$v = \frac{ER}{f}$

$\mu = \frac{v}{E}$

KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY, KUMASI
B.Sc. Biochemistry and Biotechnology
Class: BCHEM III
Second Semester Examination 2012/2013
BCHEM 264 Biophysics
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Time allowed: 3 hrs

Instructions
Answer all questions
Circle the correct answer on the question sheet

1. Phage T2 has a double stranded DNA with 2.5×10^5 bp. How many full double-helical turns are present in this DNA?

(A) 42,000 turns
(B) 2.625000 turns
(C) 25,000 turns
(D) 19,047 turns
(E) 23,809 turns

2. *Zea mays* has a double stranded DNA with 6.6×10^9 bp. What is the length of the DNA in centimeters?

(A) 194 cm
(B) 1.94 cm
(C) 224 cm
(D) 19.4 cm
(E) 2.24 cm

3. Homo sapiens has a double stranded DNA with 3.5×10^9 bp. What is the molecular mass of the this DNA?

(A) 0.23×10^9 kD
(B) 2.31×10^9 kD
(C) 0.188×10^9 kD
(D) 1.88×10^9 kD
(E) None of the above

4. Which of these secondary structures represent majority of DNA in nature?
(A) Slightly positive supercoiling in B form
(B) Slightly negative supercoiling in A form
(C) Slightly negative supercoiling in B form
(D) Extensive negative supercoiling in B form
(E) Extensive positive supercoiling in A form

5. A researcher is separating a mixture of proteins, A, B and C by polyacrylamide gel electrophoresis. The power pack is producing electricity at 200 volts. The red and black electrodes are separated by a distance of 18 cm. What is the magnitude of the electrical gradient across the electrodes?

(A) 0.90
(B) 0.09
(C) 11.1
(D) 1.11
(E) 36.0

6. Separation of the protein sample in question 5 was carried out for 60 min. Protein A migrated to a distance of 8.4 cm on the gel. What is the magnitude of the mobility of protein A?

(A) 0.01261
(B) 6.1261
(C) 1.2612
(D) 0.12612
(E) 12.6126

7. The disruption of electrophoretic separation caused by electroendosmosis involves which of the following mechanisms?

(A) Dissociation of matrix anions and their movement toward anode while sample migrates to cathode
(B) Dissociation of cations and their movement toward the cathode while sample migrates to anode
(C) Dissociation of matrix anions and their movement toward the anode together with the sample
(D) Dissociation of cations and their movement toward the anode together with the sample
(E) Dissociation of both anions and cations in the matrix with no net movement

8. Which of these gels would you use to separate 100 bp from 150 bp nucleic acid?

(A) Agarose with high –COOH content
(B) Agarose with high SO_4^- content
(C) Polyacrylamid gel or agrose with high SO_4^- and –COOH content
(D) Polyacrylamide gel or agarose with low $-SO_4^-$ content
(E) Only Polyacrylamide gel

9. Which of these gels would you use to separate proteins by immunioelectrophoresis?

(A) 12.5%: 5% acrylamide: BIS-acrylamide gel
(B) 7.5% : 5% acrylamide: BIS-acrylamide gel
(C) 3% agarose
(D) 1% agarose
(E) None of the above

10. Which of these reactions occur during polymerization of acrylamide.

(A) Vinyl addition of straight polymer in head-to-tail fashion and copolymerization of acrylamide monomers
(B) Vinyl addition of acrylamide monomers to form crosslinks and copolymerization straight polymers in head-to-tail fashion
(C) Vinyl addition of acrylamide monomers in head-to-tail fashion and copolymerization straight polymers to form crosslinks
(D) Vinyl addition of straight polymer to form crosslinks copolymerization of acrylamide monomers in head to tail fashion

(E) None of the above

11. You want to improve upon the resolution of protein separation on a polyacrylamide ge... Which of these gel systems is the right one to use?
(A) Gradient gel alone
(B) Stacking gel alone
(C) Gradient gel with stacking gel
(D) Gradient gel with resolving gel
(E) Resolving gel alone

12. Which of these gel slabs would you recommend for separating proteins having similar sizes?
(A) 5 cm × 8 cm
(B) 8 cm × 10 cm
(C) 12 cm × 15 cm
(D) 15 cm × 18 cm
(E) 20 cm × 25 cm

