VU Medical Zone

BIO 303 Final term Subjective

2 Marks

Qno1: Why Pure water carries electric current?

Pure water carries electrical current as H_3O^+ migrates toward the cathode and OH^- toward the anode.

Qno2: What is activation energy reflect the rate of reaction?

The rate of a reaction reflects this activation energy: higher activation energy corresponds to a slower reaction.

Qno3: Write down the function of cofactor in dehydrogenase coenzyme A?

The function of cofactor in dehydrogenase coenzyme A is accepts acetyl group from Lipoamide.

Qno4: What is Half reaction?

A half reaction is either the oxidation or reduction reaction component of a redox reaction. A half reaction is obtained by considering the change in oxidation states of individual substances involved in the redox reaction. A half reaction is obtained by considering the change in oxidation states of individual substances involved in the redox reaction.

Qno5: What is meant by Activation energy?

The difference between the energy levels of the ground state and the transition state is the activation energy.

Qno6: Define Bioenergetics.

Bioenergetics is the quantitative study of the energy transductions, that is.. changes of one form of energy into another—that occurs in living systems, and of the nature and function of the chemical processes underlying these transductions.

Qno7: Define Hyperchromicity of denaturation of DNA.

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Concomitant with this denaturation of the DNA molecule is an increase in the optical absorbance of the purine and pyrimidine bases—a phenomenon referred to as hyperchromicity of denaturation.

Qno8: What are isozymes?

Two or more enzymes that catalyze the same reaction but are encoded by different genes are called isozymes.

Qno9: What is Ionization?

Although many of the solvent properties of water can be explained in terms of the uncharged H2O molecule, the small degree of ionization of water into hydrogen ions (H+) and hydroxide ions (OH-) must also be taken into account.

3 Marks

Qno1: Define Upstream Processing.

Upstream processing includes formulation of the fermentation medium, sterilisation of air, fermentation medium and the fermenter, inoculum preparation and inoculation of the medium.

The fermentation medium should contain an energy source, a carbon source, a nitrogen source and micronutrients required for the growth of the microorganism along with water and oxygen, if necessary.

Qno2: Which ions are formed due to ionization of water and what happened to H^+ ion?

Water molecules have a slight tendency to undergo reversible ionization to yield a hydrogen ion (a proton) and a hydroxide ion, giving the equilibrium reaction. Hydrogen ions formed in water are immediately hydrated to hydronium ions (H_3O^+).

Qno3: What are competitive inhibitor and non competitive inhibitor?

Competitive inhibitors resemble the substrate and compete for binding to the active site of the enzyme.

Noncompetitive inhibitors do not bind at the active site. They bind either free enzyme at a site other than active site or the ES complex.

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5 marks

Qno1: What is pH? Describe the pH with respect to water at 25 degree centigrade?

The pH of a solution is defined as the logarithm to the base 10 of the reciprocal of the [H+], i. e, the negative logarithm of the [H+].

pH = log 1/ [H+]

pH = -log [H+]

The pH of water at 25°C, in which H+ and OH– ions are present in equal numbers, is 7.0

pH = -log[1 x 10-7]= 7

Qno2: What is competitive inhibitor and write down the affect on Vmax.

Competitive inhibitors resemble the substrate and compete for binding to the active site of the enzyme.

1. Effect on Vmax

• The effect of a competitive inhibitor is reversed by increasing [S].

• At a sufficiently high substrate concentration, the reaction velocity reaches the Vmax as observed in the absence of inhibitor.

• When [S] far exceeds [I], the probability that an inhibitor molecule will bind to the enzyme is minimized and the reaction exhibits a normal Vmax.

• Therefore, a competitive inhibitor does not decrease Vmax.

Qno3: Explain binding site of ligand.

A ligand may be any kind of molecule, including another protein.

• A ligand binds at a site on the protein called the binding site,

• binding site is complementary to the ligand in

• size,

• shape,

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• charge, and

• Hydrophobic or hydrophilic character.

Qno4: Explain Ligand.

In biochemistry and pharmacology, a ligand (from the Latin ligandum, binding) is a substance (usually a small molecule), that forms a complex with a biomolecule to serve a biological purpose.

In a narrower sense, it is a signal triggering molecule, binding to a site on a target protein. A molecule bound reversibly by a protein is called a ligand. Ligands include substrates, inhibitors, activators, and neurotransmitters.

Qno5: Explain oxidation of pyruvate and name three catabolic routes of glycolysis.

1. Oxidation of Pyruvate:

Pyruvate is oxidized, with loss of its carboxyl group as CO_2, to yield the acetyl group of acetyl-coenzyme A; the acetyl group is then oxidized completely to CO_2 by the citric acid cycle. The electrons from these oxidations are passed to O_2 through a chain of carriers in mitochondria, to form H_2O. The energy from the electron-transfer reactions drives the synthesis of ATP in mitochondria

1. Oxidation of Pyruvate

2. Reduction to lactate

3. Ethanol (alcohol) fermentation

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10 Marks

Qno1: Write down the steps of citric acid cycle with name its enzyme in each step?

Formation of Citrate:

• The first reaction of the cycle is the condensation of acetyl-CoA with oxaloacetate to form citrate, catalyzed by citrate synthase

• The methyl carbon of the acetyl group is joined to the carbonyl group (C-2) of oxaloacetate.

