ANSWER KEYS FOR THEORETICAL TEST
PART B
(The final version)

Mark "✓" for True or "✗" for False statements.

PART B									
Q.	**A**	**B**	**C**	**D**	**Q.**	**A**	**B**	**C**	**D**
51	✗	✗	✓	✓	76	✓	✓	✗	✓
52	✗	✓	✓	✗	77	✓	✗	✗	✗
53	✓	✓	✓	✗	78	✗	✓	✗	✓
54	✗	✓	✓	✓	79	✓	✗	✓	✓
55	✓	✓	✗	✓	80	✗	✓	✗	✓
56	✗	✓	✗	✓	81	✗	✓	✓	✗
57	✓	✓	✓	✗	82	✓	✓	✗	✗
58	✓	✗	✓	✓	83	✓	✓	✗	✓
59	✗	✓	✗	✓	84	✗	✗	✓	✗
60	✗	✗	✗	✓	85	✗	✗	✗	✓
61	✓	✓	✓	✗	86	✓	✗	✗	✗
62	✓	✓	✓	✓	87	✓	✓	✗	✓
63	✓	✓	✓	✓	88	✓	✗	✓	✓
64	✗	✓	✓	✗	89	✗	✓	✗	✓
65	✓	✓	✗	✓	90	✗	✗	✓	✗
66	✗	✓	✓	✗	91	✗	✗	✓	✓
67	✓	✓	✗	✓	92	✗	✗	✓	✓
68	✓	✓	✓	✗	93	✓	✗	✗	✓
69	✓	✓	✓	✓	94	✓	✗	✓	✓
70	✓	✗	✗	✓	95	✗	✓	✗	✗
71	✓	✓	✓	✓	96	✗	✓	✓	✓
72	✗	✗	✓	✓	97	✓	✓	✓	✗
73	✓	✗	✓	✗	98	✗	✗	✓	✓
74	✗	✓	✗	✗	99	✓	✓	✗	✓
75	✗	✗	✓	✓	100	✓	✓	✗	✓

| Country: |
| Student Code: |

27th International Biology Olympiad
17th-23rd July, 2016
Hanoi, Vietnam

THEORETICAL TEST

PART B

Total points: 50 points
Duration: 180 minutes

Dear Participants,

o Please write your **student code** in the given box.

o Write down your answers using a pen in the **Answer Sheet**. Only answers given in the **Answer Sheet will be evaluated.**

o Part B consists of 50 questions:

- Q51-Q60: Cell Biology
- Q61-Q68: Plant Anatomy and Physiology
- Q69-Q80: Animal Anatomy and Physiology
- Q81-Q83: Ethology
- Q84-Q93: Genetics and Evolution
- Q94-Q98: Ecology
- Q99-Q100: Biosystematics

o There are two types of questions: True/False multiple choice questions and gap filling questions.

- For each True/False multiple choice question, there are four statements. Mark "√" for True statements and "×" for False statements in the Answer Sheet. If you need to change an answer, you should strikethrough the wrong answer and write in the new one. See the example below:

	Statement	True	False
A		√	×
B			×
C			×
D		√	

- For each gap filling question, there are four designated spaces to fill in numbers or codes.

o Scoring for one question:

- If all four answers are correct, you will receive 1 point.
- If only three answers are correct, you will receive 0.6 point.
- If only two answers are correct, you will receive 0.2 point.
- If only one answer is correct, you will not receive any points (0).

o You can use the ruler and the calculator provided.

o Stop answering and put down your pen immediately when the bell rings at the end of the exam. Enclose the **Answer Sheet** and **Question Paper** in the provided envelope.
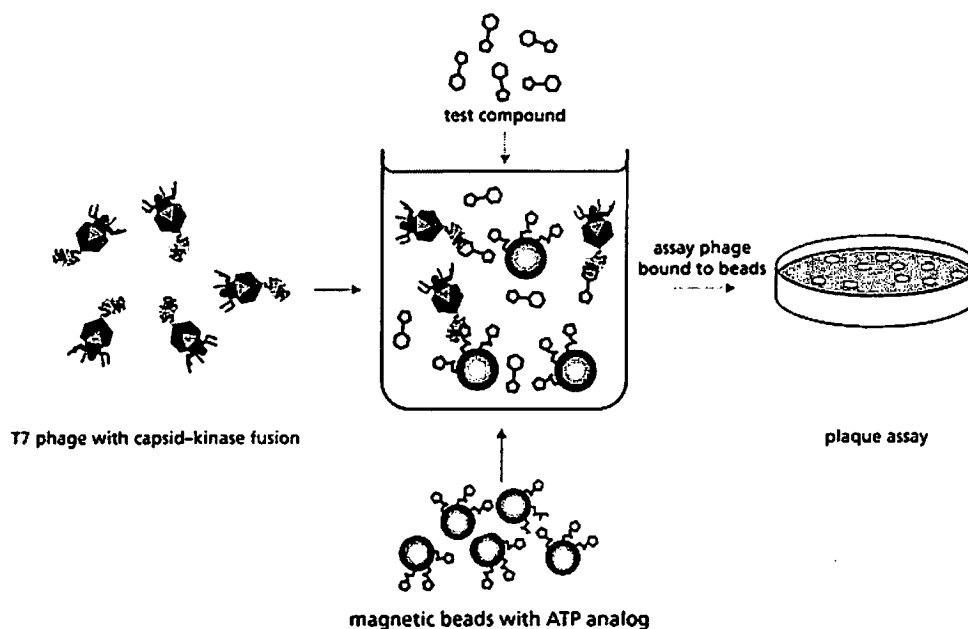
Good luck!!!

CELL BIOLOGY

Q.51

Scientist has prepared 3 essential components for high-throughput screens of protein kinase inhibitors. First, individual protein kinase genes are fused to the major capsid (head) gene of T7 phage. When expressed in bacteria, the fusion proteins are assembled into the phage capsid, with the kinases displayed on the outer surface. Second, an analog of ATP, which can bind to the ATP-binding pocket of the kinases, is attached to magnetic beads. Third, a bank of test compounds is prepared.

Fig.Q.51. Screening potential inhibitors of protein kinases

To measure the ability of a test compound to inhibit a kinase, phage displaying a specific kinase is mixed with the magnetic beads in several wells of a 96-well plate. Then the test compound is added to individual wells over a range of different concentrations. The mixtures are incubated with gentle shaking for 1 hour at 25°C, the beads are pulled to the bottom with a strong magnet, and all the free (unbound) components are washed away. Finally, the remaining, attached phage are dissociated from the beads using an excess of the same ATP analog that is attached to the beads, and counted by measuring the number of plaques they form on a bacterial lawn on a Petri dish (**Fig.Q.51**).

Indicate in the **Answer Sheet** if each of the following statements is True or False.

A. When the binding process reaches equilibrium, all potential inhibitor molecules will be bound to the kinase.

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B. Test compounds that score well in this assay bind in the ATP-binding cleft of the kinase.

C. Small differences in evolutionary conserved ATP binding sites on kinases allow targeting specific kinases.

D. A strong binding test compound will yield a low count in the plaque assay.

Answer key:

A. False, B. False, C. True. D. True

Explanation:

A. False: At equilibrium, most inhibitors can bind to the kinase, but some inhibitor molecules can dissociate from the kinase.

B. False: Some test compound could change the ATP binding site of the kinase by binding to an allosteric region of the kinase, which could be very far away from the binding site.

C. True: As the binding sites are similar but not identical between kinases, molecules that are specific for one kind of kinases can be developed.

D. True: In the presence of a strongly binding test compound, most of the phage will be attached to the test compound and will be washed away at the end of the incubation. Thus, strongly binding test compounds will give a low count in the plaque assay.
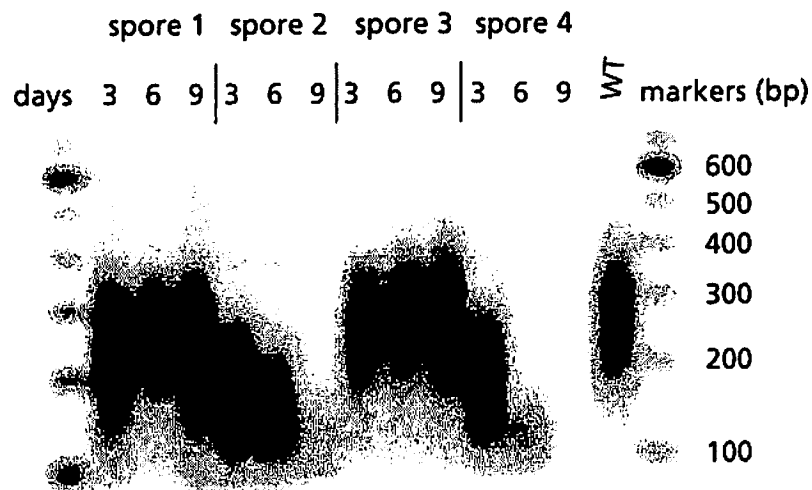
Reference: *Molecular Biolog of the cell*. B. Alberts et al

Griffin JD (2005) Interaction maps for kinase inhibitors. *Nat.Biotechnol.* 23, 308–309.

Fabian MA et al. (2005) A small molecule-kinase interaction map for clinical kinase inhibitors. *Nat. Biotechnol.* 23, 329–336.

Q.52

You identified a gene in fission yeast, homologous to a telomerase subunit from a protozoan. You then make a targeted deletion of one copy of the gene in a diploid strain of the yeast and then induce sporulation to produce haploid organisms. All four spores germinate perfectly, and you are able to grow colonies on nutrient agar plates. Every 3 days, you re-streak colonies onto fresh plates. After four such serial transfers, the descendants of two of the original four spores grow poorly, if at all. You take cells from the 3-, 6-, and 9-day master plates, prepare DNA from them, and cleave the samples at a chromosomal site about 35 nucleotides away from the start of the telomere repeats. You separate the fragments by gel electrophoresis, and hybridize them to a radioactive telomere-specific probe (**Fig.Q.52**). Assume that generation time is 6 hours.

Fig.Q.52. Analysis of telomeres from four fission-yeast spores.

WT is the normal diploid yeast

Indicate in the **Answer Sheet** if each of the following statements is True or False.

A. The average length of telomere in fission yeast is 280 nucleotides.

B. Spores 2 and 4 appear to lack telomerase.

C. Fission yeast telomeres lose less than 10 nucleotides per replication.

D. The fission yeasts that lose their telomeres will have normal size.

Answer key:

A. False; B. True, C. True. D. False

Explanation:

A. **False:** The region of intense hybridization to telomeres in the unaffected spores (1 and 3) and normal diploid yeast extends from less than 200 nucleotides to just

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over 300 nucleotides, averaging about 250 nucleotides. Since the cleavage site is 35 nucleotides from the beginning of the telomere repeats, the average length of telomere repeat in fission yeast is just over 200 nucleotides.

B. True: The descendants of spores 2 and 4 show telomere shortening with time, whereas the descendants of spores 1 and 3 remain the same size. Thus, spores 2 and 4 appear to lack telomerase

C. True: telomeres lose less than 100 nucleotides every 3 days. At four generations per day [(24 hours/day)/(6 hours/generation)] the yeast go through about 12 generations in 3 days. Thus, they lose less than 8 nucleotides per generation (100 nucleotides/12 generations).

D. False: the majority of fission yeast that lose their telomeres stop dividing but continue to grow in size, forming abnormally large cells.

Reference: *Molecular Biolog of the cell.* B. Alberts et al

Nakamura TM, Morin GB, Chapman KB, Weinrich SL, Andrews WH, Lingner J, Harley CB & Cech TR (1997) Telomerase catalytic subunit homologs from fission yeast and human. Science 277, 955–959.

Q.53

Reoxygenation after a period of lack of oxygen causes cardiomyocyte damage. One of the most potential indices evaluating myocardial functions is mitochondrial membrane potential, which is labeled by a cell permeant dye (positively-charged, grey color) readily accumulating in active mitochondria due to their relative negative charge.

The figure below illustrates hypoxia/reoxygenation (HR)-treated single myocyte model (1) with or without pre-hypoxic treatment of drug A. Myocyte images were captured at time points (a, b, c).

Fig.Q.53.

Indicate in the **Answer Sheet** if each of the following statements is True or False.

   A. As seen in **Fig.Q.53.(2)a**, cardiomyocytes are a type of striated muscle cells.

   B. Hypoxia induces acidic pH in myocardial mitochondria.

   C. Drug A pretreatment is good for cell because it prevents the collapse of mitochondrial membrane potential in HR.

   D. Captured images in drug A pretreatment group are presented in (2) and captured images in HR treatment without pretreatment of drug A are presented in (3).

Answers key:

   A. True, B. True, C. True, D. False.

Explanation:

   A. True: See Fig. (2)

   B. True: Hypoxia induces the accumulation of H^+ in the matrix.

   C. True: Drug A pretreatment is good for cell because it prevents the collapse of mitochondrial membrane potential in HR.

   - Collapse of mitochondrial membrane potential is presented with a reduction in dye intensity level. So, drug A is good for cell.

D. False: Captured images in drug A pretreatment group are presented in (2) and captured images in HR treatment without pretreatment of drug A are presented in (3).

- Capture images in drug A pretreatment group should be presented in (3) because drug A protects cell from reoxygenation injury with more dye-label mitochondria (grey color).
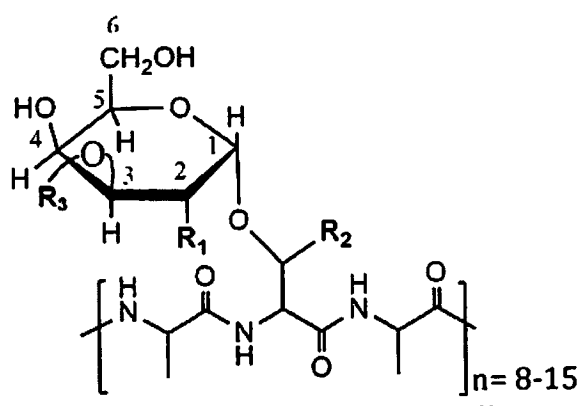
Reference

Angelos, M. G., V. K. Kutala, et al. (2006). "Hypoxic reperfusion of the ischemic heart and oxygen radical generation." Am J Physiol Heart Circ Physiol 290(1): H341-H347.

Han, J., S.-J. Park, et al. (2013). "Effects of the novel angiotensin II receptor type I antagonist, fimasartan on myocardial ischemia/reperfusion injury." International Journal of Cardiology 168(3): 2851-2859.

Thu, V. T., H.-K. Kim, et al. (2012). "NecroX-5 prevents hypoxia/reoxygenation injury by inhibiting the mitochondrial calcium uniporter." Cardiovascular Research 94(2): 342-350.

Q.54

Antifreeze glycoproteins (AFGPs) possess the ability to inhibit the formation of ice and are therefore essential to the survival of many marine teleost fishes that routinely encounter sub-zero temperatures. A typical AFGP consists of repeating tripeptide units, the alanyl-threonyl-alanyl $(Ala\text{-}Thr\text{-}Ala)_n$ unit connected to a disaccharide through a glycosidic bond at the second hydroxyl group of the threonine residue. To identify chemical groups which affect antifreeze activities of this glycoprotein, scientists synthesized numerous AFGP analogues by modifying both the structure of the sugar moieties and the peptide by replacing three groups R_1, R_2, R_3 as shown in **Fig.Q.54** with different chemical groups and recorded the antifreeze activity.

Fig.Q.54. The structure of a typical AFGP

The results of the study are shown in the following table.

R_1	R_2	R_3	Antifreeze activity
HO	CH_3	Galactosyl	No
N-Acetyl	CH_3	Galactosyl	Yes
N-Acetyl	H	Galactosyl	No
N-Acetyl	CH_3	H	Yes
O-Acetyl	CH_3	H	No
N-Acetyl	CH_3	Galactosyl-Galactosyl	No

Indicate in the **Answer Sheet** if each of the following statements is True or False.

A. A disaccharide bound to the threonine residue is required for antifreeze activity.

B. A mutant that has threonine residues replaced with serine residues significantly reduces antifeeze activities.

C. N-acetyl group at the C-2 position is required for antifreeze activity.

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D. Different numbers of repetitive motifs in AFGP genes amongst closely related species might have been caused by DNA polymerase inaccuracy.

Answer key:

A: False, B: True, C: True, D: True

Explanation:

The purpose of the question is to test comparative skill and ability to analyze table data. The question is also to test understanding on simple structure amino acids.

AFGPs display significant antifreeze activity if presence of the N-acetyl group at the C_2 position of a disaccharide, and the methyl group of the threonine residue is not modified.

A. False. AFGP has activity when R_3 is hydrogen (H), therefore the amino acid residues binding with the monosaccharide is not required for antifreeze activity.

B. True. Serine residues can form glycosidic bonds with the disaccharide but they do not have the hydrophobic group methyl (-CH_3). In this case, R_2 is H (see the table) so AFGP has no antifree activity.

C. True. If *N*-acetyl is replaced with –OH or *O*-acetyl, the activity of AFGP will be lost.

D. True. Probably a slippage of DNA polymerase happened during replication and resulted in extension of repetitive elements.
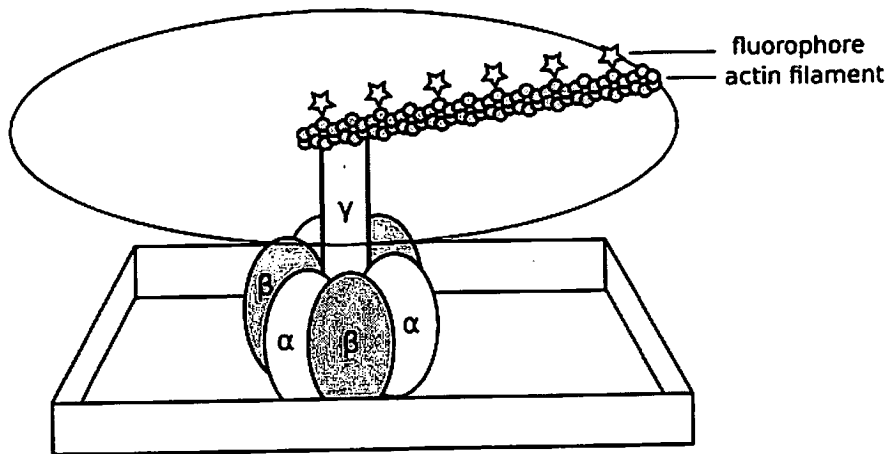
Reference

1. Jeong Kyu Bang, Jun Hyuck Lee Ravichandran N. Murugan, Sung Gu Lee, Hackwon Do, Hye Yeon Koh, Hye-Eun Shim, Hyun-Cheol Kim and Hak Jun Kim, 2013. Antifreeze Peptides and Glycopeptides, and Their Derivatives: Potential Uses in Biotechnology. Marine Drugs: 1, 2013-2041.
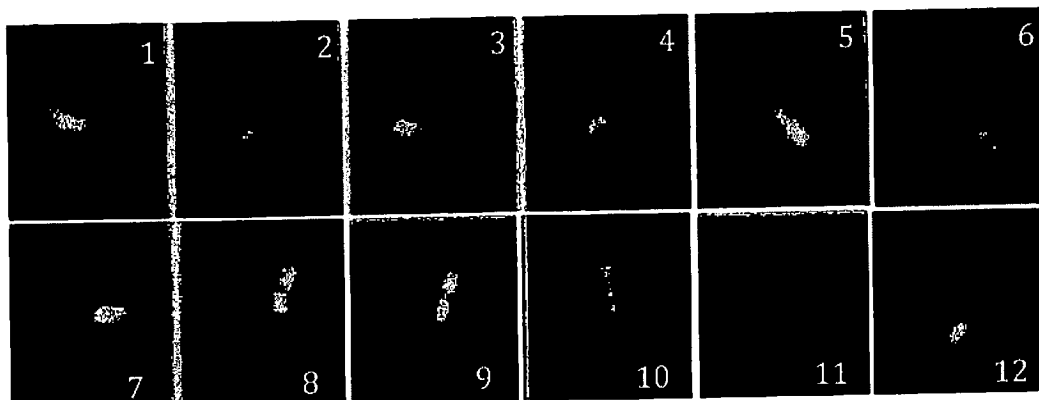
Q.55

F₁ subunit (a peripheral membrane protein) of the ATP synthase catalyses ATP synthesis using proton motive force responsible for the rotation of F_0 subunit (integral membrane protein complex) in one direction. F_1 is composed of three a and three b subunits arranged in alternating manner around a central shaft, the γ subunit.

To study the rotation, Masasuke Yoshida and his team attached a fluorescently labelled actin filament to γ and watched its movement.

Fig.Q.55A. Attachment of labelled actin filament to ATP synthase.

Rotating actin filaments were observed by an inverted fluorescence microscope after addition of 2 mM ATP into a chamber containing actin-taged $a_3b_3γ$ complex immobilized on the bottom side) as a mirror image formed on a camera. The time interval between images was 220 ms. A series of 13 images were taken and is shown in **Fig. Q.55B.**

Fig.Q.55B. Sequential image of rotating filament attatched to the sunbunit in the $a_3b_3γ$ subcomplex. The number indicate the shot image

Indicate in the **Answer Sheet** if each of the following statements is True or False.

A. Hydrolysis of ATP by F_1 leads to the conformational change of a and b subunits.

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B. From the set of figures, the filament rotated anticlockwise (looking from the membrane side).

C. Rotary rate is around 0.4 rounds per second.

D. Rotating the actin filament in the opposite direction is coupled with ATP synthesis.

Answer key:

A: True, B: True, C: False, D: True

Explanation:

To uncover many biological processes, scientists usually analyze series of images taken with time course. The idea in the question is to test ability to analyze a such series of images taken with time course. Beside, the question is designed to test ability to calculate data based on images as well as the position of ATPase on the membrane and mechanism behind generation of motive force.

A. True. The motive force rotating γ subunit results from set of conformational changes in F1 subunits directly linked to the ATP hydrolysis, mirroring the changes linked with the ATP synthesis.

B. True. From the set of figures, the filament rotated anticlockwise. The rotation is obviously anticlockwise – but viewed from opposite direction, han the membrane side and simultaneously as the mirror image. Resulting image is therefore anticlockwise.

C. False. Rotary rate is 0.375 – 0.400 rounds per second. The filament in frame 2 was at position 9 a'clock and after 9 frames (frame 12), filament was located at the same position in the frame 2. Time for one round rotation is (220 × 9)/1000 = 1.98 s, thus the rotary rate is 1/1.198= 0.50 rounds per second.

D. True. Rotating the actin filament clockwise is coupled with ATP synthesis. In the experiment, hydrolysis of ATPase couples with the filament rotated anticlockwise. The activity of ATP synthase must occur when filament rotate in the opposite direction, clockwise.

Reference

1. Albert L. Lehninger, David L. Nelson and Michael M. Cox, 2008. *Principles of biochemistry*, 5th edition. W.H. Freeman & Company.

2. Hiroyuki Noji, Ryohei yasuda, Masasuke Yoshida and kazuhiko Kinosith Jr, 1997. *Direct observation of rotation of F1-ATPase*. Nature, Vol. 386: 299-302

Q.56

Lactic fermented vegetables are traditional food in many Asian cuisines. Microorganisms commonly found in the fermentation broth are lactic acid bacteria, yeast and filamentous fungi.

