Jordan University of Science & Technology | BT232 | Department of Biotech. & Genetic Engineering

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Summer 2018
Midterm Exam

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Question 1 (20 points): Choose the most appropriate answer. Use the attached Scranton sheet to answer this question

1. Combining DNA from different sources is an example of:

A) genomics
B) bioremediation
C) recombinant DNA technology
D) nanotechnology

2. Scientists at the biotechnology company Genentech created the first recombinant DNA product for use in humans. Approved by the Food and Drug Administration in 1982, this product is ___

A) human growth hormone
B) insulin
C) gene chip
D) chymosin

3. Selective breeding involves:

A) genetic engineering of animals and plants to improve growth characteristics
B) mating organisms with desirable characteristics
C) the use of fermentation to produce biotechnology products
D) combining sperm and egg cells from different species to produce hybrid organism

4. _____ is the enzyme that copies DNA during DNA replication.

A) RNA polymerase
B) DNA polymerase
C) DNA ligase
D) RNA primase

5. _____ bind to mRNA and tRNA during translation and allow for polypeptides to be synthesized.

A) ribosomes
B) chromosomes
C) RNA polymerases
D) nuclei

6. Which of the following is a structural feature of DNA but not RNA?

A) single-stranded
B) containing phosphate groups
C) containing the nitrogenous base adenine
D) containing deoxyribose sugars

7. Reverse transcriptase catalyzes the production of

A) DNA from an mRNA template
B) mRNA from a DNA template
C) DNA from a protein template
D) tRNA from a DNA template

8. In prokaryotic organisms

A) DNA is only found in plasmids
B) transcription occurs in the nucleus
C) chromosomes are usually linear
D) translation occurs at the ribosome

9. Which of the following enzymes allows scientists to join together two DNA fragments?

A) DNA polymerase
B) restriction nuclease
C) DNA ligase
D) DNA helicase

10. Which type of point mutation creates a stop codon in a gene?

A) nonsense
B) frameshift
C) silent
D) missense

11. Which of the following is NOT used during the Roche 454 next generation pyrosequencing

A) DNA polymerase
B) Dideoxy-dNTPs (ddNTPs)
C) luciferase
D) primers

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Question 1 (Continued)

12. Which of the following vectors can hold the largest DNA insert?

(A) YAC
B) cosmid
C) Bacteriophage vectors
D) BAC

13. Which of the following is most necessary to make a cDNA library?

A) DNA polymerase
(B) reverese transcriptase
C) restriction enzymes
D) ribosome

14. Which of the following is an INCORRECT statement about restriction enzymes?

A) most restriction enzymes are isolated from bacteria
B) restriction enzymes usually recognize palindromic sequences
(C) restriction enzymes create phosphodiester bonds between pieces of dna in a cloning experiment
D) restriction enzymes can cut to create overlapping single-stranded ends of dna

15. _____ is a DNA-binding dye that fluoresces when DNA in an agarose gel is illuminated with ultraviolet light.

A) lactic acid
(B) ethidium bromide
C) IPTG
D) x-gal

16. Which of the following is true regarding TA cloning?

(A) used to clone TA-rich DNA only
B) the linear vector that has single stranded uracil on each end
C) Taq polymerase puts a single adenine nucleotide on the 3' end of all PCR products
D) uses YAC vectors

17. Transformation in a cloning experiment is _____.

A) sing PCR to clone a gene
(B) inserting DNA into bacteria cells
C) ligating pieces of foreign DNA together
D) cutting DNA with restriction enzymes

18. Which of the following is true regarding directed molecular evolution technology?

A) uses PCR to clone a gene
B) uses site-directed mutagenesis to introduce mutations into genes
(C) introduces specific, predefined alterations in the amino acid sequence
D) introduces chemical modification on histones and DNA

19. During downstream processing, proteins are stabilized in solution by which of the following?

A) addition of protease inhibitors
B) addition of anti-foaming agents
C) purifications occur at low temperatures
(D) all of the these

20. _____ is used to determine protein structure and _____ is used for protein sequencing

(A) X-ray crystallography; mass spectrometry
B) mass spectrometry; SDS-PAGE
C) mass spectrometry; protein microarray
D) X-ray crystallography; isoelectric focusing

21. Which of the following vectors is used for the high-level synthesis of eukaryotic proteins within bacterial cells?

A) YAC
(B) expression vectors
C) Ti plasmid
(D) BAC

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Question 1 (Continued)

22. A linear strand of DNA is 1,000 bp long. A recognition sequence for the restriction enzyme Eco R1 is located 300 base pairs (bp) from the 5′ end of this linear DNA molecule. Digesting this DNA molecule with Eco RI would produce _____.

