



NATIONAL ENTRANCE EXAMINATION 2025

Examination Date: Wednesday 13 August 2025

Total Time allowed: 2 hours.

This examination consists of multi-choice, true/false, numerical, and simple calculation questions. True/false question sets are marked as follows: all 4 correct is 1 mark, 3 correct is 0.6 marks, 2 correct is 0.4 marks, and 1 correct is 0.2 marks.

The exam is worth 60 marks and is delivered on Education Perfect. It may be sat at any time on the 13th of August between 8am and 8pm.

Students are advised to allocate equal time to each mark (**approximately 2 minutes per mark**). Marks will not be deducted for incorrect answers.

1. Open Google Chrome (note this technology is **NOT supported by Safari** so you **MUST NOT** use the Safari browser).
2. Login to Education Perfect using the username and password sent to the email you registered under.
3. Click on the exam and begin. Once you begin you will have exactly 2 hours to complete all questions.

The exam must be sat under exam conditions and is actively invigilated by a teacher in your school. It also uses remote monitoring, notifying NZIBO if a student is off task or having connection issues.

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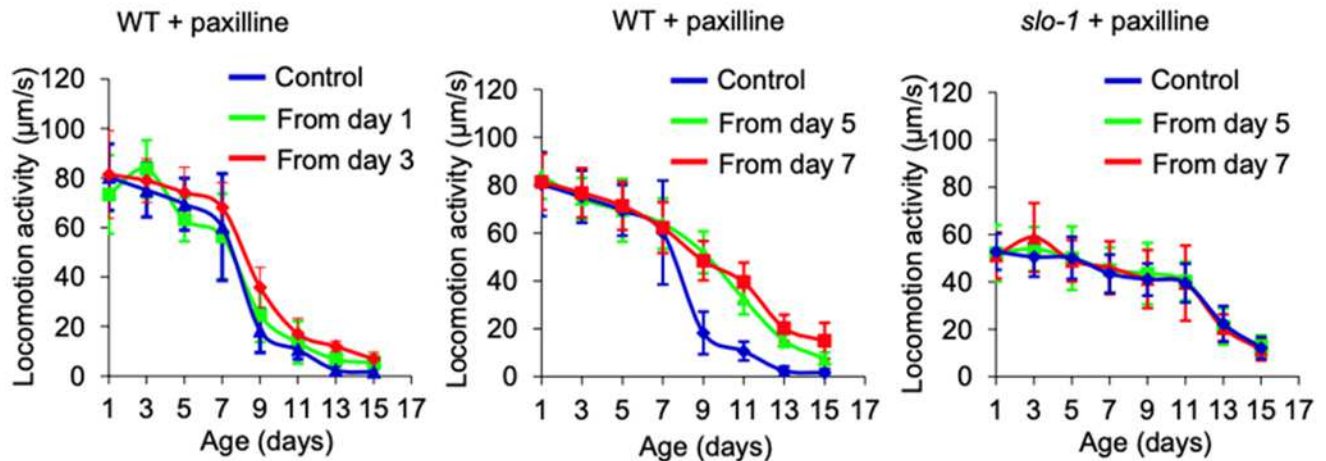
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Section A

Question 1

A recent study conducted by Li *et al.* (2019), investigated the effects of the pharmacological agent paxilline, a potassium channel blocker in neurons, on the locomotion activity of the nematode, *C. elegans*, within an aging context. The lifespan of *C. elegans* is typically 2-3 weeks under laboratory conditions, during which aging-related loss of motor function can be used as a biomarker which correlates strongly with mortality.

The figure below compares the effects of 10 nM paxilline treatment on locomotion activity in both wild-type (WT) and *slo-1* deficient mutant *C. elegans*, with treatments beginning at four developmental stages: day 1 (early adulthood), day 3 (young adulthood), day 5 (mid-life), and day 7 (late adulthood).



Source: <https://www.science.org/doi/pdf/10.1126/sciadv.aau5041>

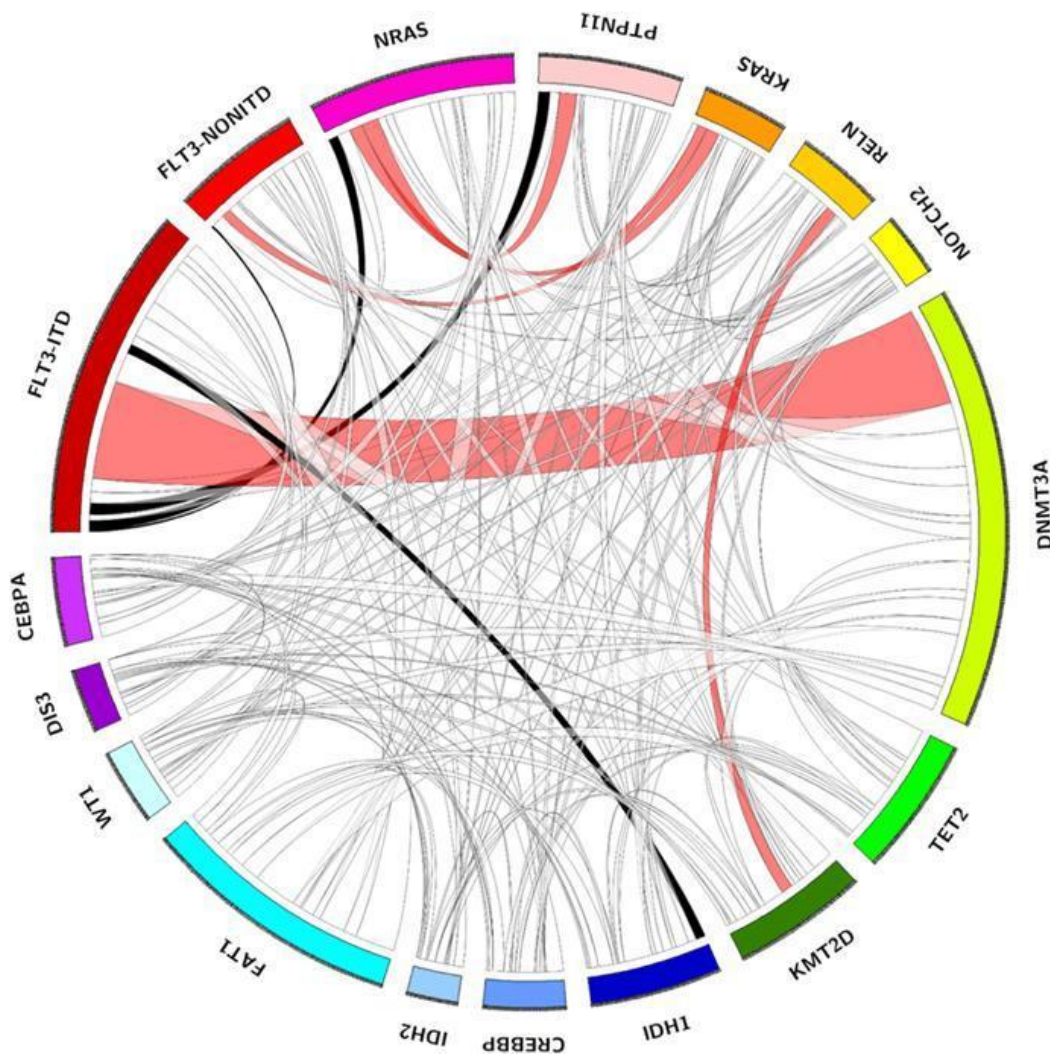
Which conclusion is best supported by the figure above?

- A. Paxilline is toxic to young worms, but beneficial in old age.
- B. Paxilline is more effective on *slo-1* mutants than WT *C. elegans* in preserving motor function.
- C. Paxilline's effects depend on *slo-1* and are age specific. *
- D. Paxilline is effective for treating motor neuron disease in humans.
- E. Paxilline's effects do not depend on *slo-1*, but are age specific in the WT.

Question 2

Acute Myeloid leukaemia (AML) typically has a genetic link to a number of commonly mutated genes. Research conducted by Wang et al., (2021) represents the association of a number of these mutations in a circos plot given below, which visualises pairwise relationships between these mutations in AML patients with the usage of coloured ribbons.

In the circos plot below, white ribbons suggest that mutations to these genes arise independently and are not associated in AML, while red ribbons suggest significant co-occurrence. Black ribbons indicate mutations that typically do not exist in the same patient together. The width of the ribbons correlates to the frequency of AML patients affected by said association.



Source:

<https://www.researchgate.net/publication/353113978> The correlation of next-generation sequencing-based genotypic profiles with clinicopathologic characteristics in NPM1-mutated acute myeloid leukemia

Which of these statements is false based on the circos plot:

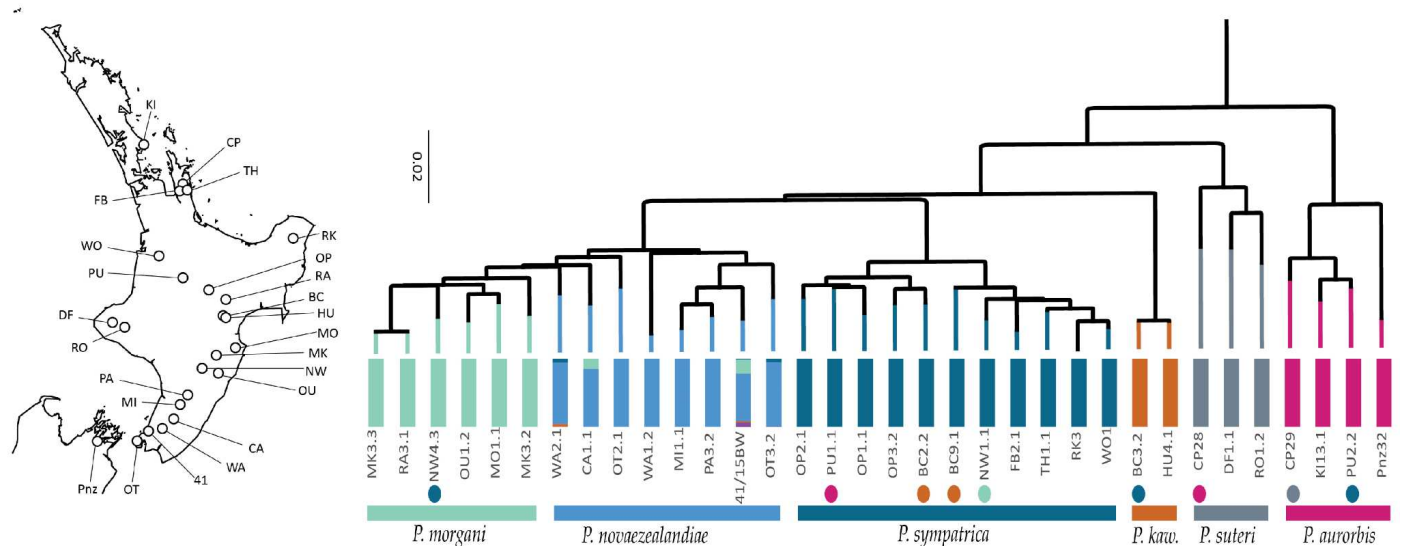
- A. *FLT3-ITD* and *DNMT3A* is the most frequent pair of mutations to occur together in patients in this study.
- B. *NRAS* and *FLT3-ITD* are mutually exclusive.
- C. *TET2* is positively correlated with *IDH1*. *
- D. *IDH1* has no association with *FLT3-ITD*.
- E. A patient is less likely to have mutations to both *PTPN11* and *FLT3-ITD*, than *RELN* and *KMT2D*.

Question 3

Ngaokeoke (velvet worms) are small annelids, native to New Zealand.



Nuclear and mitochondrial DNA from Ngaokeoke sampled from multiple sites around New Zealand were analysed. Multilocus nuclear genotypes (coloured vertical bars) and mtDNA variation (phylogeny) among a set of North Island *Peripatoides* are shown below. Individuals collected in sympatry are indicated by spots coloured for the corresponding cluster.



Source: Trewick, Koot and Morgan-Richards, 2024. Ngāokeyoke Aotearoa: The *Peripatoides* Onychophora of New Zealand.

What is the best conclusion to be made from these data?

- A. There was only one species of Ngaokeoke found at each site.
- B. For the samples with both types of data, nuclear and mitochondrial clusters correspond. *
- C. The genetic analyses support the naming of at least one new species of *Peripatoides*.
- D. It is highly unlikely for geographical intermixing of isolated populations.
- E. There are no clear conclusions to be made from these data.

Question 4

Hochsetter's Frog are an endangered New Zealand species. Scientists are still establishing aspects of their behaviour and life-cycle. The table below shows the number of froglets (baby Hochsetter's frogs) found, and their Snout-Vent Length (SVL) with respect to sampling effort in the Hunua ranges and Mahurangi forest.

	Froglets ≤ 11 mm SVL				Days searched			
	$\bar{x}$	sd	range	n	Brynderwyn Hills†	Mahurangi Hills†	Hunua Ranges†	Golden Cross*
January	9.4	0.9	6–10	34	5	3	4	5
February	9.2	1.1	6–10	19	4	4	2	0
March	9.9	0.8	7–10.8	16	1	5	4	5
April	9.9	0.8	8–10	37	0	0	8	10
May	10.0	1.1	7–10.9	12	1	0	2	8
June	8.9	0.8	7–10	29	21	0	0	0
July	9.3	1.2	6–11	20	9	0	0	8
August	9.5	0.9	7–10.5	23	6	6	0	0
September	9.4	1.0	7–11	54	9	5	4	0
October				0	0	7	2	0
November	10.9	–	10.9	1	0	1	0	1
December	10.2	0.1	10–10.3	2	0	3	0	4

*Data from Slaven (1992); † all Mahurangi, Hunua data, and some February, March and August data from the Brynderwyn Hills from Michael Crossland (pers. comm.)

Source: Beauchamp, A., Lei, P., & Goddard, K. (2010). Hochstetter's frog (*Leiopelma hochstetteri*) egg, mobile larvae and froglet development. *New Zealand Journal of Zoology*, 37(2), 167–174. <https://doi.org/10.1080/0028825X.2010.482975>

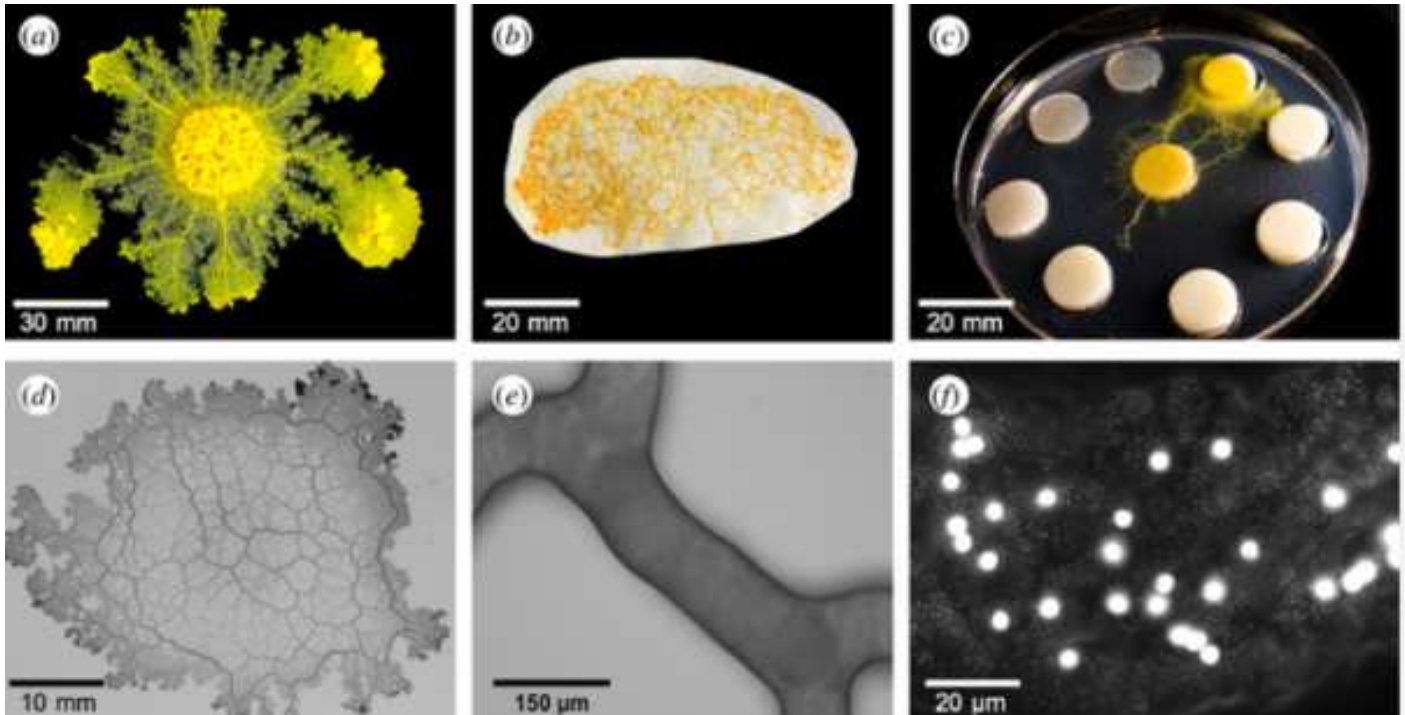
- There was no sampling completed in October
- The highest rate of frogs found per days sampled was in September
- The froglets with the largest average SVL were found in November
- The most froglets were found in the Brynderwyn Hills
- The largest froglet was found in Mahurangi

Which of the statements (i.-v.) above are true?

- ii and iii are true. *
- ii, iv and v are true.
- ii, iii, iv and v are true.
- None of the statements are true.
- All of the statements are true.

Question 5

Slime moulds are a group of unrelated eukaryotic organisms, classified within the kingdom Protista, that exhibit characteristics of both animals and fungi, and are known for their surprising similarities to the network structures of animal brains



What is the size of the largest slime mould in the images above?

Enter your answer as an integer here: **Answer 120 mm. 0.5 marks if no units are given.**

Question 6

If, in each of the numbers below, the first and last digits are swapped, how many of the resulting numbers are odd?

729 648 513 382 845 197

- A. one
- B. two
- C. three
- D. four *
- E. five

Question 7

Archaeopteryx were a genus of dinosaur that lived during the Late Jurassic Period, around 150 million years ago. Fossils show that it had feathers, asymmetrical wing structures, a wishbone, claws on its wings, teeth on its beak, and a long bony tail. They could reach up to 0.5 meters in body length and 0.7m in wingspan.