Fig.Q.56 below shows the flowchart of viable cell counts (log CFU/mL) of three different microbial groups and the pH value during the lactic fermentation course of cabbage. Oxygen dissolved in fermentation broth decreased with time and was completely consumed after the 22nd day.
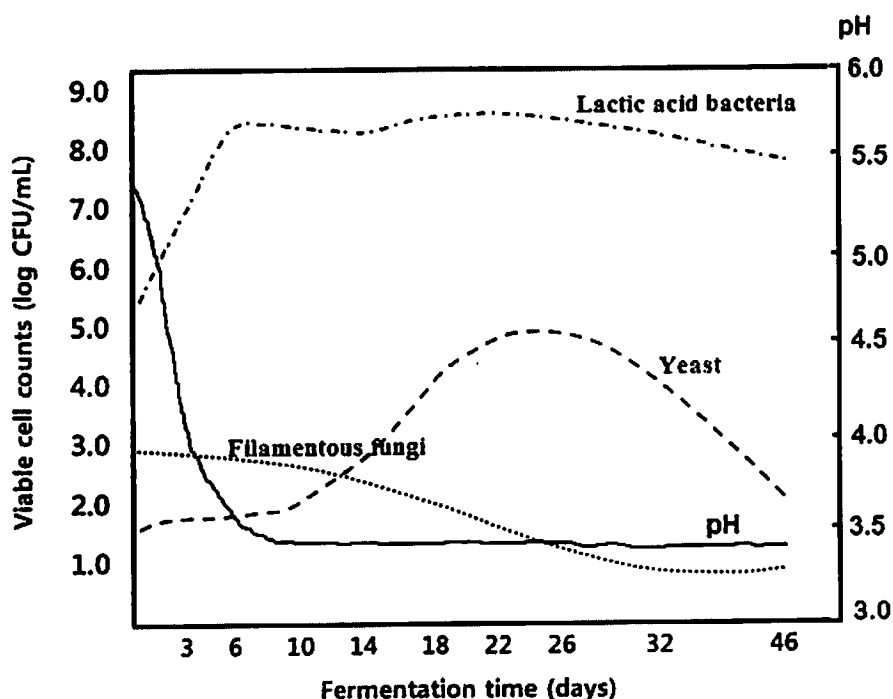

Fig.Q.56. Changes in microflora during lactic acid fermentation of cabbage.

Study **Fig.Q.56** and indicate in the **Answer Sheet** if each of the following statements is True or False.

A. The drop in pH value from day 1 to day 3 was caused by only organic acids produced by lactic acid bacteria.

B. Lactic acid produced by lactic acid bacteria favours the growth of yeast cells from day 10 till day 26.

C. Yeast cells shifted from fermentation to respiration after day 22.

D. Some filamentous fungi showed high tolerance at low pH.

Answer key:

A- False; B- True; C- False; D- True

Explanation:

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A- False: Organic acid can be produce from the respiration of various organisms. In the fermentation pile, not only lactic acid bacteria can produce organic acids but also yeast and filamentous fungi can.

B- True: Most yeast cells have optimum pH from 4 to 4.5 for their growth. Therefore, statement B is True.

C- False: After 22nd day, dissolved oxygen in fermentation broth were depressed so yeast cells must shift the respiration to the fermentation. Therefore, statement C is False.

D- True: Some filamentous fungi ($\sim$ 10 CFU/mL) found in fermented cabbage at the last stage. They are high tolerant to the low pH environment.

Reference

1- Microb Cell Factories 2011, Vol 10(Suppl 1):S5
2- Campbell R., Biology, Edition 9th

Q.57

Microorganisms that live at high salt concentration (above 2M of NaCl) are exposed to media with low water activity, and must have mechanisms to avoid water loss by osmosis. Analyses of intracellular ionic concentration of Halobacteriales living in salt lakes show that these microorganisms maintain extremely high salt (KCl) concentration inside their cells. The presence of high intracellular salt concentration requires special adaptations of the proteins and other macromolecules of the cells.

Indicate in the **Answer Sheet** if each of the following statements is True or False.

   A. Most intracellular proteins of Halobacteriales contain a large excess of negative charges on their outer surface.

   B. Halobacteriales spend a lot of ATPs to maintain osmotic pressure.

   C. Most intracellular enzymes of Halobacteriales lose their catalytic activity when suspended in solutions containing less than 1 M NaCl.

   D. Amino acids can be imported through Na^+/amino acids antiporters.

Answer key:

   A. True, B. True, C. True. D. False

Explanation

   A. True. The negative charges can help the protein maintain its proper conformation required for structural stability and enzymatic activity at high concentrations of cations.

   B. True. A lot of ATPs are used for maintaining extremely high salt (KCl) concentration inside their cells and also for the extrusion of Na^+ from the cell.

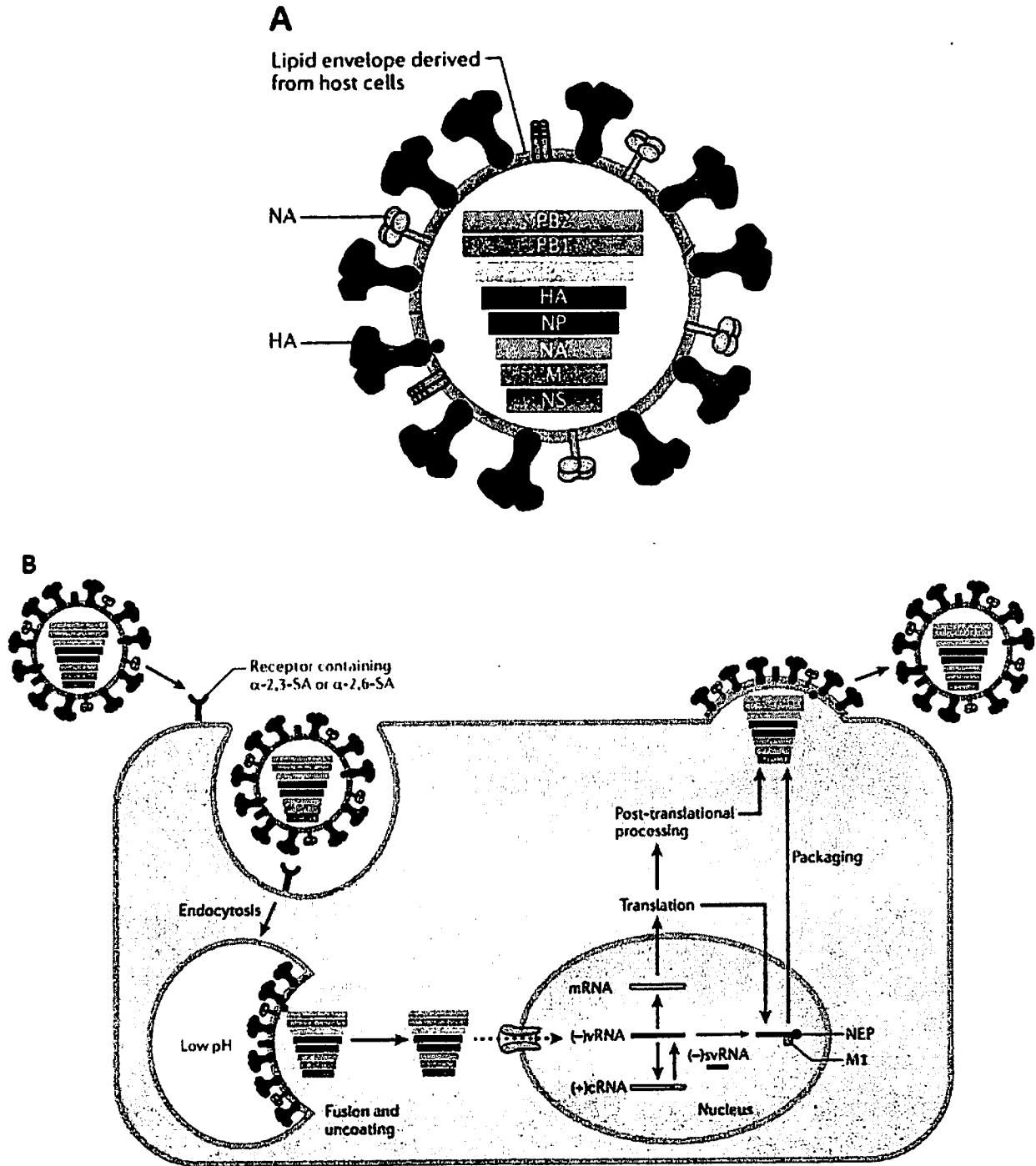   **C. True.** Most enzymes of the halobacteriales denatured when suspended in solution containing less than 1 M NaCl.

   D. False. Amino acids are imported throuth Na^+/amino acids symporters, the energy for active transport of amino acids into the cell is provided by the Na^+ gradient.

References:

Oren A. (2006). Life at high salt concentration. In: Dworkin M, Falkow S, Rosenberg E, Schleifer K-H, Stackebrandt E (eds) The Prokaryotes. A Handbook on the Biology of Bacteria: Ecophysiology and Biochemistry. Springer, New York. 2:263-28

Q.58

Influenza A genome consists of 8 separate single stranded RNA molecules, which encode a total of 11 viral proteins. Influenza A viruses are categorized by their two surface antigens, the hemagglutinin (H), of which there are 18 different subtypes (H1-18); and neuraminidase (N), of which there are 11 different subtypes (N1-11) (**Fig.Q.58A**). The influenza A virus life cycle is presented in **Fig. Q.58B**.

Fig.Q.58. Influenza A virus: (A) virus structure and (B) virus life cycle.

Indicate in the **Answer Sheet** if each of the following statements is True or False.

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A. Influenza A viruses exhibit rapid evolutionary dynamics because the genome is segmented.

B. In theory, there are 144 types of influenza A viruses.

C. Influenza A viruses exhibit high mutation rates because the genome is single strand RNA.

D. Influenza A viruses will be active only if RNA-dependent RNA polymerase is present in the virion.

Answer key

A. True, B. False, C. True. D. True

Explanation

A. True. A segmented genome provides a virus with the possibility of new gene combinations, and hence a potential for more rapid evolution.

B. False. In theory, there are 198 types of Influenza A viruses.

C. True. Influenza A virus uses RNA-polymerase for its replication but the enzyme does not possess a proof-reading-function. Hence, the error rate of viral RNA-polymerase is higher than the error rate of DNA-polymerase.
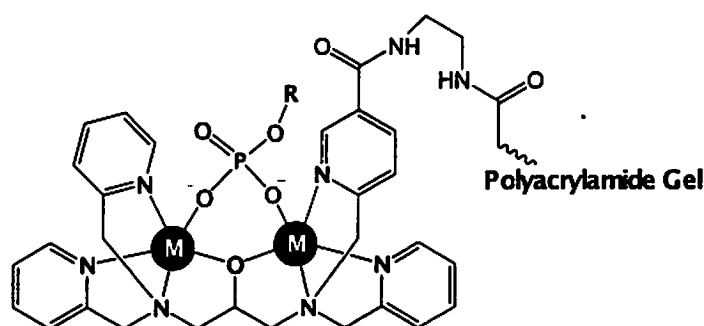
D. True. RNA-dependent RNA polymerase can only synthesize by virus, if it is not present in the virion the virus will be inactive.

References:

1. Nelson MI, Holmes EC (2007). The evolution of epidemic influenza. Nature Reviews Genetics 8:196-205.

2. Medina RA, Garcia-Sastre A (2011). Influenza A viruses: new research developments. Nature Reviews Microbiology 9:590-603.
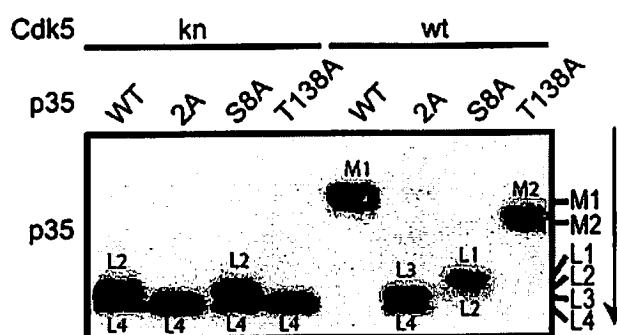
Q.59

Phosphorylation is a major post-translational modification widely used in the regulation of many cellular processes. A method to determine the phosphorylation status of proteins is to run an electrophoresis in a modified gel with a chemical group containing metal ions (M) that can reversibly bind phosphates and thus affects migration of phosphorylated proteins.

Fig.Q59A. Phospho-tag polyacrylamide gel

This technique was used to study the phosphorylation of protein p35. Three mutant forms of this protein were generated: a serine to alanine substitution in position 8 (S8A); a threonine to alanine substitution in position 138 (T138A) and both amino acid substitutions (2A). Note that serine and threonine can be phosphorylated while alanine cannot. Then two yeast strains with normal (wt) or inactive cyclin-dependent kinase 5 (Cdk5) (kn) were transformed with either the wild type version of p35 gene (wt) or one of the three mutant forms. Cell lysate of the eight resulting strains was loaded on a Phospho-tag gel. The proteins from the gel were transferred by western-blot to a membrane that was treated with anti-p35 antibodies. The result is shown below.

Fig.Q59B. Immunoblotting with anti-p35. The arrow indicates the direction of migration p35 bands are named M1, M2, L1, L2, L3, and L4. L4 band corresponds to the completely non-phosphorylated form of p35.

Based on the results of the experiment given above, indicate in the **Answer Sheet** if each of following statements is True or False.

A. Protein p35 has only two phosphorylation sites: serine 8 and threonine 138.

B. Protein p35 can be phosphorylated by a protein kinase different from Cdk5.

C. In strain Cdk5-wt p35-S8A only a few p35 molecules are phosphorylated at T138.

D. Phosphate groups attached to S8 are more accessible to phosphate binding groups of the Phospho-tag gel than phosphate groups attached to T138.

Answerkey:

A: False, B: True, C: False, D: True

Explanation:

A. False. Band L3 present in lane 6 corresponds to a phosphorylated protein. As this mutant has both S8 and T138 mutated to alanine, there should be at least one more site that can be phosphorylated.

B. True. Presence of phosphorylated form of the protein (band L2) in the kn cells lacking Cdk5 proves that protein p35 can be phosphorylated by a protein kinase different from Cdk5.

C. False. As L2 corresponds to p35 phosphorylated at T138 only and L1 corresponds to p35 phosphorylated at two sites T138 and an additional site (X) as can be seen from comparison between lines 6 and 7 all p35 molecules in this strain are phosphorylated at T138).
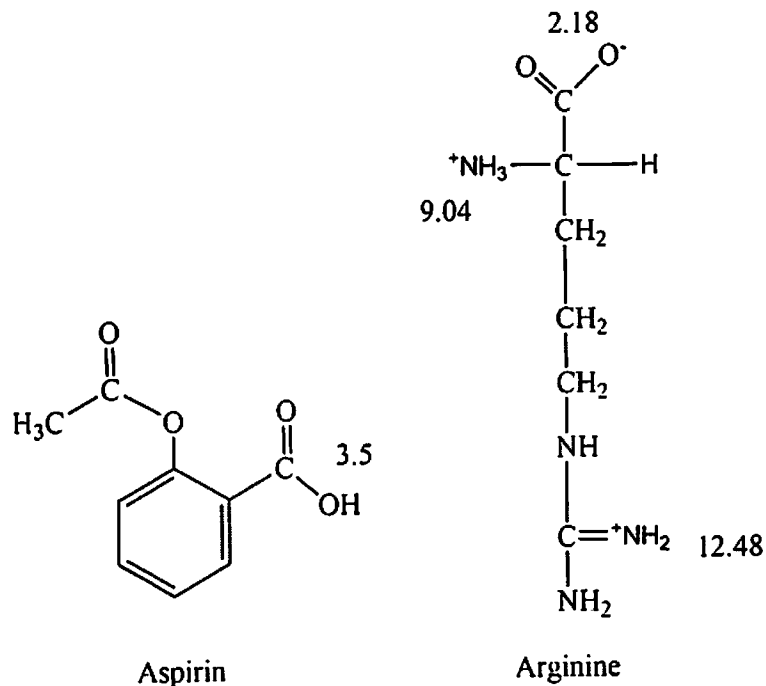
D. True. M2 band corresponds to p35 phosphorylated at S8. It might be phosphorylated at residue X as well. M2 has a much lower mobility compared to both L2 (phosphorylated at T138) and L1 (phosphorylated at T138 and X). The difference in mobility given the same number of phosphates attached per molecule makes us suppose that the mobility effect of phosphorylation of these two sites is different. As phosphorylation of S8 has a stronger effect its phosphate should interact stronger with the gel).

Reference

Mol Cell Proteomics. 2010 Jun; 9(6): 1133-43.

Q.60

Polarity, charge and molecular weight of molecules can affect their rate of passive diffusion through membranes. Amino acids and drugs like aspirin differ in both efficiency and location of absorption. In the figure below the chemical structure the pKa values of aspirin and arginine are represented.

Aspirin   Arginine

Indicate in the **Answer Sheet** if each of following statements is True or False.

A. Aspirin diffuses through membranes mainly in the stomach because more aspirin molecules are in deprotonated form at pH of about 1.6 in the stomach.

B. Arginine diffuses less efficiently than aspirin because of its higher molecular weight.

C. Optimal pH range for arginine absorption by passive diffusion is between 2.18 and 9.04.

D. The proton pump inhibitor, omeprazole, blocks the entry of aspirin into the blood in the initial few minutes after oral administration.

Answer key:
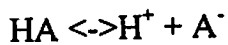
A: False, B: False, C: False, D: True

Explanation:

The purpose of the question is to test understanding factors that affect membrane transport through analysis of molecular absorption in the stomach and intestine. Because

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the pH differs in the stomach and the intestine, absorption location is different.

Stomach pH (pH$_s$) =1.6, Blood pH=7.4

pKa of aspirin = 3.4., weak acid:

HA <->H$^+$ + A$^-$

pH= pKa + lg ([A$^-$]/[HA]) -> lg ([A$^-$]/[HA]) = pH-pKa

in stomach:

lg ([A$^-$]/[HA]) =1.6-3.4= -1.8

[A$^-$]/[HA]) = 0.016

[HA] = 63 [A$^-$]

in blood:

lg ([A$^-$]/[HA]) =7.4-3.4= 4

[A$^-$]/[HA]) = 10000

[HA] = 0.0001 [A$^-$]

[HA] from stomach to blood

A. **False.** . Because at stomach pH is less than the *pI* of aspirin, the proportion of protonated form of aspirin is higher. The more charged molecules are, the less diffuse through the membrane.

B. **False.** Because the molecular weight of arginine is 174.2, smaller than that of aspirin (180. 16)

C. **False.** Arginine is neutral (zwitterionic) between pH 9.04 and 12.48.

D. **True.** Aspirin is absorbed mainly in the stomach in the initial few minutes. Omeprazole causes increase in pH in the stomach because it inhibits proton pumping into the stomach lumen.

Reference

1. Giraud N.M. et al. Effect of omeprazole on the bioavailability of unmodified and phospholipid-complexed aspirin in rats. Aliment Pharmacol Ther. 199711(5):899-906.

2. P. B. Miner Jr, J. G. Fort[†] & Y. Zhang. Intragastric acidity and omeprazole exposure during dosing with either PA32540 (enteric-coated aspirin 325 mg + immediate-release omeprazole 40 mg) or enteric-coated aspirin 325 mg + enteric-coated omeprazole 40 mg – a randomised, phase 1, crossover study:

3. William D. Masonx, Nathaniel Winer. Kinetics of Aspirin, Salicylic Acid, and Salicyluric Acid following Oral Administration of Aspirin as a Tablet and Two Buffered Solution

PLANT ANATOMY AND PHYSIOLOGY

Q.61

To study the effects of cadmium (Cd) on root development, two experiments on maize seedlings with 6-cm-long root were conducted. In the first experiment, seedlings were grown either in media supplemented with 5 μM Cd (Cd5) or without Cd (Cd0). In the second experiment, seedlings were grown either in two layers of agar without Cd (Cd0-Cd0) or unilaterally to 100 μM Cd (Cd0-Cd100). Four days later, root growth was recorded and cross-sections of roots were stained to visualize suberin lamillae in endodermis.
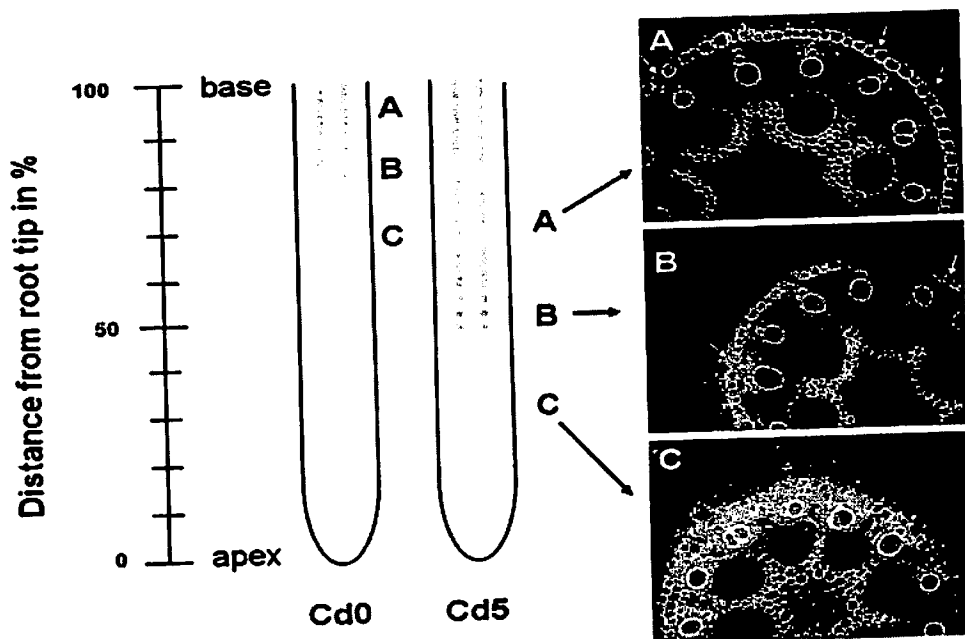

Fig.Q61-1. Distance (%) from root tip to root base is shown on the left. The presence of suberin lamillae in the roots are shown as solid and dashed lines in the center figures. White arrows in A and B indicate suberin lamellae in the endodermis.
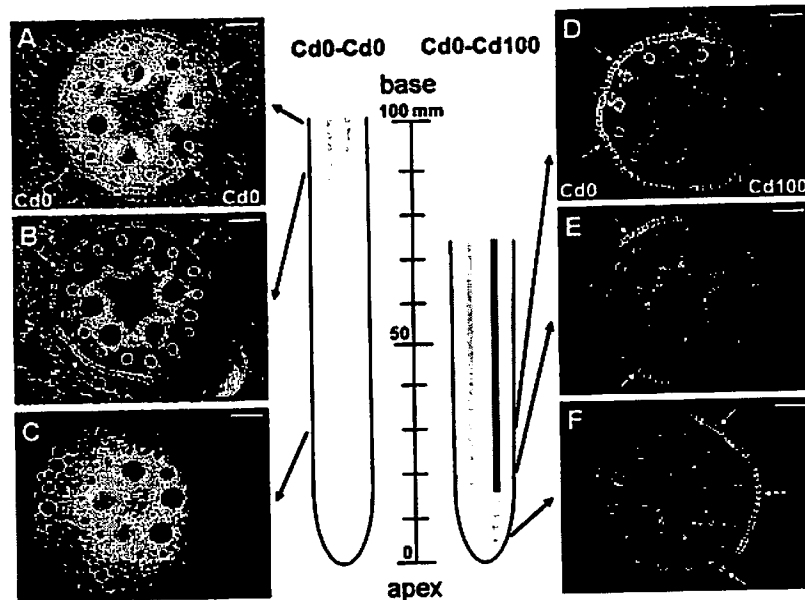
Fig.Q61-2. Distance (mm) from root apex to root base is shown on the center. The presence of suberin lamillae in the roots are shown as solid and dashed lines. White arrows in A, B, D, E and F indicate suberin lamellae in the endodermis.

Indicate in the **Answer sheet** if each of the following statements is True or False.

A. The treatment of Cd resulted in the reduction of elongation zone of the root, leading to decreased root length.

B. Endodermal cells with suberin lamellae were already present at a distance of approximately 0.5 cm from the root apex in tissues adjacent to agar containing Cd100, however, suberinized cells were found much later in the other side.

C. In roots exposed unilaterally to Cd (Cd0–Cd100), the development of the endodermis was accelerated and asymmetrical.

D. In high Cd containing media, suberin lamellae in endodermal cells were not present in older parts of the root likely due to the restriction of Cd in younger part.

Answer:

A. True   B. True   C. True   D. False

24

A. True. As shown in Fig.Q61-1, the elongation zone of control root is longer than in the treatment. In Fig.Q61-2. The root grown in the presence of Cd was much shorter than the control.

B. True. As shown in Fig.Q61-2, suberin lamellae were already present at a distance of approximately 0.5 cm from the root apex in tissues adjacent to agar containing Cd, but suberin lamellae were only present approximately 1.5 cm from the root apex at the side without Cd.

