Seoul National University

457.621.001

Biological Processes in Environmental Engineering

FINAL EXAMINATION - Solutions

TIME ALLOWED: 80 MINUTES

November 22, 2018

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1. Students may use two sheets of double-sided, A4-sized notes prepared in their own handwriting. Mechanical or electronic reproduction of any notes are not allowed.

2. Students should bring their own calculator which is not pre-programmed with formulae from the class.

3. Be aware that the cheated student will get 80% of the lowest score in class! There is no tolerance at all.

4. Make sure your answers include units if appropriate. Watch your units! Prepare your answers in a logical, easy-to-follow format.

5. This exam contains 6 questions. The total points is 130.

1. Mark true or false (T/F) for the following statements. (2 points each)

1) Classification of bacteria using the Gram staining technique is a way of taxonomic classification.

Answer) T

2) The f_s value decreases as an SRT of a CSTR-type bioreactor with cell recycle increases.

Answer) T

3) The Gibbs free energy change for ferrous (Fe^{2+}) - ferric (Fe^{3+}) half reaction is independent of pH.

Answer) T

4) The reaction to convert pyruvate to cell biomass is thermodynamically favorable in general.

Answer) F

5) No BOD removal is expected in a primary sedimentation basin of a municipal wastewater treatment plant.

Answer) F

6) A solid/water separation process (e.g., sedimentation basin, membrane filtration) is usually not necessary after an attached-growth-type bioreactor.

Answer) F

7) Water passes through a reverse osmosis (RO) membrane by diffusion.

Answer) T

8) *Nitrosomonas* is an aerobic chemoautotroph that oxidizes ammonia-nitrogen (NH_4-N) to nitrite (NO_2^-).

Answer) T

9) As long as the dissolved oxygen (DO) in an aeration tank of an activated sludge process does not significantly affect the heterotrophic microorganism growth, it is

desirable to maintain the DO level as low as possible.

Answer) F

10) Alkalinity will be reduced when ammonium-containing water goes through the process of nitrification-denitrification.

Answer) T

2. Multiple choice questions: choose one from those given in each question. (2 points each)

1) A horizontal gene transfer mechanism that a foreign DNA is introduced into a bacterial cell by a bacteriophage is called as:

A) Transformation B) Transduction C) Conjugation

Answer) B

2) Sulfur oxidizing bacteria (SOB) are a are a group of bacteria that utilizes the energy generated by the oxidation of sulfide (S^{2-}) or elemental sulfur (S^0) for growth and cell maintenance. How would you classify SOBs?

A) phototroph B) chemoorganotroph C) chemolithotroph

Answer) C

3) For an enzyme reaction without any inhibition, the reaction rate at a substrate concentration of $4K_M$ is (K_M = half-velocity constant; v_m = maximum reaction rate):

A) $2v_m$ B) v_m C) 0.8 v_m D) $0.5v_m$ E) $0.2v_m$

Answer) C

4) Select one that is NOT assumed to derive the Michaelis-Menten equation.

A) The free enzyme concentration in the system is constant.

B) The second step of the reaction, where the enzyme-substrate complex is transformed into free enzyme and products, is irreversible.

C) The enzyme-substrate complex concentration in the system is constant.

D) The first step of the reaction, where the free enzyme and the substrate is transformed into enzyme-substrate complex, is reversible.

Answer) A

5) Select one that correctly describes the effect of competitive inhibition. (v_m: maximum reaction rate; K_M: half-velocity constant)

A) decrease in v_m, increase in K_M B) decrease in v_m, no change in K_M

C) decrease in v_m, decrease in K_M D) no change in v_m, increase in K_M

Answer) D

6) Select an oxidant that results in the smallest standard Gibbs free energy change (on electron equivalent basis) for oxidation of ethanol.

A) O_2 B) NO_3^- C) SO_4^{2-} D) CO_2

Answer) D

7) At stationary growth phase of bacteria, the f_s value will be

A) positive B) close to zero C) negative

Answer) B

8) You isolated a bacterial species that reduces nitrate (NO_3^-) to nitrogen gas (N_2) using propionate ($C_2H_5COO^-$) as an electron donor. You further analyzed the growth kinetics of the bacterium to obtain the half saturation coefficient for nitrate (K_N) as 0.10 mg NO_3-N/L and the half saturation coefficient for propionate (K_P) as 50 mg propionate/L. At what condition will the specific growth rate of this bacterium be the highest? (C_N: nitrate concentration in mg NO_3-N/L; C_P: propionate concentration in mg/L)

A) C_N = 10, C_P = 10 B) C_N = 50, C_P = 5

C) C_N = 5, C_P = 50 D) C_N = 5, C_P = 5

Answer) C

9) You monitored the substrate utilization and bacterial growth rate of a heterotrophic bacterial species at an exponential growth state to obtain the maximum specific growth

rate of 10.0 d^{-1} and the maximum specific substrate utilization rate of 25 g BOD_L/g VSS-d. What is the f_s^0 value of this species? Assume that the bacterial cell can be represented by $C_5H_7O_2N$ (formula weight = 113).

