

School of Bioscience

WRITTEN EXAMINATION

Course: Molecular Biotechnology

Course code: BV703A

Credits for written examination: 4 hp

Date: 2024-01-11

Examination time: 14.15-19.30

Examination responsible: Sanja Jurcevic

Aid at the exam/appendices

- Instructions
- Take a new sheet of paper for each teacher.
 - Write your answer in the exam sheet.
 - Write only on one side of the paper.
 - Write your name and personal ID No. on all pages you hand in.
 - Use page numbering.
 - Don't use a red pen.
 - Mark answered questions with a cross on the cover sheet.

Grade points

Maximum score: 70p

Grades: A \geq 90%, B \geq 80%, C \geq 70%, D \geq 60%, E \geq 50% of the total points.

Examination results should be made public within 18 working days

Good luck!

Total number of pages 4 (excluding this page)



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Question 1

Multiple choice questions (1p for correct answer, 0p for no answer, and -1p for incorrect answer) **(4p)**

What does red biotechnology primarily involve?

- A. Transgenic modification in plants
- B. Environmental diagnostics
- C. Gene therapy
- D. Production of chemicals, mainly enzymes

Which of following is an example of green biotechnology?

- A. Engineering yeast for bread product
- B. Using fermentation in wine-making
- C. Modifying tobacco plants to produce vaccines
- D. Implementing molecular methods for environmental diagnostics

In which sector is white biotechnology primarily applied?

- A. Pharmaceutical industry
- B. Agricultural industry
- C. Environmental industry
- D. Industrial manufacturing

In your course book “An introduction to molecular biotechnology: fundamentals, methods and applications” they describe that the area of biotechnology is divided into four main different types. What are these four types of biotechnology?

- A. Grey, red, black and white biotechnology
- B. Green, white, blue and red biotechnology
- C. Red, green, white and grey biotechnology
- D. Red, black, green and grey biotechnology

Question 2

Explain the role of phenolic compounds like acetosyringone in plant infection by *Agrobacterium* (key words: acetosyringone, VirA receptor, VirG transcription factor, promoters, expression level). **(4p)**



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Question 3

- a) What are the limitations that prevent the direct use of Ti-plasmid as cloning vector, despite their natural capacity to deliver target gene into plant cell? **(3p)**
- b) Because of these limitations, Ti plasmids have to be modified to produce Ti plasmid based vectors. Briefly describe four primary components found in modified Ti plasmid based vectors that are used for plant transformation. **(4p)**
- c) What is the role of the left and right borders in a Ti-plasmid? **(2p)**

Question 4

Match one statement to one explanation below. In your answering sheet it is enough to have number and letter (1p for correct answer, 0p for no answer, and -1p for incorrect answer). **(10p)**

Statement
1. Transgenic mouse
2. Positive selection marker
3. Lentivirus
4. Adenovirus
5. <i>gag</i> gene
6. <i>tat</i> gene
7. Ganciclovir
8. Microinjection
9. Transgenic founders
10. Negative selection marker

Explanation
A) A mouse that carries a foreign gene that has been deliberately integrated into its genome
B) A laboratory mouse in which a specific gene has been inactivated
C) Is a marker used to eliminate the cells that have the DNA integrated at non-targeted sites
D) Is a gene that encodes protective core and matrix proteins for viral assembly and infection
E) Is a marker used to select the cells that have the DNA integrated anywhere in the genome
F) Is a regulatory gene that activate viral transcription
G) A method of introducing new DNA into a cell by injecting it directly into the nucleus
H) It is a chemical that is cytotoxic for recombinant ES cells carrying the thymidine kinase gene
I) It is a chemical that is cytotoxic for recombinant ES cells carrying the neomycin
J) A term used to describe a founder animal that contain the retroviral transgene
K) The introduction of any DNA molecule into any living cell
L) Is a non-enveloped virus with a linear dsDNA genome
M) Is a virus that can infect dividing and non-dividing cells



