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# **IBO 2013 Theoretical Exam 2**

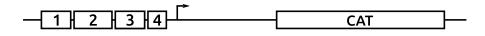
Translation version: English (Official), downloaded by: staff1



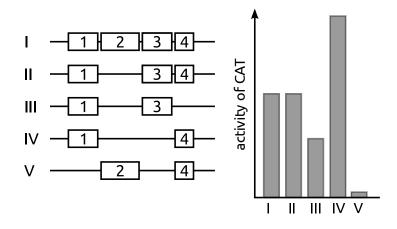
# 1

#### Cell-, molecular- and microbiology

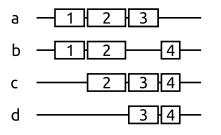
A newly discovered gene promoter is characterized by a reporter gene assay using chloramphenicol acetyltransferase (CAT) as a reporter gene. Linear pieces of double-stranded DNA of four potential promoter elements (white boxes, numbered 1 to 4) are placed upstream of the CAT reporter gene. --restored!



After transfection of individual constructs into cells, the [new text here] following CAT activities were determined.



The following constructs have not tested yet.



## Indicate if each of the following statements is true or false.

- A. Construct a is a stronger promoter than construct I.
- B. Construct a is a stronger promoter than construct b.
- C. Construct c is a stronger promoter than construct b.
- D. Construct c is a stronger promoter than construct d.
- A. False B. False C. False D. False

#### **Original commentary**

Notes I = II -> 2 plays no role, I > III -> deletion of 4 reduces the promoter strength -> 4 enhances promoter activity, I < IV -> removal of 3 increases promoter strength -> 3 reduces promoter activity, V shows almost no activity -> 1 provides basal promoter activity Correct answers A *false* 

4 enhances promoter activity, removal of it in a decreases the activity of a.

B false

This case is similar to III versus IV, since 2 does not affect promoter strength.

C false

Without 1, c will show almost no activity, further decreased by the presence of 3.

D false

2 does not play a role, c and d have the same promoter strength.

In genetic engineering, it is often desired to increase the yield of secreted proteins.

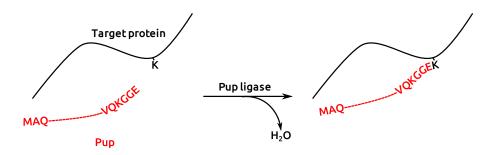
Indicate if each of the following strategies is expected to increase the yield in a mammalian cell.

- •
- .
- -
- •
- •
- A. Overexpression of chaperones (proteins assisting others to fold) present in the endoplasmatic reticulum.
- B. Deletion of the genes coding for glycosylating enzymes present in the endoplasmatic reticulum.
- C. Overexpression of proteins facilitating the fusion of secretory vesicles with the cell membrane.
- D. Duplicating the gene encoding the desired protein.
- A. True B. False C. True D. True

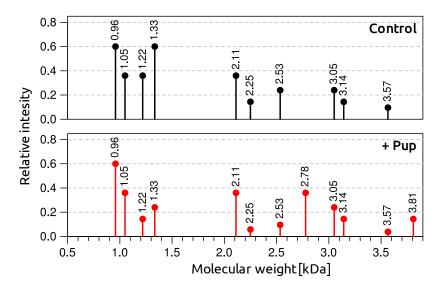
#### Original commentary Correct answers A *true* secretory proteins are folded in the ER, chaperones assist them in their folding and can increase the yield B *false* most secretory proteins are glycosylated, without glycosylation they are not recognised and are not trafficked further, and without glycosylating enzymes, the cell's membrane proteins cannot be glycosylated, the cells are not so healthy, this will also reduce the yield. C *true* secretory proteins are transported from the Golgi to the plasma membrane by vesicles D *true* Since each of the gene copies can be transcribed independently, genes with multiple copies are generally expressed more. References Peng et al, Biotechnology and Bioengineering (2009)

<u>Tigges et al, Metabolic engineering (2006)</u>

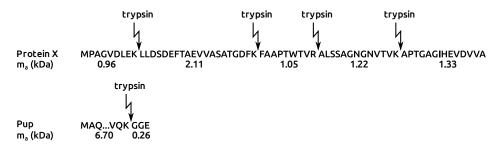
Pupylation is a post-translational protein modification found in some *Actinobacteria* in which the short protein Pup is ligated to a lysine side chain of a target protein by the Pup ligase.



To determine if a protein X is pupylated, purified X is incubated with Pup and the Pup ligase over night. Trypsin, which hydrolyses (by addition of water of 0.018kDa) proteins next to lysines (K) and arginines (R), is added and the masses of the peptides are determined by mass spectrometry (red). A control reaction where Pup was not added was processed similarly (black). The masses were rounded to two decimals. Note that the Pup fragments are out of the range to be detected.



The sequences of protein X and Pup as well as the molecular weight  $(m_a)$  of the peptides are indicated below.



## Indicate if each of the following statements is true or false.

A. Trypsin efficiency is such that several peptides are left partly uncut.

- B. Hydrolysis by trypsin is inhibited next to lysines that are pupylated.
- C. Under these conditions, pupylation is specific to a single lysine.

D. The target protein is pupylated at about 90%.

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

#### **Original commentary**

Note

The masses given in the second figure correspond to tryptic monopeptides. During trypsin hydrolysis, water ( $m_a$  18.02 Da) is added, therefore, when calculating the mass of a dipeptide, the mass of water needs to be substracted.

Correct answers

A true

Masses corresponding to dipeptides can be observed in the black sepctra, e.g. the mass 3.05 kDa corresponds to a dipeptide formed by the two first tryptic peptides. B *true* 

The peak of the pupylated peptide at 2.78 kDa corresponds to a pupylated dipeptide and no pupylated monopeptide can be observed (the peak would be at 1.46 kDa for this lysine).

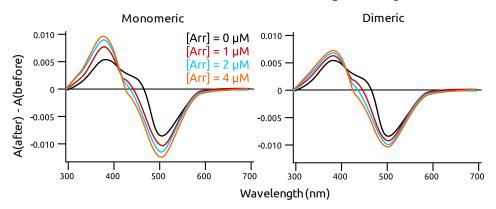
C true

Only two peaks (2.78 and 3.81 kDa) are present in the red spectra (with Pup) but not in the black one. The peak at 2.78 kDa corresponds to a pupylated dipeptide formed by the last two peptides of the protein, the peak at 3.81 corresponds to a pupylated tripeptide formed by the last three peptides of the protein.

D *false* Two peaks (1.22 and 1.33 kDa) are reduced after ligation with Pup. The peak at 1.33kDa goes from 0.6 to about 0.25 relative intensity. Therefore, the protein is pupylated to about 1-(0.25/0.6) = 58%, not 90%. Furthermore, the peak corresponding to a pupylated peptide (2.78 kDa), has a relative intensity of only 0.4, added to the peak corresponding to the pupylated tripeptide (3.81 kDa) with a relative intensity of about 0.15, the pupylation level would be only about 55%.

#### Cell-, molecular- and microbiology

Upon light activation, rhodopsin proceeds to Meta-states, of which Meta-I and Meta-II are in a dynamic equilibrium. This shifts towards Meta-II in the presence of arrestin (Arr) as some Meta-II binds to arrestin. To study the minimal functional unit of rhodopsin, the difference in absorption of rhodopsin (absorption after light activation minus absorption before light activation) is measured at different arrestin concentrations ([Arr]) for both monomeric and dimeric rhodopsin, as shown below. Meta-II is known to show a stronger absorption at 380 nm than Meta-I.



#### Based on these results, indicate if each statement is true or false.

- A. Before light activation, rhodopsin absorbs light at 500 nm.
- B. The fraction of rhodopsin in the Meta-II state increases linearly with the concentration of arrestin.
- C. Dimeric rhodopsin binds tighter to arrestin than monomeric rhodopsin.
- D. Monomeric rhodopsin is the minimal functional unit.

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

- Correct answers
- A true

B false

as can be seen from the graph, the difference in absorption between 2 and 4  $\mu M$  is smaller than the one between 1 and 2  $\mu M$ . If Meta-II would increase linearly with the arrestin, it should be equal to the double. Furthermore, one can think, that as more arrestin is added, less free rhodopsin is present, therefore, less increase in absorption is observed. C false

with monomeric rhodopsin, the increase of absorption at 380 nm is bigger than with oligomeric rhodopsin D *true* 

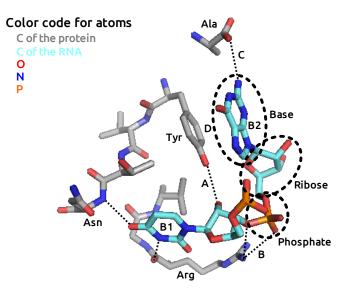
monomeric rhodopsin is enough for arrestin-binding, it is the minimal functional unit

**Original commentary** 

the negative difference absorption at 500 nm shows that dark-state rhodopsin absorbs light at 500 nm.

#### Cell-, molecular- and microbiology

The following picture depicts part of the binding site of a protein which recognizes RNA specifically over DNA, in complex with two RNA bases (B1 and B2).



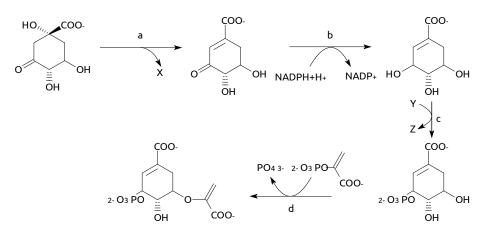
# Indicate for each of the following interactions if it provides specificity for RNA over DNA.

- A. Hydrogen bond A with Tyr
- B. Hydrogen bonds B with Arg
- C. Hydrogen bond C with Ala
- D. Hydrophobic stacking D between Tyr and B2
- A. True B. False C. False D. False

## Original commentary Correct answers A *true* Only RNA, not DNA, has a hydroxyl group at the 2' position in the sugar. B *false* Both RNA and DNA have phosphate groups in the backbone. C *false* This hydrogen bond is specific for B2, but B2 is a guanine present both in RNA and DNA (the only difference in term of bases between RNA and DNA is uracil vs. thymine, both are pyrimidines, small bases with only 1 ring). D *false* Hydrophobic stacking is a non specific interaction which is found in interactions both with DNA and RNA.

Handa et al, Nature (1999)

The following figure depicts the shikimate pathway, which is part of the synthetic pathway producing aromatic amino acids in bacteria.



## Indicate if each statement is true or false.

A. In reaction a, X represents  $H_2O$ .

- B. In reaction b, the substrate is reduced.
- C. In reaction c, Y represents ADP or GDP.
- D. Reaction d is a phosphorylation of the substrate.
- A. True B. True C. False D. False

#### **Original commentary**

Correct answers A true

A is a dehydration, the -OH group as well as a hydrogen are removed and form water.