Archaeopteryx Fossil

Source: <https://www.livescience.com/24745-archaeopteryx.html>



Artist's rendition of what *Archaeopteryx* may have looked like

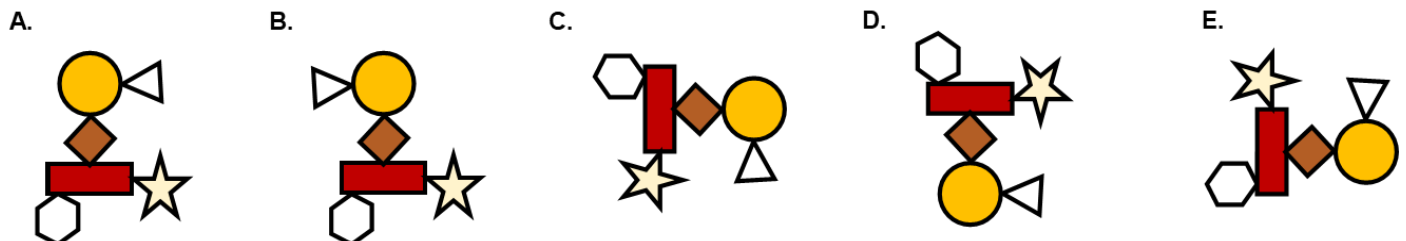
Source: <https://kids.nationalgeographic.com/animals/prehistoric/facts/archaeopteryx>

Which of the following best explains how *Archaeopteryx* contributes to our understanding of evolutionary processes?

- A. It is an example of convergent evolution, that its bird-like features evolved independently, and in a non-avian reptile lineage.
- B. It is an example of genetic drift, that random mutations led to the appearance of flight-permitting traits.
- C. It provides morphological evidence supporting the theory of descent with modification, showing how homologous traits are inherited from a common ancestor. *
- D. It proves that all features required for flight must have evolved simultaneously.
- E. It shows that birds and reptiles evolved completely separately and share no common ancestry.

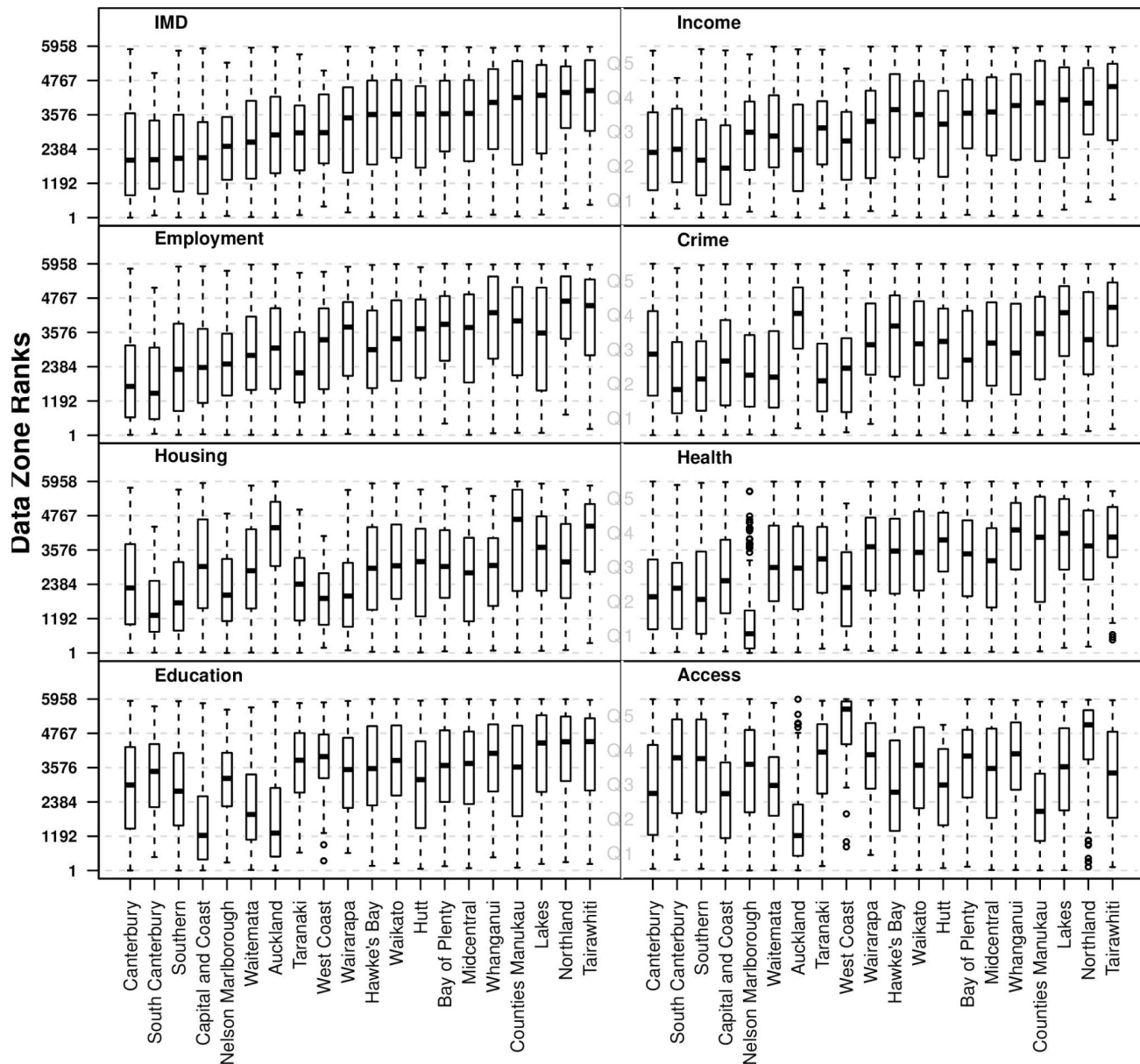
Question 8

Which of the following is the odd one out? **Answer B**



Question 9

Quality of life can be measured using multiple factors, the analysis of which provides insights into relative deprivation in New Zealand. The Index of Multiple Deprivation (IMD) gives us a measure of how regions compare with each other. The graph below shows the distribution of (A) the IMD and its Domains (B-H), ordered by median IMD rank. A higher rank in each of the graphs below, indicates a higher level of deprivation.



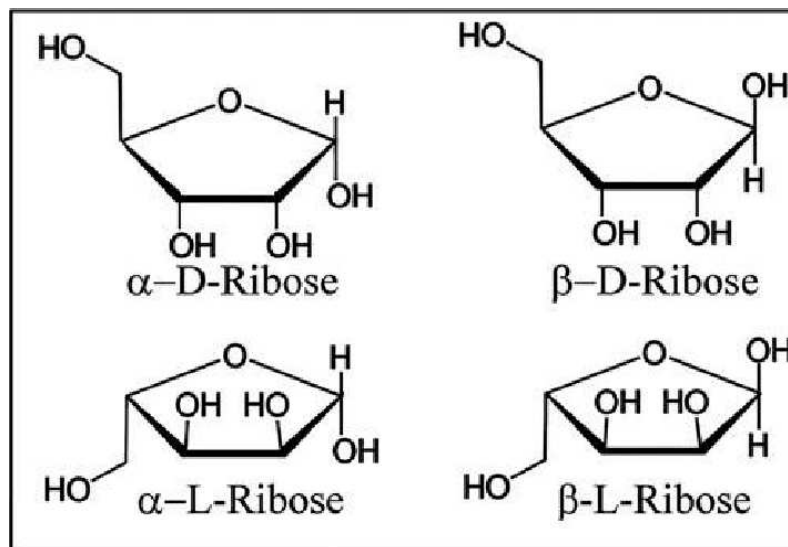
Source: The New Zealand Indices of Multiple Deprivation (IMD): A new suite of indicators for social and health research in Aotearoa, New Zealand, 2017. <https://doi.org/10.1371/journal.pone.0181260>.

Based on the data shown in the graph above, indicate whether the following statements are true or false:

- The region with the lowest relative deprivation is Canterbury. **TRUE**
- The geographical region with the highest income is Tairāwhiti. **FALSE**
- The geographical regions with the least deprivation in terms of access to education are Auckland and Capital and Coast. **TRUE**
- There is a correlation between levels of income and employment. **TRUE**

Question 10

Pentose sugars are fundamental in both RNA and DNA. The carbons in the ring are numbered clockwise with the oxygen at the top being attached to carbon 1. The configurations of cyclic D- and L-ribose with the glycosidic hydroxyl group in either α or β anomeric position are shown in the diagram below.



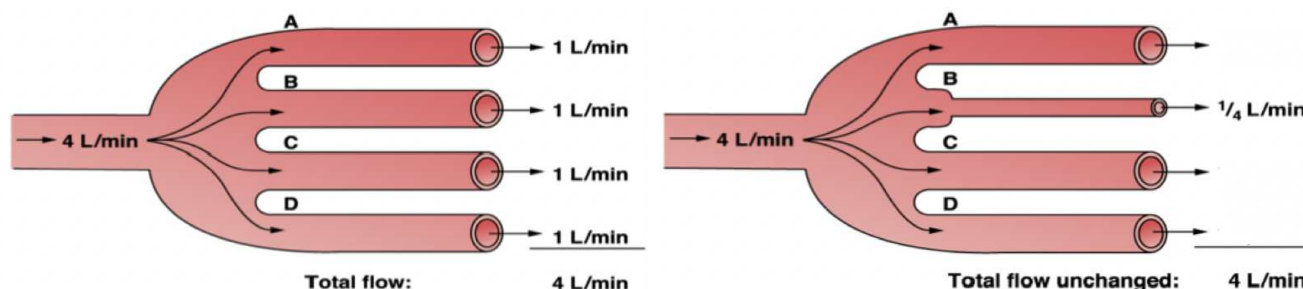
Source: Banfalvi G. Ribose Selected as Precursor to Life. DNA Cell Biol. 2020 Feb;39(2):177-186. doi: 10.1089/dna.2019.4943. Epub 2019 Dec 4. Erratum in: DNA Cell Biol. 2020 Mar;39(3):498. doi: 10.1089/dna.2019.4943.correx. PMID: 31804855.

The conformation change from α -D-ribose to β -L-Ribose is best described by (A-E)?

- A. Delocalization of the aldehyde group between the groups of the H atom and the hydroxymethyl group of the 5th carbon atom followed by anomeric con-Glycosidic hydroxyl group rotation at carbon 2. *
- B. Anomeric con-Glycosidic hydroxyl group rotation.
- C. Delocalization of the aldehyde group between the groups of the H atom and the hydroxymethyl group of the 5th carbon atom.
- D. Anomeric con-Glycosidic hydroxyl group rotation followed by flattening of the ring at carbon 3 and 4.
- E. Delocalization of the hydroxyl group at carbon 2, followed by anomeric rotation of the aldehyde group at carbon 4.

Question 11

In the diagram below, Image 1 shows the flow rate of blood through a main vessel, and 4 branching vessels. The flow through the main vessel equals the total flow rate. Image 2 shows a scenario where vessel B is partly constricted so the flow through B is 0.25 L/min.



Source: Human Physiology: An integrated approach. D Silverthorn Pearson, 6th Edition, 2013.

If the total flow rate remains the same, what is the flow rate through vessel A, if vessels A, C and D are identical?

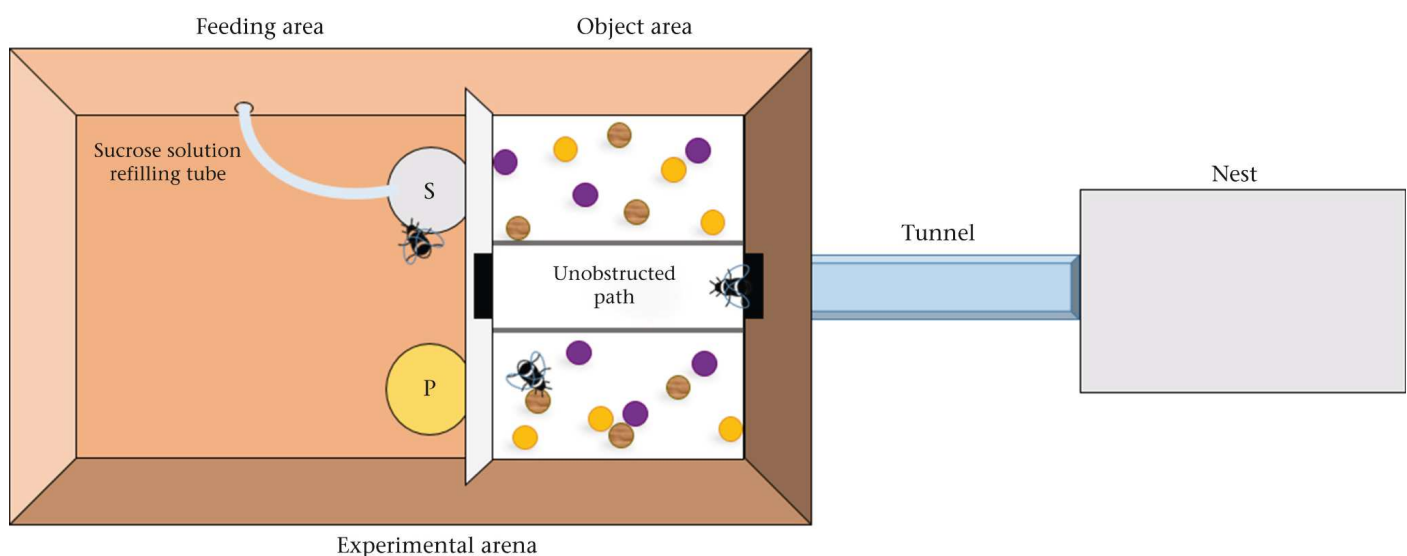
Enter your answer here to 2 decimal places: _____ answer 1.25 L/min ½ mark if no units

Questions 12 & 13

A variety of animals have been found to play - to interact with and manipulate inanimate objects 'just for fun'. A recent study investigated whether insects, the bumble bee, *Bombus terrestris* play as most clear examples of object play come from mammals and birds.

The diagram below shows an aerial view of the experimental set-up. It contains a foraging area and object area. A nestbox containing a bumble bee colony was connected to an arena via a plastic tunnel. The tunnel led to an unobstructed path in the object area with randomly distributed coloured balls on the sides of the path: nine mobile balls on the right and nine immobile balls on the left. The path led to the foraging area which contained *ad libitum* (as much as desired) 30% sucrose (S) and ground pollen (P). The location of the sucrose and pollen were alternated every day to avoid bees developing a side bias.

The bumble bees were shown to interact preferentially with the mobile balls on the right.



Entries into the areas with mobile and immobile balls were recorded. Ball rolling was defined as when an individual bee was on top of a ball and, while maintaining contact with the ball, rotated it along the floor. A total of 910 ball-rolling actions by 45 bumble bees were recorded. The bumble bees were shown to interact preferentially with the mobile balls on the right following their first ball rolling experience.

Source: Hiruni Samadi Galpayage Dona, Cwyn Solvi, Amelia Kowalewska, Kaarle Mäkelä, HaDi MaBouDi, Lars Chittka, Do bumble bees play?, *Animal Behaviour*. Volume 194, 2022, Pages 239-251, <https://doi.org/10.1016/j.anbehav.2022.08.013>.

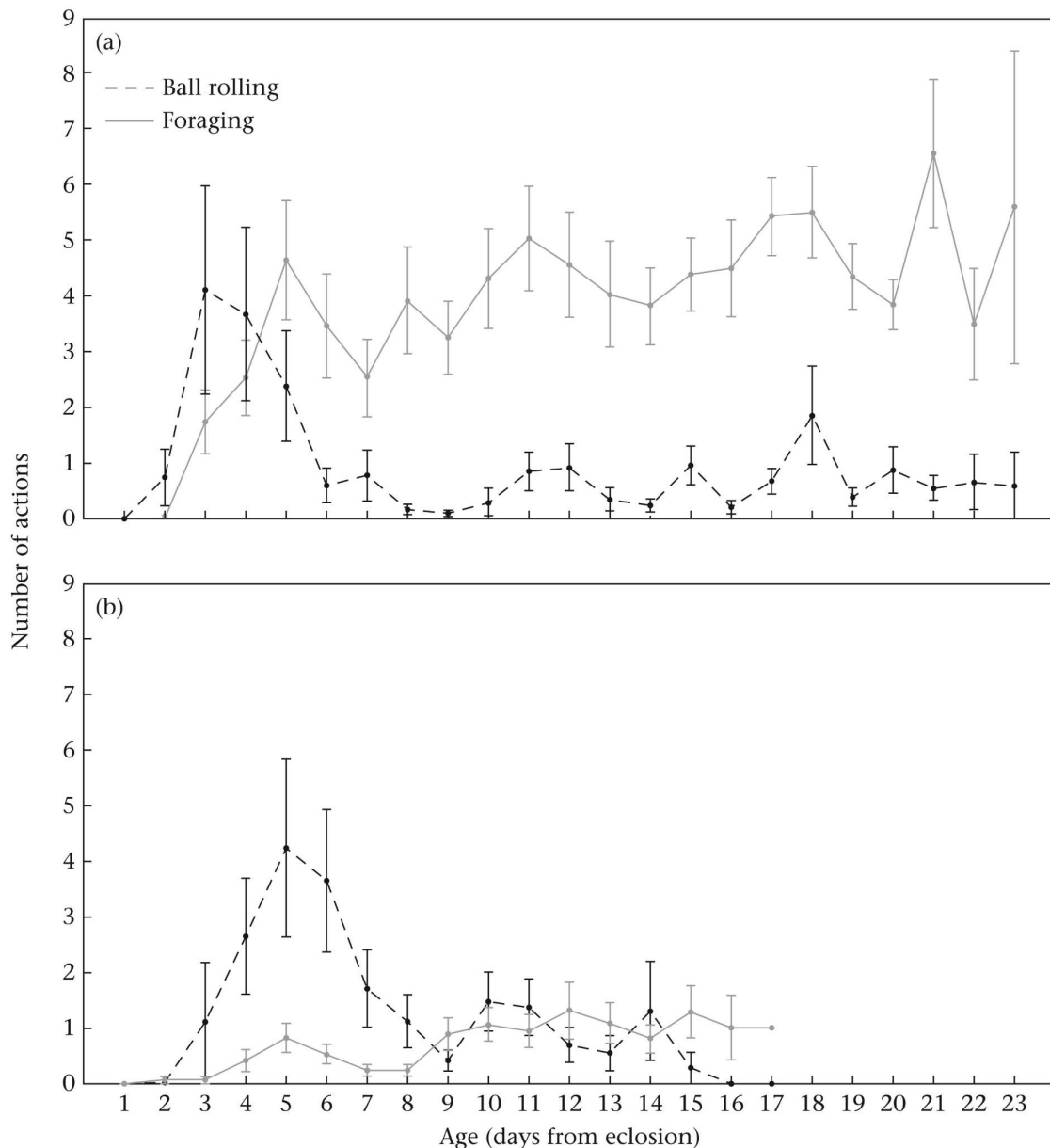
Question 12

Which of the following is the most important aspect of the experimental set-up to ensure that the bees were not engaging with the mobile balls on the right because of a preference or bias for entering that side of the arena?