C. True. In the treatment with Cd, root endodermis with suberin lamellae is present earlier than in the control. The presence of suberinized endodermal cells is unequally in two sides with Cd100 and Cd0 (Fig.Q61-2)
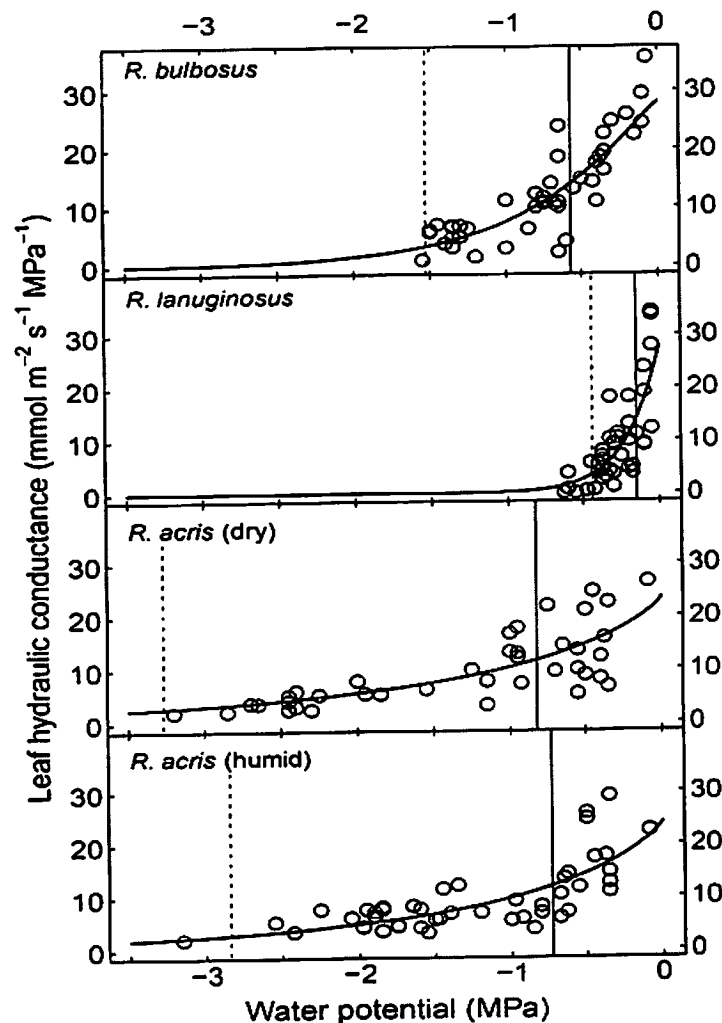
D. False. Suberin lamellae in endodermal cells disappeared early in maturation zone of the root (Fig.Q61-2C and D), but remained in the control root and the root side near the Cd0, indicating that the cells were distroyed by Cd. Younger part of root, especially cortex with parenchyma, could not restrict Cd.

Reference
Lux A., Martinka M., Vaculi'k M and White P.J, 2010. Root responses to cadmium in the rhizosphere: a review. *Journal of Experimental Botany*, Vol. 62(1), pp.21-37. doi:10.1093/jxb/erq281

Q.62

To understand the effect of desiccation on herbaceous plants and their responses, a student conducted a study on three *Ranunculus* species in their natural habitats, including *R. bulbosus* in dry medow, *R. lanuginosus* in humid medow, and *R. acris* in both habitats. He measured leaf water potential and hydraulic conductance of these species in response to dehydration (Fig.Q62). Xylem staining experiment on *R. acris* in dry habitat was used to estimate loss of conductivity due to embolism. An estimated 50% loss of xylem hydraulic conductivity due to embolism occurred at -2MPa or less. Previously, leaf hydraulic vulnerability studies found 50% reduction in leaf hydraulic conductance between − 1 and − 1.8 MPa in parennial grasses and at − 1.8 MPa in woody plant species.

Fig.Q62. Leaf hydaulic conductance of *Ranunculus* species/populations in response to dehydration. Solid and dashed vertical lines indicate, respectively, fitted 50% and 88% leaf hydraulic conductance losses.

Indicate in the **Answer sheet** if each of the following statements is True or False.

A. All species were very vulnerable to water stress. In species with narrow ecological amplitude, the drought-exposed *R. bulbosus* was less vulnerable to desiccation than the humid habitat *R. lanuginosus*.

B. Herbaceous species would be more vulnerable to water stress than woody species and perennial grasses, but also would show interspecific and intraspecific adjustments in hydraulic vulnerability based on the water availability of their respective habitats.

C. The leaf hydraulics method employs hydraulic conductance including both xylary and extraxylary pathways.

D. The effect of drought in these plant species is found to be a loss of leaf hydraulic conductance at moderate water potential based on extraxylary pathways rather than embolism formation.

Answer keys

A. True   **B.** True   **C.** True   **D.** True

Explanation

A. True. Leaf hydraulic conductance in all species/populations is reduced (50%) at water potential between - 0.2 to - 0.8 MPa (~ - 0.6 MPa for *R. bulbosus*, ~ - 0.2 MPa for *R. lanuginosus*, and ~ 0.7 and - 0.8 MPa for *R. acris* in humid and dry habitats repectively).

B. True. In this study, 50% leaf hydraulic conductance of the herbaceous species/populations is found at ~ - 0.2 to - 0.8 MPa, which is higher than woody and parennial plants. Species/populations in dry and humid habitats show responses to water stress differently as shown in the figure (particularly 50% and 88% hydraulic conductance losses).

C. True. Hydraulic conductance decreases greatly (50%) at higher than − 1 MPa, while xylem still remains conductivity at much lower than that (− 2 MPa or less), indicating a great contribution of extraxylary pathway; at lower water potentials, embolism greatly reduces hydraulic conductance. These indicate that both xylary and extraxylary pathways play role in the leaf hydraulics.
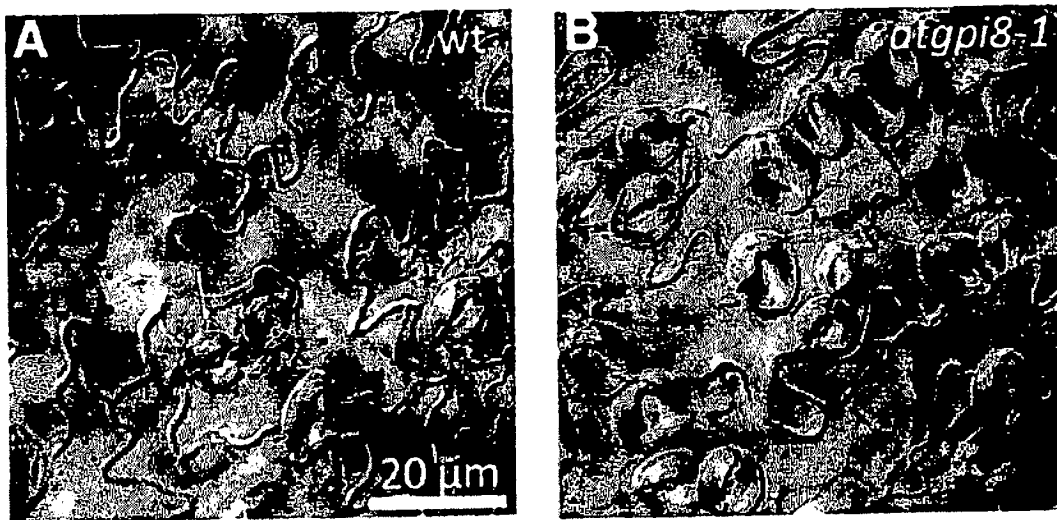
D. True. Hydraulic conductance decreases greatly at nearly -0.8 MPa (50% hydraulic conductance loss), while the embolism in xylem only occurs at water potential reaches -2 MPa, and 50% xylem conductance losses at less than -2 MPa, indicating that embolism only plays a minor role.
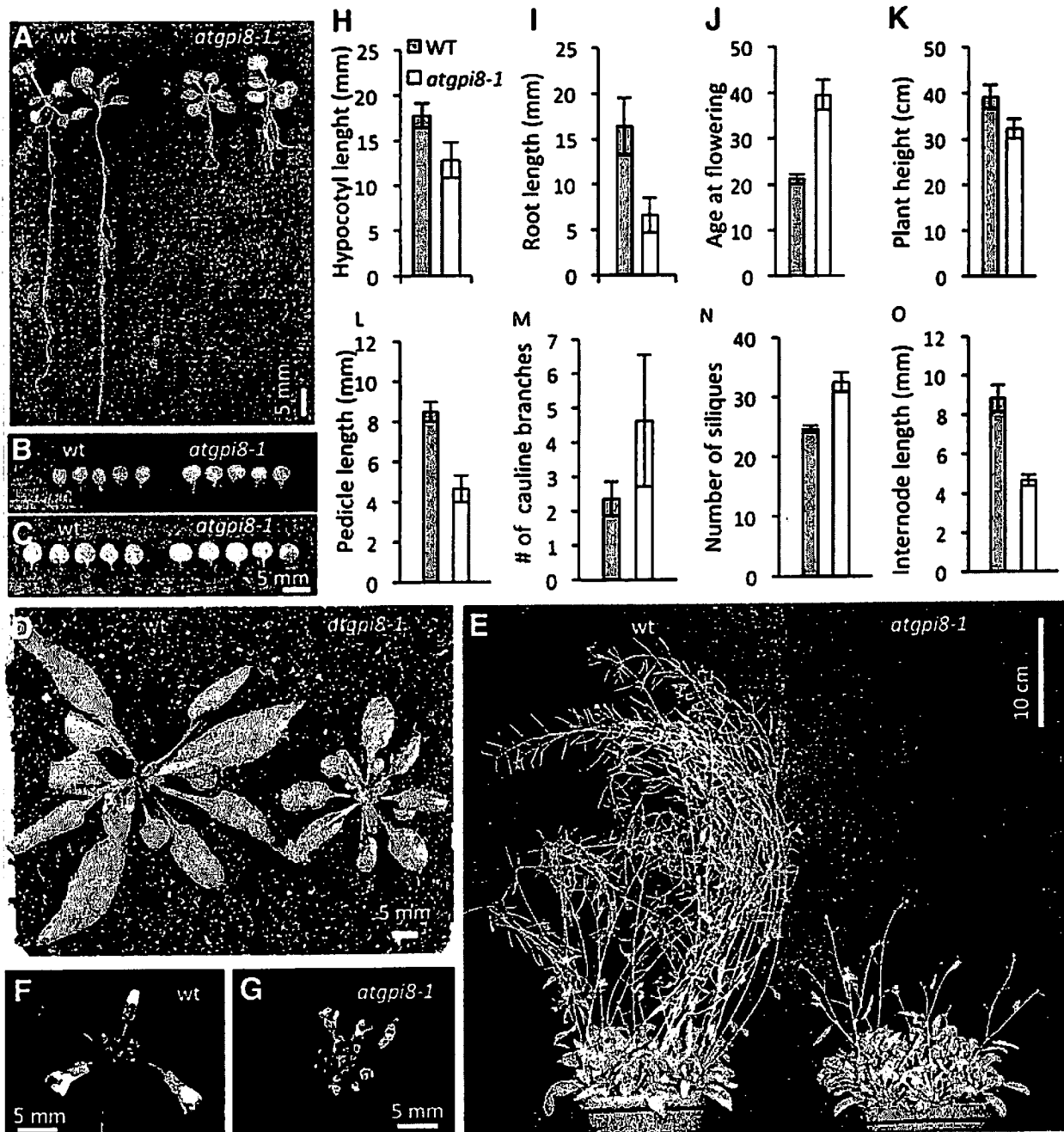
Reference:

Nolf M., Rosani A., Ganthaler A., Beikircher B., Mayr S., 2016. Herb hydraulics: Inter-and intraspecific variation in three *Ranunculus* species. *Plant physiology*, pp-01664; doi: 10.1104/pp.15.01664

Q.63

A protein can be integrated into a membrane through a polypeptide sequence or via a lipid anchor. The attachment of eukaryotic proteins to the outer leaflet of the plasma membrane occurs only via Glycosylphosphatidylinositol (GPI) anchors. The biosynthesis of GPI glycolipid is a multistep process relies on many proteins, including GPI transamidase. In *Arabidopsis* plants, *AtGPI8* gene encodes the enzyme GPI transamidase. To study the role of this gene in plant development, scientists constructed a mutant (*atgpi8-1*) plant line. He observed phenotypes of both wild type (WT) and mutant plants.

Fig.Q63-1. Cotyledonous epidermis of wild type (A) and *atgpi8-1* (B) plants

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Fig.Q63-2. Growth phenotypes of wild type and *atgpi8-1* plants. (A) Seedlings, (B)

cotyledons and (C) first two leaves of seedlings. (D) 30-day-old and (E) 60-day-old

plants. F- G: Inflorescences. H- L: Morphometric analysis of wildtype (gray bars) and

atgpi8-1 (white bars) seedlings and mature plants. All values in H to O are statistically

significant.

Indicate in the **Answer sheet** if each of the following statements is True or False.

A. The early post germination growth of cotyledons and first two leaves are not affected by the mutation. However, root growth, hypocotyl elongation and stomata differentiation are strongly affected by the mutation.

B. The data suggest that GPI anchoring promotes the growth of leaves in vegetative plants; however, it inhibits axillary shoot formation.

C. The *atgpi8-1* mutation leads to reduced internode and pedicel elongation. However, the height of *atgpi8-1* plants is only moderately reduced likely because the number of internodes is increased.

D. The results indicate that *AtGPI8* gene promotes early transition to flowering, flower formation and seed production.

Answer keys

A. True   **B.** True   **C.** True   **D.** False

Explanation

A. True. Cotyledons and first leaves are similar in both plant types (Fig.Q63-2A-C). Root and hypocotyl are longer in WT (Fig.Q63-2H-I). Number of stomata is higher in mutant plants (Fig.Q63-1).

B. True. In vegetative plants, leaves are bigger in WT compared to mutant plants (Fig.Q63-2D). Number of cauline branches is reduced in WT (Fig.Q63-2M). These indicate the role of AtGPI8 and GPI anchoring in the regulation of leaf growth (promotion) and axillary shoot formation (inhibition).

C. True. Internode and pedicel length is reduced in mutant plants (Fig.Q63-2F-G-L and O). Mutant plant height is smaller, but the difference compared to WT is not as much as internode length; hence, it must be due to the number of internodes is increased (Fig.Q63-2K and O).

D. False. Fertility and seed production are not mentioned in the data although number of cauline branches and number of siliques are increased in mutant plants, and wild type plants flowered earlier than the mutant plants did. (Fig.Q63-2E-F-G-J and N).
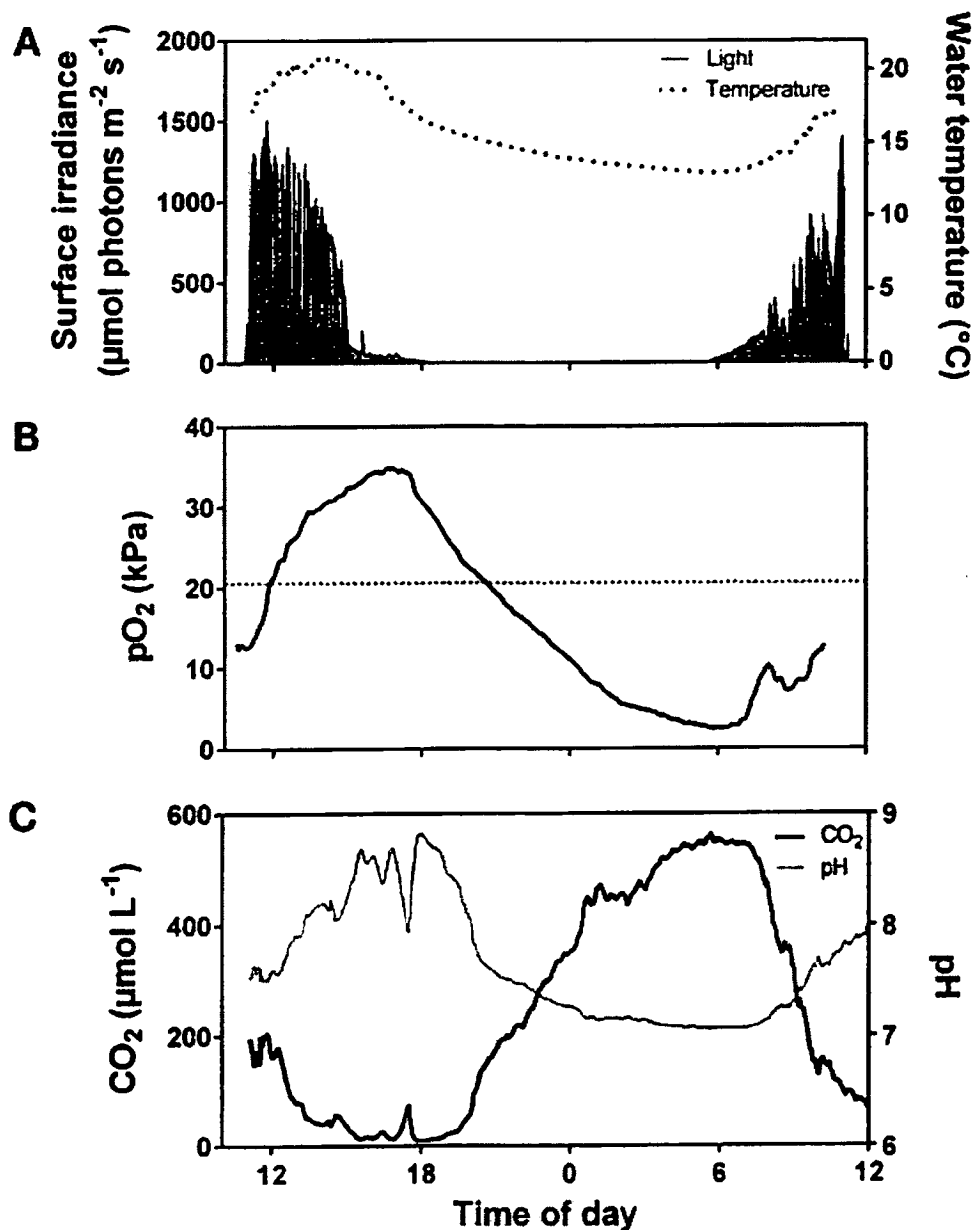
Reference.

Bundy M.G.R., Kosentka P.Z., Willer A.H, Zhang L., Miller E. and Shpak E.D., 2016. A Mutation in the Catalytic Subunit of the Glycosylphosphatidylinositol Transamidase Disrupts Growth, Fertility, and Stomata Formation. *Plant Physiol.*, Vol.171, pp. 974-985, doi: 10.1104/pp.16.00339

Q.64

Photosynthesis of submerged aquatic plants is severely impeded by many environment factors. In seawater and freshwater, light density and its spectrum is changed with depth in the water column and thus influence photosynthesis. Other factors affecting photosynthesis include level of carbon dioxide (CO_2) and oxygen (O_2).

Swamp Raspwort (*Meionectes brownie*) is a wetland plant species but can grow as a submerged aquatic plant in freshwater. An experiment was conducted to study the photosynthesis of the aquatic vegetation. Diurnal fluctuations in surface irradiance, partial pressure of O_2, CO_2 concentration and pH of the water in Swamp Raspwort-rich ponds are shown in Fig.Q64.

Fig.Q64

Indicate in the **Answer sheet** if each of the following statements is True or False.

33

A. In the underwater of ponds, light limitation appears early in the morning and co-limitation of both light and CO_2 takes place early in the afternoon.

B. The decrease in level of O_2 in water column during the night is caused by the Swamp Raspwort respiration.

C. In water column of ponds, CO_2 molecules are directly produced by respiration of Swamp Raspwort and by conversion from HCO_3^- at pH neutral results in increasing CO_2 level.

D. As indicated in the figure, temperature variation in ponds rich Swamp Raspwort is from 13 to 20°C. The alteration in temperature is mainly maintained by high density of this plant species.

Answer keys

A. False   B. True   C. True   D. False

Explanation

A. **False.** As indicated in the Fig.Q64 (A and C), limitation of both light and CO_2 occurs around 18 o'clock or late afternoon.

B. **False.** During the night, the photosynthesis of Swamp Raspwort is delayed while this species respires giving rise to decreasing O_2 level in its water column of ponds.

C. **True.** In water, CO_2 molecules are directly produced by respiration of aquatic organisms. At pH neutral, HCO_3^- is converted to CO_2 resulting in increasing CO_2 level.

D. **False.** The temperature in the water column of ponds rich Swamp Raspwort is maily controlled by solar irradiance. Therefore, the temperature of water in ponds decreases during night and increases during daylight.

Reference:

Pederson O., Colmer T.D. and Sand-Jensen K, 2013. Underwater photosynthesis of submersed plants- Recent advances and method. *Frontiers in Plant Science*, DOI: 10.3389/fpls.2013.00140.

Rich S.M., Pedersen O., Ludwig M. and Colmer T.D., 2013. Shoot atmospheric contact is of little importance to aeration of deeper portions of the wetland plant *Meionectes brownie*; submerged organs mainly acquire o2 from the water column or produce it endogenously in underwater photosynthesis. *Plant, Cell and Environment*, DOI: 10.1111/j.1365-3040.2012.02568.

Q.65

Nitrogen assimilation plays an important role in plant metabolism as well as in plant cell development. Plant cells can acquire inorganic nitrogen in the form of ammonium (NH_4^+) and nitrate (NO_3^-). When entering the plant cells through membrane-bound nitrate transporter (NRT), NO_3^- can be reduced to NO_2^- by nitrate reductase (NR) and subsequently to NH_4^+ and amino acids (AA). In addition, NO_2^- can be converted into nitric oxide (NO), then forming S-nitrosoglutathione (GSNO) by reaction with glutathione (GSH), and finally into oxidized glutathione (GSSG) and NH_4^+ under catalysis of S-nitrosoglutathione reductase 1 (GSNOR1).
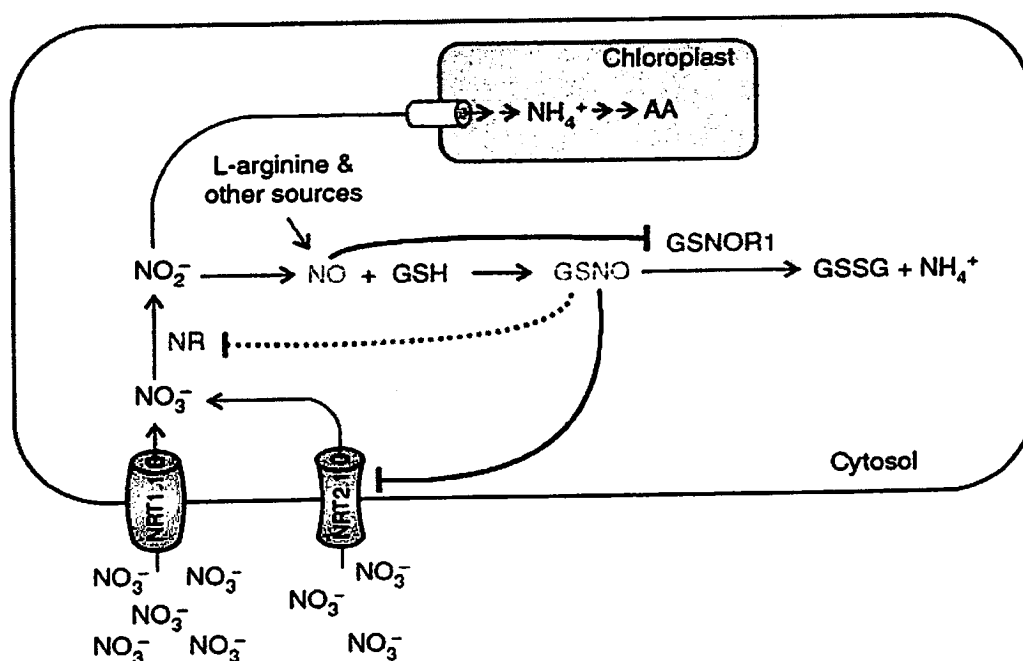

Fig.Q65. A schematic model for the control of nitrogen assimilation in plants through NO signalling.

Indicate in the **Answer sheet** if each of the following statements is True or False.

