAD(P)H-dependant oxidoreductases.

• Random Bi-Bi reaction: Example- Most kinases and dehydrogenases.

• Ping pong reaction: Example- Serine proteases, aminotransferases and pyruvate carboxylase.

Enzyme Kinetics and Regulation of Activity

Question 1:

Activation energy is the free energy difference between _____.

a) Substrate & product

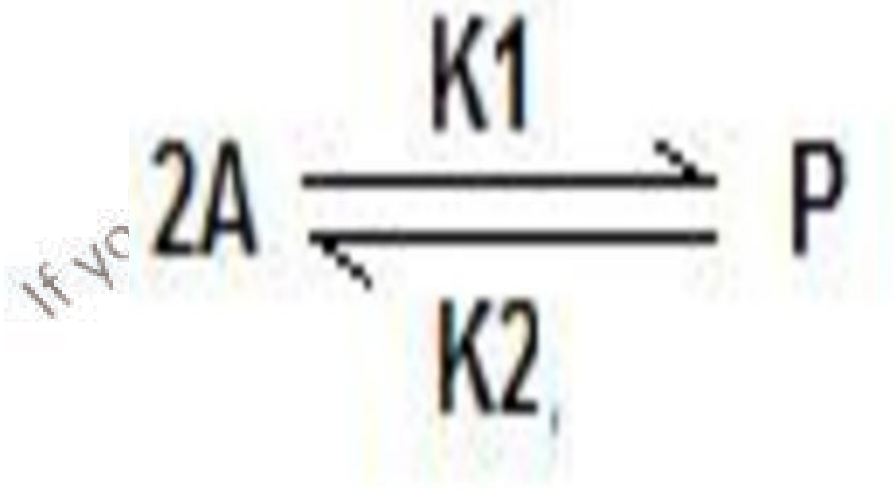
b) Substrate & transition state

c) Transition state & product

d) Sum of all the above

Question 2:

In the given reaction, two molecules of A combines to form P. What is the kinetic order of the reaction?



a) 1st order for the forward reaction & 2nd order for the reverse reaction

b) 1st order for the forward reaction & 1st order for the reverse reaction

c) 2nd order for the forward reaction & 1st order for the reverse reaction

d) 2nd order for the forward reaction & 2nd order for the reverse reaction

Question 3:

Regarding enzyme kinetics, which of the following is true?

a) Q10 = 1

b) Q10 = 2

c) Q10 = 3

d) Q10 = 4

Question 4:

Which of the following is not a type of covalent modification for regulation of enzyme kinetics?

a) Methylation

b) Acetylation

c) ADP ribosylation

d) Ionization

Question 5:

Which of the following is false regarding the effect of substrate concentration on the rate of reaction?

a) As substrate concentration increases, the initial velocity of the reaction increases until it reaches a maximum value (Vmax)

b) The substrate concentration at which the velocity of the reaction is half maximum is called Km

c) The Michaelis-Menten equation says that initial reaction velocity (Vi) = Vmax [S]/ Km+[S]

d) The plot of initial reaction velocity against substrate concentration is linear

Question 6:

The catalytic efficiency of enzyme is best expressed by which of the following kinetic constants:

a) Kcat/Km

b) Km/Kcat

c) Km/Ka

d) Ka/Km

Question 7:

In a reaction, the substrate is available in a concentration that is 1000 times the Km value of the enzyme. After 9 minutes of reaction, 1% of substrate is converted to product (12 microgram/ml). If the concentration of the enzyme is changed to 1/3 and concentration of substrate is doubled, what is the time taken to convert the substrate into the same amount of product that i.e. 12 microgram/ml?

a) 9 minutes

b) 4.5 minutes

c) 27 minutes

d) 13.5 minutes

Question 8:

Which of the following best defines the type of enzyme inhibition by malonate on succinate dehydrogenase reaction?

a) Non-competitive

b) Uncompetitive

c) Competitive

d) Allosteric

Question 9:

Which of the following is true about competitive inhibition?

a) Vmax increases

b) Vmax decreases

c) Vmax constant

d) Km decreases

Question 10:

Which of the following is false about non-competitive inhibition?

a) Vmax decreases

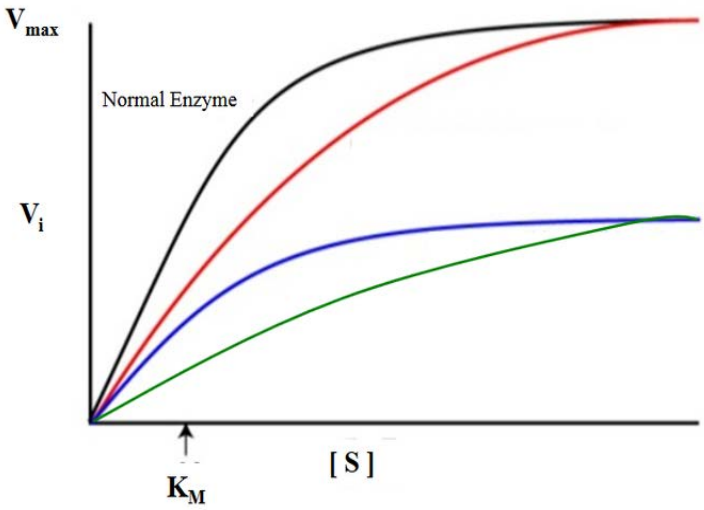
b) Km decreases

c) The inhibitor binds at a site different from the active site

d) The inhibitor does not affect the binding of the substrate

Question 11:

Which of the following situations does the red line in the following graph depict?



a) Reaction with a competitive inhibitor

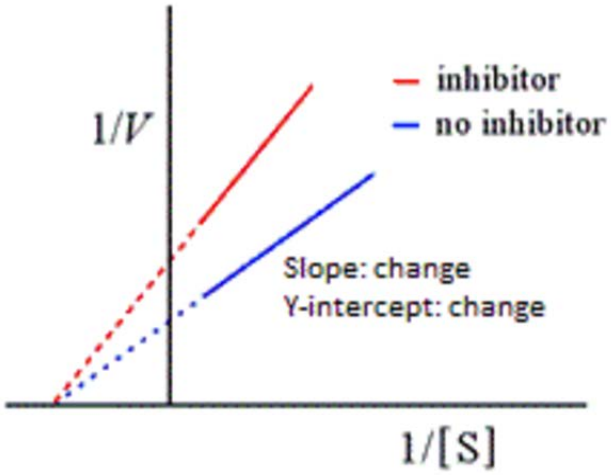
b) Reaction with a noncompetitive inhibitor

c) Reaction with a suicide inhibitor

d) None of the above

Question 12:

Which of the following situations does the red line in the following graph depict?



a) Reaction with a competitive inhibitor

b) Reaction with an allosteric Inhibitor

c) Reaction with a suicide inhibitor

d) Reaction with a noncompetitive inhibitor

Question 13:

Which of the following is true regarding an allosteric modifier?

a) Desaturates the enzyme

b) Causes the enzyme to work faster only.

c) Binds to the catalytic site

d) Participates in feedback regulation

Question 14:

Which of the following is true about allosteric regulation?

a) Allosterically regulated enzymes belong to K-series, V-series or both

b) Allosteric modifier binds to the active site of the enzyme

c) Induction and repression are examples of allosteric regulation

d) Allosteric modifier binds to the enzymes by covalent bond

Question 15:

Chymotrypsinogen is a _____

a) Zymogen

b) Carboxypeptidase

c) Transaminase

d) Clot-lysing protein

Question 16:

A 46-year-old male visits the cardiology clinic with a history of exertional chest pain. He is a known diabetic, hypertensive and hyperlipidemic which are controlled with appropriate medication. Considering his high 10 year-ASCVD risk score (atherosclerotic cardiovascular disease risk), he is started on low dose aspirin. Which of the following best defines the mechanism of aspirin action?

a) Suicide inhibition

b) Competitive inhibition

c) Allosteric activation

d) Allosteric inhibition

Question 17:

Which of the following nucleotide triphosphate targets ATCase for feedback regulation?

a) Cytidine triphosphate

b) Adenosine triphosphate

c) Guanosine triphosphate

d) A & B

Question 18:

Which of the following is true regarding non-competitive inhibition?

a) Constant Km and increased Vmax

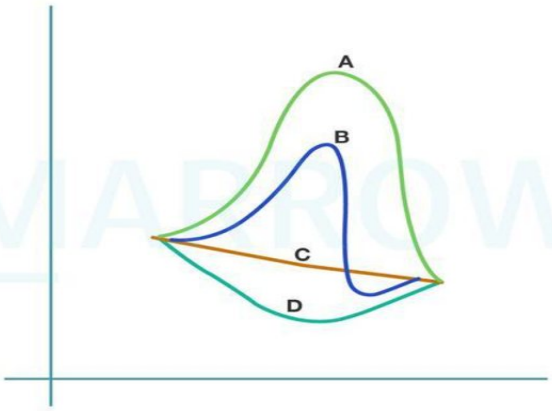
b) Constant Km and decreased Vmax

c) Decreased Km and decreased Vmax

d) Decreased Km and increased Vmax

Question 19:

Gibbs free energy curve of an enzyme-catalyzed reaction is given by



a) B

b) A

c) C

d) D

Question 20:

Which of the following enzymes has the highest catalytic efficiency?

a) Km of an enzyme = 10 micromole and Kcat = 20 per sec

b) Km of an enzyme = 2000 nanomole and Kcat = 50 per sec

c) Km of an enzyme = 2 micromole and Kcat = 200 per sec

d) Km of an enzyme = 4 micromole and Kcat = 200 per sec

Answer Key

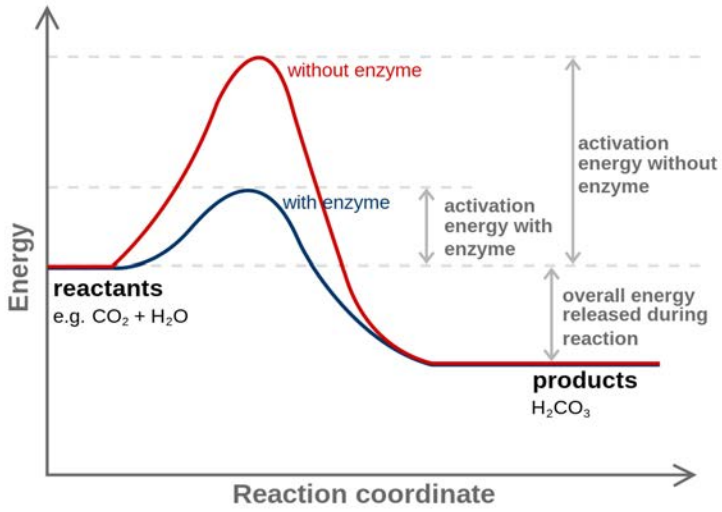
Question No.	Correct Option
1	b
2	c
3	b
4	d
5	d
6	a
7	c
8	c
9	c
10	b
11	a
12	d
13	d
14	a
15	a
16	a
17	d
18	b
19	b
20	c

Detailed Explanations

Solution to Question 1:

Activation energy is the free energy difference between the substrate and transition state.

Activation energy is defined as the minimum energy required to start a chemical reaction. Because of the high activation energy, the rates of uncatalyzed chemical reactions are often slow. In an enzyme-catalyzed reaction, the activation energy is reduced by forming a relatively stable intermediate-enzyme complex. Thus, enzymes increase the rate of chemical reactions.



Solution to Question 2:

The above reaction has 2nd order for the forward reaction and 1st order for the reverse reaction.

The sum of the molar ratios of the reactants defines the kinetic order of the reaction.

In the forward reaction, A + A ° P, the sum of reactants is 2. Thus,

Rate ∝ [A] [A] or Rate ∝ [A]2

Therefore, the reaction is said to be second order with respect to reactant A.

In the reverse reaction, P ° 2 A, the sum of reactants is 1. Thus,

Rate ∝ [P]

Therefore, the reaction is said to be first order with respect to reactant P.

Kinetic order of reactions:

• Zero-order reaction: Rate is independent of the concentration of reactants.

• First-order reaction: Rate is directly proportional to the concentration of one of the reactants.

• Second-order reaction: In the simplest kind of 2nd order reaction, the rate is proportional to the square of the concentration of one reactant.

Solution to Question 3:

Q10 = 2 is the correct statement regarding enzyme kinetics.

The temperature coefficient (Q10) is the factor by which the rate of a biologic process increases for a 10°C increase in temperature. The rates of most biological processes typically double for a 10°C rise in temperature.

Thus, Q10 (degree celsius rise) = 2 (double).

Solution to Question 4:

Ionization is not a type of covalent modification regulating enzyme kinetics.

Regulation of enzyme kinetics can be brought about by reversible covalent modifications of the enzyme. These include methylation, acetylation, ADP ribosylation, phosphorylation and partial proteolysis.

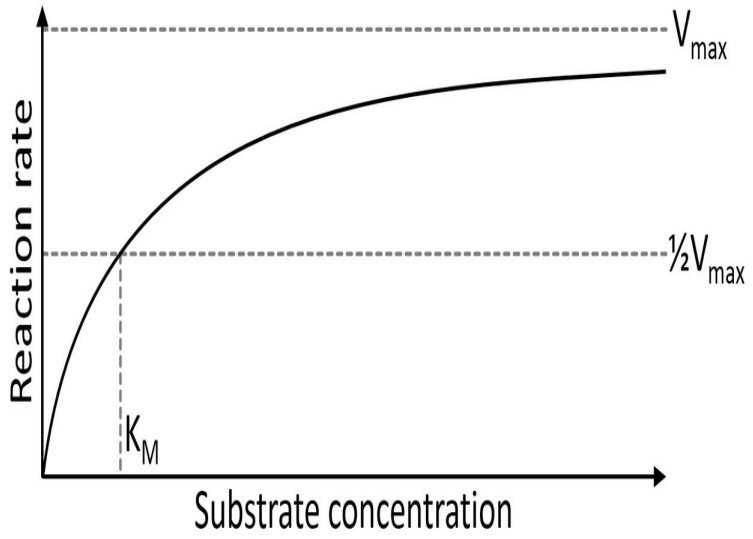
Solution to Question 5:

The plot of initial reaction velocity against substrate concentration is hyperbolic, not linear.

For a typical enzyme, the velocity of a reaction has a hyperbolic relationship with the substance concentration. As the substrate concentration increases, initial velocity (Vi) increases until it reaches a maximum value Vmax. After Vmax is reached, further increase in the substrate concentration does not change reaction velocity. This is because the enzyme is saturated with the substrate.

The Michaelis constant (Km) is the substrate concentration at which Vi is half the maximal velocity i.e. Vmax/2. The Michaelis-Menten equation illustrates the relationship between Vi and substrate concentration [S] mathematically i.e,

$Vi = Vmax \times [S] / Km + [S]$



Solution to Question 6:

The catalytic efficiency of an enzyme is expressed as Kcat/Km.

Kcat is the rate constant, which is the limiting rate of an enzyme-catalyzed reaction at saturation. It is given by Vmax divided by number of active sites of the enzyme. Km is the Michaelis-Menten constant, which is defined as the substrate concentration at which the initial velocity(Vi) is half the maximum velocity (Vmax/2).

For some enzymes, when a substrate (S) binds to the active site of the enzyme (E), and forms an enzyme-substrate complex (ES), the product is instantaneously formed. For such enzymes, the reaction of E+S° ES becomes the rate-limiting step. Its rate is determined on the rate of diffusion of the molecules in a solution. Hence, these enzymes are called diffusion-limited or catalytically perfect.

Examples of the catalytically perfect enzymes, where the catalytic efficiency reaches the diffusion limit of 108-109 M–1s–1:

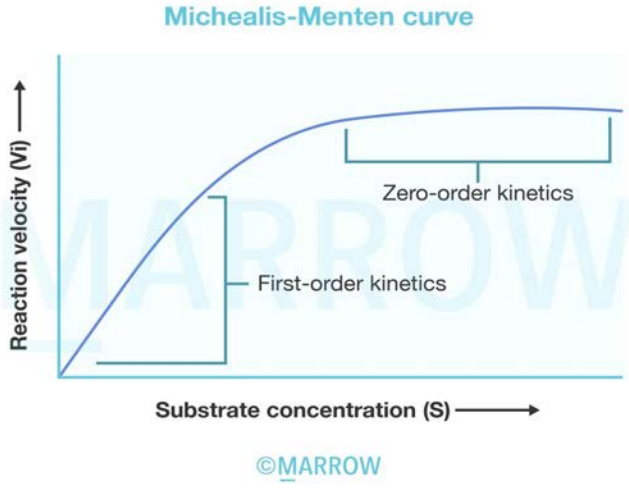
- Triosephosphate isomerase
- Carbonic anhydrase
- Acetylcholinesterase
- Adenosine deaminase

Solution to Question 7:

The time taken for the formation of the same amount of product is 27 minutes.

In this reaction, the substrate concentration becomes 1000 times i.e much greater than the Km value. Thus, the rate of this reaction is equal to Vmax and is thus independent of the substrate concentration i.e. it follows zero-order kinetics. Thus, doubling the substrate concentration does not change the velocity of the reaction i.e. it remains as Vmax.

No matter the substrate concentration, the rate of the reaction is directly proportional to the concentration of the enzyme. In this situation the enzyme concentration becomes 1/3rd. Thus, the velocity of the reaction (Vmax) also becomes 1/3rd. This means that the time taken for the formation of the same amount of product will become tripled i.e 9 × 3 = 27 minutes.



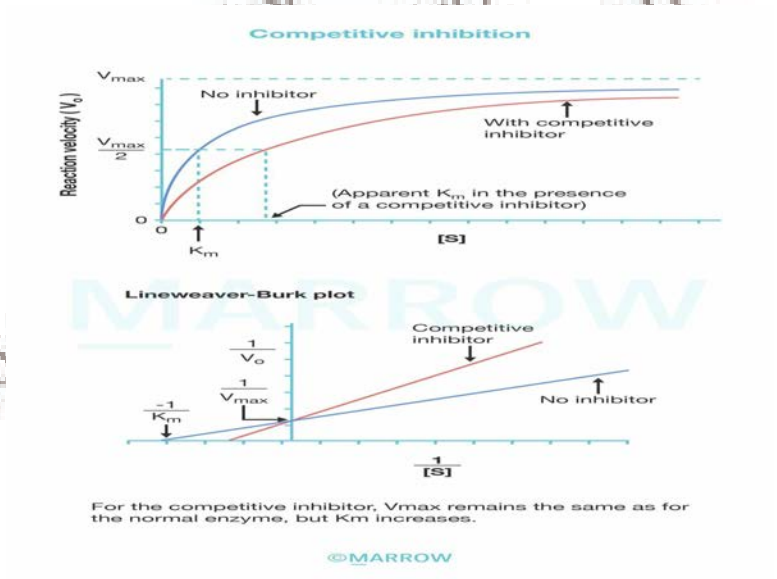
Solution to Question 8:

Inhibition of succinate dehydrogenase reaction by malonate is an example of competitive inhibition.

Solution to Question 9:

Vmax remains constant in competitive inhibition.

Competitive inhibition is when the inhibitor binds reversibly to the substrate-binding site and competes with the substrate for the active enzyme-binding site. This effect of competitive inhibition can be overcome by increasing the substrate concentration by which maximum velocity can still be achieved. Therefore, Vmax (maximum velocity of the reaction) is unaltered in this type of inhibition. However, the concentration of the substrate required to achieve half of the maximum velocity is increased. Therefore, Km is increased in this condition.

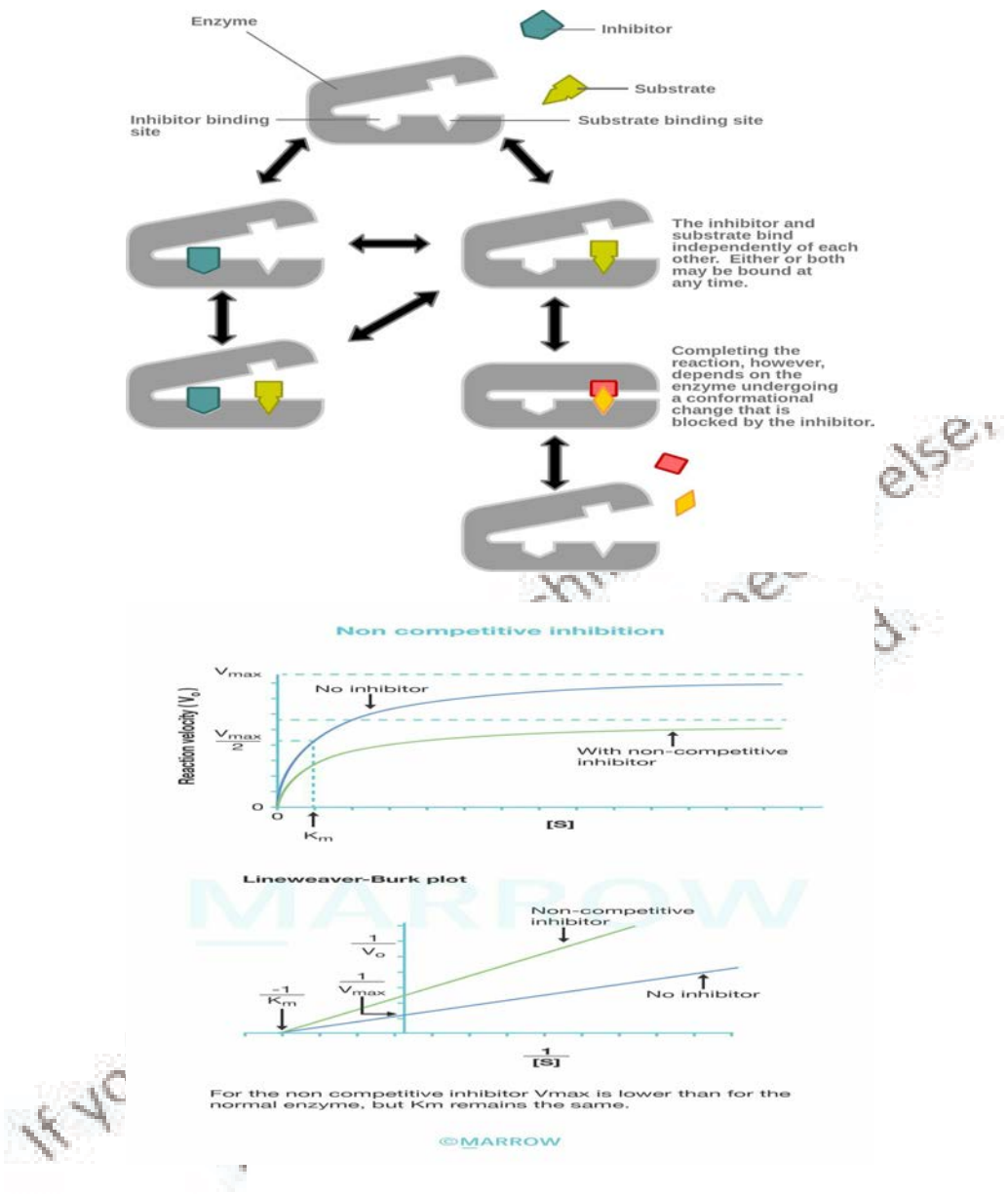


Solution to Question 10:

Km remains constant in noncompetitive inhibition.

Non-competitive inhibition of enzymes occurs by the binding of inhibitors to enzymes in locations different from the active site. The inhibition is not overcome by increasing the concentration of the substrate. Thus, noncompetitive inhibitors decrease the Vmax of the reaction.

Non-competitive inhibtiors do not resemble the substrate. Thus, they do not interfere with the binding of the substrate to the enzyme. Thus, Km of the enzyme does not change.



Solution to Question 11:

The red line indicates an enzymatic reaction taking place in the presence of a competitive inhibitor.

The graph represents the enzymatic reaction velocity in relation to substrate concentration. The black line indicates the reaction taking place in the absence of any inhibitor.

In the presence of a competitive inhibitor, as indicated by the red line,

• Vmax remains unchanged.

• Km is increased.

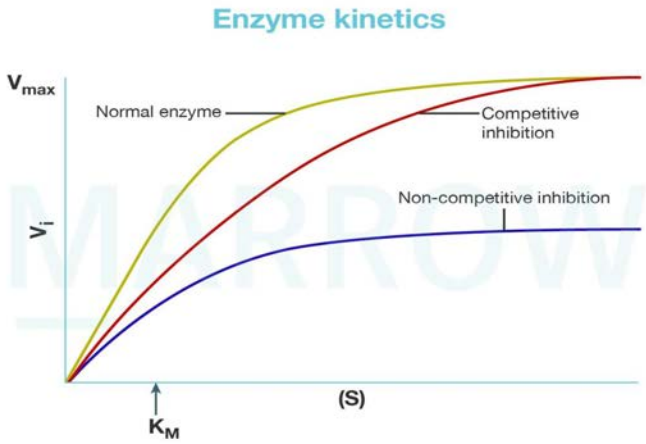
In the presence of a non-competitive inhibitor, as indicated by the blue line,

• Vmax is decreased

• Km remains unchanged

Note:

Suicide inhibitors are specialized substrate analogs. They contain a chemical group that can be transformed by the target enzyme. After binding to the active site of the enzyme, catalysis by the enzyme generates a highly reactive group that forms a covalent bond and blocks the function of the enzyme.



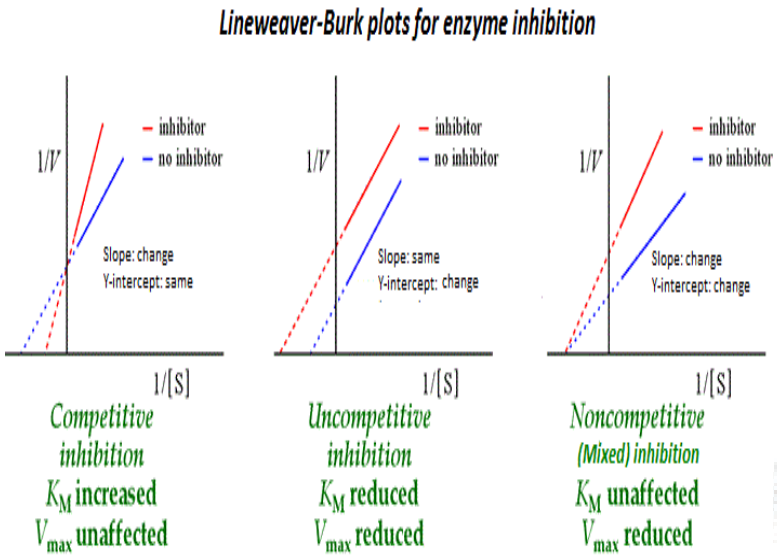
Solution to Question 12:

The red line in the graph represents an enzymatic reaction with a noncompetitive inhibitor.

The above graph is called the 'Lineweaver-Burk plot' or 'double reciprocal' plot. 1/v0 (v0 - initial velocity) is plotted versus 1/[S] ([S] - substrate concentration) and a straight line is obtained. This plot can be used to calculate Km and Vmax as well as to determine the mechanism of action of enzyme inhibitors. The x-intercept represents -1/Km while the y-intercept represents 1/Vmax.

In non-competitive inhibition, Vmax decreases (or 1/Vmax increases) but Km is unchanged. Hence the graphs of inhibited and uninhibited reactions have the same x-intercepts (converge at the same -1/Km) but different y-intercepts.

Note: An allosteric inhibitor alters the conformation of an enzyme upon binding. Enzymes affected by allosteric inhibitors exhibit a sigmoid, not hyperbolic, relationship between V0 and [S].



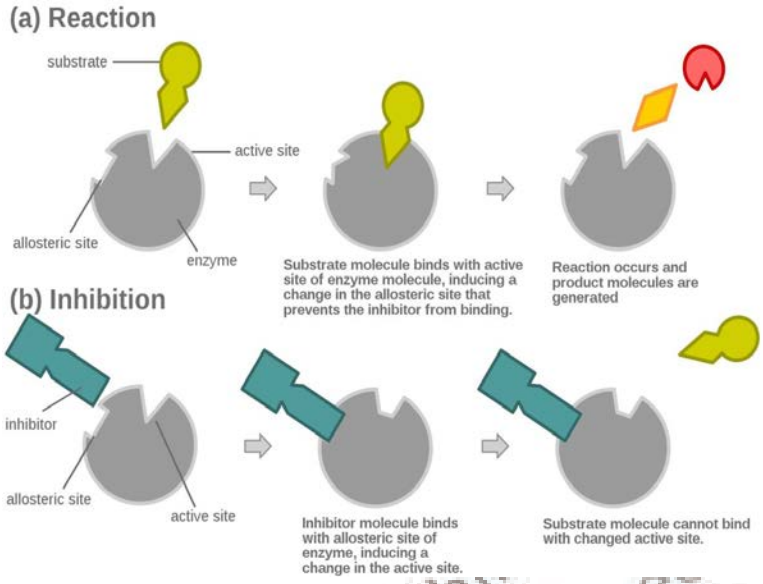
Solution to Question 13:

An allosteric modifier of an enzyme usually participates in feedback regulation.

Allosteric regulations are a natural example of control loops, such as feedback from downstream products or feedforward from upstream substrates. Allosteric regulation is also particularly important in the cell's ability to adjust enzyme activity.

An allosteric modifier or effector binds to the allosteric site to induce a conformational change in the catalytic or active site. Those modifiers or effectors that enhance the protein's activity are referred to as allosteric activators, whereas those that decrease the protein's activity are called allosteric inhibitors.

Some examples of allosteric modifiers		
Enzyme	Allosteric inhibitor	Allosteric activator
1. ALA synthase	Heme	-
2. Aspartate transcarbamoylase	CTP	ATP
3. HMG CoA reductase	Cholesterol	-
4.Phosphofructokinase	Citrate, ATP	AMP, F2,6P
5. Acetyl CoA carboxylase	Acyl CoA	Citrate
6. Citrate synthase	ATP	-
7. Carbamoyl phosphate synthase-I	-	NAG
8. Carbamoyl phosphate synthase-II	ATP	-



Solution to Question 14:

Allosterically regulated enzymes belong to either the K-series, V-series or both.

An allosteric modifier or effector binds to the allosteric site to induce a conformational change in the catalytic or active site. The kinetics of allosteric effector dos not follow Michaelis-Menten kinetics. Hence, allosterically activated enzymes can fall under the following categories:

• K-series: The allosteric effector raises the Km of the enzyme without affecting Vmax i.e. similar to competitive inhibition.

• V-series: The allosteric effector lowers the Vmax without affecting Km i.e. similar to non-competitive inhibition.

• Both: Intermediate effects can also be seen.

Option B: Allosteric modifier noncovalently binds to a site other than the active site (Allo=other, steric=space) and affects the catalysis.

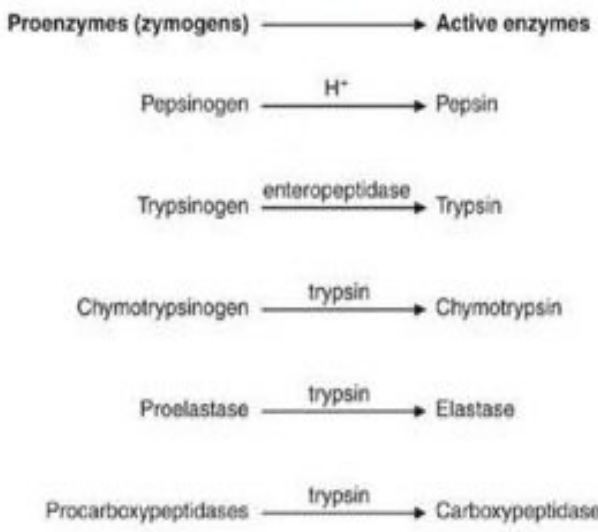
Option C: Induction and repression are methods of controlling the rate of enzyme synthesis, not regulation of activity.

Option D: Allosteric modifier binds to the enzymes noncovalently.

Solution to Question 15:

Chymotrypsinogen is a zymogen.

Some enzymes are secreted as an inactive form called as zymogens. The zymogens are converted to their active functional forms by partial, or selective proteolysis. Examples of zymogens include trypsinogen, chymotrypsinogen, proelastase, pepsinogen, prophospholipase A2 etc.



