CHAPTER – 4

Enzymes and Bioenergetics

EXERCISES

2 Mark Questions

Q1: What is relation between vitamins and enzyme cofactors?

Answer:

Vitamins help in many ways in our body for each an every enzymetic activity. There are many enzymes which require cofactors for their catalytic activity. The cofactor which is complex organic molecule is known as co enzyme. Most of the co-enzymes are derived from vitamins. Cofactors of enzyme work with vitamins and play important role in the catalytic reaction.

Q2: What is specific activity of an enzyme?

Answer: Specific activity of enzyme is the activity in which mole of product is formed by an enzyme in a given amount under the given conditions per milligram of proteins. This activity is activity which represents a measurement of enzyme purity in the mixture.

Q3: Define entropy. What is relationship between free energy and entropy.

Answer: Entropy is defined as degree of randomness or disorder of a system and it explains whether reaction takes place or not.

The free energy is the term which is obtained by combining the two laws.

G= H – TS, this equation explains the relationship between the free energy (G) and entropy in chemical reaction at constant temperature (T) and pressure (P).

Q4: Why ATP is called as universal energy currency?

Answer: The living organisms derive free energy from environment. As the photosynthetic organisms take this energy from sunlight and chemotrophs obtain it from oxidation of food stuff. The free energy is used in many processes which are synthesis of macromolecules and precursors, in active transport across the membrane, in muscle contraction and in fidelity of genetic information transfer.

Before these processes the free energy is converted into the Adenosine triphosphate which is known as ATP. This plays an important role in transfer of free energy from exergonic process to endergonic process. Thus ATP is a universal energy currency.

Q5: Explain the role of coenzymes in enzyme function.

Answer: Coenzymes are organic molecules that are not proteins but are essential for the catalytic activity of certain enzymes. They participate directly in the chemical reactions by accepting or donating electrons, protons, or functional groups, facilitating the transformation of the substrate by the enzyme.

Q6: Describe the process of cellular respiration and its main stages.

Answer: Cellular respiration is the process by which cells break down glucose to generate ATP, the primary energy currency of the cell. It occurs in four main stages: glycolysis (occurs in the cytoplasm), pyruvate oxidation (occurs in the mitochondria), Krebs cycle (also in the mitochondria), and electron transport phosphorylation (also in the mitochondria). Each stage involves specific enzymes and produces ATP or intermediate molecules that participate in further ATP generation.

4 Mark Questions

Q1: Briefly describe first and second law of thermodynamics.

Answer: 1. First law of thermodynamics: According to first law of thermodynamics the energy exchange takes place in between system and surrounding in any process. It states that energy can neither be created nor be destroyed, but it can be converted in to other forms of energy.

$E = EB – EA = Q – W$

The equation states that the change of energy depends only on the initial and final stages of system. It does not depend on the path of transformation.

2. Second law of thermodynamics: The second law of thermodynamics states that entropy or disorder of universe will always be increasing. According to this law, if

the sum of the entropies of system and it's surrounding increases then a process can occur spontaneously.

(S system+ S surroundings) $>$ 0 (for a spontaneous process)

Q2: Define Km and its significance.

Answer: Km is defined as the substrate concentration at which v0=½ Vmax, is increased in presence of inhibitor and denoted by symbol Km.

Significance:

Km substrate concentration in non competitive inhibition has no effect as a inhibitor which decrease amount of functional enzyme. In uncompetitive inhibition it does not inhibits by increasing the substrate concentration. The value is altered.

Q3: What is effect of temperature, pH and substrate concentration on catalytic activity of enzyme?

Answer: There are some important factors which effect the enzyme activity:-

1. Temperature- When the temperature increases the rate of an enzyme catalysed reaction increases. It increases up to a maximum and them fall. The graph is plotted between temperature and enzyme activity, the bell shaped curve is obtained. There is optimum temperature which at maximum rate of reaction occurs. It is different for different enzymes. Most of the enzyme are in between 40°c to 45°c.
2. pH- Each enzyme has its optimum unique pH. The graph plotted between pH and enzyme activity which results in bell shaped curve. At optimum pH the rate of reaction is greatest. The optimum pH is which is maximum at the activity of enzyme. Many of the enzymes show optimum rate of reaction at neutral pH.
3. Substrate concentration- This is the factor which influence the enzyme activity. As the concentration of substrate increases the rate of reaction increases. It is because of the interaction of substrate molecules with the enzyme molecule.

Q4: Compare and contrast the two main pathways for ATP generation in cellular respiration: glycolysis and oxidative phosphorylation. Discuss the key steps and energy yield in each pathway.