A. In the nitrogen metabolism process of the plant cells, NO is a one of the end products but plays a key role signaling regulation of NH_4^+ formation and NO_3^- assimilation.

B. NH^{4+} level in chloroplasts of plant cells is controlled by activity of GSNO and GSNOR1.

C. Reduction of NO^{2-} ions mainly occur in cytosol

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D. NO feedback regulates flux through nitrate assimilation pathway and controls its bioavailability by modulating its own metabolism.

Answer key

A. True   B. True   C. False   D. True

Explanation

A. True. In plant cell, NO_2^- is converted into nitric oxide (NO) in cytosol. NO inhibits nitrate reductase (NR) and membrane-bound nitrate transporter (NRT) to control NO_3^- uptake. In addition, by repressing GSNOR1, NO also reduces level of NH^{4+} in plant cell cytoplasms.

B. True. GSNO inhibits NO_3^- uptake and reduction giving rise to lower level of NO_2^- which subsequently converted into NO and finally into NH^{4+} by activity of GSNOR1.

C. False. NO_2^- is generally regarded as being toxic to cytosol of plant cells. It is promptly transported into chloroplasts where it is reduced by nitrite reductase (NR) into NH^{4+}

D. True. NO feedback is through GSNO

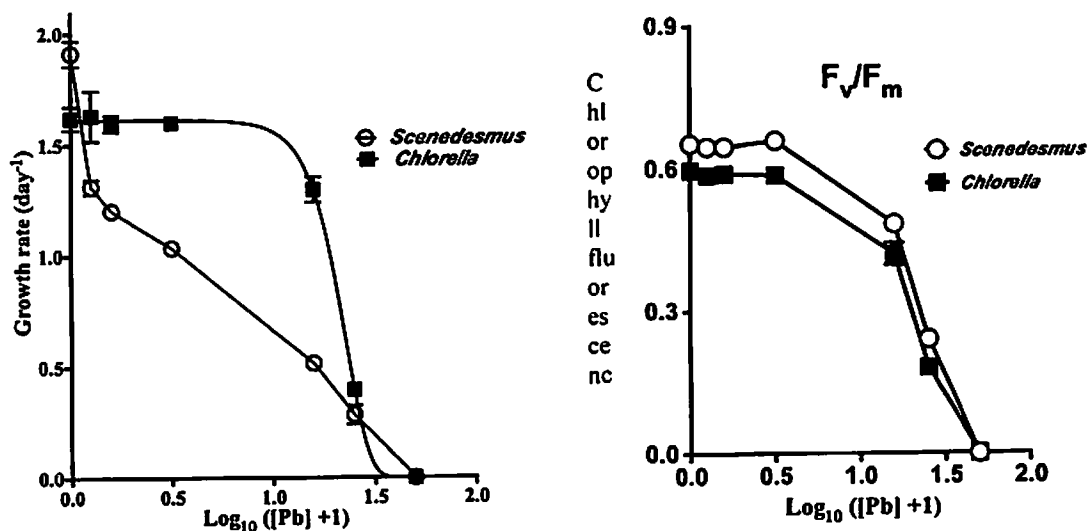**References**

Frungillo et al, *Nature Communication 2014*, DOI: 10.1038/ncomms6401

Campbell Biology 10th edition 2014, page 808

Q.66

A study on the effects of lead (Pb), a toxic heavy metal, on growth and photosynthesis of two microalgae, *Chlorella* and *Scenedesmus* was conducted. The figure on the left below shows the growth of these species responded differently to lead concentration after 4 day treatment. From growth rate (*Ke*), generation time (G) of each species at each concentration of lead can be calculated as equation: G= (ln2)/*Ke*. The right hand Fig.Q.66 is result of the effect of lead on photosynthesis of these species, indicated by Fv/Fm, a sensitive parameter that decreases when photosynthesis is impaired. The concentration of lead that gives half-maximal response, the IC_{50}, can be estimated based on response versus lead concentration plots.

Fig.Q66

Indicate in the **Answer sheet** if each of the following statements is True or False.

A. Estimated IC_{50} of growth for the *Scenedesmus* was higher than that for the *Chlorella*.

B. Photosynthetic impairment by Pb was likely responsible for the growth decrease in the *Chlorella* but this was not the scenario in the *Scenedesmus*

C. The estimated IC_{50} for effects on Fv/Fm was higher than that for growth in *Scenedesmus*

D. At lead concentration that $Log_{10}([Pb]+1)$ is 0.5, the *Scenedesmus* reproduced faster than the *Chlorella* did.

Answer keys

A. False   B. True   C. True   D. False

Explanation

A. False. Based on the y axis, half of the growth rate of each strain can be estimated. The corresponding lead concentration on x axis for the *Scenedesmus* was lower than that for the *Chlorella*.

B. True. In the *Chlorella*, growth started decreasing strongly at the same toxic threshold at which photosynthetic function was inhibited, suggesting impairment of photosynthesis was behind the decrease in growth. In contrast, in *Scenedesmus*, growth was inhibited gradually at lower lead concentrations, under which photosynthesis was not affected, implying that growth in this alga was inhibited in other ways.

C. True. Based on the y axis, half of the response (growth or Fv/Fm) of *Scenedesmus* can be estimated. The corresponding lead concentration on x axis for the growth was lower than that for Fv/Fm.

D. False. At the lead concentration, *Ke* of the *Scenedesmus* was lower than that of the *Chlorella*. Therefore, generation time of *Scenedesmus* was higher than that of the *Chlorella*. In other words, the *Chlorella* reproduced faster than the *Scenedesmus* did.

Reference

Ly H.T. Dao, John Beardall, 2016. Effects of lead on growth, photosynthetic characteristics and production of reactive oxygen species of two freshwater green algae, *Chemosphere*, vol. 147, 420-429.
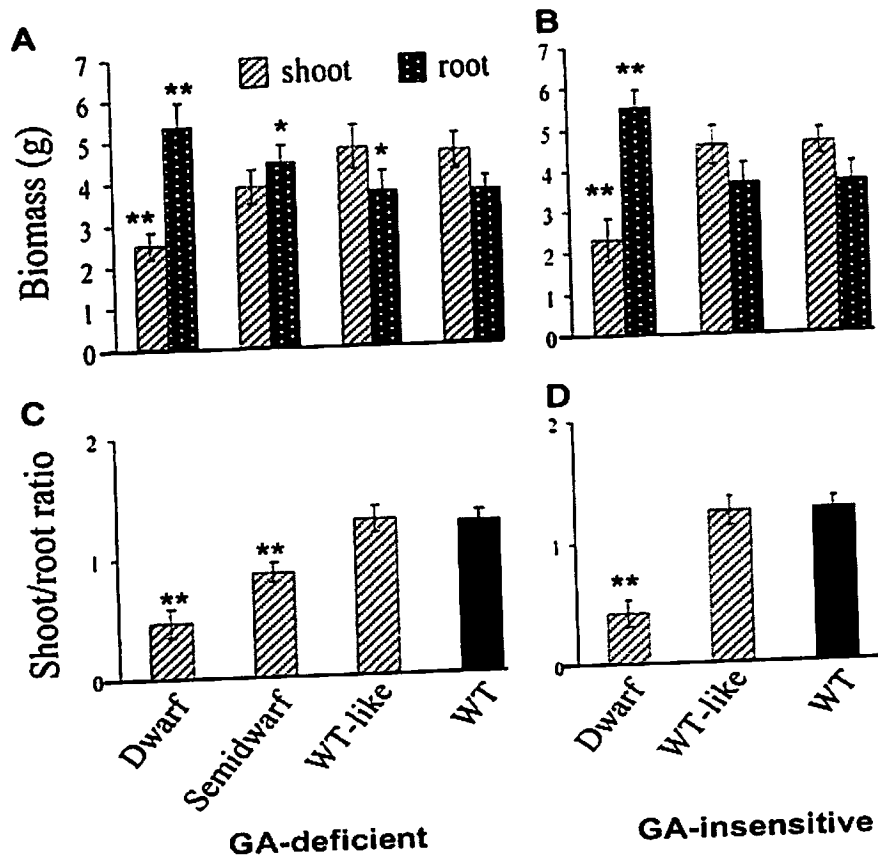
Q.67

Scientists constructed gibberellic acid (GA)-deficient and GA-insensitive transgenic lines of poplar plants. They measured concentration of phytohormones, including GA1 and GA4 and IAA, in the leaves and roots of transgenic and wildtype plants (Table Q67). They also measured the growth of plants in greenhouse and *in vitro* conditions (Fig.Q67-1 and Fig.Q67-2).
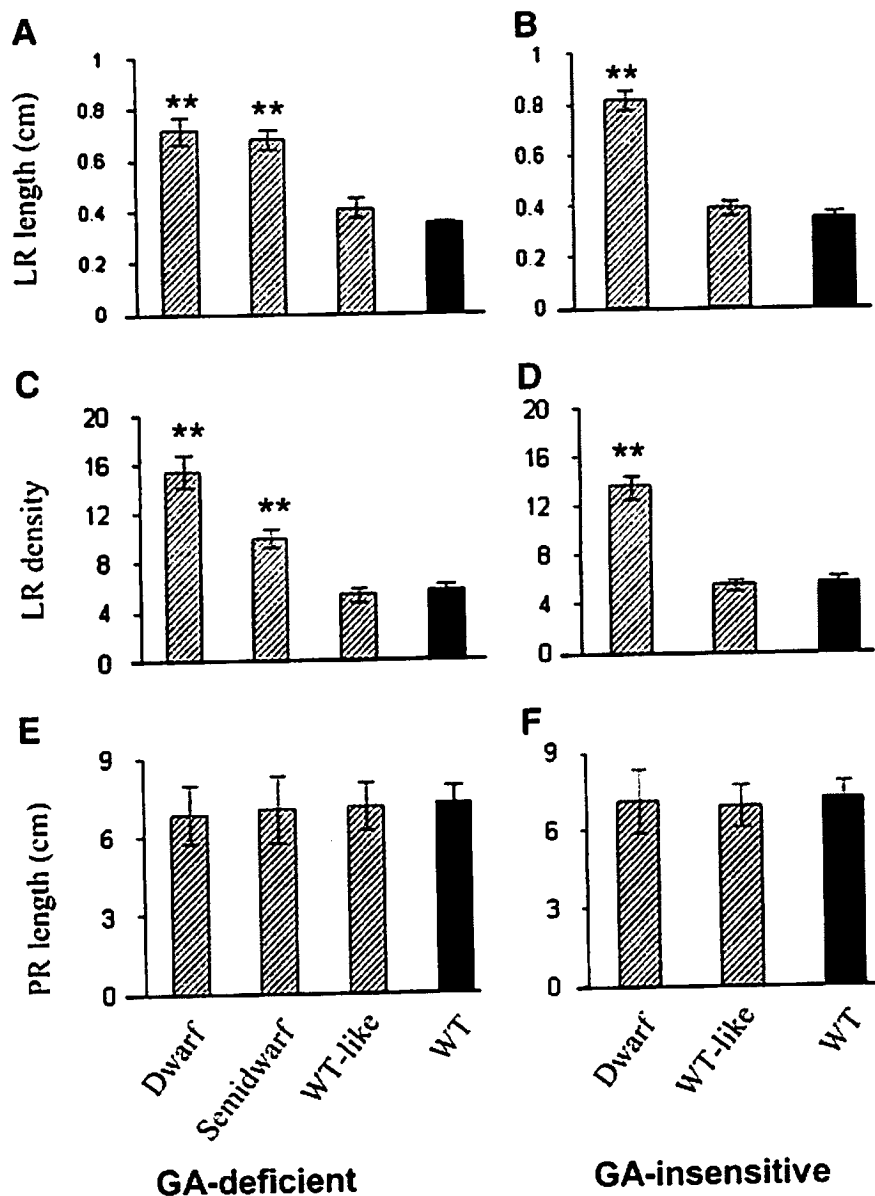
Table Q67. Phytohormone concentrations (ng/g dry weight) in leaves and roots of the wild type and the two transgenic types

Organ	Plant types	GA1	GA4	IAA
	Wild-type	58.1 ± 15.4	6.64 ± 3.18	22.5 ± 3.1
Leaf	GA-deficient	19.9 ± 9.4**	5.53 ± 2.33*	21.1 ± 5.9
	GA-insensitive	139.6 ± 21.9**	12.2 ± 3.6**	19.6 ± 3.7
	Wild-type	77.1 ± 29.3	2.24 ± 0.74	61.4 ± 4.1
Root	GA-deficient	48.8 ± 9.6**	1.15 ± 0.62**	72.9 ± 5.2*
	GA-insensitive	97.7 ± 31.5**	3.93 ± 0.68**	69.1 ± 9.7*

* and ** indicate significant differences compared to wild-type at 0.05 and 0.01 levels by Student T-test.

Fig.Q67-1. Root and shoot biomass under greenhouse conditions. Top panel shows the fresh biomass of shoots and roots in GA-deficient (A) and GA-insensitive (B) transgenics. Bottom is the shoot/root ratio in GA-deficient (C) and GA-insensitive (D) transgenics. * and ** indicate significant differences.

Fig.Q67-2. Root development in GA-deficient and GA-insensitive transgenic lines grown *in vitro*. LR - Lateral root; PR - Primary root. ** indicates significant differences compared to wild-type plants (WT).

Indicate in the **Answer sheet** if each of the following statements is True or False.

A. Greenhouse-grown dwarf and semidwarf plants of both transgenic types display a significant reduction in aerial biomass and an increase in belowground biomass, leading to a significant reduction in the shoot-to-root ratio relative to the wild-type control.

B. The most severely dwarfed plants have more, as well as longer, lateral roots than the wild-type control.

C. The degree of dwarfism in both GA-insensitive and GA-deficient lines is positively correlated with the extent of primary and lateral root formation and elongation.

D. In poplar plants, gibberellins negatively affect lateral root formation, and there may be an interaction between gibberellins and auxin that regulates lateral root formation.

Answer key

A. True   **B.** True   **C.** False   **D.** True.

Explanation:

A. True. A reduction of aerial biomass, an increase in belowground biomass, and a reduction in the shoot-to-root ratio are obtained in dwarf and semidwarf transgenic plants (Fig.Q67-1).

B. True. The most severely dwarfed events have two to three times more, as well as longer, lateral roots than the wild-type control (Fig.Q67-2).

C. False. The degree of dwarfism is not positively correlated to primary root elongation (Fig.Q67-2).

D. True. In both GA-insensitive and GA-deficient transgenic lines, lateral root formation is increased compared to wild-type, indicating that GAs inhibit lateral root formation. The increase of lateral root formation in transgenic plant is associated with the increase of IAA concentration, indicating that it promotes root formation. Taken together, GAs may interact with auxin (IAA) to regulate lateral root formation.
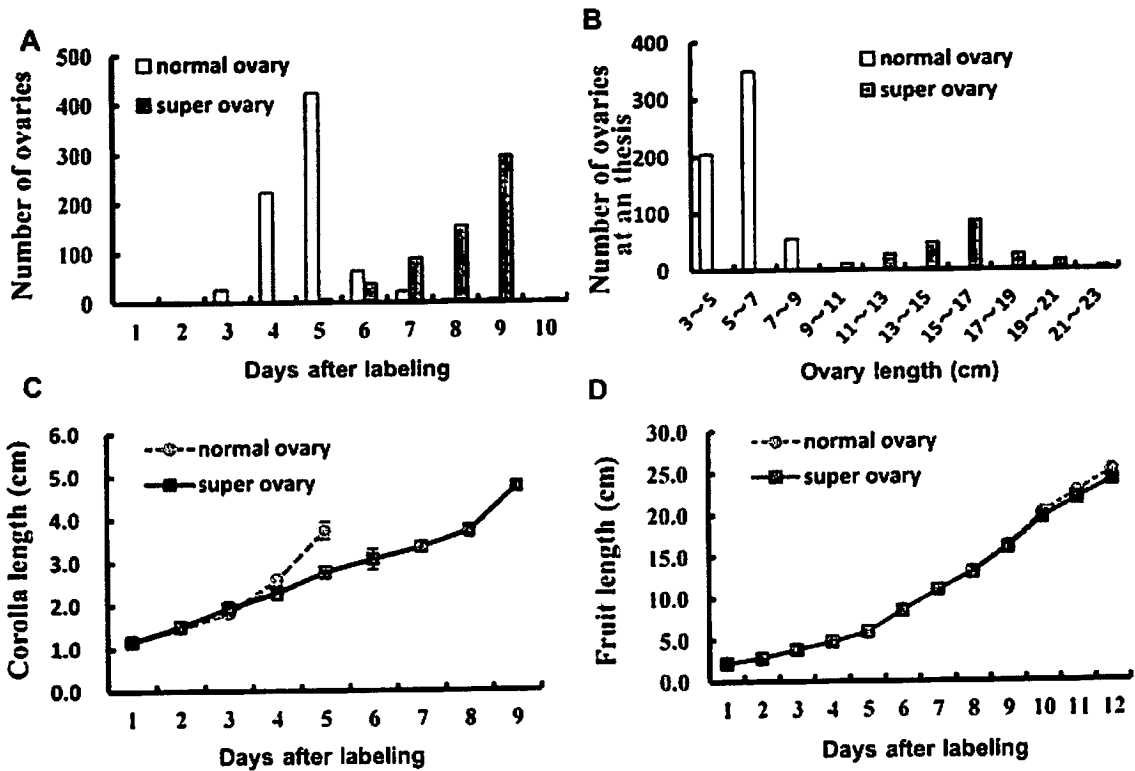
References:

Jiqing Gou et al., The Plant Cell 22.3 (2010): 623-639.

Kathleen L. Farquharson, The Plant Cell 22.3 (2010): 540-540.

Q.68

Scientists grew cucumber plants in different nutrient conditions to generate obtain either super ovary or normal ovary types. They labelled flowers when they emerged and observed the development of flowers. Based on the color and shape, corolla development was divided into four stages: green bud (G), green-yellow bud (GY), yellow bud (Y), and flowering (F). They also measured plant growth regulator concentrations in different flower developmental stages.

Fig.Q68-1. Morphological characterization of the normal ovary and super ovary types.

Fig.Q68-2. Concentration of cytokinins (IPA, ZR, DHZR), gibberellins (GA3, GA4) and auxin (IAA) in different flower developmental stages. In super ovary type, two sub-stages were included. ** indicates statistically significant differences within one developmental stage.

Indicate in the **Answer sheet** if each of the following statements is True or False.

A. The corolla progression between stages was much delayed in the super ovary

B. The ovary at anthesis was on average much longer in the super ovary than the normal ovary, while at the same time courses after labelling, fruit length was not different between two types.

C. Gibberellins were increased in the super ovary during the early stages of corolla development, which corresponds to the enlarged corolla size

D. Cytokinins appear to be the primary regulator for flower opening in cucumber, whereas auxin is probably involved in the size control of corolla and fruit.

Answer keys

A. True   B. True   C. True   D. False

Explanation

A. **True.** Corrolla onpening in normal ovary occurs mostly before 5 days after labelling, while in super ovary it occurs nearly 9 days after labelling (Fig.Q68-1A and 1C).

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B. True. Ovaries at anthesis in super ovary type are longer than those in normal ovary type (Fig.Q68-1B), while fruit length of two types in the same days is not different (Fig.Q68-1D).

C. True. Gibberellins concentrations in super ovary during GY and Y stages are generally higher than those in normal ovary type (Fig.Q68-2D and 2E).

D. False. Although flower opening is generally associated with the difference of cytokinins' concentration, auxin concentration is not different in two ovary types (Fig.Q68-2A, C, 2F).
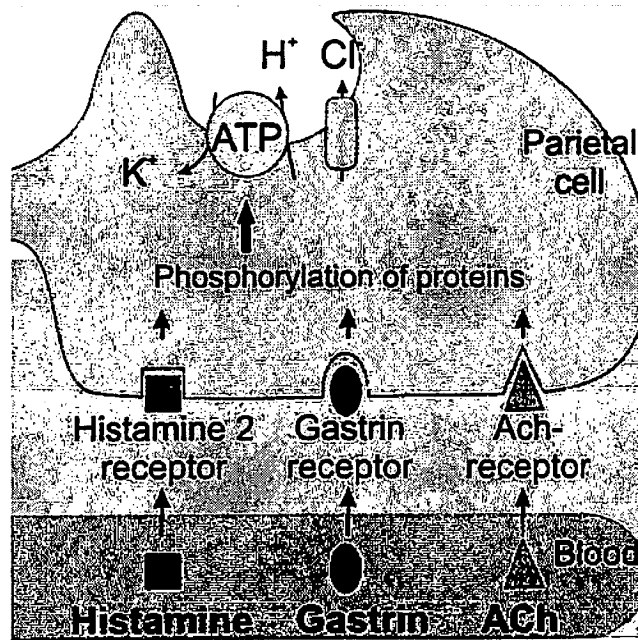
Reference

Chengzhen Sun et al., Plant Physiolog, 2016, Vol. 171, pp. 1209–1229

ANIMAL ANATOMY AND PHYSIOLOGY

Q.69

Figure Q.69 shows the regulation of HCl secretion in the parietal cell of the stomach.

Figure Q.69.

Four drugs (Drugs 1 to 4) differently inhibit gastric acid secretion *in vivo* via one of the four pathways: inactivating the H^+/K^+ATPase, blocking the Histamine 2 receptor, blocking Gastrin receptor, and blocking Acetylcholine (Ach) receptor.

A set of experiments were conducted to determine in which pathway these drugs inhibit gastric acid secretion. Parietal cells were isolated and cultured in different media. Each medium contained one of the four drugs (Drugs 1 to 4). Each drug-containing medium was added with one of three compounds (Histamine, Gastrin, Ach). The parietal cells cultured in the medium without drug and compound were the control. The HCl secretion of parietal cells in the cultures was determined and compared with the control. The following table shows the results of the experiments.

	No drug	Drug 1	Drug 2	Drug 3	Drug 4
No addition	–	–	–	–	–
Histamine added	?	?	?	?	–
Gastrin added	?	?	?	+	?

Ach added	+	−	?	?	−

(−: No HCl secretion; +: HCl secretion; ?: not shown).

Indicate in the Answer sheet if each of the following statements is True or False.

A. HCl was secreted by the parietal cells cultured in the medium containing Drug 1 and Histamine.

B. Drug 2 blocked Gastrin receptors.

C. Drug 3 blocked Histamine 2 receptors .

D. The parietal cells cultured in the medium containing Drug 4 and Ach had lower levels of intracellular K^+ than the cells cultured in the medium containing Ach.

Answer key

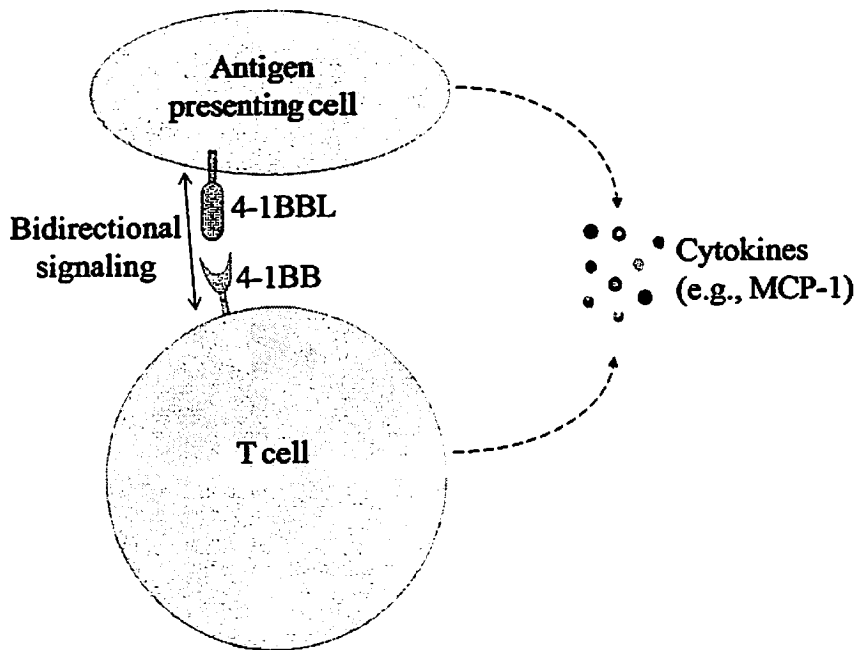   **A. True**   **B. True**   **C. True**   **D. True**

Explanation

A. True. Drug 1 blocked Ach receptors and did not block Histamine receptors.

B. True. From the data shown, we can see that Drug 1 blocked Ach receptors, Drug 4 inactivated H+/K+ APTase, Drug 3 did not block Gastrin receptors. Therefore, Drug 2 blocked Gastrin receptors.

