Question	Answer	Question	Answer	Question	Answer	Question	Answer
1	4	41	1	81	3	121	3
2	3	42	1	82	8	122	6
3	1	43	3	83	4	123	4
4	2	44	1	84	1	124	5
5	1	45	1	85	2	125	4
6	1	46	3	86	3	126	4
7	2	47	2	87	4	127	2
8	1	48	2	88	3	128	
9	1	49	4	89	8	129	
10	3	50	3	90	2	130	
11	5	51	6	91	3	131	
12	3	52	7	92	4	132	
13	2	53	3	93	5	133	
14	1	54	2	94	2	134	
15	3	55	3	95	1	135	
16	2	56	6	96	2	136	
17	2	57	3	97	3	137	
18	1	58	7	98	4	138	
19	3	59	5	99	5	139	
20	2	60	2	100	7	140	
21	2	61	4	101	3	141	
22	3	62	2	102	3	142	
23	1	63	7	103	2	143	
24	4	64	2	104	8	144	
25	1	65	5	105	5	145	
26	2	66	3	106	6	146	
27	2	67	7	107	5	147	
28	2	68	5	108	2	148	
29	3	69	8	109	3	149	
30	3	70	7	110	4	150	
31	2	71	2	111	2	151	
32	1	72	3	112	5	152	
33	1	73	2	113	3	153	
34	6	74	1	114	6	154	
35	2	75	1	115	8	155	
36	5	76	1	116	1	156	
37	1	77	2	117	6	157	
38	2	78	4	118	1	158	
39	3	79	1	119	0	159	
40	2	80	2	120	4	160	

# **Theoretical Exam 2 Answer keys**

Points of questions are indicated in the question papers.

# **Biochemistry**

Q1

No.	1	2	3	4
Answer	(4)	(3)	(1)	(2)

**Q-1** (glycogen degradation)

## Answer: (4) about 500

 $10000 / 10 = 1000 \rightarrow 1000 / 2 = 500$ 

About half of linear chains can be exposed to phosphorylase.

**Q-2** (degradation graphs)

## Answer: phosphorylase (3), debranching enzyme (1).

According to the figure 1 and Q1, phosphorylase degrades nearly half of total chains.

The debranching enzyme only acts on the branching sites, which will be exposed by action of phosphorylase.

**Q-3** (amylopectin degradation)

## Answer: (2)

Qualitatively, breakdown efficiency is dependent on the number of enzyme-accessible chains, which are smaller in amylopectin than glycogen because more branching produces more chains in glycogen than amylopectin.

10000 / 25 = 400

This means amylopectin of 10000 residues consists of 400 linear chains, half of which can be degraded by phosphorylase. Namely, number of phosphorylase-accessible linear chains in amylopectin (200), which corresponds to the speed of degradation, is less than half of that in glycogen (500). On the other hand, final breakdown extent of amylopectin (200 \* 25 = 5000) is comparable to that of glycogen (500 \* 10 = 5000).

**Q2** (endo-type and exo-type hydrolases)

No.	5	6	7	8
Answer	(1)	(1)	(2)	(1)

## Answer: A (1), B (1), C (2), D (1)

Proteases in stomach are endo-type, because they can creates large number of peptides, which are degraded quickly by exo-type proteases (aminopeptidases and carboxypeptidases in small intestine).

Although the translocation signal peptide is usually found at N-terminus of the nascent polypeptides, it is removed efficiently by specific endo-type protease, followed by full degradation due to more general exo-type proteases.

Proofreading nuclease is  $5' \rightarrow 3'$  exonuclease, which removes newly added nucleotides by DNA polymerase.

Cas9 nuclease of the CRISPR-Cas9 is the endo-type nuclease, which dissects the chromosome DNA chain according to the guide RNA as an initial step of genome editing.

Q3 (competitive inhibitor of alcohol dehydrogenase)

No.	9	10	11
Answer	(1)	(3)	(5)

## Answer: 13.5

In methanol without ethanol,

 $v_0 / V_{\text{max}} = 5 / (10 \text{ x } 1 + 5)$ 

For 90% suppression by ethanol addition

 $\{5 / (10 \times 1 + 5)\} \times 0.1 = 5 / (10 \times \alpha + 5)$ 

Thus,  $\alpha = 14.5$ 

From eq 2, [I] = 13.5 mM

## **Cell Biology**

Q4 (Centrifugation)

No.	12	13	14	15	16	17	18	19	20	21	22	23
Answer	(3)	(2)	(1)	(3)	(2)	(2)	(1)	(3)	(2)	(2)	(3)	(1)

The rate of sedimentation during the centrifugation process depends on the size of specimens. It should be acquired by the examinee as textbook knowledge. In addition, it

would be general scientific knowledge that the centrifugal force is determined by the density difference between the specimen and the solvent. Based on the students' knowledge related to the size of biomolecules, viruses and cell nuclei (nuclei>viruses>biomolecules), the following questions were prepared to determine their logical thinking.

## **Q4-1** Exp. A (sedimentation in a uniform density medium)

### Answers: (3) III – viruses (2) b / globular proteins (1) a / nuclei (3) c

As the sedimentation speed of a specimen that has a similar density (1.3 g/mL) is dependent on size (nuclei>viruses>biomolecules), <u>nuclei sedimentation is greatest with an almost constant speed</u> as shown by the straight line, c, within diagram III of Figure 2. Viruses and biomolecules then follow nuclei.