13. Which of these gel slabs would you recommend for separating proteins having diverse sizes?

(A) 1.5 cm × 3 cm
(B) 5 cm × 8 cm
(C) 15 cm × 18 cm
(D) 8 cm × 10 cm
(E) 5 cm × 8 cm

14. Electrophoretic stacking of a discontinuous buffer system is achieved via which of the sequence of events?
(A High concentration stacking gel→Cl^- leads glycinate trails→ Kolrausch discontinuity develops→protein stacks
(B) High concentration stacking gel→ glycinate leads→ Cl^- trails→ protein stacks→ Kolrausch discontinuity develops
(C) Low concentration stacking gel→glycinate leads→ Kolrausch discontinuity develop→sCl trails→ protein stacks
(D) Low concentration stacking gel→ Cl^- leads→ glycinate ions trail→Kolrausch discontinuity develops→ protein stacks
(E) High concentration stacking gel→Cl^- leads→ Kolrausch discontinuity develops→ glycinate trails→protein stacks

15.Sodium dodecyl sulfate is for denaturing of polyacrylamide gel electrophoresis of proteins as _____ and _____ are for denaturing of polyacrylamide gel electrophoresis of DNA
(A)dithiothreitol and NaOH
(B)Urea and formamide
(C)β-mercaptoethanol and urea
(D) β-mercaptoethanol and formamide
(E)uric acid and formaldehyde

16.Three proteins have the same Y intercept on a Ferguson plot. What is the interpretation?
(A)Same charge

Same Y intercept => same U
Same slope = same mass

(B) Same size
(C) Same mobility
(D) Same isoelectric pH
(E) Both A and B

17. Three proteins have the same slope on a Ferguson plot. What is the interpretation?
(A) same charge
(B) Same molecular mass
(C) Same mobility
(D) Same isoelectric point
(E) Both B and C

18. The equation which governs the mobility of macromolecules in native gel electrophoresis is
(A) Log R_m = log $Y_U T - K_R Y_0$
(B) Log $^{v}_{m}$ = log $R_0 - K_R T$
(C) Log R_m = log $Y_0 - K_R T$
(D) Log R_m = log $R_0 . K_R T$
(E) Log Y_m = log $R_o + K_R T$

Log Rm = Log Yo − KR T

19. Following SDS electrophoresis of histones that you have isolated from the nucleus of a eukaryote, you are getting one band per lane and or some lanes show no band at all. What would be the reason for such results?
(I) Anomalous migration of histones due to presence of extra charged groups on the protein
(II) No migration of histones due to absence of charged groups on the protein
(III) Retention of secondary structure due to incomplete denaturation by SDS
(IV) Complete denaturation of the histones to produce amino acids which migrate with no impedance by the gel matrix

(A) Only I
(B) Only II
(C) I and III
(D) II and IV
(E) II and IV

20. Which of these reagents and processes would you select to enhance denaturation of proteins that are difficult to denature by SDS alone?
(A) Triton X-100 + formamide + urea
(B) Triton X-100 + urea + acetic acid
(C) SDS + Triton X-100 + acetic acid with heating
(D) SDS + acetic acid + urea with heating
(E) SDS + Triton X-100 +urea

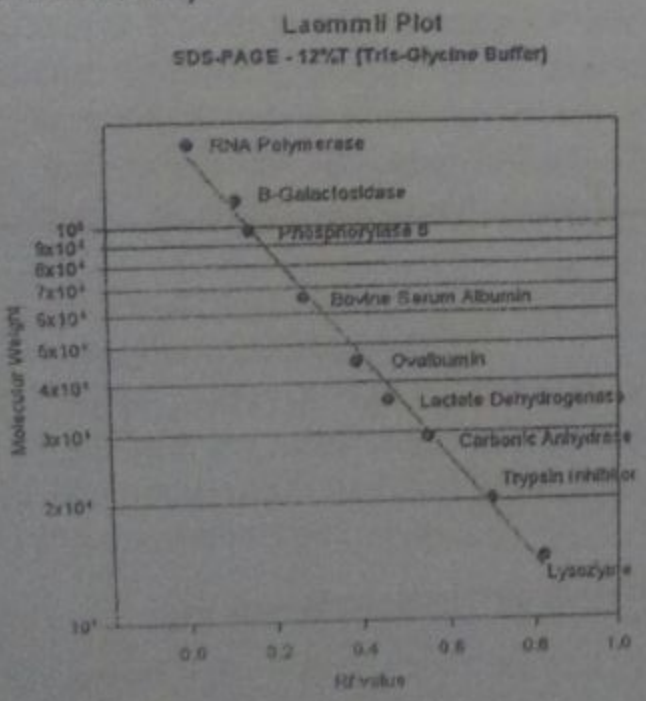
21. The contents of a good loading buffer for SDS-PAGE of plant tissue would be which of the following?
(I) More SDS, dithiothreitol, high pH
(II) More SDS, and mercaptoethanol, high pH
(III) Less SDS, CHES buffer, dithiothreitol, high pH
(IV) Less SDS, CHES buffer, mercaptoethanol, high pH