C. True. Drug 1 blocked Ach receptors, Drug 4 inactivated H+/K+ APTase, Drug 3 did not block Gastrin receptors. Therefore, Drug 3 blocked Histamine receptors.

D. True. Drug 4 inactivated H+/K+ ATPase. Thus, it decreased the flow of K+ pumped into the parietal cells in the medium containing Ach.

Reference

Gustavo Zubieta-Calleja and Poul-Erik Paulev, *2004. New Human Physiology 2nd Edition.chapter 22.*

Q.70

The glycoprotein 4-1BB is a receptor that is highly expressed on the surface of active T-cells. The 4-1BB ligand (4-1BBL) is a molecule that binds to and activates 4-1BB. It is found strongly expressed on antigen-presenting cells. Bidirectional signals of 4-1BB and 4-1BBL interaction increase the activity of white blood cells and increase the production and secretion of cytokines, such as MCP-1 that is an important factor promoting the infiltration of leukocytes (Figure Q.70). Currently, many studies have shown a relationship between the signaling pathways via 4-1BB/4-1BBL interaction and several human diseases, including those related to metabolism.

Figure Q.70.

Indicate in the Answer sheet if each of the following statements is True or False.

A. Inhibition of 4-1BB expression diminishes the development of atherosclerosis.

B. Activation of 4-1BB limits the effect of autoimmune diseases on the body.

C. All three kinds of cells, macrophages, dendritic cells and natural killer cells strongly express 4-1BBL.

D. Blockade of the 4-1BB and 4-1BBL interaction increases graft tolerance.

Answer key:

A. True   B. False   C. False   D. True

Explanation

A. True. The infiltration of leukocytes to the lipid accumulated positions in the blood vessels plays an important role in the development of atherosclerosis. The presence of white blood cells accelerates the process of fat accumulation and plaque formation in the blood vessel walls. Inhibiting expression of 4-1BB reduces the activity of leukocytes and cytokine secretion, such as MCP-1, resulting reduced activity and infiltration of those cells. These results help to prevent and limit the development of atherosclerosis.

B. False. When the immune system attacks self-molecules, autoimmune diseases occur. This is the result of an overactive immune response, including the activity of the T cells. Activation of 4-1BB enhances the activity of T cells, thus, exacerbating the effects of autoimmune diseases.

C. False. Macrophages, dendritic cells, and B cells (Not natural killer cells) are typical antigen presenting cells that strongly express 4-1BBL.
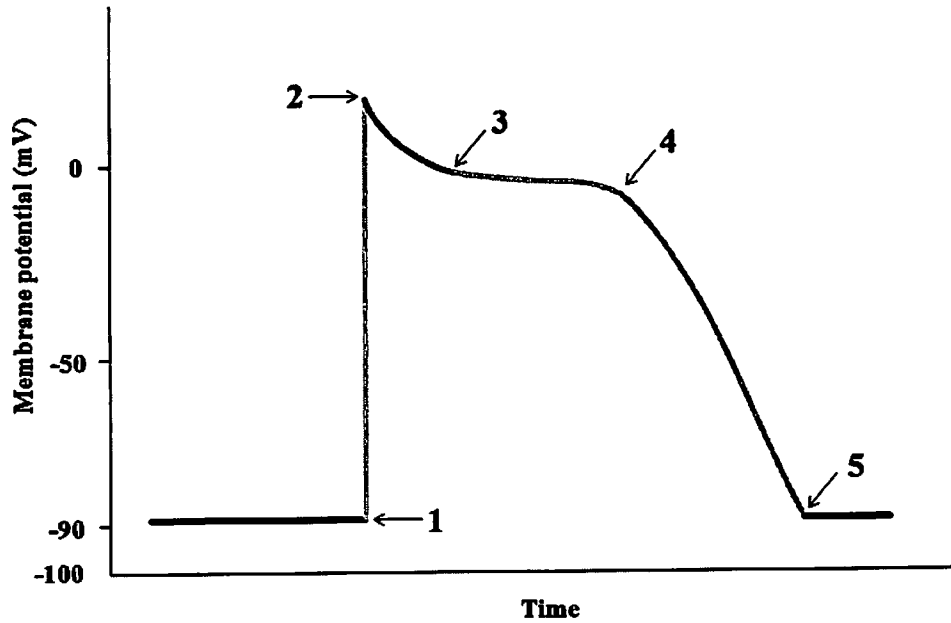
D. True. Graft rejection is due to the activity of immune cells as antigen-presenting cells presenting foreign molecules (antigens) from graft to cause T cells to disable antigens. Signals via 4-1BB and 4-1BBL enhance the activity of immune T cells and antigen-presenting cells. Therefore, blockade of 4-1BB and 4-1BBL interaction increases graft tolerance.

References

1. Byungsuk Kwon. CD137-CD137 Ligand Interactions in Inflammation. Immune Netw. 2009 Jun; 9(3): 84–89.

2. Hyung Jun Jeon et al. CD137 (4-1BB) Deficiency Reduces Atherosclerosis in Hyperlipidemic Mice. Circulation /2010; 121(9):1124-33.

3. Qin L et al. Blockade of 4-1BB/4-1BB ligand interactions prevents acute rejection in rat liver transplantation. Chin Med J (Engl). 2010 Jan 20;123(2):212-5.

Q.71

The action potential of cardiac muscle cells differs from that of other cells such as skeletal muscle cells and neurons. Figure Q.70 displays the different phases of action potential in cardiac muscle cells.

Figure Q.71

Indicate in the Answer sheet if each of the following statements is True or False.

A. A substance that inhibits the reuptake of Ca^{2+} into the sarcoplasmic reticulum increases the time interval from 3 to 4.

B. The concentration of K^+ in the sarcoplasm at position 2 is higher than that at position 3.

C. Injection of adrenaline decreases the time interval from 1 to 5.

D. The height of membrane potential (from 1 to 2) is decreased when the sarcoplasmic level of Na^+ is higher than the normal level.

Answer key

  A. True   B. True   C. True   D. True

Explanation

A. True. Inhibition of the reuptake of Ca^{2+} into the sarcoplasmic reticulum results in longer time of Ca^{2+} remaining in the sarcoplasm. This prolongs the plateau phase (phase from 3 to 4).

B. True. A mild decrease in membrane potential from 2 to 3 is due to the closure of voltage-gated Na^+ channels and the opening of voltage-gated K^+ channels. At position 3, more K^+ ions are transported out of the cells, leading to lowered concentration of K^+ in the cells

C. True. Adrenalin increases heart rate via increase in frequency of potential impulse resulting in shortening of a potential impulse time.
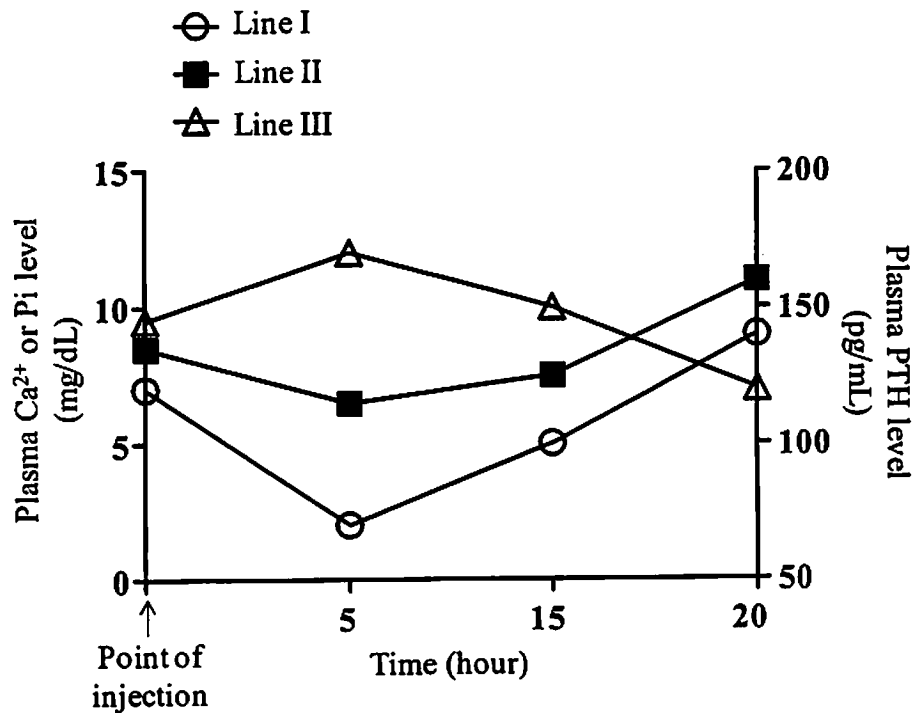
D. True. When the concentration of Na^+ inside of the cells is higher than the normal level, the flow of Na^+ from outside to inside of the cells is decreased. Thus, the amplitude of action potential is decreased.

Reference

Life: the science of biology, 9th edition, 2012. p1055.

Q.72

Parathyroid hormone (PTH) plays an important role in the regulation of plasma calcium and phosphate levels. Figure Q.72 shows the changes in levels of PTH, Ca^{2+}, and phosphate (Pi) in plasma of mice injected with a specific inhibitor of PTH secretion.

Figure Q.72.

Indicate in the Answer sheet if each of the following statements is True or False.

A. If Line I shows the level of PTH, then Line II and Line III would likely be showing the levels of Pi and Ca^{2+}, respectively.

B. PTH knock-out mice would have higher Pi levels in their urine compared with the wild type mice on the same diet.

C. Eating a calcium-rich diet decreases the plasma level of vitamin D (active form) in healthy people.

D. People with calcium-sensing receptor suppression have higher levels of plasma Ca^{2+} compared with healthy people on the same diet.

Answer key

A. False   B. False   C. True   D. True

Explanation

A. False. PTH enhances absorption of Ca^{2+} in intestine and stimulates the release of Ca^{2+} from bone. In the kidney, PTH increases reabsorption of Ca^{2+} and excretion of Pi. Therefore, if Line I were specific for PTH, Line II must be specific for Ca^{2+} and Line III for Pi.

B. False. PTH increases the release of Pi via urine. PTH knock-out mice thus would have lower Pi in their urine.

C. True. Eating a calcium-rich diet results in increased levels of plasma Ca^{2+}. This leads to inhibition of PTH secretion based on the negative feedback loop. Lack of PTH thus lowers the process that modifies vitamin D to the active form.

D. True. Increase in the levels of plasma Ca^{2+} after a meal stimulates calcium-sensing receptor in the cells of parathyroid gland leading to inhibition of PTH secretion. Thus, people with calcium-sensing receptor suppression have high levels of PTH and plasma Ca^{2+}.

References

1. Stephen J. Quinn et al. Interactions between calcium and phosphorus in the regulation of the production of fibroblast growth factor 23 in vivo. Am J Physiol Endocrinol Metab 304: E310–E320, 2013.

2. J. Ruth Wu-Wong et al. Mechanistic analysis for time-dependent effects of cinacalcet on serum calcium, phosphorus, and parathyroid hormone levels in 5/6 nephrectomized rats. Physiol Rep, 1 (3), 2013, e00046.

3. Ogo I. Egbuna. Hypercalcaemic and hypocalcaemic conditions due to calciumsensing receptor mutations. Best Pract Res ClinRheumatol. 2008 March ; 22(1): 129–148.

Q.73

The following table describes the rate of blood flow to different parts of the body including cardiac muscle, brain, skin, and intestines at rest and during strenuous exercise.

Part of the body	Rate of blood flow/cm^3/min	
	At rest	During exercise
I	250	1200
II	500	500
III	500	1000
IV	2500	90

Indicate in the Answer sheet if each of the following statements is True or False.

A. At rest, ATP of the cells of part I comes mainly from oxidation of fatty acid.

B. The activity of insulin receptors in the cells of part II is increased during exercise, enhancing glucose uptake.

C. The increase in blood flow to part III during exercise helps to regulate the body temperature.

D. Epinephrine decreases blood flow to part IV via β-receptor.

Answer key

      A. True B. False C. True D. False

Explanation

      During exercise, the sympathetic activity increases, leading to increased rate of blood flow to cardiac muscle and skins, and decreased rate of blood flow to intestines. The increase in the rate of blood flow to cardiac muscle is higher than that to skin. The rate of blood flow to brain does not change during exercise.

      A. True. Part I is cardiac muscle. At rest, the heart works constantly and requires a high amount of ATP that comes mainly from oxidation of fatty acids because fatty acids provide higher energy than glucose. Indeed, about 60% of ATP of the cardiac muscle cells comes from oxidation of fatty acid and about 35% from oxidation of glucose.

B. False. II is brain. The cells of this part do not use insulin signals to uptake glucose.

C. True. Part III is skin. The increase in blood flow to skin during exercise helps to reduce the body temperature.

D. False. Part IV is intestines. Epinephrine decreases blood flow to the digestive tract via α-receptor (NOT β-receptor).

Reference

Glenn and Susan Toole, 2013. Biology in context for Cambridge International A level. Nelson Thornes, pp.154

Rod R. Seeley, 2000. Anatomy and Physiology, pp. 618.

Q.74

A man had lost approximately 700 mL of blood in a severe injury of a major artery in a motorcycle accident. At the time of accident, his blood pressure was 90/50 mmHg. Several physiological changes should be expected in response to hemorrhage.

Indicate in the Answer sheet if each of the following statements is True or False.

A. Oxygen affinity of hemoglobin was increased getting more oxygen for cellular respiration.

B. Total peripheral resistance was increased.

C. Hyperpolarization occurred in the cells of the sinoatrial node.

D. Vasoconstriction occurred in the brain and in the coronary arteries.

Answer key

A. False   B. True   C. False   D. False

Explanation

A. False. Hemorrhage leads to decreasing oxyhemoglobin within red blood cells. This promotes red blood cells producing more 2,3-BPG, which in turn decreases the affinity of hemoglobin for oxygen, so that oxygen is unloaded from hemoglobin more easily at the tissues.

B. True. Decreased blood pressure causes an increase in activity of the sympathetic nervous system, leading to constriction of peripheral blood vessels. Hence, the peripheral resistance is increased.

C. False. Hyperpolarization did not occur in the cells of the sinoatrial node because norepinephrine released from the sympathetic nerve ending causes depolarization of the sinoatrial node.
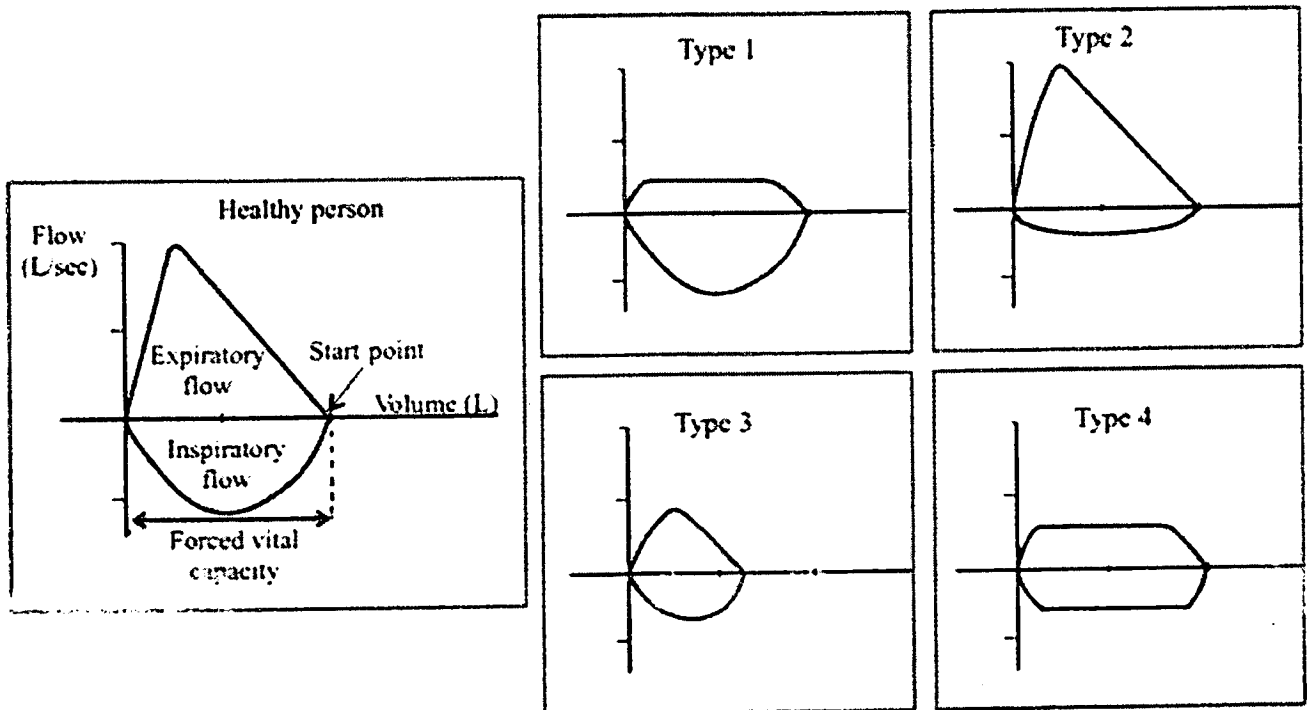
D. False. Brain and heart are key vital organs and are always guaranteed blood amount supplied. Vasoconstriction does not occur in both brain and in coronary arteries in response to hemorrhage.

Reference

Guyton, A.C and Hall, J.E, *Physiology Review*, 2006

Q.75

In a simple way, respiratory disorders can be classified as obstructive or restrictive disorders. Obstructive disorders are characterized by a reduction in the airflow rate in the respiratory tracts. Restrictive disorders are characterized by a reduction of lung volume. Figure Q.75 shows the shapes of the flow-volume loops measured during forced inspiration and forced expiration in healthy people with normal respiratory function and in four patients suffering from four common types of respiratory disorders.

Figure Q.75.

Indicate in the Answer sheet if each of the following statements is True or False.

A. The blood pH of patient with Type I is higher than that of healthy people.

B. The time of the forced inhalation of patient with Type 2 is shorter than that of healthy people.

C. A patient with Type 3 displays higher breathing rate than healthy people.

D. Residual volume in a patient with Type 4 is higher than that of healthy people.

Answer key

A. False   B. False   C. True   D. True

Explanation

A. False. Because of the decreased forced expiratory flow in Type 1, leading to increased CO_2 accumulation in the lungs. This results in decreased blood pH.

B. False. Owing to lowered inspiratory flow in Type 2, the time of forced inhalation is longer.

C. True. Type 3 displays decrease in airflow and lung volume. These result in increase in breathing rate.

D. True. Type 4 shows limitation of airflow in both inspiration and expiration so that the residual volume is increased.

Reference

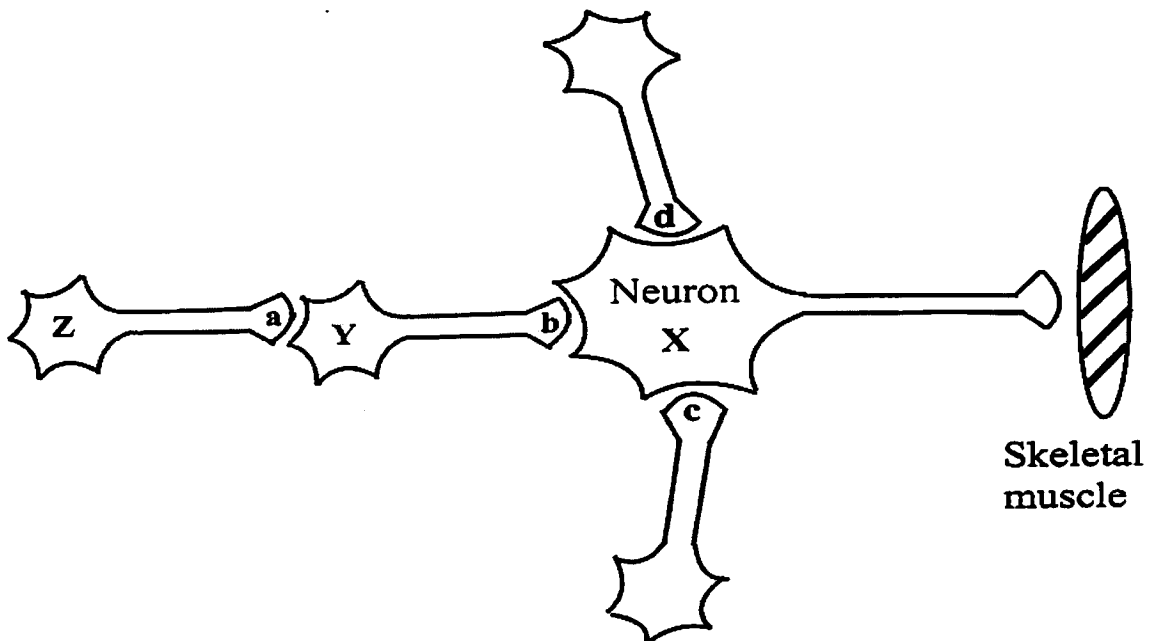Thomas R. Gildea, Kevin McCarthy, 2010. Pulmonary Function Testing.

Q.76

Figure Q.76A shows neuron X receives signals directly from three separate nerve terminals b, c and d. Neuron Y receives signals from nerve terminal a. .

Figure Q.76B shows the various postsynaptic potentials recorded in neuron X after receiving input signals directly from terminals b, c, and d.

Figure Q.76C shows the action potential recorded in neuron Y after receiving input signals from the presynaptic terminal a.

Figure Q.76A

Figure Q.76B.

Figure Q.76C.

Indicate in the Answer sheet if each of the following statements is True or False.

A. Action potentials could be generated in neuron X if nerve terminal c is stimulated rapidly.
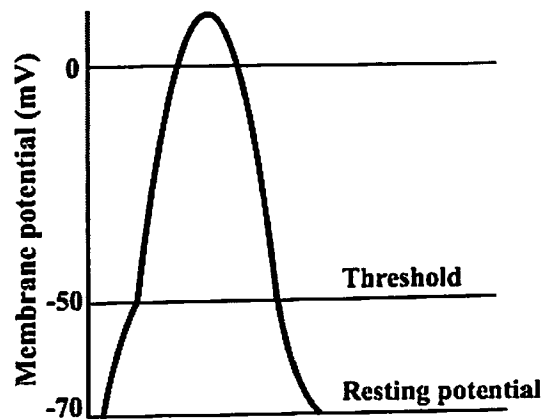
B. When three nerve terminals a, c and d are stimulated simultaneously, the postsynaptic potentials recorded in neuron X are smaller than those when the nerve terminals c and d are stimulated simultaneously.

C. Nerve terminal a releases inhibitory neurotransmitter and nerve terminal b releases excitatory neurotransmitter.

D. In the mammalian body, there are many neurons X, Y, Z and interneurons. Neurons Z, Y, X are sensory neurons, Renshaw cells (inhibitory neurons) and motor neurons, respectively. If a substance (e.g., Strychnine) injected in the body blocks glycine receptors, diaphragm contracts fully and remains contracted.

Answer key

A. True   B. True   C. False   D. True

Explanation

A. True. Action potentials could be generated in neuron X if nerve terminals c is stimulated rapidly because the resultant excitatory postsynaptic potentials (EPSPs) combine in a process called temporal summation.

B. True. When nerve terminals a, c and d are stimulated simultaneously, the postsynaptic potentials recorded in neuron X are of the same value as those recorded when only nerve terminal c or d is stimulated. When nerve terminals c and d are stimulated simultaneously, the resultant EPSPs combine in a process called spatial summation to cause higher postsynaptic potentials.

C. False. Nerve terminal a releases excitatory neurotransmitter which generates action potentials in neuron Y. This in turn causes nerve terminal b to release inhibitory neurotransmitter.

D. True. Normally interneurons in the spinal cord called Renshaw cells release neurotransmitter glycine at inhibitory synapses in somatic motor neurons. This inhibitory input to their motor neurons prevents excessive contraction of skeletal

muscles. If strychnine blocks glycine receptors, the motor neurons generate nerve impulses without restraint causing the skeletal muscles including diaphragm to contract fully and remain contracted.