## **Q4-2** Exp. B (sedimentation in a density-gradient medium)

## Answers: (2) II – viruses (2) b / globular proteins (1) a / nuclei (3) c

As the sedimentation speed of a specimen that has a similar density (1.3 g/mL) is dependent on size (nuclei>viruses>biomolecules), <u>nuclei sedimentation is greatest and stops</u> when the medium density (1.0 to 1.6 g/mL) is equal to the specimen density (1.3 g/mL) as shown by the curve, c, of diagram II within Figure 2. Viruses and biomolecules then follow nuclei.

#### **Q4-3** Exp. C (floating in a density-gradient medium)

#### Answers: (2) VI – viruses (2) b / globular proteins (3) c / nuclei (1) a

Similar to the sedimentation speed, the floating speed of a specimen that has a similar density (1.3 g/mL) is dependent on size (nuclei>viruses>biomolecules), whereby <u>nuclei</u> <u>float first and stop</u> when the medium density (1.0 to 1.6 g/mL) is equal to the specimen density (1.3 g/mL) as shown by the curve, c, of diagram VI in Figure 3. Viruses and biomolecules then follow nuclei.

**Q5** (Molecular crowding)

No.	24	25	26	27	28
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Answer (4) (1) (2) (2) (2)	
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'Molecular crowding' should be a rather new concept for the examinees. Here, we ask the students to calculate the volume ratio of hemoglobin molecules to solvent to understand how hemoglobin proteins are concentrated and crowded within red blood cells (RBCs). We also assess theoretical thinking based on the data obtained by measuring the actual rate of ion diffusion in RBCs. At the same time, we also set up a question to infer the compromising evolution of oxygen transport capacity (diffusion rate) and hemoglobin concentration (amount of oxygen transportation) found in alpaca.

#### **Q5-1** Answers: (4)

The concentration (320 g/L) of hemoglobin (MW = 64,000) corresponds to the molar concentration of 5 mM, where a single molecule is dissolved in a volume of 333 nm<sup>3</sup> on average (=1x10<sup>24</sup> nm<sup>3</sup>/Avogadro constant / 0.005 M). From the molecular weight, molecular mass is 1x10<sup>-19</sup> g. As the density of hemoglobin is 1.35 g/mL, its molecular volume is estimated to be 79 nm<sup>3</sup>. Thus, a single hemoglobin molecule should occupy 79/333 = 0.237  $\sim$  24% volume in the medium, as in the intracellular space of RBCs. We can also calculate this value using molar volume (A = 64,000/1.35 mL/mol) and occupied volume (B = 1000 mL/0.005). A/B = 0.237  $\sim$  24 %.

## Q5-2 A: True (1)

As described in the text and Figure 3, MCHC in alpaca RBCs is 450 mg/mL, approximately 1.5-fold higher than that in humans. It is also clear from Figure 3 that the  $H^+$ -diffusion rate in alpaca RBCs is less than half of that in human RBCs.

#### Q5-2 B: False (2)

RBCs increase their volume at lower osmosis as shown in Figure 2a.

#### Q5-2 C: False (2)

The relationship between the diffusion rate of ions and MCHC is almost <u>inverse</u> <u>proportionally</u> as shown in Figure 3.

#### Q5-2 D: False (2)

The high MCHC in alpaca RBCs would have been the result of evolutional adaptation to low oxygen at high altitude. However, the measured diffusion rate of ions, which may be corresponding to the diffusion rate of gas O<sub>2</sub> and CO<sub>2</sub> (not exactly the same) was low compared to those in humans and chickens owing to the effects of high molecular crowding (Figure 3). This would have been the result of compromising evolution, *i.e.*, the high capacity of  $O_2$  to be transported but the low rate of gas diffusion within RBCs.

#### Reference

S.L. Richardson & P. Sietach, Red blood cell thickness is evolutionarily constrained by slow, hemoglobin-restricted diffusion in cytoplasm. *Sci. Rep.*, **6**:36018 (2016).

Q6 (Cytoskeleton)

No.	29	30	31	32	33	34	35	36	37	38
Answer	(3)	(3)	(2)	(1)	(1)	(6)	(2)	(5)	(1)	(2)

The cytoskeleton, which is described in textbooks with a relatively large number of pages, supports the structure of cells and is an important organelle involved in intracellular transportation and cellular movement. As questions that evaluate comprehensive knowledge, we composed them to determine whether the examinees can link the shape of cells, functional roles and the type of cytoskeletons.

Description	Cytoskeleton type	<b>Figure number</b>
Α	(3)	(3)
B	(2)	(1)
С	(1)	(6)
D	(2)	(5)
Ε	(1)	(2)

#### Reference

C.M. Sahlgren et al., Mitotic reorganization of the intermediate filament protein nestin involves phosphorylation by cdc2 kinase. *J. boil. Chem.*, **276(19)**:16456-16463 (2001).

#### **Q7** (GLUT kinetics)

No.	39	40	41	42
Answer	(3)	(2)	(1)	(1)

The Michaelis–Menten equation is one of the most important concepts for understanding substrate affinity in enzymatic reactions. The examinees may not know the equation in terms of textbook knowledge, but I hoped the examinees could understand the exact meaning of  $K_m$  and  $V_m$ , from the mathematical formula



based on the experimental data showing the relationship between glucose concentration (substrate concentration) and amount of glucose transport (reaction rate). By answering the questions, I intended the students to learn how the difference in the rate of glucose transportation by GLUT is related to the differences in  $K_m$ , or substrate affinity.