A) 0.28 B) 0.42 C) 0.57 D) 0.83

Answer) C

10) Select one that correctly describes the changes in a chemostat as a function of SRT (θ_x).

A) If $\theta_x \leq \theta_x^{min}$, active biomass is washed out from the chemostat without having a chance of observable substrate degradation.

B) Effluent substrate concentration approaches zero when θ_x goes to infinity.

C) Active biomass concentration in the chemostat consistently increases with an increase in θ_x.

D) Fraction of active biomass in chemostat VSS increases with an increase in θ_x.

Answer) A

11) For a chemostat bioreactor, select one that contributes to the soluble BOD of the effluent.

A) Non-biodegradable VSS originating from the source water

B) Biodegradable fraction (f_d) of active biomass

C) Soluble microbial products

D) Non-biodegradable soluble organics originating from the source water

Answer) C

12) Select one that correctly describes the effect of the presence of the biodegradable organic particulates in the influent of a chemostat bioreactor (compare with a case when the biodegradable organic particulates do not exist).

A) Effluent substrate concentration (S) increases. (Note: do not account for the soluble microbial products)

B) The true yield (Y) increases.

C) The fraction of active biomass in total VSS (X_a/X_v) in the reactor decreases.

D) The active biomass concentration (X_a) in the reactor increases.

Answer) D

13) Select one that correctly describes the anammox process.

A) Nitrite (NO_2^-) is used as an electron acceptor.

B) The overall reaction requires NH4+ and NO2- at 1:1 molar ratio.

C) External carbon supply is needed.

D) The process is conducted by phototrophs.

Answer) A

14) Select a status of an anaerobic methane process that indicates a possibility of process failure.

A) The VFA concentration in the reactor is maintained low.

B) The pH in the reactor is maintained to be higher than 7.0.

C) Acetate concentration in the reactor steadily increases over time.

Answer) C

15) Select one that correstly explains what happens in an anaerobic tank of an enhanced biological phosphorus removal (EBPR) process.

A) Polyhydroxyalkanoates (PHAs) accumulated in the phosphorus accumulating organisms are consumed.

B) Polyphosphates are accumulated within the cell of the PAOs.

C) Increase in aqueous phosphate concentration is observed.

D) Phosphorus is removed from the system in the form of P-rich sludge.

Answer) C

16) Select one that can accomplish substantial biological phosphorus removal.

A) A^2O (Anaerobic/Anoxic/Oxic)

B) MLE (Modified Ludzak-Ettinger)

C) Contact stabilization

D) Trickling filter

Answer) A

17) Select one that correctly describes the feature of a membrane bioreactor (MBR).

A) The reactor biomass should be maintained low to prevent membrane fouling.

B) Clarifier is required to further remove SS from the MBR effluent.

C) Reverse osmosis (RO) membranes are used for effluent collection.

D) Can be operated at higher volumetric organic loading rate (OLR) compared to conventional activated sludge process.

Answer) D

18) Select one that correctly describes the anaerobic fermentation and oxidation process.

A) Acetogens are active at high H_2 partial pressure.

B) Aceticlastic methanogens consume CO_2 for CH_4 production.

C) A portion of the organic waste COD is converted to methane COD.

D) Generally acidogenesis is a rate-limiting step.

Answer) C

19) A reaction that may occur at the cathode of a microbial fuel cell (MFC) is:

A) Reduction of O_2 to H_2O

B) Reduction of proton to H_2

C) Oxidation of an organic substrate

D) Oxidation of an inorganic substrate

Answer) A

20) Select a compound of which concentration is unlikely to be reduced substantially during

wastewater treatment.