Solution to Question 16:

Low dose aspirin exerts its action by irreversibly inhibiting cyclooxygenase enzyme activity in platelets, which is an example of suicide inhibition.

A suicidal enzyme is one that undergoes self-destruction in order to terminate its own activity. In this case, in response to irreversible inhibition by aspirin, platelet cyclooxygenase loses its ability to synthesize thromboxane. It undergoes self-destruction. Unless new platelets are formed, this cyclooxygenase activity cannot be regained.

Solution to Question 17:

Both cytidine triphosphate and adenosine triphosphate target aspartate transcarbamoylase (ATCase) for feedback regulation.

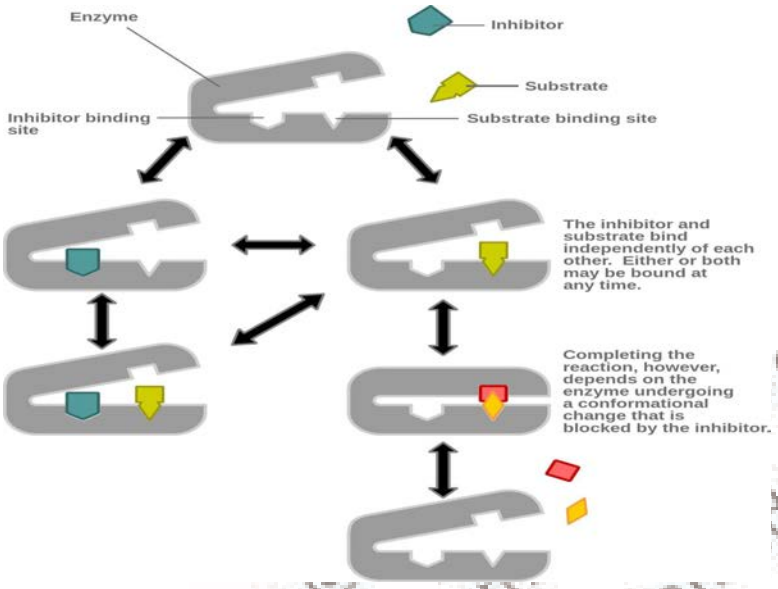
ATCase, the catalyst for the first reaction unique to pyrimidine biosynthesis, is a target of feedback regulation by two nucleotide triphosphates: cytidine triphosphate (CTP) and adenosine triphosphate (ATP). CTP is an end product of the pyrimidine biosynthetic pathway. It inhibits ATCase, whereas the purine nucleotide ATP activates it.

High levels of ATP can overcome the inhibition by CTP, enabling synthesis of pyrimidine nucleotides to proceed even when purine nucleotide (CTP) levels are elevated.

Solution to Question 18:

Noncompetitive inhibition has constant Km and decreased Vmax.

Noncompetitive inhibition occurs when the inhibitor binds at a site different from the substrate-binding site. The structure of the inhibitor does not resemble that of the substrate and the inhibitor does not affect the binding of the substrate.

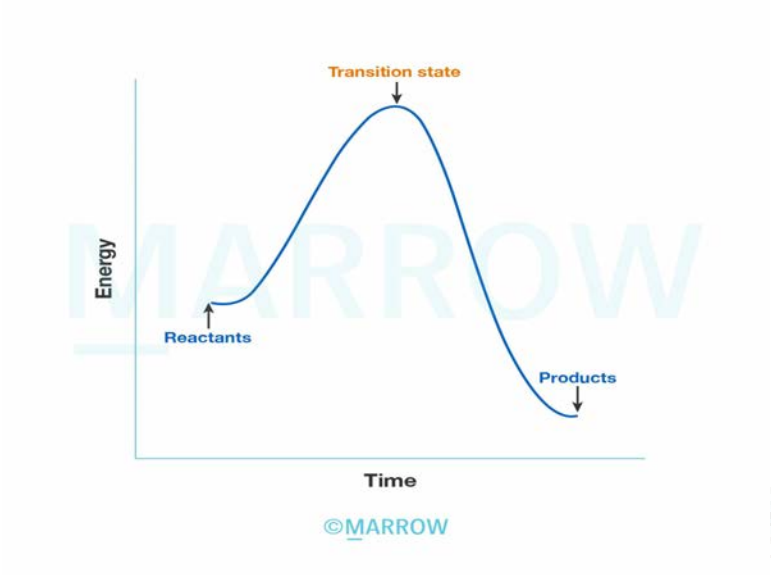


Solution to Question 19:

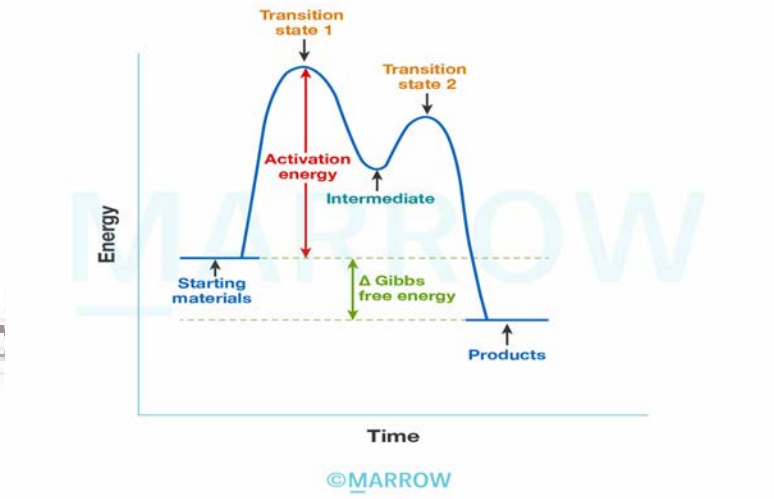
Among the given graphs, the Gibbs free energy change of an enzyme-catalyzed reaction is shown by graph A (Option B).

Graph B (Option A) cannot represent the Gibbs free energy of an enzyme-catalyzed reaction, as the energy state of an intermediate cannot be lower than the product. A lower energy state of the intermediate implies that the enzyme-intermediate complex is stable and such a complex will usually not further form a product.

During any chemical reaction, the product is formed by the breaking of substrate bonds and the conformational change of the substrate into a highly unstable transition state. This transition state is denoted by the peak of the graph. The Gibbs free energy curve of an enzyme-catalyzed reaction (with no intermediate state) is shown in the image given below:

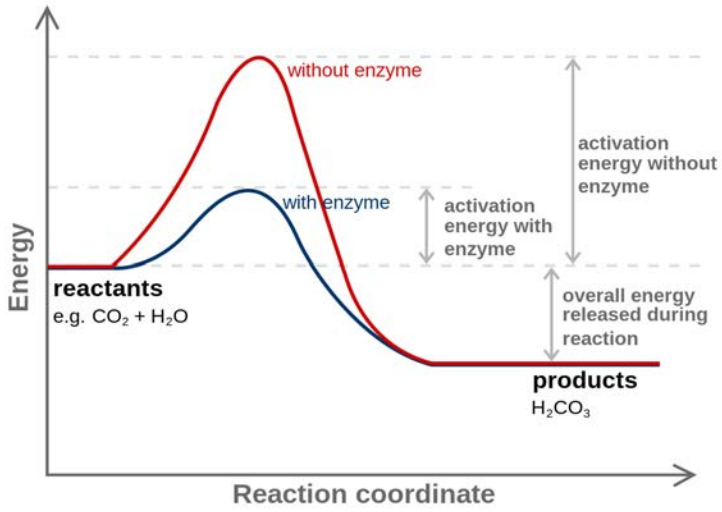


The below graph shows the normal intermediate formation with an energy state still higher than that of the product:



All the reactions have a transition state, but not all reactions have an intermediate state. Every intermediate state is again always followed by another transition state.

Activation energy is the energy difference between the free energy of the substrates and that of the transition state. Because of the high activation energy, the rates of uncatalyzed chemical reactions are often slow.



In an enzyme-catalyzed reaction, the activation energy is reduced by forming a relatively stable intermediate-enzyme complex. Enzymes provide attachment sites for the substrate and speed up the rate of reaction. They don't change the energy state of either the substrate or the product. The Gibbs free energy change remains the same.

Graph C: Free energy change cannot be a straight line. Every reaction passes through a high-energy intermediate transition state.

Graph D: The activation energy of any reaction is never lower than the free energy of the product.

Solution to Question 20:

The highest catalytic efficiency is seen when Km= 2 micromole and Kcat = 200 per sec.

Catalytic efficiency is calculated as Kcat/Km, where Km is inversely proportional to the binding affinity of the enzyme and Kcat is the catalytic constant (Vmax divided by the total number of active sites of the enzyme present). Higher values of Kcat and lower values of Km gives better catalytic efficiency. Catalytic efficiency can be used to compare the performance of 2 different enzymes in catalyzing a reaction.

Step 1: Km of each option after converting respective units to moles will be 10, 2, 2 and 4 (each multiplied by 10-6) moles.

Step 2: The catalytic efficiency is calculated as Kcat/Km. On substituting the values in various options after converting to moles, the values come out to be 2, 25, 100 and 50 (each multiplied by 106) M-1 s-1.

Some enzymes can be so efficient that they convert the substrate to product instantaneously. They are limited only by the rate at which substrate diffuses towards the enzyme in a solution. These are called diffusion-limited enzymes and their catalytic efficiency is as high as 108 M-1 s-1.

Fat soluble vitamins

Question 1:

Basal metabolic rate is closely dependent on which of the following factors?

a) Body surface area

b) Lean body mass

c) Body mass index

d) Height to weight ratio

Question 2:

Which of the following conditions will cause a decreased BMR?

a) Starvation

b) Cachexia

c) Cold environment

d) Hyperthyroidism

Question 3:

All of the following vitamins are synthesized by the bacterial flora in the intestine except?

a) Niacin

b) Biotin

c) Vitamin K

d) Pantothenic Acid

Question 4:

Which of the following represents the recommended dietary allowance of a vitamin or mineral?

a) Estimated average requirement +2 standard deviation

b) Estimated average requirement -2 standard deviation

c) Equal to estimated average requirement

d) Twice the estimated average requirement

Question 5:

What is the recommended dietary allowance of vitamin A in a 6-year-old child?

a) 700mcg

b) 800mcg

c) 400mcg

d) 300mcg

Question 6:

A patient presented with symptoms of delayed adaptation to vision in dark places, a gritty sensation in the eyes, and dry eyes. Examination shows soft corneas. Which of the following is deficient in his retina?

a) Retinol

b) Retinal

c) Retinoic acid

d) Isotretinoin

Question 7:

A young woman presents with a 2-week history of progressive severe headache and blurry vision. Examination showed bilateral papilledema. A lumbar puncture revealed elevated opening pressure with normal CSF composition and no space-occupying lesions were seen on MRI. She has recently started taking high-dose vitamin supplements. Which vitamin excess is likely to cause the above scenario?

a) Vitamin A

b) Vitamin D

c) Vitamin E

d) Vitamin K

Question 8:

An arctic explorer facing a scarcity of food resorts to hunting down a polar bear. He later develops headaches, flushing, vomiting, and desquamation of the skin. The consumption of which of the following should have been avoided to prevent this toxicity?

a) Kidneys

b) Spleen

c) Muscle

d) Liver

Question 9:

A 24-year-old obese woman presented with severe headache and exfoliative dermatitis. She was found to be on a herbal weight loss supplement for 3 years, which contained high concentrations of vitamin A. All of the following suggest toxicity except:

a) Ataxia

b) Reduced intracranial pressure

c) Hepatomegaly

d) Alopecia

Question 10:

Which of the following vitamins is also considered a hormone?

a) Vitamin E

b) Vitamin C

c) Vitamin D

d) Vitamin K

Question 11:

The first hydroxylation of vitamin D takes place in which of the following organs of the body?

a) Skin

b) Kidney

c) Liver

d) Small intestine

Question 12:

A 60-year-old woman with osteoporosis is prescribed vitamin D tablets. Which of the following is not a function of this vitamin supplement?

a) Immunomodulatory

b) Regulation of phosphate

c) Antioxidant

d) Antiproliferative

Question 13:

Which of the following vitamins is the most potent and naturally occurring antioxidant?

a) Beta carotene

b) Vitamin E

c) Vitamin C

d) Vitamin A

Question 14:

A 34-year-old woman presented with generalized weakness and lightheadedness for 2 weeks. Blood investigations were notable for decreased hemoglobin, increased indirect bilirubin, and increased LDH. Her peripheral smear showed RBC fragments. Which vitamin deficiency is associated with the above presentation?

a) Vitamin A

b) Vitamin E

c) Vitamin D

d) Vitamin K

Question 15:

Which of the following fat-soluble vitamins also has coenzyme activity?

a) Vitamin A

b) Vitamin D

c) Vitamin E

d) Vitamin K

Question 16:

Which of the following statements is true about vitamin K?

a) Deficiency of vitamin K leads to hypercoagulability.

b) It is a water soluble vitamin.

c) Prolonged use of antibiotics leads to vitamin K deficiency.

d) Menaquinone is the dietary source of vitamin K found in green vegetables.

Question 17:

A 45-year-old woman who is a known case of DVT is on long-term warfarin therapy. The levels of which of the following clotting factors will be unaffected in her?

a) Factor II

b) Factor VII

c) Factor VIII

d) Factor X

Answer Key

Question No.	Correct Option
1	b
2	a
3	a
4	a
5	c
6	b
7	a
8	d
9	b
10	c
11	c

12	c
13	b
14	b
15	d
16	c
17	c

Detailed Explanations

Solution to Question 1:

Basal metabolic rate (BMR) is closely dependent on lean body mass (fat-free mass).

Lean body mass is the difference between total body weight and body fat weight. It is the metabolically active tissue in the body.

The BMR decreases with increasing age, even when body weight remains constant. This is the result of muscle tissue replacement by adipose tissue, which is metabolically less active. Women have a significantly lower BMR than men of the same body weight and age. This is because women's bodies contain proportionally more adipose tissue.

Solution to Question 2:

Decreased basal metabolic rate (BMR) is seen in starvation.

Solution to Question 3:

Niacin is not synthesized by the bacterial flora in the intestine.

Endogenously synthesized vitamins can be derived from 2 sources:

1. Synthesized by the body

• Niacin: synthesized from an amino acid tryptophan

• Vitamin D: synthesized from 7 Dehydrocholesterol by exposure to UV-B rays (290-315 nm) in the skin.

2. Synthesized in the body (by bacterial flora in the intestine)

• Vitamin K

• Biotin

• Pantothenic Acid

Solution to Question 4:

Recommended dietary allowance (RDA) is calculated by using the formula, RDA = EAR + 2SD.

Estimated average requirement (EAR): The EAR is the average daily nutrient intake level estimated to meet the requirement of one-half of healthy individuals in a particular life stage and gender group.

Recommended dietary allowance (RDA): The RDA is the average daily dietary intake level that is sufficient to meet the nutrient requirements of nearly all individuals in a life stage and gender group. The RDA is calculated based on EAR and is set above the EAR to provide a margin of safety for most individuals. So, RDA is set 2SDs above the EAR.

Solution to Question 5:

Recommended dietary allowance (RDA) of vitamin A in a 6-year-old child is 400mcg/day.

Sources of vitamin A:

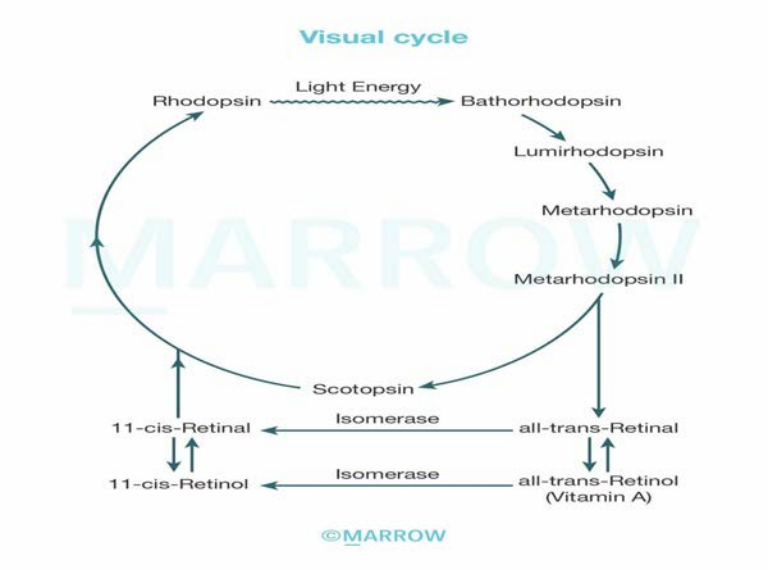
- Animal source: liver, egg, butter, milk, fish, meat
- Richest plant source: carrot
- Richest source: halibut liver oil (fish oil)

Solution to Question 6:

This patient with difficult dark vision and gritty sensations in the eye with findings of soft corneas suggests vitamin A deficiency. In the retina, a form of vitamin A called retinal (11-cis retinaldehyde) plays an important role in Wald's visual cycle. Its deficiency leads to vision defects.

Rhodopsin, the visual pigment of the rod cells in the retina, consists of 11-cis retinal specifically bound to the protein opsin. When rhodopsin is exposed to light, a series of photochemical isomerizations occurs. This results in the bleaching of the visual pigment and release of all-trans-retinal and opsin. This process triggers a nerve impulse that is transmitted by the optic nerve to the brain.

The image given below shows Wald's visual cycle.



Solution to Question 7:

The clinical scenario of increased intracranial pressure in the absence of space-occupying lesion points towards the diagnosis of pseudotumor cerebri. Vitamin A excess is the likely cause.

Clinical features of hypervitaminosis A:

• CNS: Symptoms of raised intracranial pressure (pseudotumor cerebri) such as headache, nausea, vomiting, blurring of vision, ataxia, anorexia

• Liver: Hepatomegaly and hyperlipidemia

• Musculoskeletal: Thickening of the long bones (bony exostosis), hypercalcemia, soft tissue calcification

• Dermatological: Dryness, exfoliative dermatitis, alopecia

• Teratogenesis: Pregnant women should not ingest excessive quantities as it can cause congenital malformations in the developing fetus. Isotretinoin(13-cis retinoic acid) is a drug that is teratogenic and absolutely contraindicated in pregnancy.

Solution to Question 8:

The clinical scenario is suggestive of vitamin A toxicity. Polar bear livers should not be consumed as they can cause vitamin A toxicity and death.

Vitamin A and other fat soluble vitamins are stored particularly in the liver and adipose tissue.

Vitamins can be broadly classified into two categories:

• Fat-soluble vitamins: Vitamins A, D, E, K

• Water-soluble vitamins: Vitamin B complex and vitamin C

Properties of fat-soluble vitamins:

• Hydrophobic in nature

• Transported in blood by the lipoproteins, such as chylomicrons

• Stored in the liver and the adipose tissue

Solution to Question 9:

The clinical scenario is suggestive of vitamin A toxicity. It causes increased intracranial pressure, not reduced intracranial pressure.

In cases of chronic ingestion, the accumulation of vitamin A goes beyond the capacity of intracellular binding proteins producing hypervitaminosis.

Clinical features of hypervitaminosis A:

• CNS: Symptoms of raised intracranial pressure (pseudotumor cerebri) such as headache, nausea, vomiting, blurring of vision, ataxia, anorexia

• Liver: Hepatomegaly and hyperlipidemia

• Musculoskeletal: Thickening of the long bones (bony exostosis), hypercalcemia, soft tissue calcification

• Dermatological: Dryness, exfoliative dermatitis, alopecia

• Teratogenesis: Pregnant women should not ingest excessive quantities as it can cause congenital malformations in the developing fetus. Isotretinoin (13-cis retinoic acid) is a vitamin A derived drug that is teratogenic and absolutely contraindicated in pregnancy.

Solution to Question 10:

Vitamin D is also considered a hormone.

Vitamin D encompasses a group of sterols, which have a hormone-like function.

Functions of vitamin D:

• Regulation of calcium and phosphate: It increased blood calcium and phosphate level

• Immunomodulatory action: Vitamin D increases the ability of the innate immune system to fight against pathogens

• Antiproliferative: Because of the antiproliferative function, deficiency (<20ng/ml) is associated with an increased incidence of colon cancer, breast cancer, prostate cancer

• Bone development

Solution to Question 11:

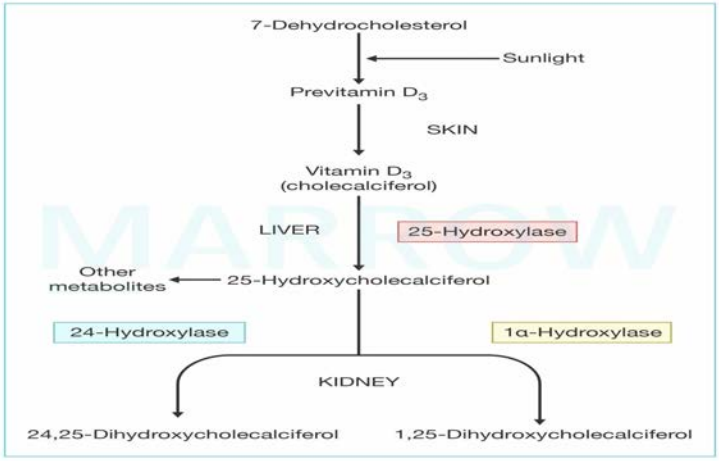
The first hydroxylation of vitamin D takes place in the liver to form 25-hydroxycholecalciferol.

Vitamin D, as either cholecalciferol or ergocalciferol does not have significant biological activity. It is activated in the following steps:

• Skin: 7-Dehydrocholesterol (an intermediate in the synthesis of cholesterol in the skin) undergoes a nonenzymic reaction on exposure to ultraviolet light (290nm to 315nm), yielding cholecalciferol.

• Liver: Cholecalciferol is hydroxylated to 25-hydroxycholecalciferol (the most abundant circulatory form of Vitamin D) by the enzyme 25-hydroxylase.

• Kidney: 25-hydroxycholecalciferol is converted to 1,25 dihydroxycholecalciferol (calcitriol), the biologically active form by the enzyme 1α-hydroxylase.



Various steps in formation and hydroxylation of Vitamin D3

Calcitriol acts on the bone, intestine, and kidney to maintain calcium and phosphate homeostasis. If the body does not require vitamin D, in the kidney, 24-hydroxylase converts vitamin D into 24,25 dihydroxycholecalciferol, which is an inactive metabolite.

Solution to Question 12:

Vitamin D is not an antioxidant.

Functions of vitamin D:

• Regulation of calcium and phosphate: It increases blood calcium and phosphate level

• Immunomodulatory action: Vitamin D increases the ability of the innate immune system to fight against pathogens

• Antiproliferative: Because of the antiproliferative function, deficiency(<20ng/ml) is associated with an increased incidence of colon cancer, breast cancer, prostate cancer

• Bone development

Solution to Question 13:

Vitamin E is the most potent and naturally occurring antioxidant.

Current therapeutic use of vitamin E is recommended in diseases that cause fat malabsorption, severe pancreatic insufficiency, and cholestatic liver disease.

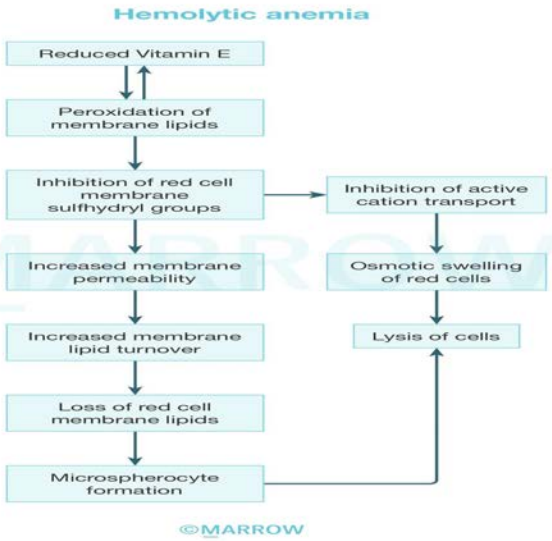
Protective antioxidant nutrients:

- Vitamin E
- Vitamin C
- Vitamin A and the carotenoids
- Selenium
- Polyphenolic compounds derived from plant foods

Solution to Question 14:

The clinical scenario of a patient with fatigue and decreased hemoglobin, increased indirect bilirubin, and increased LDH is suggestive of hemolytic anemia. It is associated with a deficiency of vitamin E.

The most important function of vitamin E is its chain-breaking antioxidant role. Vitamin E inhibits lipid peroxidation by free radicals, thereby stabilizing cell membranes. Hence, its deficiency can result in erythrocyte membranes being abnormally fragile leading to hemolytic anemia.



Note: Vitamin K deficiency does not cause hemolysis. It causes manifestations of impaired coagulation, such as easy bruisability, mucosal bleeding, etc. However, vitamin K toxicity can

cause hemolysis.

Solution to Question 15:

The only fat-soluble vitamin that has a coenzyme function is vitamin K.

The co-enzyme role of vitamin K is in the γ-carboxylation reaction of clotting factors.

Many of the water-soluble vitamins also have a coenzyme function, such as:

• Vitamin B1 as thiamine pyrophosphate (TPP)

• Vitamin B6 as pyridoxal phosphate (PLP)

Solution to Question 16:

Prolonged use of antibiotics leads to vitamin K deficiency. This is due to the loss of normal bowel flora that synthesizes menaquinones (vitamin K2).

Vitamin K is a fat-soluble vitamin. The following are the vitamers of vitamin K:

• Phylloquinone (Vitamin K1) is the normal dietary source of Vitamin K that is found in green vegetables.

• Menaquinones (Vitamin K2) are synthesized by intestinal bacteria.

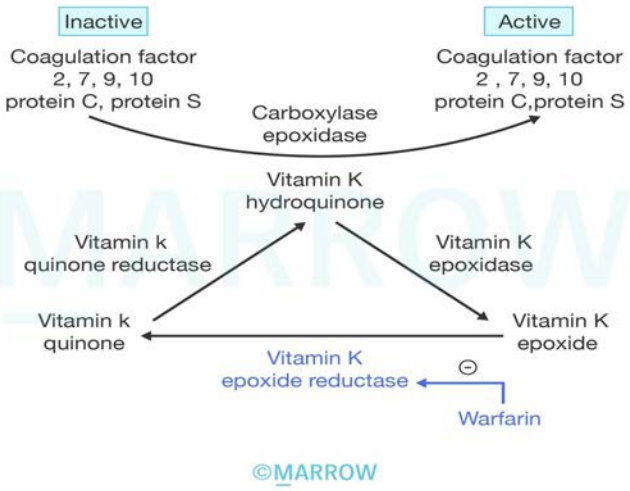
• Menadione (Vitamin K3) is a synthetic compound that is metabolized to phylloquinone.

Solution to Question 17:

Warfarin, which is an oral anticoagulant drug inhibits gamma-carboxylation of clotting factors II, VII, IX, and X, but not factor VIII. Thus, the levels of clotting factor VIII will be unaffected in her.

Warfarin inhibits vitamin K epoxide reductase, an enzyme required for the reduction of vitamin K. Reduced vitamin K is involved in the post-translational modification of blood clotting factors such as prothrombin (factor II), factor VII, factor IX, factor X, protein C, and protein S.

Therefore, warfarin indirectly inhibits the gamma-carboxylation reaction or post-translational modification of blood clotting factors.



Energy releasing vitamins

Question 1:

Which of the following multienzyme complexes catalyzing decarboxylation reactions does not require thiamine as a coenzyme?

a) Pyruvate dehydrogenase

b) α-ketoglutarate dehydrogenase

c) Branched-chain keto-acid dehydrogenase

d) Isocitrate dehydrogenase

Question 2:

Which of the following co-enzyme deficiencies makes a patient prone to lactic acidosis?

a) Thiamine

b) Riboflavin

c) Biotin

d) Niacin

Question 3:

What is the reason behind decreased energy production in thiamine deficiency?

a) It is required for transamination reactions

b) It is the coenzyme for pyruvate dehydrogenase

c) It is the coenzyme for transketolase

d) It is required in the process of oxidative reduction

Question 4:

A man with chronic alcoholism presented with orthopnea and pedal edema. On investigation, cardiomegaly is noted. Which vitamin deficiency can lead to this presentation?

a) Vitamin B1

b) Vitamin B2

c) Vitamin B5

d) Vitamin B6

Question 5:

Which of the following enzyme activity assays can be used for the diagnosis of a patient with suspected thiamine deficiency?

a) Pyruvate dehydrogenase

b) Erythrocyte transketolase

c) α-ketoglutarate dehydrogenase

d) RBC glutathione reductase

Question 6:

A middle-aged man with a history of chronic alcohol comsuption presented to the casualty with confusion, ataxia and diplopia. What is the protocol to treat this patient?

a) Thiamine injection followed by glucose infusion

b) Glucose infusion followed thiamine injection

c) Glucose infusion alone

d) Thiamine injection alone

Question 7:

FMN and FAD coenzymes are derived from which of the following vitamin?

a) Niacin

b) Vitamin B1

c) Riboflavin

d) Vitamin B5

Question 8:

A young lady presented with a tingling sensation in her legs and hands. On examination, she had fissured tongue, lips, and lesions in the angle of the mouth. On investigation, she had low RBC glutathione reductase activity. Which vitamin is most likely deficient in this patient?

a) Vitamin B12

b) Vitamin B3

c) Niacin

d) Riboflavin

Question 9:

A man presents with rashes around the neck, as shown in the image below. Dietary history reveals that he has consumed jowar for a long duration. Which vitamin is most likely deficient in this patient?

a) Vitamin B5

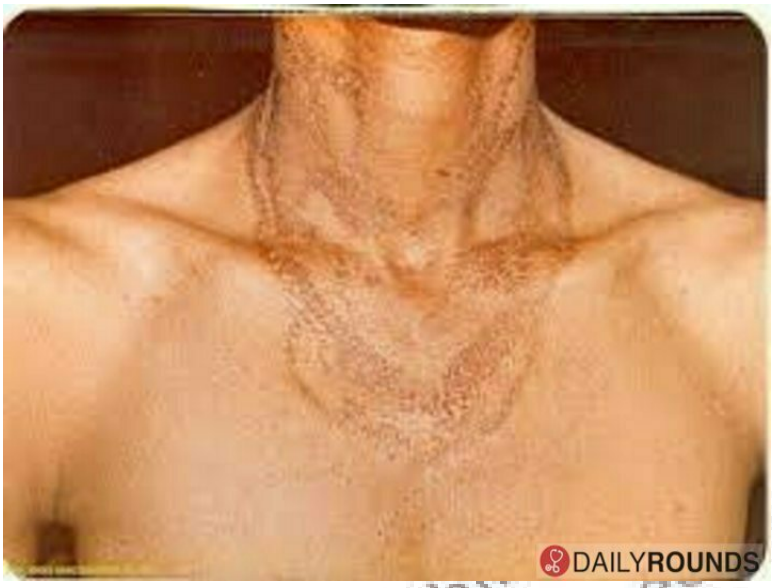
b) Vitamin B1

c) Vitamin B3

d) Vitamin B7

Question 10:

A farmer presents with loose stools, depression, and photosensitive dermatitis as shown in the image. On examination, a red beefy tongue is noted. Which of the following is not associated with this presentation?



a) Hartnup disease

b) Carcinoid syndrome

c) Wernicke's encephalopathy

d) Pyridoxine deficiency

Question 11:

A 45-year-old woman with hypertriglyceridemia is considered for niacin therapy. She is a known diabetic managed adequately with sugars well under control. Why should niacin be used cautiously in this patient?

a) It can cause hyperglycemia.