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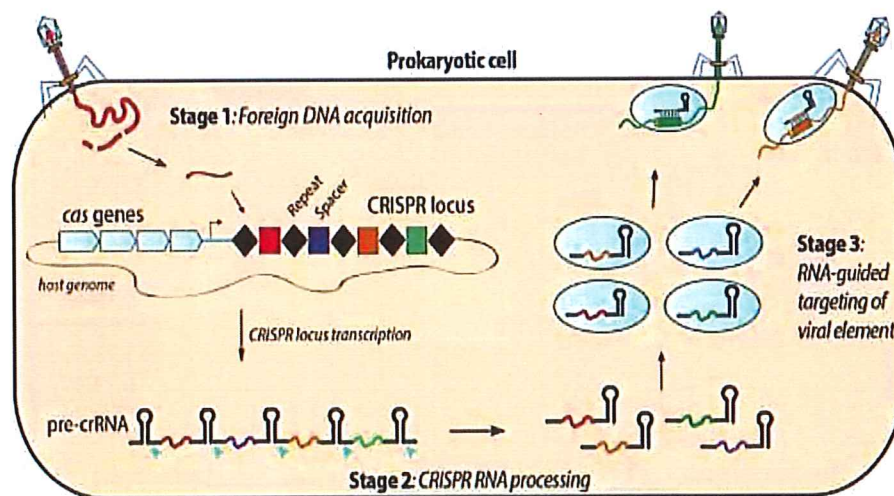
Question 5

True or false (4p)

- One of the major challenges with transgenic animals is the lengthy duration it takes to conform a transgenic offspring.
- Transgenic animals can serve as exact replicates of human system due to high genome similarity, facilitating disease study and development research
- The primary purpose of using transgenic animals is to solely improve animal production for human consumption.
- The knockout process involves a single crossover event in the crucial region of the gene, resulting in the disruption of its function.
- A knockout construct replaces a crucial part of the gene with an expression cassette that codes for a drug resistance marker, facilitating the disruption of the gene's function.
- The viral RNA contains three essential genes, *gag*, *pol*, and *env*, and is flanked by long terminal repeats (LTRs).
- Adeno-associated viruses are known to target the genetic material to human chromosome number 21.
- Once the adenovirus enters the host cell, the first gene to be expressed is the E1A.

Question 6

In detail describe the adaptive anti-viral immune system, CRISPR-Cas systems (as shown in the figure below). (13p)





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Question 7

Explain the differences between miRNA and siRNA in terms of their origin, processing pathway and their interactions with mRNA targets. **(4p)**

Question 8

Describe six things, three of each, you need to know about a **gene/vector** and the resulting **protein** when planning a cloning and expression experiment and why each one is important. **(6p)**

Question 9

Number the following steps in a protein expression and purification experiment from start **(1)** to end **(8)**: polishing, induction, cell lysis, SDS-PAGE, harvesting, functional analysis, chromatography, homogenization. **(2p)**

Question 10

Explain why or why not mammalian should be first choice of host for protein expression. **(2p)**

Question 11

Maple Syrup Urine Disease (MSUD) is a rare autosomal recessive metabolic disorder characterized by a deficiency in an enzyme complex crucial for breaking down the three branched-chain amino acids (BCAAs) – leucine, isoleucine, and valine – within the body. This metabolic failure results in the abnormal accumulation of all three BCAAs, and if left untreated, MSUD can lead to seizures, coma, and death. The occurrence of MSUD can be attributed to a point mutation in the BCKDHA gene. Explain in detail a method to identify carriers of MSUD, where carriers are individuals with a point mutation in one of the BCKDHA alleles. Consider that the mutation neither destroys nor creates a new restriction site. Present a comprehensive, step-by-step description of the genetic testing process, emphasizing the key laboratory techniques involved. **(8p)**

Question 12

What three things should a useful diagnostic test fulfill? Choose the best alternative below. **(1p)**

- A) Specific, Simple, Cheap
- B) Sensitive, Specific, Simple
- C) Sensitive, Fast, Cheap
- D) Specific, Sensitive, Fast
- E) Simple, Specific, Fast
- F) Cheap, Simple, Sensitive
- G) Fast, Simple, Sensitive

Question 13

As a genetic counselor at a Genetic Diagnostic Center, you encounter a family with a 2-year-old child displaying symptoms of developmental delay, seizures, and dysmorphic facial features. You suspect a very rare syndrome caused by a genetic mutation. In this context, explain your decision-making process regarding whether to utilize Whole-Exome Sequencing (WES) or Whole-Genome Sequencing (WGS) to identify the underlying cause of the child's symptoms. Provide a brief differentiation between WES and WGS, and discuss the advantages of choosing WES over WGS when investigating an unknown genetic disease or syndrome. **(3p)**