B true

NADPH+ $H^+$  is a reducing agent. The oxidation number of the carbon of the carboxyl in the substrate changes from +2 to +1 in the product of b: this is a reduction.

C false

GDP cannot phosphorylate substrates, Y represents here ATP.

- D false
- In d, a phosphate group is released as part of the condensation of the 2 substrates.

To separate DNA fragments on an agarose gel, one liter of 10x TAE buffer (consisting of Tris base, Acetic acid and EDTA) has to be prepared. The desired concentrations for TAE as well as the available stocks are as follows:

Chemical	Desired concentration	Available stocks Powder (121 g/mol)	
Tris base	0.40 M		
Acetic acid	1.14%	Solution 100 %	
EDTA 0.01 M		Solution 0.50 M	

Distilled water

## Indicate if each of the following statements is true or false.

A. 4.84 g of Tris base are required.

B. 11.4 ml of acetic acid are required.

C. 0.2 ml EDTA are required.

D. Tris base, EDTA and acetic acid have to be added to 1 l of distilled water.

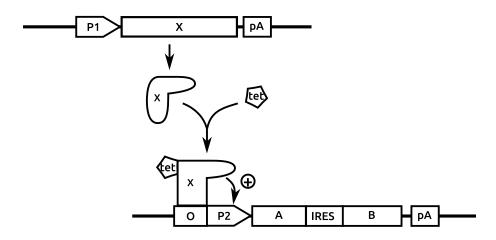
A. False B. True C. False D. False

## **Original commentary**

Correct answers A *false* Tris base: 0.4 mol/l \* 1 l \* 121 g/mol =48.4 g B *true* acetic acid: dilution: V1 = c2\*V2/c1 = 1.14% \* 11/100% = 11.4 ml C *false* EDTA: dilution: c1\*V1 = c2\*V2 <=> V1 = c2\*V2/c1 = 0.01 mol/l \* 1 l / 0.5 mol/l D *false* Tris, EDTA and acetic acid are first added to a smaller amount of water. When Tris is completely dissolved, the volume is adjusted to 1 l with water. If they were added to 1 l of water, the final volume would be greater than 1 l and the concentrations would be wrong.

#### Cell-, molecular- and microbiology

A protein complex consisting of two polypeptides A and B needs to be expressed. To ensure proper folding and complex formation, a eukaryotic host is employed. To achieve this, eukaryotic cells are co-transfected with two artifical gene constructs. In the first construct (above), the gene coding for Transcription Factor X is under the control of Promoter P1. In the second construct (below), the genes coding for A and B are separated by an internal ribosomal entry site (IRES) and under the control of a specific Promoter P2. In the presence of tetracycline, X can bind to the Operator O and activate P2, as shown below (pA indicates polyadenylation sites).



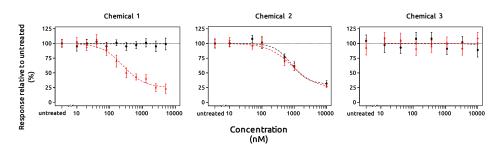
## Indicate if each of the following statements is true or false.

- A. The expression of the genes coding for A and B can only be induced by tetracycline if P1 is active.
- B. In the presence of tetracycline, the gene coding for B is expressed at higher levels than the gene of A if the ribosome binds stronger to the 5'-cap than to the IRES.
- C. If the DNA-binding domain of X is removed, the genes coding for A and B are expressed even in the absence of tetracycline.
- D. The length of the polyadenine tail of the mRNA of X influences the amount of synthesis of protein X in the cell.
- A. True B. False C. False D. False

Original commentary Correct answers A <i>true</i> A and B can only be expressed if X is present in the cell, for this, P1 needs to be active. B <i>wrong</i> gene B would be more expressed than A if the ribosome would bind stronger to the IRES than the 5'-cap C <i>false</i> if the DNA-binding domain of X was removed, X could not bind the DNA and activate gene expression, so neither A nor B would be produced. D <i>true</i> an mRNA with a longer or shorter poly-A tail may survive longer in the cell and can be translated during a longer time,
so more X-protein could be produced.
References <u>Fussenegger et al, Biotechnol. Prog. (2001)</u>

#### Cell-, molecular- and microbiology

The toxicity of three different chemicals 1-3 was tested *in vitro* on neural crest cells by counting the number of viable cells (black circles) and the number of cells migrating (red triangles). The average and standard deviation of multiple replicates obtained at different concentrations are plotted relative to the average counts from untreated cultures.



## Indicate for each of the following statements if it is true or false.

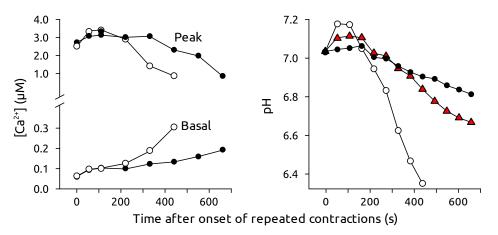
- A. These results are compatible with Chemical 1 affecting receptors receiving migration signals.
- B. These results are in agreement with Chemical 2 affecting cell viability only.
- C. 100 to 500 nM of Chemical 3 are likely beneficial for the viability of neural crest cells.
- D. Measuring viability of neural crest cells is sufficient to establish maximum permissible concentrations of these chemicals.

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

#### **Original commentary** Correct answers A true Chemical 1 affects only migration, not viability of neural crest cells. This could be explained e.g. by chemical 1 inhibiting receptors involved in migration. B true Even if the migration is reduced with increasing concentrations of chemical 2, it is so with the same amplitude as the viability is reduced. The reduction in viability is enough to explain the reduction in migration (since dead cells do not migrate). C false Even if the viability in the presence of 100 to 500nM of chemical 3 is higher than the viability of untreated neural crest cells, this is due to the large standard deviation in the measurements, not to a beneficial effect of chemical 3. D false As for chemical 1, the response in cell viability and migration is not always similar. Since during embryonic development, neural crest cells need to migrate to create different tissues, it is important to take cell migration into account when establishing maximum permissible concentrations.

#### Animal anatomy and physiology

The contraction of a muscle fiber is triggered by an electric impulse that leads to the release of  $Ca^{2+}$  ions from the sarcoplasmic reticulum (SR) into the cytosol. After contraction, the  $Ca^{2+}$  ions are pumped back to the SR by sarcoplasmic reticulum  $Ca^{2+}$  ATPases (SERCA). To better understand the relative energy consumption of the actual contraction and the pumping of  $Ca^{2+}$  ions, skeletal muscle fibers of the frog species *Xenopus laevis* were treated with the chemicals BTS and cyanide. BTS inhibits cross-bridging between myosin and actin and cyanide inhibits the cytochrome c oxidase. The figure below shows the basal and peak cytosolic  $Ca^{2+}$  concentration as well as the pH of individual fibers during repeated contractions under natural conditions (open circles), in the presence of BTS (filled circles) and in the presence of both BTS and cyanide (red triangles).



## Indicate if each of the following statements is true or false.

A. The hydrolysis of ATP at both myosin heads and SERCA contributes to tiring of muscle fibers.

- B. In these fibers, aerobic respiration is an almost exclusive source of ATP after about 200 sec of repeated contractions.
- C. The phosphorylation of ADP from creatine phosphate releases H<sup>+</sup> ions.
- D. In the presence of cyanide only, the acidification of the cytosol over time was expected to be intermediate between the reduction observed under natural conditions and in the presence of both BTS and cyanide.
- A. True B. False C. False D. False

Original commentary Correct answers A true
This can be seen easily from the right figure where the pH decreases over time (due to anaerobic glycolysis) is substantial even when no ATP is hydrolyzed at actin heads (in the presence of BTS).
B <i>false</i> The decrease in pH over time is due to anaerobic glycolysis. Since the pH decreases well beyond 200 seconds, aerobic respiration may be contributing substantially to the production ATP, but is far from being the only important source. In the first 60 seconds, substantial sources of ATP are the stock of ATP and the phosphorylation of ADP from creatine
phosphate. C false
It actually absorbs $H^+$ ions (ADP + PCr + $H^+$ = ATP + Cr). This can easily be seen in the right figure since the pH is increasing at first. D false
In the absence of aerobic respiration, the drop in pH is expected to be even quicker than for the natural conditions.
References
Nogeira et al, AJPRICP (2013)

Walsh et al, Experimental Physiology (2008)



*Myasthenia gravis* is an autoimmune disease caused by autoantibodies that competitively bind and block postsynaptic nicotinic acetylcholine receptors on neuromuscular junctions of skeletal muscles.

## Indicate for each of the following statements if it is true or false.

- A. Reduced motility of the intestines is a likely symptom of this disease.
- B. Repetitive muscle contractions without relaxation (tetanic contractions) are a likely symptom of this disease.
- C. Drugs slowing down the degradation of acetylcholine in the synaptic clefts are likely to reduce symptoms.
- D. Drugs inhibiting cytotoxic T-cell proliferation are likely to reduce symptoms.

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

#### **Original commentary**

Correct answers

A false

The guts contain no skeletal but smooth muscles. The latter are usually not affected by Myastenia gravis due to the absence of neuromuscular junctions.

B false

The antibodies block the access of acetylcholine to the post-synaptic receptor (as mentioned in the stem), which cause a paralyzation or a reduced postsynaptic neuronal stimulation. C true

Slowing down the degradation of Acetylcholin (for instance by inhibiting the Acetylcholinesterase) allows it to act longer on the post synaptic receptors, which leads to a stronger signal since antibodies bind reversible on the ACh Receptors (they are in competition). D false

B-Lymphozytes not T-Lymphozytes produce antibodies.

#### Animal anatomy and physiology

In blood capillaries, fluid movement  $(J_\nu)$  across the capillary membrane, between the lumen of the capillary and the interstitial space, depends on the difference in hydrostatic and oncotic pressures between these two compartments. (Oncotic pressure is a form of osmotic pressure exerted by proteins.)

- $\bullet$  The following equation applies for  $J_{\nu}$
- $J_v = K_f x [(P_c P_i) \sigma(\pi_c \pi_i)]$
- with
- $P_c$ : the capillary hydrostatic pressure
- P<sub>i</sub>: the interstitial hydrostatic pressure
- $\pi_c$ : the capillary oncotic pressure
- $\pi_i$ : the interstitial oncotic pressure
- $K_{f}$ : the filtration coefficient
- $\sigma$ : the reflection coefficient

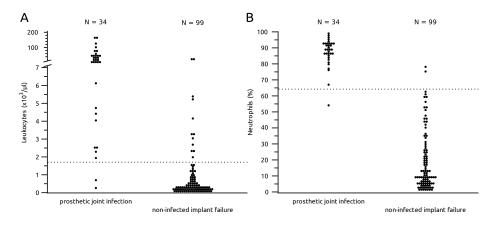
# Indicate for each of the following situations if the risk of edema (accumulation of fluid in the interstitial space) is increased.