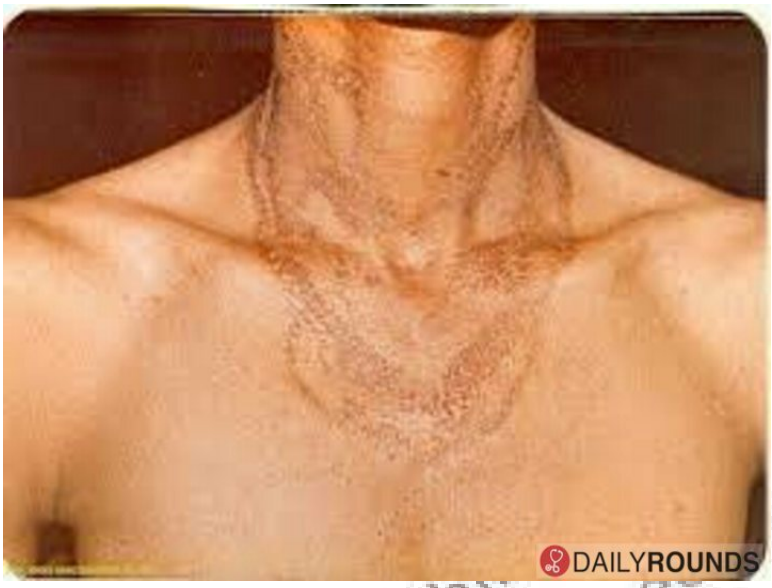
b) It can cause scleroderma which makes injecting insulin difficult.

c) It can increase the metabolism of oral hypoglyemic drugs.

d) It can cause hypoglycemia.

Question 12:

An alcoholic patient presented with itchy lesions as shown in the image, He also has a red-colored beefy tongue. Which of the following histories would you like to additionally ask/ elicit in this patient?



a) Ask for diarrhea and history of memory loss

b) Ask for diarrhea, thiamine deficiency features, and detailed dietary history

c) History of allergy

d) Ask for diarrhea and conjunctivitis

Question 13:

A middle aged man comes for routine follow-up. His lipid profile shows hypercholesterolemia, hypertriglyceridemia, elevated LDL and VLDL. Which of the following vitamins can be used to treat this patient?

a) Riboflavin

b) Pantothenic acid

c) Nicotinic acid

d) Pyridoxine

Question 14:

A patient is prescribed niacin for treating his hyperlipidemia. All of the following statements about this drug are true except:

a) It inhibits lipolysis in adipose tissue

b) It causes a reduction in the liver TAG synthesis

c) It reduces the level of HDL

d) It reduces the level of LDL

Question 15:

Which of the following vitamins constitutes a part of the coenzyme A?

a) Thiamine

b) Riboflavin

c) Pantothenic acid

d) Pyridoxine

Question 16:

Burning feet syndrome is caused by a particular vitamin deficiency. Which of the following amino acids is contained in this vitamin?

a) Phenylalanine

b) Arginine

c) Beta-alanine

d) Tryptophan

Question 17:

Which of the following vitamins serves as a cofactor in carboxylation reactions?

a) Thiamine

b) Riboflavin

c) Pyridoxine

d) Biotin

Question 18:

A 20-year-old bodybuilder presented with fatigue on moderate exercise. He had recently started eating raw eggs for protein. Which of the following enzymes is likely deficient in him?

a) Glucose-6-phosphatase

b) Glycogen phosphorylase

c) Pyruvate carboxylase

d) Phosphoenolpyruvate carboxykinase

Question 19:

Vitamin B12 is found maximum in:

a) Animal products

b) Green leafy vegetables

c) Roots and tubers

d) Vegetarian diet

Question 20:

Which vitamin, when infused in supraphysiological doses, causes macular edema and macular cysts?

a) Vitamin A

b) Vitamin D

c) Vitamin E

d) Vitamin B3

Answer Key

Question No.	Correct Option
1	d
2	a
3	b
4	a
5	b
6	a
7	c
8	d
9	c
10	c

11	a
12	a
13	c
14	c
15	c
16	c
17	d
18	c
19	a
20	d

Detailed Explanations

Solution to Question 1:

Thiamine pyrophosphate (TPP) does not serve as the coenzyme for isocitrate dehydrogenase.

Isocitrate dehydrogenase, an enzyme of the citric acid cycle, uses Mg^{2+} or Mn^{2+} ions as a coenzyme.

Thiamine pyrophosphate (TPP), from vitamin B_1, serves as the coenzyme for three multienzyme complexes that catalyze oxidative decarboxylation reactions:

- Pyruvate dehydrogenase - carbohydrate metabolism
- α-ketoglutarate dehydrogenase - citric acid cycle
- Branched-chain keto-acid dehydrogenase - metabolism of leucine, isoleucine, valine

Most of these enzymes are involved in carbohydrate metabolism, so thiamine is required in excess during a high carbohydrate diet. TPP also serves as the cofactor for transketolase in the HMP shunt.

Solution to Question 2:

Thiamine (Vitamin B_1) deficiency makes a patient prone to lactic acidosis.

Thiamine pyrophosphate (TPP) serves as a coenzyme for the pyruvate dehydrogenase enzyme. In a state of thiamine deficiency, there is an impaired conversion of pyruvate to acetyl CoA. When the patient consumes a high carbohydate diet, there is thus an accumulation of pyruvate. Pyruvate then gets converted into lactate by the enzyme lactate dehydrogenase. This results in increased plasma concentrations of lactate and pyruvate, leading to a state of lactic acidosis.

Solution to Question 3:

Decreased energy production is seen in thiamine deficiency because it acts as a coenzyme for pyruvate dehydrogenase and α-ketoglutarate dehydrogenase.

Both pyruvate dehydrogenase and α-ketoglutarate dehydrogenase are required for the production of NADH, which is used for ATP production. In thiamine deficiency, the reaction catalyzed by pyruvate dehydrogenase cannot go forward. Hence, there is no formation of acetyl CoA, causing decreased substrate for the citric acid cycle. As a result, energy (ATP) production is grossly reduced and instead, lactic acidosis is seen.

Solution to Question 4:

Cardiac failure (orthopnea and pedal edema) and cardiomegaly in a patient with chronic alcoholism suggest a diagnosis of wet beriberi. Thiamine (vitamin B1) deficiency leads to this condition.

Chronic alcohol consumption is associated with thiamine deficiency due to inadequate dietary intake, malabsorption of thiamine from the gastrointestinal tract and impaired utilization of thiamine in the cells. This deficiency presents as:

• Wet beriberi: The cardiovascular system is affected. Patients present with high output cardiac failure (orthopnea, pedal edema), dyspnea, cardiomegaly, and pulmonary edema.

• Dry beriberi: Both the peripheral and central nervous systems are affected. Patients present with symmetric motor and sensory neuropathy (pain, paresthesia, loss of reflexes) especially in lower limbs. In severe cases, muscle atrophy may also be seen.

Solution to Question 5:

Erythrocyte transketolase activity can be measured to diagnose thiamine deficiency.

Thiamine pyrophosphate (TPP) is the cofactor of erythrocyte transketolase, which is involved in the non-oxidative phase of the HMP pathway. In thiamine or vitamin B1 deficiency, the value of erythrocyte transketolase activity will be decreased.

Solution to Question 6:

Diplopia, ataxia and confusion in a patient with chronic alcohol consumption is suggestive of Wernicke's encephalopathy. The current standard of treatment for patients with symptoms of Wernicke's encephalopathy is to give thiamine 100 mg intravenously before administering glucose-containing IV fluids.

Thiamine deficiency leads to the following:

• Wernicke's encephalopathy: It is more commonly seen in alcoholics with chronic thiamine deficiency. It is characterized by global confusion, truncal ataxia, ophthalmoplegia and horizontal

nystagmus.

• Korsakoff psychosis: Symptoms in addition to those of Wernicke's encephalopathy include memory loss and confabulation

• Beriberi: It can be either -

• Dry beriberi: Sensorimotor peripheral neuropathy, muscle weakness

• Wet beriberi: Tachycardia, cardiomegaly, high-output cardiac failure

Solution to Question 7:

Coenzymes flavin mononucleotide (FMN) and flavin adenine dinucleotide (FAD) are derived from riboflavin (vitamin B2).

FMN-coenzyme is part of complex I (also known as NADH-Q oxidoreductase, NADH dehydrogenase) in electron transport chain (ETC). It also acts as a coenzyme for L-amino acid oxidase.

FAD-coenzyme is part of complex II in ETC. It acts as a coenzyme for succinate dehydrogenase (Kreb's cycle), acyl CoA dehydrogenase (fatty acid oxidation), D-amino acid oxidase and xanthine oxidase.

Solution to Question 8:

Tingling sensation of legs and hands, fissured tongue and lips, and lesions in the angle of the mouth combined with low RBC glutathione reductase activity is consistent with riboflavin (vitamin B2) deficiency.

Deficiency state of riboflavin, a water soluble vitamin, is characterized by the following signs and symptoms:

• Cheilosis: Pallor in the angle of mouth and fissuring of lips, which extends radially to skin

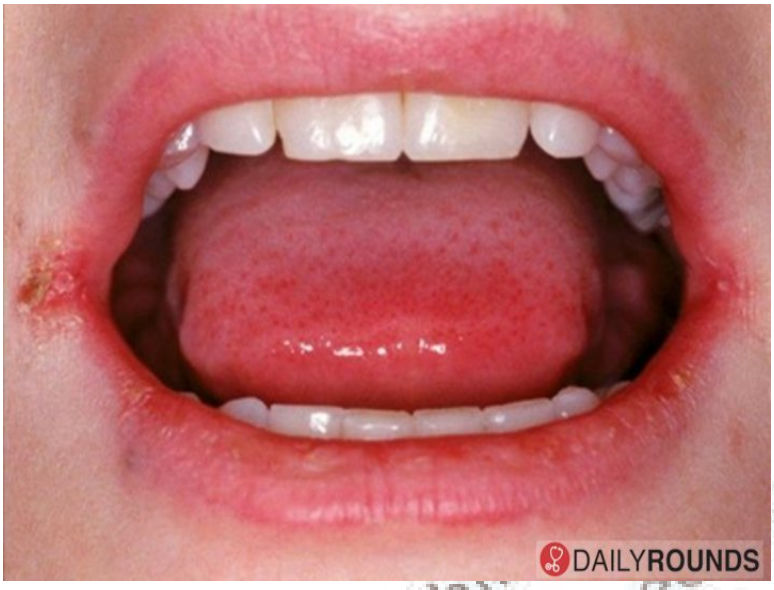
• Glossitis: Tongue becomes smooth, loss of papilla causing magenta-colored tongue

• Eyes: Keratitis, conjunctivitis and corneal vascularisation

• Other features: Seborrheic dermatitis, anemia

The nutritional status of riboflavin can be assessed by measuring the activity of the RBC glutathione reductase, by FAD added in vitro. This test is specific for riboflavin deficiency.

The image below shows cheilosis and stomatitis due to riboflavin deficiency.



Solution to Question 9:

A patient with high intake of jowar and characteristic Casal's necklace pattern rash is most likely suffering from pellagra. It is caused due to a deficiency of vitamin B3, known as niacin.

Pellagra, or niacin deficiency, presents with 3 classical Ds - dermatitis (photosensitive), diarrhea and dementia. Patients often have the Casal collar or Casal necklace. It is a clinical sign in which there is an erythematous pigmented skin rash in the distribution of a broad collar (dermatomes C3 and C4). If left untreated, pellagra leads to the 4th D of death.

Jowar (sorghum) has high leucine content. This inhibits QPRTase, an enzyme responsible for the synthesis of niacin from amino acid tryptophan. Excessive consumption of maize-based diets can also cause pellagra due to their low levels of niacin and tryptophan.

Solution to Question 10:

Loose stools, depression, beefy red tongue and Casal's necklace, as seen in the image, is suggestive of pellagra. Pellagra-like symptoms are not seen in Wernicke's encephalopathy.

Pellagra is caused by niacin deficiency, either due to dietary deficiency or deficient conversion of tryptophan to niacin. Hence, pellagra-like symptoms are seen in:

• Hartnup disease: It is an autosomal recessive disease caused by mutations in the SLC6A19 gene leading to defective membrane transport of tryptophan across PCT (kidney) and enterocytes (intestine). This leads to malabsorption, and hence deficiency of tryptophan.

• Carcinoid syndrome: Due to excess conversion of tryptophan to serotonin, there is a decreased level of tryptophan availavle for niacin synthesis

• Pyridoxine deficiency: Pyridoxine is required as the co-factor of kynureninase, an enzyme in the pathway of niacin synthesis.

• Maize/corn rich diet: In these grans, niacin is in bound form (niacytin form) and is not absorbed.

• Jowar diet (Sorghum): The increased leucine content inhibits QPRTase enzyme, an enzyme in the pathway of niacin synthesis.

Solution to Question 11:

Niacin should be used cautiously in diabetics as it can cause hyperglycemia.

Niacin induces insulin resistance and also decreases glucose-stimulated insulin secretion, which may result in severe hyperglycemia. When niacin is prescribed for patients with known or suspected diabetes, blood glucose levels should be monitored at least once a week.

Solution to Question 12:

An alcoholic patient with a red-colored beefy tongue, itching and the characteristic Casal's necklace, shown in the image, is suggestive of pellagra. It is caused by niacin/vitamin B3 deficiency. Hence, one should ask for a history of diarrhea and memory loss in this case.

Pellagra presents with 4 Ds: Diarrhea, Dermatitis, Dementia, and if untreated, Death. Early symptoms of pellagra are loss of appetite, generalized weakness and irritability, abdominal pain, and vomiting. Photosensitive dermatitis is seen over sun-exposed areas. Diarrhea occurs due to proctitis or malabsorption. Bright red glossitis (beefy tongue) and Casal's necklace (a pigmented, scaly skin rash that forms a ring around the neck) follow. Depressive psychosis and dementia also commonly occur.

Corn/ maize/ jowar based diets are the main causes of niacin deficiency. Corn/maize lacks niacin and tryptophan. The excess leucine in jowar and maize disrupts the conversion of tryptophan to niacin. Niacin deficiency is also seen in alcoholism. Hence, additional histories of diarrhea and memory loss will aid in supporting the diagnosis of pellagra (along with dietary history).

Treatment includes oral supplementation with 100–200 mg of nicotinamide or nicotinic acid three times daily for 5 days.

Option B: Thiamine deficiency (vitamin B1) features include CVS manifestations like heart failure and edema (wet beriberi), peripheral neuritis (dry beriberi), and Wernicke encephalopathy (in alcoholics).

Option C: Dermatitis seen in pellagra is due to dietary deficiency of niacin and so allergy history is not relevant.

Option D: Conjunctivitis is not a feature of niacin deficiency.

Solution to Question 13:

A man with hypercholesterolemia, hypertriglyceridemia, elevated LDL and VLDL should be treated with a lipid-lowering agent. Thus, the vitamin that can be used to treat this patient is

nicotinic acid (niacin or vitamin B3).

Niacin causes a reduction in LDL, VLDL and triglycerides along with increasing HDL. It is the only lipid-lowering agent that lowers lipoprotein (a) levels significantly. The main adverse effect of niacin is cutaneous flushing and pruritus. It can be controlled by pre-treatment with aspirin.

Solution to Question 14:

Niacin increases the level of HDL.

Niacin (vitamin B3) is known to inhibit lipolysis within the adipose tissue. Hence, the circulating free fatty acids (FFAs) are reduced. The liver normally uses these circulating FFAs as a precursor in the synthesis of triacylglycerol (TAG). As a result, niacin causes a reduction in liver TAG synthesis. TAGs are in turn required for very low density lipoprotein (VLDL) and low density lipoprotein (LDL) synthesis. Therefore both plasma VLDL and LDL values are reduced. Overall, niacin decreases LDL, VLDL and TAG levels, and increases high-density lipoprotein (HDL) levels. Thus it is particularly useful in the treatment of type IIb hyperlipoproteinemia, in which both VLDL and LDL are elevated.

Solution to Question 15:

Pantothenic acid constitutes a part of the coenzyme A.

The primary function of coenzyme A (pantothenic acid) is to transfer acyl groups. Pantothenic acid as coenzyme A is involved in:

- Fatty acid oxidation
- TCA cycle (as succinyl Co-A and acetyl Co-A)
- Cholesterol synthesis (as HMG Co-A)
- Acetylation reactions

Solution to Question 16:

The vitamin that is deficient in burning feet syndrome is vitamin B5 (pantothenic acid) which contains the amino acid beta-alanine.

Pantothenic acid deficiency is rarely seen as the vitamin is abundantly found in food (egg yolks, whole frains, vegetables). Burning feet syndrome is classically seen in pantothenic acid deficiency. The other symptoms of pantothenic deficiency are non-specific and include GI disturbances, paraesthesia, muscle cramps and ataxia.

Beta-alanine is an amino acid seen in:

- Pantothenic acid (vitamin B5)
- Coenzyme A

- Carnosine
- Anserine
- Acyl carrier protein

Solution to Question 17:

Biotin serves as an important cofactor in carboxylation reactions

Biotin (vitamin B7) functions as a carrier for the activated carbon dioxide molecule in carboxylation reactions. The enzymes using biotin as a cofactor are acetyl CoA carboxylase, pyruvate carboxylase, propionyl CoA carboxylase and methyl crotonyl CoA carboxylase.

Malic enzyme and carbomyl phosphate synthetase I and II do not use biotin as a coenzyme. Gamma carboxylation by vitamin K is also biotin-independent.

Solution to Question 18:

The patient developed pyruvate carboxylase enzyme deficiency.

Individuals who eat an excess of uncooked egg-white develop biotin deficiency and as a result carboxylase enzyme deficiency. This is because a protein called avidin present in the egg-white binds to biotin in the diet and makes biotin unavailable for intestinal absorption. Biotin deficiency can cause dermatitis, mental status changes (depression, hallucinations), paraesthesia and alopecia.

Biotin serves as a co-enzyme for the synthesis of several carboxylase enzymes such as acetyl-CoA carboxylase, pyruvate carboxylase, propionyl CoA carboxylase and methylcrotonyl-CoA carboxylase. Thus these enzymes are deficient in biotin deficiency.

Solution to Question 19:

Vitamin B12 is found maximum in animal products.

Vitamin B12 or cobalamin is synthesized only by microorganisms. Thus, the only source of cobalamin for humans is animal products. It is not found in plant sources. Therefore, individuals who are strict vegetarians are usually at the risk of developing vitamin B12 deficiency.

Solution to Question 20:

Macular cysts and edema may develop in patients due to the infusion of supraphysiological doses of vitamin B3 (niacin toxicity).

The toxic effects of niacin include:

• Flushing associated with skin dryness, itching, paresthesia, and headache is seen. These effects may be seen with even a low dose of 30mg/day. Pre-medication with aspirin alleviates these symptoms as these symptoms are mediated by prostaglandins.

• Macular edema and macular cysts

• Nausea, vomiting, and abdominal pain.

• Hepatic toxicity presents as jaundice with elevated AST and ALT levels.

• Fulminant hepatitis requiring liver transplantation.

• Glucose intolerance

• Hyperuricemia

Hematopoietic and other vitamins

Question 1:

Which of the following vitamins contains cobalt as a part of their structure?

a) Vitamin B1

b) Vitamin B2

c) Vitamin B6

d) Vitamin B12

Question 2:

Which of the following patients is likely to develop vitamin B12 deficiency?

a) A 40-year-old male with a segmental resection of primary duodenal cancer

b) An intestinal obstruction case with jejunectomy due to necrotic segment

c) A 30-year-old patient who got his ileum removed due to Crohn's disease

d) A 62-year-old patient who underwent colectomy for ulcerative colitis

Question 3:

A 45-year-old patient came with complaints of easy fatiguability and palpitations. On examination, she had pallor. Methylmalonic acid excretion in the urine was estimated as a part of evaluation. This can be utilized to assess the nutritional status of which vitamin?

a) Vitamin B12

b) Folic acid

c) Niacin

d) Iron

Question 4:

Folate trap occurs due to the deficiency of which of the following vitamins?

a) Pyridoxine

b) Vitamin B12

c) Niacin

d) Folic acid

Question 5:

A young woman presents with increasing fatigue, numbness, and tingling in the legs. She started following a vegan diet 4 years ago. On examination, glossitis is seen, and hyperreflexia is present in the lower limbs. Lab investigations revealed the following. Which of the following findings is not seen in this condition?

a) Decreased homocysteine levels

b) Positive Romberg sign

c) Optic atrophy

d) Dementia

Question 6:

Which of the following are vitamin-B12 dependent enzymes?

a) 1, 2 , 3 and 4

b) 1 and 4 only

c) 1 and 3 only

d) 1, 2, and 3

Question 7:

A patient presented with symptoms of peripheral neuropathy and macrocytic anemia. She was given folate 5mg and the blood picture improved. However, the neurologic manifestations were aggravated. What is the most likely cause?

a) Malabsorption of folate

b) Treatment with folate unmasking pyridoxine deficiency

c) Deficiency of folate reductase in CNS

d) Folate therapy using the B12 stores

Question 8:

Which of the following vitamins serves as a carrier for one carbon atom?

a) Thiamine

b) Niacin

c) Folic acid

d) Pantothenic acid

Question 9:

Which of the following vitamins serves as a coenzyme in transamination reactions?

a) Thiamine

b) Riboflavin

c) Pyridoxine

d) Biotin

Question 10:

Which of the following vitamins is not required in the TCA cycle?

a) Thiamine

b) Niacin

c) Riboflavin

d) Pyridoxine

Question 11:

A 45-year-old man is undergoing treatment for pulmonary tuberculosis. During one of his follow-ups, it was noted that he is taking pyridoxine tablets in excess. Which of the following symptoms is he most likely to develop?

a) Dementia

b) Hallucinations

c) Sensory neuropathy

d) Nystagmus

Question 12:

Which of the following enhances iron absorption?

a) Vitamin A

b) Vitamin C

c) Thiamine

d) Riboflavin

Question 13:

Which of the following is false about scurvy?

a) Microcytic anemia can be seen

b) The beading on ribs is non-tender and round

c) Patient can present with pseudoparalysis

d) There is impaired formation of mature connective tissue

Answer Key

Question No.	Correct Option
1	d
2	c
3	a
4	b
5	a
6	b
7	d
8	c
9	c
10	d
11	c
12	b
13	b

Detailed Explanations

Solution to Question 1:

Vitamin B12 contains corrin rings with cobalt as a part of its structure, hence this group of vitamins are referred to as cobalamins.

Vitamin B12 is derived mainly from animal products such as meat, fish, eggs and fortified cereals. There are no plant sources as such for vitamin B12 / cobalamine, hence strict vegetarians are at risk of developing a nutritional deficiency of vitamin B12.

Vitamin B12 deficiency is associated with defects in RBC production leading to megaloblastic anemia.

Solution to Question 2:

A 30-year-old patient who got his ileum removed due to Crohn's disease is likely to develop vitamin B12 deficiency. Vitamin B12 is absorbed from the distal third of the ileum via receptors known as cubulin, which binds the intrinsic factor-vitamin B12 complex.

Dietary vitamin B12 is normally bound to proteins in food. Pepsin and gastric acid in the stomach degrade these food proteins and release vitamin B12. The free vitamin B12 then binds to a salivary glycoprotein called haptocorrin (R-binder).

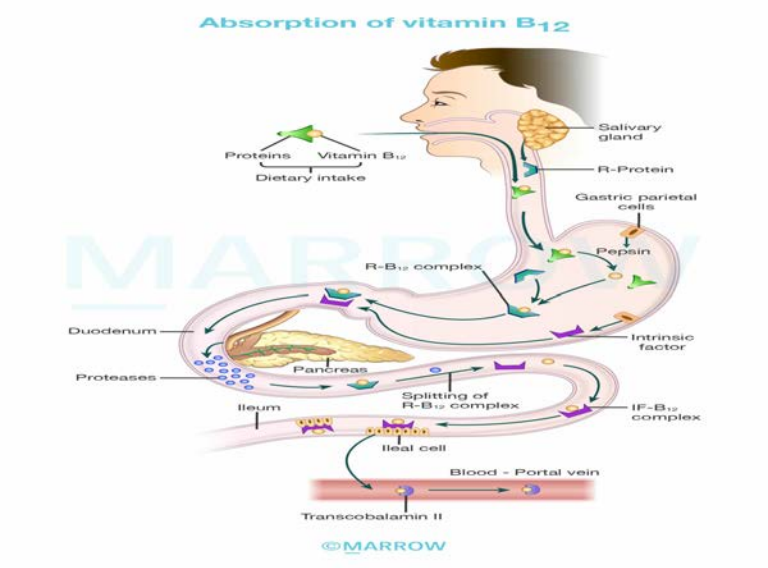
In the alkaline pH of the duodenum, pancreatic proteases degrade the haptocorrin, and vitamin B12 is released again. It then binds tightly to intrinsic factor (IF), a glycoprotein secreted by the parietal cells of the stomach and this binding takes place in the duodenum. The IF-Vitamin B12 complex reaches the terminal ileum and attaches to a receptor on the enterocyte, from where it is absorbed. Vitamin B12 is transported in the blood bound to a carrier protein called transcobalamin II and delivered to the peripheral tissues.

Absorption sites:

Iron- Duodenum

Folate- Jejunum

Vit B12- Ileum



Solution to Question 3:

The above scenario suggests anemia and methylmalonic acid excretion in the urine can be utilized to assess the nutritional status of vitamin B12.

Methylmalonyl-CoA mutase is a vitamin B12-dependent enzyme. The activity of methylmalonyl CoA mutase is greatly reduced in vitamin B12 deficiency, leading to an accumulation of methylmalonic acid and its excretion in urine.

There are two vitamin B12-dependent enzymes:

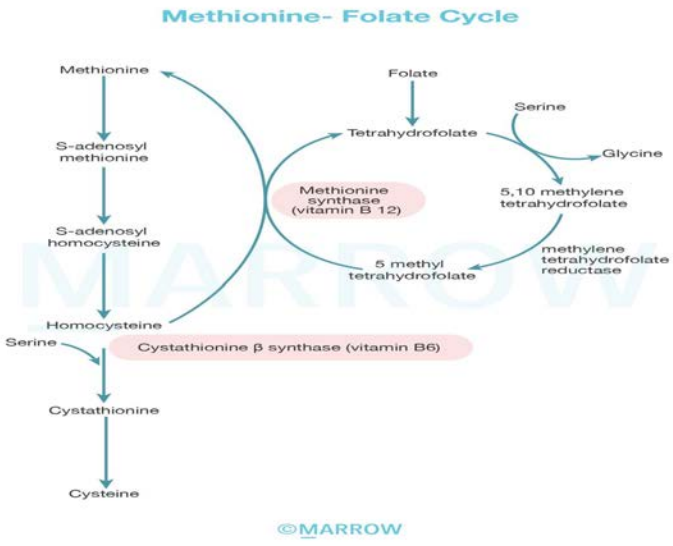
• Methionine synthase (homocysteine methyltransferase): Catalyzes the conversion of homocysteine to methionine

• Methylmalonyl CoA mutase: methylmalonyl CoA, formed as an intermediate in the catabolism of valine, isoleucine, and propionyl CoA is converted to succinyl CoA

Note: Urinary excretion of methylmalonic acid is also used to differentiate between megaloblastic anemia due to folate and vitamin B12 deficiency.

Solution to Question 4:

Folate trap occurs in the deficiency of vitamin B12.



The conversion of 5,10-methylene-THF into 5-methyl-THF(methyltetrahydrofolate) is an irreversible process. The major source of tetrahydrofolate for tissues is 5-methyl-THF. The only way to make further use of 5-methyl-THF and to maintain the folate cycle is by the vitamin B12-dependent remethylation of homocysteine to methionine by methionine synthase enzyme.

Decreased activity of methionine synthase in vitamin B12 deficiency results in the accumulation of methyltetrahydrofolate that cannot be used, resulting in a folate trap. The folate cycle provides a link between the functions of folate and vitamin B12, where vitamin B12 deficiency causes a functional deficiency of folate.

Solution to Question 5:

The clinical scenario of a patient on a vegan diet now presenting with glossitis of the tongue, hyperreflexia, and numbness and tingling in the legs is suggestive of vitamin B12 deficiency. Decreased homocysteine levels are not seen in this condition. Rather, increased homocysteine levels are seen.

Vitamin B12 is a cofactor of methionine synthase and methylmalonyl CoA mutase. Methionine synthase is required for the conversion of homocysteine to methionine, which is the principal methyl donor. In vitamin B12 deficiency, defective methionine synthase leads to the following:

1. In the absence of methionine, there is decreased methylation in the CNS. This leads to defective synthesis of myelin and neurotransmitters, which results in the following symptoms :

• Posterolateral cord degeneration: It presents as loss of proprioception and vibration, positive Romberg sign, ataxia, hyperreflexia, spasticity

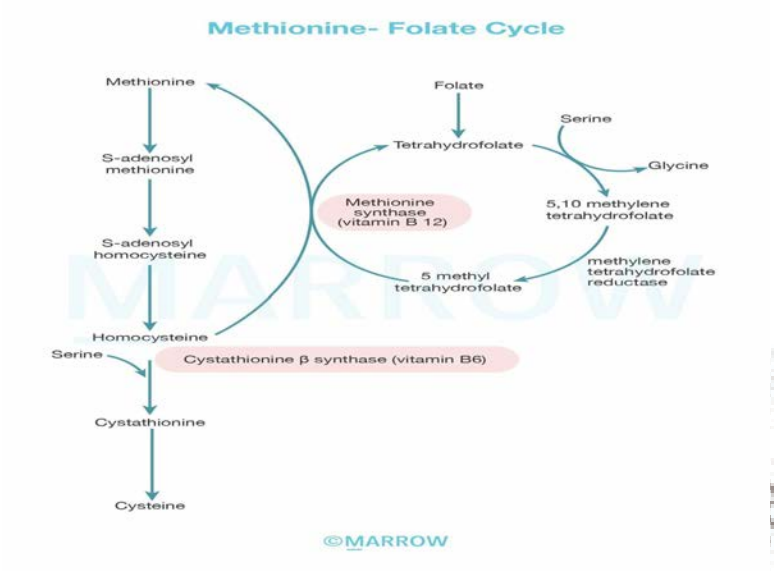
• Dementia leading to cognitive impairment

• Peripheral neuropathy: presenting as muscle weakness, paraesthesia

• Optic atrophy

2. Homocysteine accumulates in the blood conferring an increased risk of cardiovascular disease.

3. Megaloblastic anemia: The availability of 5,10-methylene THF is reduced, which is required in the conversion of deoxyuridine monophosphate (dUMP) to thymidine monophosphate (TMP) for DNA synthesis. This leads to macrocytic anemia with megaloblasts in the bone marrow.



Solution to Question 6:

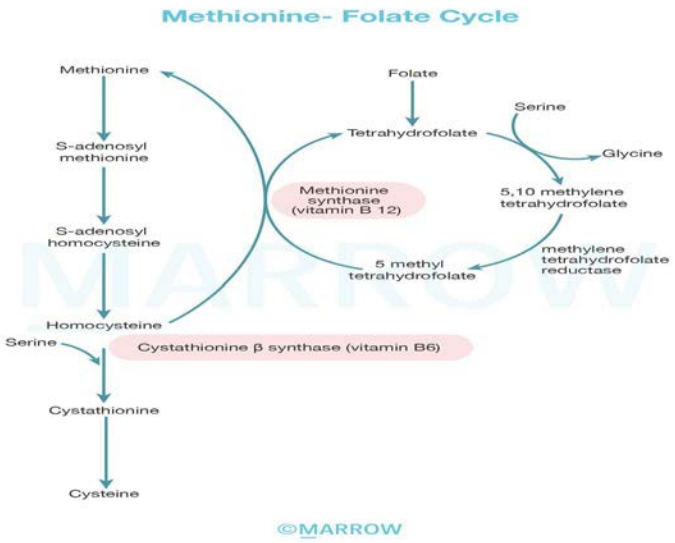
Among the given enzymes, methyl malonyl CoA isomerase and methionine synthase are vitamin B12 dependent enzymes.

There are two vitamin B12-dependent enzymes:

• Methionine synthase (homocysteine methyltransferase): Catalyzes the conversion of homocysteine to methionine

• Methylmalonyl CoA mutase: methylmalonyl CoA, formed as an intermediate in the catabolism of valine, isoleucine, and propionyl CoA is converted to succinyl CoA

The below figure shows B12 serving as a cofactor of methionine synthase. Its deficiency may cause a folate trap.