Answer: While both glycolysis and oxidative phosphorylation contribute to ATP production, they differ significantly in their location, mechanisms, and efficiency:

Glycolysis:

- Occurs in the cytoplasm without requiring oxygen.
- Breaks down glucose molecule into two pyruvate molecules via a series of enzyme-catalyzed reactions.
- Generates a net of 2 ATP molecules and 2 NADH molecules per glucose molecule.
- Relatively quick but less efficient in ATP production.

Oxidative phosphorylation:

- Takes place in the mitochondria and requires oxygen as the final electron acceptor.
- Pyruvate molecules from glycolysis enter the Krebs cycle, generating NADH and FADH2 molecules.
- Electrons from NADH and FADH2 enter the electron transport chain, transferring energy to pump protons across the mitochondrial membrane.
- Proton gradient created by the electron transport chain drives ATP synthesis through ATP synthase.
- Generates around 36-38 ATP molecules per glucose molecule, much more efficient than glycolysis.

7 Mark Questions

Q1: A newly discovered enzyme exhibits maximal activity at 40°C and pH 7.0. However, its activity significantly decreases at higher temperatures and acidic pH. Design an experiment to determine the Km and Vmax of this enzyme for its substrate. Explain the rationale behind each step of your experiment.

Answer: Experiment Design:

1. Prepare a series of reaction mixtures: Vary the substrate concentration in multiple reaction tubes while maintaining other factors like temperature and pH at optimal conditions (40°C and pH 7.0).

2. Monitor the initial reaction rate: Measure the change in product concentration or any other suitable indicator of reaction progress over a short time interval (initial linear phase) for each reaction mixture.
3. Plot a Michaelis-Menten graph: Create a graph with substrate concentration on the x-axis and initial reaction rate on the y-axis.
4. Analyze the graph: Determine the Km and Vmax from the graph. Km is the substrate concentration at half the Vmax, and Vmax is the maximum reaction rate when all active sites are saturated with the substrate.

Rationale:

- By varying the substrate concentration, we can observe how the enzyme's activity changes based on substrate availability.
- Measuring the initial reaction rate avoids complications from product inhibition or enzyme denaturation at later stages.
- The Michaelis-Menten graph provides a visual representation of the relationship between substrate concentration and reaction rate, allowing for easy calculation of Km and Vmax.
- Understanding Km and Vmax allows for further characterization of the enzyme, such as its substrate affinity and catalytic efficiency.

Q2: Explain the concept of enzyme regulation in metabolic pathways. Discuss two different mechanisms used by cells to regulate enzyme activity and their significance in maintaining cellular homeostasis.

Answer: Enzyme regulation is crucial for controlling metabolic pathways and maintaining cellular homeostasis. Cells utilize various mechanisms to fine-tune enzyme activity based on their needs and environmental conditions.

Two common mechanisms:

1. Allosteric regulation:

- Regulatory molecules (activators or inhibitors) bind to specific allosteric sites on the enzyme, causing conformational changes that affect the active site and alter its catalytic activity.

- Example: ATP as an inhibitor for phosphofructokinase in glycolysis, slowing down the pathway when energy levels are sufficient.

2. Covalent modification:

- Specific enzymes can be modified by phosphorylation or dephosphorylation, adding or removing phosphate groups, respectively. This alters the enzyme's structure and activity.
- Example: Protein kinase A phosphorylating and activating glycogen synthase in response to glucagon signaling, promoting glycogen synthesis when glucose levels are high.

Significance of enzyme regulation:

- Ensures efficient utilization of cellular resources by only activating pathways as needed.
- Maintains a balance between anabolic and catabolic processes, preventing energy waste or depletion.
- Allows for rapid adaptation to environmental changes, such as nutrient availability or stress conditions.

By understanding these mechanisms, researchers can develop drugs and therapies targeting specific enzymes for treating various diseases or metabolic disorders.

Q3: Discuss the role of microorganisms in environmental bioremediation and their potential applications in cleaning up polluted environments. Explain the types of metabolic pathways involved in bioremediation processes.

Answer: Microorganisms play a vital role in bioremediation, the natural process of using living organisms to degrade and remove pollutants from the environment. Their diverse metabolic capabilities allow them to break down contaminants like:

- Hydrocarbons: Oil spills and contaminated soil can be remediated by bacteria with alkane-degrading pathways.
- Heavy metals: Certain bacteria can accumulate and detoxify heavy metals like mercury or chromium.

- Pesticides and herbicides: Microbial enzymes can degrade these agricultural chemicals, preventing their persistence in the environment.