- A. Tennis competition in the sun leading to dehydration
- B. Wearing anti thrombotic stockings
- C. Inflammation leading to increased blood vessel permeability
- D. Proteinuria (excessive protein excretion by the kidneys)
- A. False B. False C. True D. True

#### Original commentary Correct answers A false Excessive sweating causes a reduction of intravasal fluid which lowers the hydrostatic pressure in the capillaries. B false Anti thrombotic stockings increase the interstitial hydrostatic pressure and peripheral edema are decreased consecutively. C true Increased blood vessel permeability/leak -> plasma proteins move to the interstitium increasing the interstitial oncotic pressure -> more water movement to the interstitial space. D true Proteinuria reduces the concentration of proteins in the blood resulting in a reduced capillary oncotic pressure.

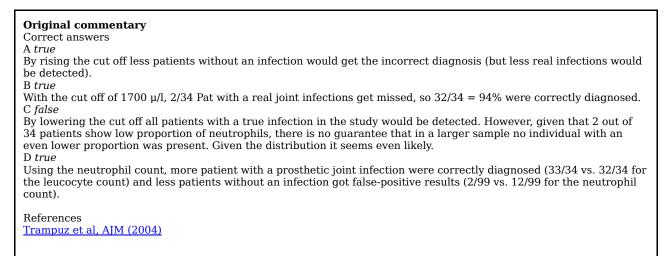
#### Animal anatomy and physiology

Proper treatment of chronic bacterial infections of artificial implants (e.g. prosthetic joints) requires expensive and demanding surgery. Unfortunately, such infections are often difficult to distinguish from non-infected implant failures. To propose new diagnostic tests, the absolute leucocyte count (A) and the fraction of neutrophils among white blood cells (B) were measured in 34 patients with true prosthetic joint infections and 99 patients with known non-infected implant failures. For each test, dotted lines indicate proposed cut off values to diagnose a true infection.



# Indicate if each of the following statements is true or false.

- A. Raising the cut off of absolute leucocyte counts to  $7000/\mu$ l would lower the risk of false positive results for a prosthetic joint infection.
- B. With the leucocyte cut off at 1500/ $\mu$ l, more than 90% of patients with a prosthetic joint infection are expected to be correctly diagnosed.
- C. Lowering the cut off of the fraction of neutrophils to 50% would prevent missing prosthetic joint infections in the future.
- D. These results suggest that the fraction of neutrophils is a better diagnostic test than leucocyte count.
- A. True B. True C. False D. True





While the glomerular filtration rate (GFR) is determined by blood pressure in the glomerulus, it does not directly reflect the systemic blood pressure. Instead, a stable GFR is maintained by either dilation or constriction of the afferent (flow in) and efferent (flow out) arterioles of glomeruli by an autoregulation mechanism. Several drugs interfere with this mechanism as side-effects. Those include non-steroidal anti-inflammatory drugs (NSAIDs) that reduce the capability of the afferent arterioles to dilate, as well as angiotensin-converting-enzyme inhibitors (ACEIs) that inhibit the production of Angiotensin II, and therefore reduce the capability of the efferent arterioles to constrict.

## Indicate for each of the following statements if it is true or false.

- A. NSAID intake reduces glomerular blood flow.
- B. ACEI intake reduces glomerular blood pressure.
- C. The effects of NSAID and ACEI on the GFR may compensate each other when taking both drugs together.
- D. While a chronic overproduction of aldosterone can be treated with ACEIs, using an Aldosterone antagonist affects the autoregulation mechanism less.

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

#### **Original commentary**

Correct answers A true

A side effect of NSAIDs is to inhibit dilation of the afferent arterioles. Hence the glomerulus is unable to increase blood flow through that mechanism.

B true

If the glomerular filtration pressure is not high enough, Renin is produced, which coverts the pre-hormone Angiotensinogen into Angiotensin I, which is then converted into Angiosin II by the Angiotensin-converting-enzyme. By inhibiting this enzyme, the ability to constrict the efferent arteriols is reduced (as mentioned in the stem). In addition, the production of Angiotensin II rises the blood pressure by rising the vascular pressure and rising the production of Aldosterone, which itself rises the renal reuptake of Natrium and water. Inhibiting this further decreases blood pressure system wide.

C false

While NSAID decrease the blood flow into the glomerulus, ACEI increase the outflow of blood. Hence both lead to a reduction on the glomerular pressure and lead to a reduction of the GFR.

D true

An Aldosteron overproduction can be treated by inhibiting the Angiotensin-converting-enzyme, as this leads to a lower level of Angiotensin II, which is a stimulant for the production of Aldosteron. However, due to the larger role of Angiotensin, a direct antagonist of Aldosteron implies a smaller effect, including the one described in this question.

References Campell Biology

#### Animal anatomy and physiology

The respiratory quotient (RQ) of an adult woman was measured at 0.7, along with an oxygen concentration of 170ml/l in her exhaled air. The RQ is the ratio between the eliminated amount of carbon dioxide and the absorbed amount of oxygen of the body. The metabolization of glucose and palmitic acid is as follows:

- Glucose: C<sub>6</sub> H<sub>12</sub>O<sub>6</sub> + 6 O<sub>2</sub>  $\rightarrow$  6 CO<sub>2</sub> + 6 H<sub>2</sub>O
- Palmitic acid:  $C_{16}H_{32}O_2 + 23 O_2 \rightarrow 16 CO_2 + 16 H_2O$

# Indicate for each of the following statements if it is true or false.

A. The woman adds about 119ml of  $CO_2$  to every liter of exhaled air.

- B. If the woman was metabolizing solely glucose, her RQ would be larger than when solely metabolising palmitic acid.
- C. The measurements are consistent with the woman solely metabolizing palmitic acid.
- D. If the woman was forced to sprint for a few minutes, her RQ is expected to decrease rapidly.

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

#### **Original commentary**

Correct answers A *false* 

The adult woman is exhaling 170ml of  $O_2$  per liter of air. Since the concentration of oxygen in the atmosphere is about 210 ml/l, the woman is absorbing around 40 ml of oxygen per liter of air. Given an RQ of 0.7, 28 ml of carbon dioxide (and not 119ml) must be eliminated per liter of exhaled air.

B true

An RQ of about 1 is expected when metabolizing glucose, but only 0.7 in the case of palimitc acid. Note that the students do not need to make the full calculations to answer this question, but just observe that there is an imbalance of  $O_2$  absorbed and  $CO_2$  eliminated when metabolizing palimitc acid, compared to a balance when metabolizing glucose.

C true

Metabolizing solely palmitic acid requires an absorption of 23  $O_2$  per 16  $CO_2$  eliminated (see equation 2). Hence the expected RQ is 16/23=0.69565, or 0.7 when rounded to the significant digits provided in the question. D *false* 

Short but heavy exercise leads to a large usage of anaerobic metabolism by muscle cells. At first, this is not expected to change the RQ at all as lactate fermentation does neither eliminate  $CO_2$  nor absorb  $O_2$ . However, through time, Lactate build up leads to an increase in RQ due to an inhibition of fatty acid metabolism. While the students are not required to know the latter, they should know that anaerobic metabolism cannot lead to a decrease in RQ as less  $O_2$  is absorbed.

#### Animal anatomy and physiology

Mammalian herbivores use different strategies to digest cellulose. Ruminants (e.g. cattle) use multiple stomachs, whereas monogastrics rely on an extended caecum or colon.

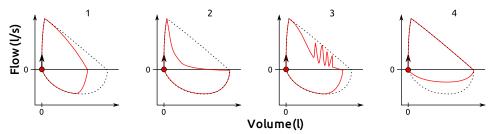
## Indicate if each of the following statements is true or false.

- A. The relative abundances of different amino acids in the small intestine of ruminants differ from the relative abundances of the swallowed food.
- B. Ruminants eat their faeces digested in the caecum to cover their nutritional need.
- C. In monogastric herbivores, the absorption of nutrients occurs primarily in the colon.
- D. The majority of the bacteria in the stomach of monogastric herbivores are able to produce cellulase.

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

Original commentary Correct answers
A true
Microorganisms metabolise anorganic nitrogen to build their own proteins which have a different amino-acid-pattern
than the food they swallowed. In the abomasum, microorganisms are killed by the HCL and their proteins are digested
from the ruminant.
B false
Not ruminants but monogastric herbivores like rabbits have to eat their faeces from the coecum.
C false
In most monogastric herbivores, it is still the small intestine where most nutrients are absorbed. That is why several monogastric herbivores eat their feaces. D <i>false</i>
Monogastric stomaches are not supposed to host bacterias.

To assess respiratory function, the flow and the volume of the exhaled air are measured during a forced expiration (positive flow) followed by a full inspiration (negative flow). Shown below are the measurements from four different patients with an airway problem. The black dotted line indicates normal respiratory function.



# Indicate if each of the following statements is true or false.

- A. The results of Patient 1 are expected if he had his left lung removed.
- B. The effect of the medication to reduce the symptoms of Patient 2 on the airways is similar to that of the parasympathetic nervous system.
- C. No diagnosis can be established for Patient 3 because of a cough attack.

D. Patient 4 is likely suffering from an airway obstruction in the thorax.

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

#### **Original commentary** Correct answers

A true

Restrictive lung diseases reduce the lung volume which is shown/documented in patients flow/volume graph #1. B false

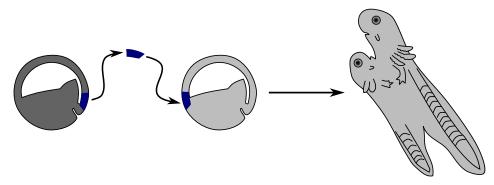
The patient suffers from a obstructive disease. He's volume is normal, but the flow is reduced. Sympathic effect on the bronchial system causes an dilatation, the parasympathic effect an obstruction. An adaequate drug has either to inactivate the parasympathetic nervous system or activate the sympathetic nervous system. C true

The results shown are indeed typical for a cough attack in which a rhythmic pulse of fast expiration are observed. D *false* 

As mammals have a negative pressure breathing (rise of intrathoracal volume causes a negative intrathoracal pressure leading to inspiration), an extrathoracal stenosis (obstruction) causes a fixed air-flow reduction apparent during inspiration.

The opposite is true for an intrathoracal obstruction, because the negative pressure during inspiration widens the intrathoracal air ways. Vice-versa in expiration.

The following figure shows an experiment in which a dorsal lip from a darkly pigmented donor embryo was transplanted to the ventral ectoderm of a lightly pigmented recipient embryo which was allowed to develop into a tadpole. The developing second body axis consisted mostly of non-pigmented cells.