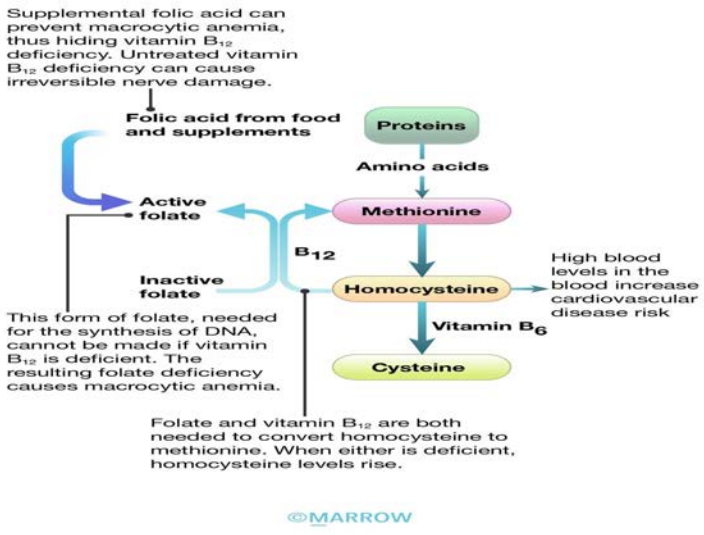


Solution to Question 7:

The most likely cause is folate therapy which used up the B12 stores, aggravating the symptoms.

Administration of folic acid for patients with macrocytic anemia can partially reverse the hematologic abnormalities associated with vitamin B12 deficiency. However, the neurologic manifestations of vitamin B12 deficiency are not treated by folic acid.

Administration of folic acid to an individual with vitamin B12 deficiency can potentially mask untreated vitamin B12 deficiency or even worsen the neurologic complications. Hence, testing for and/or treatment of vitamin B12 deficiency is required in all patients being treated with folic acid.



Solution to Question 8:

Folic acid serves as a carrier of one carbon atom.

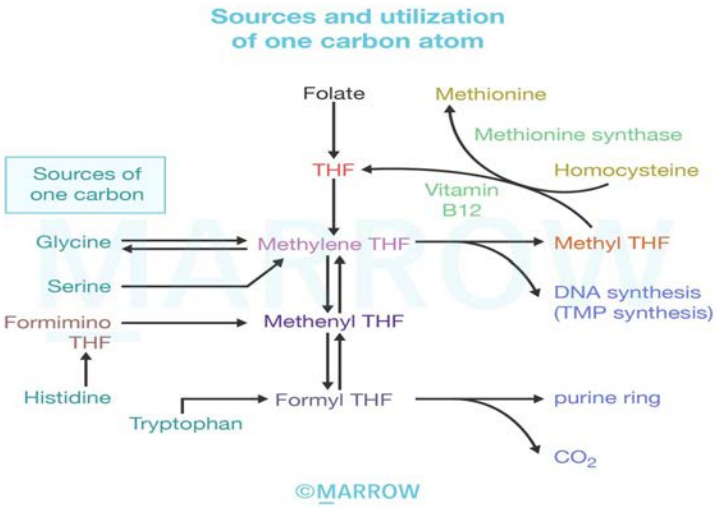
Tetrahydrofolate receives one-carbon fragments from donors such as serine, glycine, and histidine and transfers them to intermediates in the synthesis of amino acids, purines, and pyrimidines.

The manifestations of folic acid deficiency includes:

• Megaloblastic anemia

• Homocystinemia

• Neural tube defect (spina bifida, anencephaly)



Note: The major point of entry for one-carbon fragments, in one-carbon metabolism, is methylenetetrahydrofolate.

Solution to Question 9:

Pyridoxal phosphate (PLP), an active form of pyridoxine serves as a coenzyme in transamination reactions.

Pyridoxal phosphate (PLP) is a coenzyme for enzymes involved in the following:

• Transamination

• Decarboxylation

• Glycogenolysis: Glycogen phosphorylase

• Transsulfuration reaction: Cystathionine β-synthase and cystathionase

• Heme synthesis: ALA synthase (deficiency causes microcytic hypochromic anemia)

• Tryptophan metabolism: Kynureninase (deficiency causes pellagra-like symptoms)

Note: Pyridoxal phosphate (PLP) serves as a coenzyme in many amino acid metabolisms, so its RDA mainly depends on daily protein intake.

Solution to Question 10:

Pyridoxine is not required in the TCA cycle.

Vitamins, in the form of their respective coenzymes, are required for the proper functioning of the TCA cycle.

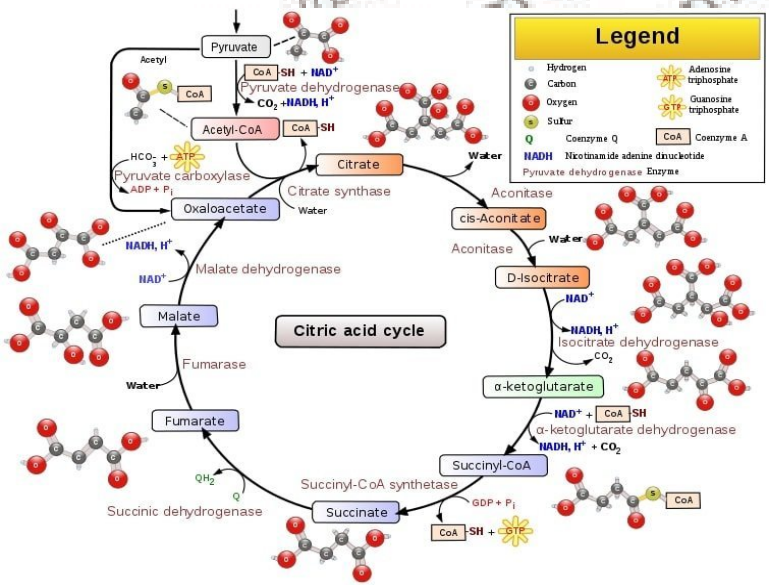
The following vitamins are needed in the TCA cycle:

• Thiamine as TPP, for decarboxylation in the α-ketoglutarate dehydrogenase reaction

• Riboflavin as FAD and FMN, for succinate dehydrogenase

• Niacin as NAD and NADH, for isocitrate dehydrogenase, α-ketoglutarate dehydrogenase, and malate dehydrogenase

• Pantothenic acid as a part of Coenzyme A helps to form acetyl-CoA and succinyl-CoA



Solution to Question 11:

Excess pyridoxine (vitamin B6) causes sensory neuropathy. It occurs at intakes above 2 g/day.

There are only two B-complex vitamins that cause toxicity:

• Niacin: It is associated with:

• Prostaglandin-mediated cutaneous flushing

• Fulminant hepatitis (most fatal manifestation)

• Glucose intolerance

• Pyridoxine: It is associated with sensory neuropathy.

Solution to Question 12:

Iron absorption is enhanced by vitamin C.

Vitamin C reduces the ferric iron to its ferrous form, thereby helping in the process of absorption of dietary iron from the intestine.

Vitamin C deficiency causes

• Decreased absorption of iron leading to microcytic-hypochromic anemia

• Decreased hydroxylation of proline and lysine, which results in defective collagen synthesis and bleeding manifestations.

Conversely, vitamin C toxicity (>2000 mg/ day) can lead to iron overloaded states.

Note: Alcohol and fructose also enhance iron absorption. However, the absorption of both inorganic and heme iron is impaired by calcium. So, a glass of milk with a meal significantly reduces iron availability.

Solution to Question 13:

The beading of ribs in scurvy is sharp and tender.

A deficiency state of vitamin C results in scurvy. The clinical features are:

• Fatigue, irritability, loss of appetite

• Loose and spongy gums along with loose teeth

• Swollen joints

• Perifollicular hemorrhages

• Bleeding into the skin - ecchymosis, purpura, petechiae

• Pseudoparalysis

• Microcytic anemia (due to decreased absorption of iron from the intestine)

• Scorbutic rosary - sharp, tender beading at the costochondral junctions.

Many of the deficiency symptoms can be attributed to deficient collagen synthesis caused due to an inability to hydroxylate the prolyl and lysyl residues of collagen.

Antioxidants & Minerals

Question 1:

Which of the following is not a reactive oxygen species?

a) Superoxide

b) Glutathione

c) Hydroxyl radical

d) Oxygen singlet

Question 2:

Which of the following method is not used for the measurement of body free radical burden?

a) Ferrous oxidation in xylenol assay

b) Measurement of pentane and methane in exhaled air

c) Estimation of dialdehydes

d) Direct assay of free radicals

Question 3:

Which of the following is a chain-breaking antioxidant?

a) Glutathione peroxidase

b) Vitamin E

c) Catalase

d) Selenium

Question 4:

Which of the following kills lipid-engorged macrophages?

a) Esterified cholesterol

b) Unesterified cholesterol

c) Unesterified lipoprotein

d) Esterified lipoprotein

Question 5:

A child is brought to the clinic due to repeated episodes of ear and lung infections. History and investigations have revealed a deficiency of the enzyme responsible for respiratory burst in the neutrophils. Which of the following enzyme is deficient?

a) Catalase

b) NADPH oxidase

c) Superoxide dismutase

d) Myeloperoxidase

Question 6:

Which of the following acts as a lipid soluble radical-trapping antioxidant?

a) Ascorbate

b) Uric acid

c) Polyphenols

d) Ubiquinone

Question 7:

A middle-aged obese woman had elevated liver enzymes on routine follow-up. A USG showed grade 2 fatty liver. She is non-diabetic, non-hypertensive and has never consumed alcohol. Which of the following can be considered as a first-line pharmacological treatment option for this patient?

a) Vitamin B6

b) Selenium

c) Vitamin E

d) Vitamin C

Question 8:

A homeless man is brought to the ER by the police after he was found shouting unnecessarily at people in a nearby area. He is confused and has a slurred speech. His blood ethanol levels are 220 mg/dl and his glucose levels are 45 mg/dL. The toxic effects of the substance consumed by this patient are due to which of the following?

a) Decreased lactate/pyruvate ratio

b) Stimulation of gluconeogenesis

c) Increased NADH/NAD+ ratio

d) Stimulation of fatty acid oxidation

Question 9:

An infant is brought to the pediatric clinic with symptoms of diarrhea, hair loss, and failure to thrive. On clinical examination, there is inflammation of the skin around the mouth and the perianal region. Which mineral deficiency is responsible for this disease?

a) Zinc

b) Selenium

c) Chromium

d) Copper

Question 10:

Zinc is a cofactor of which of the following enzymes?

a) Xanthine oxidase

b) Glutathione peroxidase

c) Carbonic anhydrase

d) Lysyl oxidase

Question 11:

Which of the following enzyme action is not affected in a patient with Menke's disease?

a) Superoxide dismutase

b) Dopamine hydroxylase

c) Glutathione peroxidase

d) Amine oxidase

Question 12:

An exchange student from China presented to the ER with features of heart failure. On treatment, he was stabilized and later reported that he is a known case of Keshan disease. This disease is due to deficiency of which of the following?

a) Thiamine

b) Selenium

c) Zinc

d) Copper

Question 13:

Magnesium does not have a role in which of the following processes?

a) Calcium homeostasis

b) Maintenance of normal cardiac rhythm

c) Copper metabolism

d) Neuromuscular function

Question 14:

An adolescent boy presents with jaundice and tiredness. On investigation, his AST was 150 IU/L, ALT was 175 IU/L and bilirubin was 4.5mg/dl. Serum ceruloplasmin levels were found to be low. Which of the following minerals is used in the treatment of this condition?

a) Copper

b) Zinc

c) Magnesium

d) Molybdenum

Answer Key

Question No.	Correct Option

1	b
2	d
3	b
4	b
5	b
6	d
7	c
8	c
9	a
10	c
11	c
12	b
13	c
14	b

Detailed Explanations

Solution to Question 1:

Glutathione is not a reactive oxygen species (ROS).

Glutathione is an antioxidant. Reduced glutathione is used for detoxification of hydrogen peroxide.

Bridges–Good syndrome is a chronic granulomatous disease due to a deficiency of NADPH oxidase. In this syndrome, the lysosomes are unable to produce hydrogen peroxide to kill the bacteria. Such patients have increased susceptibility towards catalase-positive organisms.

Note:

• The most powerful oxygen-free radical is hydroxyl radical (OH-)

• The least powerful reactive oxygen species is hydrogen peroxide

• The precursor of all reactive oxygen species is superoxide radical (O_2^-)

Solution to Question 2:

Direct assay of free radicals is not used for the measurement of total body free radical burden.

This is because free radicals are extremely reactive and short-lived. Free radical activity is usually assessed by indirect methods. Thus, the total body free radical burden can be estimated by measuring the products of lipid peroxidation using the following methods:

• FOX (Ferrous oxidation in xylenol orange) assay

• Estimation of dialdehydes: The dialdehydes formed from lipid peroxides can be measured by reaction with thiobarbituric acid.

• Measurement of pentane and methane in exhaled air

Note: Radical damage to unsaturated fatty acids in cell membranes and plasma lipoproteins leads to the formation of lipid peroxides.

Solution to Question 3:

Vitamin E is a chain-breaking antioxidant.

Antioxidants are used to control and reduce lipid peroxidation during which reactive oxygen species (ROS) are formed. Antioxidants thus fall into two classes:

• Preventive: These reduce the rate of chain initiation of lipid peroxidation.

• Chain-breaking: These interfere with the chain propagation phase of lipid peroxidation.

Solution to Question 4:

Lipid-engorged macrophages are killed by unesterified cholesterol.

Free radicals can damage the proteins and lipids in plasma low-density lipoprotein (LDL). They are chemically modified to form abnormal LDL which are not recognized by the liver LDL receptors. Thus, abnormal LDL is not cleared by the liver and is instead taken up by macrophage scavenger receptors. These lipid-engorged macrophages infiltrate the blood vessel endothelium. These macrophages are then killed by the high content of unesterified cholesterol they have accumulated. This causes the development of atherosclerotic plaques which, over time, can completely occlude a blood vessel.

Solution to Question 5:

The enzyme responsible for the respiratory burst in neutrophils is NADPH oxidase, which is deficient in this child.

NADPH oxidase is an enzyme in the inflammatory cells (neutrophils, eosinophils, monocytes, and macrophages) which produces superoxide anion by reducing oxygen. It does so by a process of respiratory burst during phagocytosis. Most of the enzymes that produce and require superoxide are contained in the peroxisomes, together with superoxide dismutase, catalase, and peroxidases.

$NADPH + 2O_2 \circ \quad NADP+ + 2 \cdot O_2- + 2H+$

NADPH deficiency can lead to chronic granulomatous disease (CGD), which is characterised by an inability of neutrophils and monocytes to kill catalase-positive organisms. Children present with recurrent ear, lung or bone infections.

Note:

There are multiple sources of oxygen radicals in the body:

• Ionizing radiation (x-rays and UV) causes lysis of water, leading to the formation of hydroxyl radicals

• Transition metal ions (Cu^{+}, Co^{2+}, Ni^{2+}, and Fe^{2+}) can react nonenzymatically with oxygen or hydrogen peroxide to form hydroxyl radicals

• Nitric oxide can react with superoxide to yield peroxynitrite, which decays to form hydroxyl radicals

• NADPH oxidase in the inflammatory cells produces superoxide

Solution to Question 6:

Ubiquinone acts as a lipid soluble radical-trapping antioxidant.

Lipid-soluble radical-trapping antioxidants like ubiquinone and carotenes trap radicals in plasma membranes and plasma lipoproteins. The water soluble radical-trapping antioxidants are ascorbate, uric acid and polyphenols derived from plant foods.

Solution to Question 7:

An obese woman with grade 2 fatty liver in the absence of alcohol consumption is suggestive of non-alcoholic steatohepatitis (NASH). Vitamin E is considered as a first-line pharmacological treatment option for NASH in non-diabetics when diet and lifestyle changes have failed.

Vitamin E is well tolerated and shows modest improvements in aminotransferase levels, radiographic features of hepatic steatosis, and/or histologic features of NASH. It is important to note that vitamin E must be considered only for non-diabetic patients due to cardiovascular risks with chronic vitamin E therapy.

Note: The FDA in 2024 has approved Resmetirom, a thyroid hormone receptor-beta (THR-beta) agonist for noncirrhotic NASH with moderate to advanced fibrosis.

Solution to Question 8:

Confusion and slurred speech with elevated blood ethanol levels suggests ethanol intoxication. The toxicity of ethanol is due to the increased NADH/NAD+ ratio.

Ethanol is metabolised in the liver by alcohol dehydrogenase. This reaction generates NADH. A high concentration of NADH leads to a high NADH/NAD+ ratio. Due to this, the metabolic reactions which require NAD+ are inhibited. This is the basic cause of all the metabolic effects of alcohol.

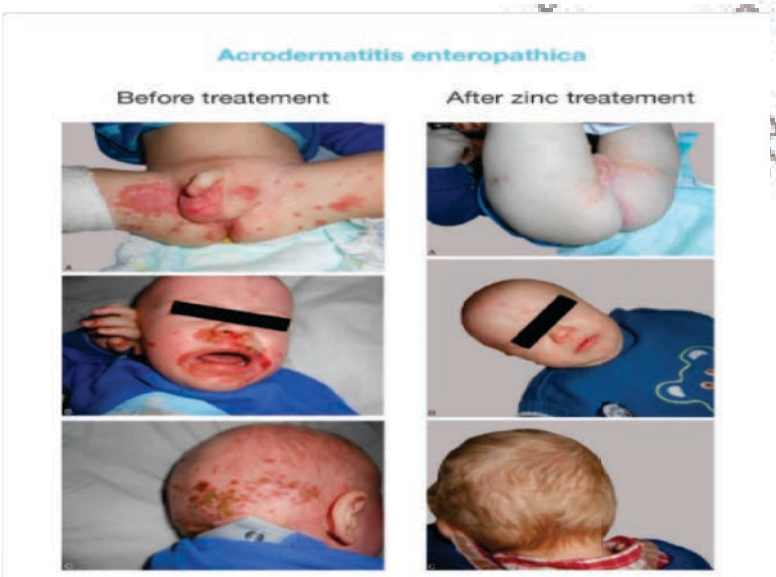
Glycolysis, citric acid cycle and fatty acid oxidation are inhibited. Ketogenesis is favoured. Pyruvate is shunted for lactate synthesis due to abundance of NADHand lactate/pyruvate ratio increases. Thus gluconeogenesis, which requires pyruvate as a substratre, is inhibited. This leads

to hypoglycemia.

Solution to Question 9:

An infant with diarrhea, hair loss, failure to thrive and dermatitis around orifices likely has acrodermatitis enteropathica which is due to the deficiency of zinc.

Acrodermatitis enteropathica is an autosomal recessive disease due to a defect in zinc absorbing transmembrane protein. Affected infants present with growth retardation, diarrhea, alopecia, gonadal atrophy, muscle wasting and impaired wound healing. A rash is classically present. It involves the extremities, face, and perineum and is characterized by vesicular and pustular crusting with scaling and erythema.



Solution to Question 10:

Zinc acts as a cofactor for carbonic anhydrase.

Zinc is a component of many metalloenzymes including carbonic anhydrase II. Zinc also has antioxidant activity as extracellular superoxide dismutase is zinc-dependent.

Chronic zinc deficiency can cause stunted growth, hypogeusia (decreased taste sensation) hypogeusia, impaired immune function, hypogonadism and hypopigmented hair. Acrodermatitis enteropathica is an autosomal recessive disorder characterized by abnormalities in zinc absorption.

Solution to Question 11:

In a patient with Menke's disease, glutathione peroxidase action is not affected.

Menke's disease, or kinky/steely hair syndrome is an X-linked recessive condition, where there is a mutation in the ATP7A gene. This leads to defective copper-binding P-type ATPase and copper is not mobilized from the intestine. Thus copper deficiency occurs and enzymes containing copper are affected, like superoxide dismutase, dopamine hydroxylase and amine oxidase.

Glutathione peroxidase is not affected since it requires selenium in the form of selenocysteine.

Other copper deficiency disorders include Wilson's disease and MEDNIK syndrome. Wilson's disease is an autosomal recessive disorder, due to ATP7B mutation, a gene encoding for copper transporting ATPase in the cells. There is defective biliary copper excretion from liver cells, leading to copper deposits in the liver and brain. MEDNIK syndrome is a rare multisystem disorder of copper metabolism with features of both Wilson's and Menke's disease.

Note:

Other enzymes requiring copper are ferroxidase (ceruloplasmin) , cytochrome c oxidase, tyrosinase (melanin synthesis), lysyl oxidase (cross-linking in collagen). It is also a component of ferroportin, which is involved in iron absorption.

Solution to Question 12:

Keshan disease is due to a deficiency of selenium.

Selenium is an important trace element that is a part of the 21st amino acid selenocysteine. There are many selenium-containing enzymes such as glutathione peroxidase, de-iodinase and thioredoxin reductase. Selenium is also an intracellular antioxidant that complements the role of Vitamin E. Dietary sources of selenium include seafood, muscle meat, and some cereals.

Selenium deficiency leads to Keshan disease and Kashin-Beck disease. Keshan disease is an endemic cardiomyopathy found in children and young women residing in regions of China where dietary intake of selenium is low. Kashin-Beck disease is a chronic, endemic osteoarthropathy seen in regions of China and Russia.

Chronic ingestion of high amounts of selenium leads to selenosis or selenium toxicity. It is characterized by hair and nail brittleness, garlic breath odor, myopathy, irritability, and other abnormalities of the nervous system.

Solution to Question 13:

Magnesium has no role in copper metabolism.

Magnesium is a major intracellular cation required for the function of various enzymes such as hexokinase, phosphofructokinase, alkaline phosphatase, enolase. The role of magnesium can be seen in the following processes:

• Calcium homeostasis: One-half of the total body magnesium is located in bone. Magnesium is important for parathormone secretion which controls serum Ca^{2+} level.

• Neuromuscular function:

• Hypomagnesemia may cause generalized alterations in neuromuscular function including tetany, tremor, seizures.

• Hypermagnesemia is known to cause symptoms of neuromuscular blockade such as hypoactive tendon reflexes, GI hypomotility, paralysis.

• Maintenance of cardiac rhythm:

• Hypomagnesemia can lead to supraventricular and ventricular arrhythmias.

• Hypermagnesemia can cause bradycardia, heart block, and asystole.

Magnesium deficiency symptoms are similar to those of calcium deficiency, but symptoms will be relieved only when magnesium is given. Oral administration of magnesium may lead to diarrhea, hence intravenous magnesium sulfate is given.

Solution to Question 14:

An adolescent with jaundice, elevated AST, ALT and low serum ceruloplasmin likely has Wilson's disease. Zinc is used in the treatment of this condition.

Wilson's disease is an autosomonal recessive disorder. It is charactered by mutations of the ATP7B gene, resulting in decreased biliary copper excretion and ceruloplasmin formation. The accumulation of copper leads to liver disease, hemolysis, a movement disorder neurologic disease, and/or psychiatric illness.

Elemental zinc is commonly used as a maintenance therapy along with chelating agents. Zinc acts by blocking the intestinal absorption of copper. It can also be used as first-line therapy in pregnancy.

Basics of genetics - Nucleotide metabolism and its disorders

Question 1:

Which of the following nitrogenous base contain amino group on the 6th carbon?

a) Adenine

b) Guanine

c) Cytosine

d) Thymine

Question 2:

Which of the following process do not contribute to purine nucleotide biosynthesis?

a) Synthesis from amphibolic intermediates

b) Phosphoribosylation of purines

c) Phosphorylation of purine nucleosides

d) Phosphoribosylation of purine nucleosides

Question 3:

Which of the following molecule doesn't contribute to the synthesis of a purine ring?

a) CO_2

b) Aspartate

c) Glutamine

d) Lysine

Question 4:

What is the rate-limiting step of de novo purine synthesis?

a) PRPP to Phosphoribosyl amine by PRPP glutamyl amidotransferase

b) PRPP to Phosphoribosyl amine by PRPP synthetase

c) Ribose 5-phosphate to PRPP by PRPP synthetase

d) Ribose 5-phosphate to PRPP by PRPP glutamyl amidotransferase

Question 5:

The parent purine nucleotide made from de novo purine synthesis is _____

a) AMP

b) GMP

c) IMP

d) TMP

Question 6:

A 4-year-old girl is started on rasburicase before initiating chemotherapy for acute lymphoblastic leukemia. What is the end product of the breakdown of purine nucleotides in her?

a) Allantoin

b) Amino acid

c) Ammonia

d) Uric acid

Question 7:

Which of the following diseases is not associated with purine degradation?

a) Gout

b) Severe combined immunodeficiency syndrome

c) Purine nucleoside phosphorylase deficiency

d) Orotic aciduria

Question 8:

Which of the following bases can't be salvaged to the corresponding nucleotide?

a) Uracil

b) Adenine

c) Hypoxanthine

d) Guanine

Question 9:

A 4-year-old child presents with hyperuricemia and delayed developmental milestones. He also has the habit of biting his lips, fingers, and feet because of which he is always strapped to the bed. Urine examined showed orange sands. Which is the most probable enzyme deficiency?

a) Adenine phosphoribosyltransferase

b) Hypoxanthine-guanine phosphoribosyltransferase

c) Adenosine Deaminase

d) Purine nucleoside phosphorylase

Question 10:

Which of the following is not required for pyrimidine ring synthesis?

a) Aspartate

b) CO_2

c) Glycine

d) Glutamine

Question 11:

The only mitochondrial step in pyrimidine synthesis is catalyzed by the enzyme _____

a) Carbamoyl phosphate synthetase-2

b) Aspartate transcarbamoylase

c) Dihydroorotase

d) Dihydroorotate dehydrogenase

Question 12:

The rate-limiting enzyme for pyrimidine synthesis in human is _____

a) Aspartate Transcarbamoylase

b) Carbamoyl Phosphate Synthetase I

c) Carbamoyl Phosphate Synthetase II

d) PRPP glutamyl amidotransferase

Question 13:

Which is the first true pyrimidine nucleotide formed in pyrimidine synthesis?

a) UDP

b) CTP

c) UMP

d) TMP

Question 14:

Which of the following is not a product of pyrimidine catabolism?

a) NH_3

b) β-alanine

c) β-aminoisobutyrate

d) Uric acid

Question 15:

The blood report of a patient shows increased uric acid levels. Which of the following diagnosis can be ruled out?

a) von Gierke's disease

b) Xanthinuria

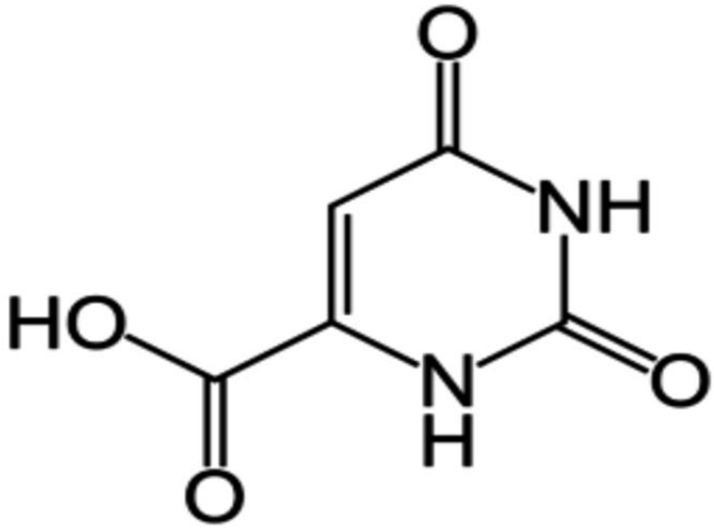
c) Lesch-Nyhan syndrome

d) Kelley-Seegmiller syndrome

Question 16:

An 11-month-old infant presented with growth failure and developmental delay. Her peripheral smear showed macrocytic anemia. Vitamin B12 or folic acid levels in plasma were

normal. No evidence of Transcobalmin -2 deficiency was found. The compound given in the picture is excreted in the urine. What is the most likely diagnosis?



a) Gout

b) Lesch Nyhan Syndrome

c) Xanthinuria

d) Orotic aciduria

Question 17:

A 14 year old male is brought with the complaints of multiple hyperpigmented papules over the face and other sun exposed areas with persistent erythema. He had history of similar episodes since childhood. Which of the following mechanisms is likely to be defective in this condition?

a) Removal of mismatched base during DNA replication

b) Removal of oligonucleotides containing damaged bases by endonuclease

c) Removal of the altered base by base specific glycosylase

d) Repair of double stranded breaks

Answer Key

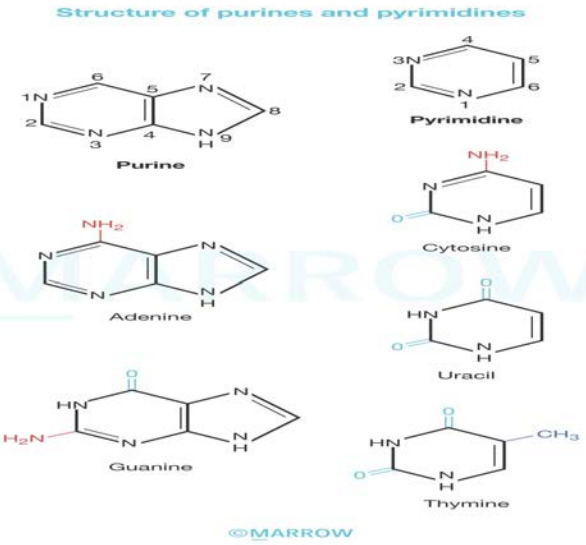
Question No.	Correct Option

1	a
2	d
3	d
4	a
5	c
6	a
7	d
8	a
9	b
10	c
11	d
12	c
13	c
14	d
15	b
16	d
17	b

Detailed Explanations

Solution to Question 1:

Adenine has an amino (NH_2) group on the 6th carbon.



Solution to Question 2:

Phosphoribosylation of purine nucleosides does not contribute to purine nucleotide biosynthesis.

The three processes that contribute to purine nucleotide biosynthesis in the order of decreasing importance are :

• Synthesis from amphibolic intermediates.

• Phosphoribosylation of purine bases.

• Phosphorylation of purine nucleosides.

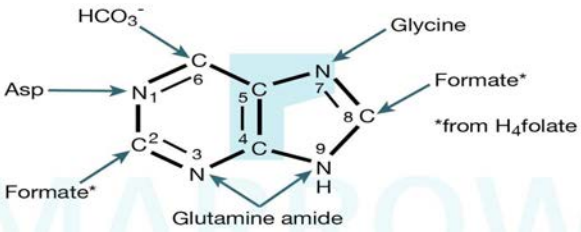
De novo purine nucleotide synthesis doesn't occur in the brain, bone marrow, erythrocytes, and leucocytes.

Solution to Question 3:

Lysine does not contribute to the synthesis of a purine ring.

Glycine, aspartate & glutamine are the amino acids that contribute to synthesizing a purine ring. CO2, N10-Formyl-THFA and N5N10-Methenyl-THFA are the other molecules that influence the structure.

The image below shows the origin of the atoms in the purine ring:



Sources of the atoms in purine ring

N1 :	Derived from NH_2 group of aspartate
C2 and C8 :	From formate group
N3 and N9 :	Amide group of glutamine
C4, CS, N7 :	Glycine amino acid
C6 :	From CO_2

Note: Glycine does not participate in pyrimidine ring synthesis.

Solution to Question 4:

The rate-limiting step of de novo purine synthesis is the conversion of 5-phosphoribosyl-1-pyrophosphate (PRPP) to phosphoribosylamine by PRPP Glutamyl amidotransferase.

The first intermediate formed in the de novo pathway for purine biosynthesis is 5-phosphoribosyl 1-pyrophosphate (PRPP). The rate-limiting enzyme is PRPP Glutamyl amidotransferase. This is the committed step in purine nucleotide biosynthesis.

Note: PRPP Glutamyl amidotransferase is activated by PRPP and glutamine and inhibited by AMP and GMP.

Solution to Question 5:

Inosine monophosphate (IMP) is the parent purine nucleotide made from de novo purine synthesis.

Ribose 5-phosphate is converted to inosine monophosphate (IMP) by series of reactions. The regulatory enzymes of purine synthesis are

• PRPP Synthetase

• PRPP glutamyl amidotransferase

The rate-limiting enzyme is PRPP glutamyl amidotransferase.

Following the synthesis of IMP, separate branches lead to AMP and GMP synthesis.

Solution to Question 6:

Allantoin is the end product of purine catabolism in a patient on rasburicase, a recombinant uricase enzyme.

Uric acid is the natural end product of purine catabolism in humans. So, in conditions with high cell turnover, such as leukemia, lymphomas, and other malignancies, there is increased uric acid production. To prevent hyperuricemia, medications such as rasburicase are administered prophylactically. It is a recombinant uricase enzyme that converts uric acid to a water-soluble end product, allantoin that is easily excreted by the kidneys.