(A) Only I
(B) Only II
(C) I and III
(D) II and IV
(E) I, III, and IV

22. Four proteins of sizes, , 10 kD, 12.5 kD 25 kD and 100 kD were loaded in lanes 1 to 4, respectively, of SDS-PAGE gel and run with Tris-glycine buffer in the tank. Electrophoresis was stopped when bromophenol blue dye front was halfway down the gel. Some lanes did not show any band. What are these lanes?
(A) Lanes 1 and 2
(B) Lanes 1 and 3
(C) All lanes
(D) Only lane 1
(E) Only lane 4

23. The tank buffer in question 22 is changed to Tris-Tricine buffer, samples reloaded and SDS-PAGE run again. Which lanes would show bands?
(A) Lanes 1 and 2
(B) Lanes 2 and 3
(C) All lanes
(D) Only lane 1
(E) Only lane 4

24. Use the standard curve below to answer this question. The R_f value of a standard protein, X having relative molecular weight 7×10^4 was found to be 0.6 on SDS-PAGE. What explanation can you give for this anomaly?



(I) Protein X may be heterogeneous, such as glycoprotein
(II) Protein X may be heterogeneous, such as phosphorylated protein
(III) Excessive denaturation of protein X by SDS

(A)Only I
(B)Only II
(C)Only III
(D) II and III
(E) I, II and III

25.Which method of electrophoresis would you use to identify multiple proteins from a gene expression study?
(A) Isoelectric focussing
(B)Blue native PAGE
(C)2-dimensional gel electrophoresis
(D) Cellulose acetate electrophoresis
(E) Quantitative preparative native continuous PAGE

26.You are using isozyme marker to determine victim, suspect, and a pepetrator in a crime scene where a spot of blood was left. Which method of electrophoresis would you use?
(A) Isoelectric focussing
(B)Clear native PAGE
(C) 2-dimensional gel electrophoresis
(D) Cellulose acetate electrophoresis
(E) Quantitative preparative native continuous PAGE

27. Which is the correct sequence of the events listed below, which occur during fixation of proteins on a gel by methanol:water:glacial acetic acid reagent?
(I) Protein complex trapped inside gel
(II) Low pH disrupts hydrogen bonds
(III) Hydrophobic portions of protein exposed by organic solvent
(IV) Uncoiling of peptide chains
(V) Irreversible association between peptide chains

(A) I, III, IV, II, V
(B) I, II, III, IV, V
(C) III, IV, II, I, V
(D) IV, I, V, III, II
(E) II, III, IV, V, I

28. Which stain would you use to detect proteins up to 200 ng concentration on a polyacrylamide gel?
(A) Silver stain 2-5 ng
(B) Fluorescent dye 1-2 ng
(C) Coomassie Brilliant Blue G250 0.1 ug
(D) Coomassie Brilliant Blue R250 0.5 ug
(E) Ethidium bromide 20 ng 25 – 100 ng
glycoprotein

29. You performed electrophoresis on glycoproteins, loaded in two lanes and used horseradish peroxidase as a positive control in the third lane. Only one sample lane showed a band and there was no band in lane 3. Which of these not may be a good reason for your observation?
(A) Samples were contaminated