- A. The nestbox tunnel led to an unobstructed path in the object arena.
- B. Balls of three different colours were used.
- C. Mobile balls were always on the right of the central path.
- D. The location of sucrose and pollen were alternated each day. *
- E. The coloured balls were randomly placed on either side of the central path.

Question 13

The graph below shows ball-rolling activity and foraging activity with age for (a) female and (b) male bees. The number of actions ($\pm$ SE) for each day since the emergence from the larval cell is shown.



These data show that:

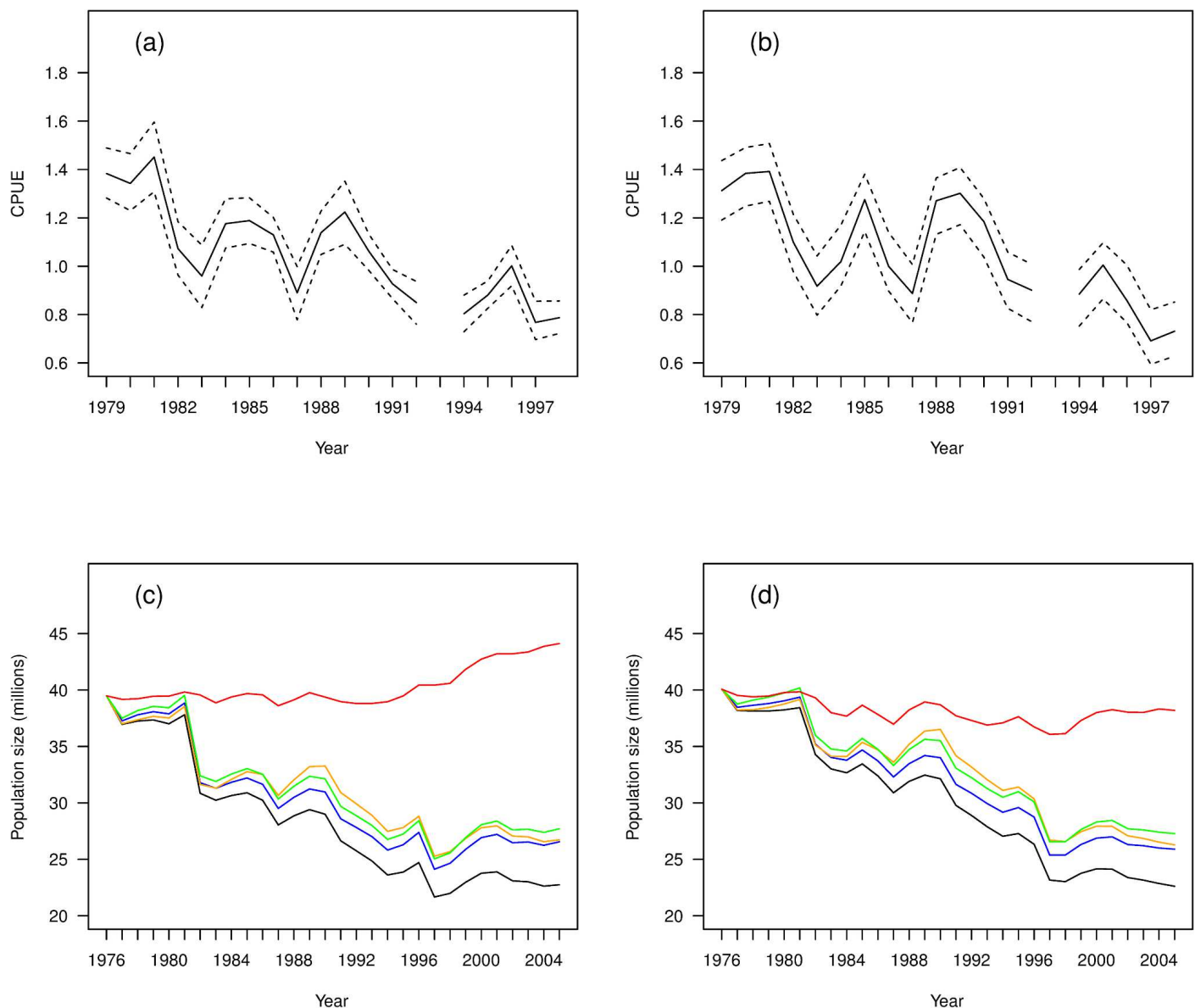
- A. Bees rolled balls more when younger. **TRUE**
- B. The pattern of object manipulation was a result of a general increase in activity related to foraging. **FALSE**
- C. Foraging activity increased with age for females whilst ball-rolling activity peaked earlier and then decreased. **TRUE**
- D. Males showed similar patterns to females for ball rolling and foraging activity. **FALSE**

Overall the results from this study suggest that bees had a different motivation to move objects compared to that of foraging, that bumble bees 'play'.

Question 14

The numbers of tītī (sooty shearwater; *Ardenna grisea*) in the New Zealand region have been declining since at least the 1960s. Researchers have been using mathematical modelling to investigate the relative importance of factors that may have contributed to this decline. The best-fitting models explained much of the variation in the abundance index when survival and fecundity were linked to the Southern Oscillation Index (SOI), with strong decreases in adult survival, juvenile survival and fecundity being related to El Niño-Southern Oscillation (ENSO) events.

The graphs below show effects of including process error in the best-fitting model on the estimation of Catch Per Unit Effort (CPUE) and the effects of climate, harvesting, bycatch, and weka-depredation with a) CPUE from the best model (S-1F0) and b) CPUE from the modified best model, plus likely mean population size based on both prior beliefs about tītī population dynamics and the observed population size (black), predicted population size in the absence of harvest (blue), the absence of bycatch (orange), the absence of weka (green), and the absence of a relationship between SOI and adult survival (red) c) from the best model (S-1F0) and d) from the modified best model.



Source: McKechnie S, Fletcher D, Newman J, Bragg C, Dillingham PW, Clucas R, et al. (2020) Separating the effects of climate, bycatch, predation and harvesting on tītī (*Ardenna grisea*) population dynamics in New Zealand: A model-based assessment. PLoS ONE 15(12): e0243794. <https://doi.org/10.1371/journal.pone.0243794>

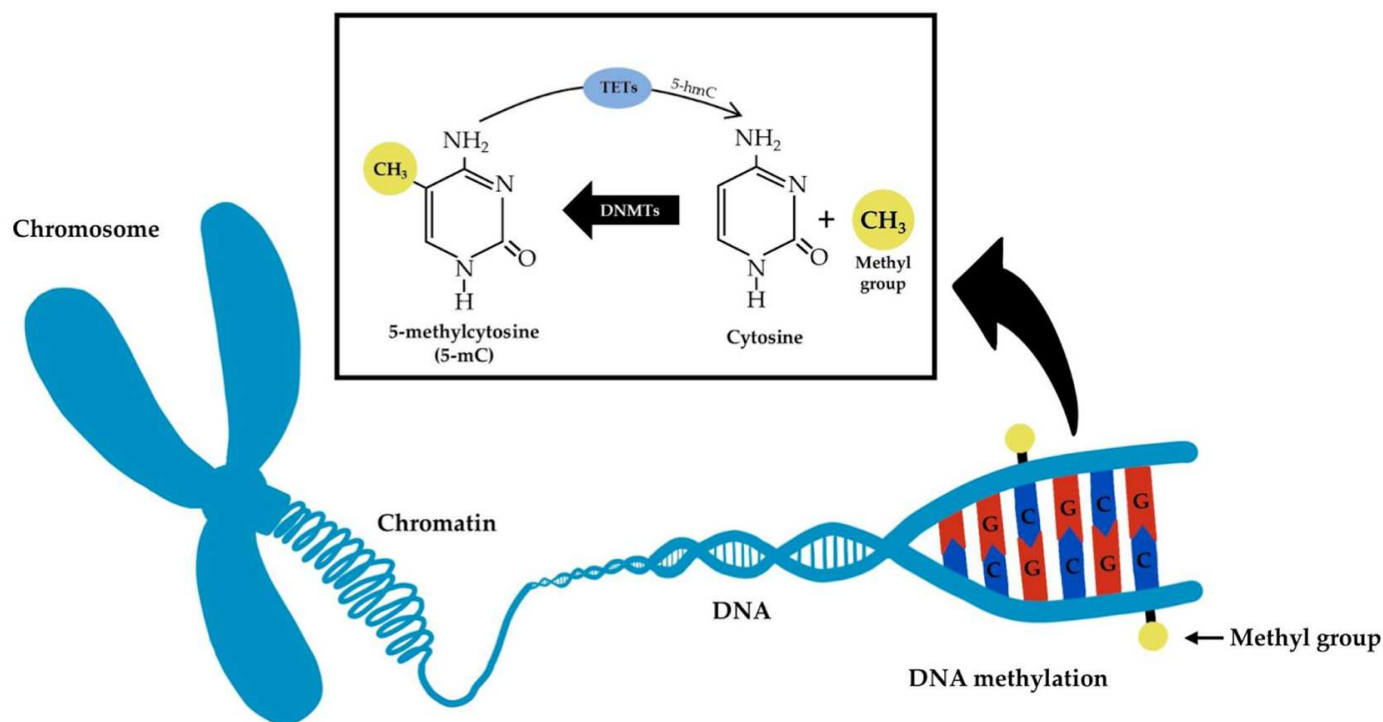
Which of the following statements is **false**?

- A. The best-fitting models explained much of the variation in the abundance index.
- B. There is no relationship between climate (SOI) and tītī population size. *
- C. Predation by introduced animals, harvesting by humans, and bycatch in fisheries also appear to have contributed to the population decline.
- D. Removal of the effects of both depredation by introduced predators and harvesting by humans is likely to have fewer benefits for the population than alleviating climate effects.
- E. Predation by introduced animals, harvesting by humans, and bycatch in fisheries also appear to have contributed to the population decline.

Question 15

DNA methylation is an essential epigenetic modification that involves the addition of a methyl group to the 5th carbon of cytosine residues. This modification plays a crucial role in regulating gene expression, development, and genomic stability. DNA methylation typically represses gene expression by inhibiting the binding of transcription factors or by recruiting methyl-binding proteins that compact chromatin, rendering it less accessible for transcription.

A number of enzymes are involved in this process. Crucial are a family of enzymes known as DNA methyltransferases (DNMTs) and ten-eleven translocation (TET) enzymes which oxidize 5-methylcytosine (5-mC) to 5-hydroxymethylcytosine (5-hmC). The process is summarised in the diagram below.



Source: Valente, A.; Vieira, L.; Silva, M.J.; Ventura, C. The Effect of Nanomaterials on DNA Methylation: A Review. *Nanomaterials* 2023, 13, 1880.

What is the function of TET enzymes in the process of DNA methylation?

- A. Methylation.
- B. Addition of CH₃ to the 5th carbon of cytosine.
- C. Conversion of cytosine to 5-mC.
- D. Conversion of 5-hydroxymethylcytosine (5hmC) to 5-methylcytosine (5-mC).
- E. Demethylation. *

Question 16

Given that:

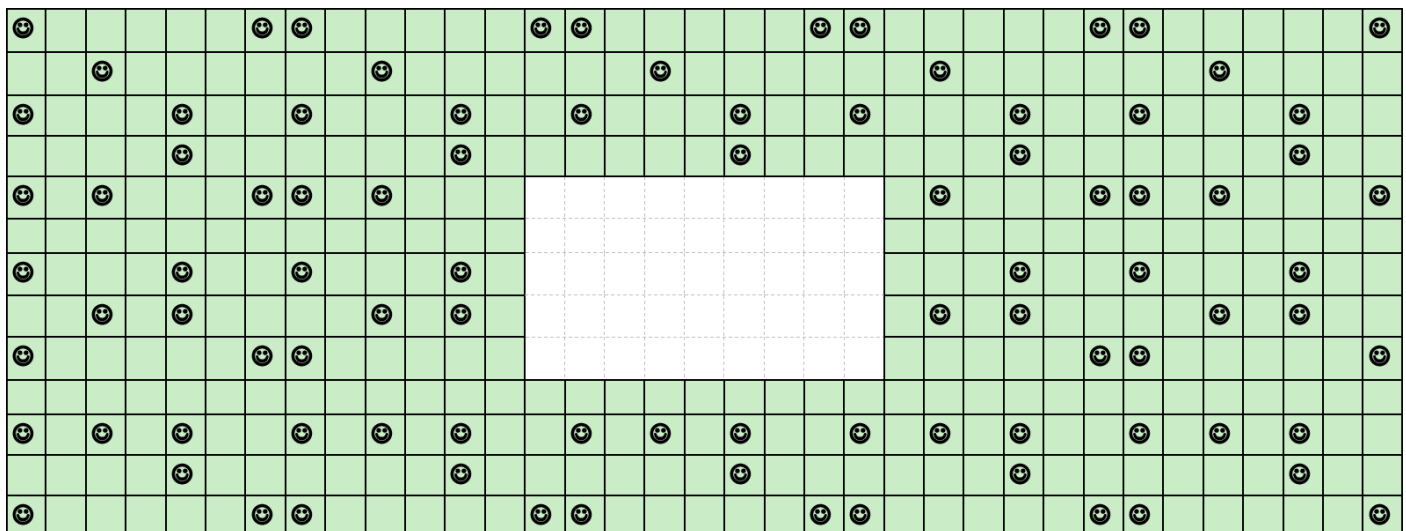
- All Firmicutes are Gram-positive bacteria.
- All Proteobacteria are Gram-negative bacteria.
- All *Clostridium* sp. are bacteria.
- No species of bacteria have a membrane-bound nucleus.
- All Gram-positive bacteria have a thick peptidoglycan layer in their cell wall.

Indicate whether each of the following statements are **true** or **false** using the dropdown box.

- A. All bacteria with a thick peptidoglycan layer are Firmicutes. **FALSE**
- B. *Clostridium* sp. can have a thick peptidoglycan layer. **TRUE**
- C. Proteobacteria do not have a thick peptidoglycan layer. **TRUE**
- D. If penicillin can kill bacteria with thick peptidoglycan cell walls by disrupting the normal formation of the cell wall, then penicillin can kill Firmicutes. **TRUE**

Question 17

How many smiley face squares are missing from this design?



Answer 14

Questions 18 & 19

Blueberry Earth

“What if the entire Earth was instantaneously replaced with an equal volume of closely packed, but uncompressed blueberries?”

This weird yet still intellectually provoking scientific premise was explored by Anders Sandberg from the University of Oxford in 2018. Here are some fun and interesting conclusions:

- Gravity falls to 0.16 g which is about the same as the Moon.
- Surface light levels fall to 4% due to the thick (blue) atmosphere.
- Earth will be converted into an ocean of boiling jam with large impressive geysers of air bubbling to the surface.

Obviously, it's all a bit absurd. But by applying fundamental principles, we can understand and predict the surprising consequences too.

The following information will help you to tackle the next two questions:

- Initial density of closely packed, uncompressed blueberries: $\rho_{\text{berries}} \approx 700 \text{ kgm}^{-3}$
- Estimated density of blueberry pulp: $\rho_{\text{pulp}} \approx 1000 \text{ kgm}^{-3}$
- Average radius of Earth (initial state): $r_{\text{earth}} \approx 6.371 \times 10^6 \text{ m}$
- The relationship derived from mass conservation (where r_{pulp} is the radius of the new pulp sphere):

$$r_{\text{pulp}} = (\rho_{\text{berries}} / \rho_{\text{pulp}})^{1/3} r_{\text{earth}}$$
- Average radius of a single blueberry: $r_{\text{blueberry}} \approx 0.0075 \text{ m}$
- Volume of a sphere: $V = 4/3 \pi r^3$

Question 18

To truly grasp the immense scale of "Blueberry Earth," determine the approximate total number of individual blueberries required to fill the entire volume of our planet, assuming they are closely packed.

Express your final answer rounded to the nearest septillion (10^{24} - one followed by 24 zeros!).

$$\text{Volume of a single blueberry} = 4/3 * \pi * (0.0075)^3$$

$$\text{Volume of Earth} = 4/3 * \pi * (6.371 \times 10^6)^3$$

$$\text{Number of blueberries} = \text{Vol of Earth} / \text{Vol of blueberry} = 6.1296 \dots \times 10^{26}$$

$$\text{Rounded to nearest septillion: } 6.1296 \dots \times 10^{26} / 10^{24} = 612.9697 \approx 613 \text{ septillion}$$

Question 19

The coalescence of blueberries leads to a reduction in the planet's size. Calculate the ratio of the new pulp sphere's radius (r_{pulp}) to the original Earth's radius (r_{Earth}).