Note: In mammals other than primates, uricase is naturally occurring and allantoin is the end product of purine catabolism.

Solution to Question 7:

Orotic aciduria is not associated with purine degradation.

Orotic aciduria is an autosomal recessive disorder caused by the deficiency of orotidylate decarboxylase or UMP synthase. It is the most common metabolic error in the synthesis of pyrimidines.

Gout, severe combined immunodeficiency syndrome (SCID) and purine nucleoside phosphorylase (PNP) deficiency are associated with purine degradation.

Solution to Question 8:

Uracil cannot be salvaged to its corresponding nucleotide.

Pyrimidine bases cannot be salvaged to their corresponding nucleotides. Purine bases that are salvaged are adenine, hypoxanthine, and guanine.

Salvage pathway - This pathway involves the conversion of purines to nucleosides and later to mononucleotides. It requires less energy than de novo synthesis. It usually occurs in places where de novo synthesis of purines is absent like brain, erythrocytes, etc. The enzymes that catalyze this reaction are :

• Adenine phosphoribosyltransferase (APRT)

• Hypoxanthine-guanine phosphoribosyltransferase (HGPRT).

Solution to Question 9:

The given clinical scenario is highly suggestive of Lesch-Nyhan syndrome. It is caused due to the complete deficiency of Hypoxanthine-guanine phosphoribosyltransferase (HGPRT).

Lesch-Nyhan syndrome is an X-linked recessive disorder. This deficiency results in an inability to salvage hypoxanthine or guanine, from which excessive amounts of uric acid is produced and excreted in urine as orange sands.

Clinical features include compulsive self-mutilation, hyperuricemia, intellectual disability, delayed milestones, dystonic movements and megaloblastic anemia.

Diagnosis is made by deficient HGPRTase enzyme activity in the erythrocyte. Partial deficiency of HGPRTase cause Kelley-Seegmiller syndrome.

Other options:

Option A: Adenine phosphoribosyltransferase enzyme deficiency causes urinary calculus formation with crystalluria.

Option C: Adenosine deaminase enzyme deficiency is one of the 2nd major cause of SCID.

Option D: Purine nucleoside phosphorylase causes a severe deficiency of T cells but B cells are normal.

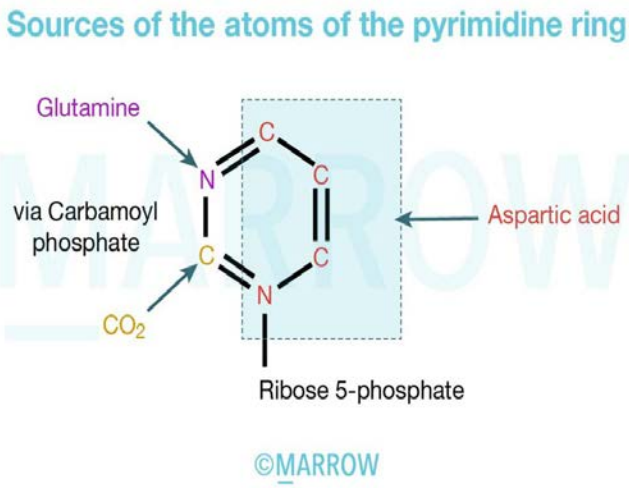
Solution to Question 10:

Glycine is not required for pyrimidine ring synthesis.

The sources of the atoms in the pyrimidine ring are glutamine, CO_2, and aspartic acid. The pyrimidine ring is synthesized before being attached to ribose 5-phosphate, which is donated by PRPP.

Note: Glycine is required in the synthesis of purine ring and not in the pyrimidine synthesis.

The image below shows the origin of the various atoms in the pyrimidine ring:



Solution to Question 11:

The only mitochondrial step in pyrimidine synthesis is catalyzed by the enzyme dihydroorotate dehydrogenase.

Pyrimidine synthesis primarily happens in the liver in both cytoplasm and mitochondria. The conversion of dihydroorotic acid to orotic acid catalyzed by the enzyme dihydroorotate dehydrogenase is the only step that occurs in mitochondria. The rest of the reaction occurs in the cytoplasm.

CAD enzymes are multifunctional enzymes present in a single polypeptide chain. It includes:

- Carbamoyl phosphate synthetase-2
- Aspartate transcarbamoylase
- Dihydroorotase

Solution to Question 12:

In humans, the rate-limiting enzyme for pyrimidine synthesis is carbamoyl phosphate synthetase-II (CPS-II).

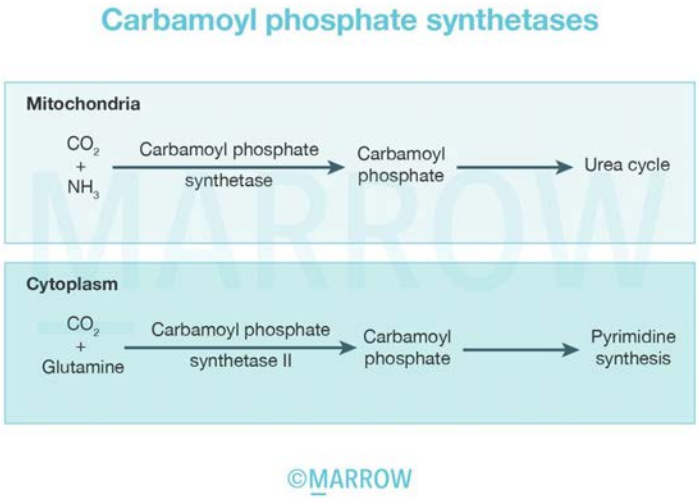
Carbamoyl phosphate synthetase II catalyzes the conversion of glutamine to carbamoyl phosphate. CPS-II is inhibited by UTP and activated by PRPP.

Other options:

Option A: Aspartate Transcarbamoylase is the rate-limiting enzyme for pyrimidine synthesis in bacteria(prokaryote).

Option B: Carbamoyl Phosphate Synthetase I is the rate-limiting enzyme of the urea cycle.

Option D: PRPP glutamyl amidotransferase is the rate-limiting enzyme of De Novo purine synthesis.



Solution to Question 13:

The first true pyrimidine nucleotide formed in pyrimidine synthesis is UMP.

OMP formed from orotic acid by the enzyme orotate phosphoribosyltransferase (OPRTase). OMP is then immediately decarboxylated using orotidylic acid decarboxylase to produce UMP. Nucleotides CTP and TMP are then formed subsequently from UMP via CTP synthetase and thymidylate synthase.

Methotrexate inhibits Dihydrofolate reductase (DHFR) which stops TMP synthesis. 5-Fluorouracil inhibits thymidylate synthase.

OPRTase and orotidylic acid decarboxylase are two enzymes which are considered as a single polypeptide enzyme - UMP synthase. Deficiency of UMP synthase leads to orotic aciduria.

Solution to Question 14:

Uric acid is not a product of pyrimidine catabolism, it is a product of purine catabolism. All other options are products of pyrimidine catabolism.

Unlike the low solubility products of purine catabolism, the catabolism of the pyrimidines forms highly water-soluble products, thus pyrimidine overproduction results in few clinical features.

Cytosine and uracil form β-alanine, CO2, and NH3. Thymine forms β-amino isobutyrate, CO,2 and NH3.

Solution to Question 15:

Hyperuricemia is not seen in xanthinuria.

Xanthinuria occurs due to a defect in the enzyme xanthine oxidase. Xanthine oxidase catalyzes the conversion of hypoxanthine to xanthine and xanthine to uric acid. It is associated with hypouricemia and xanthine lithiasis.

Lesch-Nyhan syndrome is caused by complete deficiency of HGPRTase, whereas Kelley-Seegmiller syndrome is caused by the partial deficiency of HGPRTase. Purines accumulate, causing hyperuricemia.

In von Gierke's disease, the oxidation of glucose-6-phosphate via the pentose phosphate pathway leads to increased production of ribose-5-phosphate which activates the de novo synthesis of purine nucleotides. Overproduction of these nucleotides leads to their degradation, which causes hyperuricemia.

Solution to Question 16:

The given clinical picture is highly suggestive of orotic aciduria. The compound given in the image is orotic acid.

Orotic aciduria is a rare autosomal recessive condition. The deficiency of UMP synthase causes type-1 orotic aciduria. Type-2 orotic aciduria is caused by the deficiency of orotidylate decarboxylase. It manifests usually in the first year of life and presents with growth failure, developmental retardation, megaloblastic anemia and increased urinary excretion of orotic acid.

Severe megaloblastic anemia with normal serum B12 and folic acid levels suggests this condition. A presumptive diagnosis is made by checking increased urinary excretion of orotic acid. Confirmation is done by enzyme assays of transferase and decarboxylase of erythrocytes. The anemia only responds to the administration of uridine. Allopurinol and 6-azauridine may precipitate this condition.

Other options:

Option A and B: In gout and Lesch Nyhan syndrome uric acid crystals are excreted.

Option C: In xanthinuria, xanthine crystals are excreted.

Solution to Question 17:

The above given clinical findings is suggestive of xeroderma pigmentosum. Defect in the removal of oligonucleotides containing the damaged bases by endonucleases is seen in xeroderma pigmentosum.

Xeroderma Pigmentosum (XP) is a rare autosomal recessive genetic disorder characterized by defective DNA repair which leads to clinical and cellular hypersensitivity to ultraviolet radiation and other carcinogenic agents.

Important clinical features are:

Intense cutaneous photosensitivity, xerosis, poikiloderma, actinic keratosis, acute burning under minimal sun exposure, erythemas, hyperpigmented lentiginous macules, and malignant lesions in sun-exposed areas, including basocellular carcinoma, squamous cell carcinoma, and melanoma.

The final diagnosis of XP can be confirmed by special laboratory tests by examining the DNA damage repair in cells from cultures exposed to ultraviolet radiation. The most common tests are skin biopsy and culture of skin fibroblasts.

There is no cure for XP. Treatment consists of minimizing exposure to sunlight and regular dermatological care, as well as surgery for recurrent tumors excision. Patients should be followed up every 3 months. Genetic counseling is important in a family that has an affected child and is considering having more children.

DNA organization, replication and repair

Question 1:

Which of the following is true about DNA?

a) Each nucleotide pair includes two purines.

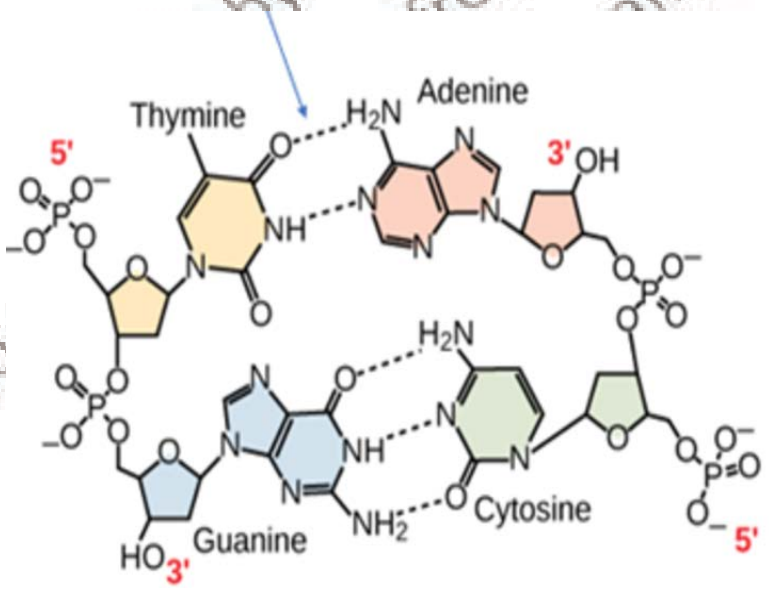
b) The information from DNA is copied in the form of tRNA.

c) The nucleotide of one strand binds with the nucleotide of opposite strand.

d) Cytosine and uracil differ by one ribose sugar.

Question 2:

Identify the marked bond:



a) Ionic bond

b) Covalent bond

c) Hydrogen bond

d) Phosphodiester Bond

Question 3:

What is the percentage of cytosine in DNA if the percentage of thymine residues is 28%?

a) 22

b) 28

c) 36

d) 44

Question 4:

Which of the following is true about the types of DNA?

a) Z-DNA is a right handed double helix.

b) B-DNA is found in conditions of high salt concentration and low humidity.

c) A-DNA is found in high humidity and low salt concentration.

d) B-DNA is the physiologically most common form

Question 5:

Which of the following is a feature of denatured DNA?

a) Breaking of phosphodiester bond

b) Alteration of primary structure

c) Increase in viscosity

d) Hyperchromicity at 260 nm

Question 6:

Which of the following is not a histone found in nucleosome?

a) H1

b) H2A

c) H2B

d) H3

Question 7:

What is the role of the (H3–H4)2 tetramer in the nucleosome?

a) Central role in the formation of the nucleosome

b) Stabilizes the primary particle

c) Firmly binds two additional half-turns of DNA

d) Stabilize the 30-nm chromatin fiber

Question 8:

Which among the following statements is incorrect?

a) Acetylation of H3 and H4 is associated with the activation of gene transcription

b) Acetylation of core histones is associated with chromosomal assembly during DNA replication

c) ADP-ribosylation of histones is associated with DNA repair

d) De-phosphorylation of histone H1 is associated with chromosome condensation during replication

Question 9:

Which of the following statements is true regarding heterochromatin?

a) It is transcriptionally active

b) It is less densely packed

c) It stains densely

d) It is replicated earlier than euchromatin

Question 10:

Which of the following is true about DNA organization in chromosomes?

a) Centromere is an adenine-thymine (A–T)-rich region containing repeated DNA sequences

b) Telomeres consist of short TTAGGG-rich repeats

c) Each sister chromatid contains one dsDNA molecule

d) All of the above

Question 11:

What percentage of cellular DNA is in mitochondria?

a) 1%

b) 10%

c) 0.1%

d) 20%

Question 12:

True regarding mitochondrial DNA is:

a) One set from each parents

b) It has a higher mutation rate than nuclear DNA

c) It codes for more than 25% of oxidative phosphorylation proteins

d) It has 3 X 10^9 base pairs

Question 13:

Klenow fragment lacks the activity of

a) 3'° 5' Exonuclease

b) 5'° 3' Exonuclease

c) 5'° 3' DNA polymerase

d) 3'° 5' DNA polymerase

Question 14:

DNA synthesis occurs in which phase?

a) G1 Phase

b) G2 Phase

c) S Phase

d) Go Phase

Question 15:

Which of the following is not a feature of DNA replication?

a) Semi-discontinous

b) Template independent

c) Bidirectional

d) RNA primer is required.

Question 16:

Which of the following is true about DNA replication?

a) Replication can occur only from a single stranded DNA template

b) Replication can occur only from a double stranded DNA template

c) Deoxynucleotide polymerization is catalyzed by helicases

d) Only DNA polymerase functions at the replication fork

Question 17:

Which of the following initiates local denaturation and unwinding of DNA during replication?

a) Helicase

b) Topoisomerase

c) Single strand binding protein

d) Ori-binding protein

Question 18:

Which of the following is the function of the replication protein-A during eukaryotic DNA replication?

a) Unwinds DNA template at replication fork

b) Create a single- or double-stranded break in the helix to add or remove supercoils

c) Prevent strands from reannealing

d) Catalyzes the formation of a phosphodiester bond within a strand of double-stranded DNA

Question 19:

Which of the following is synthesized by DNA primase during replication?

a) DNA primer

b) RNA primer

c) Okazaki fragments

d) Leading strand

Question 20:

Which of the following is true about DNA polymerase I?

a) It is involved in the creation of okazaki fragments.

b) It is found in prokaryotes.

c) It synthesises RNA primers.

d) It is the primary enzyme of DNA synthesis.

Question 21:

Which of the following is not a function of the DNA polymerase complex?

a) Chain elongation

b) Processing

c) Proofreading

d) Unwinding of DNA

Question 22:

Which of the following is two-base alteration DNA damage?

a) Oxidative free-radical formation

b) Alkylation of base

c) Base-analog incorporation

d) Bifunctional alkylating agent cross-linkage

Question 23:

A patient who has been diagnosed with multiple gastrointestinal polyps and carcinoma, with a positive family history/history of hereditary non-polyposis colon carcinoma has a defect in which of the following?

a) Mismatch repair

b) Nucleotide excision repair

c) Base excision repair

d) Point mutation

Question 24:

A 10-year-old girl is brought to the dermatologist by her parents. She has many freckles on her face and limbs as shown below. She is also unusually sensitive to sunlight. Early changes of squamous cell carcinomas are also noted on her face. Which of the following processes is most like to be defective in this patient?

a) Defective DNA nucleotide excision repair

b) Defective DNA base excision repair

c) Defective DNA mismatch repair

d) Defective homologous repair

Question 25:

What is the defective DNA repair mechanism in a patient with SCID?

a) Mismatch repair

b) Nucleotide excision repair

c) Non-homologous end joining repair

d) Homologous recombination

Question 26:

By which mechanism does gene repair occur when mediated by CRISPR-Cas9?

a) Base excision

b) Non-homologous end joining

c) Homologous end joining

d) Mismatch repair

Question 27:

Chimeric DNA is associated with:

a) Paternity test

b) Maternity test

c) Personal identification

d) Organ transplantation

Question 28:

Which of the following is not a true statement regarding telomerase?

a) Has reverse transcriptase activity

b) Present only in eukaryotes

c) Maintain the chromosome length

d) Involved in DNA repair

Answer Key

Question No.	Correct Option
1	c
2	c
3	a
4	d

5	d
6	a
7	a
8	d
9	c
10	d
11	a
12	b
13	b
14	c
15	b
16	a
17	d
18	c
19	b
20	b
21	d
22	d
23	a
24	a
25	c
26	b
27	d
28	d

Detailed Explanations

Solution to Question 1:

The true statement about DNA is that the nucleotide of one strand binds with the nucleotide of the opposite strand.

DNA (deoxyribonucleic acid) is a polymeric molcule containing genetic information. It is a double-stranded molecule consisting of nucleotide pairs. Each nucleotide pair includes one purine and one pyrimidine. The nucleotide of one strand binds with the nucleotide of the opposite strand by hydrogen bonds.

Other options:

Option B: The information from DNA is copied in the form of mRNA.

Option D: Cytosine and uracil differ by one amino group.

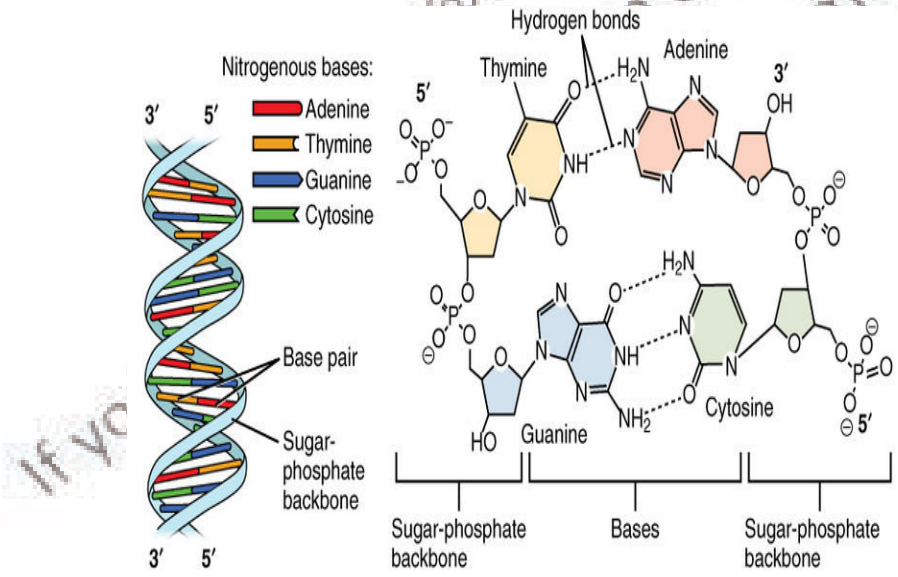
Solution to Question 2:

The marked bond represents a hydrogen bond between thymine and adenine.

The complementary nitrogenous bases of the 2 strands of the DNA double helix are linked by hydrogen bonds. Adenine and thymine are linked by two hydrogen bonds (A=T) whereas three hydrogen bonds are present between cytidine and guanine (C≡G). Van der Waals and hydrophobic interactions are present between the stacked adjacent base pairs.

A β-N-glycosidic bond is present between the nitrogenous base and deoxyribose sugar. C1 of deoxyribose/ribose sugar is joined covalently to N1 of pyrimidines and N9 of purines by β-N-glycosidic bond. An ester bond is present between the sugar and phosphate group of the same nucleotide. An acid anhydride bond is present between two phosphate groups.

A polymer of nucleotides is joined by 3'-5' phosphodiester bond. The bond is present between the sugar of one nucleotide and the phosphate group of another.



Solution to Question 3:

The percentage of cytosine in DNA will be 22%.

According to Chargaff's rule in cellular DNA, regardless of species,

A = T and G = C

Purines = Pyrimidines i.e. A+G = C+T.

In the question it is given that thymine is 28%, therefore adenine is also 28%. So, adenine + thymine = 56% and guanine + cytosine = 44%. Since guanine = cytosine, the percentage of cytosine = 44/2 = 22%.

Solution to Question 4:

The correct statement is that B-DNA is the physiologically most common form.

There are 6 types of DNA- A, B, C, D, E and Z. All are right-handed except Z-DNA. Z-DNA is a left-handed double helix in 5' end of chromosome and has phosphodiester backbone in a zigzag form. A-DNA is found in conditions with low humidity and high salt concentration. B-DNA is the physiologically most common form. It is found in conditions with high humidity and low salt concentration.

Solution to Question 5:

Hyperchromicity at 260nm is a feature of denatured DNA.

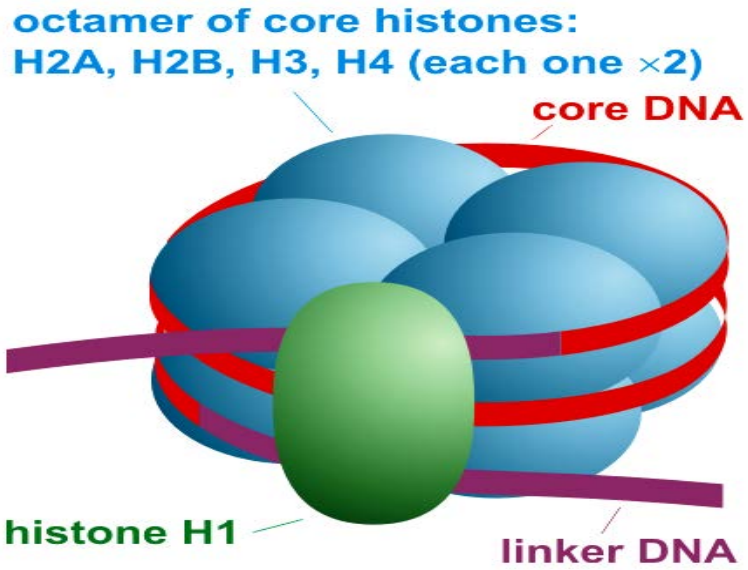
Denaturation or melting of DNA is a process that separates the two strands into component strands. During denaturation, the hydrogen bonds are broken. The phosphodiester bond is not broken. DNA's secondary and teriary structures are also broken, butthe primary structure is not altered. As DNA gets denatured, its viscosity decreases. Denatured DNA also shows hyperchromicity at 260nm, which is the increase in absorbance of UV light showed by nitrogenous bases.

Solution to Question 6:

H1 is not a histone found in nucleosomes.

Chromatin is formed by nucleosomes, which are composed of DNA wound around an octameric complex of histone molecules. They contain four major types of histones: H2A, H2B, H3, and H4. Histones are a small family of closely related basic protein composed of arginine and lysine. H3 and H4 form a tetramer while H2A and H2B form dimers; these histone oligomers associate to form the histone octamer. These four core histones are subject to at least six types of covalent modification or post-translational modifications - acetylation, methylation, phosphorylation, ADP-ribosylation, monoubiquitylation and sumoylation.

H1 histone is seen in the linker region between nucleosomes and is bound loosely to the nucleosome.



Solution to Question 7:

The (H3–H4)2 tetramer has a central role in the formation of the nucleosome.

Type	Function
H1 histones	Stabilize the 30-nm chromatin fiber
H2 histones	H2A–H2B dimers stabilize the primary particle and firmly bind two additional half-turns of DNA
H3 histones	(H3–H4)2tetramer has a central role in the formation of the nucleosome. It confers nucleosome-like properties on DNA.
H4 histones	

Solution to Question 8:

Roles of modified histones:

• Phosphorylation of histone H1 is associated with the condensation of chromosomes during the replication cycle.

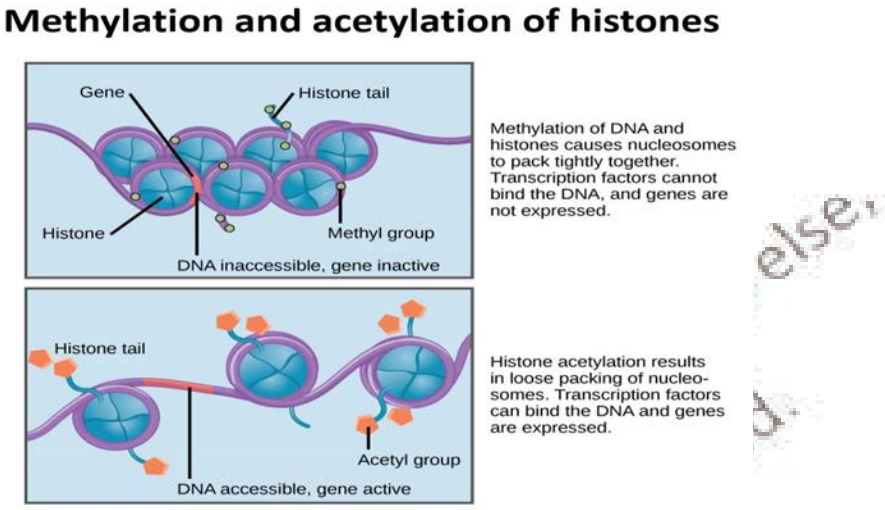
• Acetylation of H3 and H4 is associated with the activation or inactivation of gene transcription.

• Acetylation of core histones is associated with chromosomal assembly during DNA replication.

• Methylation of histones is correlated with activation and repression of gene transcription

• ADP-ribosylation of histones is associated with DNA repair.

• Monoubiquitylation is associated with gene activation, repression, and heterochromatic gene silencing.



Solution to Question 9:

The true statement is that heterochromatin stains densely.

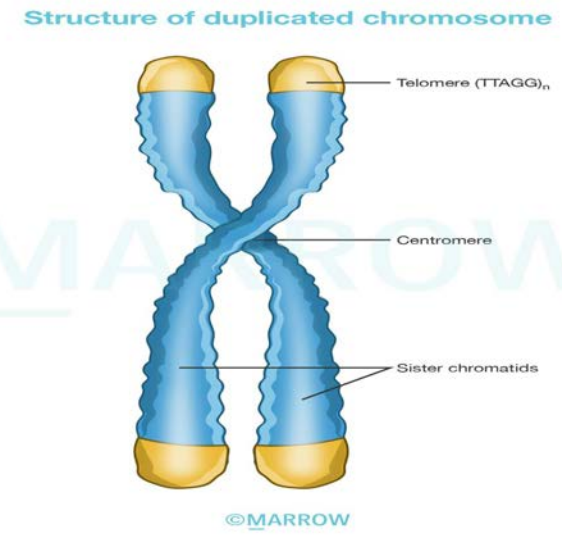
Differences between heterochromatin and euchromatin:

Heterochromatin	Euchromatin
Transcriptionally inactive	Transcriptionally active
Densely packed	Less densely packed
Chromatin stains densely	Chromatin stains less densely
The chromatin in the region i s high in meC content (methy lation of deoxycytidine residu es decreases gene transcripti on)	Generallyreplicated earlierth an heterochromatin
Histone deacetylation increas es heterochromatin	Histone acetylation increases euchromatin

Solution to Question 10:

All of the given statements are correct.

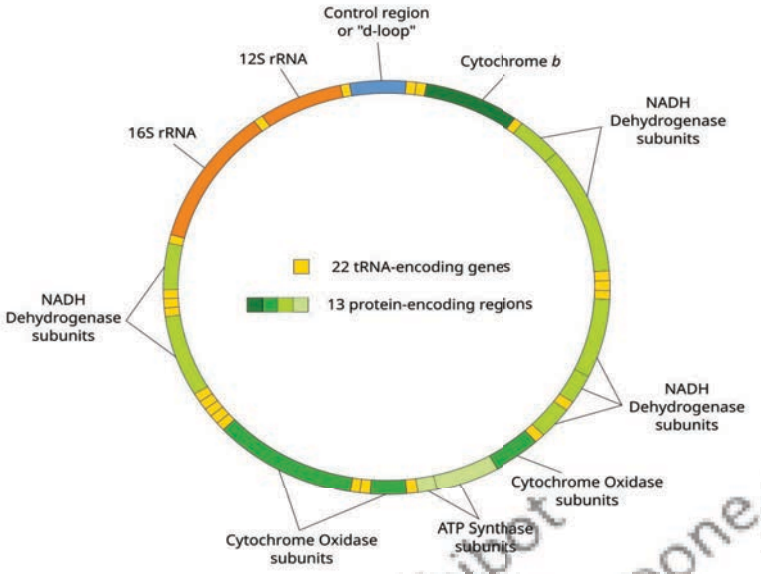
Chomosomes can be visualised during metaphase of mitosis. At this point, they are duplicated with two-fold symmetry. The duplicated, identical sister chromatids are connected at the centromere. Each sister chromatid consists of a single dsDNA moleule. The centromere is an adenine-thymine (A–T) - rich region containing repeated DNA sequence. The ends of each chromosome contain structures called telomeres, which have a variable number of repeats of the sequence 5'-TTAGGG-3'. Telomere shortening is associated with aging and malignant transformation.



Solution to Question 11:

1% of cellular DNA is in mitochondria.

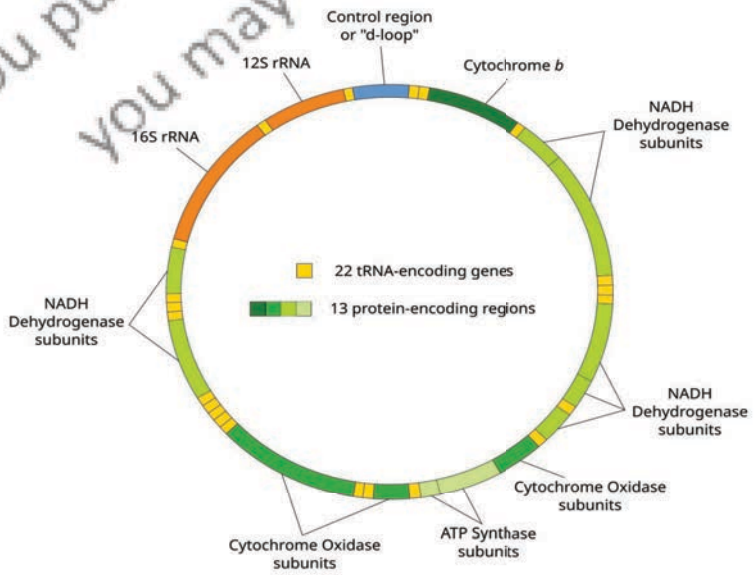
Human mitochondria contains 2 to 10 copies of a small circular ~16 kbp (kilo base pair) dsDNA molecule that makes up approximately 1% of total cellular DNA. Out of 37 genes in human mitochondrial DNA (mtDNA), 22 genes codes for transfer RNAs, 2 for ribosomal RNAs, and 13 for proteins that play key roles in the respiratory chain. A feature unique to human mitochondrial mtDNA is maternal non-mendelian inheritance because all mitochondria are contributed by the ovum during zygote formation. Thus, mothers transmit mtDNA to all their offspring.



Solution to Question 12:

The true statement is that mitochondrial DNA has a high mutation rate.

