National Exams May 2016

04-BS-13, Biology

3 hours duration

NOTES:

1. If doubt exists as to the interpretation of any question, the candidate is urged to submit with the answer paper, a clear statement of any assumptions made.

2. This is a CLOSE BOOK EXAM. One aid sheet allowed written on both sides. Approved calculator is permitted.

3. FIVE (5) questions constitute a complete exam paper. The exam comes in two parts answer (3) questions from Part 1 and answer (2) questions in Part 11.
The first (3) questions in Part 1 and the first (2) questions in Part 11 will be marked as they appear in the answer book.

4. Each question is of equal value.

5. Some questions require an answer in essay format. Clarity and organization of the answer are important.

Part I: Solve any 3 questions out of the following 5 questions (20 marks for each)

Note: For some questions in order to calculate molecular weights of biomasses, products and substrates, elemental atomic masses will be needed. These are: for C = 12, for H = 1, for N = 14, and for O = 16.

1. (a) Describe with suitable diagrams typical structures of plant and animal cells. What are the differences in both structures? (8 marks)

(b) Explain the structure of a typical plant cell wall with a diagram. Describe cell contents and how these contents affect rheological properties, elasticity, swelling and shrinkage, rigidity and tensile strength of the plant material. (12 marks)

2. A feed stream containing glucose enters fed-batch fermenter at constant flow rate (F_i). The initial volume of liquid in the fermenter is V_o. Cells in the fermenter consume glucose at a rate given by $r_s = k_1$ s, where k_1 is the rate constant (1/h) and s is the concentration of glucose in the fermenter (g/L). (a) Assuming constant density (ρ), derive an equation for the total mass balance. What is the expression relating liquid volume (V) in the fermenter and time (t)? (10 marks) (b) Derive the differential equation for the rate of change of substrate concentration (s). (10 marks).

OR

A sewage disposal plant has a concrete holding tank of 100 000 L capacity. It is 75% full of suspension (water and organic material) to start with and contains 15000 kg of organic material in the suspension. Water enters into the holding tank at the rate of 20000 L/h and the suspension leaves at the rate of 15000 L/h. (a) How much (kg) organic material is in the tank at the end of 3 h? (12 marks) (b) After how much time, the tank will be full and find organic material concentration at that time. (8 marks). Consider density of the suspension constant at 1 kg/L. Note: Use unsteady state mass balance to solve this question.

3. The growth of an organism on glucose can be described by the following stoichiometric equation:

$$C_6H_{12}O_6 + a\ O_2 + b\ NH_3 \text{ -----> } c(C_{4.4}H_{7.3}N_{0.86}O_{1.2}) + d\ H_2O + e\ CO_2$$

Experiments have shown that cells convert 2/3 (w/w) of the substrate carbon to biomass. Calculate (a) the stoichiometric coefficients for the above biological equation (10 marks), (b) calculate the yield coefficients Y_{xs} (g biomass/g substrate), and Yx/O_2 (g biomass/g O_2) (10 marks). Neglect the ash content of biomass in calculating its molecular weight.

4. Production of recombinant protein ($CH_{1.55}O_{0.31}N_{0.25}$, MW = 22.03) by a genetically engineered strain *E. coli* ($CH_{1.77}O_{0.49}N_{0.24}$, MW = 25) is proportional

to cell growth. Ammonia (NH_3) is used as nitrogen source for aerobic respiration of glucose ($C_6H_{12}O_6$, MW = 180.2). The yield of biomass from glucose (Y_{XS}) is 0.48 g/g; and the yield of recombinant protein from glucose (Y_{PS}) is 20% that for cells. (a) How much ammonia is required using N elemental balance, g ammonia/g glucose? (6 marks) (b) What is the oxygen demand using electron balance, g oxygen/g glucose? (7 marks) (c) If the biomass yield remains at 0.48 g/g, how much different are the ammonia and oxygen requirements for wild-type *E. coli* unable to synthesise recombinant protein, i.e. f = 0? (7 marks). The biological reaction can be written as:

$C_6H_{12}O_6 + a\ O_2 + b\ NH_3 \rightarrow c\ CH_{1.77}O_{0.49}N_{0.24} + d\ CO_2 + e\ H_2O + f\ CH_{1.55}O_{0.31}N_{0.25}$

Glucose degree of reduction (γ_S) = 4.0; biomass degree of reduction (γ_B) = 4.07.

5. Immobilised cells of a genetically improved strain of *Brevibacterium lactofermentum* are used to convert glucose to glutamic acid for production of MSG (monosodium glutamate). The immobilized cells are unable to grow, but metabolise glucose according to the equation:

$$C_6H_{12}O_6 + 1.5O_2 + NH_3 \rightarrow C_5H_9O_4N + CO_2 + 3H_2O$$

A feed stream of 4% glucose in water enters a 25000 L reactor at 25°C at a flow rate of 2000 kg/h. A gaseous mixture of 12% NH_3 in air is sparged into the reactor at 1 atmospheric pressure and 15°C at a flow rate of 4 vessel volume per min. The product stream from the reactor contains residual sugar at a concentration of 0.5%. (a) Estimate the cooling requirements. (15 marks) (b) What will be the temperature increase in the fermentation if no cooling is provided? (5 marks)
Assume temperatures of product, reactor and off gases equal to 25°C.
Standard heat of combustion for the compounds (Δh_c^o) at 25°C and atmospheric pressure are: for glucose = -2805 kJ/gmol; for ammonia = -382.6 kJ/gmol; for oxygen = 0; for glutamic acid = -2244.1 kJ/gmol. Consider specific heat of all components mixture and water equal to 4.18 kJ/(kg K).

Part II. Answer any 2 questions out of the following 3 questions (20 marks for each question)

6. (a) List 4 techniques that are classed as "rapid methods" for microbiological analysis. (4 marks)

(b) What is the underlying principle of the Most Probable Number technique used to enumerate microbial levels? (4 marks)

(c) A food sample was delivered to a laboratory to perform coliform counts. **10 g** of each sample were added to **90 ml** of saline. A dilution series were prepared down to 10^{-4} and **0.1 ml** plated onto MAC agar (duplicate plates). After incubation at 35°C for 24 h, the following counts were obtained.
Calculate the cfu/g (show calculations) (8 marks)

Plate	10^{-2}	10^{-3}	10^{-4}
I	250	155	10
II	228	176	5

(d) Draw and label the structure of i) enveloped and ii) non-enveloped virus. (4 marks)

7. A mouse hybridoma cell line is used to produce monoclonal antibody. Growth in batch culture is monitored with the following results:

Time, days	Cell concentration, cells/ml x(10^{-6})	
0.0	0.45	
0.2	0.52	
0.5	0.65	
1.0	0.81	
1.5	1.22	
2.0	1.77	
2.5	2.13	
3.0	3.55	
3.5	4.02	
4.0	3.77	
4.5	2.20	

(a) Determine the specific growth rate during the growth phase. (15 marks)
(b) What is the culture doubling time? (5 marks)

Use the graph paper provided for data plotting.

8. (a) What conditions would lead to **low** growth yields of yeast cells? (3 marks)

(b) Differentiate among bacteria, yeasts, molds, algae and protozoa. Compare and contrast their general characteristics. (8 marks)

(c) Explain Monod growth kinetics of cells. What are other forms of growth kinetics (give only 2 more)? (5 marks)

(d) Explain the thermal death kinetics of cells. (4 marks)