A) one DNA fragment, 1,000 bp long
B) three DNA fragments, two of them 300 bp long and one 400 bp long
(C) two DNA fragments, one 300 bp long and one 700 bp long
D) two fragments, each 500 bp long

23. Approximately how many genes are present in the human genome?

A) 3 billion genes
B) 20 billion genes
(C) 20,000 genes
D) 100,000 genes

24 A _____ consists of cloned DNA fragments for all expressed genes in a particular tissue

A) genomic DNA library
(B) cDNA library
C) PCR library
D) mRNA library

25. Which of the following techniques is the best choice for amplifying DNA?

A) affinity chromatography
B) Southern blot
C) microarray analysis
(D) PCR

26. Which of the following techniques is most commonly used to separate and analyze DNA by size?

A) DNA libraries
B) Northern blot
(C) agarose gel electrophoresis
D) PCR

27. During molecular cloning, a gene of interest (insulin) is inserted into a bacterial structure called a _____ and enters a bacterial cell through a process called _____.

A) chromosome; electrophoresis
B) plasmid; transcription
(C) plasmid; transformation
D) nucleus; transformation

28. During library screening, PCR, Southern blotting, and other techniques, binding two pieces of DNA to each other by hydrogen bonding is called _____.

(A) hybridization
B) DNA ligation
C) autoradiography
D) polyadenylation

29. 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

30. Studying proteins and enzymatic pathways involved in cell metabolism

A) metagenomics
B) glycomics
(C) metabolomics
D) transcriptomics

31. All of the following are pharmaceutical products produced as recombinant proteins <u>except</u>

A) antibodies
B) human growth factor
C) interferons
(D) cellulase

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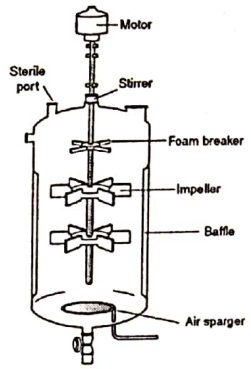
Question 1 (Continued)

32. Which of the following is NOT true regarding the device shown on the right?



A) it is called a bioreactor
B) used to grow large quantities of biological cells
C) computers monitor the environment inside the device to keep oxygen levels and temperature ideal for cell growth
(D) used for downstream processing

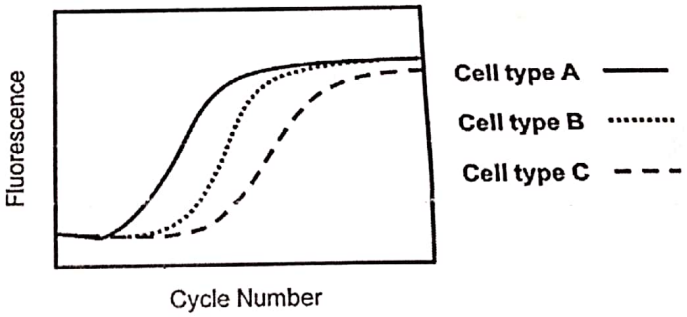
33. RT-PCR is a method that is used for:

(A) forensic analysis of DNA
B) amplification of genomic DNA sequences
C) analysis of mRNA expression
D) amplification of mRNA sequences

34. Protein glycosylation

A) can increase proteins solubility
B) orient proteins into membranes
C) extend the active life of proteins
(D) all of these are correct

35. The graph on the right shows the results from a real-time PCR experiment done to study the expression of a gene in 3 different cell types (A, B and C). It can be concluded from the figure that:



A) the gene is not expressed in any cell type
(B) highest expression of the gene is found in cell type A
C) highest expression of the gene is found in cell type B
D) highest expression of the gene is found in cell type C

36. All of the following techniques separate proteins from each other based on their charge except

A) ion-exchange chromatography
(B) affinity chromatography
C) isoelectric focusing
D) A & C

37. All of the following techniques require the use of electrophoresis to separate molecules except

(A) Southern blot
B) Sanger Sequencing (dideoxy chain termination)
C) SDS-PAGE
D) microarray

38. All of the following techniques can be used to study gene expression except

(A) Northern blot
B) FISH
C) real-time PCR
D) microarray

39. All of the following techniques separate different proteins from each other based on their size except

A) size-exclusion chromatography
(B) affinity chromatography
C) dialysis
D) ultrafiltration

40. Which of the following sequences is most commonly found at eukaryotic promoters?

(A) TATAAT
B) Poly(A) tail
C) 7-methyl G cap
D) 5′-GU . . . AG-3′

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Question 2 (10 points): Choose from the following list to fill in the blank. Some terms might not be used at all.