Provide your answer as a percentage, rounded to 3 significant figures, to complete the following statement: The Blueberry Earth collapses to ____% of its original radius!

$$r_{\text{pulp}} = (\rho_{\text{berries}} / \rho_{\text{pulp}})^{1/3} r_{\text{Earth}} = (700/1000)^{1/3} * 6.371 \times 10^6 = 5656836.395.$$

$$\text{Ratio: } r_{\text{pulp}} / r_{\text{Earth}} = 5656836 / 6.371 \times 10^6 = 0.88790$$

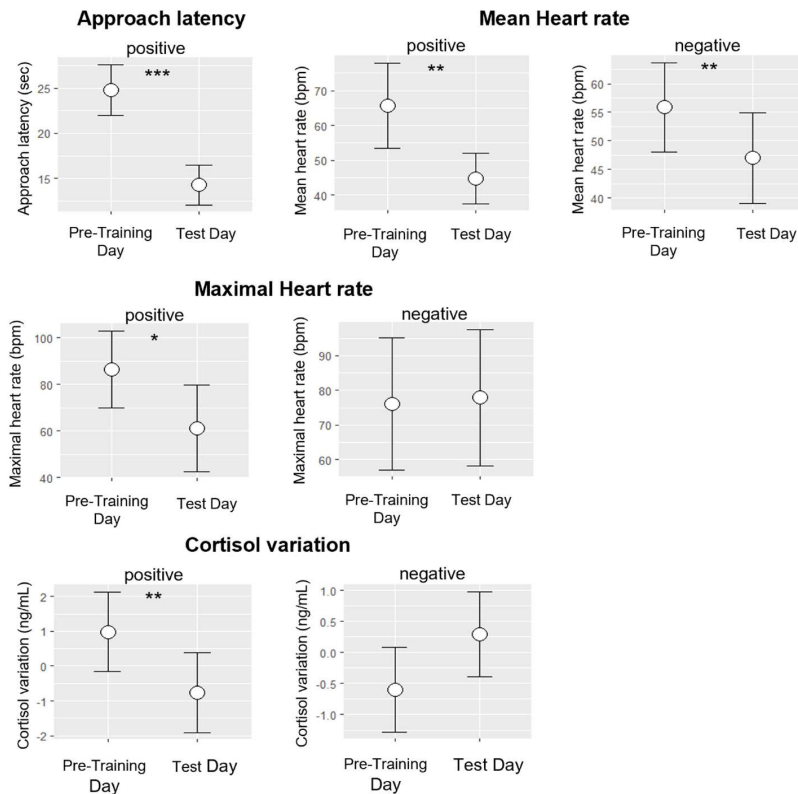
$$\text{Therefore } 88.8\%$$

Source: <https://arxiv.org/pdf/1807.10553>

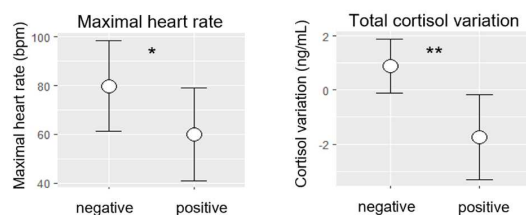
Question 20

Research was undertaken to distinguish between a horse's physiological response to positive and negative stimuli. The positive environment involved being allowed onto fresh pasture. The negative stimuli involved going alone to a novel environment. Horses were first exposed on a 'pre training day' then their responses measured again on a 'test day.' The diagrams below show a summary of the data collected. Statistical significance is indicated by asterisks. * p value = 0.05; ** p value = 0.01; *** p value = 0.005.

A) Evolution of the indicators between the Pre-Training Day and the Test Day



B) Comparison of the indicators between conditions on the Test Day



Source: Characterisation of facial expressions and behaviours of horses in response to positive and negative emotional anticipation using network analysis | PLOS One Published: May 14, 2025

<https://doi.org/10.1371/journal.pone.0319315>

Based on the results of the experiment graphed above, indicate whether the following statements are true or false:

- There is no significant difference in horses' physiological response between positive and negative stimuli. **FALSE**
- There is a significant difference between the horse's response to positive and negative stimuli in all physiological variables measured on test day. **FALSE**
- The only variable to show a difference between the horse's response to positive and negative stimuli is the Maximal Heart Rate. **FALSE**
- Horses' mean heart rate is lower when exposed to both positive and negative stimuli that they have experienced before. **TRUE**

Questions 21 - 22 The Microbial Multiplication Mystery

In a petri dish, a colony of *E. coli* bacteria doubles every 20 minutes. At 8:00 a.m., the colony contains exactly 125 bacteria.

By 10:00 a.m., the population had reached exactly 32,000 bacteria.

Question 21

How many times did the population double between 8:00 a.m. and 10:00 a.m.?

Enter your answer here: 10:00 am – 8:00 am = 2 hours = 120 minutes. Doubling time = 20 minutes.

Number of doublings = $120 \div 20 = 6$ doublings

Question 22

Given that the formula for exponential growth is: $N = N_0 \times 2^t$

Where:

- N = population at time t
- N_0 = initial population
- t = number of doublings
- 2^t reflects doubling every 20 minutes

At what time was the colony exactly 1,000 bacteria?

Enter your answer here:

Answer:

$$N = 125 \times 2^t$$

We solve:

$$125 \times 2^t = 1,000$$

$$2^t = 1,000/125 = 8$$

$$2^t = 2^3 = 8$$

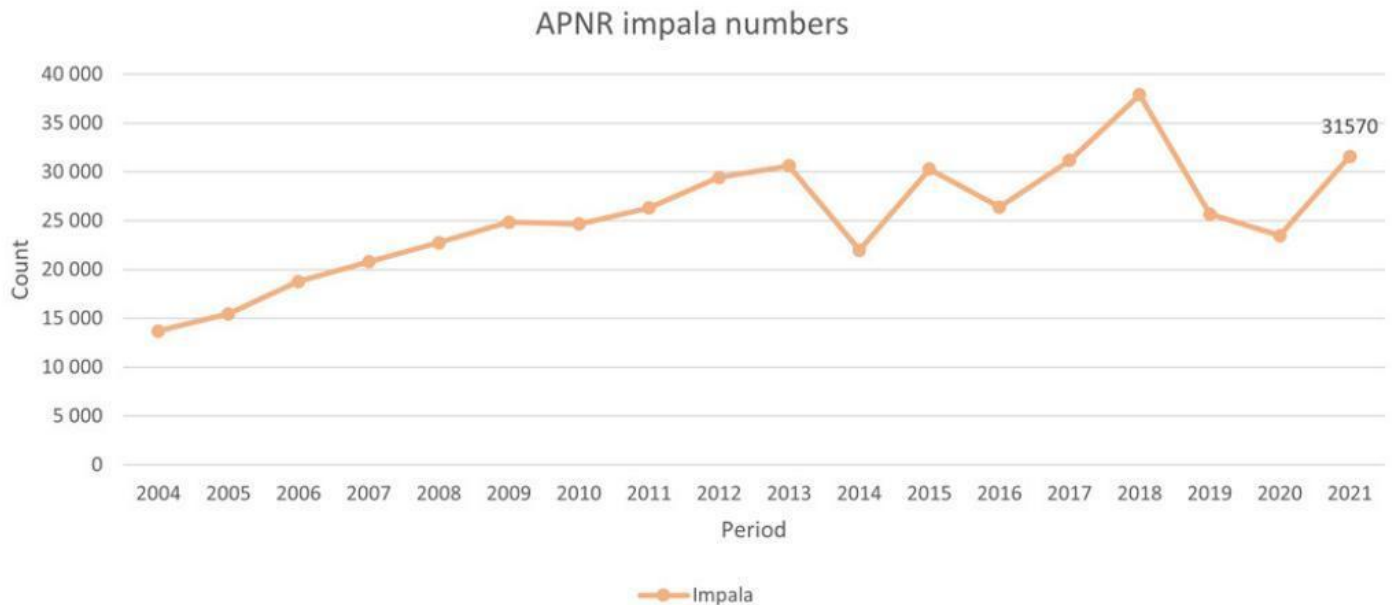
$$t = 3$$

Each doubling = 20 minutes, 3 doublings $\times$ 20 mins = 60 minutes, 8:00 am + 60 minutes = **9:00 am**

Question 23

Adjacent to Kruger National Park in South Africa, 5 private nature reserves are home to an abundance of protected wildlife, including giraffes, lions, elephants, and wildebeests, and are collectively called APNR (Associated Private Nature Reserves). They cover a total land area of 1850 square kilometres. They allow hunting of large African game animals under controlled conditions, meaning that population numbers of these animals remain stable, and in most cases increase, and they end uncontrolled trophy hunting and poaching in the area.

The graph of population count of impala within these nature reserves is shown below.



Source: <https://africageographic.com/stories/keeping-count-wildlife-populations-in-kruger-private-nature-reserves/>

What is the change in population density of Impala in the APNRs between 2005 and 2021?

- A. 12 impala per square kilometre
- B. 31 impala per square kilometre
- C. 15 impala per square kilometre
- D. 9 impala per square kilometre *
- E. 6 impala per square kilometre

Question 24

In a controlled transgenesis experiment, only plants that received both transgene A and transgene B showed resistance to the leaf blight fungus. Some plants received transgene A, some received transgene B, and some received neither.

If the statement above is true, which of the following conclusions must be true?

- A. No plant that received only transgene A was resistant to the fungus. *
- B. All plants that received transgene B were resistant to the fungus.
- C. Any plant that was resistant to the fungus must have received either transgene A or transgene B.
- D. Some plants that received neither transgene still showed resistance to the fungus.
- E. Any plant that did not show resistance did not receive transgene A.

Question 25

Alveolar minute ventilation (VA) is the volume of air, per minute, that reaches the alveoli and participates in gas exchange. Respiration Rate is the number of breaths taken per minute. Tidal volume is the amount of air breathed in and out with each normal breath. Dead Space is the amount of air that doesn't participate in gas exchange.

$$VA = \text{Respiratory Rate} \times (\text{Tidal Volume} - \text{Dead Space})$$

Source: <https://www.respiratorytherapyzone.com/respiratory-therapy-formulas-calculations/>

Indicate whether each of the following statements are **true** or **false** using the dropdown box.

- A. If the average Tidal Volume is 500mL in a normal adult, and the average Dead Space is 150 mL, 30% of the air breathed in is participating in gas exchange. **FALSE**
- B. Increasing the Tidal Volume without changing Dead Space or Respiratory rate will increase VA. **TRUE**
- C. A person who has an above average Dead Space will have a higher than average respiratory rate to compensate if they are to keep breathing with a normal Tidal Volume and VA. **TRUE**
- D. An adult at rest has a respiratory rate of 16 breaths per minute. If their Tidal Volume is 500 mL and their Dead Space is 150 mL, their VA is 5.6 L/min. **TRUE**

Questions 26 & 27

Mammals that live in cold-water environments are large and have adaptations such as thick blubber or fur to prevent them losing heat. For example, polar bear fur has a dense undercoat and the outer coat is made of individual hairs that are actually transparent and hollow, with a core that traps air. The fur's transparency allows sunlight to penetrate to the black skin underneath, where it is absorbed and helps warm the bear. Together with their relatively small surface area as a result of their large size, polar bears stay warm in the arctic cold.

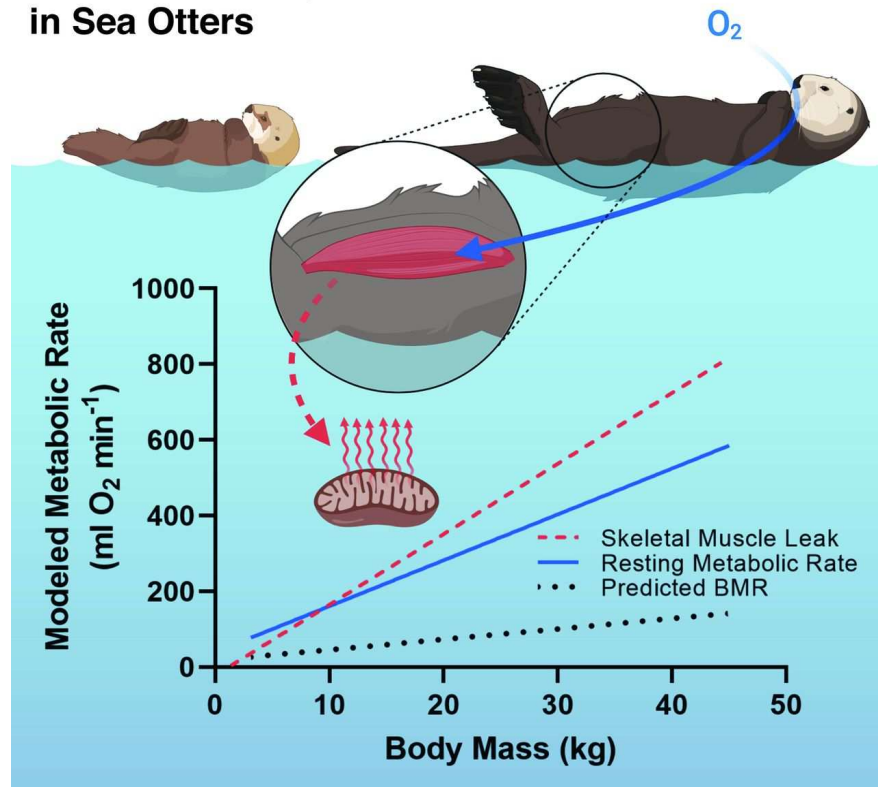
Sea otters (*Enhydra lutris*) are the smallest marine mammals, living in cold North Pacific waters (0° to 15°C). Because water draws heat from the body 23 times faster than air, staying warm is a major challenge, especially for a small animal with no blubber. While their dense fur provides important insulation, a recent *Science* paper explores how sea otters may use internal metabolic adaptations to stay warm.

Like all warm-blooded (endothermic) mammals, sea otters use energy not only for basic bodily functions but also to maintain a constant core temperature of around 37°C. This process happens in the mitochondria, where fats and carbohydrates are broken down using oxygen to produce energy. Some of this energy is used to make ATP (the cell's energy currency), while some is released as heat through a process called *proton leak*. This “leak” helps generate warmth without the muscles doing any physical work.

Young sea otters face extra challenges. As small, undeveloped animals, they lose heat quickly and have less muscle mass to help produce warmth. Notably, neonatal pups have dense natal fur and spend most of their time resting on the mother's abdomen, so there is little heat loss to water until the age of 1 month.

To better understand how sea otters use their muscles to stay warm, researchers measured how much heat-producing proton leak occurs in their muscle cells and combined this with known data on sea otter muscle mass. This allowed them to estimate how much heat skeletal muscles can produce across the otter's body at rest.

Cold-Adapted Hypermetabolism in Sea Otters



Source: J.A. Estes, J.L. Bodkin, M. Ben-David, Otters, Marine, Editor(s): William F. Perrin, Bernd Würsig, J.G.M. Thewissen, Encyclopedia of Marine Mammals (Second Edition). Academic Press, 2009, Pages 807-816, <https://doi.org/10.1016/B978-0-12-373553-9.00189-9>.

Question 26

Considering the information provided and the data in the graph determine whether each of the following statements are true or false.

- A. An otter's resting metabolic rate is much greater than that expected for their size. **TRUE**
- B. Generation of heat via skeletal muscle leak is present even in infants with immature muscles, providing these animals with sufficient internal warmth from birth to maintain their body temperature. **FALSE the skeletal muscle leak is not independently adequate to fully account for the resting metabolic rate owing to the lower relative muscle mass, seen in the skeletal muscle leak red line dropping below the resting metabolic rate.**
- C. In sea otters with body mass greater than ~9 kg, skeletal muscle leak capacity exceeds whole-body resting oxygen consumption and can therefore account for the elevated resting metabolic rate. **TRUE**
- D. Otters are internally warmed by thermogenic leak from skeletal muscle. **TRUE**

Question 27

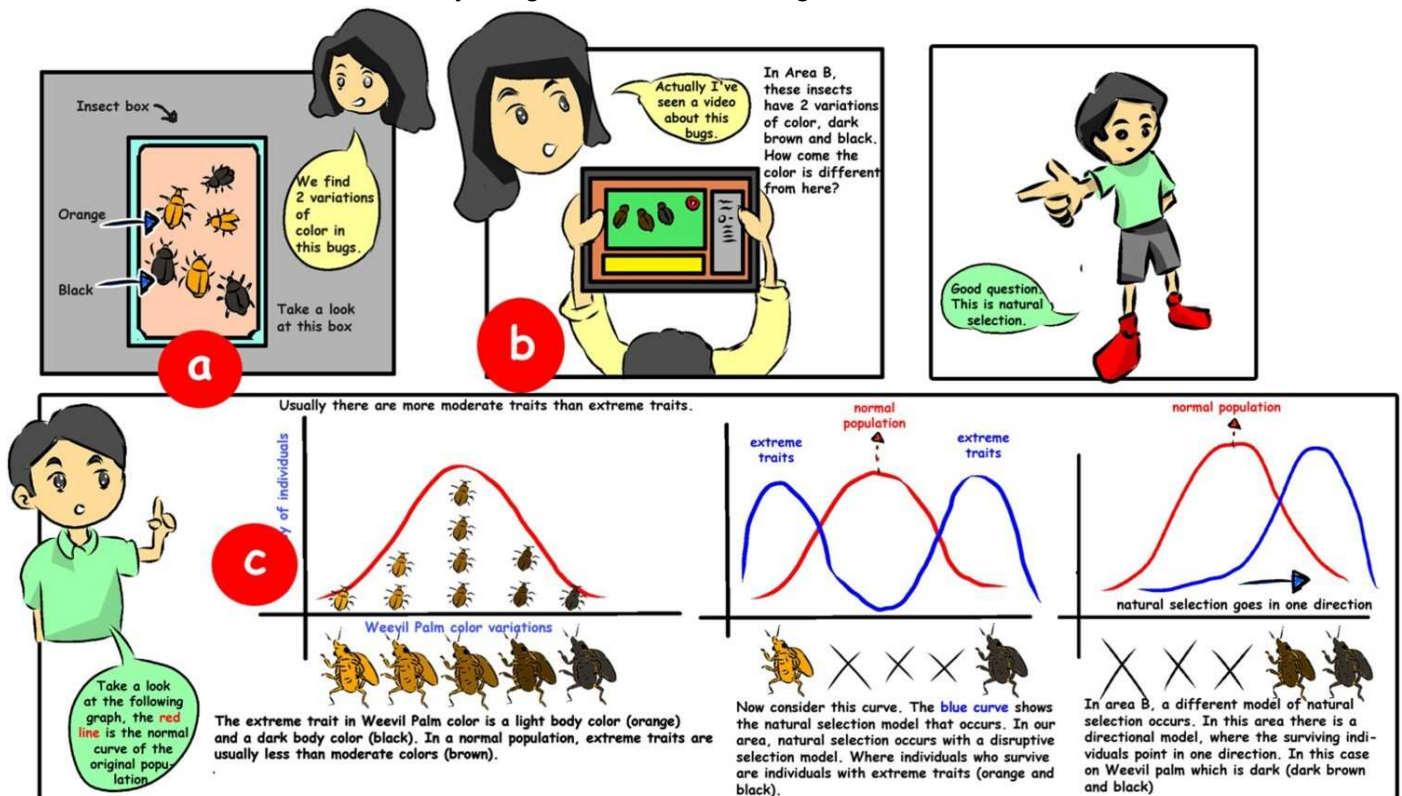
Which of the factors below (I. - V.) do infant otters use to maintain their body temperature?