Mitochondrial DNA is circular and double-stranded. It has 16,569 bp (that makes up approximately 1% of total cellular DNA) while nuclear DNA has 3.3 billion bp. It has 37 genes that encode 13 protein subunits of the respiratory chain (out of a total of 67 proteins) i.e. about 20%. It mutates at a rate 5-10 times that of nuclear DNA. Mitochondrial DNA is transmitted by maternal non-mendelian inheritance.



Solution to Question 13:

Klenow fragment lacks 5' ° 3' exonuclease activity.

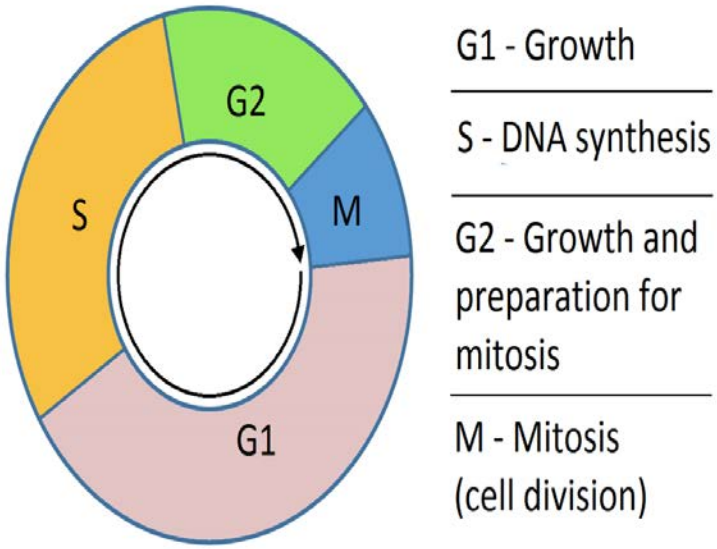
DNA polymerase I is an enzyme involved in DNA replication. It has polymerase activity along with both 3' ° 5' and 5' ° 3' exonuclease activity. When the 5' ° 3' exonuclease domain is removed, the remaining fragment is called Klenow fragment. It retains the polymerization and proofreading activities of DNA polymerase I.

Solution to Question 14:

DNA synthesis occurs in the S phase.

In the cell cycle, replication of the DNA genome occurs only at a specified time during the synthetic or S phase. During the S phase, mammalian cells contain greater quantities of DNA polymerase than during the nonsynthetic phases of the cell cycle.

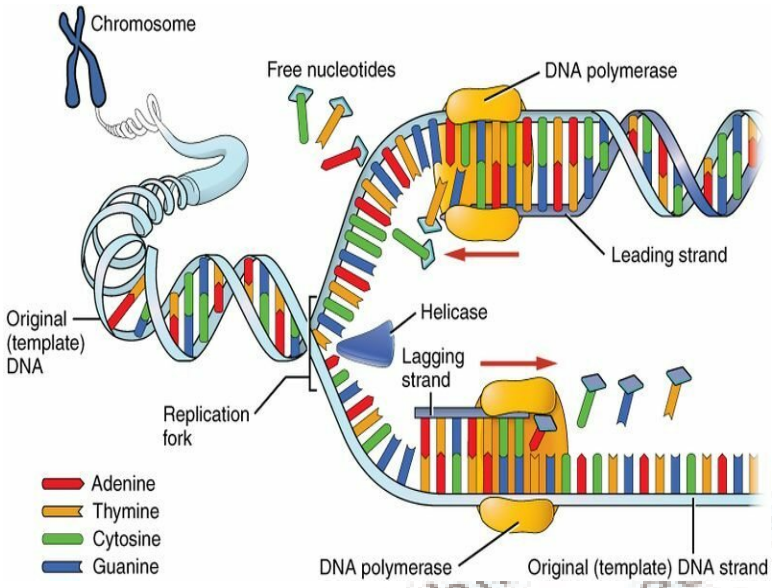
Gap 1 (G1) phase consists of cell growth and preparation for DNA synthesis. It occurs before the S phase. Gap 2 (G2) phase occurs after S phase, during which preparation for mitosis (M phase) occurs. Cyclins tightly regulate the transition between phases of the cell cycle.



Solution to Question 15:

DNA replication is not template independent.

DNA replication occurs in S-phase of cell cycle. It is semi-conservative, semi-discontinuous and bidirectional. DNA strands separate and each acts as a template strand on which new complementary strand is synthesized. RNA primer is required. The new strand is synthesized always in 5'-3' direction and thus, the synthesis of DNA in both strands is not the same.



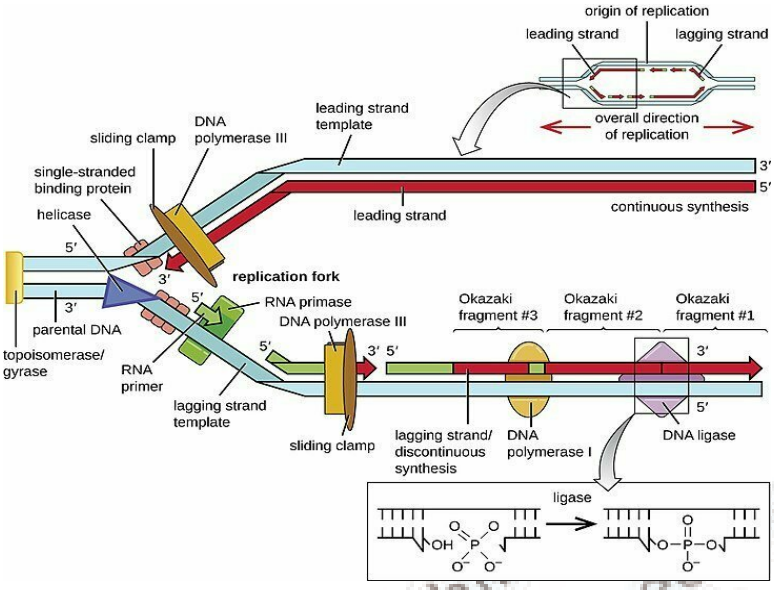
Solution to Question 16:

In all cells, replication can occur only from a single-stranded DNA (ssDNA) template.

Steps of DNA replication:

- Identification of the origins of replication
- ATP hydrolysis-driven unwinding of dsDNA to provide an ssDNA template
- Formation of the replication fork; synthesis of RNA primer
- Initiation of DNA synthesis and elongation
- Formation of replication bubbles with ligation of the newly synthesized DNA segments
- Reconstitution of chromatin structure

The replication fork is formed when DNA helicase unwinds a short segment of the parent dsDNA. Primase initiates the synthesis of an RNA primer. DNA polymerase then initiates DNA synthesis (deoxynucleotide polymerisation) in the 3' to 5' direction. Hence, helicase, primase and polymerases act at the replication fork.



Solution to Question 17:

Ori-binding protein initiates local denaturation and unwinding of DNA during replication.

Splitting of the two DNA strands during replication is initiated when the ori-binding protein (eg: DnaA protein) binds to the origin of replication (ori). This leads to positive supercoiling of the ori. The adjacent A+T rich region, known as DNA unwinding element (DUE), undergoes local denaturation and separation. A short region of ssDNA is formed, to which helicases bind and further unwind DNA.

SSB or replication protein-A prevents reannealing of the ssDNA strands generated by helicases.

Topoisomerase stabilizes the strain caused during unwinding.

Solution to Question 18:

Replication protein-A prevents strands from reannealing during the process of DNA replication.

Replication protein-A is a type of ssDNA-binding protein (SSB). During DNA replication, it binds to the ssDNA generated by helicases. It shifts the equilibrium between dsDNA and ssDNA in the direction of ssDNA and thus preve ts reannealing of strands. It also protects the ssDNA from degradation by nucleases.

Option A: Helicase unwinds the DNA template at the replication fork.

Option B: DNA topoisomerases create a single- or double-stranded break in the helix to add or remove supercoils.

Option D: DNA ligase catalyzes the formation of a phosphodiester bond within a strand of double-stranded DNA.

Solution to Question 19:

DNA primase synthesizes RNA primers during replication of DNA.

During DNA replication, DNA is synthesized using an ssDNA template. This is done by DNA polymerases in the 5' to 3' direction. However, DNA polymerases require an RNA primer, which is a short (10 - 200bp) double stranded region consisting of RNA paired to the DNA templete. Thus, RNA primers serve as scaffolds to which deoxyribonucleotides are added. The RNA in the primers is later replaced by DNA.

Other options:

Option C: Okazaki fragments are lagging strands built on RNA primers by DNA polymerase δ.

Option D: Leading strands are synthesized by DNA polymerase ε.

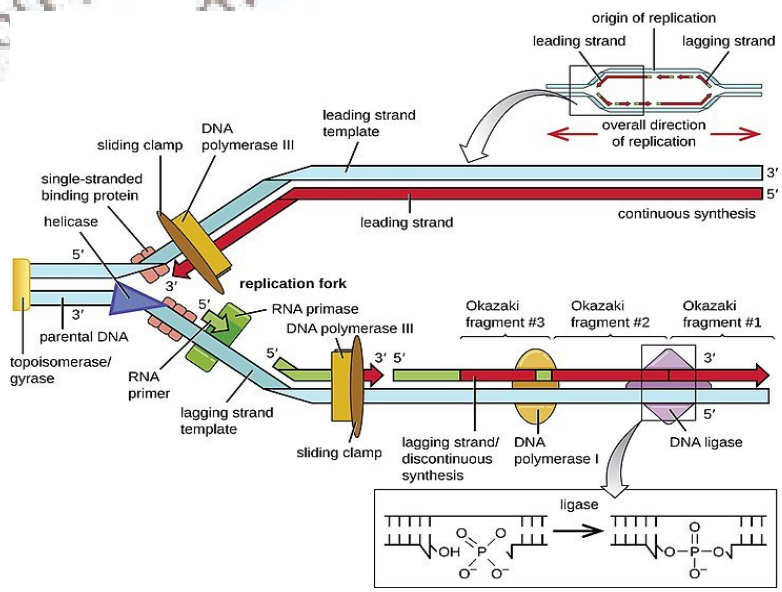
Solution to Question 20:

The true statement is that DNA polymerase I (pol I) is found in prokaryotes.

The prokaryotic polymerases are DNA polymerases I, II, and III. DNA polymerase I recognize a “nick” or break in the phosphate backbone, and then removes each RNA primer and fills the gaps with DNA. It does this through its 5'° 3' exonuclease activity. It has DNA repair activity through 3'° 5' exonuclease activity. DNA polymerase II carries out DNA proofreading and repair. DNA polymerase III is the primary enzyme for the synthesis of the leading strand of DNA.

Option A: DNA polymerase δ (eukaryotic polymerase) synthesizes Okazaki fragments which are short, newly synthesized DNA fragments that are formed on the lagging template strand during DNA replication. They are complementary to the lagging template strand.

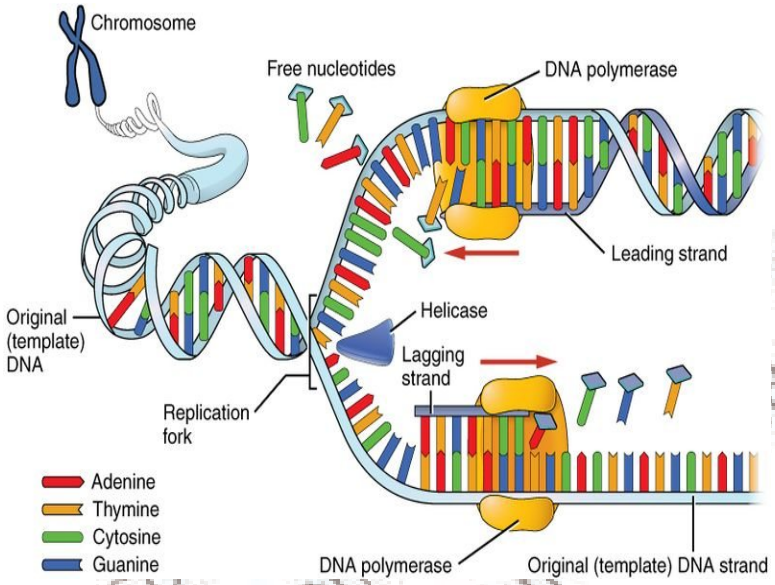
Option C: Primase synthesizes RNA primers to start the replication.



Solution to Question 21:

Unwinding of DNA is not a function of the DNA polymerase complex.

A number of different DNA polymerase molecules engage in DNA replication that share three important properties: Chain elongation, processivity, and proofreading. Unwinding of DNA is done by enzyme helicase.



Solution to Question 22:

Bifunctional alkylating agent cross-linkage is a type of two-base alteration DNA damage.

Bifunctional alkylating agents, like cyclophosphamide and chlorambucil, are commonly used anti-cancer drugs. They contain two reactive sites and thus, can cause intrastrand and interstrand cross-links between two nucleotides.

Solution to Question 23:

Mismatch repair will be defective in a patient with multiple gastrointestinal polyps and carcinoma, with a positive family history/history of hereditary non-polyposis colon carcinoma (HNPCC)

Mismatch repair is a DNA repair mechanism that corrects single-nucleotide mismatches, insertions, and deletions introduced by the DNA polymerase during DNA replication.

HNPCC or Lynch syndrome is an autosomal dominant genetic condition. It occurs due to defects in DNA mismatch repair genes like MLH1 and MSH2. The genomes of these patients show microsatellite instability. HNPCC is associated with a high risk of colon cancer. It may also be associated with a risk of developing carcinomas of the endometrium, stomach, small bowel, ovary, pancreas, and transitional cell carcinoma of the ureter and renal pelvis.

The diagnostic criteria for HNPCC are referred to as the Amsterdam criteria or the 3-2-1-0 rule. It includes:

• Three or more relatives with HNPCC-associated cancer (colorectal cancer, endometrial cancer, cancer of the small bowel, ureter, or renal pelvis), one of whom is a first-degree relative of the other two.

• At least two successive generations affected

• At least one case diagnosed before age 50 years

• Familial adenomatous polyposis excluded

• Tumors verified by pathologic examination.

Other options:

Option B: Patients with xeroderma pigmentosum have a defect in the nucleotide excision repair pathway.

Option C: Defect in base excision repair pathway is seen in patients with MUTYH-associated polyposis.

Option D: Sickle cell disease and beta-thalassemia are examples of diseases seen in patients with point mutations.

Solution to Question 24:

Photosensitivity, freckling and skin cancer on the face in a child aare suggestive of xeroderma pigmentosum. It is an example of defective DNA nucleotide excision repair.

In xeroderma pigmentosum, there is an inability to repair DNA pyrimidine dimers caused by UV exposure. It is an autosomal recessive disease characterized by exaggerated sunburns and pigmentary changes on exposure to UV radiations. There is increased risk of developing non-melanoma and melanoma skin cancers, and neurodegeneration. They are also more prone to developing smoking-induced lung cancers.

Diagnosis is mainly clinical and it can be confirmed definitively by cellular tests for defective DNA repair. Management includes sun avoidance and regular follow-up to assess and treat any skin cancers. There is no cure for xeroderma pigmentosum.

Solution to Question 25:

SCID is associated with a defective non-homologous end-joining (NHEJ).

NHEJ is a predominant pathway for DNA double-stranded break repair. NHEJ also functions in class switch recombination, enhancing T and B cell diversity. Thus, NHEJ deficiency leads to combined immunodeficiency due to a failure to carry out recombination efficiently.

Severe combined immunodeficiency (SCID) is characterised by defects in both humoral and cell-mediated immune responses. Genes involved are JAK3, RAG1, and IL-7R. Persons with SCID are extremely susceptible to recurrent, severe infections by a wide range of

pathogens, including Candida albicans, Pneumocystis jirovecii, Pseudomonas species, cytomegalovirus, varicella, and a host of other bacteria.

X-linked SCID is the most common form, accounting for 50% to 60% of cases, is X-linked, and hence, SCID is more common in boys. The genetic defect is a mutation in the common γ-chain (γc) subunit of cytokine receptors. In these patients, the thymus contains lobules of undifferentiated epithelial cells, resembling fetal thymus.

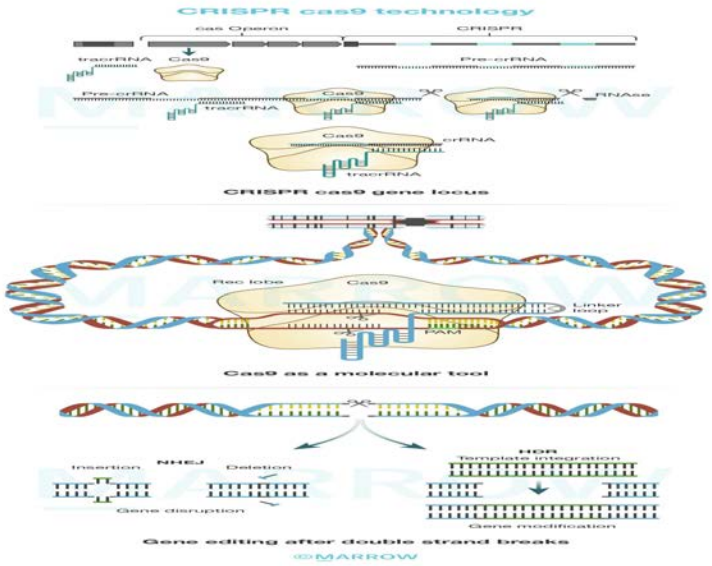
Autosomal recessive SCID is commonly caused by a deficiency of adenosine deaminase (ADA) enzyme. In these patients, the thymus contains remnants of Hassall's corpuscles.

Solution to Question 26:

Non-homologous end joining (NHEJ) is the major mechanism used by CRISPR-Cas9 for gene repair.

The CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats) are clustered palindromic sequences found in many bacteria. It is a form of acquired immunity to infection by bacteriophages which complements the system of restriction endonucleases and methylases. CRISPR uses RNA-based targeting to bring the Cas9 endonuclease to foreign (or any complementary) DNA. Within bacteria this CRISPR-RNA-Cas9 complex then degrades and inactivates the targeted DNA.

The CRISPR system has been adapted for use in eukaryotic cells for gene deletion, gene editing and even modulation of gene transcription.



Solution to Question 27:

Chimeric DNA is associated with organ transplantation, especially bone-marrow transplantation.

In organ transplantation, chimeric DNA refers to the mixture of donor and recipient genetic material that occurs when donor cells begin functioning within the recipient's body. It is used for long-term monitoring of bone marrow transplant recipients. Detecting changes in the levels of donor-derived cells can provide early warning signs of graft failure, rejection, or relapse of underlying disease.

Note: The practical applications of recombinant DNA technology include:

• Gene mapping using techniques such as fluorescence in situ hybridization (FISH).

• Producing proteins such as:

• Interferons, tPA (tissue plasminogen activator)

• Insulin, growth hormone

• Hepatitis B vaccine

• Molecular analysis of disease, to detect gene variations and mutations.

• Restriction Fragment Length polymorphism (RFLP) and single nucleotide polymorphism (SNP)

• Microsatellite DNA polymorphisms

• Gene therapy and bone marrow transplantation

Solution to Question 28:

The false statement is that telomerase is involved in DNA repair.

Telomeres are the end of chromosomal DNA. Since prokaryotic DNA is circular, it does not have free ends and hence no telomeres. Thus, telomeres are present only in eukaryotes Telomeres have TG- rich repeats and usually do not code for any proteins. With every DNA replication during cell division, telomere shortening occurs. Once telomeres are almost negligible or non-existent in length, the DNA is unable to replicate and the cell dies. Hence, there is a limit to the number of times a cell can divide.

In stem cells and cancer cells, an enzyme telomerase having reverse transcriptase activity (RNA dependent DNA polymerase) adds telomeres to the chromosome ends, thereby preserving chromosomal length. It allows the stem cells and cancer cells to multiply a lot more number of times than a normal adult cell.

RNA synthesis, processing and modification

Question 1:

Which among the following is a protein-coding RNA?

a) mRNA

b) tRNA

c) rRNA

d) snRNA

Question 2:

Which of the following RNA is not directly involved in protein synthesis?

a) mRNA

b) rRNA

c) tRNA

d) miRNA

Question 3:

Which of the following is true?

a) Primer is involved in RNA synthesis

b) Highly active efficient proofreading function in RNA transcription

c) Deoxyribonucleotides are used in RNA synthesis

d) U replaces T as the complementary base for A in RNA

Question 4:

Which enzyme is responsible for the polymerization of ribonucleotides into a sequence complementary to the template strand of the gene?

a) RNA-dependent RNA polymerase

b) DNA-dependent RNA convertase

c) RNA-dependent replicase

d) DNA-dependent RNA polymerase

Question 5:

Which enzyme prevents the formation of superhelical tensions in the DNA during transcription?

a) Topoisomerase

b) RNA polymerase

c) Pyrophosphatases

d) Unwindases

Question 6:

Which of the following is not a function of RNA polymerase II?

a) Synthesis of mRNA

b) Synthesis of rRNA

c) Synthesis of miRNA

d) Synthesis of snRNA

Question 7:

The role of the sigma factor in bacterial RNA polymerase is to _____.

a) Catalyse RNA synthesis

b) Position RNA polymerase correctly on the template DNA

c) Terminate RNA synthesis

d) Unwind the DNA template

Question 8:

During a field trip, an 11-year-old boy consumes a wild mushroom which was later identified to be a death cap mushroom. Which of the following enzymes is inhibited by the toxin from this mushroom?

a) DNA polymerase

b) RNA polymerase

c) Topoisomerase

d) Helicase

Question 9:

Which enzyme gives signals for termination of RNA transcription?

a) RNA Polymerase I

b) RNA Polymerase II

c) RNA Polymerase III

d) Topoisomerase

Question 10:

RNA produced from a fragment of DNA has the sequence of UAAGGC. The sequence of the non-template strand in the DNA that gave rise to this sequence is which one of the following?

a) CGGAAT

b) UAAGGC

c) TAAGGC

d) ATTCCG

Question 11:

What is a primary transcript?

a) RNA product synthesized in the 5′-3′ direction

b) RNA product synthesized in the 3′-5′ direction

c) DNA product synthesized in the 3′-5′ direction

d) DNA product synthesized in the 5′-3′ direction

Question 12:

7-methyl-guanosine triphosphate capping of the primary transcript is necessary for _____.

a) Processing of the primary transcript to mRNA

b) Translation of the mRNA

c) Protection of the mRNA against nucleolytic attack by 5′-exonucleases

d) All of the above

Question 13:

The attachment of Eukaryotic mRNA to the ribosome is mediated through _____

a) Poly A tail

b) tRNA

c) Guanyl Cap

d) Shine-Dalgarno Sequence

Question 14:

After synthesis, eukaryotic mRNA undergoes extensive modifications including which of the following?

a) Capping

b) Polyadenylation

c) Splicing

d) All of the above

Question 15:

Which of the following does not require 5’ capping?

a) tRNA of Alanine

b) U6 snRNA

c) mRNA for Histone

d) siRNA

Question 16:

Which of the following statements about mRNA splicing is true?

a) The existence of split genes has no advantage

b) Self-splicing introns do not require the help of any protein for splicing to occur accurately

c) β-thalassaemia results from a genetic defect in the spliceosome

d) Splicing occurs in the cytosol

Question 17:

Which of the following does not happen in 5' to 3' direction?

a) Transcription

b) DNA replication

c) DNA repair

d) RNA editing

Question 18:

The eukaryotic initiation factor (eIF) that helps in cap binding is?

a) eIF 1

b) eIF 2

c) eIF 3

d) eIF 4

Question 19:

Which statement best explains the function of eEF-2?

a) It is required for the translocation of peptidyl-tRNA during translation

b) It is required for the initiation of protein synthesis

c) It is the agent that binds to, and is inactivated by, chloramphenicol

d) It functions as a peptidyl transferase

Question 20:

All of the following statements about stop codons and amino acids are false except:

a) Selenocysteine is coded by UAA

b) Pyrrolysine is coded by UAG

c) Selenocysteine is considered the 22nd amino acid

d) Other name of UGA is ochre

Question 21:

The process of apolipoprotein B48 synthesis involves which of the following?

a) RNA alternate splicing

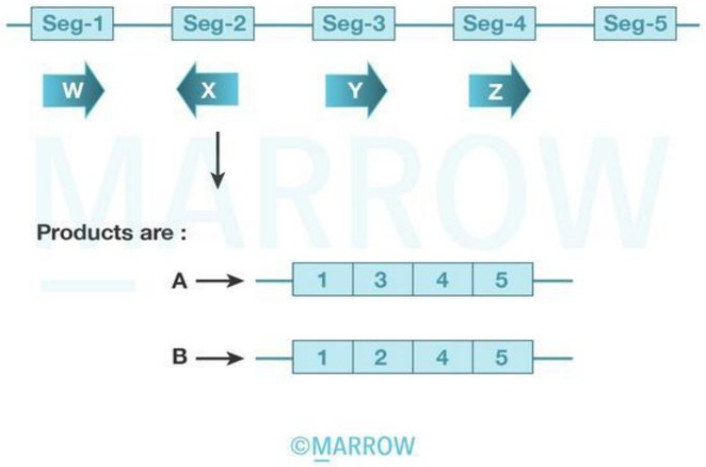
b) RNA editing

c) DNA editing

d) RNA interference

Question 22:

The illustration below shows alternative splicing of a gene which has intron segments 1, 2, 3, 4, and 5. It yields two products A and B, by the action of different promoters. You are provided with the promoters w, x, y, and z, each of which binds to a different segment as shown below and acts in the marked direction. Which of the following promoters will you use to get product A?



a) W,X,Y

b) W,Z

c) W,X

d) W, Y

Answer Key

Question No.	Correct Option
1	a
2	d
3	d
4	d
5	a
6	b
7	b
8	b
9	b
10	c
11	a
12	d
13	c
14	d
15	a
16	b
17	d
18	d
19	a
20	b
21	b
22	d

Detailed Explanations

Solution to Question 1:

mRNA is a protein-coding RNA.

The mRNA convey genetic information from DNA to the ribosome, where they specify the amino acid sequence of the protein products of gene expression.

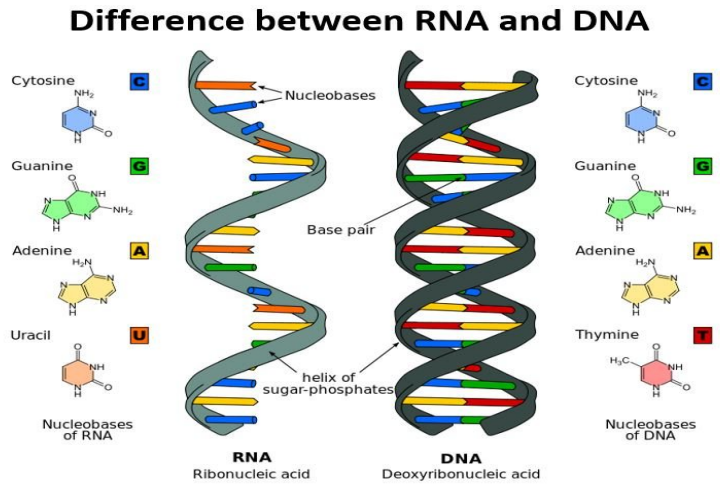
Solution to Question 2:

miRNA is not directly involved in protein synthesis. miRNA participates in the modulation of gene expression by altering mRNA function.

The mRNAs, rRNAs, and tRNAs are directly involved in protein synthesis. Other RNAs participate in either mRNA splicing (snRNAs) or modulation of gene expression by altering mRNA function (mi/siRNAs) and/or expression (lncRNAs).

Solution to Question 3:

Uracil (U) replaces thymine (T) as the complementary base for adenine (A) in RNA.



Solution to Question 4:

DNA-dependent RNA polymerase is the enzyme responsible for the polymerization of ribonucleotides into a sequence complementary to the template strand of the gene.

The enzyme attaches at a specific site, the promoter, on the template strand. This is followed by the initiation of RNA synthesis at the starting point, and the process continues until a termination sequence is reached.

Solution to Question 5:

Topoisomerase prevents the formation of superhelical tensions in the DNA during transcription.

During transcription, supercoils form when the DNA is unwound and can add stress to the molecule. Topoisomerase activity enables RNA elongation to proceed further without being

stopped. This action of topoisomerase also takes place during DNA replication.

Solution to Question 6:

Synthesis of rRNA is not a function of RNA polymerase II. This is done by RNA polymerase I

Solution to Question 7:

The σ subunit (sigma factor) enables RNA polymerase to recognize promoter regions on the DNA and thus helps to position it correctly on the template DNA.

Solution to Question 8:

The mushroom death cap (also known as Amanita phalloides) produces the toxin, α-amanitin which inhibits RNA polymerase.

In eukaryotes, α-Amanitin forms a tight complex with the polymerase, thereby inhibiting mRNA synthesis and, ultimately, protein synthesis.

Note: Rifampicin inhibits the prokaryotic DNA-dependent RNA polymerase.

Solution to Question 9:

RNA polymerase II gives signals for the termination of RNA transcription. Proteins catalyzing RNA processing, termination, and polyadenylation appear to load onto RNA polymerase II soon after initiation.

Termination of transcription can be:

• ρ dependent: When the termination signals are met, ρ factor binds to RNA. ρ factor has ATP dependent helicase activity. It detaches the RNA from the DNA.

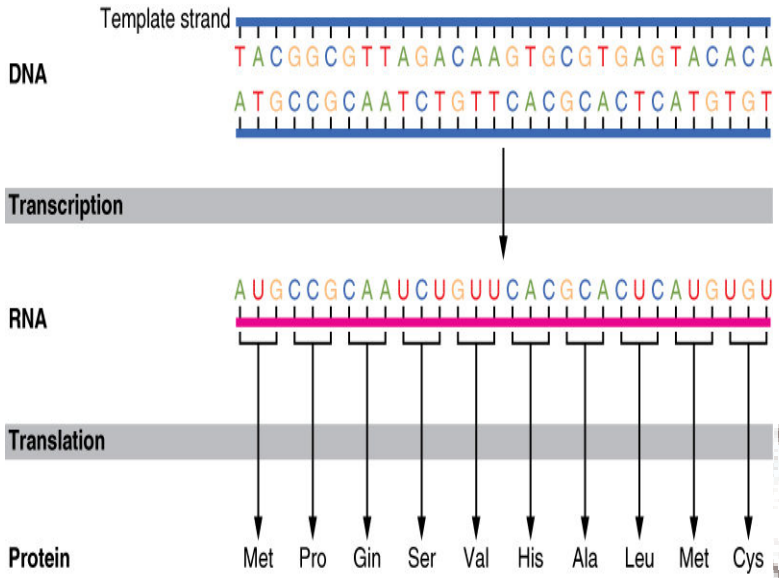
• ρ independent: In the 3' end of RNA, there is a series of U. RNA detaches from DNA due to weaker A=U at the end.

Solution to Question 10:

The non-template strand or coding strand in the DNA will have the same sequence of the RNA that is produced, except that the non-template or coding strand has T is in place of U.

As all sequences are written in the 5′ to 3′ direction, unless otherwise specified, an RNA sequence of UAAGGC would correspond to a DNA sequence, on the non-template strand, of TAAGGC. Both the non-template strand and RNA produced will be a complementary sequence to the template

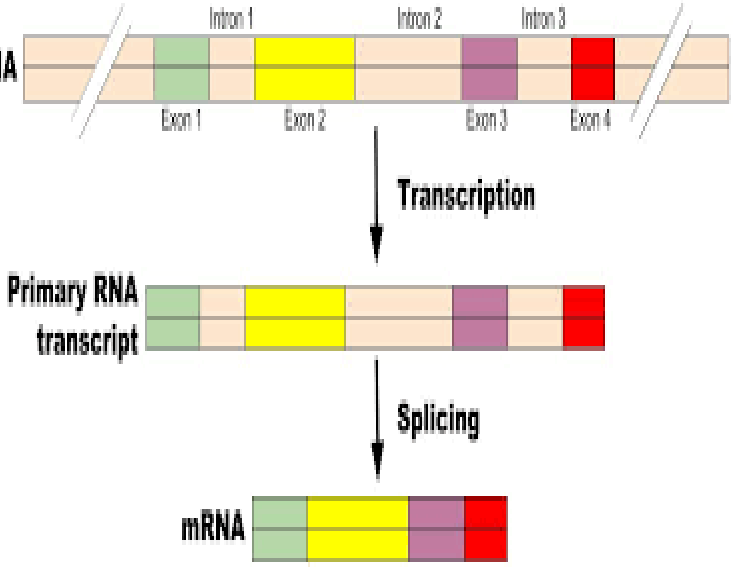
strand. Therefore, the template strand would be ATTCCG (written 5′ to 3′).