- Q7-1  $V_{max} = 3 [nmol/min/cell]$
- **Q7-2** Km = 2 [mM]

**Q7-3** True: Calculated rates are 2.1, 0.71 and 0.74 [nmol/min/cell] for GLUT1, GLUT2 and GLUT4, respectively.

Q7-4 True

References

- A.M. Navale & A.N. Paranjape, Glucose transporters: physiological and pathological roles. Biophys. Rev., 8:5-9 (2016).
- H. Katagiri et al., Replacement of intracellular C-terminal domain of GLUT1 glucose

transporter with that of GLUT2 increases V<sub>max</sub> and K<sub>m</sub> of transport activity. *J. boil. Chem.*, **267(31)**:22550-22555 (1992).

H. Nishimura et al., Kinetics of GLUT1 and GLUT4 Glucose transporters expressed in *Xenopus* oocytes. *J. boil. Chem.*, **268(12)**:8514-8520 (1993).

**Q**8

No.	43	44	45	46
Answer	(3)	(1)	(1)	(3)

**A.** Answer: (3)

From (a) and (c) conditions, the carboxylase activity of Rubisco is not in saturating situation in the air. Since oxygen molecule works as the competitive inhibitor against CO<sub>2</sub>, the carboxylase activity increases when O<sub>2</sub> concentration in the air decreases.

## **B.** Answer: (1)

Addition to (a) and (c) conditions in the air, turn over rates of  $CO_2$  and  $O_2$  is found out from (b) condition. Thus, by using all conditions, it is supposed that the carboxylase activity of Rubisco is higher than the oxygenase activity.

## **C.** Answer: (1)

Turn over rate of  $CO_2$  in Rubisco is very low. Also, from (a) and (c) conditions, the carboxylase activity is low. Thus, to maintain the full capacity of photosynthesis, large amounts of Rubisco is necessary.

## **D.** Answer: (3)

From (a) and (c) conditions, the carboxylase activity of Rubisco is not in saturating situation in the air. The carboxylase activity increases when  $CO_2$  concentration in the air increases.

## Q9

No.	47	48
Answer	(2)	(2)

## **Q9-1** Answer: (2).

Experiments were carried out under nitrogen starvation. Under this condition, since supply of nitrogen is critical, protease becomes essential for yeast proliferation compared to the other enzymes.

## **Q-2** Answer: (2).

Yeast does not have Chloroplast and Melanosome. Cell wall are usually not digested by the autophagy in the yeast cells.

No.	49	50
Answer	(4)	(3)

#### Q10-1 Answer: (4).

This is a question in which knowledge about cell pattern in plants is asked.

## **Q10-2** Answer: (3).

The orientation of cellulose microfibrils of the cell wall (CW) usually coincide with the orientation of cortical microtubules (MT). Further the cells grow to direction perpendicular of the orientation of cell wall microfibrils.

#### Q11

No.	51	52	53
Answer	(6)	(7)	(3)

**Q11-1** Answer: somatic cell A: (6) somatic cell B: (7)

Somatic cell A has increased  $115.2 \div 7.2 = 16 (2^4)$  times in 48 hours. The cell cycle is 48  $\div 4 = 12$  hours. Somatic cell B has increased  $77.6 \div 9.7 = 8 (2^3)$  in 48 hours. The cell cycle is  $48 \div 3=16$  hours.

#### **Q11-2** Answer: (3).

Somatic cell A divides 8 times (96 / 12) in 4 days, and the number of cells increases  $2^8$  times. Somatic cell B divides 6 times (96 / 16) in 4 days, and the number of cells increases  $2^6$  times. If the mixture was mixed at 1:1, it would be  $1 \times 2^8 : 1 \times 2^6 = 4 : 1$ . However, in fact, it was 2:1, so at the initial, the number of somatic cell A is thought to be one-half the number of somatic cells B. The ratio of the initial number of cells is A : B = 1:2

Q12

No.	54	55	56
Answer	(2)	(3)	(6)

**Q12-1** Answer: Condition A: (2) Condition B: (3)

In condition A, since there is no O2 absorbtion, any aerobic respiration does not occur. In condition B, when the amount of O2 absorbed is 30 mL, the amount of CO2 released by aerobic respiration supposed from the aerobic respiration reaction formula is 30 mL. Since the amount of CO2 released is 40 mL, both aerobic respiration and alcohol fermentation occur.

## Q12-2 Answer: (6)

In condition A, alcohol fermentation only occurs. If the ATP produced is 100 equivalents, the glucose consumed is 50 equivalents because 100 equivalents is divided by 2. In condition B, since the amount of O2 absorbed is 30mL, the 30 mL CO2 is consumed. 10 mL of CO2 is released by the alcoholic fermentation. The amount of glucose consumed when 10 mL of CO2 is released, is half of condition A, so 25 equivalents of glucose are consumed. The remaining 25 equivalents are metabolized by aerobic respiration. From the reaction equation, the ATP produced by aerobic respiration is  $25 \times 32 = 800$  equivalents

The amount of ATP produced by alcoholic fermentation is  $25 \ge 2 = 50$  equivalents Therefore, the amount of ATP produced in condition B is 800 + 50 = 850 equivalents

# Genetics

## Q13

No.	57	58	59
Answer	(3)	(7)	(5)

## Answer: [ 3 ][ 7 ].[ 5 ]%

The genotype of the H gene of daughter Lisa is hh. Carl and Jane are neither O-type, so the genotype is Hh. From the parents' blood types, the genotypes of Carl and Jane are (BO, Hh) and (AB, Hh), respectively.