- I. Increased muscle activity, physical work.
 - II. Thermogenic leak from skeletal muscle.
 - III. Additional muscle thermogenesis via shivering.
 - IV. By seldom entering the water, lying on their mother's abdomen most of the time.
 - V. Dense neonatal fur.
- A. They use all the factors I. - V listed above.
 - B. They only use factors II. - V. *
 - C. They only use factors III. - V.
 - D. They use factors II., IV., and V only.
 - E. They only use factors IV. and V.

Question 28

Natural selection is a fundamental process in evolution where organisms whose phenotype (observable characteristics) is better adapted to their environment tend to survive and produce more offspring, passing on their advantageous traits. Over time, this leads to a population evolving and becoming better suited to its environment. A recent publication looked at the use of comics to foster students' data visualization skills and understanding of natural selection.

In this comic story two children catch Red Palm Weevil (*Rhynchophorus ferrugineus*) insects at several locations where they live. These weevils live on the trunk of sago (dark brown trunk), coconut (light greyish-brown trunk), and sugar (dark reddish-brown to ebony trunk) palms and salak trees (dark reddish-brown trunk). Insects caught have two colours bright orange and dark black. The girl then asked the boy if he had watched a video about this insect, and in the video, this insect had dark brown and black colours, different from the ones they caught, which were orange and black.



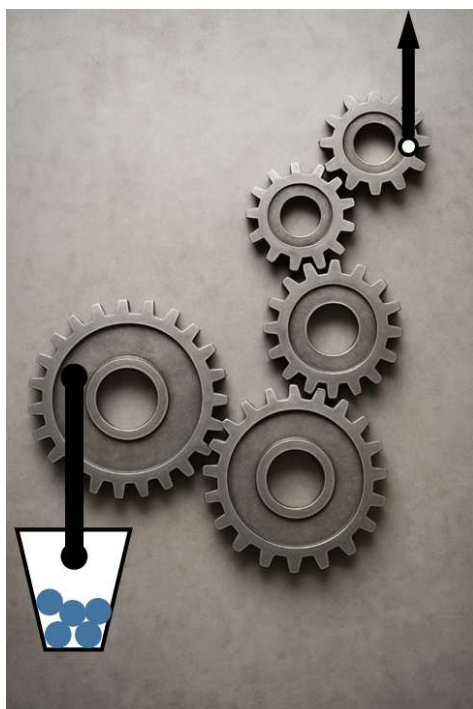
Source: Abrori, F. M., Tejera, M., & Lavicza, Z. (2024). Combining biology and mathematics in educational comics to explain evolution. *Journal of Mathematics and the Arts*, 18(1–2), 154–167. <https://doi.org/10.1080/17513472.2024.2386646>

Which of the following models and explanations can describe the data about weevil coloration that the children collected.

- The weevils were caught on different types of palms and trees and the presence of orange and black weevils is an example of disruptive selection. *
- Disruptive selection has resulted in two species of weevil being found in the area the children studied.
- The colouration of the trunks of the palms in the area the children studied resulted in directional selection of weevil coloration, with orange weevils found on palm species with light coloured trunks and dark weevils found on palm species with dark trunks.
- The dark weevils in the video are a different species to those collected by the children.
- A-D can all describe aspects of the data collected by the children.

Question 29

If the black arrow pulls in the direction indicated, will the load rise or fall?



Choose the correct answer from the dropdown here: Rise, Fall *

Question 30

In 2024, the NZIBO team competed at the International Biology Olympiad (IBO) in Kazakhstan. During their visit, they explored the Astana Botanical Garden and made some interesting ecological observations.

Kazakhstan's forest steppe ecoregion lies between the taiga forests of Siberian Russia to the north and the expansive Kazakh steppe grasslands to the south. This zone consists mainly of flat, sandy lowland plains, with wetlands in the hollows and tree-lined watercourses. Common tree species include birch, aspen, and pine.

While examining the native pine trees, the team noticed that the trees were monoecious, meaning they bore both male and female cones on the same individual. The male cones, which produce yellowish pollen, were located on the lower branches, while the larger, woody female cones were found higher up in the tree (below left). When mature, the scales of the female cones open, allowing the seeds to be dispersed by the wind or by birds or other herbivorous animals.



The team discussed why male and female cones were positioned in such distinct parts of the tree. Which of the following is the most likely explanation for this observed distribution of pine cones?

- A. To protect developing seeds – Positioning female cones higher in the tree helps shield them from birds and other animals that might eat the seeds.
- B. To enhance seed dispersal – The elevated position of female cones allows seeds to be carried farther by the wind once they are released.
- C. To reduce pollen loss to rain – Male cones are located lower in the tree canopy, where they are less exposed to rain falling from above, protecting the pollen.
- D. To increase the likelihood of cross-pollination – Wind-blown pollen from the lower-positioned male cones is more likely to reach the higher female cones on other trees, reducing the chances of self-pollination. *
- E. To optimise nutrient allocation – Separating male and female cones spatially may help the tree manage its energy resources more efficiently during reproduction.

Section B

CUTTING-EDGE BIOLOGICAL RESEARCH

Biological research is vital to Aotearoa New Zealand's economy, environment, and the wellbeing of all New Zealanders. Using cutting-edge tools in DNA science, genomics, and biostatistics, researchers are driving breakthroughs in ecosystem management, the protection of biodiversity, human and animal health, and the advancement of our primary industries.

This research is characterised not only by its scientific excellence but also by its strong foundation in collaboration—between institutions, across disciplines, and with communities. Aotearoa's scientists are at the forefront of international efforts to understand the complexities of life at molecular, organism, and ecological scales, and their work contributes meaningfully to global scientific knowledge.

A distinctive strength of New Zealand's research landscape is its commitment to integrating mātauranga Māori, Māori knowledge, values, and worldviews, into biological inquiry. This approach enriches the way research questions are framed, how data are interpreted, and how solutions are developed, ensuring that science in Aotearoa is both globally relevant and deeply rooted in local context and responsibility.

This section of the NZIBO exam explores real-world applications of biological science in Aotearoa. The questions that follow are inspired by current research initiatives that harness advanced biological techniques to solve complex challenges. You will be asked to apply your understanding of molecular biology, ecology, genetics, bioinformatics, and evolutionary principles to investigate scenarios modelled on cutting-edge New Zealand research.

Through this, we celebrate the role of young scientists, like yourselves, in shaping the future of biological science in Aotearoa and beyond.

UNLOCKING THE SECRETS OF THE ESTUARY USING eDNA

Estuaries are special places where rivers meet the sea, creating rich and dynamic environments. In New Zealand, estuaries range from large wetland harbours like Tauranga Harbour to small lagoons that only open to the sea during storms. These shallow, sunlit waters are packed with nutrients, making them perfect for phytoplankton growth and ideal nurseries for young fish and shellfish. Estuaries also support migratory birds and help maintain biodiversity. They naturally clean water by trapping sediments and breaking down pollutants, and they protect coastal areas by absorbing floodwaters and reducing erosion.

Many New Zealand cities, such as Auckland, Christchurch, and Dunedin, are built around estuaries, making them vulnerable to pollution and degradation from human activity. With about 350 estuaries nationwide, these areas are vital habitats and treasured sources of kai moana. Protecting our estuaries is essential for the wildlife, the environment, and the people of Aotearoa.

Question 31

Key features of estuarine environments are listed below;

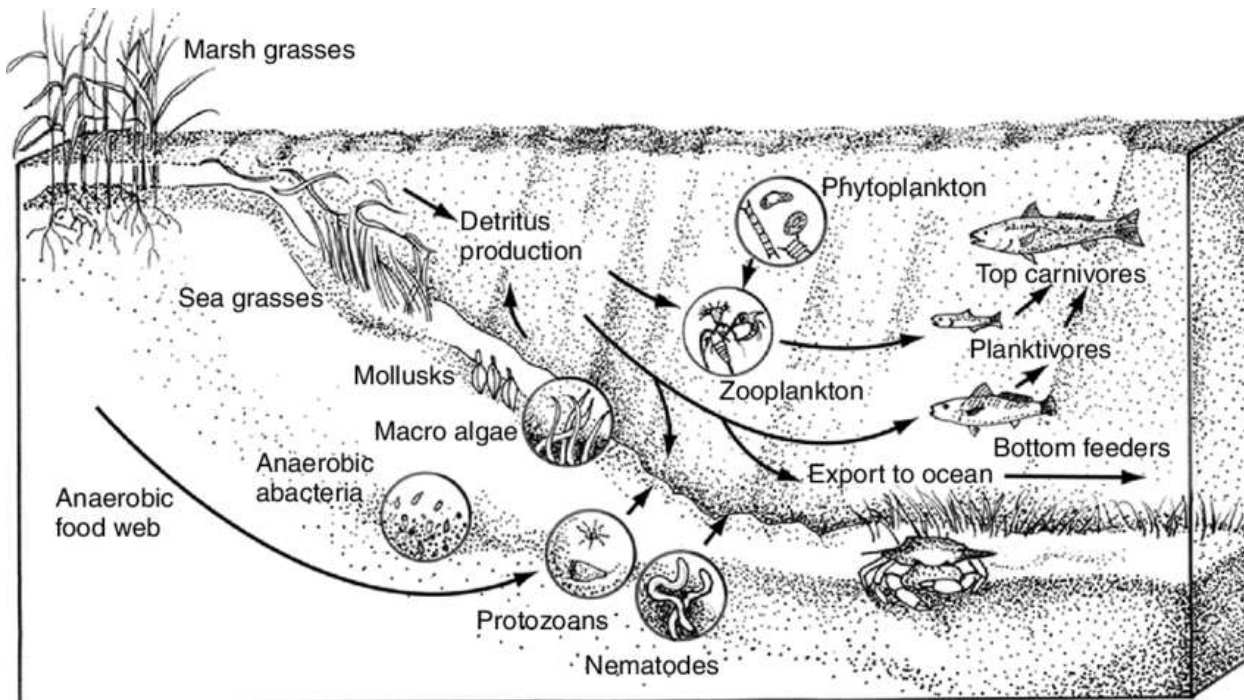
- I. Warm shallow water.
- II. Nutrient inflow from upstream land runoff.
- III. Uninterrupted sunlight.
- IV. High rates of phytoplankton growth.
- V. Nursery conditions for young fish and shellfish.

What are the likely abiotic (physical) factors that make the estuary so productive

- A. all of the above.
- B. I-III only. *
- C. III - V only.
- D. IV - V only.
- E. none of the above.

Question 32

Consider the following food web diagram for a typical estuarine ecosystem showing some feeding links among some of the major trophic (feeding) groupings. Black lines and arrows indicate flow of food from source to consumer.



Source: Estuarine Ecology - Scientific Figure on ResearchGate. Available from:

https://www.researchgate.net/figure/Food-web-diagram-for-a-typical-estuarine-ecosystem-showing-some-feeding-links-among-some_fig4_39012861 [accessed 1 May 2025]

Which of the groupings below is a major trophic grouping?

- A. Seagrass, marsh grass and phytoplankton. *
- B. Detritus, zooplankton, and planktivores.
- C. Anaerobic bacteria, nematodes, and protozoans.
- D. Phytoplankton, zooplankton, and detritus.
- E. Bottom feeders, planktivores, and top carnivores.

Question 33

Biodiversity is a key feature of a healthy estuarine ecosystem. This will include migratory birds, sea mammals and their young, and flatfish which are not shown on this food web. In what ways would bioaccumulation from either pollutants or biological toxins, eg. from algal blooms, affect these organisms? (Choose the best answer from A - E).

- A. The estuary will naturally clean water by trapping sediments and breaking down pollutants.
- B. Estuaries protect coastal areas by absorbing floodwaters and reducing erosion.
- C. Cities are commonly built around estuaries, making them vulnerable to pollution and degradation from human activity.
- D. Problem chemicals may be consumed by zooplankton, e.g via detritus, and then be concentrated through the food chain via planktivores and carnivorous fish. *
- E. Climate change is bringing more frequent heat waves to estuaries, threatening marine life.

Questions 34 - 36

One estuarine example is the Ōngātoto/Maketū Estuary in the Bay of Plenty, where the Kaituna River once flowed naturally. In the late 1950's the Kaituna River was diverted directly into the Pacific Ocean at the Western end of the estuary, creating the Kaituna River Mouth and preventing the river from flowing through the estuary and out through the Maketu Estuary Entrance. The purpose of cutting a new path for the river was to prevent flooding and to allow wetland areas to be drained for farming.

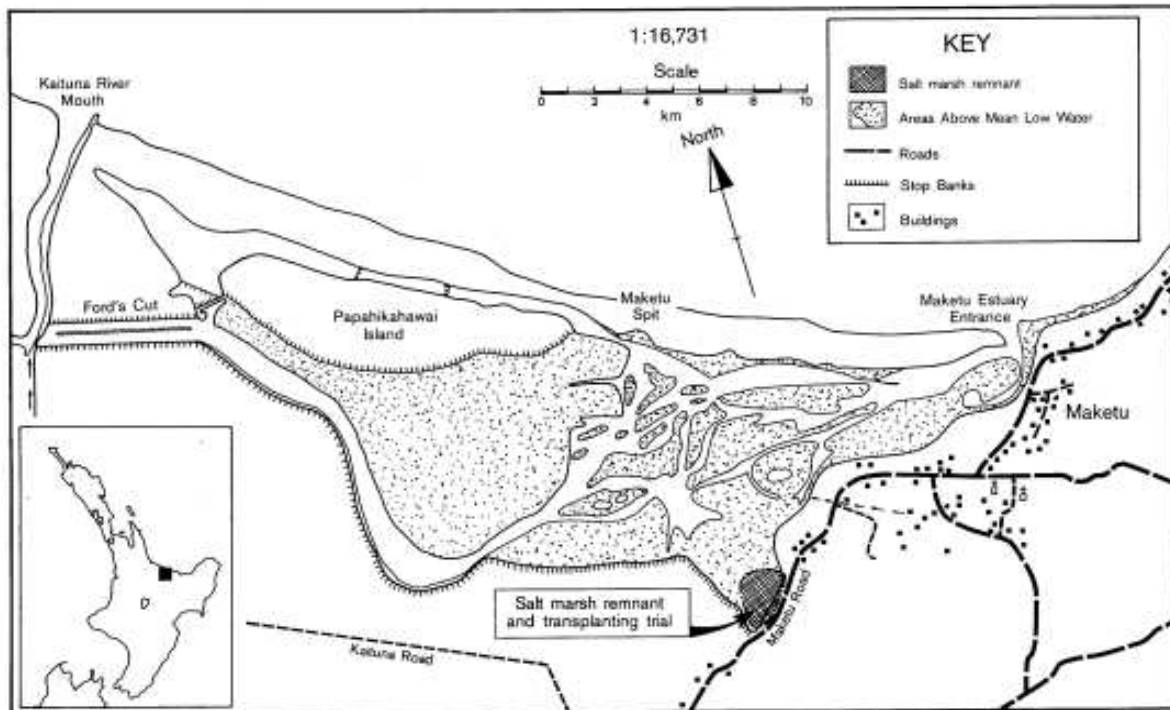
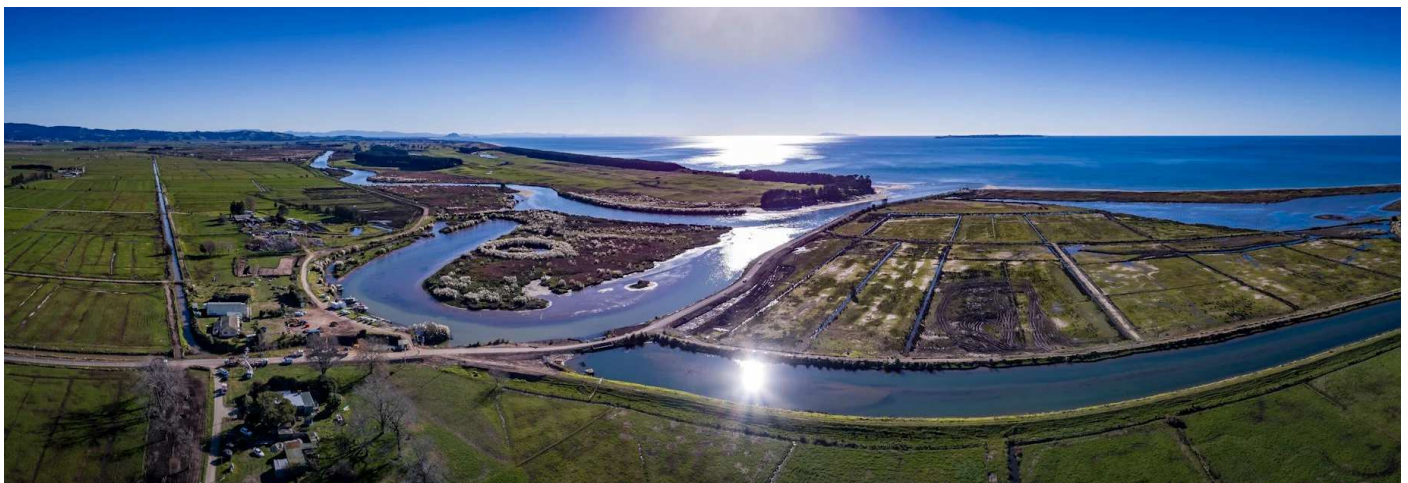


Figure 1: Location of trial area and main features of Maketu Estuary

Source: Bergin, D.O (1994). Performance of transplanted indigenous salt marsh species, Maketu Estuary. Indigenous Forest Management Section, Forest Technology Division, New Zealand Forest Research Institute, Rotorua.

This led to poor water quality, algae overgrowth, and damage to ecological and cultural values in the Ōngātoto/Maketū estuary.



This image shows Te Tumu Cut, which directs Kaituna River water to the sea rather than through the Ōngātoto/Maketū Estuary. Rights: Andy Belcher, Bay of Plenty Regional Council Toi Moana.

Source: Science Learning Hub

<https://www.sciencelearn.org.nz/resources/2700-working-together-to-restore-the-ongatoro-maketu-estuary>

Ford's Cut has now been reinstated to re-divert the Kaituna river flow via the estuary. Today, the Bay of Plenty Regional Council, alongside iwi, hapū, and local communities, is restoring the estuary with the re-diverted river flow, by replanting wetlands, and honouring the mauri of the land and water. Their shared vision is to protect the estuary as a taonga. This long process shows how co-governance and community collaboration can help restore and protect Aotearoa's 350 estuaries, which support biodiversity, kai moana, and natural defences against climate change.

