UNIVERSITY OF JOHANNESBURG

FACULTY	: Science
DEPARTMENT	: Biochemistry
CAMPUS	: APK
MODULE	: BICX04 CURRENT ADVANCES IN BIOTECHNOLOGY
SEMESTER	: Second
EXAM	: SSA January 2020

DATE	: January 2020	**SESSION**	: 08:00-11:00
ASSESSOR(S)	: PROF LA PIATER & Guest Lecturers		
MODERATOR	: DR L STEENKAMP		
DURATION	: 3 HOURS	**MARKS**	: 100

NUMBER OF PAGES: 5 PAGES (including this page)

INSTRUCTIONS:

1. Answer ALL THE QUESTIONS.
2. Number your answers clearly

QUESTION 1: Genes and Genomes **[10]**

Humans have tens of thousands of genes, and the development of DNA microarrays by Patrick O. Brown, Joseph DeRisi, David Botstein, and colleagues in the mid-1990s has led to many applications. Describe how microarrays work, and discuss how it has been used in cancer diagnosis and treatment thus far? (10)

QUESTION 2: Yeast Biotechnology **[10]**

2.1 Briefly discuss how a yeast cell may be engineered into (i) producing useful plant secondary metabolites and (ii) "humanizing" (authentic glycosylation) proteins. (7)

2.2 Briefly discuss the application of each of the following techniques:

2.2.1 TAR (1)

2.2.2 Yeast-two hybrid system (2)

QUESTION 3: Plant Biotechnology **[10]**

What are the genetic engineering strategies to create the following traits in transgenic crops. Where applicable, give an example of each:

a) Herbicide tolerance (5)

b) Virus resistance (5)

QUESTION 4: Molecular Diagnostics **[10]**

"HIV RNA (viral load) and CD4 T lymphocyte (CD4) cell count are the two surrogate markers of antiretroviral treatment (ART) responses and HIV disease progression that have been used for decades to manage and monitor HIV infection. Viral load is a marker of response to ART. A patient's pre-ART viral load level and the magnitude of viral load decline after initiation of ART provide prognostic information about the probability of disease progression. The key goal of ART is to achieve and maintain

durable viral suppression. Thus, the most important use of the viral load is to monitor the effectiveness of therapy **after** initiation of ART."

4.1. Design an experiment in which you measure the viral load of HIV in blood plasma of patients that have received ART treatment. (7)

4.2. "The co-circulation of different HIV types and groups can lead to dual infections and recombinants, which hinder diagnosis and therapeutic management". Suggest and justify ways in which you would modify your experiment approach in 4.1 in order to detect and measure different HIV types and groups HIV in ARV-treated vs. pre-ART treated patients. (3)

QUESTION 5: Biotechnology-based Drug Discovery **[10]**

5.1. State the drug discovery process in sequence and indicate how long does this process take on average. (6)

5.2. What considerations should be made when selecting a suitable screening assay? (3)

5.3. What is the parameter during lead identification that measures the efficacy of the lead compound? (1)

QUESTION 6: Vaccines **[10]**

List 4 differences between OPV and IPV polio vaccines, and the reason/s why we are slowly withdrawing OPV.

QUESTION 7: Stem Cells **[10]**

7.1 In stem cell-based therapies, the cells that are used within the therapy can be sourced in two different methods, either autologous or allogeneic. What is meant by these two terms? (2)

7.2 CAR-T therapy is a combination of genome engineering and cellular replacement therapy that is at the forefront of precision medicine. Discuss the concept behind the therapy including advantages and disadvantages of the treatment. (8)

QUESTION 8: Biofuels **[10]**

8.1 Various feedstocks are used to produce biofuels. Using corn as a feedstock, explain in detail the two processes employed for corn processing.

(10 X ½ = 5)

8.2 Biotechnology is the exploitation of biological processes for industrial and other purposes, especially the genetic manipulation of microorganisms to produce desired products. Using any two genetic manipulation approaches, explain how you will enhance both cellulose break down and sugar fermentation. Use 1 approach for cellulose and 1 approach for sugar fermentation (5)

QUESTION 9: Crystallography **[15]**

9.1 In your crystallography practical in July 2019, you attempted purification of two protein components that together were meant to crystallize.

9.1.1 The target protein MAGEB1 has a molecular weight of 29kDa, and the nanobody has 13k kDa. After purification, you have 80ul of MAGEB1 at a concentration of 5 mg/ml, and 130ul of the nanobody at 3 mg/ml. What volumes of each solution must you mix together to yield a solution where the nanobody is in 2x molar excess? Show your calculations. (2)

9.1.2 Explain the role of the Nickel in the IMAC purification. (1)

9.1.3 During purification, after adding the crude lysate to the IMAC column and doing several wash steps, you had to do a pre-elution with low

concentrations of imidazole. Explain why this was necessary, and how the imidazole addresses this. (3)

9.2 Apart from intrinsic properties of a protein, describe aspects of the experiment that are in your control that can improve chances of getting the protein to crystallize. (3)

9.3 Discuss how entropy drives both micelle formation and therefore protein folding. (3)

9.4 Describe a general strategy for achieving larger crystals simply by changing concentrations of crystallization components, and explain the principle; sketch a schematic phase diagram if it simplifies your answer. (3)

QUESTION 10: Application of knowledge in project [5]

During your individual BICX00 research project, you made use of “omics” (genomics, proteomic, metabolomics, *etc*.) method(s) or aspects thereof (genetics using qPCR, *etc.*). Suggest an alternative approach (one not used in your project) to answer your scientific research question, and briefly give an outline of the experimental design you will follow.