## Indicate for each of the following statements if it is true or false.

A. The second body axis came solely from the transplanted dorsal lip.

- B. Transplanting the presumptive ectoderm to a host in the neurula stage would likely result in a tadpole with two body axes.
- C. Cell fate is predetermined and results from cell-intrinsic properties.
- D. If the presumptive endoderm was transplanted instead of the ectoderm, the secondary body axis would consist mostly of pigmented cells.
- A. False B. False C. False D. False

#### **Original commentary**

Correct answers A *false* 

The experiment shows, that the resulting tadpole on the ventral side had a lightly pigmented surface too, so this cells originate from the receiving embryo.

B false

A second body axis can only be induced when the cells are still able to differentiate in all tissues, this is not anymore the case at the neurula stage. C false

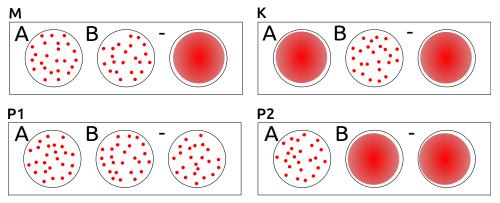
The grafted cells are able to induce neurulation in the receiving embryo where "cell fate" was not to neurulate. D false

The endoderm would not undergo neurulation and hence would not induce the cells to develop a secondary body axis at all.

Three blood groups have been characterized in cats, all of which are encoded by a single gene with three alleles, of which allele A is dominant over allele B, and allele AB is dominant over B, but recessive to A. Most cats with blood groups A or B have anti-B or anti-A antibodies, respectively. Cats with blood group AB do not produce either antibodies anti-A nor anti-B.

Antigens	Produced antibodies		
	Anti-A	Anti-B	Anti-AB
Α	-	+	-
В	+	-	-
AB	-	-	-

The figure below shows the results of blood transfusion compatibility tests performed for a mother cat (M), her kitten (K) and two potential father cats (P1 and P2). The cards consist of three circles that contain anti-A (A) and anti-B (B) antibodies, or no antibodies at all as a negative control (-). When adding a drop of blood to the circles, the occurrence of an agglutination reaction becomes visible (red dots).



## Indicate if each of the following statements is true or false.

- A. Mixing blood of kitten K with serum from P2 should lead to agglutination.
- B. M could receive erythrocytes from P2.
- C. A back-cross between mother M and kitten K might donate erythrocytes to P2.
- D. These results suggest that P1 is the more likely father of K than P2.
- A. True B. True C. False D. False

Original commentary Correct answers A true
P2 produces anti-B, which are present in its serum and would cause an agglutination of erythrocytes of kitten K. B true
The P2 serum has no anti-A and causes therefor no agglutination with antigens of erythrocytes in M. C false
A cat with the blood group A has anti-B, which react both with AB and B antigens on erythrocytes. Since the mother has genotype AB/B and the kitten B/B, any offspring of them has either blood group AB or B. D false
The negative control indicates that the test failed. So the genotype of P1 is unknown and hence these results do not suggest anything. Note, however, that if P1 had indeed blood group AB, he would have the same probability as P2 to

be the father of K. The corresponding probability for P2 is either 0% if he had genotype A/B or A/AB or 25% if he had genotype A/B.



The urea-to-creatinine ratio is used to assess renal function. It is calculated by dividing blood urea concentration by blood creatinine concentration. Urea and creatinine are both able to freely pass the glomerular filtration barrier. However, while creatinine is not reabsorbed, a percentage of urea is reabsorbed in the collecting ducts. An elevated rate of reabsorption is only observed when the total blood volume is increased.

# Indicate if each of the following statements is true or false. Compared to a healthy individual, the urea-to-creatinine ratio is expected to be higher...

A. ... in a patient suffering from an acute obstruction of the urethra (urinary retention).

B. ... in a patient suffering from an acute necrosis of the collecting duct epithelium.

C. ... in a patient suffering from dehydration.

D. ... in a healthy individual after intensive exercise but with sufficient water intake.

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

#### **Original commentary**

Correct answers A false

B false

Less urea reabsorption leads to a decrease in the ratio.

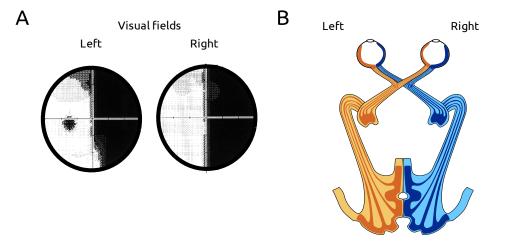
C true

Due to volume depletion, a higher percentage of urea is reabsorbed in the kidney, which leads to a larger ratio. D *false* 

In case of intensive exercise, muscles release more creatinine, and hence the ratio is decreased.

Urinary retention affects urea and creatinine equally, and hence does not lead to a change in the ratio.

Figure A illustrates the results of an examination of the visual field of a patient's left and right eyes, whereby dark areas indicate poor, and white areas good visual reception. The way sensory information is received by the eyes and transferred to the visual cortex is presented in figure B.



## Indicate if each of the following statements is true or false.

- A. After an injury of the left visual cortex, a patient would lose vision on its left side.
- B. A tumor of the hypophysis, situated below the optic chiasma, usually causes a loss of view of the lateral visual fields of both eyes.
- C. The visual field of the patient above is most likely caused by a problem between its optic chiasm and both retinae.
- D. Total visual loss from only one eye may be caused by a trauma of the eyeball or an inflammation of the optic nerve of the blind eye.

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

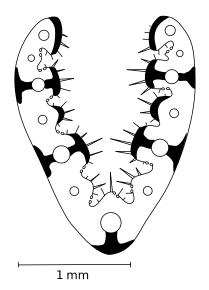
#### **Original commentary** Correct answers

A false It causes a loss of function of the retinal receptors on the left side of both eyes which causes a visual loss of the right side from patients view. B true The hypophysis is situated right below the optic chiasm. Tumors of the hypophysis consequently mainly affect those nerves crossing at the optic chiasm, which are the nerves innervating the retinal receptors an the medial part of the retina causing lateral visual field defects. C false a lesion between the optic chiasm and retinae would affect both eyes but would most likely cause blindness in both visual fields (right and left) of both eyes. A dysfunction of both visual pathways between the optic chiasm and the retinae is much more unlikely to be the reason for the indicated visual field than a single or multiple lesion(s) affecting both orange part of the optic nerve (and not affecting the blue ones at the same time). The visual fields given are typical for a lesion between the optic chiasm and the left visual cortex. D true A lesion affecting 100% visual field of only one eye is usually located between the optic chiasm and the retina. References

Duanes's Ophtalmology: E-book: Evaluation of Visual Function

#### Plant anatomy and physiology

The figure shows a schematic and representative cross section through a leaf of an angiosperm plant. Vascular bundles are represented by circles and sclerenchyma fibers by black surfaces. Additionally, the position of trichomes and stomata is indicated. The relative position of the vascular bundles is constant along the leaf.



## Indicate if each of the following statements is true or false.

- A. The leaf shown most likely represents a monocotyledon plant.
- B. The plant is most likely to be found in wetlands.
- C. The overall leaf shape is expected to be long and thin rather than oval or round.
- D. In a freshly cut section of this leaf the sclerenchyma fibers can be recognized by its intensely green color.
- A. True B. False C. True D. False

## **Original commentary**

Note The section belongs to a fescue (*Festuca*) out of the family of *Poaceae*.

Correct answers

A true

The regular position of vascular bundles that is constant all along the leaf indicates a parallel pattern of leaf veins without branching. This is a typical trait of monocotyledons. B *false* 

The stomata are situated in cavities of the leaf surface and protected by trichomes. This is a typical trait of drought adapted plants, reducing transpiration. C true

The amount of supporting tissue (sclerenchyma) within the thin leaf is a hint to a very long length. A relatively short (laceolate or round) leaf would not need such a tissue.

D false

 $\dot{Sc}$  lerenchyma contains dead cells without cytoplasma. Therefore they do not contain chlorophyll and appear colorless in a fresh section.

#### Plant anatomy and physiology

Plant organelles can be isolated from plant lysate by multiple rounds of centrifugation and washing. To distinguish between different organelles, centrifugation fractions are subjected to simple assays before and / or after incubation for 30 minutes under specific conditions. Possible assays include:

- 1) measuring the concentration of glucose and other aldoses using a Fehling reaction,
- 2) detecting the presence of DNA by measuring absorption at 260 nm,
- 3) observing gas bubbles.

## Indicate for each of the following statements if it is true or false.

- A. A Fehling assay before and after incubating in light distinguishes fractions of chloroplasts from those of amyloplasts.
- B. A Fehling assay before and after incubating with glucose distinguishes fractions of Golgi from those of mitochondria.
- C. Fractions of endoplasmic reticulum and nuclei can be distinguished by incubating with lipases and proteases, followed by centrifugation and measuring absorbance at 260 nm in both supernatants.
- D. The presence or absence of gas bubbles after incubating with  $H_2O_2$  distinguishes fractions of peroxisomes from those of endosomes.