Types of metabolic pathways:

- Aerobic biodegradation: Requires oxygen for the complete mineralization of pollutants into carbon dioxide, water, and inorganic minerals.
- Anaerobic biodegradation: Occurs in the absence of oxygen, often resulting in methane and other organic compounds as end products.
- Cometabolism: Pollutants are degraded indirectly as byproducts of the microorganism's primary metabolic pathway.

Applications of bioremediation:

- Cleaning up oil spills and contaminated sites.
- Treating wastewater from industrial and agricultural activities.
- Removing persistent organic pollutants (POPs) from soil and water.
- Developing eco-friendly methods for waste management and resource recovery.

Fill in the Blanks

1. The three-dimensional structure of the enzyme that allows it to bind to its specific substrate is called the _____.

Answer: active site

2. The process by which enzymes increase the rate of chemical reactions without being consumed themselves is called _____.

Answer: catalysis

3. Coenzymes are organic molecules that bind to enzymes and participate in the chemical reactions by accepting or donating _____.

Answer: electrons, protons, or functional groups

4. The process of cellular respiration that occurs in the mitochondria and requires oxygen is called _____.

Answer: oxidative phosphorylation

5. The molecule that carries protons across the inner mitochondrial membrane during oxidative phosphorylation is _____.

Answer: ATP synthase

6. The two main products of glycolysis are _____ and _____.

Answer: pyruvate and NADH

7. The electron transport chain in oxidative phosphorylation is located in the _____ of the mitochondria.

Answer: inner membrane

8. Competitive inhibition occurs when a molecule similar to the substrate binds to the _____ of the enzyme.

Answer: active site

9. Non-competitive inhibition alters the _____ of the enzyme, reducing its catalytic activity.

Answer: conformation

10. The process by which microorganisms break down pollutants in the environment is called _____.

Answer: bioremediation

Multiple Choice Questions

1. Which molecule provides the shape and specificity for an enzyme's active site?

(a) Substrate

(b) Cofactor

(c) Apoenzyme

(d) Holoenzyme

Answer: (c) Apoenzyme

2. What happens to an enzyme's activity when the temperature is increased beyond its optimal range?

(a) It increases.

(b) It remains constant.

(c) It decreases.

(d) It becomes irreversible.

Answer: (c) It decreases.

3. Which type of inhibition binds directly to the enzyme's active site?

(a) Competitive inhibition

(b) Non-competitive inhibition

(c) Allosteric inhibition

(d) None of the above

Answer: (a) Competitive inhibition

4. What is the main function of ATP in cellular respiration?

(a) To break down glucose molecules

(b) To transport electrons across the mitochondrial membrane

(c) To store and transfer energy

(d) To accept protons from the electron transport chain

Answer: (c) To store and transfer energy

5. Which of the following is NOT a product of glycolysis?

(a) ATP

(b) NADH

(c) Pyruvate

(d) Oxygen

Answer: (d) Oxygen

6. Which enzyme is responsible for the rate-limiting step in glycolysis?

(a) Hexokinase

(b) Phosphofructokinase

(c) Pyruvate kinase

(d) Lactate dehydrogenase

Answer: (b) Phosphofructokinase

SUMMARY:

This chapter delvers into the fascinating world of enzymes and bioenergetics, understanding how these crucial elements powers the machinery of life:

Enzymes:

- Proteins with a specific three-dimensional structure that acts as a catalyst, speeding up biochemical reactions without being consumed themselves.
- Possess an active site that perfectly fits the substrate molecule, facilitating the reaction.
- Activity influenced by various factors like temperature, pH, substrate concentration, and inhibitors.
- Examples: Hexokinase in glycolysis, Pepsin in digestion, DNA polymerase in replication.

Bioenergetics:

- Deals with the energy transformation and utilization in living organisms.
- Focuses on cellular respiration, the process by which cells break down glucose to generate ATP, the primary energy currency of the cell.
- Divided into two main stages:
  - Glycolysis: Occurs in the cytoplasm, generates 2 ATP and 2 NADH per glucose molecule.
  - Oxidative phosphorylation: Takes place in the mitochondria, utilizes oxygen to produce around 36-38 ATP per glucose molecule through the electron transport chain and ATP synthase.

Additional highlights:

- Understanding enzyme regulation is key for maintaining cellular homeostasis. Mechanisms like allosteric and covalent modification control enzyme activity based on cellular needs.
- Microorganisms play a vital role in bioremediation, using their diverse metabolism to degrade pollutants like hydrocarbons and heavy metals from the environment.