When Carl and Jane have another child, there are four genotypes, AB, BB, AO, and BO, and the phenotype can be B type in the cases of BB type and BO type. However, since the H gene becomes recessive homozygous hh and the phenotype becomes Bombay O type with a probability of 1/4, the probability that the phenotype eventually becomes B type is as follows:

 $2/4 \ge (1 - 1/4) = 3/8$  37.5%

No.	60
Answer	(2)

#### Answer: (2)

The sequence to be amplified is 100 bp, and a 20-mer primer is paired at both ends. The extension reaction extends 100 - 20 = 80 bases for one strand. From this, 160 dNTPs are consumed in two strands in one reaction. Since the template DNA is 4 copies, dNTPs consumed in the first cycle is  $160 \times 4 = 640$  base.

Since one primer pair is required for each molecule of template DNA, the number of primer pairs required in the first cycle is  $1 \times 4 = 4$  sets.

Since 2 DNA polymerases are required for 1 molecule of template DNA (double-stranded DNA),  $2 \times 4 = 8$  molecules of DNA polymerase are required in the first cycle.

After the second cycle, each component is required twice. For DNA polymerase, the same molecule is used for many times. Since the set of primer pair and dNTPs are consumed for each reaction, accumulative total is calculated.

For each component, the required numbers and totals for each cycle are shown in the table below.

#### Q15

No.	61	
Answer	(4)	

#### Answer: (4)

Since the activity of the xylose-metabolizing enzyme is proportional to the copy number of the corresponding gene, AT36 strain is presumed to have no alternative xylosemetabolizing system.

In the disrupted strain A, one *XYL1* gene is disrupted so that the XR activity is halved, but the XDH activity and the XK activity remain unchanged, so the produced xylitol is metabolized and the final xylitol production amount is reduced.

In the disrupted strain B in which both *XYL1* genes are disrupted, xylitol cannot be metabolized and xylitol is not produced at all.

In the disrupted strain C in which one of the *XYL2* genes is disrupted, the XR activity does not change, so the xylitol production does not change, but the XDH activity that metabolizes xylitol is halved, so the final xylitol content may be expected to increase. Be expected. Of the four disrupted strains, only disrupted strain C is expected to increase xylitol production. Graph (B) is the pattern of disrupted strain C.

In the disrupted strain D in which both XYL2 genes were disrupted, the host cannot utilize xylose, which is the only carbon source, so the yeast can no longer maintain the growth when glucose is consumed, and the xylitol production is significantly decreased. Graph (C) is the pattern of disrupted strain D.

#### Q16

No.	62	
Answer	(2)	

## Answer: (2)

During the replication process of the DNA chain, DNA polymerase III, DNA polymerase I, and DNA ligase work in this order.

In the result of Mr. B, the leading strand was replicated and the primer was not removed from the replicated lagging strand, so it is considered that DNA polymerase III worked but DNA polymerase I did not worked (R1).

In the result of Mr. C, it is considered that DNA ligase is not working because the ligation of the lagging strand has not occurred (R6).

#### Q17

No.	63
Answer	(7)

#### Answer: (7)

In general, the size of the genome correlates with the size and complexity of the organism.

A. Homo sapiens (human); 3000 Mb

B. Daphnia pulex (daphnia); 200 Mb

- C. Sahharomyces cerevisiae (baker's yeast); 12 Mb
- D. Acetobacter aceti (acetic acid bacteria); 2.9 Mb
- E. Caenorhabditis elegans (nematode);100 Mb

No.	64
Answer	(2)

## Answer: (2)

XY-3C strain ( $a/\alpha$ , ura3/URA3, leu2/LEU2, LEU1/leu1, HIS3/his3) is formed by conjugating XY-1A strain (a, ura3, leu2) and XY-2B strain ( $\alpha$ , his3, leu1). There are 2<sup>5</sup> = 32 spore genotype patterns obtained by meiosis of the XY-3C strain, depending on the combination of five genes,  $a/\alpha$ , URA3, LEU2, LEU1, and HIS3. Spores of all genotypes can grow on a medium containing uracil, leucine and histidine.

A strain that can grow on a medium containing only uracil has a probability of  $1/2^3 = 1/8$  because all three genes *LEU1*, *LEU2*, and *HIS3* must be dominant. Of the 160 spores, 160/8 = 20 is viable.

## Q19

No.	65
Answer	(5)

## Answer: (5)

Wild type should be amplified by the primer of (1+2), but not by the combination of (1+3).

Homozygous should not be amplified by the primers (1+2), but amplified by the combination of (1)+(3).

Hetero should be amplified by any combination of (1+2) and (1+3).

No.	66	67	68
Answer	(3)	(7)	(5)

#### Answer [ 3 ][ 7 ].[ 5 ]%

The blue flower trait is incompletely dominant because the intermediate light blue flower appears due to the cross between the blue flower and the white flower. The trait of red flowers is considered to be completely dominant over white flowers.

When the blue pure line BB strain and the red pure line RR strain are crossed, purple flowers with both traits appear, so the blue flower trait and the red flower trait are considered to be co-dominant. Individuals with a dominant red flower genotype It is considered that individuals with a heterozygous blue flower genotype bloom reddish purple flowers.