Solution to Question 11:

The RNA product, which is synthesized in the 5′-3′ direction, is the primary transcript. The primary transcripts are generated by RNA polymerase II.

The longest primary transcript is produced by the dystrophin gene.



Solution to Question 12:

7-methyl-guanosine triphosphate capping of primary transcript is necessary for:

• Processing of the primary transcript to mRNA

• Translation of the mRNA

• Protection of the mRNA against nucleolytic attack by 5′-exonucleases

Solution to Question 13:

The attachment of mRNA to the ribosome is mediated through the guanyl cap at the 5' end. Eukaryotic mRNAs have 7-methylguanosine caps at the 5' end and poly-A tails at the 3' end.

The 7-methylguanosine caps (Guanyl caps) are necessary for:

• Processing of primary transcripts to mRNA.

• Mediating the attachment of mRNA to the ribosome for translation.

• Protection of mRNA against nucleolytic attack by 5'exonucleases.

Other options:

Option A: AAUAAA is present at the 3' end, which is the polyadenylation sequence (poly-A tail). They help stabilize the mRNA, facilitate its exit from the nucleus and aid in translation. After the mRNA enters the cytosol, the poly-A tail is gradually shortened.

Option B: tRNA molecules serve as adapters for the translation of the information in the sequence of nucleotides of the mRNA into specific amino acids.

Option D: Shine-Dalgarno sequence initiates translation in prokaryotes.

Note: The guanyl cap and poly-A tail aid in attaching the mRNA to the ribosome for translation, however, it is initiated and mediated first through the Guanyl cap. The poly(A) tail stimulates recruitment of the 40S ribosomal subunit to the mRNA, after which the ribosomal subunit binds to the guanyl cap at 5'end of mRNA.

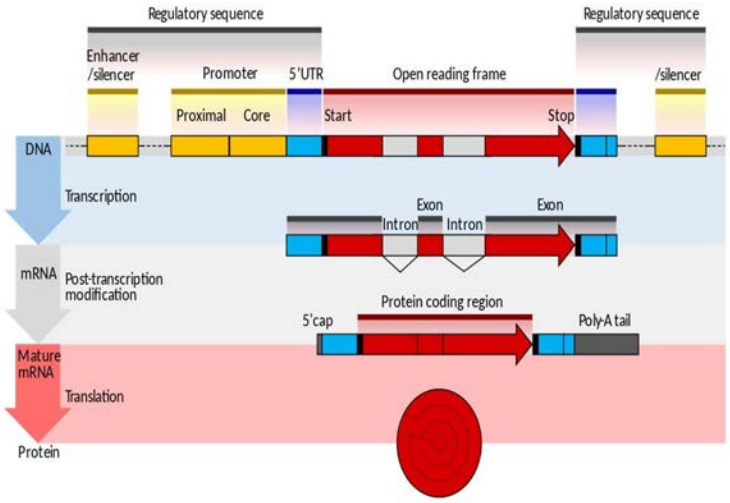
Solution to Question 14:

After synthesis, eukaryotic RNA undergoes extensive post-transcriptional modifications including 7-methylguanosine capping, polyadenylation forming poly A tail and splicing

• 5' Capping: This is the first of the processing reactions for pre mRNA. The cap is a 7-methylguanosine attached backward to the 5'-terminal end of the mRNA, forming an unusual 5'° 5' triphosphate linkage.

• Poly-A tail is added after transcription by the nuclear enzyme, polyadenylate polymerase, using ATP as the substrate. These tails help stabilize the mRNA, facilitate its exit from the nucleus, and aid in translation.

• Maturation of eukaryotic mRNA usually involves the removal of introns & joining of exons together to form the mature mRNA. The process of removing introns and joining exons is called splicing. snRNA facilitates the removal of introns by forming base pairs with the consensus sequences at each end of the intron.



Solution to Question 15:

tRNA of Alanine does not require 5' capping.

Capping process protects the 5′ end of mRNA from the action of exonucleases and polyadenylation protects the 3′ end of the mRNA. All mRNA, snRNA, and siRNA undergo this process, but tRNA are comparatively stable and do not require a capping at the 5' end.

Note: rRNAs also do not require 5' capping.

Solution to Question 16:

Self-splicing introns do not require a spliceosome or any other proteins for the splicing reaction.

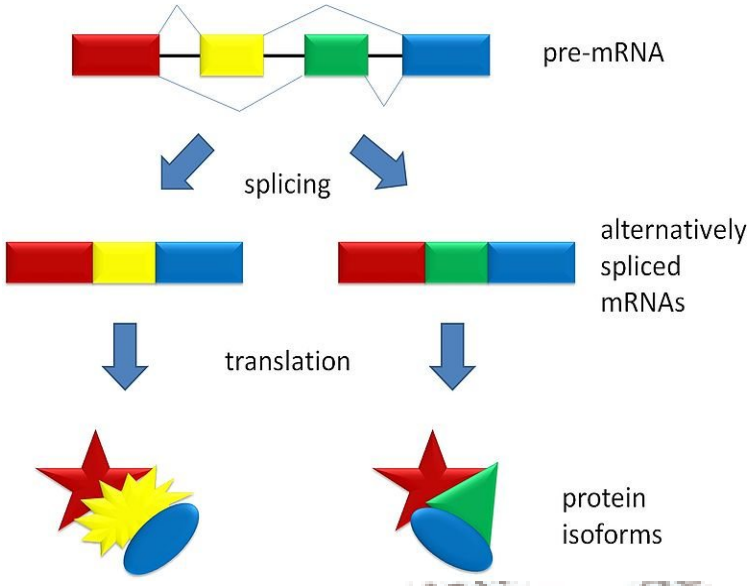
Self-splicing introns fold into complex loops and undergo transesterification reactions, thus undergoing self splicing.

Other options:

Option A: Split genes have the advantage that they allow alternative splicing so that a single gene can code for variant forms of the same protein.

Option C: β-thalassemia results from the faulty splicing of the β-globin transcript. However, the spliceosome is normal and the reason for the fault is a mutation in the β-globin gene itself.

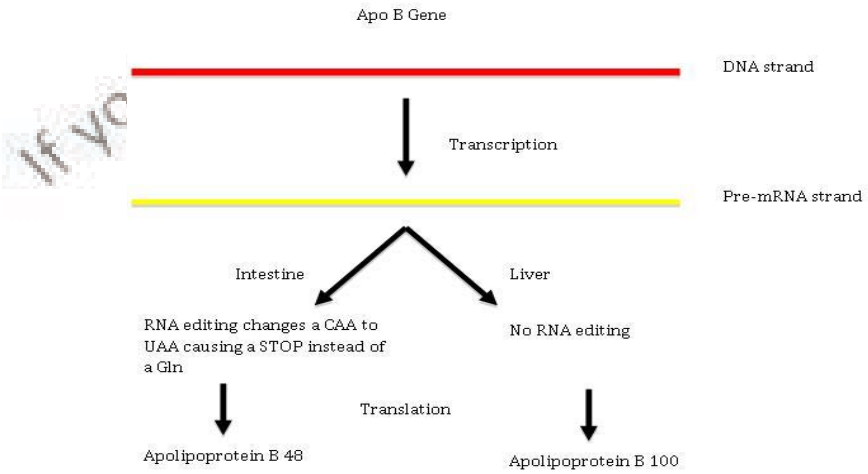
Option D: Splicing occurs in the nucleus and mature transcripts are then transported into the cytosol for translation (protein synthesis) to occur.



Solution to Question 17:

RNA editing occurs in 3' to 5' direction. It is the process by which the coding sequence is changed at the mRNA level before translation.

RNA editing occurs by insertion of additional Us at the 3' end of the mRNA and proceeds in a 3' to 5' direction. The guide RNA (gRNA) acts as a template for the editing process. An example of RNA editing is apoB gene and mRNA shown in the image below:



Solution to Question 18:

The eukaryotic initiation factor (eIF) that helps in cap binding is eIF 4.

eIF are accessory proteins that help initiate protein synthesis.

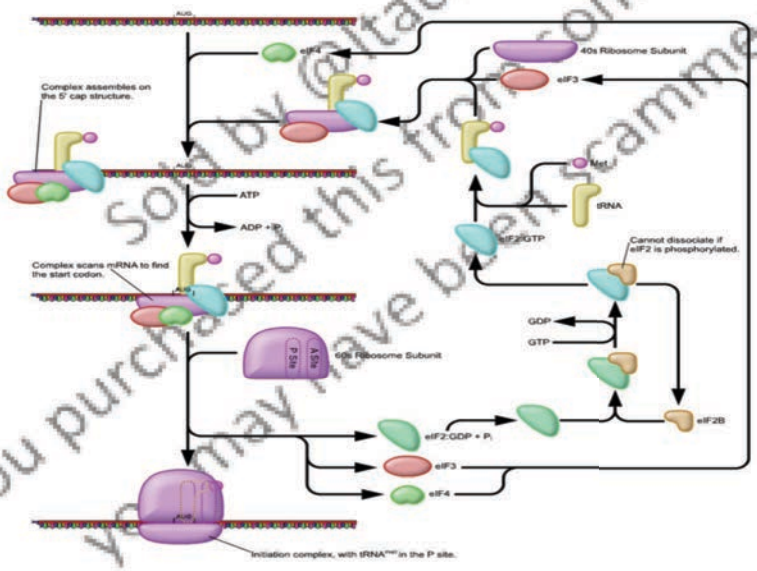
Functions of eIF:

• eIF3 & eIF 1A: First step of initiation involves dissociation of the ribosome into its 40S and 60S subunits. eIF3 & eIF 1A bind to newly dissociated 40s ribosomal subunit and hence delays the association of 40s and 60s.

• eIF2: It is involved in the formation of binary complex (GTP+eIF). This complex binds to met t RNA. Further, the GTP-eIF-2-tRNAi ternary complex binds to the 40S ribosomal subunit to form the 43S preinitiation complex. This is the second step of initiation.

• eIF4: It forms the cap binding complex. It binds to 5′ terminal of mRNA molecules which are capped. The mRNA would then bind to the 43S preinitiation complex to form the 48S initiation complex, in the third step of initiation.

• eIF5: It is involved in the hydrolysis of GTP bound to eIF2. This results in binding of 60s subunit with 48s initiation complex to form the 80s initiation complex. This is the fourth step of initiation.



Solution to Question 19:

Eukaryotic elongation factor 2 (eEF-2) is required for the translocation of peptidyl-tRNA along the mRNA during translation.

The elongation reaction requires GTP hydrolysis. The corresponding elongation factor in bacteria is EF-G. Diphtheria toxin ADP-ribosylates eEF-2, leading to its inactivation.

Other options:

Option B: Initiation of eukaryotic protein synthesis requires several initiation factors, designated eIF-1, eIF-2, etc.

Option C: Chloramphenicol inhibits prokaryotic peptidyl transferase.

Option D: Peptidyl transferase is an rRNA (within the large ribosomal subunit) involved in formation of the peptide bond between the amino acid groups within the A and P sites of the ribosome.

Solution to Question 20:

Pyrrolysine is coded by UAG is the correct statement.

Other options:

Option A: Selenocysteine is coded by UGA and not UAA

Option C: Selenocysteine is considered as the 21st amino acid

Option D: Other names of stop codons are as follows:

• UAA: Ochre

• UAG: Amber

• UGA: Opal

Solution to Question 21:

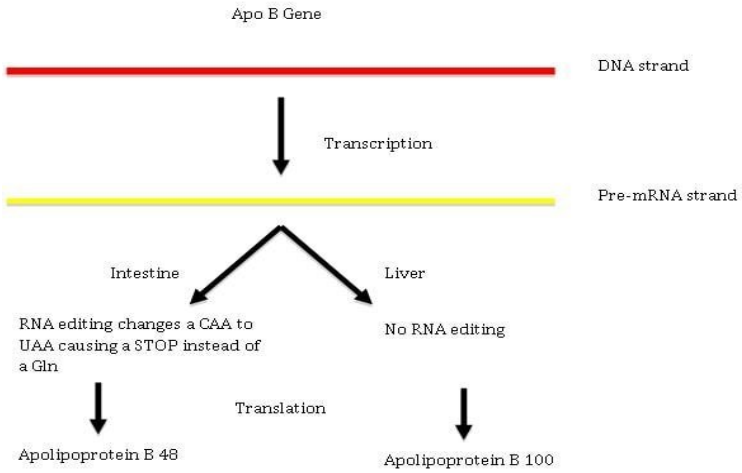
The process of apolipoprotein B48 synthesis involves RNA editing.

RNA editing is the process by which the coding sequence of the mRNA is changed before translation. As a result, the mRNA template differs from the DNA from which it was transcribed. This leads to the formation of a different protein. This occurs by insertion of additional uracils at the 3' end of the mRNA and proceeds in 3' to 5' direction.

RNA editing in apoB synthesis:

• In the liver, the apoB gene is transcribed to mRNA which is then translated to apoB 100.

• In the intestine, the same apoB gene is transcribed to the same mRNA. But now, the mRNA undergoes editing by an enzyme cytidine deaminase which converts the CAA codon in the mRNA to UAA at a single specific site. This edited mRNA translates to apoB 48.



Solution to Question 22:

Among the given options, using primers W and Y will yield the desired product A.

Promoters or DNA regulatory elements usually precedes the desired exon to be transcribed. It helps redirect transcription of only the desired exons of a gene to yield a product. Alternative splicing utilizes different promoters to code slightly different products from the same gene.

In this question, the desired product A has segments 1,3,4,5 whilst the other product B has 1,2,4,5. The difference between both is the presence of seg 3 in product A and the presence of seg 2 in product B. As the product in question is A, only seg 3 is required while excluding seg 2 and ensuring seg 1,4 and 5 intact.

• Seg 1- Needs to be transcribed and so W is essential

• Seg 2- Does not need to be transcribed.

• Seg 3- Needs to be transcribed and so Y is essential

• Seg 4- Needs to be transcribed but Z is not essential as Y continues the transcription in the direction shown by the arrow.

The answer is hence promoter W and Y.

Regulation of gene expression

Question 1:

All of the following are examples of housekeeping genes except _____

a) Ribosomal protein genes

b) rRNA genes

c) Beta galactosidase

d) RNA polymerase

Question 2:

Which protein is produced by a regulatory gene?

a) Operon

b) Inducer

c) Promoter

d) Repressor

Question 3:

What does the operon model attempt to explain?

a) The coordinated control of gene expression in bacteria

b) Bacterial resistance to antibiotics

c) How genes move between homologous regions of DNA

d) The mechanism of viral attachment to a host cell

Question 4:

In the Lac operon model, how does the presence of lactose activate transcription?

a) Lactose directly activates RNA polymerase

b) Lactose binds to the operator region and blocks the attachment of RNA polymerase to the promoter

c) Lactose binds directly to the promoter region and activates transcription

d) Lactose binds to repressor protein and inactivates it

Question 5:

The lactose operon is likely to be transcribed when _____

a) There is more glucose in the cell than lactose

b) Cyclic AMP levels are low

c) There is glucose but no lactose in the cell

d) Cyclic AMP and lactose levels are both high within the cell

Question 6:

A mutation in which section of DNA could influence the binding of RNA polymerase to the DNA?

a) Operon

b) Inducer

c) Promoter

d) Corepressor

Question 7:

The tryptophan operon is a repressible operon that is _____

a) Permanently turned on

b) Turned on only when tryptophan is present in the growth medium

c) Turned off only when glucose is present in the growth medium

d) Turned off whenever tryptophan is added to the growth medium

Question 8:

Two potential devices that eukaryotic cells use to regulate transcription are _____

a) DNA methylation and histone amplification

b) DNA amplification and histone methylation

c) DNA acetylation and methylation

d) DNA methylation and histone acetylation

Question 9:

The phenomenon in which RNA molecules in a cell are destroyed if they have a sequence complementary to an introduced double-stranded RNA is called _____

a) RNA interference

b) RNA obstruction

c) RNA blocking

d) RNA targeting

Question 10:

Which of the following describes the function of an enzyme known as dicer?

a) It degrades single-stranded DNA

b) It degrades single-stranded mRNA

c) It degrades mRNA with no poly(A) tail

d) It trims small double-stranded RNAs into molecules that can block translation

Question 11:

Which of the following does not favor euchromatin formation due to changes occurring at cytosine residues at CpG islands in DNA?

a) Methylation

b) Phosphorylation

c) Alkylation

d) Sumoylation

Question 12:

DNA methylation is not involved in?

a) 1 and 2

b) 2 and 3

c) 1 and 4

d) 2 and 4

Answer Key

Question No.	Correct Option
1	c
2	d
3	a
4	d
5	d
6	c
7	d
8	d
9	a
10	d
11	a
12	d

Detailed Explanations

Solution to Question 1:

The gene expressing beta-galactosidase is not a housekeeping gene.

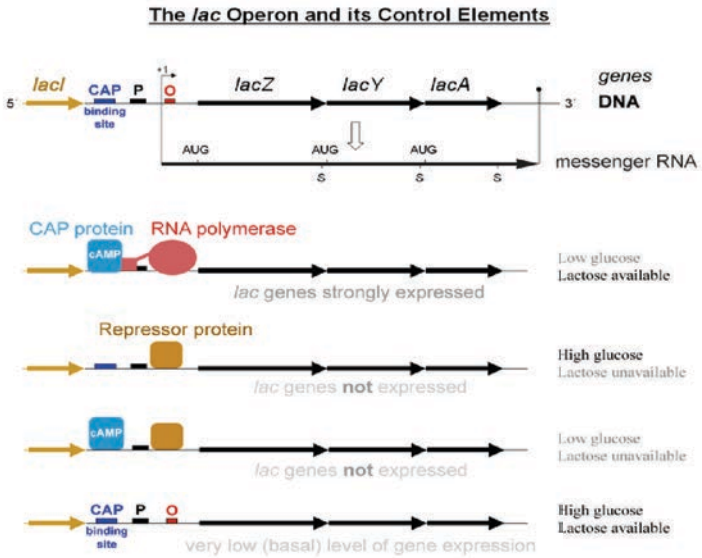
Housekeeping or constitutive genes are those which are expressed at a constant rate in almost all the cells of the body. These genes are required for the basal cellular function. Examples include genes encoding RNA polymerase, ribosomal protein, rRNA, enzymes for glycolysis, etc.

In contrast, inducible genes are those which are expressed under special circumstances. The expression of these genes increase in response to an activator or inducer, or decrease in response to a repressor.

Solution to Question 2:

A repressor protein is produced by a regulatory gene.

In bacterial operons, a regulatory gene, like the lac I gene, which is present upstream of a set of structural genes, codes for a repressor protein (a transacting factor). The repressor protein binds to the operator site and interferes with the progress of RNA polymerase. This blocks transcription of the structural genes and is an example of negative regulation of gene transcription.



Solution to Question 3:

The operon model attempts to explain the coordinated control of gene expression in bacteria.

In bacteria, the structural genes that code for proteins involved in a particular metabolic pathway are often found sequentially grouped on the chromosome. The cis-acting regulatory elements that determine the transcription of these genes are also grouped with the structural genes.

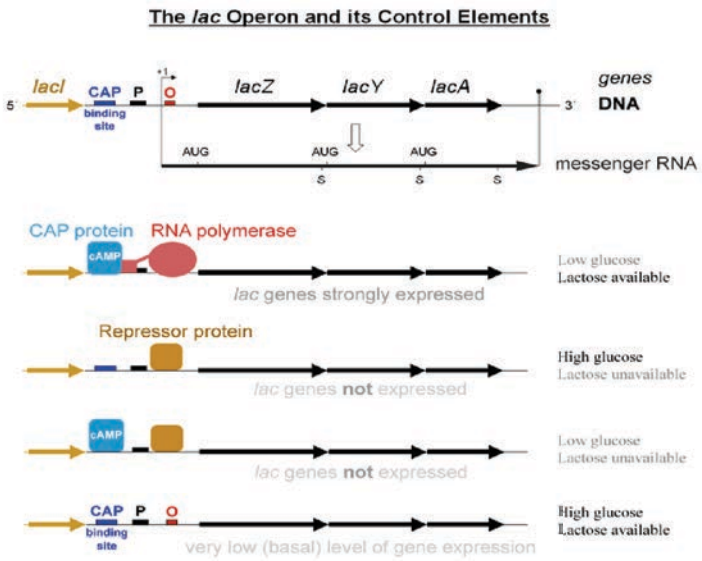
The transcription product of the entire operon is a single polycistronic messenger RNA (mRNA). The structural genes described above are coordinately controlled, that is, turned on or off as a unit. This entire package, of structural and regulatory genes, is referred to as an operon. An example of this is the lac operon in Escherichia coli.

Note: Gene regulation in bacteria is important in order to:

• Enable response to environmental stimuli

• Allow cells to adjust promptly to changes in the growth medium

• Alter biochemical capabilities appropriate for available substrate



Solution to Question 4:

In the Lac operon model, lactose binds to the repressor protein and inactivates it, thus activating transcription.

When lactose is present in abundance, a small amount of lactose is converted to allolactose. This compound binds to the repressor protein of the lac operon and creates a conformational change. As a result, the repressor cannot bind to the operator and transcription of the structural genes occurs. This is an example of negative regulation.

This can only occur when glucose is absent. In the absence of glucose, adenylyl cyclase is active. This generates sufficient quantities of cAMP which bind to catabolite activator protein (CAP).The cAMP–CAP trans-acting complex binds to the CAP-binding site, another regulatory region present upstream of the structural genes of the operon. This causes RNA polymerase to more efficiently initiate transcription at the promoter site. This is an example of positive regulation.

Solution to Question 5:

The lac operon is induced only when cyclic AMP and lactose levels are both high within the cell.

When lactose is present in abundance, a small amount of lactose is converted to allolactose. This compound binds to the repressor protein of the lac operon and creates a conformational change. As a result, the repressor cannot bind to the operator and transcription of the structural genes occurs. This is an example of negative regulation.

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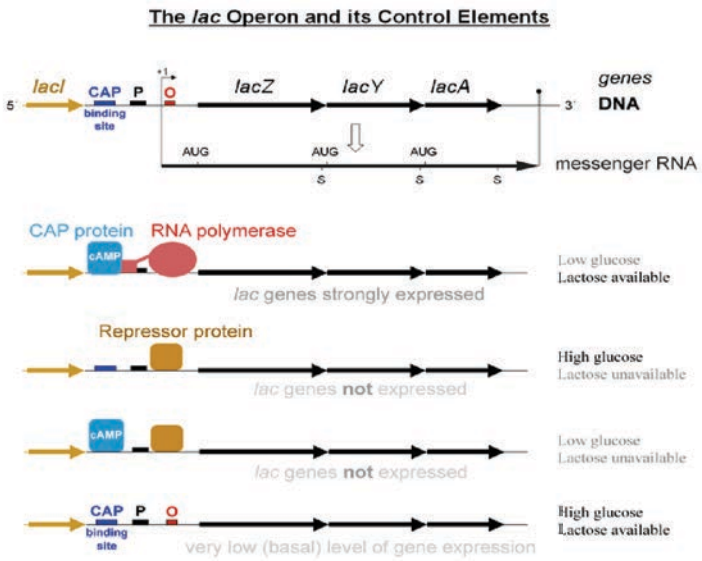


Solution to Question 6:

A mutation in the promoter section of DNA could influence the binding of RNA polymerase to the DNA.

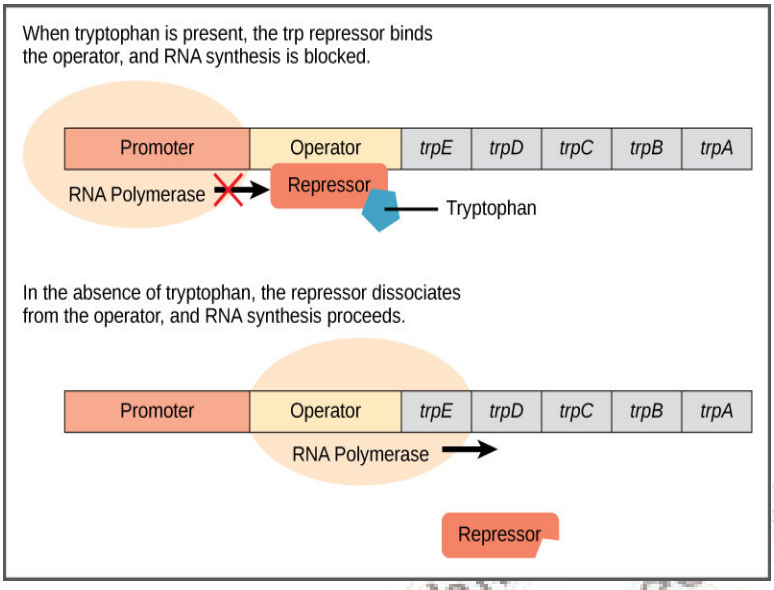
The regulatory portion of bacterial operon is upstream of its structural genes, and consists of the promoter (P) region where RNA polymerase binds and two additional sites, the operator (O) site and the CAP site, where regulatory proteins bind.

Corepressor is a substance that facilitate the inhibition of gene expression. In prokaryotes, corepressors are small molecules whereas, in eukaryotes, corepressors are proteins. A corepressor binds to the repressors and activates the repressor transcription factor. The repressor, in turn, binds to the operator site, thereby blocking the transcription from the structural genes. A lack of corepressor would result in the inability of the cell to ■turn off■ genes.

Solution to Question 7:

The tryptophan operon is a repressible operon that is turned off whenever tryptophan is added to the growth medium.

The tryptophan operon contains five structural genes: trp E, trp D, trp C, trp B, trp A, which encode enzymes required for the synthesis of tryptophan. It also contains a promoter that binds to RNA polymerase. Its operator blocks transcription when it is bound to repressor protein synthesized by the repressor gene (trp R). In this, tryptophan is a corepressor i.e. it facilitates binding of the repressor protein to the operator.



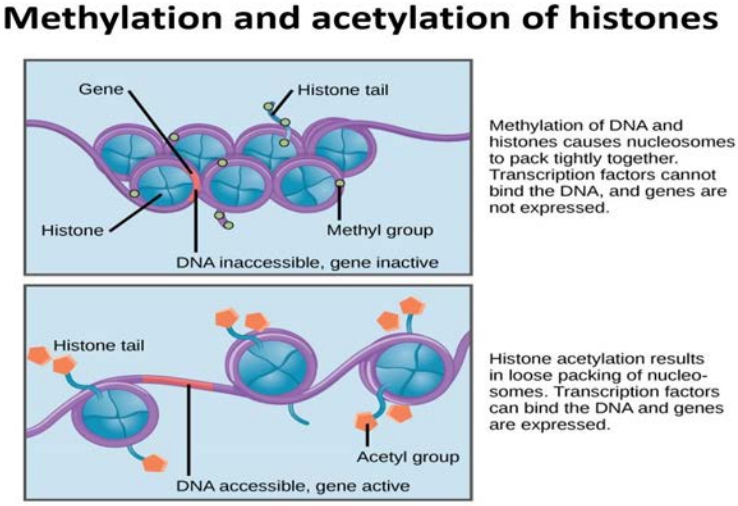
Solution to Question 8:

Two potential devices that eukaryotic cells use to regulate transcription are as follows DNA methylation and histone acetylation.

DNA methylation occurs by methyltransferases that use S-adenosyl-methionine as the methyl donor. It silences gene expression.

Histone acetylation decrease the positive charge on histone proteins, thus decreasing the strength of their association with negatively charged DNA. This chromatin remodeling relaxes the nucleosome, allowing transcription factors access to specific regions on the DNA. Thus, histone acetylation improves gene expression.

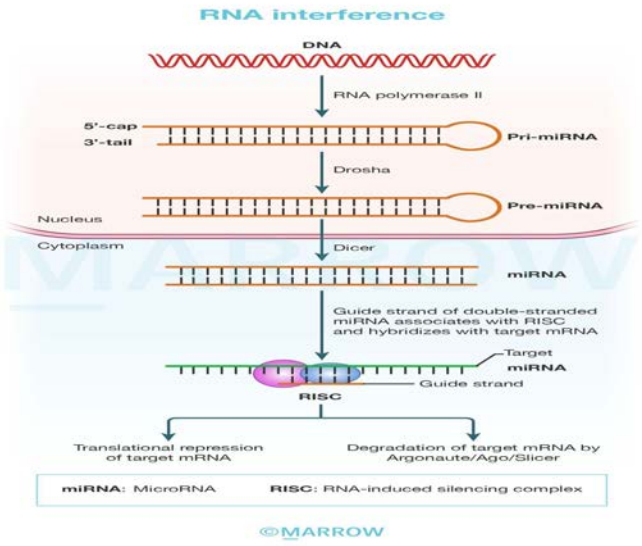
Note: In both eukaryotes and prokaryotes, gene expression is primarily regulated at the level of transcription.



Solution to Question 9:

The phenomenon in which RNA molecules in a cell are destroyed if they have a sequence complementary to an introduced double-stranded RNA is called RNA interference.

RNA interference is a mechanism of gene silencing through decreased expression of mRNA, either by repression of translation or by increased degradation. It is thought to play a key role in such a fundamental process as cell proliferation, differentiation, and apoptosis.



Solution to Question 10:

Dicer is an endonuclease that trims small double-stranded RNA into molecules that can block translation.

Dicer is an enzyme that cleaves the cytosolic dsRNA encoded by the genome and generates short, double-stranded (ds) miRNA. One of the two strands (the guide or antisense strand) selected for loading into the RNA- induced silencing complex (RISC) which is composed of one of four argonaute proteins (Ago 1° 4). The RNA then forms a mature, functional 21–22 nt single-stranded miRNA.

In the RISC complex, miRNAs can modulate mRNA function by one of three mechanisms:

(1) Promoting mRNA degradation directly

(2) Stimulating CCR4/NOT complex-mediated poly(A) tail degradation

(3) Inhibition of translation by targeting the 5′-methyl cap-binding translation factor

Solution to Question 11:

Methylation does not favour euchromatin formation due to changes cytosine residues at CpG islands in DNA.

The transcriptional regulatory regions of active genes often contain a high frequency of CpG dinucleotides (referred to as CpG islands) and they are normally unmethylated. Therefore, methylation is an epigenetic modification that inactivates DNA, and hinders the formation of euchromatin (transcriptionally active chromatin). Epigenetic changes include alterations of chromatin structure by:

• Methylation of cytosine residues in CpG dinucleotides

• Modification of histones by acetylation or methylation

• Changes in higher-order chromosome structure

Sumoylation of histones (SUMO - Small Ubiquitin-related MOdifier) is associated with transcription repression. Other possible roles of posttranslationally modified histones:

• Acetylation of core histones is associated with chromosomal assembly during DNA replication.

• Phosphorylation of histone H1 is associated with the condensation of chromosomes during the replication cycle.

• ADP-ribosylation of histones is associated with DNA repair.

• Methylation of histones is correlated with activation and repression of gene transcription.

• Acetylation of histones H3 and H4 is associated with the activation or inactivation of gene transcription.

• Monoubiquitylation is associated with gene activation, repression, and heterochromatic gene silencing.

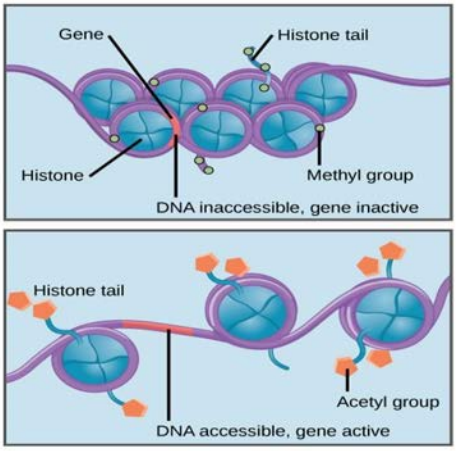
Solution to Question 12:

DNA methylation is not involved in the base pair excision repair mechanism (statement 4) and RNA splicing (statement 2).