Question 34

The initial diversion of the Kaituna River will have meant that the Maketū estuary was deprived of fresh water and nutrient in-flow. This would have led to:

- A. An increase in algal mass which, on death and decay, will increase biological oxygen demand in the seawater column.
- B. Increased anaerobic bacteria.
- C. A reduction of zooplankton biomass due to nutrient overload from decayed material.
- D. Loss of molluscs as zooplankton mass decreases.
- E. All of these things. *

Question 35

It was evident to iwi that there were profound effects on biodiversity after the diversion of the river to flow directly to the Pacific Ocean at Kaituna River mouth. Which of the effects (I - V) are likely to have occurred following the diversion of the river?

- I. Increased biodiversity as decomposers and anaerobic bacteria numbers increased.
- II. Increased biodiversity as fish and marine mammals were able to enter via both the Maketu estuary entrance and Kaituna River Mouth.
- III. Decreased biodiversity as invertebrate and vertebrate species were lost.
- IV. Decreased biodiversity as migratory birds, juvenile fish, and mammals were excluded due to a lack of food in the estuary.
- V. Increased biodiversity as a result of wetland replantings.

- A. All of the above.
- B. I - III only.
- C. III - IV only. *
- D. III - V only.
- E. None of the above.

Question 36

What factors were important in order to restore the mauri of the Ōngātoto/Maketū Estuary:

- A. Scientists and engineers worked within the participatory process with iwi to incorporate mātauranga Māori as well as cutting edge eDNA and statistical analysis tools.
- B. The waters of the Kaituna River were returned to the Ōngātoto/Maketū Estuary by increasing flow via Ford's Cut.
- C. Cultural and environmental considerations played an important part in the management and use of the land and the sea.
- D. Iwi, hapū, local government and other stakeholders agreed to a vision "to ensure that as a wider community - policies and plans, activities and actions celebrate and honour Kaituna River and Ongatoro/Maketū Estuary life as taonga – Whakanuia, whakamawawatia te mauri o te Kaituna me Ongatoro hei taonga".
- E. All of these things would contribute to the restoration of the mauri of the Ōngātoto/Maketū Estuary. *

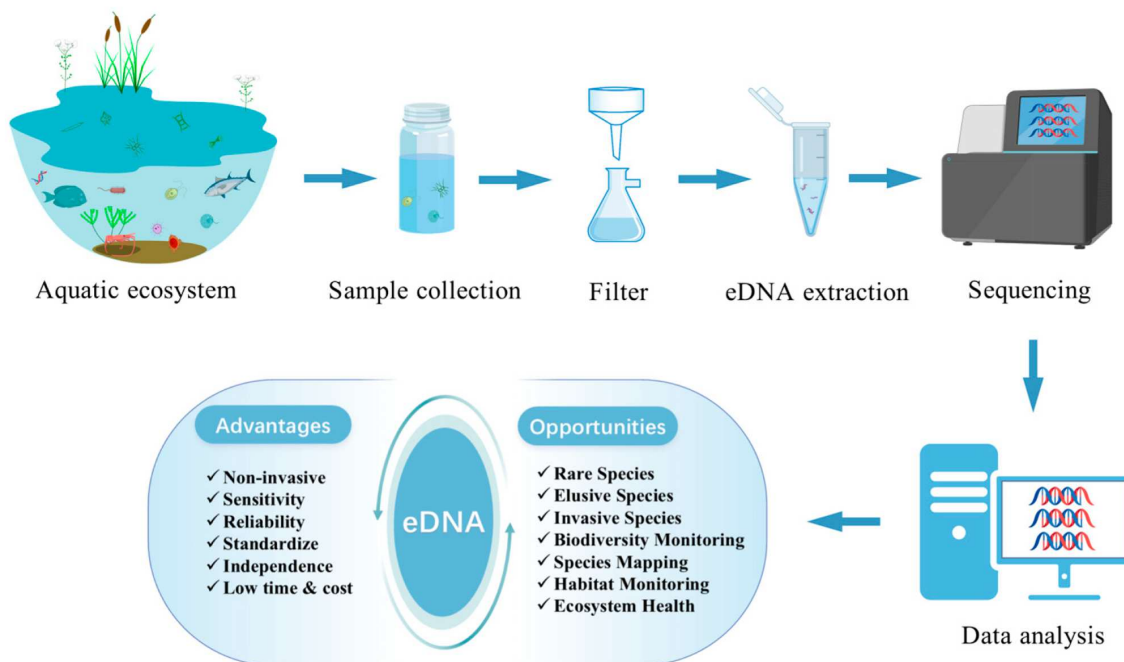
Dana Clark is a marine ecologist who leads the Restoration Ecology Team at the Cawthron Institute in Nelson. She studies how and why coastal environments change, so we can look after them better and help protect the ecosystem services they provide, like access to kai moana (seafood), recreation, and a healthy environment.



To do this, she uses cutting edge science in DNA analysis, biostatistics, and digital mapping to understand what's happening in estuaries and along the coast. Some of her recent projects include creating models to check how healthy estuaries are, using environmental DNA (eDNA) to monitor marine life, measuring the combined effects of human activities in Tauranga Harbour, and making maps that show where nature is providing useful services to people.

eDNA are the tiny traces of DNA that organisms like shellfish, worms, and bacteria leave behind in estuaries. Studying these traces is an important and cost-effective tool to detect changes in ecosystems. eDNA sampling can be faster and cheaper than traditional methods like catching fish or collecting macroinvertebrate samples. Dana's findings include identification of 100's of species in the estuary studied.

The diagram below shows how eDNA is used in biological research.



Source: Hang, H., Ye, T., Xie, Z., & Liu, X. (2025). Application of Environmental DNA in Aquatic Ecosystem Monitoring: Opportunities, Challenges and Prospects. *Water*, 17(5), 661. <https://doi.org/10.3390/w17050661>

Question 37

In order to successfully use eDNA Dana would need to:

- A. Be able to identify and classify hundreds of estuarine species.
- B. Have a robust data bank of the DNA of estuarine species to check the estuarine sample against. *
- C. Know the biomass of a wide range of estuarine species.
- D. Know the base sequences of a range of organisms.
- E. Use a control sample of eDNA from non-estuarine areas.

Question 38

Features of estuarine eDNA that could indicate poor ecosystem health may include:

- A. Evidence of eDNA from invasive species.
- B. eDNA linked to anthropogenic influences such as *E. coli* from wastewater, or from farm runoff.
- C. High levels of eDNA from anaerobes (organisms that do not require oxygen for growth).
- D. Decreasing levels of vertebrate eDNA.
- E. All of these things. *

Question 39

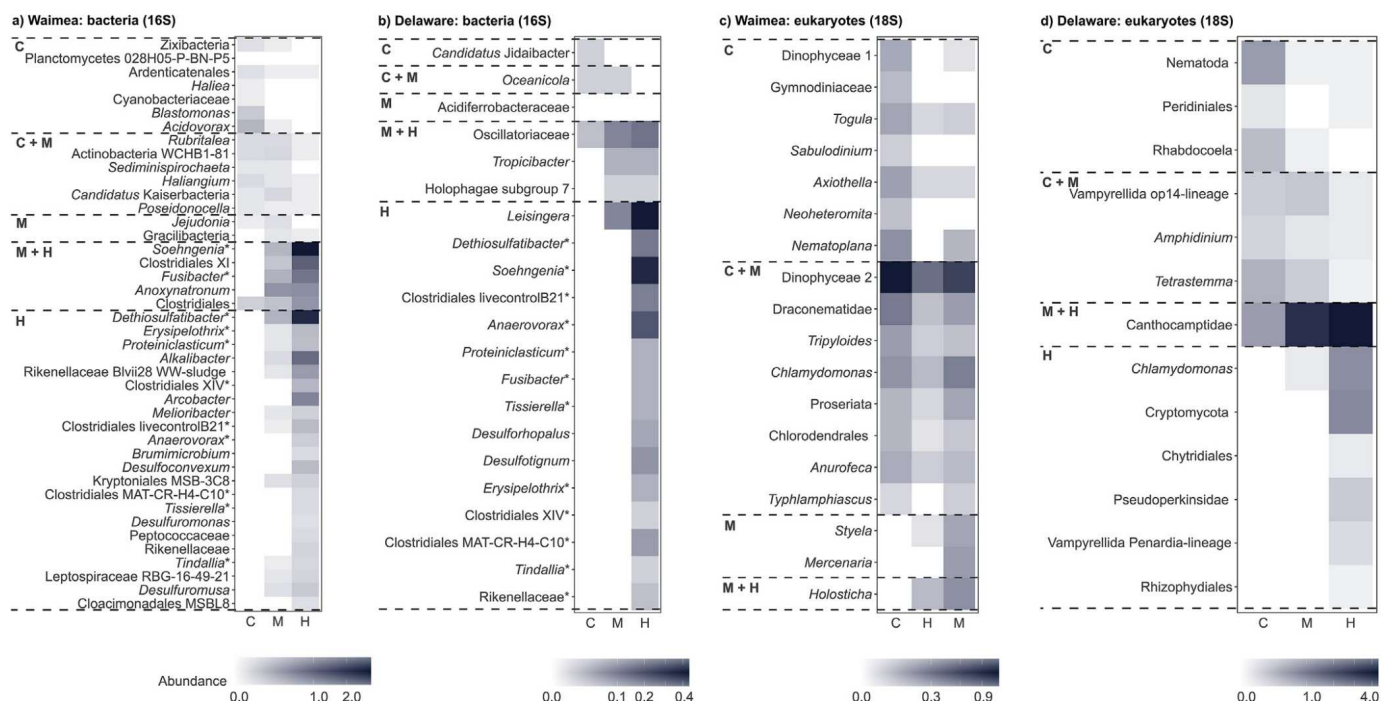
Use of eDNA in ecological studies is a game changer in conservation because:

- A. Detection of eDNA prevents the introduction of invasive pests and weeds.
- B. eDNA reduces the effect of anthropogenic change.
- C. Use of eDNA to test wastewater can be a way of assessing the amount of disease, such as Covid 19, there is in the community.
- D. eDNA assessment allows hundreds of species to be identified without the requirement for extensive taxonomic knowledge. *
- E. Use of eDNA does not require any scientists to visit the area to conduct ecosystem research.

It is critical to have good monitoring tools to detect early signs of environmental degradation in estuaries. In 2020, Dana's research group published the results of an in-situ experiment on the effects of nutrient enrichment on eukaryotic, diatom, and bacterial communities in two New Zealand estuaries.

In their study nutrient concentrations were manipulated by the addition of 0, 150, and 600 g nitrogen (N) fertiliser per m² in two estuaries in New Zealand to assess the effects of nutrient loading on benthic communities. After seven months of enrichment, environmental DNA (eDNA) metabarcoding was used to examine the response of eukaryotic (18S rRNA), diatom only (rbcL) and bacterial (16S rRNA) communities.

Some of these data are shown in the figure below of the average abundance of indicator taxa for bacterial (a, b) and eukaryotic (c, d) communities (including diatoms) across three nutrient enrichment treatments (C - control, M - medium, H - high) at sites in the Waimea and Delaware estuaries. An asterisk beside the name denotes indicator taxa shared by both sites.



The authors concluded that 31 eukaryotic, 4 diatom, and 52 bacterial taxa that were significantly ($p < 0.05$) associated with different nutrient enrichment treatments with both eukaryotic and bacterial communities showing a clear shift in response to nutrient loading.

Question 40

Which of the statements below provides evidence to support this statement?

- I. The bacterial taxon, *Tindallia*, was only present in the high nutrient treatments at Waimea and Delaware.
- II. Twelve bacterial indicator taxa associated with high nutrient enrichment were shared between sites.
- III. *Gymnodiniaceae*, a eukaryotic taxon, is only found in the control samples at Waimea.
- IV. The eukaryote group *Canthocamptidae* was present in higher abundances in the medium and high nutrient treatments at Delaware.
- V. All eukaryotic indicator taxa were site-specific.

- A. All of the statements, I – V.
- B. Only statement I.
- C. Statements I, III, and IV only. *
- D. Statements I, II, III, and IV only.
- E. Statements I and IV only.

Question 41

Indicate whether each of the following statements are true or false.

- A. Eukaryotic communities showed changes in response to nutrient enrichment at both sites despite differing environmental conditions. **TRUE**
- B. Clear shifts were seen in eukaryotic indicator taxa but not in bacterial indicator taxa because many bacterial taxa were found in both sites. **FALSE**
- C. Community changes were discernible (seen) between 0 and 150 g N/m² suggesting that eDNA estuary health tools could be developed to detect early signs of environmental disturbance and degradation. **TRUE**
- D. Similar changes were seen in the macrofaunal communities identified using traditional morphological techniques, confirming concordance between disturbance indicators detected by eDNA and current monitoring approaches. **TRUE**

THE KĀKĀPŌ RECOVERY PROGRAMME

The kākāpō is a taonga species. Te Rūnanga o Ngāi Tahu, in partnership with the Department of Conservation's Kākāpō Recovery Team, is working to realise a shared vision: restoring the mauri (life force) of the kākāpō and ensuring its long-term survival.



<i>Kākāpō</i>	Parrot of the night
<i>Mai a Rangi</i>	You came from Rangi
<i>Mai a</i>	You came from Mother Earth
<i>Papatūānuku</i>	You came from Tāne
<i>Mai a Tāne</i>	For the flowering of Rimu
<i>Ka puawai Rimu</i>	A sustenance for Kākāpō
<i>Ka hua a</i>	From this world
<i>Kākāpō</i>	To the world beyond We are
<i>Ki te Whai ao</i>	bound together in conserving
<i>Ki te Ao mārama</i>	nature
<i>Ka here tāngata</i>	We are bound together in
<i>Ka here whenua</i>	conserving land
<i>Ka tū te Pō</i>	Let night come
<i>Ka tū te Ao</i>	Let day come
<i>Thei mauri ora</i>	Such is life

Bringing the kākāpō back from the brink of extinction requires the application of cutting-edge biological science at every stage of conservation. The decline of the species was driven by the introduction of mammalian predators, cats, rats, and stoats, that are highly effective ground hunters. Kākāpō, being flightless, nocturnal, and evolutionarily adapted to a predator-free environment, are particularly vulnerable.

Unlike any other parrot, the kākāpō is quiet, well-camouflaged in mottled green plumage, and nests in burrows on the forest floor. When threatened, it instinctively freezes, an otherwise effective defence that fails against scenting predators. Kākāpō breeding is uniquely tied to the masting of rimu trees, which fruit only every few years, typically following a warm summer succeeded by a colder one. During these mast years, kākāpō breed and raise one to three chicks, feeding them nutrient-rich rimu fruit. Similarly, kākā breeding aligns with beech mast cycles, relying on the availability of beech seed.

Today, kākāpō conservation is a sophisticated endeavour. It combines traditional knowledge with modern tools such as genomic sequencing, reproductive technologies, bioacoustic monitoring, and population modelling. This collaborative, science-driven approach is helping one of Aotearoa's most iconic species make a slow but remarkable comeback.

Question 42

Which statement below may explain the evolution of many of the very “non-parrot” like attributes of a Kākāpō in Aotearoa?

- A. The introduction of rats and stoats resulted in the Kākāpō’s camouflage so that the introduced predators will not easily see them in the bush.
- B. Ample food is available on the ground for Kākāpō in Aotearoa bush so Kākāpō did not need to fly.
- C. Kākāpō nests on the ground because it uses less energy than building a nest in a tree.
- D. There was a lack of mammalian predators in early Aotearoa. *
- E. Freezing when confronted by rats and stoats means that these predators will not be able to detect the Kākāpō in the bush.

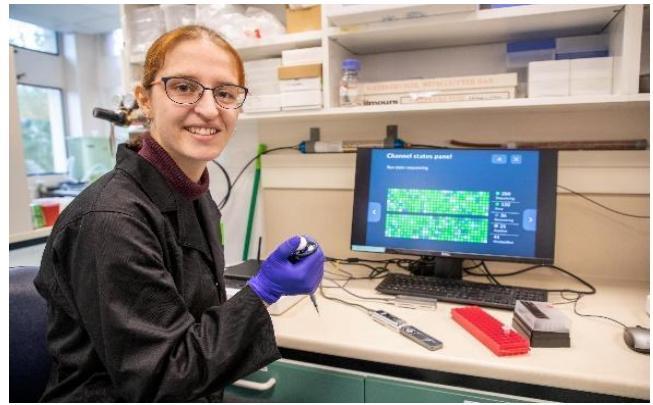
Question 43

From a biological point of view which of the following statements could be true?

- I. The Kākāpō strategy of freezing likely evolved when hunted by aerial predators such as the Haast’s Eagle – the biggest Eagle in the world.
- II. By utilising different foods for feeding chicks, today’s Kākāpō and Kākā will breed in different years when seed masts provide plentiful high nutrient food.
- III. The Kākāpō fills a niche somewhat similar to that of a rabbit in Europe.
- IV. The nocturnal habit of the Kākāpō make it likely that it is related to the Ruru, endemic Aotearoa owl.
- V. Kākāpō are likely endangered because of their difficulty finding mates in dense bush.

- A. Statements I. – V. are all true
- B. Only I., III., and V. are true.
- C. Only I., II., and III. are true. *
- D. Only II. and IV. are true.
- E. Only IV. and V. are true.

Marissa Le Lec gained her PhD at the Otago University Biochemistry Department, working with the Kākāpō Recovery Programme. Her research tracks parentage and genetic diversity in the critically endangered kākāpō using advanced DNA analysis techniques. After completing her Level 3 NCEA at Mt Aspiring College in Wanaka and an Honours degree at Otago, she now conducts both lab and field work, visiting remote offshore islands and presents her findings internationally.



Marissa developed a haplotype-based pedigree analysis method to monitor genetic diversity in small populations like the kākāpō. She has been tracking loss or change of genetic diversity over generations in relation to different management tools, modelling the effects of breeding interventions, such as artificial insemination, to ensure underrepresented individuals contribute genetic diversity to future generations. Because kākāpō breed infrequently, her work analysing multi-season data is vital to understanding long-term genetic trends.