When the blue pigment gene is P and the red flower gene is Q, the genotype of the WW strain is (ppqq), the genotype of the BB strain is (PPqq), and the genotype of the RR strain is (ppQQ). The genotype of the BR strain of reddish purple flowers produced by the cross between the BB strain and the RR strain is (PpQq).

From the above, the genotypes and phenotypes of nest generation individuals are as shown in Punnet Square below. Reddish purple flowers are heterogeneous (Pp) for the P gene and at least one dominant gene for the Q gene (QQ) and (Qq).

Gamete	PQ	pQ	Pq	pq
PQ	PPQQ	PpQQ*	PPQq	PpQq*
	Purple	Reddish purple	Purple	Reddish purple
pQ	PpQQ*	ppQQ	PpQq*	ppQq
	Reddish purple	Red	Reddish purple	Red
Pq	PPQq	PpQq*	PPqq	Ppqq
	Purple	Reddish purple	Blue	Light blue
pq	PpQq*	ppQq	Ppqq	ppqq
	Reddish purple	Red	Light blue	White

The genotype of Reddish purple flowers is indicated by \*.

The probability of becoming a magenta flower is indicated below;

#### Q20

 $6 / 16 \ge 100 = 37.5\%$ 

6 / 16 x 100 = 37.5%

#### Q21

No.	69
Answer	(8)

## Answer: (8)

Since animal cells do not have RNA polymerase and reverse transcriptase that use RNA as a template, these genes must be included in the viral genome.

In the (-) single-stranded RNA viruses, whose RNA serves as templates for mRNA, mRNA encoding the capsid or the polymerase cannot be synthesized unless the RNAdependent RNA polymerase is included in the capsid.

When retroviruses infects animal cells, the viral genomic RNA should be reverse transcribed into DNA by reverse transcriptase and integrate into the cell's genome, so the reverse transcriptase must be encapsulated in the capsid.

## Q22

No.	70
Answer	(7)

## Answer: (7)

The gene A fragment obtained by digesting plasmid 1 with EcoRI and ClaI is inserted into the EcoRI and ClaI sites of the pBR322 vector to obtain the first-stage recombinant plasmid. When plasmid 1 and pBR322 are cleaved with EcoRI and BamHI, the next gene cannot be cloned because the ClaI site is deleted.

If the gene B is firstly cloned by cleaved with ClaI, BamHI or ClaI, SalI (or XhoI), the gene A cannot be cloned because the EcoRI site exists inside the gene B gene.

Next, the Gene B fragment obtained by cleaving plasmid 2 with ClaI and XhoI, and is inserted into the ClaI and SalI sites of the pBR322 vector to obtain the desired plasmid. Since SalI and XhoI produce the same cleavage terminals, the fragments can be ligated.

## Animal biology

Q23

No.	71	72	73
Answer	(2)	(3)	(2)

## Q23-1

Answer: False Oocytes that decrease by birth are at meiosis I, not meiosis II.

## **Q23-2** Answer: (3)

The graph shows that the duration from menarche to menopause is about 37 years. During that time, the number of ovulation is about 450 ( $37 \times 12 = 444$ ). The number of egg cells at menarche is about 500,000. So, the ratio of ovulation during the menstrual periods is about 0.08% of total germ cells.

## **Q23-3** Answer: (2)

In the ovary, the primary oocytes remain in the diplotene stage of first meiotic prophase until oocyte maturation just before ovulation. During diplotene, the homologous chromosomes remain attached at various points and paired in the oocytes. From this reason, the risk of aneuploidy due to errors in chromosome segregation increases as maternal age increases, causing Down's syndrome.

Q24

No.	74	75	76	77
Answer	(1)	(1)	(1)	(2)

## Q24-1 Answer: True (1)

The section from E to F indicates the period from the contraction of the atrial muscle and transmission of excitation into the apex of the ventricle via the Purkinje fiber, until the contraction of the ventricular muscle begins. The slope of the pacemaker potential at the sinoatrial node largely contributes to the increase of heart rate, and the time during this period does not significantly change. Thus, the statement is True.

#### Q24-2 Answer: (1)

In the period from H to I, the intraventricular pressure decreased, but the volume did not change. The corresponding period in the graph is (1).

## Q24-3 Answer: True (1)

After the point I, the intraventricular pressure is lower than the atrial pressure, and

the atrioventricular valve opens. Thus blood can flow into the atria to the ventricles. Therefore, this statement is True.

## Q24-4 Answer: (2)

When a physical exercise is performed, the heart's contraction force increases, resulting in the elevation of internal pressure. However, the change of the left ventricle volume with one beat is expected to become smaller. Therefore, (2) is the correct answer.

## Q25

No.	78	79	80	81
Answer	(4)	(1)	(2)	(3)

## A. Answer: (4)

Point mutations of actin at which it binds to tropomyosin increase Ca2+ sensitivity and facilitate contraction.

## **B.** Answer: (1)

Botulinum toxin inhibits muscle contraction by blocking the release of Ach from the synapse.

## C. Answer: (2)

When Ca2+ pump in skeletal muscle is impaired, muscle relaxation may be inhibited and it is difficult to extend the arm.

#### **D.** Answer: (3)

Depolarization by depletion of Ca and Mg in the blood is expected to direct convulsions.