Reference

Reece. J.B.. Campbell biology 10th edition, 2013. pp.1073.

Sadava. D. The Life 9th edition, 2011, pp. 958-959.

Q.77

A set of experiments on the regulation of hormone secretion and effects of various drugs on the activities of endocrine glands were conducted on rats. Rats were divided to different groups and each group was injected with a hormone or a drug. Some physiological parameters were collected and analysed.

Indicate in the Answer sheet if each of the following statements is True or False.

A. The group of rats injected with the drug that reduces the sensitivity of hypothalamus to cortisol resulted in higher plasma levels of both glucose and insulin than those in the group of rats injected with the drug that reduces the sensitivity of adrenocorticotropic hormone (ACTH) receptors.

B. The group of rats injected with the drug that increases the sensitivity of hypothalamus to thyroxine resulted in higher metabolic rate and body temperature than those in the group of rats injected with the drug that increases the sensitivity of target cells to thyroid releasing hormone (TRH).

C. The group of rats injected with propylthiouracil (which blocks thyroid hormone synthesis) resulted in smaller thyroid gland and body weight than those in the group of rats injected with placebo.

D. The group of rats injected with thyroid stimulating hormone (TSH) had smaller pituitary gland and bigger adrenal glands compared with the group of rats injected with corticotrophin-releasing hormone (CRH).

Answer key

A. True   B. False   C. False   D. False

Explanation

A. **True.** If the sensitivity to cortisol is reduced, hypothalamus increases CRH secretion causing the pituitary gland to increase ACTH secretion. This results in increased cortisol secretion. High levels of cortisol cause increasing plasma glucose concentration, which in turn simulates insulin secretion. If the sensitivity of ACTH receptors is reduced, the adrenal glands become smaller leading to decrease in both

plasma cortisol and glucose concentration. Low level of plasma glucose concentration reduces insulin secretion.

B. False. If sensitivity to thyroxine is increased, hypothalamus reduces TRH secretion causing the pituitary gland to reduce TSH secretion, which in turn reduces thyroxine secretion. Low levels of thyroxine causes both low metabolic rate and low body temperature. If the sensitivity to TRH is increased, the pituitary gland increases TSH secretion, which in turn stimulates thyroxine secretion. High level of thyroxine cause both high metabolic rate and high body temperature.

C. False. Propylthiouracil causes the levels of thyroxine to fall, and this in turn increases TSH secretion. High levels of TSH cause hypertrophy of the thyroid gland. Low levels of thyroxine lead to higher body weight.

D. False. Increased plasma level of CRH tends to increase the size of pituitary gland and ACTH secretion, and this in turn stimulates the adrenal gland to develop. Increased plasma level of TSH tends to increase thyroxine secretion. High level of thyroxine inhibits the secretion of TRH leading to decrease in the size of the pituitary gland, without affecting the adrenal gland.

Reference

Guyton, A.C and Hall, J.E, *Physiology Review*, 2006, pp.235.

Q.78

Some researchers studied the changes in the level of saliva cortisol and 2-AG (2-arachidonoylglycerol) concentration in blood in two groups of people with motion sickness and without motion sickness (no sickness) during parabolic flight maneuvers (PFs). During PFs, the saliva cortisol levels and 2-AG blood concentrations were measured from samples taken in-flight before start of the parabolic maneuvers (T0), after 10 parabolas (T1), 20 parabolas (T2), and 30 parabolas (T3), termination of PFs (T4) and 24 h later (T5). The results are shown in Figure Q.78.
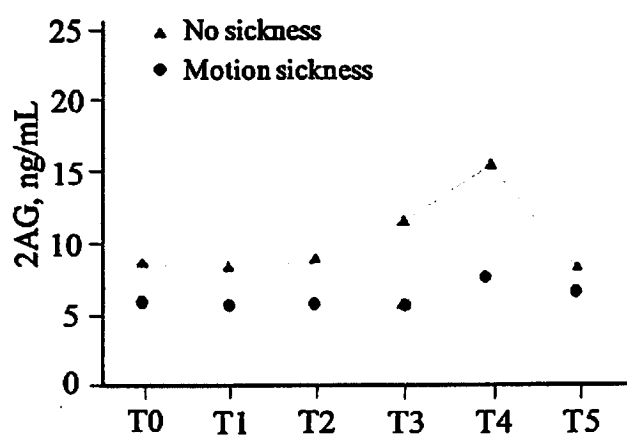

Figure Q.78A.

Figure Q.78B.

Indicate in the Answer sheet if each of the following statements is True or False.

A. A 2-AG inhibitor can be used to reduce motion sickness.

B. In motion sickness group, the blood glucose level at T4 was higher than at T1.

C. At T2, the blood ACTH (adrenocorticotropic hormone) level in the no sickness group was higher than that in the motion sickness group.

D. In motion sickness group, the blood CRH (corticotropin-releasing hormone) level at T5 was lower than that at T2.

Answer key

A. False   B. True   C. False   D. True

Explanation

A. **False.** The graph shows that the 2-AG level in the motion sickness group is lower than that in the non-motion group; so 2-AG inhibitor cannot be used to treat motion sickness.

B. **True.** In the motion sickness group, the cortisol level at T4 is higher than at T1. Cortisol stimulates gluconeogenesis in the liver leading increase the blood glucose level.

C. **False.** The graph shows that the saliva cortisol level in the no sickness group is lower than in motion sickness group, indicating that ACTH level in the no sickness group is lower than that in the motion sickness group. This means that at T2, the blood ACTH level in the no sickness group is lower than that in the motion sickness group.

D. **True.** The graph shows that in the motion sickness group, the saliva cortisol level increases from T1 to T4, and at T5 it decreased to normal level. This means that the blood CRH level at T5 is lower than at T2.

References

1. Alexander Choukèr, Ines Kaufmann, Simone Kreth, et al., 2010. *Motion Sickness, Stress and the Endocannabinoid System*. PLoS One 5(5): e10752.

2. Rorbert M. Stern, Kenneth L. Koch, and Paul L. R. Andrews, 2011. *Nausea: Mechanisms and management*. Oxford university press. P. 136.

Q.79

When a person born and brought-up in a region at sea level moves to a village at an altitude of 3000 metres above sea level by helicopter, some adaptations of his body occur to compensate for the decreased oxygen pressure at high altitude.

Indicate in the Answer sheet if each of the following statements is True or False.

A. The moment the person arrives at the high altitude, oxyhemoglobin dissociation curve shifts to the left (indicating greater affinity of hemoglobin for oxygen).

B. After several days living at the high altitude, the person's blood viscosity is decreased, enabling his blood to deliver more oxygen to his tissues.

C. After several weeks living at the high altitude, the person's lung cells of this person produce more nitric oxide (NO).

D. Many people who ascend rapidly to high altitude experience some degree of acute mountain sickness (e.g., headache, malaise, and nausea). Which may be treated with a drug that causes bicarbonate to be excreted in the urine.

Answer key

A. True   B. False   C. True   D. True

Explanation

A. **True.** At the time of arrival at the high altitude, the ventilation increases causing respiratory alkalosis. This in turn causes the oxyhemoglobin dissociation curve to shift to the left (indicating greater affinity of hemoglobin for oxygen), despite the antagonistic effect of increased 2,3-DPG concentration.

B. **False.** The number of red blood cells is increased because the oxygen pressure at high altitude is lower than that at sea level. This leads to an increase in blood viscosity, thereby increasing resistance to blood flow.

C. **True.** Chronically hypoxic people who live at high altitude have high levels of NO in their lungs/blood. NO produced in their lungs is vasodilator.

D. **True.** At 3000 metres above sea level, the decrease of arterial Po_2 stimulates the carotid bodies to produce an increase in ventilation. This produces a respiratory alkalosis that contributes to acute mountain sickness. A drug that causes

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bicarbonate to be excreted in the urine can be used to produce metabolic acidosis that partially compensates for the respiratory alkalosis caused by hyperventilaton.

Reference

Fox. S.I, *Human Physiology*, 2011. pp563-565.   ,

Q.80

Figure Q.80 demonstrates the relationship between oxygen concentration and oxygen partial pressure (Po_2) in blood of two species of vertebrates (species a and b). Each sample was subjected to two levels of carbon dioxide pressure (Pco_2): curve I represents the values measured at normal Pco_2 and curve II represents the values measured at elevated Pco_2. The blood having passed through the lungs of the two species normally has a Po_2 of 100 mm Hg and the deoxygenated blood leaving the tissues has a Po_2 of 40 mm Hg.
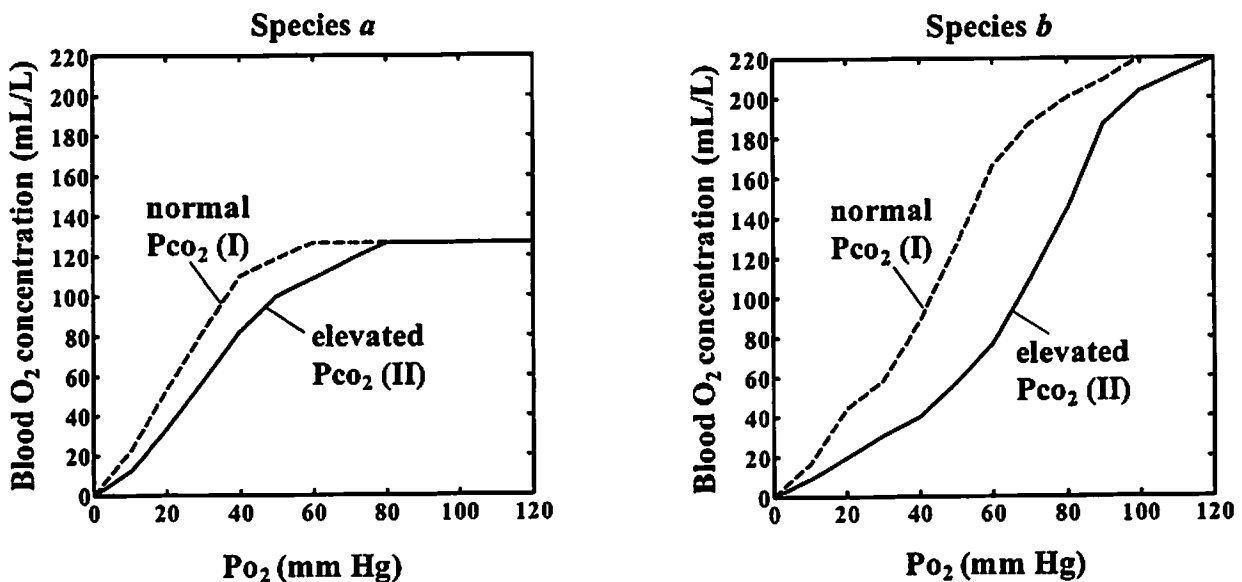

Figure Q.80.

Indicate in the Answer sheet if each of the following statements is True or False.

A. While comparing curve I of species a with curve I of species b, you can predict that the O_2 concentration of the blood in the lungs of species a will be higher than that of species b.

B. If you expose deoxygenated blood of the two species at the same level of Pco_2 to increasing Po_2, the first blood to become saturated with O_2 would be that of species a.

C. In species b, if curves I and II represent the Pco_2 of oxygenated and deoxygenated blood, respectively, there will be less than 160 mL of O_2 released from a litre of blood as it passes through the tissues.

D. In species a, an increase in Pco_2 in the blood reduces the affinity of hemoglobin for oxygen but has no effect on the maximum oxygen-carrying capacity in the blood.

Answer key

A. False   B. True   C. False   D. True

Explanation

A. False. The Po_2 is 100 mm Hg in the blood of the two species. At this value, the O_2 concentrations in the blood of species a and b are approximately 120 and 220 mL/L, respectively.

B. True. The Po_2 is 40 mm Hg in the deoxygenated blood. At this value, the O_2 concentration in the blood of species a is higher than that of species b. Furthermore, the level of saturated O_2 in the blood of species a is lower than that of species b. Therefore, if the deoxygenated blood of the two species at the same level of Pco_2 was exposed to increasing Po_2, the fist blood to become saturated with O_2 would be that of species a.

C. False. The Po_2 in arterial blood is 100 mm Hg. At this value in curve I, the blood O_2 concentration is 220 mL/L. The Po_2 in venous blood is 40 mm Hg. At this value in curve II, the blood O_2 concentration is 40 mm Hg. Thus, 220 - 40 = 180 mL of O_2 will be released from a litre of blood as it passes through the tissues in species b.

D. True. An increased in Pco_2 shifts the O_2 dissociation curve toward the right (the Bohr effect). This means that the O_2 affinity of hemoglobin in the blood is decreased. However, the maximum oxygen-carrying capacity in the blood of species a is the same in both normal and elevated Pco_2 conditions (e.g., $Po_2 \geq 80$ mm Hg, as shown in Figure Q.80).
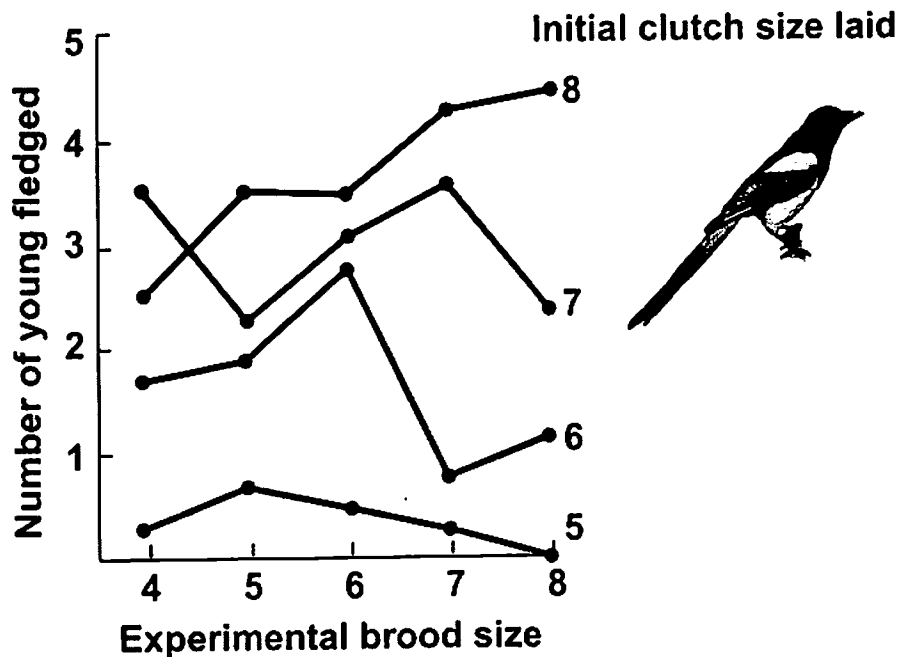
Reference

Reece, 2013. Campbell Biology 10th edition, pp 924.

ETHOLOGY
Q. 81

In an experimental study on magpies *Pica pica*, conducted in Sweden, Goran Högstedt manipulated the original clutch size (shown on the right of the graph) of experimental birds to generate a number of different clutch sizes for each category of bird, as shown in the X-axis. The number of young fledged successfully by the birds under these different conditions is indicated by the Y-axis. Food abundance and quality of territory are thought to be associated with clutch size. Predation is lower in large clutches.

Analyse the following statements, with reference to the data provided and indicate in the answer sheet if each of the statements is true or false.

A. The birds, in general, did better with experimentally manipulated, larger broods.

B. The reproductive rate of birds is closest to that which maximises individual breeding success.

C. Birds in high quality territory tend to have larger clutches.

D. Experimentally-manipulated clutches experience higher starvation.

Answer key:

A. False   B. True   C. True   D. True

Explanation

A. Statement (a) is false because it is clear from the data that different individuals do better with different clutch sizes.

B. Statement (b) is true because this naturally follows from the data

C. Statement (c) is an inference from the stem given.

D. Statement (d) is true as predation is lower in larger clutches (given in stem) so lack of food accounts for increased mortality of exp. enlarged clutches.

References:

Krebs, J R and Davies, N B. 1981. *An Introduction to Behavioural Ecology*, 3rd Edition. Blackwell Publishing, Oxford, pp. 20–21.

Högstedt, G. 1980. Evolution of clutch size in birds: Adaptive variation in relation to territory quality. *Science*, 210, 1148–1150.

Q. 82

The behaviour of two similar-sized species of fiddler crabs (*Uca latimanus* and *U. musica*) that intermingle in the same habitat was studied. Males build hoods over their burrows for mate attraction. Mate searching is a dangerous activity for fiddler crab females, so these females may be forced to make suboptimal choices for their own safety, especially in areas where their conspecifics are in lower densities. The figures Fig.Q.82 below show the approaches made by females of the two species to male crabs as well as to burrows (with and without hoods) of conspecific males.

(A) Mean (±SE) proportion of resident *Uca latimanus* and *U. musica* males approached by wandering
(a) *U. latimanus* females and (b) *U. musica* females.

(B) Mean (±SE) proportion of resident conspecific males with and without hoods approached by wandering (a) *U. latimanus* females and (b) *U. musica* females. $*p < 0.05$.

Indicate in the answer sheet if each of following statements is true or false.

A. Females of both species approached a greater proportion of the conspecific males than the heterospecific males they encountered.

B. Attraction of *U. musica* females to hoods is not as strong as that of *U. latimanus* females.

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C. A male fiddler crab's willingness to court all females, regardless of species, is made used of by females of both species for shelter-seeking and avoidance of predators.

D. An overlap in habitat use between these two similar-sized fiddler crabs has no impact on both signalers and receivers.

Answer key

A. True;   B. True;   C. False;   D. False
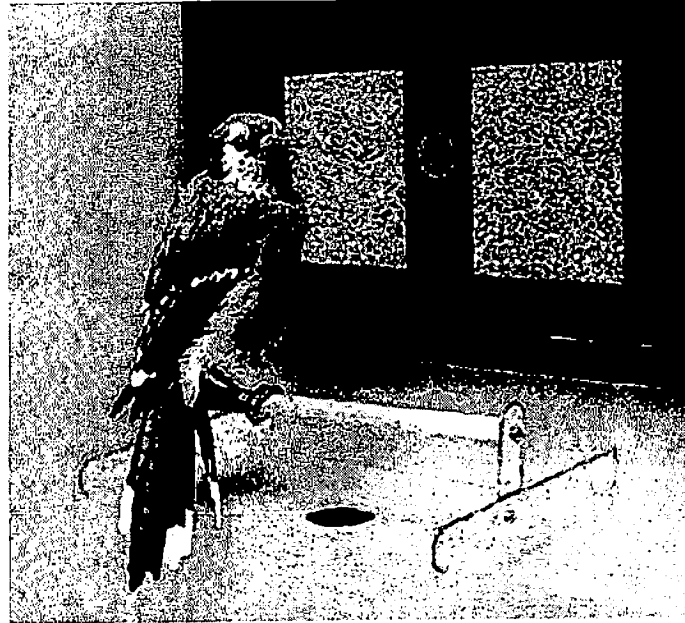
Explanation

A. True. *Uca latimanus* females were never seen to approach heterospecific males (Fig. A(a) but *U. musica* females approached mostly conspecifics (Fig. A(b)).

B. True. *U. latimanus* females approached significantly more burrows with hoods but *U. musica* approached almost equal numbers of both hooded and non-hooded burrows (marginal non-significance, p = 0.063).

C. False. Only *U. musica* takes advantage of the male fiddler crab's willingness to court all females, regardless of species, for shelter-seeking and avoidance of predators.

D. False. The presence of heterospecific males and hoods potentially causes more visual noise and may complicate receivers' ability to discriminate among conspecific signals. There are obvious impacts on both signalers (male *U. latimanus* are visited by *U. musica* females) and receivers (*U. muscia* females take advantage of the burrows of *U. latimanus* males for refuge).

Reference: Pope and Haney (2008) Interspecific signalling competition between two hood-building fiddler crab species, *Uca latimanus* and U. *musica*. Animal Behaviour 76:2037-2048.

Q. 83

Three morphs of a polymorphic species of *Catacola* moths differ only in the patterns on the forewings.

Six experienced blue jays (*Cyanocitta cristata*) searched for prey on a computer screen in a series of trials.

Each trial involved a screen showing the presence or absence of a moth from three distinct morphs of a moth population. If the bird found the moth, it was rewarded with food. Each bird had 36 prey and 84 no-prey trials and lasted for 50 days. Three replicates were carried out, with the second having a larger population of morph 2 and third replicate starting off with a larger relative abundance of morph 3.

A. Population numbers of the three morphs.
B. Prey detectability

a. Population numbers of the three morphs.
b. Prey detectability

(Bond & Kamil, 1998 - doi:10.1038/26961)

Indicate in the answer sheet if each of the following statements is true or false.

A. Morph 1 was the most cryptic morph.

B. Relative numbers that escaped detection determine the abundance of each prey type.

C. Preferential feeding behavior of the blue-jays of the most prevalent morph maximizes their foraging success.

D. Polymorphisms are maintained in the population through frequency-dependent selection by predators.

Answer key

A. True   B. True   C. False   D. True

Explanation

A. Escaped detection so the most cryptically coloured.

B. If they are not detected they stay in the population.

C. Blue-jays appear to prefer more obviously coloured morphs but there is nothing about foraging success.

D. This is apostatic selection where predators determine frequency-dependent selection.

GENETICS AND EVOLUTION

Q. 84

An organism has four genes, A, B, C and D with two alleles each. An individual heterozygous for these genes was bred with one that is homozygous recessive. The cross produced 3288 offspring with phenotypes shown in the table below:

Phenotypes	Number of Individuals
ABCD	675
ABCd	83
ABcD	1
ABcd	74
AbCD	73
AbCd	1
AbcD	84
Abcd	670
aBCD	655
aBCd	86
aBcD	1
aBcd	73
abCD	71
abCd	1
abcD	87
abcd	653

Indicate in the answer sheet if each of the following statements is true or false

A. The four loci are genetically linked.

B. The distance between gene B and gene D is 9 cM.

C. The distance between gene D and gene C is 10.5 cM.

D. Interference happened with a value less than 0.25

Answer key

A. False   B. False   C. True   D. False

Explanation

A. False. Genes BCD are linked but gene A is unlinked with them.

B. False. The distance B-D: 100% (83+74+73+84+86+73+71+87)/3288 = 19 m.u.

C. True. The distance C-D: 100% (87+1+1+86+84+1+1+83)/3288 = 10.5 m.u.

D. False. Interference (I) = 1- [(observed DCO)/(expected DCO)] = 1-{[(1+1+1+1)/3288]/[(0.0897) (0.105)]} = 0.89.

References: McCauley, S. J., Rowe, L., & Fortin, M.-J. (2011). The Deadly Effects of "Nonlethal" Predators. *Ecology*, *92* (11), 2043-2048.

Q. 85

A plant is normally red-flowered. Plant breeders obtained three genetically different pure mutated lines of white-flowered plants (designated as a, b and c). They performed crosses and observed progeny phenotypes as follows:

Cross	Parents	Progeny
1	Line a × line b	F_1 all white
2	Line a × line c	F_1 all red
3	Line b × line c	F_1 all white
4	Red F_1 from cross 2 × line a	¼ red : ¾ white
5	Red F_1 from cross 2 × line b	1/8 red : 7/8 white
6	Red F_1 from cross 2 × line c	1/2 red : ½ white

Indicate in the answer sheet if each of the following statements is true or false.

A. Line (a) is homozygous for one mutated gene.

B. Line (b) shares two mutated homozygous genes with line c.

C. Line (c) shares one mutated homozygous gene with line a.

D. Line (b) has three mutated homozygous genes.

Answer key

A. False   B. False   C. False   D. True

Explanation

Note: To solve this problem, based on complementation tests students should determine how many genes involved.

Cross 2 tells that there are at least two genes involved because white crossed white yields red flowers. However, cross 5 indicates that there are at least three genes involved because a 1: 7 ratio is observed. Therefore, F_1 red must be heterozygous for 3 genes and line b must be homozygous for three defective genes located in different chromosomes (**D is true**).