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

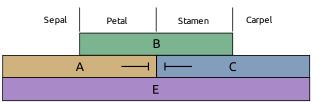
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Original commentary
Correct answers
A true
Amyloplasts convert glucose in starch that does not react with the Fehling reagent, whereas in light, chloroplasts
produce sugars that will turn the Fehling solution blue.
B false
Neither Golgi nor Mitochondria are involved in the central sugar metabolism.
C true
Nuclei treated with lipases and proteases will liberate DNA that stays in the supernatant after pelleting the nuclei
debris. Endoplasmic reticulum does not contain DNA, therefore even if lipases break it down, no change in absorption
at 260nm will be observed.
D true
Peroxysomes transform H<sub>2</sub>O<sub>2</sub> in H<sub>2</sub>O and O<sub>2</sub>, thereby liberating O<sub>2</sub> bubbles, whereas endosomes do not.
References
Lang, Plant Cell Rep (2011)
```

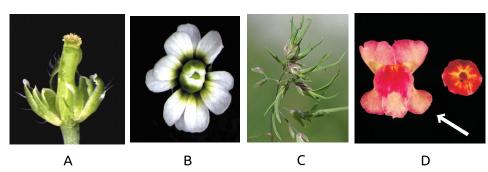
Peroxisome Database



#### Plant anatomy and physiology

According to the ABCE-Model of flower development, activity of genes from different classes A, B, C or E determines the identity of floral parts. Expression of class A genes is needed to determine future sepals and petals, class B genes to determine future petals and stamen and class C genes to determine future stamen and carpels. A and C genes inhibit each other's expression. Differentiation of each floral part additionally requires activity of class E genes. The figure illustrates the ABCE-model and shows flower samples of *Arabidopsis* (A and B), the alpine grass *Poa alpina* (C) and two flowers of the snapdragon *Antirrhinum majus* (D; the arrow indicating the bilateral wildtype, while the radial symmetric to the right is a mutant).





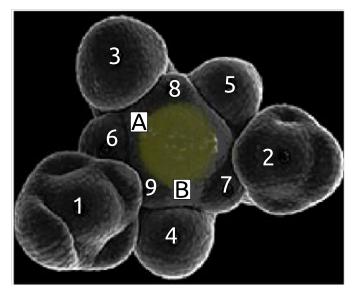
## Indicate if each of the following statements is true or false.

- A. The phenotype of *Arabidopsis* A is best explained by a loss of function of class B genes.
- B. The phenotype of *Arabidopsis* B is best explained by a loss of function of class A and C genes.
- C. The phenotype of Grass C is best explained by a loss of function of class E genes.
- D. The symmetry of the mutant flower of Snapdragon D is best explained by a loss of function of class C genes.
- A. True B. False C. False D. False

**Original commentary** Correct answers A true B false The best explanation is the loss of only class C genes. C false The leaves emerging from the flowers represent grass seedlings and not altered flowers with leaves at the position of floral parts. The picture shows a viviparous plant with grains germinating before falling off the mother plant. D false The mutant shows a loss of bilateral symmetry and not an alteration in the differentiation of floral parts. References Krizek, Nature (2005)

#### Plant anatomy and physiology

The pattern of leaf primordia (future leaves) at the apical meristem is determined by active auxin transport. Auxin is transported towards the meristem tip. Young primordia act as auxin sinks through the auxin efflux carriers PIN1 and thus decrease the auxin level in nearby meristem cells. A new primordium will be induced at the place with the highest remaining auxin level. The image below shows the meristem of *Arabidopsis* with the primordia 1-9, with 1 being the oldest.



# Indicate if each of the following statements is true or false.

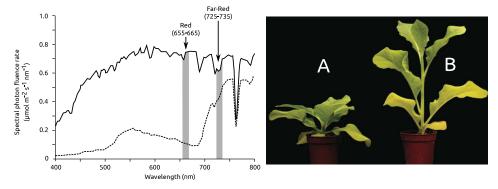
- A. Assuming PIN1 was only present in the two youngest primordia, its activity must be different in them.
- B. The next primordium will emerge at position A.
- C. If PIN 1 is inhibited at the stage shown in the figure, the next primordium is most likely to emerge at position B.
- D. In a mutant where only the youngest primordium is acting as an auxin sink, leaves will grow opposite to each other.
- A. True B. False C. False D. True

Original commentary Correct answers A true
The characteristic angle of 137° between two subsequent primordia can be only explained with the youngest primordium being a stronger auxin sink than the second youngest.
B false According to the order of older primordia the next primordium will emerge between primordium 2 & 5. C false
In this mutant the primordial pattern would be aberrant, as well as the leaf shape. But it would emerge at a random position. There is no reason to assume that it will be position B.
D <i>true</i> In this case the highest auxin concentration would be present directly opposite of the youngest primordium.
References <u>Reinhard, Nature (2003)</u>



#### Plant anatomy and physiology

Many plants use, among other means, the ratio of red/far-red light to detect other plants competing for light and react with adaptive growth to avoid shade. The ratio of red to far-red light is detected by the photoreceptor phytochrome. Phytochrome is converted between two forms  $P_X$  and  $P_Y$ , depending on the wavelength of the photon it has absorbed. The ratio between both forms reflects the red/far-red ratio in the environment. A high proportion of  $P_X$  mediates the expression of genes responsible for shade-avoiding growth. The figure shows the spectra of normal daylight (solid line) and daylight filtered through a tobacco canopy (dashed line). Tobacco plant A has been grown under normal sunlight, plant B under a canopy of older plants.



## Indicate if each of the following statements is true of false.

- A. The genes leading to the shade-avoiding phenotype are expressed when the red/far-red ratio is between 3 : 1 and 4 : 1.
- B. A high proportion of red light increases the proportion of  $P_X$  to  $P_Y$ .
- C. Upward orientation of leaves is a typical feature for shade-tolerant species.
- D. The shade-avoiding phenotype includes activation of lateral meristems.

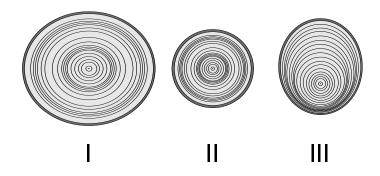
A. False B. False C. False D. False

# Original commentary Correct answers A false The maximal ratio is observed in normal light and is slightly above 1. B false The opposite is true. Plant B has the genes activated with a red/far-red ration of about 0.5. As the far-red-converted phytochrome form is dominant, this must be the active form. C false Shade-tolerant plants do not need to express the plant B-phenotype of growing out of the shade. In the shade they keep the normal leaf exposition, allowing to maximize the amount of photons collected by the leaves. D false A plant trying to avoid shade by enhanced growth will invest in few elongated stems and not in a multitude of branches that stay in the shade. Plant B doen't show any branching. References

Vandenbussche, Current Opinion in Plant Biology (2005)

#### Plant anatomy and physiology

In the wood of trees of temperate climates, annual growth rings are present and reflect different growth conditions between years and individuals. Three coniferous trees of the same species were cut at the same stem height in the same year. The stem cuts in the figure are drawn to the same scale.



# Based on these stem sections, indicate for each of the following statements if it is true or false.

- A. Trees I and II likely grew in the same region, whereas Tree III likely grew in a more distant region.
- B. Tree III is likely to have experienced more climate variation between years than Tree I.
- C. Trees I and II may originate from the same forest.
- D. The asymmetric pattern of Tree III may be due to constant exposure to strong wind beginning roughly ten years ago.
- A. True B. False C. True D. True

#### **Original commentary** Correct answers

A true

I and II show the same pattern of two periods of narrow rings (bad growth conditions) 2-6 and 9-13 years before they have been cut. Thus they have been growing in the same climatic conditions present in one same region. Tree III shows a different, much more regular pattern and did not suffer those two periods of bad years. B *false* 

Tree III has relatively regular rings throughout the lifetime on one side of the section. The asymmetric pattern can be explained by very local effects like a physical obstacle or shadow on one side of the tree. *C true* 

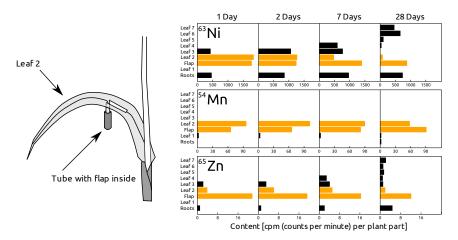
Local ecological factors such as available light or soil conditions can result in very different growth rates even for neighboring trees.

D true

The first rings are regular, indicating an equilibrated light supply. The last rings are more and more asymmetric. A likely explanation is that faster growing trees are competing for sunlight on one side of the tree, whereas on the other side the tree is still sufficiently exposed to sunlight.

#### Plant anatomy and physiology

The second leaf (Leaf 2) of a young and growing plant of wheat (*Triticum aestivum*) was fed via a rectangular flap cut symmetrically in the middle of the lamina and brought into a tube with a feeding solution containing radioactive nickel ( $^{63}$ Ni), manganese ( $^{54}$ Mn) and zinc ( $^{65}$ Zn). After 1, 2, 7 and 28 days, the contents of radioactive elements were measured in different parts of the plant. Concentrations measured in the flap and Leaf 2 are shown in orange in the figure.



# Indicate if each of the following statements is in agreement with the results shown above.

A. The plants absorbed the entire feeding solution provided before the first measurement.

B. Nickel is mostly transported to growing organs.

C. Manganese has higher phloem mobility than zinc or nickel.

D. Leaf 2 is turning into a net sugar exporter after Day 1.

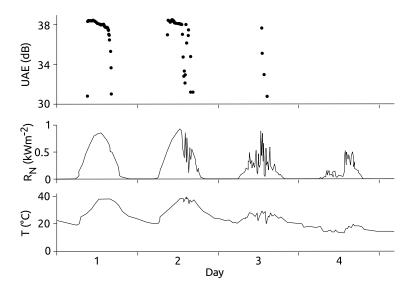
A. True B. True C. False D. False

#### **Original commentary** Correct answers A true B true Nickel is first accumulated in leaf 3, then in leaf 4. After several days when these leaves are grown up nickel is exported again towards leaves 5-7. C false Manganese (known to have low phloem mobility) stays in the second leaf, whereas Zn and Ni is reduced in leaf 2 and appears in significant amounts in other parts of the plant. D false Leaf two is an "adult" leaf already at the beginning of the experiment. At day one a fraction of nickel already has been transferred from leaf 2 to 3. As the transfer is done by phloem transport, leaf 2 must already have a net sugar export. References Riesen, Journal of Plant Nutrition (2003) Plant Physiology 5th ed., Lincoln Taiz and Eduardo Zeiger, section "Essential nutrients, deficiencies, and plant disorders" page 108 and following.

Campbell, Biology (9th ed.), table 37.1 page 791.

#### Plant anatomy and physiology

In xylem, water conduits occasionally undergo embolism, characterized by an inflow of air into the conduit lumen, followed by collapse of the water column. Each event of embolism emits a sound called ultrasonic acoustic emission (UAE) that can be detected with adequate sensors. The figure below shows such measurements of an oak tree (*Quercus pubescens*) during four subsequent summer days. Radiation of sunlight ( $R_N$ ) and air temperature (T) were measured simultaneously.



## Indicate if each of the following statements is true or false.

- A. Embolisms occur during periods of elevated water tension in xylem conduits.
- B. The sound intensity of a single UAE event is correlated with the difference in water potential between the air and the leaves.
- C. The decrease in UAE events over time is likely due to an increase in cloud cover.
- D. Embolisms reduce the conductivity of xylem and therefore the water supply of distal tissues.

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