## Q26

No.	82
Answer	(8)

## Answer: (8)

Gene A is a gene that regulates chondrocyte differentiation and is essential for cartilage formation (so choice 1 is excluded). Gene A works as a negative factor in the process of ossification because this gene supports cartilage survival, thus Loss of function of gene A inhibits cartilage formation, resulting in no bone formation. When gene A is upregulated, both cartilage and bone are formed, but bone formation is suppressed/delayed. Thus, the choices 2-5 are excluded.

Gene B is a gene that regulates osteoblast differentiation and is essential for induction of mineralized bone. A loss of gene B function does not affect cartilage formation but inhibits bone formation. In upregulation of gene B, both cartilage and bone are formed, but bone formation is promoted. Together with these results, the correct answer is 8.

No.	83	84
Answer	(4)	(1)

#### Q27-1 Answer: (4)

The difference in Waggle duration between experiment 2 and 4 is 529-441 = 88 ms. This duration corresponds to the difference in the distance flown outside (35-6 = 29 m). Considering from this, the waggle duration for outside flight is  $88 \text{ ms}/29 \text{ m} \cong 3.03 \text{ ms/m}$ . The Waggle duration for a 35 m or 6 m outdoor flight can be estimated to be 106.05 ms and 18.18 ms, respectively. The Waggle duration for the tunnel flight can be estimated to be about 423 ms by 529 - 106.05 ms (experiment 2) or 441 - 18.18 (experiment 4).

In this case, the tunnel is twice as long, so the Waggle dance duration for the tunnel flight can be estimated to be 846 ms (423 ms x 2).

The correct answer is 952 ms: 846 ms + 106 ms (3.03 ms/m x 35 m: the Waggle duration corresponding to 35 m in the outside).

#### Q27-2 Answer: (1)

- 1. The mosaic has a change of scenery when moving in the tunnel, but the horizontal stripe does not change the scenery when moving. It is possible to expect that the number of Waggle dancers will increase when the vertical stripes are used because the change of scenery can easily recognized, compared with the horizontal ones. **Ture**
- 2. Considering from experiments 2 and 3, bees do not decide the duration of Waggle dance by the absolute distance from the nest to the feeding site. Therefore, there are different feeding places for which the Waggle dance duration is same, the time required for arrival is not the same even if the flight speed is the same. False
- **3.** Experiments 2 and 3 show that the distance recognized by bees varies with the surrounding landscape. Therefore, even if you go to the same feeding place, the Waggle duration changes if the route is different. **False**

No.	85	86	87	88
Answer	(2)	(3)	(4)	(3)

#### **Q28**

## **A.** Answer: (2)

The drug enters the liver from the intestine through the portal vein, then, it is detoxified, and is delivered to the heart through the inferior vena cava.

#### B. Answer: (3)

The lower the metabolism, the better the effect of the drug.

### C. Answer: (4)

From Figures 2 and 3, Japan has a highest proportion of poor metabolizers and intermediate metabolizers, so omeprazole remains in the blood for a long time without being detoxified.

### **D.** Answer: (3)

It is expected that the effect will be further reduced by adding the competing omeprazole to the patient, who originally has low metabolism by CYP2C19.

#### Q29

No.	89	90
Answer	(8)	(2)

## Q29-1 Answer: (8)

Generally, larger genome is more likely to have new mutations. Also, RNA are chemically more unstable compared with DNA. Therefore, they are susceptible to mutation. Segmented genome with polycistronic genes will have better chance to recombine with other type of viruses to exchange the genome segments without losing any gene function. This happens in the body of natural reservoir, when the individual is infected with multiple type of virus simultaneously.

## **Q29-2** Answer: (2)

The free movement of animals that are infected with the virus without development of the disease is a major factor in spreading the infection. Also, the emergence of diverse viruses with new mutations is responsible for the emergence of new hosts to infect. In addition, the migration of animals that have been deprived of their original habitat due to climate change are increasing the risks for humans and livestock to contact with emerging infectious diseases.

## **Plant biology**

Q30

No.	91	92	93	94	95	96
Answer	(3)	(4)	(5)	(2)	(1)	(2)

## Q30-1 Answer: (3) > (4) > (5) > (2) > (1)

Because this plant forms only red leaves after the formation of the first red leaf, the number of red leaves can be calculated from the timing of the appearance of 'plants with red leaf' and the increase of 'total leaf number' thereafter.

- (1) 16L8D: no red leaf formation.
- (2) 14L10D: 'Plants with red leaf' appeared at day 64 to 65 on average. 'Total leaf number' was about 19 at this time and about 21.5 at day 70. Then the average number of red leaves of 70-day-old plants is calculated as 21.5 19 + 1 = 3.5.
- (3) 12L12D: 18 10 + 1 = 9.
- (4) 8L16D: 10 4.5 + 1 = 6.5.
- (5) 7L8D1L8D: 10 6 + 1 = 5.

## **Q30-2** Answer: (2)

When a high level of signal X sufficient to induce fully red leaf formation is suddenly, instead of gradually, applied to the shoot apex of plants with no red leaf, hypothesis I will be supported if newly expanded leaves are all fully red, and hypothesis II will be supported if the first few newly expanded leaves are partially red and then fully red leaves are formed. Among experiments (1) to (4), only (2) can make such situation.

Reference

K. Iwamoto, H. Fukuda, M. Sugiyama (2001) Elimination of POR espression correlates with red leaf formation in *Amaranthus tricolor*. *The Plant Journal* 27, 275–284.