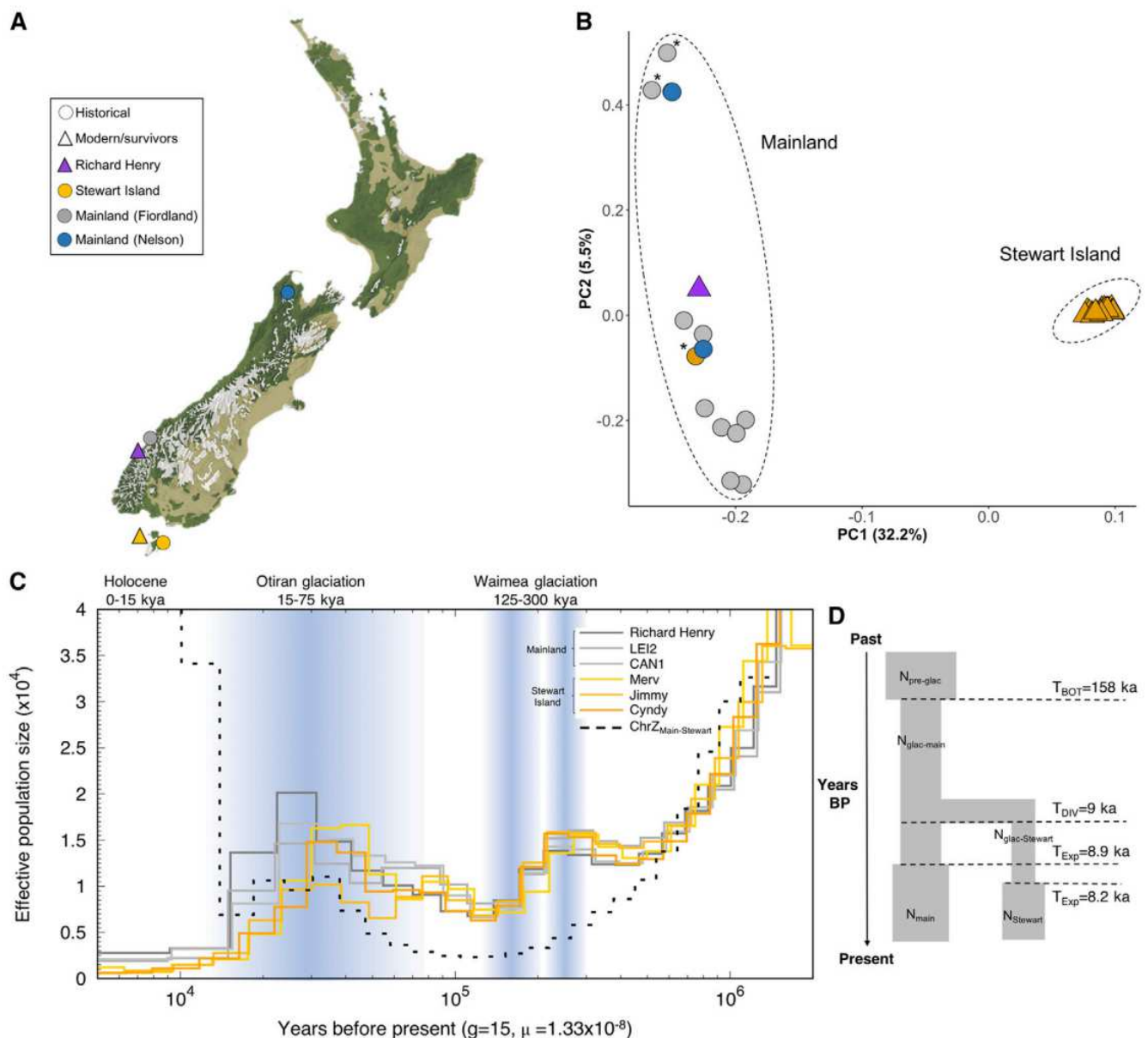
She also uses nanopore sequencing, a portable tool that quickly identifies paternity, sex, and disease status from blood samples in the field. This includes detecting *Aspergillus* fungus, responsible for a deadly 2019 outbreak that killed nine kākāpō on Pukenui Island.

Before human arrival, kākāpō were widespread across Aotearoa New Zealand. The population on Rakiura (Stewart Island) had likely been isolated for around 10,000 years and became genetically distinct. When the species was rediscovered there after being presumed extinct, just 50 birds remained, along with a single Fiordland male, Richard Henry. These individuals formed the basis of the current recovery effort, which has increased the population to over 200 birds on predator-free islands.

Genetic research has played a vital role in this recovery. Early work by Bruce Robertson's team at the University of Otago used DNA from museum specimens, fossils, and the surviving birds to reconstruct the species' genetic history. They found that the Rakiura birds, while inbred, carried relatively few harmful mutations, likely due to genetic drift and random allele loss rather than natural selection.

Marissa Le Lec has built on this research, refining our understanding of genetic drift, mutation load, and the genetic legacy of the founding population. Her work provides critical insight for conservation management, helping guide breeding strategies to maintain genetic diversity.

The figure below presents the DNA analysis of the original Rakiura founders and Richard Henry, the sole surviving Fiordland kākāpō.



(A) Sampling locations for historical and modern specimens on a map showing the vegetation cover circa 1840. (B) Principal-component analysis (PCA) for 14 mainland and 35 Stewart Island kākāpō. Asterisks indicate museum samples likely to have been mislabeled. (C) Demographic history and divergence time between the mainland and Stewart Island population inferred using the PSMC method. Each colored curve represents an individual bird. The black dashed curve represents the sex chromosome comparison (i.e., Z chromosome), with population size reaching infinity at the time of divergence between the two populations. (D) Parameter estimates for a scenario of post-glacial population divergence and expansion using ABC.

Question 44

What do Figures A and B suggest about kākāpō populations?

- A. There are significant genetic differences between birds from the mainland and those from Stewart Island. *
- B. Mainland and Stewart Island kākāpō are genetically unrelated.
- C. Aotearoa was once entirely forested.
- D. Richard Henry shows greater genetic similarity to the Stewart Island kākāpō than the fossil remains do.
- E. There are three distinct species of kākāpō.

Question 45

Why was it important that this data set was generated using kākāpō DNA from samples collected before the breeding programme began?

- A. It enabled scientists to identify the sex of birds before introducing them into the breeding programme.
- B. It helped scientists determine the parentage of the chicks born in recent years.
- C. It allowed researchers to assess the genetic diversity present in the original population from which today's birds have descended. *
- D. It meant kākāpō from different regions of Aotearoa could be moved to Rakiura to increase genetic diversity.
- E. It enabled tracking and removal of harmful mutations from the emerging gene pool during the breeding programme.

Question 46

This genetic data also provides insight into how a flightless bird like the kākāpō could have ended up isolated on an offshore island. Which of the following is the most likely explanation?

- A. Kākāpō flew to Rakiura 10^5 years ago and then evolved to become flightless on the island.
- B. The Otiran glaciation created a land bridge, allowing kākāpō to walk to the island. *
- C. The Waimea glaciation created a land bridge, enabling movement to offshore islands.
- D. The phylloclade shows Rakiura kākāpō became flightless 10,000 years before mainland populations.
- E. Kākāpō from Nelson crossed Cook Strait during a glaciation period.

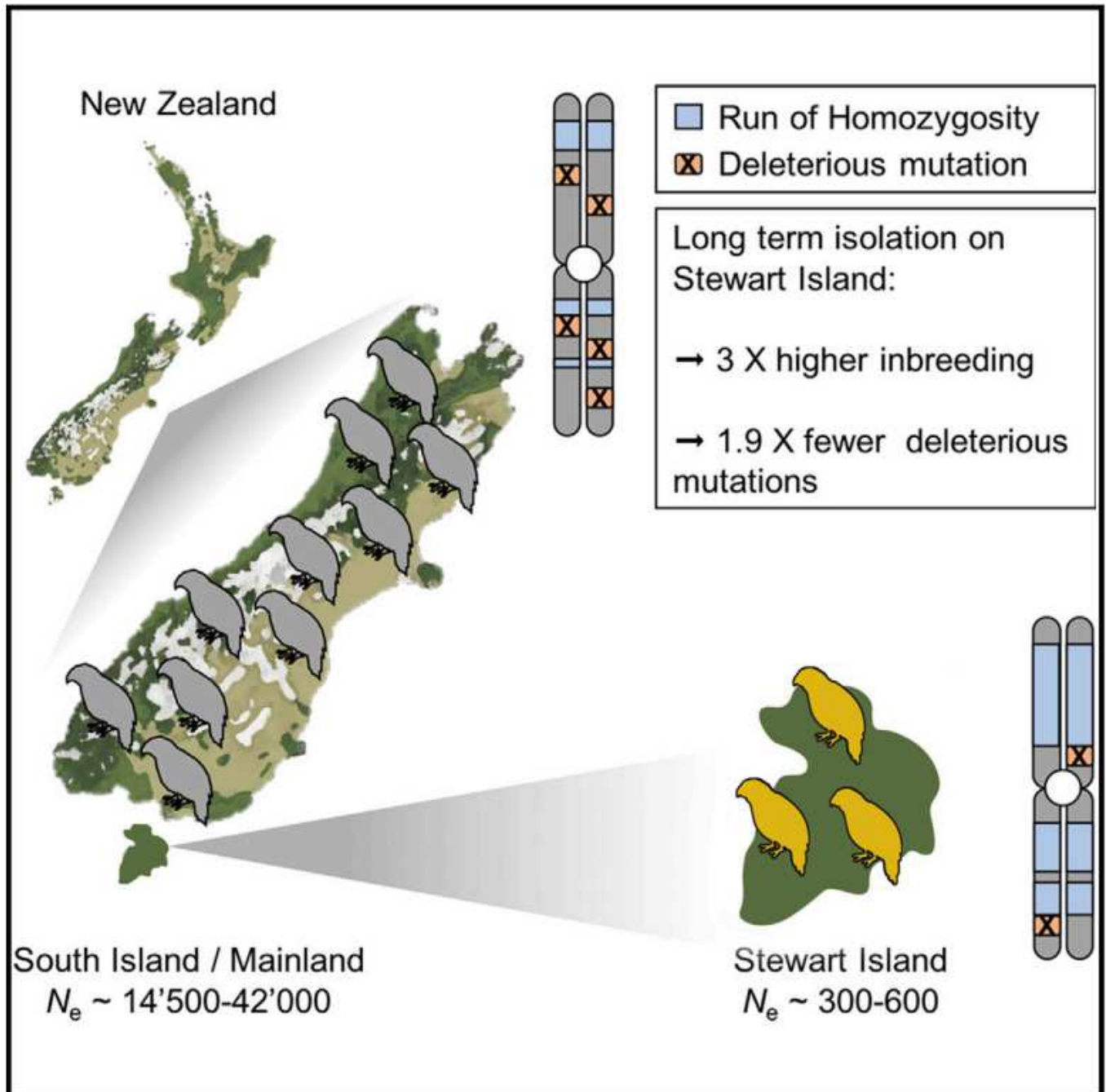
Question 47

It was once hypothesised that Māori or even early European settlers may have moved kākāpō from Fiordland to Rakiura. However, the data in Figure D suggests...

- A. This theory is supported, as the data shows the two populations diverged 9,000 years ago.
- B. This theory is supported, as the data shows two distinct demes present at the time of divergence.
- C. This theory is not supported, as the populations diverged 9,000 years ago, before humans arrived in Aotearoa. *
- D. This theory is not supported, as the bird populations actually converged 158,000 years ago.
- E. The theory is neither supported nor refuted, as DNA evidence cannot reveal movement patterns.

Genetic tools are invaluable in the conservation of endangered species. They help identify diseases present in the environment, maintain genetic diversity, and reduce inbreeding by avoiding pairings between closely related individuals, thereby limiting the spread of harmful traits. DNA analysis revealed that Richard Henry, a kākāpō from Fiordland, was genetically distinct from the remnant population, providing a valuable opportunity to introduce new genetic diversity to the remnant kākāpō population.

The infographic below graphically summarises this research on the genetics of the kākāpō.



Question 48

Conservation and restoration programmes aimed at saving endangered species often involve managing very small populations. This can result in several genetic consequences, including:

- I. Increased homozygosity of rare alleles.
- II. Reduced genetic diversity.
- III. Random loss of alleles due to chance rather than natural selection.
- IV. Increased inbreeding.
- V. A rise in the frequency of deleterious mutations.

In the case of the Kākāpō which of these consequences appears to have occurred?

- A. All of the above, I - V.
- B. I, II, III, and IV only. *
- C. I, III, and IV only.
- D. II, and IV only.
- E. IV and V only.

Question 49

How has the application of DNA sequencing most directly contributed to the success of the kākāpō conservation breeding programme in Aotearoa?

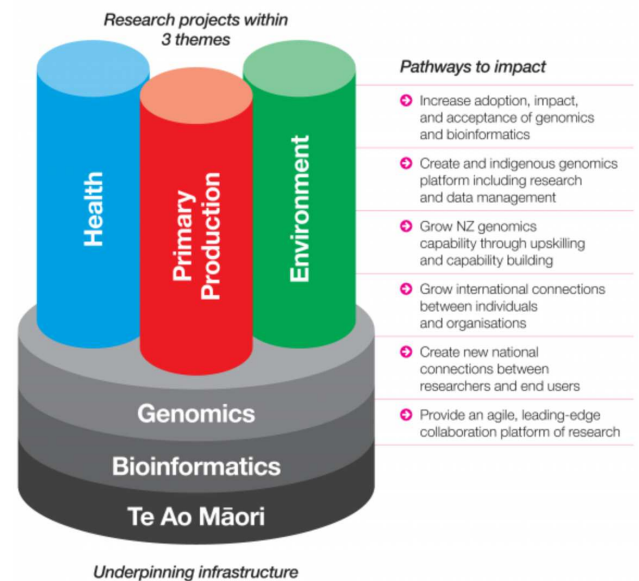
- A. By enabling the integration of fossil-derived genetic material into artificially inseminated individuals to enhance allelic diversity.
- B. By revealing historical flight patterns to offshore islands and supporting hypotheses regarding the evolution of flightlessness.
- C. By substantiating links between climate-driven seed masting events and fluctuations in reproductive output.
- D. By facilitating biosecurity measures to inhibit the transmission of *Aspergillus* spores within translocated island populations.
- E. By enabling precise determination of chick parentage to effectively manage inbreeding risk. *

Sources: Marissa Le Lec *pers com*. Miriam Sharpe Biochemistry Department University of Otago *Photography*, Dussex *et al.*, 2021, Cell Genomics 1, 100002. <https://doi.org/10.1016/j.xgen.2021.100002>

DON'T IGNORE THE 'OMICS

Genomics is the study of genomes, the complete set of genetic material (DNA) within a single cell or organism. In Aotearoa New Zealand, Genomics Aotearoa is a key player in ensuring the country actively contributes to and leads international research in this rapidly advancing field, unlocking a wide range of benefits. Its work is guided by three central themes: Health, Primary Production, and the Environment. These are supported by both technical and cultural foundations, comprising genomics, bioinformatics (the tools and methods used to analyse genomic data), and Te Ao Māori. Genomics Aotearoa also champions research that is “by, for, and with Māori”, and upholds Māori governance over indigenous genomic data.

Source: <https://www.genomics-aotearoa.org.nz>



Source:

<https://www.genomics-aotearoa.org.nz/our-work>

Genomics Aotearoa is focused on improving the reliability and quality of genome sequencing by enhancing existing technologies and bioinformatics tools, as well as developing new ones. High-quality genome sequences are essential for researchers to more accurately predict an organism's physical traits, behaviour, and susceptibility to disease, all of which influence both its economic value and its conservation status.

Rewarewa (*Knightia excelsa*) is a native taonga species, valued both for its cultural significance to Māori and its commercial importance to New Zealand's honey industry. Rewarewa honey, produced by bees that forage on its flowers, is known for its dark, malty, and complex flavours with a sweet finish. Researchers from the University of Otago and Manaaki Whenua – Landcare Research have collaborated to produce a high-quality reference genome for rewarewa. They also developed tools to evaluate genome assembly quality for future projects, ensuring that iwi and hapū consent was sought and respected throughout the research process.

Source: <https://pmc.ncbi.nlm.nih.gov/articles/PMC8362059>

Question 50

Why is it important to give greater consideration to Māori in genomics research?

- Initiatives that support Māori involvement in research, particularly in areas connected to Te Ao Māori, help ensure that the benefits of genomics reach Māori communities.
- Genomic data gathered from Māori individuals or culturally significant species must be managed in ways that uphold Māori values and respect tikanga.
- Māori are often underrepresented in international genomic datasets, meaning that emerging medical technologies based on those datasets may be less effective for Māori.
- Integrating mātauranga Māori (traditional knowledge) into genomics research can generate new research pathways and offer insights into existing challenges.
- All of the above are valid reasons for ensuring Māori are meaningfully considered in genomics research. *

Question 51

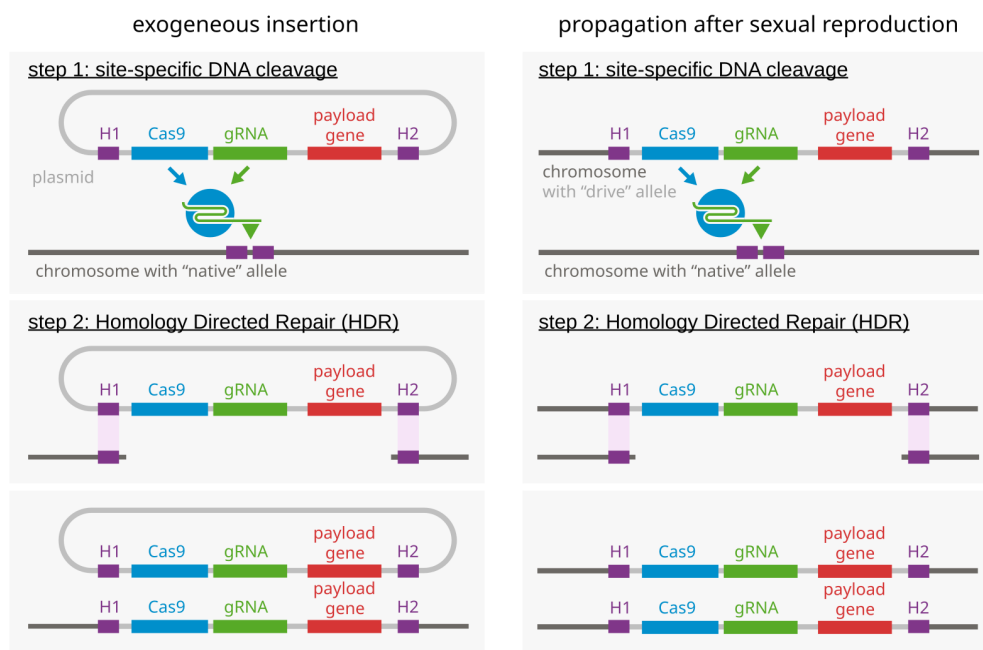
This research produced a reference genome by *de novo* sequencing, which is performed when there is no reference sequence for that species against which other sequences can be compared. Why might reference sequences be important in genomics?