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• Citroyl-CoA is a transient intermediate formed on the active site of the enzyme …It rapidly undergoes hydrolysis to release CoA and citrate

• The hydrolysis of this high-energy thioester intermediate makes the forward reaction highly exergonic. • The CoA liberated in this reaction is recycled to participate in the oxidative decarboxylation of another molecule of pyruvate by the PDH complex.

2. Isomerization of Citrate by Aconitase

• The enzyme aconitase (aconitate hydratase) catalyzes the reversible transformation of citrate to isocitrate

• Reaction Intermediate of the tricarboxylic acid is formed..known as cis-aconitate

• Cis-aconitate does not dissociate from the active site.

• Aconitase can promote the reversible addition of H2O to the double bond of enzyme-bound cis-aconitate in two different ways, one leading to citrate and the other to isocitrate.

• Under physiological conditions in the cell.. Conc. Of isocitrate is very little , the reaction is pulled to the right because isocitrate is rapidly consumed in the next step of the cycle.

3. The Isocitrate Dehydrogenase Reaction

• Oxidation of Isocitrate to a-Ketoglutarate and CO2 In the next step

• isocitrate dehydrogenase catalyzes oxidative decarboxylation of isocitrate to form aketoglutarate

• Mn2+ in the active site interacts with the carbonyl group of the intermediate oxalosuccinate, which is formed transiently but does not leave the binding site until decarboxylation converts it to a-ketoglutarate.

• Mn2+ also stabilizes the enol formed transiently by decarboxylation.

4. Oxidation of α-ketoglutarate:

• The next step is another oxidative decarboxylation

• Oxidation of a-Ketoglutarate to Succinyl-CoA and CO2

• a-ketoglutarate is converted to succinyl-CoA and CO2 by the action of the a-ketoglutarate dehydrogenase complex

• NAD+ serves as electron acceptor and CoA as the carrier of the succinyl group.

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• The energy of oxidation of a-ketoglutarate is conserved in the formation of the thioester bond of succinyl-CoA

• This reaction is somehow identical to the pyruvate dehydrogenase reaction and to the reaction sequence responsible for the breakdown of branched chain amino acids

5. Conversion of Succinyl-CoA to Succinate:

• Succinyl-CoA like acetyl-CoA, has a thioester bond with a strongly negative standard free energy of hydrolysis (G= -36 kJ/mol).

• Energy released in the breakage of this bond is used to drive the synthesis of a phosphoanhydride bond in either GTP or ATP, with a net G= - 2.9 kJ/mol.

• The enzyme that catalyzes this reversible reaction is called succinyl-CoA synthetase or succinic thiokinase, names indicate the participation of a nucleoside triphosphate in the reaction.

6. Oxidation of Succinate to Fumarate:

• Succinate Dehydrogenase reaction

• The succinate formed from succinyl-CoA is oxidized to fumarate by the flavoprotein succinate dehydrogenase

• In eukaryotes, succinate dehydrogenase is tightly bound to the mitochondrial inner membrane; in bacteria, to the plasma membrane.

• The enzyme contains three different iron-sulfur clusters and one molecule of covalently bound FAD

• Electron flow from succinate through these carriers to the final electron acceptor, O2, is coupled to the synthesis of about 1.5 ATP molecules per pair of electrons (respirationlinked phosphorylation).

7. Hydration of Fumarate to Malate:

• The reversible hydration of fumarate to L-malate is catalyzed by fumarase (fumarate hydratase).

• The transition state in this reaction is a carbanion

• This enzyme is highly stereospecific; it catalyzes hydration of the trans double bond of fumarate but not the cis double bond of maleate (the cis isomer of fumarate).

• In the reverse direction (from L-malate to fumarate), fumarase is equally stereospecific: D-malate is not a substrate.

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8. Oxidation of Malate to Oxaloacetate:

• Oxidation of Malate to Oxaloacetate Is the last reaction of the citric acid cycle

• NAD-linked L-malate dehydrogenase catalyzes the oxidation of L-malate to oxaloacetate

• In intact cells oxaloacetate is continually removed by the highly exergonic citrate synthase reaction

• This keeps the concentration of oxaloacetate in the cell remains extremely low pulling the malate dehydrogenase reaction toward the formation of oxaloacetate.

Qno2: Describe the mechanism of enzyme action and describe how enzyme lowers the activation energy?

Enzymes bind and chemically transform other molecules— they catalyze reactions.

• The molecules acted upon by enzymes are called reaction substrates rather than ligands.

• Enzymes are highly effective catalysts, commonly enhancing reaction rates by a factor of 105 to 1017 . • The distinguishing feature of an enzyme-catalyzed reaction is that it takes place within the confines of a pocket on the enzyme called the active site.

• The surface of the active site is lined with amino acid residues with side chains that complement and bind the substrate and catalyze its chemical transformation.

• Often, the active site encloses a substrate, sequestering it completely from solution.

• The enzyme substrate complex, whose existence is central to the action of enzymes.

When an **enzyme** binds to its substrate, we know it **lowers the activation energy** of the reaction, allowing it to happen more quickly. The **enzyme**-substrate complex can also **lower activation energy** by bending substrate molecules in a way that facilitates bond-breaking, helping to reach the transition state.

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