~~Epigenome~~	Gene	~~Alternative splicing~~	~~Inclusion bodies~~
~~Lipases~~	~~RNA interference (RNAi)~~	~~Model organisms~~	~~Plaques~~
~~Karyotyping~~	~~2D electrophoresis~~	~~Primers~~	~~Probes~~
~~Paleogenomics~~	Biomarker protein	~~Fusion protein~~	~~Anticodon~~
~~Glycosylation~~	~~Cohesive (sticky) ends~~	Genetic engineering	~~Restriction map~~
Metagenomics	~~Fermenters~~	~~Lyophilization~~	Chromosome

1. Model organisms are Non-human organisms that scientists use to study biologic processes in experimental laboratory conditions.
2. The process of altering an organism's DNA is called Genetic engineering
3. Fermenters are large containers used for growing cultures of microorganisms or mammalian cells in a batch process.
4. The sequence of nucleotides that provides cells with the instructions to synthesize a specific protein or a particular type of RNA is called a Primers.
5. Karyotyping is a laboratory procedure for analyzing the number and structure of chromosomes in a cell.
6. Alternative splicing allows several different protein products to be produced from the same gene sequence.
7. The three-nucleotide sequence at the end of a tRNA molecule that binds to a specific codon in an mRNA is called Anticodon.
8. Epigenome consists of all chemical modifications in an organism's genome that are not due to mutations in DNA sequence.
9. Cohesive (sticky) end are overhanging single-stranded ends of a DNA molecule created by the action of certain restriction enzymes.
10. Plaques are small clear spots of dead bacteria appearing on a culture plate, caused by bacterial cell lysis by bacteriophage.
11. Probes are short oligonucleotides complementary to specific sequences of interest; used in PCR reactions to amplify DNA and DNA-sequencing reactions.
12. The analysis of ancient DNA such as DNA from fossils is called Paleogenomics
13. Restriction map is an arrangement of the number, order, and types of restriction-enzyme cutting sites in a DNA molecule.
14. RNA interference (RNAi) is a RNA-based mechanism of gene silencing involves the formation of protein–RNA complex called the RNA-induced silencing complex (RISC).
15. Glycosylation is a natural process of adding sugar units to proteins by complex cells.
16. Fusion Protein is a "hybrid" recombinant protein consisting of a protein from a gene of interest connected (fused) to another, well-known protein that serves as a tag for isolating recombinant proteins..
17. Inclusion bodies are made of expressed foreign proteins that concentrate in an insoluble form inside transformed cell.
18. The process of freeze-drying proteins is called Lyophilization.
19. 2D electrophoresis is a technique used to separate proteins based on their electrical charge and size.
20. Fat-digesting enzymes are called Lipases

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Question 3 (3 points):
List 3 advantages and 3 disadvantages of using *E coli* for the production of recombinant proteins.

Advantage	Disadvantage
it's genetics well Known	can't Folded protein as in eukaryotic
it's reprduction processes well known	most of protein inactive insid bacteria

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Question 4 (2 points):
Write down 4 basic concepts scientists have learned from the Human Genome Project:

1: not all genome are coding protein

2: different protiens ar produce from the same gene sequence

3: ✗

4: ✗

(1)

Question 5 (6 points):
Write the correct order of steps (Left to right) for the following techniques/procedures (3.25 points)

Genomic library preparation (1.5)

A) Inserting DNA fragments into plasmids by DNA ligase
B) Introduction of plasmids into bacteria
C) Genomic DNA digestion with restriction enzyme

Correct order is C, A, **and** B ✓

Southern blotting (2.5)

A) Filter (blot) is baked or exposed to UV light to permanently attach the DNA
B) Gel is treated with alkaline solution to denature the DNA
C) Fragments are transferred onto a nylon or nitrocellulose filter (called blotting)
D) DNA fragments are separated by agarose gel electrophoresis
E) Filter (blot) is incubated with a labeled probe and exposed to film by autoradiography

Correct order is ✗, ✗, ✗, ✗, **and** ✗

4.5

Basic Steps in protein purification (2)

A) Concentration and initial purification
B) Purification by chromatography techniques
C) Stabilization of final product and adjustment of activity to required level
D) Extraction of crude protein (cell lysis)