A) Volatile, refractory, and hydrophilic

B) Non-volatile, biodegradable, and hydrophobic

C) Non-volatile, refractory, and hydrophobic

D) Volatile, refractory, and hydrophilic

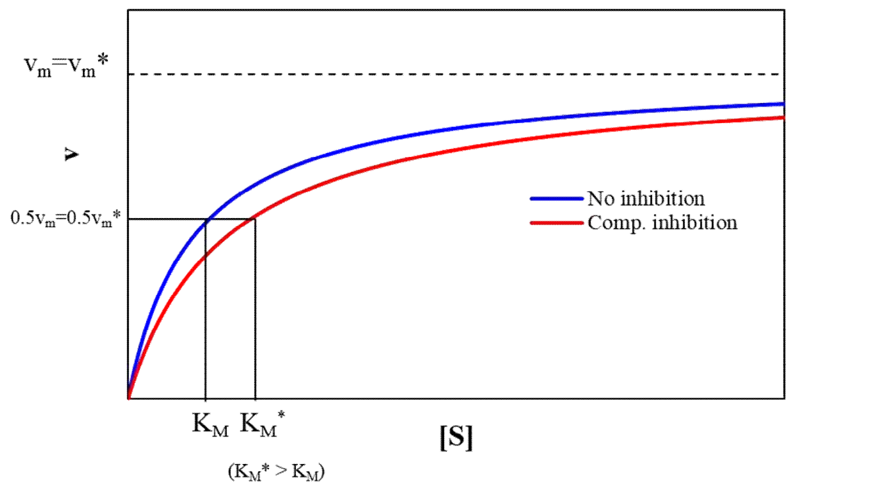
E) Non-volatile, refractory, and hydrophilic

Answer) E

3. Answer the following questions.

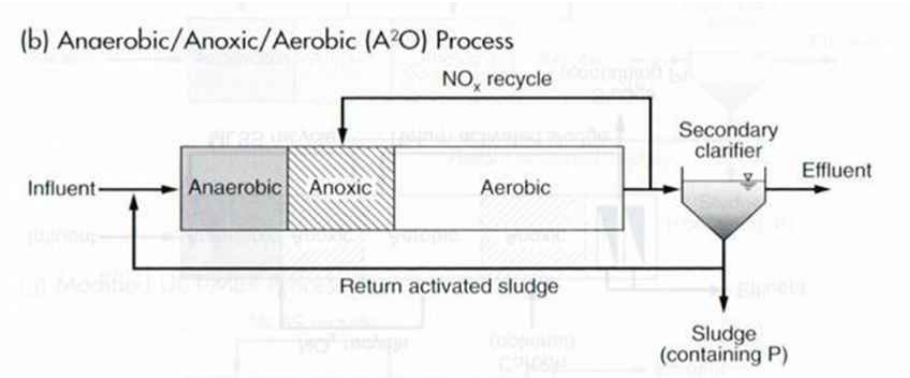
1) Plot the rate of an enzyme reaction (v) as a function of substrate concentration ([S]) i) in the absence of any inhibitors and ii) in the presence of a competitive inhibitor. Mark the maximum rates (v_m, v_m*) and half-velocity constants (K_M, K_M*) in the plot. (7 points)

Answer)



2) Draw a process diagram for A^2O process and describe what occurs in each reactor. (7 points)

Answer)



Anaerobic reactor: PAO synthesizes PHAs from VFAs by utilizing energy of polyphosphate, releasing P into the water

Anoxic reactor: Denitrifying bacteria consumes BOD to convert NO_3-N fed from the internal recycle stream to produce dinitrogen gas. The PAO metabolism described below also takes place with NO_3-N as an electron acceptor to produce dinitrogen gas.

Aerobic reactor: PAO utilizes PHAs stored in cells as well as wastewater BOD with O_2 supplied, resulting in cell growth. Phosphate in the wastewater is taken up by PAOs reducing the P concentration in the water. Nitrifying bacteria converts NH_4-N to NO_3-N.

3) Describe two possible ways by which trichloroethylene (TCE) can be biotransformed to innocuous (i.e., non-toxic) compounds. (6 points)

Answer)

i) reductive dehalogenation (dechlorination): TCE and its degradation intermediates are utilized as electron acceptors by certain bacteria species to be reduced serially to dichloroethylene (DCE), vinyl chloride (VC) and then ethylene (C_2H_4). Ethylene is easily

mineralized in (an)aerobic condition.

ii) cometabolism: TCE oxidation is mediated by monooxygenase enzyme produced in the presence of an organic substrate such as methane and toluene. The product is TCE-epoxide, which is easily mineralized under aerobic condition.

4. Answer the following questions for mineralization of caproic acid ($C_5H_{11}COOH$).

1) Write the half reaction in an electron-equivalent form using caproic acid ($C_5H_{11}COOH$) as a reactant and CO_2 a mineralized carbon product. (10 points)

Answer)

Following the 7 steps we studied,

1) $CO_2 \rightarrow C_5H_{11}COOH$

2) $CO_2 + H_2O + e^- \rightarrow C_5H_{11}COOH$

3) $6CO_2 + H_2O + e^- \rightarrow C_5H_{11}COOH$

4) $6CO_2 + e^- \rightarrow C_5H_{11}COOH + 10H_2O$

5) $6CO_2 + 32H^+ + e^- \rightarrow C_5H_{11}COOH + 10H_2O$

6) $6CO_2 + 32H^+ + 32e^- \rightarrow C_5H_{11}COOH + 10H_2O$

7) $\frac{3}{16}CO_2 + H^+ + e^- \rightarrow \frac{1}{32}C_5H_{11}COOH + \frac{5}{16}H_2O$