#### Original commentary

# Correct answers A true

The UAEs are recorded during periods of high radiation and temperatures. During this time the water potential in the surrounding air is very low, leading to increased evaporation. During increased evaporation the difference in water potential between leafs and roots is big and therefore the tension forces within xylem is high. B *true* 

The audibility is indicated by the decibel value. At mornings and evenings the few measured UAEs have lower decibel values and are therefore less loud. Keep in mind that the decibel scale is logarithmic. The difference between the water potential of air and leaves depends on radiation and temperature and is maximal at noon and early afternoon but lesser at morning and evening. Therefore a relation is given. *C true* 

The radiation diagram shows how the cloud covers increases (huge variation within short time), leading to a decrease in overall radiation, and hence to a decrease in water tension within water conduits. D true

Water transport depends on a permanent water clomn. An embolism is leading to the interruption of a xylem conduit. Until it is refilled, further water transport through this conduit will be null or very limited.

References Zweifel et al, New Phytologist (2008)

#### **Genetics and evolution**

In a plant species, the level of anthocyanin pigments produced is controlled by a single gene G, for which only a "dark" and a "light" allele are present. To more accurately map the position of gene G on chromosome 3, two inbred lines (P1 and P2) are crossed and F2 individuals (X1 through X5) are genotyped at five single nucleotide variant loci (SNV1 through SNV5) on the same chromosome.

	SNV1	SNV2	SNV3	SNV4	SNV5	Anthocyanin [mM]
P1	A/A	C/C	A/A	т/т	C/C	120
P2	т/т	т/т	C/C	G/G	G/G	25
X1	A/T	с/т	A/C	T/G	C/G	115
X2	A/A	C/C	A/A	T/G	C/G	123
Х3	A/A	т/т	C/C	T/G	C/G	22
X4	A/A	с/т	C/C	T/G	C/C	29
X5	A/T	с/т	A/A	т/т	C/C	118

#### Based on these results, indicate if each of the following statements is true or false.

A. One recombination event happened in each parent of X2 between the genotyped loci.

B. F1 individuals are likely showing intermediate levels of anthocyanin.

C. Among the studied loci, SNV3 is closest to gene G.

D. The phenotype ratio in the progeny of a cross between X4 and X5 is 2:1.

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

#### **Original commentary**

Correct answers A false

Most likely, a single recombination event happened in only one of the parents, as one of the haplotype is parental (ACATC) and the other shows a recombination between SNV3 and SNV4.

B false

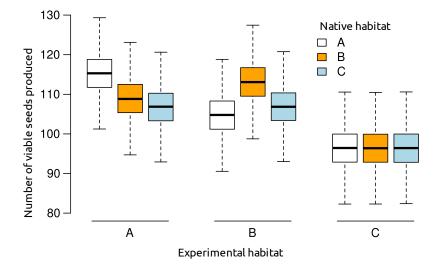
Gene G is linked to SNV3, and the table lists a heterozygous individual (X1) with an elevated anthocyanin level (which is the dominant allele). But note that the students do not need to identify the most closely linked locus as for each of them heterozygous individuals are given and the conclusion would remain unchanged. *C true* 

This is the only locus for which the genotypes match the phenotypes in a Mendelian fashion.

D false

A 2:1 ratio is impossible for any crossing, as we clearly have dominant-recessive inheritance. So it is possible to answer this question even if the truly linked locus is not identified.

A plant species grows in three different habitats A, B and C, and populations from different habitats also differ genetically. To test if some of these genetic differences are driven by local adaptation, a so-called reciprocal transplant experiment is carried out where seeds collected from different habitats are grown in all three habitats. After a year, the fitness of the plants are measured by counting the number of viable seeds produced per individual.



# Based on these results, indicate if each of the following statements is true or false.

A. Plants from Habitat A are locally adapted to their native environment.

- B. Plants from Habitat B are locally adapted to their native environment.
- C. Plants from Habitat C are locally adapted to their native environment.
- D. Habitat C is less suitable for these plant species than habitats A or B.

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

#### **Original commentary**

Correct answers A true

Plants grown from seeds collected in habitat A show, on average, a much higher fitness when grown in habitat A than seeds collected form other habitats.

B true

Plants grown from seeds collected in habitat B show, on average, a much higher fitness when grown in habitat B than seeds collected form other habitats. Note that the fact that seeds from habitat B perform better in habitat A than in habitat B is likely due generally better conditions in habitat A.

C false

Plants grown from seeds collected in habitat C follow the habitat quality, but do not indicate that they are specifically adapted to environment C since they do not perform better there than plants adapted to different environments. D *true* 

All plants perform much worse in environment C than environments A or B, inclusing the individuals originating from this environment.



Consider a large and constant population of a diploid organism with non-overlapping generations and sexual reproduction happening in spring. In addition, there is no difference in allele frequencies between sexes and there is no migration or natural selection acting. In such a population, the frequencies of alleles fluctuate at a given rate between generations due to the random nature of reproduction.

# Indicate if each of the following statements is true or false. Allele frequencies are expected to fluctuate at ...

A. ... a higher rate if the population was growing exponentially.

B. ... a lower rate if all individuals had the same number of offspring.

C. ... a similar rate even if there was strong inbreeding.

D. ... a higher rate if the population crashed every winter.

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

### **Original commentary**

Correct answers

A false

If a population is growing exponentially, stochasticity in reproduction is reduced due to a larger number of offspring "sampling" alleles from the parent generation, and hence allle frequencies fluctuate at a lower rate. B *true* 

If all individuals had the same number of offspring, allele frequencies are almost constant since each individual leaves a fixed number of copies of his alleles in the population. The only stochastic variation left comes from heterozygous individuals randomly passing one of their alleles per offspring.

C true

While inbreeding decreases the frequency of heterozygotes, it does not lead to a faster change in allele frequencies. Dtrue

A population crash in winter leads to a recurrent bottleneck which removes alleles randomly from the population and leaves a smaller number of individuals reproducing in spring. This leads to an increased stochasticity.

The effect of various mutations in a gene x coding for a protein X, essential for the synthesis of leucine, is studied in a haploid yeast. The beginning and the end of the complete sequence of the coding strand of x is given below.

# Indicate if each of the following statements is true or false.

- A. Cells with a C  $\rightarrow$  T mutation at position 13 produce shorter mRNA of x.
- B. Cells with a A  $\rightarrow$  T mutation at position 16 are able to grow on a medium lacking leucine.
- C. Cells with a T  $\rightarrow$  A mutation at position 31 only produce an alternative protein X\* missing the first 10 amino acids.
- D. Cells with an additional G between positions 33 and 34 produce functional X.

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

#### **Original commentary**

Note

Students are expected to know the start and stop codons. Nevertheless, for those who don't, they can find out by looking at the sequence, that ATG is a start codon and TAG a stop codon.

Correct answers

A false

Introducing the mutation C13T results in the TAG stop codon, but the transcription stops only at the transcription termination sequence independently of the codons  $\frac{1}{2}$ 

B false

Introducing the mutation A16T results in the TAG stop codon. With a stop codon at the beginning of the sequence, no functional protein is produced and no leucine can be synthesized by the yeast that would need to take it up from the medium.

C false

Introducing the mutation T31A results in the ATG start and methionine codon. This will simply result in a methionine at this position. The eukaryotic ribosome binds at the 5'-cap of the mRNA and starts translation at the first AUG encountered. Further AUG are simply translated into methionine.

D false

Introducing a G between positions 33 and 34 results in a shift of the translation frame. Instead of reading ...GAT AGC... (positions 34-39), the ribosome will read ...GGA TAG... and reaches a stop codon. Translation stops there, the protein will not be functional.



**34** 

An operon encoding enzymes 1 and 2 is regulated by metabolite X and consists of four sequences A, B, C and D of unknown function. To elucidate their function, the effect of mutations in the sequences A-D on the synthesis of the enzymes is assessed in the presence and absence of X.

	Хрг	esent	X absent		
Mutation in sequence	Enzyme 1	Enzyme 2	Enzyme 1	Enzyme 2	
no mutation	-	-	+	+	
А	-	-	-	+	
В	+	+	+	+	
С	-	-	+	-	
D	-	-	-	-	

# Indicate if each of the following statements is true or false.

A. Enzymes 1 and 2 are likely part of the synthetic pathway for X.

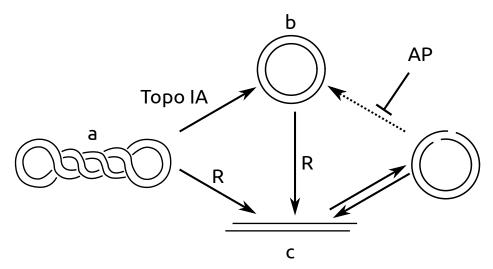
- B. The sequence A codes for enzyme 2.
- C. The sequence B is the promoter region.
- D. The sequence D is the regulatory gene.

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

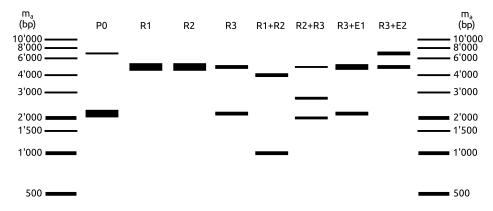
Original commentary Correct answers A *true* This operon is repressed by X. This kind of negative transcriptional regulation is mostly found in biosynthetic pathways. B *false* In the absence of X and mutation in A, enzyme 2 is still produced, enzyme 1 isn't, therefore A codes for enzyme 1. C *false* In the case of a mutation in B, enzymes 1 and 2 are produced even in the absence of X, therefore B corresponds to the regulatory gene. D *false* In the case of a mutation in D, neither enzyme is produced, even in the absence of X, therefore D corresponds to the promoter region.

## **Genetics and evolution**

In cells, most plasmids are supercoiled (a in figure below). While such plasmids can be uncoiled to relaxed circles (b) using Topoisomerase IA (TopoIA), using restriction enzymes (R) linearizes plasmids through cutting (c). A linearized plasmid may spontaneously self-anneal and subsequently be ligated to form a relaxed circle. The ligation reaction can be inhibited by the addition of phosphatase (AP).



In an experiment, a plasmid was treated with different restriction enzymes (R1, R2, R3) under similar conditions and separated on an agarose gel together with an untreated sample (P0) and a marker consisting of linear DNA pieces. Topoisomerase IA and AP treatments in combination with R3 were also analyzed, but the tubes were mixed up (R3+E1 and R3+E2 on gel).



# Indicate if each of the following statements is true or false.

A. On this plasmid, the restriction site for R2 is closer to the one for R3 than to the one for R1.

- B. The plasmid is about 5000 bp long.
- C. R1 cuts more efficiently than R3.
- D. E1 represents AP, E2 represents Topo IA.
- A. False B. True C. True D. True