## Q31

No.	97
Answer	(3)

Answer: (3)

- A. After culture of the wild-type protonemata, both auxin and cytokinin were detected in the culture medium, which had not contained any plant hormones before culture. This result indicates that the wild-type protonemata secrete both auxin and cytokinin
- **B.** Bud formation in the wild type was inhibited by continuous medium exchange but rescued by addition of both auxin and cytokinin, indicating that both auxin and cytokinin are involved in bud formation. Bud formation did not occur in mutant x under the standard condition but was rescued by addition of cytokinin, indicating that mutant x is normal in sensing and responding to both auxin and cytokinin but is defective in biosynthesis and/or secretion of cytokinin.
- **C.** Culture of protonemata at a higher density is expected to increase the concentrations of auxin and cytokinin in the medium and thus increase the sites where auxin and cytokinin levels exceed the thresholds required for bud formation.

#### References

- N. W. Ashton, D. J. Cove, D. R. Featherstone (1979) The isolation and physiological analysis of mutants of the moss, *Physcomitrella patens*, which over-produce gametophores. *Planta* 144, 437–442.
- N. W. Ashton, N. H. Grimsley, D. J. Cove (1979) Analysis of gametophytic development in the moss, *Physcomitrella patens*, using auxin and cytokinin resistant mutants. *Planta* 144, 427– 435.
- D. J. Cove (1984) The role of cytokinin and auxin in protonemal development in *Physcomitrella patens* and *Physcomitrium sphaericum*. *Journal of the Hattori Botanical Laboratory* 55, 79–86.

## Q32

No.	98
Answer	(4)

#### Answer: (4)

- A. Comparison of the bending of unpretreated fruit valves between air, water, and 4M NaCl conditions shows that fruit valves bend more at higher turgor pressure.
- B. In the intact fruit valve, the outer layer is under stress to shrink longitudinally.

Therefore, if a shallow cut was made perpendicular to the stress direction, the stress is released to cause a longitudinal shrinkage, which appears as an opening of the cut.

**C.** Recall the turgor movement of a guard cell. Increased turgor pressure not necessarily leads to isotropic expansion of a cell. Anisotropic cell wall stiffness or geometry sometimes lead to shrinkage in a specific direction. However, the increased turgor pressure should lead to a volume increase, which should accompany an extension in at least one direction.

#### Reference

H. Hofhuis, D. Moulton, T. Lessinnes, A.-L. Routier-Kierzkowska, R. J. Bomphrey, G. Mosca,
H. Reinhardt, P. Sarchet, X. Gan, M. Tsiantis, Y. Ventikos, S. Walker, A. Goriely, R. Smith,
A. Hay (2016) Morphomechanical innovation drives explosive seed dispersal. *Cell* 166, 222–233.

#### Q33

No.	99
Answer	(5)

## Answer: (5)

Characters associated with the organ function are greatly modified in a tendril from the original organ-specific characters, and thus are not informative for judging whether a given tendril is a modified leaf or a modified stem. In contrast, characters determined by developmental constraints are not much changed in a tendril from the original organ-specific characters and thus informative. Among observations (1) to (8), only (5) is an observation of such developmentally constrained characters.

#### Q34

No.	100
Answer	(7)

Answer: (7)

- A and B. If the position of a new leaf is determined by the inhibitory effect from only its immediately preceding leaf, a new leaf should always arise at the site opposite to the immediately preceding leaf. This is not the case in this plant. However, when the effect from  $P_0$  was intercepted by microsurgical isolation of it,  $P_2$  was formed at the site almost opposite to  $P_1$ . This indicates that the second preceding leaf in addition to the immediately preceding leaf are critical in determination of new leaf position.
- C and D. Microsurgical isolation on  $P_0$  changed the position of  $P_2$ , while the position of  $P_1$  was unchanged.
- E and F. Like in normal seedlings of this plant, the leaf arrangement pattern is expected to settle into the Fibonacci spiral.

#### Reference

D. Reinhardt, M. Frenz, T. Mandel, C. Kuhlemeier (2005) Microsurgical and laser ablation analysis of leaf positioning and dorsoventral patterning in tomato. *Development* 132, 15–26.

Q35

No.	101	102
Answer	(3)	(3)

#### Q35-1 Answer: (3)

Competition for reductants between nitrogen assimilation and carbon assimilation lowers CAQ. As application of ammonium skips its upstream steps of nitrogen assimilation,  $\Delta$ CAQ correlates positively with the activity of the nitrate-to-ammonium process of nitrogen assimilation.

## Q35-2 Answer: (3)

In C4 plants, unlike in C3 plants,  $\Delta$ CAQ is unaffected by Ci and always very low. This indicates that the nitrate-to-ammonium process of nitrogen assimilation does not compete with the Calvin cycle, which occurs in bundle sheath cells, for reductants.

#### Reference

A. J. Bloom, J. S. R. Asensio, L. Randall, S. Rachmilevitch, A. B. Cousins, E. A. Carlisle (2012)
 CO<sub>2</sub> enrichment inhibits shoot nitrate assimilation in C<sub>3</sub> but not C<sub>4</sub> plants and slows growth

under nitrate in C<sub>3</sub> plants. Ecology 93, 355-367.

## Evolution

#### Q36

No.	103	104	105	106
Answer	(2)	(8)	(5)	(6)

Answer: 20 years old: 0.28, 40 years old: 0.56

Age 20: Among 40 mutations derived from paternal side, the number of mutations within genes is  $40 \times 1$ kb $\times 10000/1$ Gb =  $40 \times 10^3 \times 10^4/10^9 = 4 \times 10^8/10^9 = 0.4$ . Since the probability that a mutation within a gene is deleterious is 70%, the number of deleterious genes derived from paternal side is  $0.4 \times 0.7 = 0.28$ .