2) Calculate the COD value for 1 mole of caproic acid. (5 points)

Answer)

1 e^- equivalent = 8 g COD, 1 mole caproic acid = 32 e^- equivalent

Therefore,

COD value for 1 mole caproic acid = 32×8 = *256 g COD*

5. A complete-mix activated sludge process with solids recycle is used to treat wastewater at a flowrate of 4000 m^3/day with soluble, biodegradable organic concentration of 200 mg COD/L and dissolved oxygen (DO) concentration of 3.0 mg/L. The aeration tank volume is designed to be 1000 m^3. Using the following growth parameters, answer the questions. Neglect the production of soluble microbial products, hydrolysis of particulate COD, and any inert VSS in the influent.

$\hat{q} = 6\ mg\ COD/mg\ VSS-d$

$Y = 0.4\ mg\ VSS/mg\ COD$

$K = 100\ mg\ COD/L$

$b = 0.05/d$

$f_d = 0.85$

1) Determine the solids retention time (SRT) required to just meet the effluent standard of 10 mg COD/L. Determine the active biomass concentration in the aeration tank at that SRT. (10 points)

Answer)

$$S = \frac{K_s(1+b\theta_x)}{Y\hat{q}\theta_x - (1+b\theta_x)} = \frac{100(1+0.05\theta_x)}{0.4 \cdot 6 \cdot \theta_x - (1+0.05\theta_x)} \quad (\theta_x \text{ in days})$$

$$10\ mg/L = \frac{100+5\theta_x}{2.35\theta_x - 1}\ mg/L$$

$$18.5\theta_x = 110$$

$$\theta_x = 5.95\ days$$

$$X_a = \frac{\theta_x}{\theta} \cdot \frac{Y(S^0 - S)}{1+b\theta_x}$$

$$\theta = \frac{V}{Q} = \frac{1000\ m^3}{4000\ m^3/d} = 0.25\ d$$

$$X_a = \frac{5.95\ d}{0.25\ d} \cdot \frac{0.4(150-10)}{1+0.05 \cdot 5.95} = 1030\ mg\ VSS/L$$

2) At the SRT derived in 1), determine the total VSS concentration in the aeration tank. (5 points)

Answer)

$$X_v = X_a + X_i = X_a + \frac{\theta_x}{\theta}\left[X_i^0 + X_a(1-f_d)b\theta\right] = X_a\left[1+(1-f_d)b\theta_x\right] \quad (X_i^0 = 0)$$

$$= 1030 \cdot [1+(1-0.85) \cdot 0.05 \cdot 5.95] = 1080\ mg\ VSS/L$$

3) At the SRT derived in 1), calculate how much fraction of the soluble COD utilized is converted to particulate COD by the process. (10 points)

Answer)

Total VSS production rate,

$$r_{VSS} = \frac{X_v V}{\theta_x} = \frac{1080\ mg\ VSS/L \cdot 1000\ m^3 \cdot 10^3\ L/m^3}{5.95\ d} = 1.82 \times 10^8\ mg\ VSS/d = 182\ kg\ VSS/d$$

Mass rate of substrate utilization,

$$R_{ut} = Q(S^0 - S) = 4000\ m^3/d \cdot 10^3\ L/m^3 \cdot (200-10)\ mg\ COD/L = 7.6 \times 10^8\ mg\ COD/d = 760\ kg\ COD/d$$

$$\text{Fraction converted} = \frac{182\ kg\ VSS/d \cdot 1.42\ kg\ VSS/kg\ COD}{760\ kg\ COD/d} = 0.34$$

6. Calculate the maximum amount of methane that can be produced (in kg) by anaerobic digestion of 1000 m^3 of liquid waste containing 1000 mg BOD_L/L. (10 points)

Answer)

Because the question asked the maximum amount, we would assume i) no net biomass growth and ii) full utilization of BOD by anaerobic digestion.

COD value per g of CH_4 = $64\ g\ COD/mole\ CH_4 \times \frac{1\ mole\ CH_4}{16\ g\ CH_4} = 4.0\ g\ COD/g\ CH_4$

$$1000\ mg\ COD/L \times 10^3\ m^3 \times 10^3\ L/m^3 \times 10^{-3}\ g\ COD/mg\ COD = 10^6\ g\ COD$$

Therefore, the liquid waste can produce a maximum of

$$\frac{10^6\ g\ COD}{4.0\ g\ COD/g\ CH_4} = 2.5 \times 10^5\ g\ CH_4 = 250\ kg\ CH_4$$