```
Original commentary
Correct answers
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A false

R1 and R2 are distant from 1000 bp, R2 and R3 from 2000 bp, even if 2 vector maps are possible: R1 - 1000 bp - R2 - 2000 bp - R3 - 2000 bp -R1 - 1000 bp - R2 - 3000 bp - R3 - 1000 bp -

B true

Since the shape of DNA plays a role in the migration, the length of DNA can only be determined by comparing fragments with the same shape. The plasmid is linearized when using only 1 restriction enzyme. The supercoiled plasmid from P0 (thick band) is converted to linearized DNA which migrates around 5000 bp (see R1 or R2). C true

In R3, some uncut supercoiled is still present, only about half was cut, whereas in the same time, R1 could cut everything

D true

Treatment with AP removes prevents re-formation of relaxed circles, the slowest migrating band disappeared in R3+E1. Treatment with E2 converted the uncut supercoiled plasmid into relaxed circles, this corresponds to Topo IA.

36

A child affected by a rare genetic disease is born to two healthy parents. The child has a healthy sister.

# Indicate if each of the following statements is true or false.

- A. If the disease was known to be segregating in the family of the father, the disease is more likely autosomal than sex-linked recessive.
- B. If the disease was sex-linked recessive, the probability of the sister being a carrier is 50%.
- C. If the disease was autosomal recessive, the probability of the sister being a carrier is 50%.
- D. If the disease was not known to be segregating in either family, the causal mutation is either autosomal recessive or, within the family, unique to the child.

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

**Original commentary** 

Correct answers

A true

X-linked recessive would imply that the father was affected.

B true

In this case, the mother is carrier and the father healthy. Hence the sister inherits an unaffected X chromosome from the father and has a 50% chance to inherit the affected chromosome from the mother.

C false

In this case, both parents are carriers. But since we know that the sister is healthy, the probability is 2/3 (and not 1/2). D true

A rare autosomal recessive disease is likely not to show up in an outbred family for many generations. An alternative explanation for the disease, however, might indeed be a spontaneous mutation being unique to the child.



In a small pasture, 500 individuals of two closely related snail species were sampled. Genetic analyses detected a locus at which none of the individuals were found to be heterozygous, despite the presence of two alleles in each species. The two species and all genotypes were randomly distributed in the homogenous habitat.

Snail species	Genotype	Number of snails
A. sulfis	AA	126
A. sulfis	BB	125
A. andea	СС	122
A. andea	DD	127

# Indicate if each of the following statements is a likely explanations of the observed pattern.

A. These snails generally self-fertilize.

B. Both species experience strong genetic drift due to low population sizes.

C. These snails reproduce hermaphroditically.

D. These snails mate preferably with individuals of the same genotype.

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

## **Original commentary**

Correct answers

A true

Self-fertilization would indeed lead to all snails being homozygous.

B false

If the populations were experiencing strong drift through low population sizes, this would lead to a reduction in diversity. However, there is no reason why this should affect heterozygous individuals preferentially. C *false* 

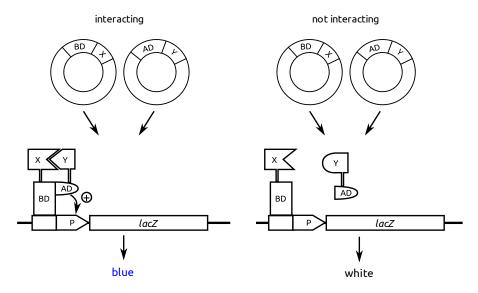
Hermaphroditic reproduction does not have any impact on allele frequencies as long as mating is random.

D true

If snails mate only with snails of the same genotype, heterozygous individuals would be very rare in the population.



A yeast-two-hybrid assay (Y2H) allows to test if a protein X interacts with another protein Y. In this assay, the gene coding for X is fused to the gene of a DNA-binding domain (BD) of a transcription factor T. The gene coding for Y is fused to the gene of the activating domain (AD) of T. The resulting plasmids are transformed into a yeast strain containing the *lacZ* gene under the control of a promoter P, which is specifically recognized by BD. Plated on agar with X-gal, the colonies turn blue if they are expressing LacZ, which is only the case if BD and AD are in close proximity and hence if X and Y interact with each other.



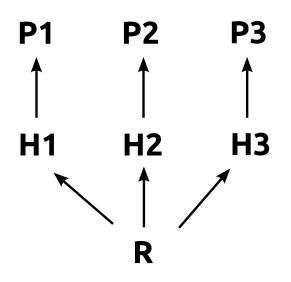
# Indicate if each of the following statements is true of false.

A. Y2H also works if BD alone is sufficient to activate transcription.

- B. Y2H allows studying interactions between integral plasma membrane proteins.
- C. Y2H can give false-positive results if X and Y interact indirectly via a third protein.
- D. Y2H can give false-negative results if the binding site for Y on X is situated close to the terminus at which BD is attached.
- A. False B. False C. True D. True

#### Original commentary Correct answers A *false* The assay works correctly only if BD and AD by themselves are not enough to activate transcription, but are both needed. B *false* This in vivo assay works only for soluble proteins. To study interaction of membrane proteins, the membrane proteins need to be properly folded, which is only possible if they are in lipidic environment. C *true* If I binds to a third protein which binds to II, the assay can give positive results, even if I and II do not interact directly. D *true* The binding site can be distorted if it is too close to the junction to the transcription factor domain.

Consider a currently stable system in which three predatory insect species P1, P2 and P3, feed exclusively on three herbivore insect species H1, H2 or H3, respectively, all of which feed on the same limited plant resource R.



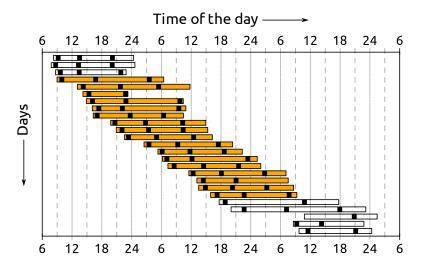
# Indicate if each of the following statements is true or false.

- A. If the plant resource was doubled, the abundance of P2 will increase.
- B. If H1 is a stronger competitor than H2, removing P1 leads to a increase of P2.
- C. If H3 was severely limited by its predator, the removal of P3 would lead a decrease in the abundance of P2.
- D. The introduction of a top-predator feeding on P1 and P2 is likely to increase the abundance of P3.
- A. True B. False C. True D. False

Original commentary Correct answers
A <i>true</i> Doubling the resource should lead to a doubling of all abundances in the system in the long term since the interactions remain unchanged.
B <i>false</i> Population size of predators depend on populations size of herbivores. H1 now lacking a predator will increases on cost of H2. As a consequence the abundance of P2 decreases as well. This is expected unless H2 is limited almost completely by its predator and not by competition with H1, but even in that case the abundance is expected to remain
unchanged, but not increase. C <i>true</i> Population size of predators depend on populations size of herbivores. H3 when having its predator removed, is able to
reduce H2, P3 must be a limiting factor for H3. H2 when having its predator removed does not seem to be able to affect H3. Therefore the effect of the predator is not limiting enough to lead to significant loss of competition against H3.
D <i>false</i> The top-predator will decrease the abundance of P1 and P2 and therefore lead to an increase of H1 and H2. As both those herbivores are in competition with H3, the abundance of H3 is likely to decrease, leading to a dcrease in the abundance of P3.



The following figure illustrates the result of an experiment during which a person was alone in a room and was allowed to freely choose the awake and sleep periods by turning a bright light on and off. The consecutive time of light for each day is shown as a rectangle with times at which the person chose to eat a meal indicated by black bars. While the person had no time cues from the outside world during the days shown in orange, the room was exposed to natural light during the days shown in white.



# Indicate if each of the following statements is true or false.

A. Without external cues, the person chose increasingly longer periods of light.

- B. The endogenous clock of this person cycles on a 28.5 h rhythm.
- C. These results are in agreement with bright light being a cue to delay the sleeping phase.
- D. These results suggest that the endogenous clock of this person can readjust completely within two days.
- A. False B. False C. True D. False

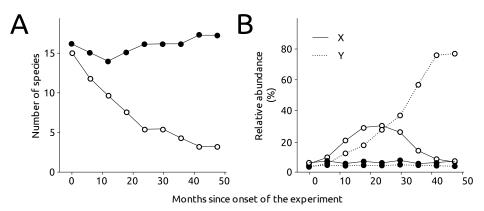
## **Original commentary**

Correct answers A false While the periods of light were extended in the absence of external cues, there us no indication the those periods got longer during the experiment. B false Firstly, this experiment does not measure the endogenous clock since the bright light is an effective, external cue to delay the endogenous clock. Secondly, the the observed rhythm is on the order of about 26 hours (average over the 17 days). C true The person chose extended periods of light, without extending the periods of sleep much. This is an indication that the bright light resets the clock and delays the desire to go to sleep. A good example can seen at the beginning of the experiment where the person stay up for more than 20 hours on the first two days and apparently got very tired on the third day. D false The transition back to normal conditions took the person at least four days. This can be seen best with the irregular eating times. References Charles et al, Science (1999)



A herbivorous insect H is known to exclusively feed on the seedlings of two tree species X and Y. In an experiment, patches of a forest were subjected to a treatment, or not. In the treatment, seedlings of X and Y were protected from being fed on by H (open circles). Patches in which X and Y were unprotected served as a control (filled circles). Panel A shows the average number of all species for which seedlings were found in the patches. Panel B shows the average relative abundance of seedlings of X and Y observed in the patches.

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



A. Seedlings of species Y are weak competitors.

B. The regulation of this tree community involves a top-down process.

C. Seedlings of species X are strongly regulated by an additional herbivore.

D. The herbivore insect functions as a key stone species.

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

#### **Original commentary** Note

The two prey species must be excellent competitors, not inferior ones; otherwise they would not be able to dominate the experimental plot communities in the absence of the predator (78% vs. 27%). When community organization is regulated in this way, as opposed to the availability of nutrients — recall seedling growth was not limited by light availability — it follows a "top-down model" and not a "bottom-up model" (p. 1206). A top-down regulatory role for the predator is predicated on there being interspecific competition between prey and non-prey species for space when the prey are largely sessile, which plants are (p.1205 top of Fig. 54.17). In this way, the predator pre-empts interspecific competition among seedlings to promote local alpha diversity in the forest.

#### Correct answers

A false

Competition of species Y is only controlled by herbivore H. Without this control it dominates the tree seedlings in the forest within a few years. Moreover it is clearly able to outcompete species X when both species are lacking herbivore H.

The diversity of tree species is regulated by the presence of herbivore H, a species at a higher position in the food chain. This feature qualifies a top-down process. C false

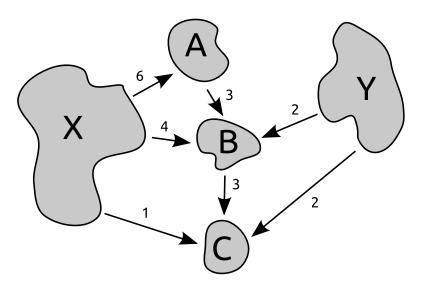
After removing herbivore H, species X increases rapidly over two years, before diminishing again when being exceeded by species Y. This feature is best explained by competition between species X and Y. A potential regulation by another herbivore would be likely to prevent the initial strong increase of species Y. D true

The predator not very abundant in the community, as both its host species only have a cumulative abundance of about 10%, but it clearly plays a pivotal ecological role in structuring the community by preventing potential dominance by prey, so it therefore qualifies as a "keystone species" (p. 1204; Campbell Biology 9th ed., 2010)

B true

## Ecology, ethology and systematics

The meta-population concept describes the population size in a habitat patch as a function of birth rate, mortality and migration. Consider the hypothetical meta-population given below, consisting of two big source patches X and Y where the birth rate exceeds mortality and three sink patches (A, B and C) where reproduction does not occur. Yearly net migration (individuals) between different sub-populations is constant and indicated with arrows in the figure. In each sink patch, 6 individuals die at the end of each migration season. Individuals cannot migrate further than to the next patch within a year. The initial population sizes in the sink patches are A=22, B=9 and C=12.