Correct order is D, A, B, **and** C

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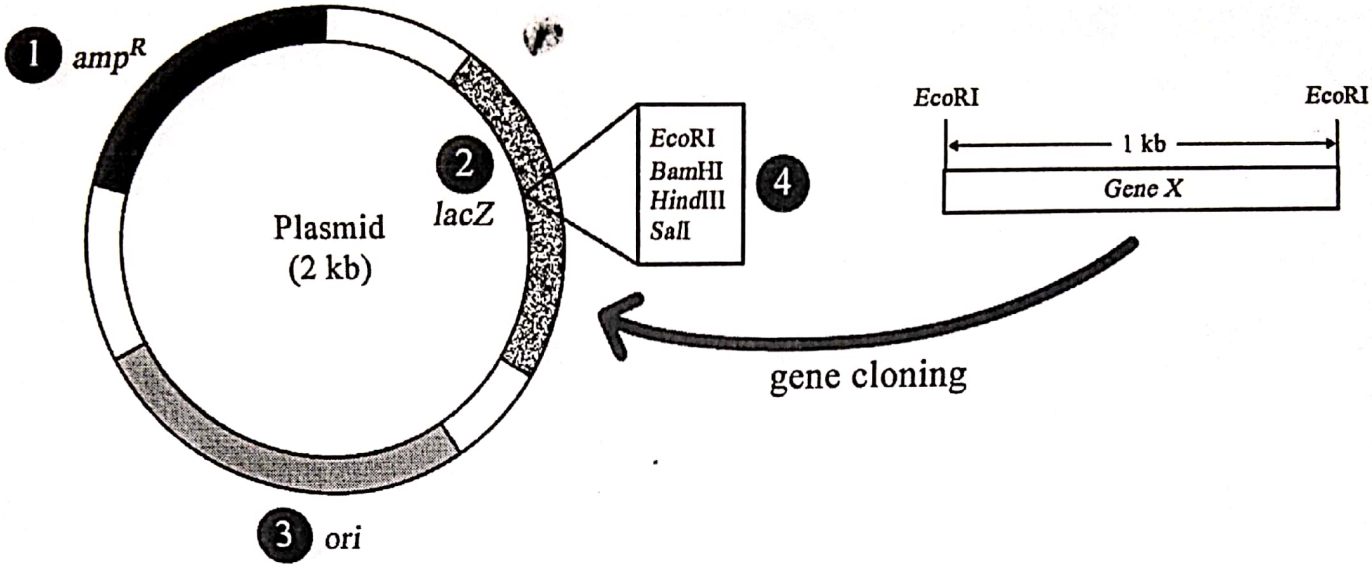
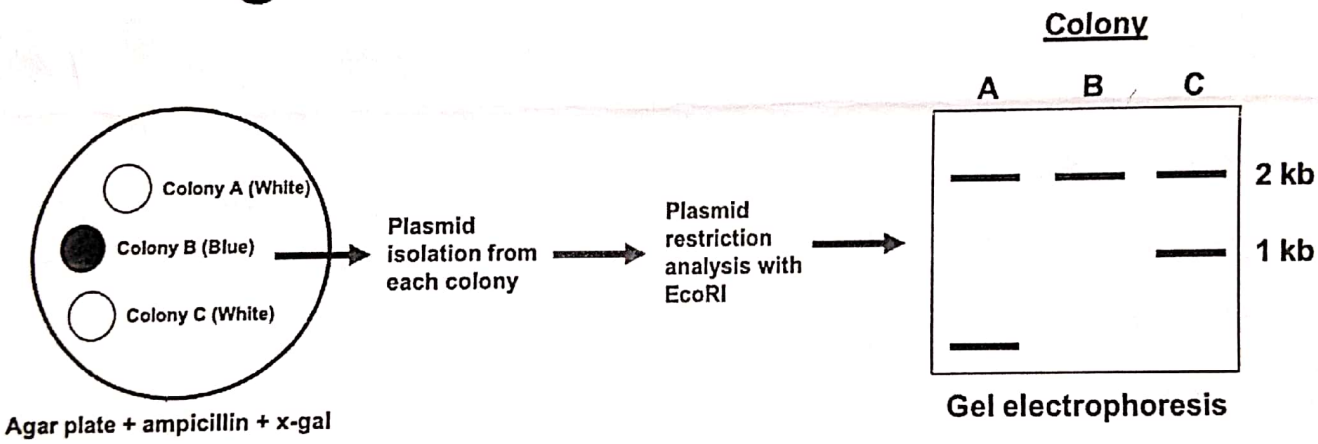
Question 6 (3 points):

A 1 kb DNA fragment containing gene X was cloned into the plasmid (size is 2 kb) below. After cloning, the resulting plasmid was transformed into *E.coli*. After transformation, three colonies were obtained on agar plate containing the antibiotic ampicillin and x-gal, the substrate for the enzyme Beta-galactosidase (LacZ).

The plasmids from the 3 colonies were isolated, each plasmid was digested with *Eco*RI and the products were run on gel electrophoresis. After taking a photo for the gel, the gel showed the plasmid banding patterns shown below.





A. On the plasmid, region number 4 (1, 2, 3, or 4) represents the multiple cloning site.

B. On the plasmid, region number 2 (1, 2, 3, or 4) represents selectable marker gene.

C. On the plasmid, region number 3 is important for replication

D. To clone gene X into the plasmid, both gene X and the plasmid must be cut with the restriction enzyme EcoRI

E. X-gal and the enzyme Beta-galactosidase are used for the blue - white selection

F. Plasmid(s) from colony/colonies A and C (A, B and/or C) contain(s) the inserted gene X

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