Compare crosses 2 and 3. Lines (a) and (c) can compensate for each other's defects but lines (b) and (c) cannot. This suggests that line (b) shares a defective homozygous gene with line (c) (**B is false**) and that line (a) is normal for the defective gene seen in line (c) but not in line (b). Line a must be homozygous for 2

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defective genes (**A is false**). Line (a) and line (c) compensate for each other's defect so these lines cannot share any defective genes (**C is false**). Line (c) is normal for the two defective genes in line (a). Genotypes of the three lines should be: Line a: aaBBcc; Line b: aabbcc; line c: AAbbCC.

References: Adapted from:

Anthony J.F. Griffiths; Jefferey H. Millerr; David T. Suzuki; Richard C. Lewontin and William M. Gelbart. An Introduction to genetic analysis, fifth edition 1993.

Q. 86

Indicate in the answer sheet if each of the following statements is true or false.

A. A completely recessive allele is lethal in homozygous condition and if its dominant allele mutates to recessive allele at a rate of 10^{-6}, then frequency of the lethal allele when the population reaches mutation-selection equilibrium is 0.001.

B. If frequency of a completely recessive lethal allele is 0.2 and it remains unchanged from generation to generation due to the superior fitness of heterozygotes, then the intensity of selection against the dominant homozygotes should be 0.025.

C. Selection for recessive alleles is less effective than selection against recessive alleles.

D. In a large, randomly mating population, the frequency of an autosomal recessive lethal allele is 0.2. Frequency of this allele in the next generation will be 0.07 if the lethality takes place before reproduction.

Answer key

A. True	B. False	C. False	D. False

Explanation

A. True. At mutation-selection equilibrium frequency of recessive allele can determine by formula $q = u/s$, where u = mutation rate; s = selection coefficient.

Because s =1 so q = 0.00.1

B. False. If selection coefficient of dominant homozygotes is t and selection coefficient of recessive homozygotes is s then frequency of recessive allele at equilibrium, $q = s/(s+t)$. In this case, s = 1, q = 0.2 then t = 0.25.

C. False. Selection for recessive alleles (selection against dominant phenotypes) is more effective than selection against recessive alleles.

D. False. When selection against recessive homozygotes, frequency of recessive allele in next generation change a value $\Delta = (- spq^2)/ (1-sq^2)$. In our case, s = 1; q=0.2 then $\Delta = 0.03333$. Frequency of recessive allele in next generation is = 0.2 - 0.333 = 0.1667.

Reference: Genetics A conceptual approach 4th edition, Pierce 2012.

Q. 87

Indicate in the answer sheet if each of the following statements about cancers is true or false.

A. A particular type of colon cancer can be caused by recessive alleles even though its inheritable pattern appears similar to that of a dominant trait.

B. In one patient, normal cells have only one mutated *p53* allele but cancer cells have two identical mutated *p53* alleles. Then it can be concluded that the second mutated *p53* allele is formed by gene conversion.

C. Some cancers have been effectively treated with drugs that cause demethylation. Then it can be concluded that genes causing those cancers are more likely to be oncogenes.

D. Chromosome inversions can produce novel oncogenes.

Answer key

A. True   B. True   C. False   D. True

Explanation

A. True. At the level of the organism, this type of colon cancer be inherited in dominant fashion but the mutant tumor-suppressor allele is recessive to the wild-type allele. This is because all somatic cells of the patient carry one recessive defective copy of the gene, the strong likelihood that in at least one of the hundreds of thousands of colon cells heterozygous for the tumor-suppressor gene, a subsequent genetic event will disable the single remaining functional tumor-suppressor allele, resulting in a mutant cell with no functional tumor-suppressor gene. This one cell then multiplies out of control, eventually generating a clone of cancerous cells.

B. True. This can be explained if a wild-type *p53* allele is degraded and then resynthesized with template strand by DNA carrying the mutated *p53* allele.

C. False. Dominant mutations that change proto-oncogene to oncogene may overactive expression of protein that promotes proliferation. Demethylation cause gene became active so cancer gene in this case is tumor-suppressor gene.
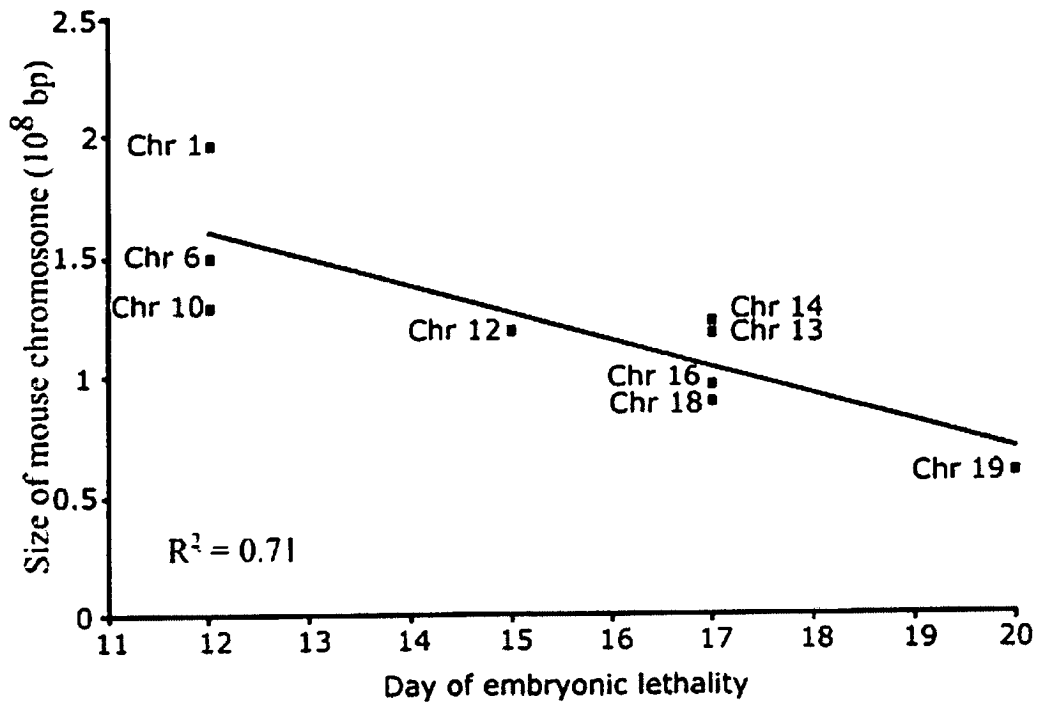
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D. True. Inversion can produce a novel oncogene by combining different parts of genes.

Reference: Principle of genetics 6th-Sunstad and Simmons 2012.

Q. 88

Mice that were trisomic for each of the 20 different chromosomes were monitored during embryonic development. Their survival time was plotted against the size of trisomic chromosome in Fig.Q.88.

Fig.Q.88

Based on this information, indicate in the answer sheet if each of the following statements is true or false.

A. Chromosome 19 is likely the smallest chromosomes with respect to the numbers of transcripts that they encode.

B. The total amount of additional genetic material determines the severity of the defects associated with the chromosome imbalance.

C. Gene density on chromosome 1 is probably lower than that on chromosome 10.

D. Genes on chromosomes 12 are probably more important for embryo development than those on chromosome 13.

Answer key

A. True   B. False   C. True   D. True

Explanation

A. True. Trisomies of other chromosomes died earlier than trisomy 19.

B. False. Chromosomes 1, 6, 10 are different in size but their trisomies died at the same day (day 12th) indicate that total amount of additional genetic material do

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not determines the severity of the defects associated with the chromosome imbalance. Only number of genes on chromosomes determines the lethality.

C. True. Chromosome 1 is the largest but trisomy 1 died at the same time as trisomy 10.

D. True. Chromosomes 12 and 13 are of similar size. However trisomy of chromosome 12 dies earlier than that of chromosome 13. Therefore genes on chromosome 12 are probably more important for embryo development than those on chromosome 13.

Reference: Eduardo M. Torres, Bret R. Williams and Angelika Amon. Aneuploidy: Cells losing their balance.

Q. 89

mRNA in cytoplasm of eukaryote cells sometimes forms closed loop by circularization. Indicate in the answer sheet if each of the following statements concerning closed loop is true or false.
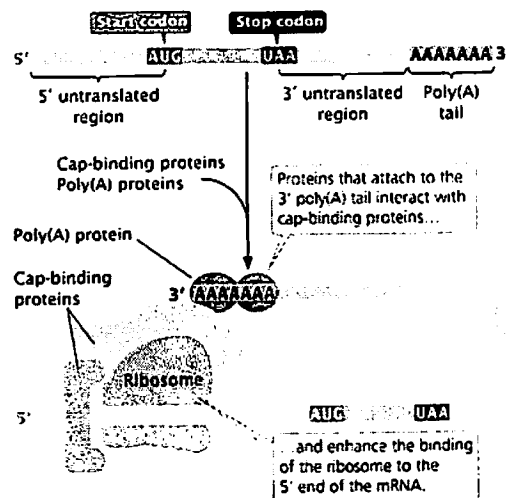
    A. Circularization is due to a phosphodiester bond between the 5'end and the 3'end of mRNA.

    B. Circularization increases stability of mRNAs.

    C. Circularization enhances translocation speed of the ribosomes.

    D. Controlling circularization is a mechanism of post-transcriptional regulation.

Answer key

    A. False   B. True   C. False   D. True

Explanation

    A. False. Circularization is mediated by protein-protein interaction between proteins binding to the 5'-cap and polyA-binding proteins.

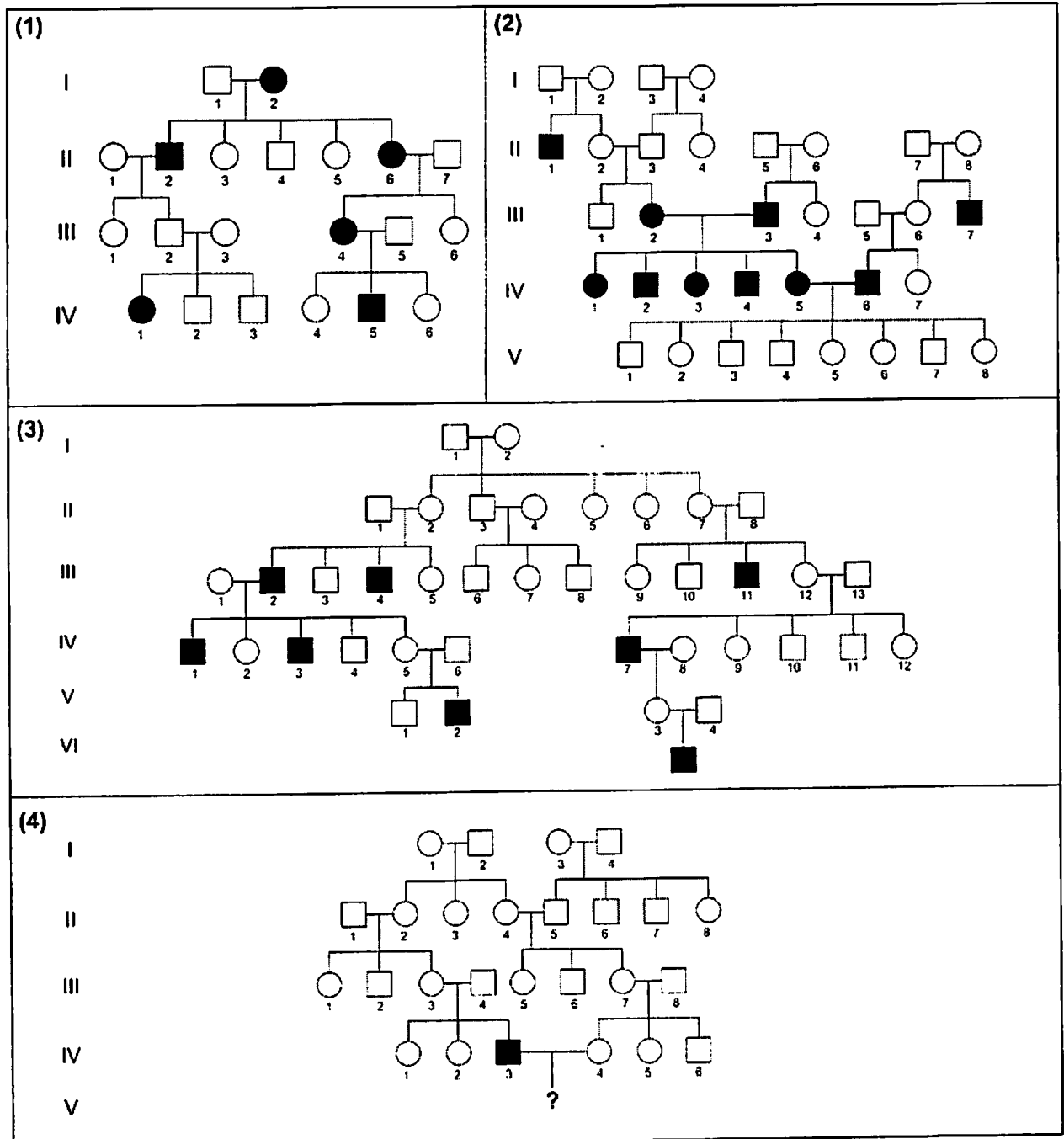**15.18** The poly(A) tail of eukaryotic mRNA plays a role in the initiation of translation.

    B. True.

    C. False. Interaction between 5'-end and 3'-end binding proteins enhances formation of translation initiation complex, thus increasing translation rate. However, it does not affect the speed of ribosome moving on mRNA.

    D. True.

Reference: Genetics A conceptual approach 4th. E. Pierce 2012

Q. 90

Pedigrees 1-4 show the inheritance of four different rare disorders. It is known that the disease in pedigree 4 is X-linked recessive.

Studying the pedigrees and indicate in the answer sheet if each of the following statements is true or false.

A. The disorder in pedigree 1 is most likely caused by a recessive allele.

B. Person III$_2$ and III$_7$ in pedigree 2 have the same genotype.

C. Pedigree 3 shows the inheritance of a rare disorder can be caused by a recessive allele on X-chromosome.

D. If the affected man and his unaffected wife in pedigree 4 have a son then the probability of this son be affected is 0.125.

Answer key

A. False   B. False   C. True   D. False

Explanation

A. False. If the disorder is autosomal recessive then three people I_1, II_7 and III_5 must be carriers. Heterozygotes should not be so common because it is a rare disorder. Therefore, it is not likely the disorder is caused by a recessive allele. The disorder is actually caused by a dominant allele.

B. False. The disorder is caused by recessive alleles. Persons IV_5 and IV_6 were recessive homozygotes but none of their children was affected. It indicates that there is complementation between recessive alleles of different genes. Therefore, the genotype of person III_2 must be different from the genotype of person III_7.

C. True.

D. False. Pedigree 5 shows that the disorder was caused by a recessive allele on X-chromosome. In the pedigree 4, the male IV_3 is affected therefore females III_3, II_2 and I_1 must be carriers. Chance of the recessive alleles be passed on from female I_1 to female II_4 is ½, from female II_4 to female III_7 is ½, from female III_7 to female IV_4 is ½ and from female IV_4 to her son is ½. Therefore we can calculate the probability for the son to be affected is ½ * ½ * ½ * ½ = 0.0625.

Q. 91

Polymorphic DNA sequences are widely used for molecular identification. Short tandem repeat (STR) is composed of multiple repeats of 2-8 nucleotides flanking by two conserved sequences. Each STR locus normally has more than two alleles. Single nucleotide polymorphism (SNP) is a variation at a single position in a DNA sequence among individuals. Each SNP usually has only two alleles. Seven individuals were genotyping for two autosomal and two mitochondrial (mtDNA) SNPs, two autosomal and two Y-linked (NRY) STRs (Table Q.91).

Table Q.91

Individuals	Autosomes				NRY		mtDNA	
	SNP1	SNP2	STR1	STR2	STR1	STR2	SNP1	SNP2
Ind_1	A/A	A/A	13/15	18/20	13	12	C	A
Ind_2	A/C	A/G	12/14	18/21	13	15	T	A
Ind_3	C/C	A/G	14/15	18/21	13	15	C	G
Ind_4	A/C	G/G	13/15	19/19	11	14	T	G
Ind_5	C/C	A/G	14/15	18/19	-	-	C	G
Ind_6	A/C	G/G	14/14	18/19	-	-	T	G
Ind_7	C/C	G/G	14/16	19/21	-	-	C	A

Indicate in the answer sheet if each of the following statements is true or false.

A. If the same number of SNPs or STRs is used, SNPs are better marker to distinguish individuals than STRs.

B. Ind_6 is more likely a child of Ind_2 and Ind_5 than Ind_3 is.

C. Ind_4 is possibly a brother of Ind_6.

D. It is possible that Ind_7 is a granddaughter of Ind_1 and Ind_6.

Answer key

A. False   B. False   C. True   D. True

Explanation

A. False. SNPs are less polymorphic than STRs therefore they are not as good as STRs to differentiate individuals of the same species.

B. False. All autosomal markers of Ind_3 and Ind_6 can be inherited from Ind_2 and Ind_5. Ind_3 share Y chromosome markers with Ind_2 and mitochondrial markers with Ind_5 while Ind_6 does not share mitochondrial marker with Ind_5.

C. True. Ind_4 and Ind_6 share autosomal SNP markers and mitochondrial markers. Their autosomal STR1 and STR2 can be inherited from parents of 13/14 and 14/15, and 18/19 and 19/19, respectively.

D. True. It is not necessary that Ind_7 inherits mitochondrial marker from Ind_6. It is possible that Ind_7 inherits all autosomal markers from her father who is a son of Ind_1 and Ind_6.

Reference: Jobling, M., Hollox, E., Hurles, M., Kivisild, T., Tyler-Smith, C., 2014. Human evolutionary genetics, 2nd edition. ed. Garland Science, Taylor & Francis Group, LLC, New York.

Q. 92

A wildtype female Drosophila was mated with a wildtype male that had been X-ray irradiated. One of F_1 females was mated with a male that had recessive phenotype (caused by recessive allele a). Progenies of the second mating were unusual in two aspects. Firstly, there were twice as many females as males. Secondly, while all males were wild type, ½ females were wild type, and the other ½ exhibited the recessive phenotype a.

Indicate in the answer sheet if each of the following statements is true or false.

A. X-rays converted a dominant allele (A) on chromosome X coding for the wild type to a recessive allele (a).

B. X-rays produced a chromosomal translocation.

C. A loop could be seen on chromosome during prophase of meiosis 1.

D. If a female from the second mating exhibiting recessive phenotype (a) was crossed to a wild type male then her progenies compose of females and males at the ratio of 2 females to 1 male.
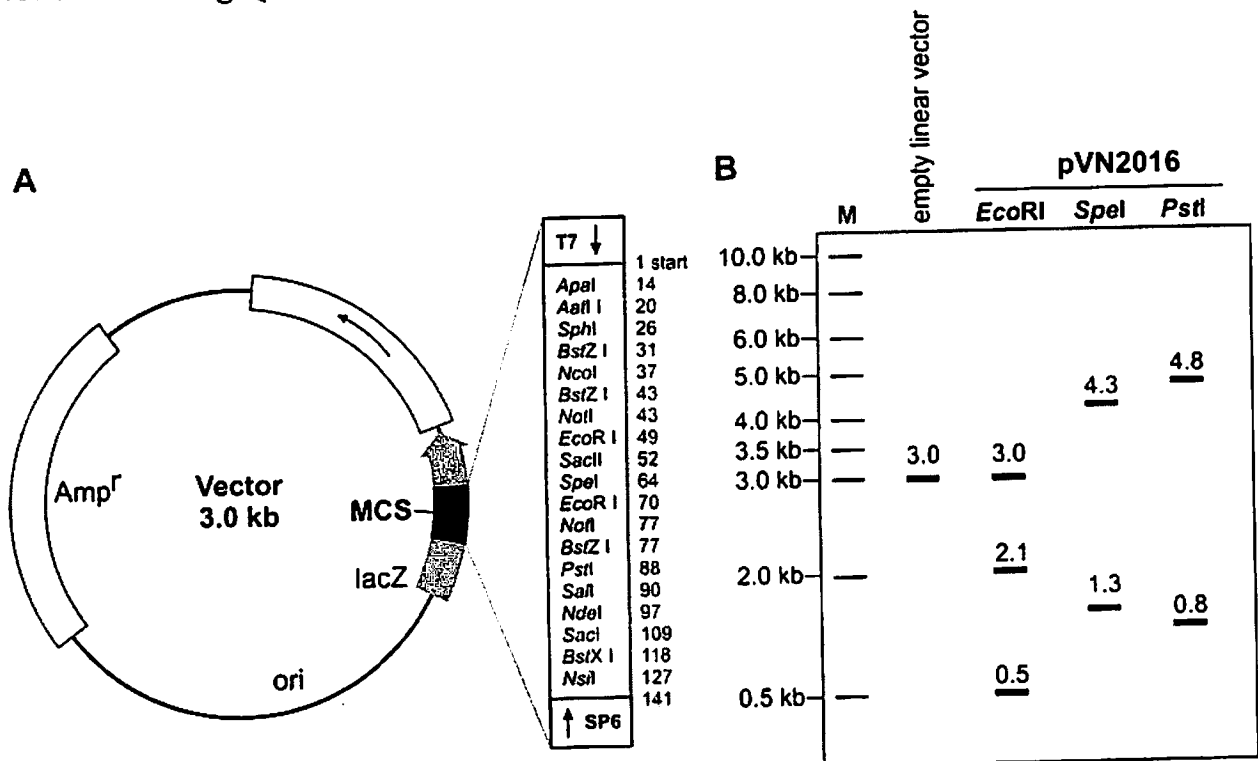
Answer key

A. False   B. False   C. True   D. True

Explanation

A. False. Recessive alleles are not lethal so they cannot cause lethality in males.

B. False. Translocation heterozygote is not lethal.

C. True. One X-chromosome with a deletion synapsed with normal X-chromosome resulted in a loop in salivary polytene chromosomes.

D. True. Female with recessive phenotype from the second cross has genotype of X^aX^- where X^- denote a deletion containing gene A on X-chromosome). Wildtype male is X^AY. Combination of their gametes would generate four different genotypes ¼ X^AX^a : ¼ X^AX^- : ¼ X^aY : ¼ X^-Y. All X^-Y males were dead therefore female to male ratio is 2 to 1.

Q. 93.

Trung cloned a coding sequence (CDS) of a gene into a vector, named pVN2016. This fragment was inserted at the *Sac*II recognition site which is located in the multi cloning site (MCS) region within the *lacZ* gene of the vector (Fig.Q.93A). The inserted fragment has a *Pst*I restriction site located 0.8 kb upstream of its stop codon. To identify the size and direction of the inserted CDS (3'-end to 5'-end of template strand), Trung digested this plasmid by different restriction enzymes, and the results of the digestions are shown in Fig.Q.93B.

Fig.Q.93. (A) A schematic map of the vector, numbers indicate positions of restriction enzyme recognition sites located in the vector (B) A schematic electrophoresis of digestive products using different restriction enzymes, M: 1 kb DNA ladder.

Based on above data, indicate in the answer sheet if each of the following statements is true or false.

A. The CDS is 2.6 kb in length and has an *Eco*RI recognition site at about 0.5 kb from one of its ends.

B. *Spe*I can be used to determine the orientation of the CDS.

C. The CDS is oriented in reverse direction compared to *lacZ* gene.

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D. If plasmid pVN2016 is digested by both enzymes *Spe*I and *Eco*RI in Tango buffer (*Eco*RI and *Spe*I cut at 100% and 20% efficiency, respectively), five fragments of 0.5, 0.8, 1.3, 2.1 and 3.0 kb could be detected by gel electrophoresis assuming that fragments smaller than 50 bp are not visible.

Answer key:

A. True   B. False   C. True   D. True

Explanation

A. **True.** Length of the plasmid pVN2016 equals to sum of length of digested fragments, which is 5.6 kb. The empty vector's length is 3.0 kb therefore the inserted CDS is 2.6 kb long.

There are two *Eco*RI recognition sites next to cloning *Sac*II site. Because *Eco*RI digestion generated three fragments, there must be another *Eco*RI recognition site in the inserted CDS. The 3.0 kb digested fragment corresponds to the empty vector. Two fragments of 2.1 kb and 0.5 kb are resulted from digested CDS. Therefore, the *Eco*RI recognition site in the CDS must be located about 0.5 kb from either of its ends.