# Indicate if each of the following statements is true or false.

- A. The subpopulation in Patch A dies out for the first time after 8 years.
- B. Subpopulations X and Y will be genetically isolated within few years.
- C. If in Patch C, 50% of the individuals (instead of 6 individuals) die each year, the Subpopulation C will not decrease below 7 individuals.
- D. A conservation measure to reduce mortality in A by 50% (3 individuals a year) is sufficient to preserve all subpopulations.
- A. True B. True C. False D. True

A true

Net migration for patch A is 3. With a mortality of 6 individuals a year the sub-population declines 3 individuals each year. After 8 years, the population will reach 0.

B true

With the given migration and mortality, all sink population have a negative long-term trend and will die out. First, population A will die out so migration between A and B will be interrupted. Without this migration, population B and subsequently population C will die out as well. So no individual will be able any more to migrate between X and Y. C *false* 

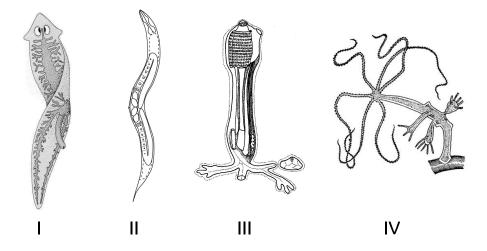
With a constant net migration of 6 individuals into patch C, the population will fall below 7 individuals within three years, but will not fall below 6 individuals.

D true

This reduction in mortality prevents A from dying out. Therefore all other subpopulations will not die out either.

**Original commentary** Correct answers

You are given four drawings referring to typical representatives of four major groups of metazoa.



# Indicate if each of the following statements is true or false.

- A. Organism I belongs to a taxon characterized by a digestive system with a single opening and the lack of a specialized respiratory system.
- B. Organism II belongs to a taxon characterized by a hydrostatic skeleton, a thick cuticle, a tubular digestive system with openings at both ends and often a genetically fixed number of somatic cells.
- C. Organism III belongs to a taxon characterized by a larval stage with nerves forming a neural tube and a sessile adult stage with a body surrounded by a tunic.
- D. Organism IV belongs to a taxon characterized by an endoskeleton with calcareous pads and a vascular system of fluid-filled canals used for various functions, including locomotion by tubular feet.
- A. True B. True C. True D. False

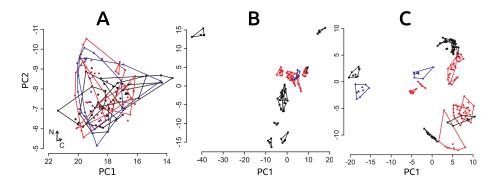
Original commentary Correct answers A *true* Both the drawing and the description are referring to the group of *Plathelminthes* (*Tubularia*). B *true* Both the drawing and the description are referring to the group of Nematoda. C *true* Both the drawing and the description are referring to the group of *Tunicata* (*Chordata*) D *false* The drawing shows a representative of the group of *Cnidaria* (*Hydra*) with typical polyps, whereas the description refers to a typical representative of the group of *Echinodermata*.

## Ecology, ethology and systematics

The following figure shows the phylogenetic relationship among several sympatric fish species of the family Mormyridae, which are known to use weak electric communication signals no predator is capable of sensing and the transmission of which does not depend on environmental factors.



For each species, measurements informative about the trophic level (A), body shape (B) and the shape and frequency of their electric signals (C) were determined for several individuals. The figure below shows the position of each measured fish individual in a principal components space for each of the three groups of measurements where all individuals of a species are enclosed in a polygon. The colors refer to the phylogenetic positions shown above. Principal components analysis is a statistical procedure that maximizes the variance on the first few axes.



# Indicate if each of the following statements is true or false.

- A. Among these species, the phylogenetic distance is highly informative about the divergence in communication systems between two taxa.
- B. Speciation of recently diverged sister species was likely driven by sexual selection on the communication system rather than by natural selection by ecological differences.
- C. The morphological variation among taxa is in agreement with an increase in habitat types at the onset of the radiation of the red and blue clade.
- D. Shape or frequency of communication signals in these species is strongly constrained by morphological trade-offs.
- A. False B. True C. False D. False

**Original commentary** 

Correct answers

A false

Recently evolved sister species are as different or even more different in their communication signals than phylogenetically distinct pairs.

B true

Currently diverged sister species are very distinct in their communication signals but show only limited differences in trophic ecology and morphology. Information (no predator with receptors able to track EOD's, no effect of typic ecological parameters like water turbidity and pH on communication system) given in the text make ecological driven effects responsible for the pronounced differences in signals very unlikely.

C false

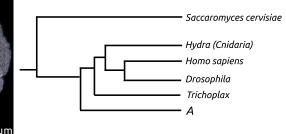
Morphospace is mainly determined by phylogenetically very distinct taxa and therefore not created by recent changes of habitats. Recently evolved taxa do differ only marginally in body shape. D *false* 

There is no obvious correlation between body shape and electric signals of *Mormyridae*. In addition, morphologically very similar species differ heavily in their communication signals.



*Trichoplax adhaerens* is the only known animal (metazoa) of the phylum *Placozoa*. It appears as a flat disc with a very simple structure made of only very few distinct cell types. Nerves, sensory cells and muscle cells are absent. Shown below is an electron microscope image of *T. adhaerens* and a dendrogram based on molecular data indicating the phylogenetic position of *T. adhaerens* in relation to other taxa.





# Indicate if each of the following statements is true or false.

- A. *Drosophila* is more closely related to *Trichoplax* than humans are.
- B. Taxa A listed in the dendrogram is more likely to be a sponge (*Porifera*) than a snail (*Mollusca*).
- C. Trichoplax has no coelom and no gastric tube system.
- D. *Trichoplax* is likely to be a representative of bilateria (animals with distinct dorsal and ventral sides as well as front and back side).
- A. False B. True C. True D. False

## **Original commentary**

Correct answers

## A false

As Placozoa are a sister group of all Eumetazoa, they are equally related to both humans and Drosophila.

B true

Taxa A has a more basal position than *Cnidaria*. *Porifera* are known to be the most basal group of metazoa, whereas *Mollusca* are representatives of bilateria and therefore in the same group as humans or *Drosophila* are. *C true* 

Coelom and gastric tube are traits of higher *Metazoa*. *Cnidaria* do not yet have those organs, and *Placozoa* have even a more basal position than *Cnidaria*. Moreover, these organs would demand for more distinct cell types than *Placozoa* has.

D false

*Placozoa* have a more basal position than *Cnidaria*, which is a group of organisms that do not yet belong to *Bilateria*.

References Srivastava, Nature (2008)



While some mammals are known for their large difference in body size between males and females, there is no apparent difference in size between sexes in other mammal species. This sexual dimorphism can often be well explained by the ecology and mating system of a species.

# Indicate if each of the following statements is true of false. A significant sexual size dimorphism with larger males is expected in ...

- A. ... a very small antelope (< 5 kg), in which a male and a female together defend their small area (< 10 ha) with rich food resources, water and shelter.
- B. ... a seal species where males are known to travel large distances offshore to feed on dispersed food and to copulate with any oestrous female they encounter.
- C. ... an antelope species, in which males gather on a sandy lake shore during the dry season, where each of them defends a 20 m  $^2$  area vigorously.
- D. ... a small carnivore (about 20 cm in length) living in a predator-rich habitat in mixed sex groups. The offspring rely heavily on parental care from both sexes and males are known for their remarkably large testicles.

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

## **Original commentary**

Correct answers

A false

Both sexes defend the small territory as a full time job year-round against conspecifics, therefore selection pressures are the same for both sexes.

B false

This particular case of fission-fusion mating selects more for agile, fast travelling males and resource-rich females than for heavily built competitive males.

This territory is evidently too small and of bad food quality to serve as home range. Male-male interaction is permanent and highly competitive, females choose under many males - selection to show good genes and heavy bodies.

D false

This species is social and group members have similar functions. Male-male competition is limited since cooperation is very important to limit own mortality and to increase survival rate of offspring. In this system it is likely that females mate with several partners that cause sperm competition.

References

Floyd, Journal of Mammology(1998)

## Ecology, ethology and systematics

In ecology, two different measures of biodiversity are commonly used: alpha diversity characterizes the biodiversity at a given location and beta diversity characterizes the diversity found between habitats. A good estimate of alpha diversity is the Shannon index, which is computed as

$$H = -\sum_{i=1}^{S} p_i \cdot ln(p_i)$$

where the sum runs over all species 1, ..., S present in a habitat and  $p_i$  is the relative abundance of Species i.

The table shows the abundance of adult trees of eight tree species (A through H) in four plots of a temperate ecosystem in both pristine and disturbed states.

Plot	State	А	В	С	D	Е	F	G	Н
1	pristine	19	0	56	332	0	76	0	0
2	pristine	3	0	12	456	0	5	0	0
3	disturbed	13	135	0	101	5	0	0	0
4	disturbed	0	143	12	178	0	4	13	7

# Indicate if each of the following statements is true of false.

A. Alpha diversity is higher in Plot 1 than in Plot 2.

- B. Disturbance seems to increase beta diversity in this ecosystem.
- C. Species B is likely to be a pioneer species.
- D. Seedlings of species D perform best in the presence of adult D trees.

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

Original	commentary
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Correct answers

A true

While the number of species present is the same in both plots, their relative abundance is rather different in that species D dominates the system much more in plot 2 than plot 1, leading to a lower alpha diversity in plot 2. B *true* 

Beta diversity is much higher among plots 2 and 3 than plots 1 and 2. The students should be able to see this without calculating anything since exactly the same species are present in plots 1 and 2, but several species are restricted to either plot 3 or 4.

C true

Pioneer species are the first species to colonize a damaged / disturbed habitat. While other species are also present uniquely in either plots 3 and 4, none of them is present in both nor in that number, suggesting that species B. D *true* 

This is a typical characteristics of a species capable of dominating tree ecosystems, as species D is capable of doing in the pristine state. Seedling of other trees only get a chance after disturbance through external factors.