Age 40: Among  $40 + 20 \times 2 = 80$  mutations derived from paternal side, the number of mutations within genes is  $80 \times 1$ kb $\times 10000/1$ Gb =  $80 \times 10^3 \times 10^4/10^9 = 8 \times 10^8/10^9 = 0.8$ . Since the probability that a mutation within a gene is deleterious is 70%, the number of deleterious genes derived from paternal side is  $0.8 \times 0.7 = 0.56$ .

Q37
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No.	107	108
Answer	(5)	(2)

#### A. **Answer:** (5)

The numbers of Hox clusters in zebrafish and medaka are logically expected to 8, however, they lost one of them after the last (3rd) whole genome duplication.

#### B. Answer: (2)

Gene duplication occurred in the common ancestor of X, and the common ancestor of Y and Z.

**Q38** 

No.	109	110	111
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Answer (3)	(4)	(2)
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## A. **Answer:** (3)

In protein X, 400 x 0.625 x  $10^{-9}$  x 8 x  $10^7$  <u>x 2</u> = 40 In protein Y, 600 x 1.25 x  $10^{-9}$  x 8 x  $10^7$  <u>x 2</u> = 120

B. Answer: (4)

 $0.625 \ge 10^{-9} \ge 400 \ge T \ge 2 = 6$ , T = 1.2  $\ge 10^{7}$ 

## C. Answer: (2)

The substitution rate of A-B domain is smallest because this domain is active insulin and is expected to be most important in function.

#### Q39

No.	112
Answer	(5)

#### Answer: (5)

The number of substitutions between sequences of bottlenose dolphin and the other species reflect the distances (divergence times). We can infer the species name based on these distances. Students can also refer to question Evolution Q42 in Exam 1 for phylogenetic relationships of cetartiodactyls.

No.	113	
Answer	(3)	

This figure was modified from Keeling (2004) Am. J. Botany

#### Answer: (3)

In the first symbiotic uptake of a cyanobacterium, phagosomal membrane was lost. In the second symbiotic uptake of a green alga, phagosomal membrane was retained.



# Ecology

## Q41

No.	114	115	116	117	118	119	120
Answer	(6)	(8)	(1)	(6)	(1)	(0)	(4)

## Q41-1 Answer: 68, 16, 10

The numbers of individuals are calculated as follows.

$$N_{J,2} = (100 \times 0.2 \times 0.5 \times 2) + (100 \times 0.2 \times 3 \times 0.2 \times 3) + (20 \times 0.5 \times 2 \times 0.2 \times 3) = 20 + 36 + 12 = 68$$
$$N_{A1,2} = 100 \times 0.2 \times 3 \times 0.2 + 20 \times 0.5 \times 2 \times 0.2 = 12 + 4 = 16$$
$$N_{A2,2} = 100 \times 0.2 \times 0.5 = 10$$

### Q41-2 Answer: 4

To maintain the constant number of individuals through time, the number of juveniles should be 100 in the next generation. This means that the following equation needs to be satisfied.

 $100 \times 0.2 \times m_1 + 20 \times 0.5 \times 2 = 100,$ 

which results in  $m_1 = 4$ 

No.	121
Answer	(3)

#### Answer: (3)

Responses to a - d are described as follows.

a. Fertilization increases the total biomass.

b. Damage on apical meristem cause the growth of lateral shoot.

c. Shade condition induces shade avoiding response, such as spindly growth and internode elongation.

d. Trampling pressure suppresses the vertical growth.

#### Q43

No.	122	123
Answer	(6)	(4)

#### Answer: Q43-1(6), Q43-2(4)

Direct fitness is one-half of the number of offsprings produced.

Inclusive fitness represents the sum of direct and indirect fitness. The indirect fitness of an individual is calculated by adding the relations of that individual multiplied by the degree of relatedness. For example, the inclusive fitness of genotype A is 13.875, calculated by 6 (direct fitness) +  $3 \times 7 \times 0.375$  (relatedness of full sibling's offspring in haplodiploidy;  $0.5 \times 0.75$ ).

Q44

No.	124	125	126
Answer	(5)	(4)	(4)

### Q44-1 Answer: 54

The pathways from herbivores or detritivores to passerine birds include direct link and indirect link through spiders. Loss of energy across trophic levels needs to be taken into

account for the indirect link. Since conversion efficiency across trophic levels is 10%, the energy flow of the indirect link should be 10 times larger than the direct link. The contribution of the pathway from detrital food web to passerine birds is calculated as follows.

 $(0.2+0.3\times0.6\times10)$  /((0.2+0.3×0.6×10)+(0.5+0.3×0.4×10)) = 0.54051

#### Q44-2 Answer: (4)

Since spiders consume both herbivores and detritivores, the decreasing pattern of cesium concentration of spiders should be intermediate between grasshoppers (herbivores) and earthworm (detritivores). As soil Cs is difficult for vascular plants to absorb while fungi can accumulate Cs, earthworms continue to have higher levels of Cs, while grasshoppers retain lower Cs with time.

#### Q45

No.	127
Answer	(2)

### Answer: (2)

Soil nitrogen starts from almost zero in the primary succession and increases with time. Phosphorus, in contrast, is lost through time and is not accumulated with time.