B. **False.** There is only one *Spe*I recognition site in the MCS region. Because *Spe*I digestion of pVN2016 results in two fragments, there must be another *Spe*I recognition site in the inserted CDS. The 1.3 kb fragment is mainly composed of DNA from the inserted CDS while the 4.3 kb fragment should be composed of 3.0 kb empty vector and 1.3 kb fragment of the inserted CDS. It means that *Spe*I cuts the CDS into two fragments which almost identical in length. With either orientation of the inserted CDS, *Spe*I digestion of pVN2016 always results in two fragments of 1.3 kb and 4.3 kb. Therefore cannot be used to determine the orientation of the CDS.

C. **True.** There is a *Pst*I recognition site 36 bp upstream of the cloning *Sac*II site (regarding to *lacZ* gene direction) in the MCS region. There is another *Pst*I recognition site in the CDS 0.8 kb upstream of its stop codon (regarding to the

CDS direction). If the inserted CDS orients in the same direction as the *lacZ* gene, *Pst*I digestion should result in two fragments of 1.8 kb and 3.8 kb. If the CDS orients in reverse direction of the *lacZ* gene, *Pst*I digestion should result in two fragments of 0.8 kb and 4.8 kb. Result in Fig.Q.93 indicates that the inserted CDS orients in reverse direction of *lacZ* gene

D. **True**. In Tango buffer, all DNA containing *Eco*RI recognition site is cut but only about 20% DNA containing *Spe*I recognition site is cut. Because DNA fragment which is smaller than 50 bp is not visible, five fragments of 0.5, 0.8, 1.3, 2.1 and 3.0 kb could be observed on electrophoresis gel.

References:

Promega Resources (https://worldwide.promega.com/resources/product-guides-and-selectors/protocols-and-applications-guide/cloning/)

Scientists constructed models for four threatened tree species in sub-tropical forests in Vietnam, and used these models to estimate tree ages (figure Q.94). Tree age is measured by ring count and trunk diameter at breast height (DBH). Rates of growth were categorised using changes in DBH from 10 to 1000, with 10 at the finest-grain measure of change.
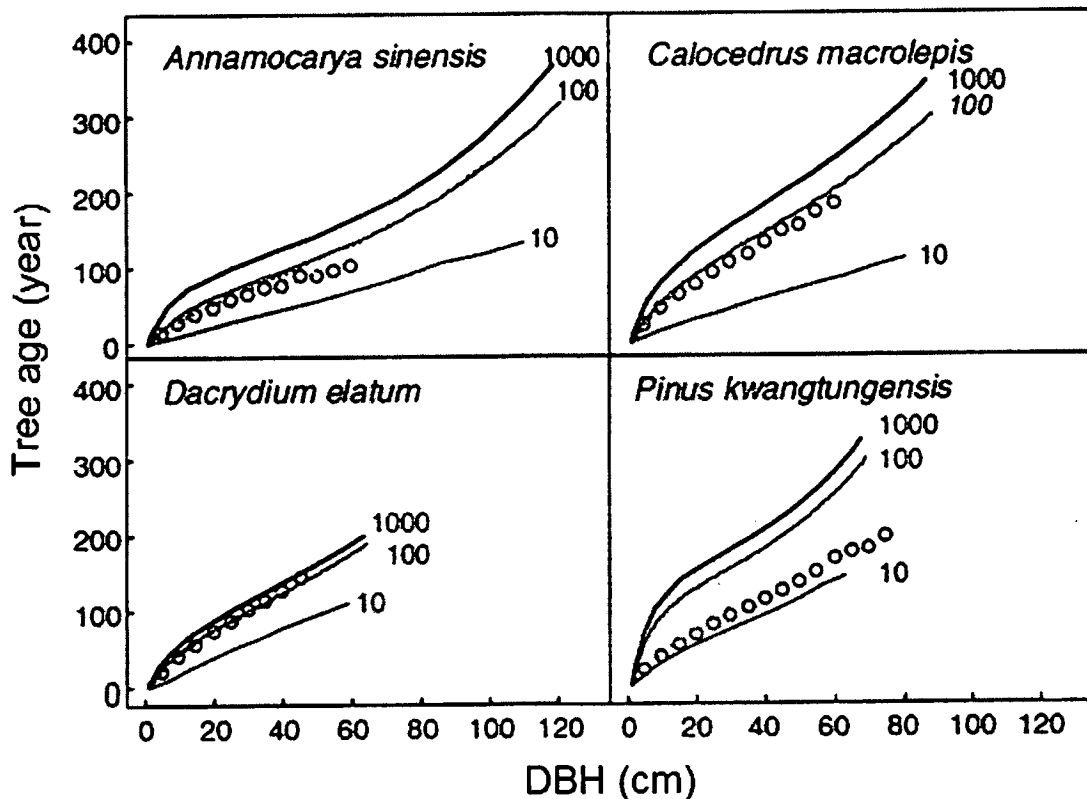

Figure Q. 94

Estimated (lines) and observed (circles) ages for DBH categories of four tree species.

Indicate in the **Answer sheet** if each of the following statements is True or False.

A. Using the smallest category gives the most accurate information of tree age of *P. kwangtungenesis*.

B. Age estimates increase particularly strongly from 100 to 10-category model in *D. elatum*.

C. Model with just 10 DBH categories underestimate the observed ages for three species.

D. For *D.elatum*, measuring DBH using either 100 or 1000 will give an accurate estimate of tree age, whereas to estimate the age of *C. macrolepis*, only 100 gives a reliable estimate.

Answer Key

A. True   **B. False**   C. True   **D.** True

Explanation

A. True. For *P. kwangtungenesis*, the model with 10 categories gives accurate estimate which is close to ring count.

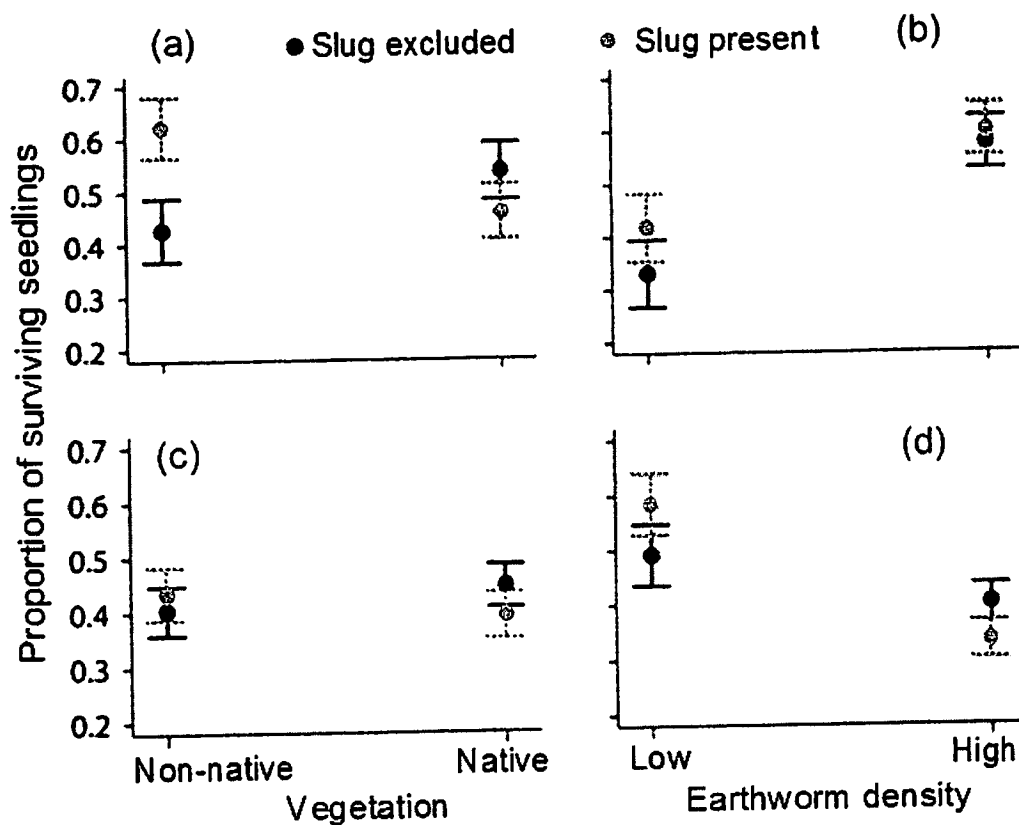**B. False.** Age estimate is higher using the model with 100 DBH categories.

C. True. When comparing age estimates to observed ages from ring counts, it becomes clear that model with just 10 DBH categories strongly underestimate the observed ages for three (*A. sinensis, C. macrolepis, D. elatum*) of the four species.

D. True. As shown in the figure, tree ages estimated using models with 100 and 1000 categories are close to observed ages in *D.elatum*, while for *C. macrolepis*, only the model with 100 categories give accurate estimate.

Reference: Zuidema, Pieter A. et al., *Journal of Ecology* 98.2 (2010): 345-355.

Q. 95

To understand the effects of several factors on plants (*Agrimonia rostellata* and *Trillium erectum*) in forest ecosystems, students transplanted seedlings into experimental sites and observed the proportion of surviving seedlings growing with native or non-native vegetation, with or without slugs, and with low or high earthworm density. The results are shown in the figure below.

Figure Q.95. Proportion of surviving seedlings. *Agrimonia rostellata* (a, b) and *Trillium erectum* (c, d)

Indicate in the **Answer sheet** if each of the following statements is True or False.

A. There are either positive effects or interactive effects with slug exclusion.

B. Slug effects are dependent on other stressors, especially on interactions with non-native plants and earthworms.

C. Earthworms have positive effects on *Agrimonia rostellata* and *Trillium erectum*.

D. Non-native plants and slugs synergistically decrease seedling survival through increased competition and consumption.

Answer key

A. True   B. True   C. False   D. False

Explanation

A. True. There are either positive effects (*Agrimonia*) or interactive effects with slug exclusion (survival of *Agrimonia* and *Trillium*). Figures 95 (a) & (c).

B. True. Slug effects on seedlings survival are in interactions with non-native plants and earthworms.

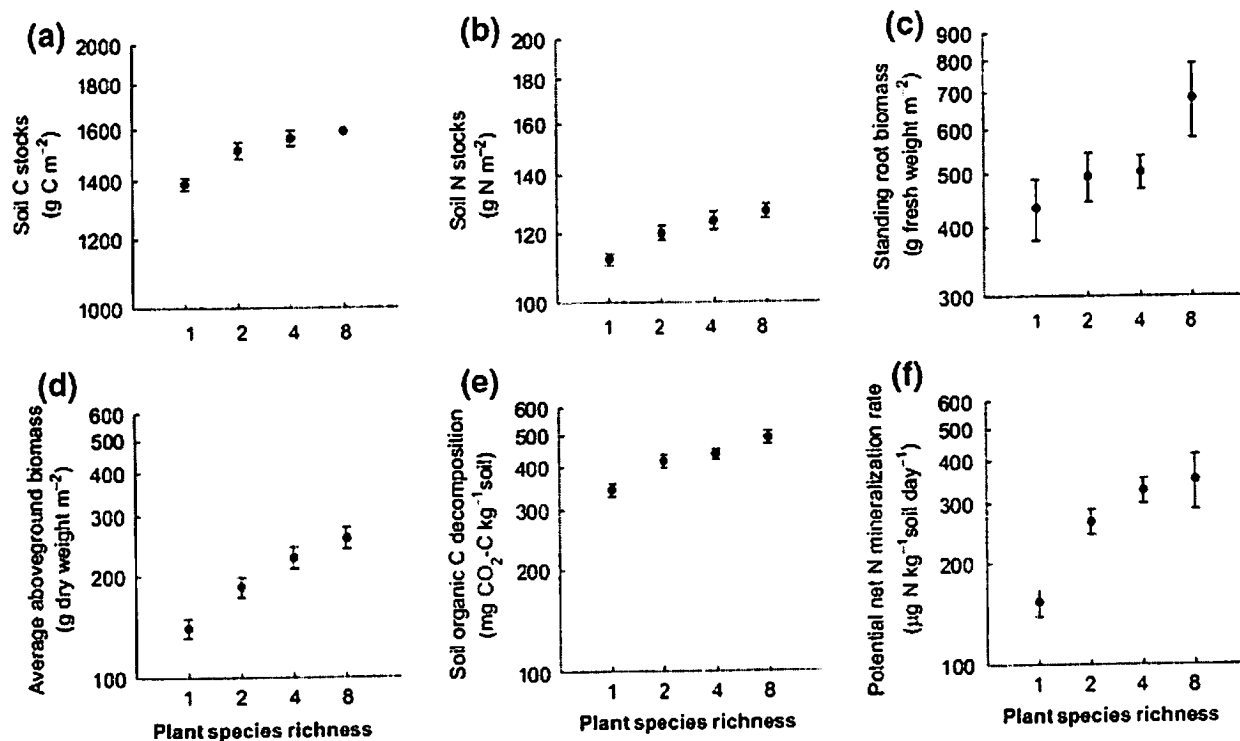**C. False.** Earthworms have positive effects on *A. rostellata*, but negative effects on *T. erectum*.

D. False. Non-native plants and slugs synergistically increase seedling survival through increased competition and consumption.

Q. 96

Students cultured plants, including four grass species (*A. capillaris, A. odoratum, F. rubra,* and *H. lanatus*) and four herbs (*C. jacea, L. vulgare, P. lanceolata,* and *R. acetosa*) without legumes, in different blocks with treatments of monoculture and mixtures of two, four or all eight species. They then measured different parameters as functions of plant species richness, as shown in the figure below. Values are shown in $\log_2$ scale.

Figure Q.96

Indicate in the **Answer sheet** if each of the following statements is True or False.

A. In monoculture, both above ground and standing root biomass are lower than those in the mixtures of two, four or eight species.

B. Plant species richness promotes soil C stocks mainly through enhanced plant productivity, despite accelerated soil organic C decomposition.

C. Greater soil N stocks at higher species richness is mainly attributed to increased N retention, rather than N input, with enhanced plant productivity.

D. More diverse ecosystems can increase the potential for C sequestration in terrestrial ecosystems.

Answer key

A. True   B. True   C. True   D. True

Explanation

A. True. Aboveground and standing root biomasses are higher at higher species richness, Figure 96 (c) and (d).

B. True. See figure 96. a and c-e

C. True. Plant productivity is increased in higher species richness treatments. Greater soil N stocks at higher species richness is due to N retention rather than N input since legume plant is absent in all treatments.

D. True. The results from the study indicates that more diverse ecosystems can increase the potential for C sequestration in terrestrial ecosystems.

Reference: W.-F. Cong *et al.* (2014), Journal of Ecology 2014, 102, 1163–1170

Q. 97

A rocky shore contains many shallow rock pools dominated by macroalgae and grazing gastropods, comprising primarily *Patella ulyssiponensis* (P), *Littorina littorea* (L) and *Gibbula umbilicalis* (G). The experiment is designed to test the interaction between grazer species and the additive interaction with nutrient enrichment. Pools contained either none, one, two or all three of grazer species at realistic densities (*Patella*, *Littorina* and *Gibbula*). Another complete set of all the manipulation grazer treatments was also established concurrently where nutrient concentrations were enhanced to compare the simultaneous effects of grazer treatments at ambient and enriched nutrient conditions. Gross ecosystem productivity (GEP), number of algal taxa, and the biomass (dry weight) of all algal species were measured.

Figure Q.97

Indicate in the **Answer sheet** if each of the following statements is True or False.

A. Gross ecosystem productivity is enhanced by nutrient enrichment and is greater in pools where *Littorina* is present.

B. The effects of grazer species loss on accumulated algal biomass are regulated by nutrient conditions, grazer identity and grazer diversity.

C. The effects of loss of grazer species on ecosystem functioning depend upon both the diversity and identity of the species present.

D. The presence of all grazers results in lower algal diversity and biomass in both nutrient conditions.

Answer key

A. True   **B.** True   **C.** True   **D.** False

Explanation

A. True. Gross ecosystem productivity is higher in treatments with nutrient enrichment and it is much higher in pools where *Littorina* is present compared to those when *Littorina* is absent (Fig. 97 a).

B. True. In non-enriched nutrient condition, algal biomass in the pools loss P, L or all species increases, and decreases in pools loss G; however, in pools with nutrient enrichment, algal biomass generally are not affected by species loss (L, G or all P-L-G, exept P) (Fig. 97 b).

C. True. Based on data given in the figure, we can see that the effects of loss of grazer species on ecosystem functioning depend upon both the diversity and identity of the grazer species present.

D. False. In enriched nutrient conditions, the presence of all grazers does not affect algal biomass (Fig. 97 b).

Reference: O'Connor et al., 2015, *Journal of Ecology*, 103: 862–870.

Q. 98

Gall aphids (*Pemphigus betae*) live in poplar plants. Adult females produce galls on poplar leaves. Some fraction of these galls will emerge and survive to adulthood. Female aphids complete their life cycle after laying eggs in the leaves. All the progeny of a single female aphid are contained in one gall. A student recorded observation on several aphid populations, shown in the table below. All environmental parameters are assumed constant.

Population	Number of aborted galls	Number of successful galls	Female/Male ratio in adult stage
1	35	70	1/1
2	25	75	1/2
3	21	63	Not given
4	16	32	1/1

An equation representing number of female aphids in t^{th} generation is established as below:

$$N_t = [f \times r \times (1-m)]^t \times N_0$$

Whereas:

N_t – number of adult female aphids in the tth generation

N_0 – number of adult female aphids in the initial generation

m – fraction mortality of the young aphids

f – number of progeny per female aphid

r – ratio of female aphids to total adult aphids.

Theoretically f, m and r are constant.

Indicate in the **Answer sheet** if each of the following statements is True or False.

A. Population 1 has a constant number of adult females across generations when each female produces 4 progeny.

B. When every female in population 2 produces 3 progeny, this population will have a constant number of adult females across generations.

C. When population 3 has a constant number of adult females across generations and each female aphid produces 4 progeny, the female/male ratio of the population in adult stage is 1/2.

D. Given that each female in population 4 produces 6 aphids and taking the offspring of population 4 to be in the first generation, the number of adult females in the third generation will be 384.

Answer key

A. False   **B.** False   **C.** True   **D.** True

Explanation

Based on the given data and the equation, several values are computed as shown in the table below:

Population	Number of aborted galls	Number of successful galls	Female/ Male ratio	f	m	1-m	r	Number of adult aphids
1	35	70	1/1		1/3	2/3	0.5	
2	25	75	1/2	3	0.25	0.75	1/3	
3	21	63	Not given	4	0.25	0.75		
4	16	32	1/1	6	1/3	2/3	0.5	192

A. False.

Population 1 has a constant number of adult females across generations when $N_t/N_0 = 1$; hence,

$f \times r \times (1\text{-}m) = 1$

$f \times (0.5) \times (2/3) = 1$

$f = 3$

B. False.

For the population 2:

$N_t/N_0 = [3 \times (1/3) \times (0.75)]^t = [0.75]^t$

$N_t/N_0 < 1$

So, the population does not have a constant number of adult females across generations.

C. True.

For the population 3: $N_t/N_0 = 1$; thus,

$1 = 4 \times r \times (0.75)$

$r = 1/3$

So, the female/male ratio is **1/2**.

D. True.

For population 4, in the first generation, the population size in adult stage is 192 and the number of females is **96**; hence, in the third generation number of adult females is:

$N = [6 \times (2/3) \times 0.5]^{(3-1)} \times (96) = \mathbf{384}$

References:

- Leah Edelstein-Keshet (1988). Mathematical models in Biology. Random House, New York, pp. 7-8
- Thomas G. Whitham (1980). *The American Naturalist*, Vol. 115, No. 4, pp. 449-466.

BIOSYSTEMATICS

Q.99

Information about the relationships among organisms is a useful source of data for scientists investigating a wide variety of biological questions. Indicate in the answer sheet if each of the following statements about using the phylogenetic trees is true or false.

A. Phylogenetic trees can be used to determine how many times a particular trait independently evolved.

B. Phylogenetic trees can suggest a particular trait is the ancestral state.

C. Phylogenetic trees can be used to determine the order in which evolutionary lineages split but not the timing of those splits.

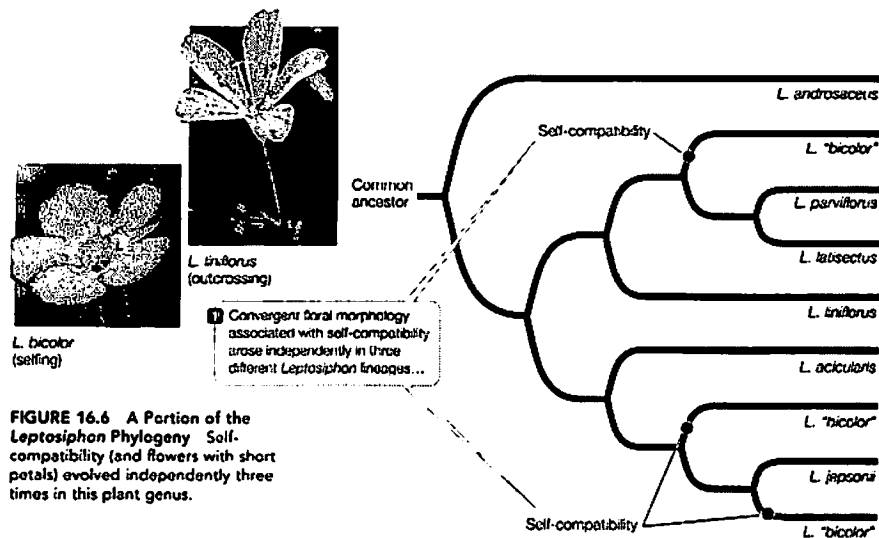D. Phylogenetic trees can be used to determine the virus's origins in human populations.

Answer key:

A. True.   B. True.   C. False.   D. True.

Explanation:

A. True. If a particuliar trait found in different taxa of the tree but these taxa do not have most rencent common ancestor then the number of taxa having this trait idicates number of time the trait independently evoleved. For example, the reconstructed phylogeny suggests that self-incomatibility is the ancestral state and that self-compatibility evolved three times within the group of *Leptosiphon* (see Fig below).

FIGURE 16.6 A Portion of the
Leptosiphon Phylogeny Self-
compatibility (and flowers with short
petals) evolved independently three
times in this plant genus.

B. True. See the explanation I (A). In addition to using phylogenetic methods to infer evolutionary relationships, biologists can use these techniques to reconstruct the morphology, behavior, or nucleotide and amino acid sequences of ancestral species.

C. False. A molecular clock uses the average rate at which a given gene or protein accumulates changes to gauge the time of divergence for a particular split in the phylogeny. Molecular clocks must be calibrated using independent data such as fossil record, known times of divergence, or biogeographic dates (e.g., the time of separations of continents). Using such calibrations, time of divergence have been estimated for many groups of species that have diverged over millions of years.

D. True. The molecular clock was used to project back to the common ancestor of a group HIV-1 sample. Extrapolation suggests a date of origin for the group of viruses of about 1930.

Reference: Principle of Life by David Hills 2010

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Q.100

The analysis of DNA and protein sequences nowadays is widely used in constructing phylogenetic trees.

Indicate in the answer sheet if in the answer sheet each of the following statements is true or false.

A. The number of differences in DNA or RNA nucleotide sequences or in amino acid sequences in two organisms reflects how much time has passed since the groups branched from a common ancestor.

B. If two polypeptides from two organisms sharing a most recent common ancestor differ from each other in only one amino acid, then one substitution had occurred during the evolution of the two polypeptides.

C. rRNA sequence analysis is useful not only for phylogenetic analysis of organisms belonging to different domains but also for phylogenetic relationships among species within genera.

D. Pseudogenes often used for constructing phylogenetic trees.

Answer key:

A. False. B. False.   C. False.   D. True.

Explanations:

A. False. This can be true only for certain sequences if the molecular changes that occur at a steady rate-specific macromolecules can be used as molecular clocks. Biology 9th Solomon et al. 2011.

B. False. At least two substitutions had occurred.

C. False. Variation in rRNA sequences is not big enough for identification of phylogenetic relationships among species within genera.

D. True. Molecular clock hypothesis hypothesized that rates of molecular change were constant that they could be used to predict evolutionary divergence times. Pseudogenes are functionless, so substitution rate is constant.

Reference: Principle of genetics 6th, (chapter 24 and appendix E)