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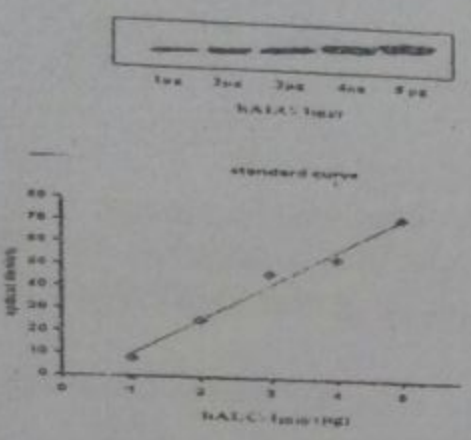
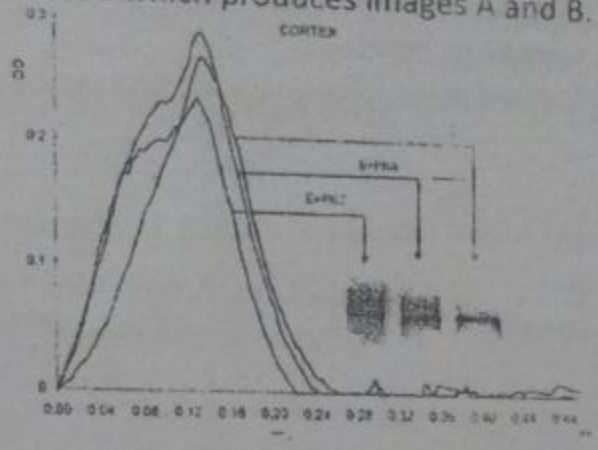
(B) Lane with no band means sample concentration was below detection limit
(C) The horseradish peroxidase was inactive
(D) The method did not work well
(E) All of the above

30. Identify the protein quantification method which produces images A and B.



A



B

(A) Gel documentation and Absorbance of eluted bands
(B) Absorbance of eluted bands and scanning densitometry
(C) scanning densitometry and Absorbance of eluted bands
(D) scanning densitometry and Gel documentation
(E) None of the above

31. Which of these is the correct sequence of immunostaining of western blots. *Note: CSPD is ChloroPhosphoPhenyl Dioxetane; BCIP/NBT is BromoChloroIndoyl Phosphate coupled with Nitro Blue Tetrazolium.*

(A) Blot→ Blocking→Horseradish peroxidase→Antibody→ CSPD→Bue color
(B) Blot →Blocking→Antibody→Horseradish Peroxidase→BCIP/NBT→Blue color
(C) Blot →Antibody→ Blocking→Horseradish Peroxidase→CSPD→ Luminiscence
(D) Blot→antibody→Blocking→Alkallne Phosphatase→BCIP/NBT→Bue color
(E) Blot→ Blocking→Alkaline Phosphatase→Antibody→BCIP/NBT→Luminiscence

32. Which of these do not cause electroendosmosis?

I) Electrophoresis above pH 3
II) A region of charge separation at the capillary wall/electrolyte interface
III) Cations in the electrolyte near the capillary wall migrate towards the cathode
IV) Electrolyte solution pulled toward the anode
V) Electrophoresis below pH 3

(A) I and II
(B) I and IV
(C) I alone
(D) V alone
(E) II and V

33. Select the correct statement in preparation of gels for electrophoresis
(A) Agarose crosslinks are formed by cooling of heated agarose

B) Agarose crosslinks are formed by heating
C) Acrylamide crosslinks are formed by heating
(D) Acrylamide crosslinks are formed by copolymerization and vinyl addition
E) None of the above

34. Which of the methods listed below would you choose to resolve up to about 4,000 proteins?
A) Agarose gel electrophoresis
B) cellulose acetate electrophoresis
(C) 2D gel electrophoresis
D) Isoelectric focusing
E) SDS-PAGE

35. Two proteins have same relative molecular mass of 12,000. How can you detect this on a Ferguson plot?
(A) Same slope
B) Same intercept on Y axis
C) A slope of zero for each protein
D) A Y intercept of zero for each protein
E) None of the above

36. Which method is best for separating acidic water-soluble and membrane proteins?
A) Quantitative Preparative Native Continuous (QPNC) PAGE
B) Isoelectric Focusing
C) Blue Native PAGE
D) SDS-PAGE
(E) Clear Native PAGE

37. Arrange the following fixatives in order of denaturation ability
A) Alcohol:acid:water> Glutaraldehyde> Trichloroacetic acid
B) Glutaraldehyde> Trichloroacetic acid> Alcohol:acid:water
C) Alcohol:acid:water> Glutaraldehyde >Trichloroacetic acid
D) Glutaraldehyde> Alcohol:acid:water> Trichloroacetic acid
(E) Trichloroacetic acid> Alcohol:acid:water> Glutaraldehyde

38. You have separated a mixture of membrane proteins on SDS-PAGE. Which of these fixatives would you use in your preparation for staining?
A) Isoprpanol:acetic acid:water
B) Formaldehyde
(C) Ethanol:acetic acid:water
D) TCA
E) None of the above