DNA methylation is an epigenetic modification used as a gene expression regulation mechanism. It causes gene silencing by inactivating DNA and hindering the formation of euchromatin (transcriptionally active chromatin). Thus, it also aids in chromatin remodelling.

It however is not involved in the base pair repair mechanism or RNA splicing.

Methylation and acetylation of histones



Methylation of DNA and histones causes nucleosomes to pack tightly together. Transcription factors cannot bind the DNA, and genes are not expressed.

Histone acetylation results in loose packing of nucleosomes. Transcription factors can bind the DNA and genes are expressed.

Molecular genetics, recombinant DNA & genomic technology

Question 1:

Enzymes that cut DNA at specific DNA sequences within the molecule are _____

a) Helicase enzyme

b) Polymerase enzyme

c) Restriction enzyme

d) Gyrase enzyme

Question 2:

Restriction enzymes are found with _____

a) Site-specific DNA methylases

b) RNA-inducible interferon system

c) Thermostable DNA polymerases

d) Terminal transferase

Question 3:

Restriction endonuclease will act on which of the following:

a) AAGAAG

b) GAGAGG

c) AAGCTT

d) TACGAG

Question 4:

Which of the following tools of recombinant DNA technology is incorrectly paired with its use?

a) DNA polymerase - copies DNA sequences in the polymerase chain reaction

b) DNA ligase - enzyme that cuts DNA, creating sticky ends

c) Reverse transcriptase - production of cDNA from mRNA

d) Electrophoresis - RFLP analysis

Question 5:

Gene cloning uses _____.

a) Plasmids

b) Noncoding nucleotide sequences

c) Bacteriophage

d) A & C

Question 6:

Which of the following cloning vectors has linear DNA?

a) Phages

b) Plasmid

c) Cosmid

d) All of above

Question 7:

Genomic library is a term used to describe _____

a) A computerized listing of known DNA sequences

b) Bacterial plasmids containing DNA fragments representing the majority of the genetic information from a plant or animal

c) A library of complementary DNA copies of the population of mRNAs in a tissue

d) A compilation of the amino acid sequences of protein coding genes

Question 8:

Northern blotting is used for detection of which of the following?

a) DNA

b) RNA

c) Proteins

d) None

Question 9:

What is the correct sequence of events in Southern blotting?

a) 1, 2, 3

b) 2, 1, 3

c) 3, 1, 2

d) 2, 3, 1

Question 10:

Approximately how many polymerase chain reaction (PCR) products are produced from a single template DNA molecule in a PCR reaction that continues for 30 cycles?

a) 64

b) 128,000

c) Approximately 1 million

d) Approximately 1 billion

Question 11:

Which of the following is NOT required for a PCR reaction?

a) A thermostable DNA polymerase

b) Dideoxy-dNTPs (ddNTPs)

c) Primers

d) Template DNA

Question 12:

Which of the following is a function of PCR?

a) To detect infectious agents, especially latent viruses

b) Make prenatal genetic diagnoses

c) Detect allelic polymorphisms

d) All

Question 13:

RT-PCR is a method that is used for _____

a) Forensic analysis of DNA

b) Amplification of genomic DNA sequences

c) Amplification of mRNA sequences only

d) Analysis of mRNA expression and its amplification

Question 14:

Which of the following statements about forensic analysis of DNA is correct?

a) A DNA profile using short tandem repeats is not unique to an individual

b) Forensic analysis makes use of SNPs in coding sequences to distinguish between individuals

c) PCR is used for DNA profiling (DNA fingerprinting)

d) DNA fingerprinting cannot be used for paternity testing

Question 15:

In which technique of gene localization is a radioactive probe used?

a) Somatic cell hybridization

b) In situ hybridization

c) Fluorescence in situ hybridization (FISH)

d) All of the above

Question 16:

Which of the following diseases is an example of point mutation?

a) Hemoglobin Lepore

b) β-Thalassemia type III

c) Diabetes

d) Sickle cell disease

Question 17:

Restriction fragment length polymorphism (RFLP) is used to _____

a) Construct high resolution linkage map

b) Identify single gene disease

c) Construct QTL maps

d) All of the above

Question 18:

Which of the following is correct regarding CRISPR-Cas9?

a) It is a system used by bacterial cells to recognise and destroy viral DNA as a form of adaptive immunity

b) It is a tool which is designed to detect TB

c) It is tool designed to test air pollution

d) It is a tool used in DNA sequencing

Question 19:

Which of the following site does the restriction enzyme act when RFLP is used in-order to identify the five different species of staphylococci in a surgical ICU?

a) ATGGAC-TACGTG

b) GATATC- CTATAG

c) TAGATA-ATCTCT

d) AATATA-TATAAT

Question 20:

Which of the following is the best screening investigation for metabolic disorders?

a) Western blot

b) Tandem mass spectrometry

c) PCR

d) Gel electrophoresis

Question 21:

The best method to measure HbA1C is _____

a) Isoelectric focusing

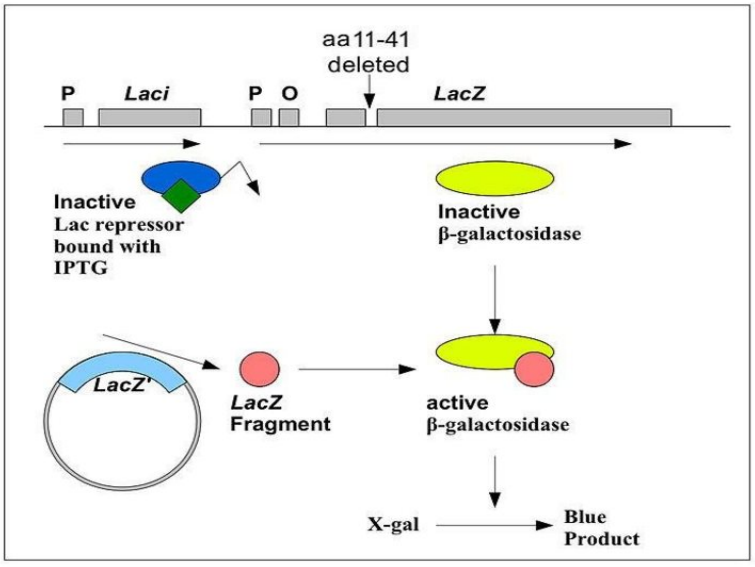
b) Affinity chromatography

c) Ion exchange chromatography

d) Electrophoresis

Question 22:

Identify the phenomenon shown below.



a) Translation

b) Transformation

c) Complementation

d) Conjugation

Question 23:

In a patient with chronic myeloid leukemia, the translocation in Philadelphia chromosome is best studied by

a) G banding

b) SKY

c) Southern blotting

d) DNA sequencing

Question 24:

Which of the following techniques is used for the detection of variations in DNA sequences and gene expression?

a) Microarray technique

b) Northern blot

c) Southern blot

d) South western blot

Question 25:

Array CGH can detect all except?

a) Mutation in cancers

b) Subtelomeric deletion

c) Balanced translocation

d) Aneuploidy

Question 26:

The correct order of steps in Polymerase Chain Reaction (PCR) is:

a) Hybridisation, Annealing, Elongation, Denaturation

b) Elongation, Denaturation, Hybridisation, Annealing

c) Denaturation, Annealing, Extension

d) Annealing, Extension, Denaturation

Question 27:

Wobble hypothesis explains which of the following concepts of the genetic code?

a) Universal

b) Unambiguous

c) Degeneracy

d) Non-punctuated

Answer Key

Question No.	Correct Option
1	c
2	a
3	c
4	b
5	d
6	a
7	b
8	b
9	c
10	d
11	b
12	d
13	d
14	c
15	b
16	d
17	d
18	a
19	b
20	b
21	c
22	c
23	b
24	a
25	c

26	c
27	c

Detailed Explanations

Solution to Question 1:

Enzymes that cut DNA at specific DNA sequences within the molecule are called restriction enzymes.

These defensive enzymes protect the host bacterial DNA from the DNA genome of foreign organisms (primarily infective phages) by specifically inactivating the invading phage DNA by digestion.

Solution to Question 2:

Restriction enzymes are found with site-specific DNA methylases.

Restriction endonucleases are present in cells that also have a companion enzyme that site-specifically methylates the host DNA, thus rendering it an unsuitable substrate for digestion by that particular restriction enzyme.

Thus, site-specific DNA methylases and restriction enzymes that target the same sites always exist in pairs in a bacterium.

Solution to Question 3:

Restriction endonucleases can act on AAGCTT.

Restriction endonucleases act and cleave at the site of a palindromic sequence. A palindromic sequence is a sequence that reads the same backward and forward. An example is listed below.

5' --AAGCTT-- 3'

3' --TTCGAA-- 5'

Solution to Question 4:

DNA ligase is not paired correctly with its use for cutting DNA and creating sticky ends. DNA ligase joins the adjacent nucleotides in a covalent linkage.

Restriction endonucleases cut DNA at specific sites creating sticky ends.

Solution to Question 5:

Gene cloning uses both plasmids and bacteriophages.

Hybrid DNA molecules can be constructed in cloning vectors typically using bacterial plasmids, phages, or cosmids. A clone is a large population of identical molecules, bacteria, or cells that arise from a common ancestor. Molecular cloning allows for the production of a large number of identical DNA molecules, which can then be characterized or used for other purposes.

Solution to Question 6:

Phages have linear DNA into which foreign DNA can be inserted at several restriction enzyme sites.

Amplification of DNA (cloning) is done using cloning vectors - bacterial plasmids, phages, or cosmids.

Bacterial plasmids are small, circular, duplex DNA molecules whose natural function is to confer antibiotic resistance to the host cell. Cosmid is a plasmid into which the DNA sequences from bacteriophage lambda (λ cos sites) have been inserted. This enables the packaging of plasmid DNA in vitro. They have a small, circular, duplex DNA. (Options B and C)

Solution to Question 7:

A genomic library is bacteria with plasmids containing DNA fragments representing the majority of the genetic information from a plant or animal.

Gene library describes the combination of restriction enzymes and various cloning vectors allows the entire genome of an organism to be individually packed into a vector. A collection of these different recombinant clones is called a library.

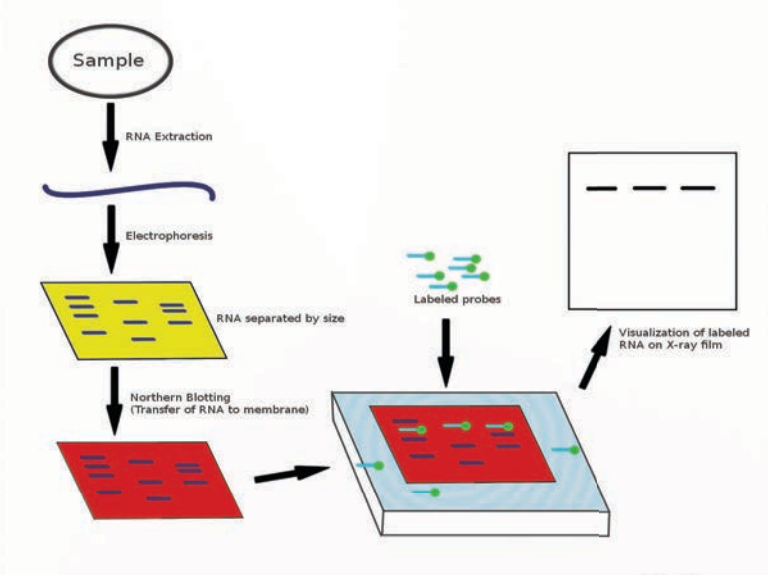
A genomic library is prepared from the total DNA of a cell line or tissue. A cDNA library comprises complementary DNA copies of the population of mRNAs in a tissue. (option C)

Genomic DNA libraries are often prepared by performing partial digestion of total DNA with a restriction enzyme that cuts DNA frequently.

Solution to Question 8:

Northern blotting is used for the detection of RNA.

Southern blotting is used for the detection of DNA and Western blotting is used for the detection of protein.



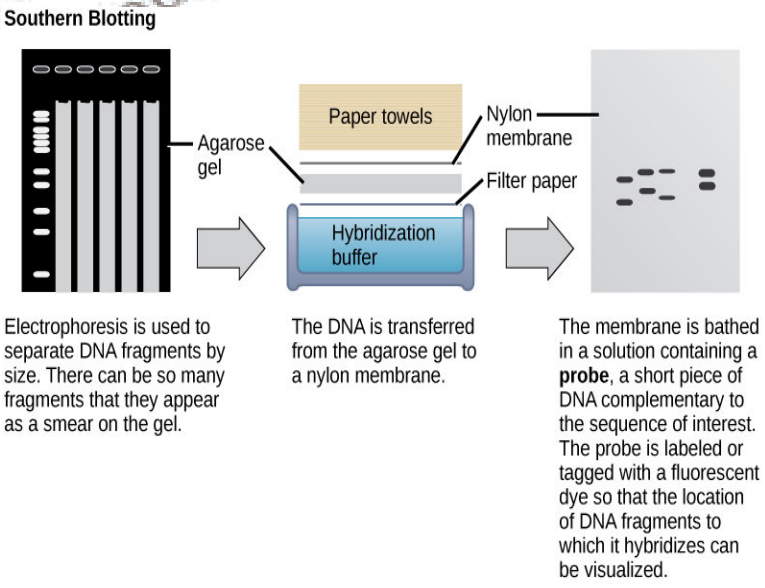
Solution to Question 9:

In Southern blotting DNA fragments are first separated by electrophoresis.

• The DNA in the gel is then made single-stranded by exposure to dilute alkali, and transferred by capillary action to the nylon membrane, which is laid over it. The relative positions of the different DNA fragments are thus preserved on the membrane.

• The membrane is then soaked in buffer containing the labelled probe, which base pairs with the DNA fragment of interest.

• The position of that fragment is then determined by autoradiographic (exposure of the membrane to X-ray film), or fluorescent detection of the hybridized probe.



Solution to Question 10:

A PCR reaction that continues for 30 cycles will produce approximately 1 billion PCR products from a single template DNA molecule.

Polymerase chain reaction (PCR) is a method of amplifying a target sequence of DNA. DNA sequences as short as 50 to 100 bp and as long as 10 kb can be amplified by PCR. The amplification is exponential.

• 20 cycles provide amplification of 220 = 106 or 1 million copies

• 30 cycles provide an amplification of 230 =109 or 1 billion copies

Solution to Question 11:

Dideoxy-dNTPs (ddNTPs) is not required in PCR reaction.

In a PCR reaction, the DNA sample is first heat-denatured (>90°C) to separate the two strands of the template DNA containing the target sequence. The primers, added in excess, are allowed to anneal to the DNA (typically 50–75°C); and each strand is copied by a DNA polymerase, starting at the primer sites in the presence of all four dNTPs(again in vast excess).

DNA synthesis is catalyzed by a heat-stable DNA polymerase purified from one of a number of different thermophilic bacteria, organisms that grow at 70–80°C.

Solution to Question 12:

All of the listed options are applications of PCR.

The applications of PCR to forensic medicine are as follows:

• To detect infectious agents, especially latent viruses

• Make prenatal genetic diagnoses

• Detect allelic polymorphisms

• Establish precise tissue types for transplants

• To study evolution, using DNA from archeological samples

• For quantitative RNA analyses after RNA copying and mRNA quantitation by the so-called RT-PCR method (cDNA copies of mRNA generated by a retroviral reverse transcriptase)

• To score in vivo protein-DNA occupancy using chromatin immunoprecipitation assays

Solution to Question 13:

RT-PCR is a method that is used for analysis of mRNA expression and its amplification.

Reverse transcriptase polymerase chain reaction (RT-PCR) is a variant of PCR and includes 2 steps:

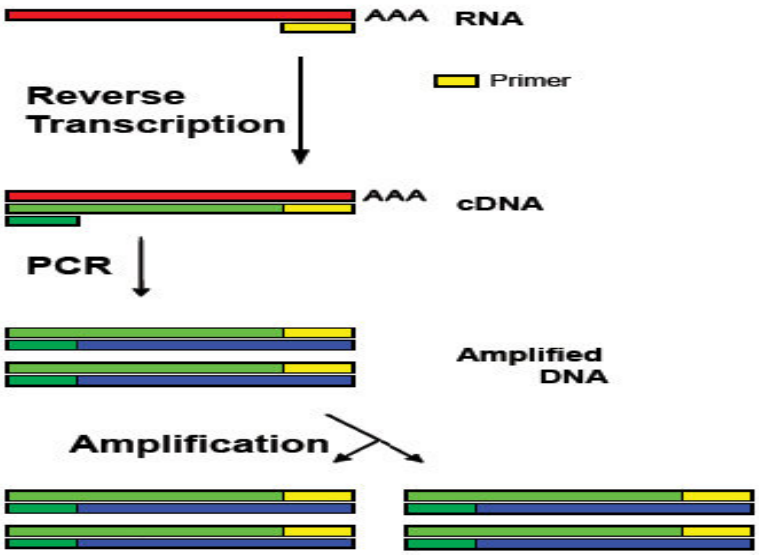
• Generation of complementary DNA (cDNA copies) from mRNA using reverse transcriptase - used to qualitatively detect gene expression

• Amplification of newly synthesized cDNA by traditional PCR -This can be quantitatively measured by using fluorescent dyes (qRT-PCR).

Uses:

• Determination of viral load in disease

• Screening of genetic diseases and cancers



Solution to Question 14:

PCR is used for DNA profiling (DNA fingerprinting).

The PCR allows the DNA in a single cell, hair follicle, or spermatozoon to be amplified and analyzed. Thus, the applications of PCR to forensic medicine are obvious. DNA fingerprinting, or DNA profiling, is used for forensic analysis of crime scenes and paternity testing (Option D).

Other options:

Option A: Short Tandem Repeat (STR) are short sequences of DNA at scattered locations in the genome, repeated in tandem (one after the other). E.g., CG-CG-CG-CG-CG. The number of these repeat units varies from person to person but is unique for any given individual and, therefore, serves as a molecular fingerprint.

Option B: Single Nucleotide Polymorphism(SNP) refers to variations that involve just one base at a given locus (Eg., base C replaced by base A), which can occur within exons, introns, intergenic or coding regions. They are almost always biallelic and are majorly used in the definition of inherited diseases where the functional deficit is unknown. Each of these variations is present to some appreciable degree within a population and hence not unique to an individual.

Solution to Question 15:

In situ hybridization uses a radioactive probe for gene localization.

It is added to a metaphase spread of chromosomes on a glass slide. The exact area of hybridization is localized by layering photographic emulsion over the slide and after exposure, lining up the grains with some histologic identification of the chromosome.

Fluorescence in situ hybridization (FISH) utilizes fluorescent probes, not radioactive probes. Fluorescent probes have greater safety, stability, and ease of detection (Option C).

Solution to Question 16:

Sickle cell disease is caused by a point mutation.

This is caused by mutation of a single base out of the 3 × 109 in the genome, a T-to-A DNA substitution, resulting in an A-to-U change in the mRNA corresponding to the sixth codon of the β-globin gene.

Defective production of β-globin results in a variety of diseases and is due to many different lesions in and around the β-globin gene.

Solution to Question 17:

Restriction fragment length polymorphism (RFLP) is used to construct a high-resolution linkage map, QTL maps, and identify single gene-disease.

An inherited difference in the pattern of restriction enzyme digestion (e.g., a DNA variation occurring in more than 1% of the general population) is known as a restriction fragment length polymorphism (RFLP). RFLPs result from single-base changes (e.g., sickle-cell disease) or from deletions or insertions (CNVs) of DNA into a restriction fragment (eg, the thalassemias) and have proved to be useful diagnostic tools.

SNPs(single nucleotide polymorphisms)/RFLPs can be used to establish linkage groups, which in turn, by the process of chromosome walking, will eventually define the disease locus.

Solution to Question 18:

The CRISPR-Cas9 system, found in many bacteria, represents a form of acquired, or adaptive immunity to infection by bacteriophages, which complements the system of restriction endonucleases and methylases.

In recent years, a novel DNA editing/gene regulatory system termed Clustered Regularly Interspersed Short Palindromic Repeats- CRISPR-associated gene 9 (CRISPR-Cas9) has been

developed. CRISPR uses RNA-based targeting to bring the Cas9 nuclease to foreign (or any complementary) DNA.

Within bacteria, this CRISPR-RNA-Cas9 complex then degrades and inactivates the targeted DNA.

Solution to Question 19:

Restriction endonuclease enzymes act at specific palindromic sequences that create DNA fragments of different lengths. By analyzing the fragments, specific species of bacteria can be identified. Option B is the only palindromic sequence here, hence it is the answer.

RFLP is a restriction fragment length polymorphism (RFLP) analysis technique that involves cutting a particular region of DNA with known variability, with restriction enzymes, then separating the DNA fragments by agarose gel electrophoresis and determining the number of fragments and relative sizes.

Cleavage of a sample of DNA with a restriction enzyme produces a characteristic set of smaller DNA fragments. Deviations in the normal product pattern, produce RFLPs that occur if a mutation renders a restriction site unrecognizable and alternatively, generates a new recognition site.

Solution to Question 20:

Tandem mass spectrometry (MS) is the technique of choice for screening neonates for inherited metabolic diseases.

Tandem MS employs the equivalent of two mass spectrometers linked in series (MS-MS or MS2). It can be used to screen blood samples from newborns for the presence and concentrations of amino acids, fatty acids, and other metabolites.

Abnormalities in metabolite levels indicate inherited metabolic diseases, such as phenylketonuria, ethylmalonic encephalopathy, and glutaric acidemia type 1.

Solution to Question 21:

The best method to measure HBA1C is ion-exchange chromatography.

In ion-exchange chromatography:

• Proteins interact with the stationary phase by charge-charge interactions.

• Proteins with a net positive charge at a given pH will tightly adhere to beads with negatively charged functional groups such as carboxylates or sulfates (cation exchangers).

• Proteins with a net negative charge adhere to beads with positively charged functional groups, typically tertiary or quaternary amines (anion exchangers).

• Non-adherent proteins flow through the matrix and are washed away.

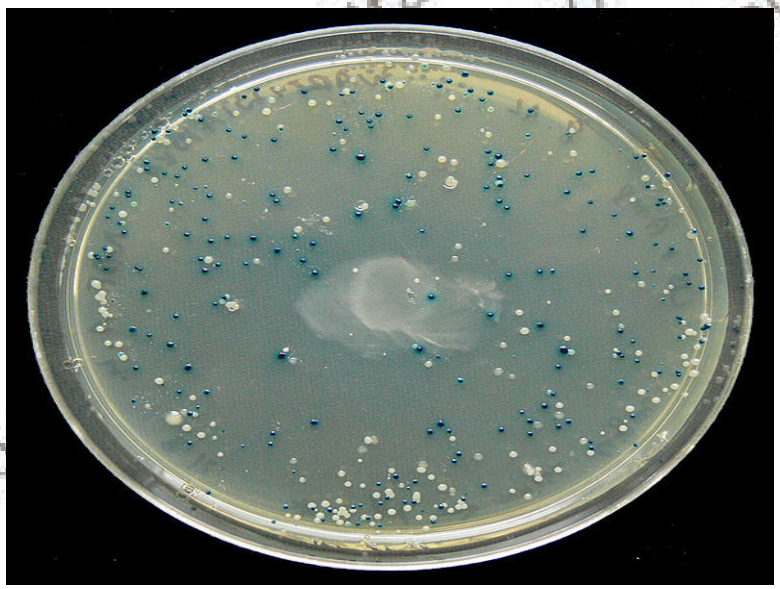
• Proteins elute in inverse order of the strength of their interactions with the stationary phase.

Solution to Question 22:

The image shows a schematic representation of the blue-white assay used in the screening of recombinant vectors for inserted DNA fragments. It uses the phenomenon of complementation (αlpha-complementation of the β-galactosidase gene).

Blue white assay: The DNA of interest is ligated into a vector. The vector is then inserted into a competent host cell viable for transformation, which is then grown in the presence of X-gal (X-gal detects active β-galactosidase). Cells transformed with vectors containing recombinant DNA will produce white colonies whereas cells transformed with non-recombinant plasmids (i.e. only the vector) grow into blue colonies.

The image below shows the result of a blue-white assay.



Solution to Question 23:

Translocation in the Philadelphia chromosome (a reciprocal translocation between chromosomes 9 and 22) is best studied by SKY.

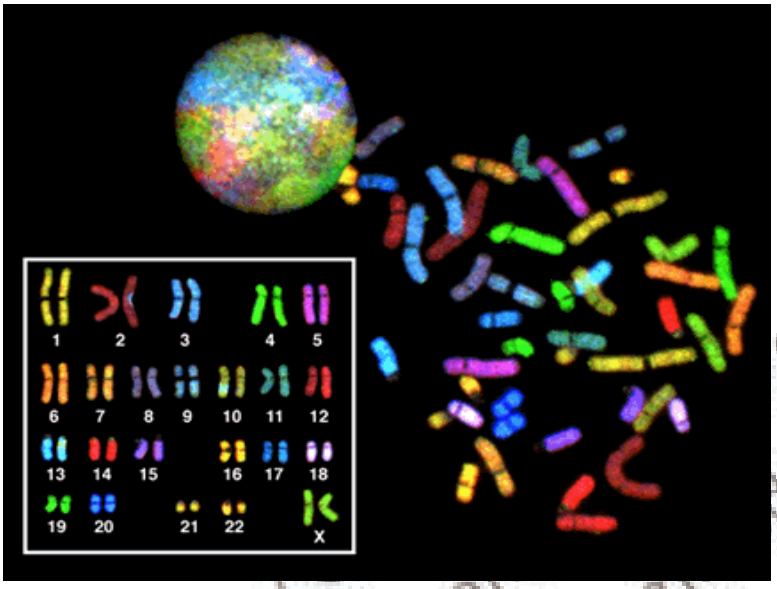
SKY stands for spectral karyotyping or multicolor FISH (M Fish). This technique allows one to display all the chromosomes together and thus help identify chromosomal defects in conditions such as chronic myeloid leukemia.

Other options:

Option A: G banding is also a karyotyping technique but SKY is a better option between the two.

Option C: Southern blotting is a technique used to detect a specific DNA sequence in a tissue sample.

Option D: DNA sequencing cannot detect structural abnormality in a chromosome.



Solution to Question 24:

DNA microarray can be used to detect DNA sequence variations.

There are various microarray techniques used in clinical practice which are described below:

DNA Microarray technique or DNA chip:

• To the DNA chip containing known oligonucleotide sequence, flourescent labelled unknown oligonucleotide is added.

• The computerised algorithms can decode the unknown oligonucleotide, by detecting the location of flourescent hybridization pattern on the chip.

• This technique is used for genotyping or genome sequencing.

RNA microarray: RNA microarray can detect gene expression.

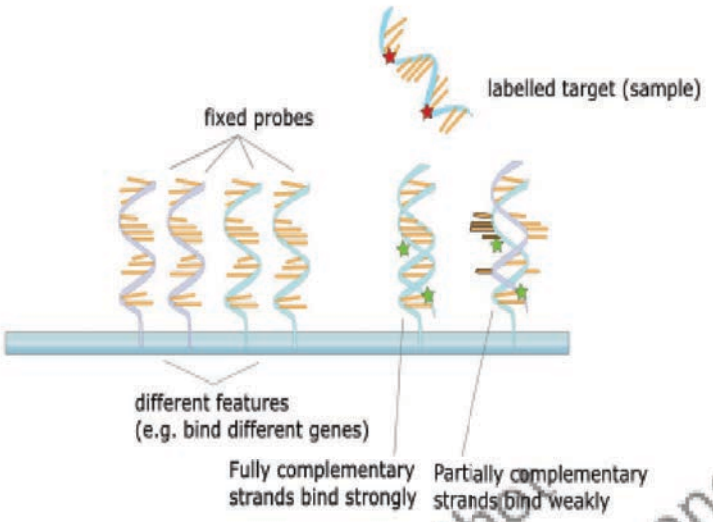
• To the known array of oligonucleotide, flourecently labelled cDNA prepared from unknown mRNA is added, and the unknown cDNA is decoded

• This technique is used for gene expression studies

Protein microarray:

• Known antibodies placed on glass slide and flourescently tagged target protein added. Target protein is detected by antigen antibody interaction.

• This technique is used in study of proteomics



Solution to Question 25:

Array CGH cannot detect balanced translocation.

In Array-based CGH, two genomes are compared. The test DNA and the reference DNA are labelled with two different flourescent dyes (red and green respectively) and are added to a DNA chip spotted with entire human genome.

• If the contribution of both sample are equal for a given chromosomal region, then all spots on the array will flouresce yellow.

• If the test sample has amplification, that spot will fluoresce red

• If the test sample has a deletion, that spot will fluoresce green

Uses of Array CGH:

• To detect gene amplification

• To detect gene deletion

• To detect copy number variations

Solution to Question 26:

The correct sequence of events for PCR among the given options is denaturation, annealing, and extension.

The process of PCR is as follows:

The DNA sample is first heat-denatured at >90°C (90-96 degrees Celsius for 3 minutes) to separate the two strands of the template DNA containing the target sequence; the primers (which attach to the 3' end of the flanking sequence) are added in excess, are allowed to anneal to the

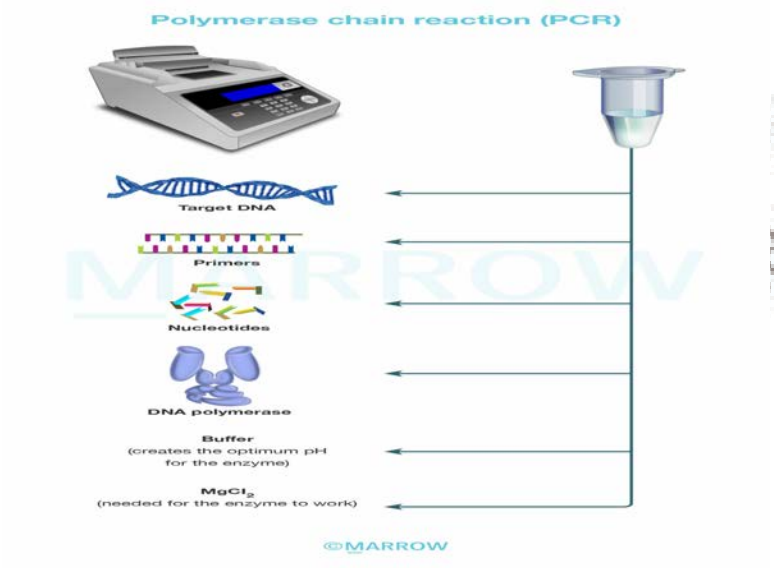
DNA (typically at 50-75°C) in order to generate the required template-primer hybridization complex. Under suitable conditions and presence of deoxynucleotides, elongation of the DNA stand occurs.

The number of copies produced in the presence of an unlimited supply of deoxynucleotides and enzymes is given by - 2 to the power n (where n is the number of PCR cycles)

• 20 cycles provide an amplification of 220 = 106 or 1 million copies

• 30 cycles provides an amplification of 230 =109 or 1 billion copies

Below is an image showing the various ingredients for carrying out PCR.



Solution to Question 27:

The wobble hypothesis explains the degeneracy of the genetic code.

The wobble hypothesis states that codon-anticodon pairing follows the traditional Watson–Crick rules (G pairs with C and A pairs with U) for the first two bases of the codon but can be less stringent for the third/last base. It has been observed that a tRNA can recognize more than one codon for a specific amino acid, especially in the last nucleotide of the codon triplet. This is referred to as degeneracy and its mechanism can be explained by the wobble hypothesis.

The first base of the anticodon moves to permit nontraditional pairing with the last base of the codon. This movement is called wobble and is possible because the first base of the anticodon is not as spatially defined as the other two bases. Due to the wobbling, 61 tRNA species are not required to read the 61 codons.

For example, three codons for glycine—GGU, GGC, and GGA—can form a base pair from one anticodon, 3′ CCI 5′ ie, I, inosine, can base pair with U, C, and A, as shown in the diagram below.

Other options:

Option A: The genetic code is universal because it is the same for all living organisms.

Option B: The genetic code is unambiguous because each codon is specific for only one amino acid. Example: GGA only codes for Glycine.

Option D: The genetic code is non-punctuated. This means that once the reading commences at a specific start codon, it is continuous and is read continuously as triplets until a stop codon is reached without stopping.