- A. To eliminate the need for genome sequencing.
- B. To ensure that all sequences subsequently collected by later researchers from other species are correct.
- C. To act as a baseline from which differences between individuals or species can be identified. *
- D. To allow researchers to directly analyse mutations without needing to make comparisons.
- E. To predict the function of all of an organism's genes.

Similar genome assemblies have been produced for invasive social wasps, including the common wasp (*Vespula vulgaris*), which is an introduced pest in New Zealand. The wasp is an example of a haplodiploid insect, where males have a single set of chromosomes and are produced from unfertilised eggs, whereas females have a full set of chromosomes and develop from fertilised eggs.

To control these wasps, pesticides and herbicides are costly and frequently environmentally-damaging methods of pest control and elimination are used. Genome editing has been investigated as a potential alternative. Researchers, with funding from Genomics Aotearoa, assessed the potential of artificial 'gene drives' using CRISPR-Cas9 gene editing technology as an alternative.

A CRISPR gene drive is a genetic engineering technique that uses the CRISPR-Cas9 system to bias inheritance. Normally, an organism has a 50% chance of passing on any given allele (gene variant) to its offspring. But with a gene drive, the CRISPR-Cas9 system cuts the DNA of the non-modified allele in the offspring. The cell then repairs this cut using the modified allele as a template, essentially copying the desired gene onto both chromosomes so the organism is homozygous (has two identical alleles) for that gene (heterozygous refers to having two different alleles for the same gene). This means nearly all offspring inherit the modified allele, allowing the trait to spread rapidly through a population, even if it doesn't offer a survival advantage.



Source: https://commons.wikimedia.org/wiki/File:Molecular_mechanism_of_gene_drive.svg

Question 52

How might gene drives be used to artificially alter inheritance?

- A. Genes are cut out of DNA and the remaining pieces are stuck back together.
- B. The CRISPR-Cas9 system is inserted into an organism's DNA. *
- C. Gene drives cause DNA to coil up very tightly so that it is inaccessible to enzymes that read it.
- D. The CRISPR-Cas9 system is removed from an organism's DNA.
- E. None of the above (A–D) are correct.

Question 53

A key advantage of the gene investigated by researchers for this gene drive is that it makes the process of eliminating *V. vulgaris* self-propagating. Which gene target might researchers choose to remove?

- A. Genes coding for spermatogenesis (producing sperm). *
- B. Genes coding for olfactory receptors (smell detecting cells).
- C. Genes coding for photoreceptors (light detecting cells).
- D. Genes coding for aerobic respiration (production of glucose using oxygen).
- E. Genes coding for wing muscles.

Question 54

What is the likely consequence of the probability of inheritance for the modified gene increasing above 50%?

- A. The modified DNA is likely to be eliminated from the population, so *V. vulgaris* will be eliminated.
- B. The modified DNA is likely to be eliminated from the population because heterozygotes will be converted to homozygotes.
- C. The modified DNA is likely to be propagated in the population, so fewer *V. vulgaris* will have it.
- D. The modified DNA is likely to be propagated in the population because heterozygotes will be converted to homozygotes. *
- E. The modified DNA is not likely to have any effect because individuals expressing it are not viable.

Question 55

All ecosystem interventions, especially those designed for direct human benefit, must consider both potential harms and benefits. Which of the following (I - V) are possible risks associated with using CRISPR gene drives to control *Vespula vulgaris* populations in Aotearoa New Zealand?

- I. *V. vulgaris* may be driven to extinction within New Zealand.
- II. Other wasps may increase in number.
- III. Genetic variation within *V. vulgaris* populations could reduce the gene drive's effectiveness.
- IV. The gene drive could unintentionally increase the evolutionary fitness of *V. vulgaris*, leading to a population increase.
- V. Modified *V. vulgaris* could spread beyond New Zealand, potentially causing global extinction of the species.

- A. I, II, III, IV and V.
- B. II, III, and V only. *
- C. I, II, and IV only.
- D. III and V only.
- E. III only.

Question 56

One of the key concerns in developing gene drives is ensuring they are highly specific to the target species. In this case, researchers assessed whether CRISPR-Cas9 modified *Vespula vulgaris* (common wasp) could unintentionally affect other species, especially through hybridisation.

They used a bioinformatic analysis to compare five designed sgRNA sequences (targeting the *boule1*, *ocnus*, and *sdic* spermatogenesis genes) against genomes of closely related species (*V. germanica*, *V. pensylvanica*), a paper wasp (*Polistes dominula*), and two bee species, the bumble bee (*Bombus terrestris*) and the honey bee (*Apis mellifera*). Their data is summarised in the table below.

Insect		Gene region and sgRNA version				
		<i>boule v1</i>	<i>ocnus v1</i>	<i>ocnus v2</i>	<i>sdic v1</i>	<i>sdic v2</i>
<i>Vespula vulgaris</i>	Equivalent gene found?	✓	✓	✓	✓	✓
(Common wasp)	Gene cleaved using sgRNA?	✓	✓*	✓	✓	✓*
<i>Vespula germanica</i>	Equivalent gene found?	✓	✓	✓	✓	✓
(German wasp)	Gene cleaved using sgRNA?	✓	✗	✓	✗	✓
<i>Vespula pensylvanica</i>	Equivalent gene found?	✓	✓	✓	✓	✓
(Western yellowjacket)	Gene cleaved using sgRNA?	✓	✗	✓	✗	✓
<i>Polistes dominula</i>	Equivalent gene found?	✓	✗	✗	✓	✓
(European paper wasp)	Gene cleaved using sgRNA?	✗	✗	✗	✗	✗
<i>Bombus terrestris</i>	Equivalent gene found?	✓	✗	✗	✓	✓
(Buff-tailed bumble bee)	Gene cleaved using sgRNA?	✗	✗	✗	✗	✗
<i>Apis mellifera</i>	Equivalent gene found	✗	✗	✗	✓	✓
(Western honey bee)	Gene cleaved using sgRNA	✗	✗	✗	✗	✗

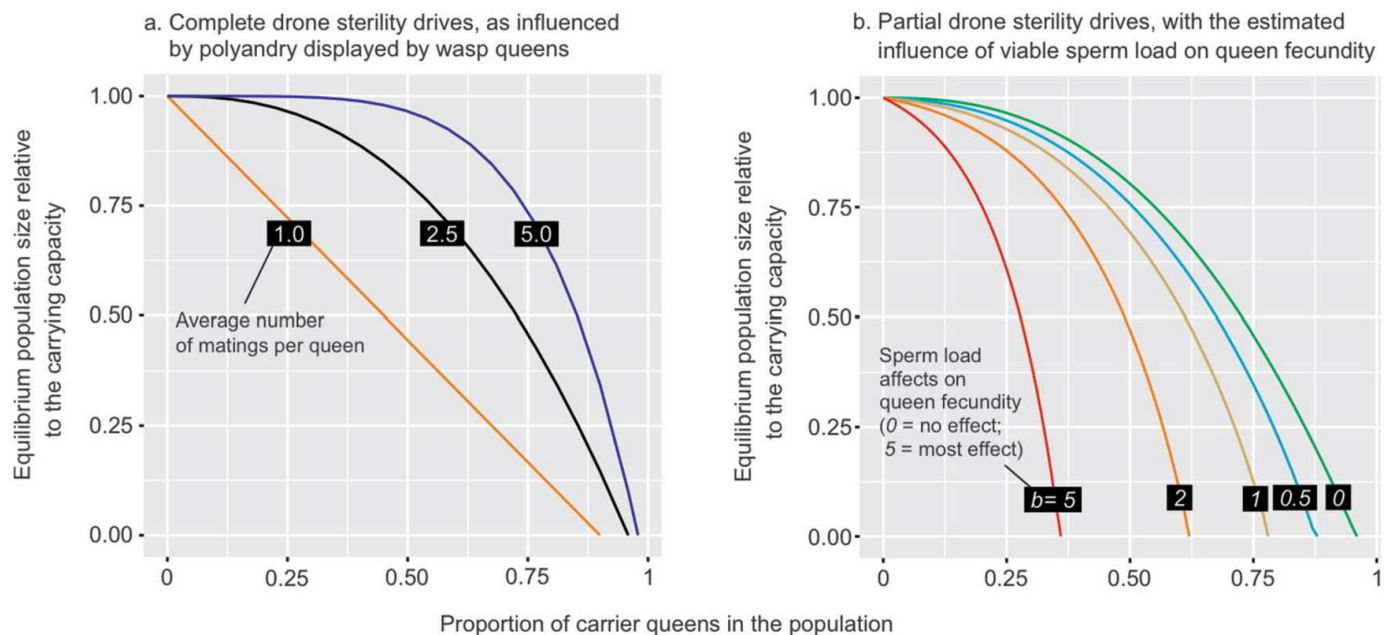
For each of the following statements, indicate whether it is True or False.

- Two sgRNAs for *boule 1*, one for *sdic*, and one for *ocnus* were found to have identical target sequences across all three *Vespula* species suggesting potential cross-species effects within the *Vespula* genus of a gene drive targeted these genes. **TRUE**
- The sgRNA designed to target the indel in the *sdic* gene was highly specific and did not match any other genome examined, including other *Vespula* species. **FALSE**
- sdic v1* is an appropriate target for a gene drive to manage the pest populations of the common wasp as it is unlikely to unintentionally affect other species of wasps or bees. **TRUE**
- V. vulgaris* is closely related to other *Vespula* species, but more distantly related to paper wasps, bumble bees and honey bees. **TRUE**

Question 57

Gene drives that cause partial male (drone) sterility may reduce wasp populations more effectively than those causing complete sterility. This is because allowing some sterile males to breed enables the gene drive to spread before reducing the population, provided the homing rate is high enough. The extent of population reduction from partial sterility depends on how many males each queen mates with (polyandry) and how relative sperm load affects queen reproduction.

The graphs below present data on the effect on population size of the proportion of queens in the population that carry a genetic modification for complete or partial drone sterility. (a) as influenced by the degree of polyandry displayed by the queens and (b) as influenced by the viable sperm load on queen fecundity..



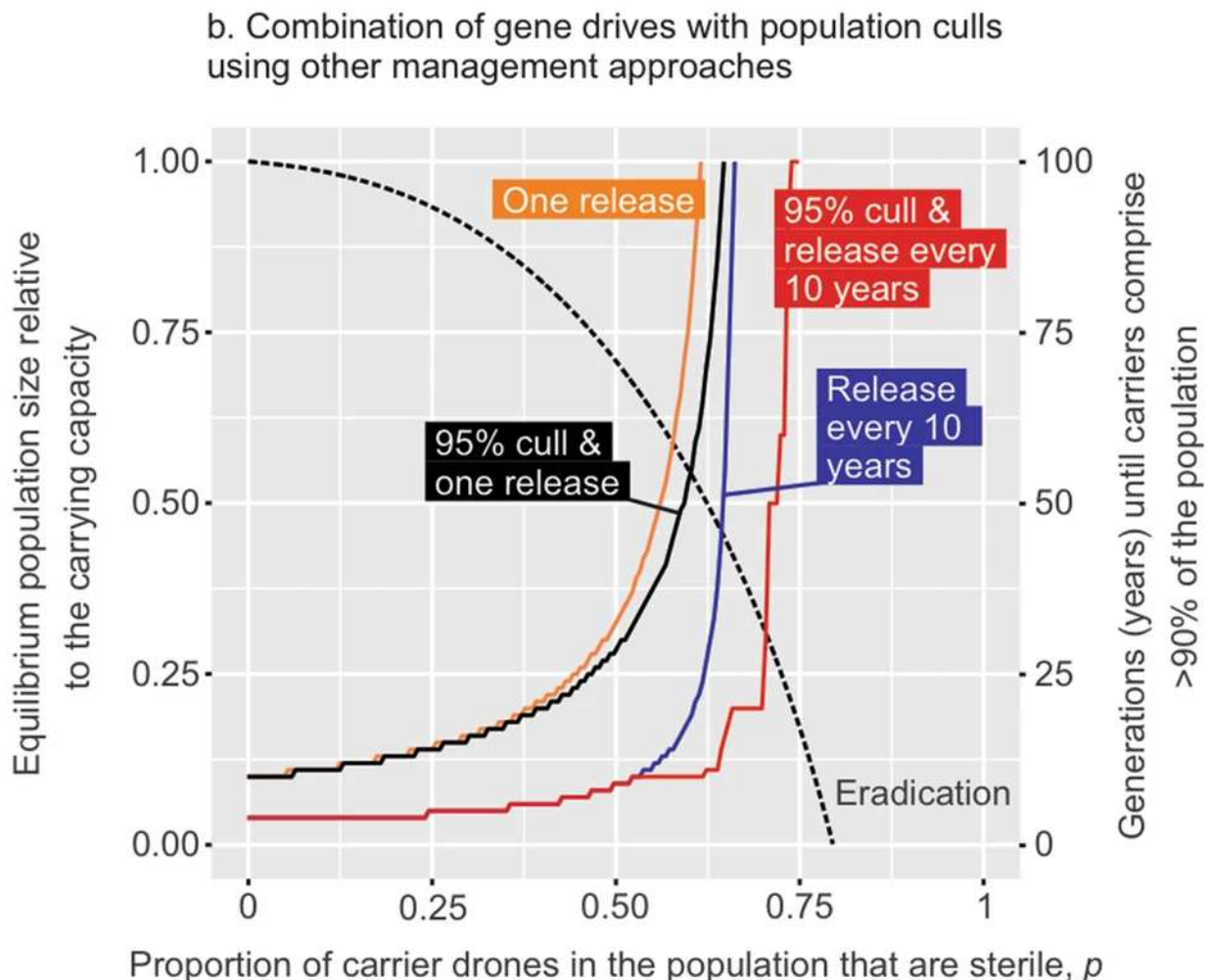
Source: <https://www.nature.com/articles/s41598-020-69259-6>

Which of the following statements about gene drives causing drone sterility in *Vespula* wasps is **NOT** correct?

- A. A higher degree of polyandry means more queens must carry the gene drive to achieve the same level of population suppression.
- B. In New Zealand, queen wasps mate with between 1 and 5 males.
- C. The volume of viable sperm collected by queens has no impact on nest size or fecundity. *
- D. When viable sperm load has a strong effect on queen fecundity, fewer gene drive-carrying queens are needed to reduce population size.
- E. Gene drives causing partial sterility can be effective even when not all queens are carriers, especially if sperm load limits fecundity.

Question 58

Gene spread can be facilitated by other management interventions. In the graph below a gene drive with $h = 80\%$ homing is released once (orange line), once immediately following a 95% cull (black line), every 10 years (blue line), or every ten years after a 95% cull (red line).



Considering all the data about gene-drives in wasps, determine whether each statement (A-D) is true or false.

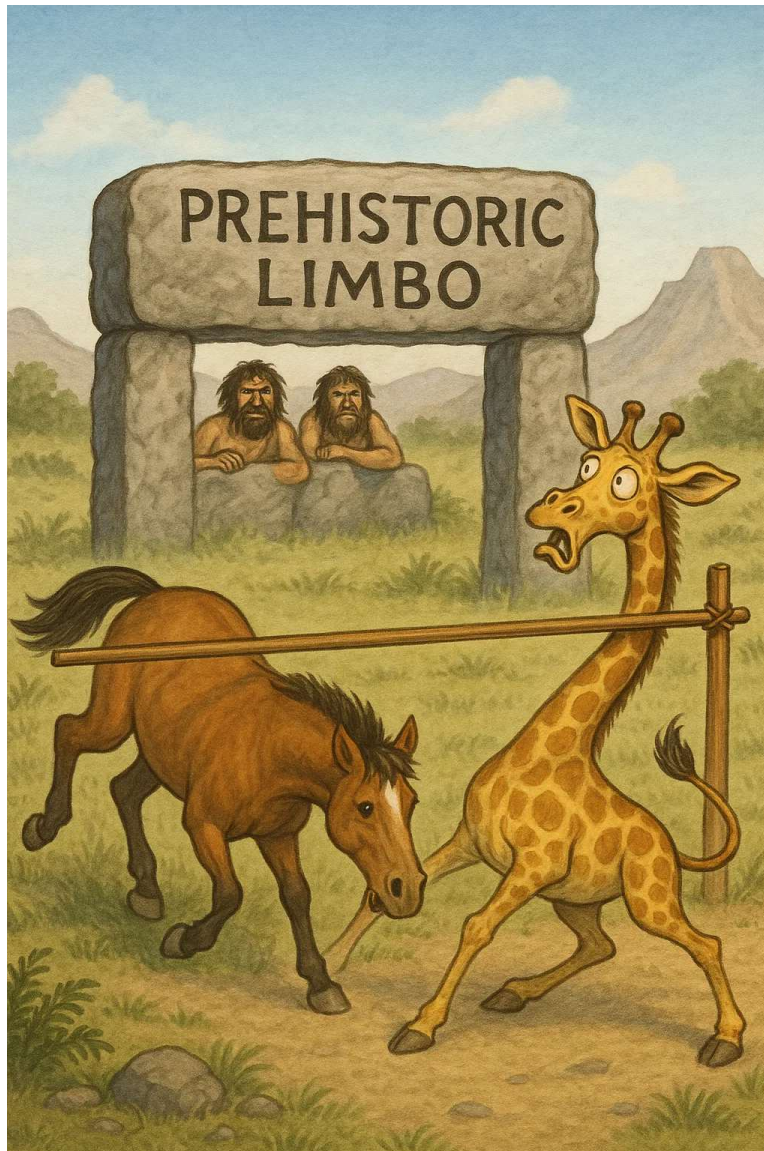
- A.** Releasing gene drive-carrying queens into a population that has been temporarily reduced may help overcome the gene drive's limited ability to spread. **TRUE**
- B.** A 95% effective poisoning event, repeated every ten years, is sufficient on its own to cause long-term wasp population decline. **FALSE**
- C.** Achieving extinction with a drone sterility gene drive requires reaching a critical threshold where the sterilising effect outweighs the wasps' reproductive capacity. **TRUE**
- D.** Model predictions suggest that population eradication can occur even if fewer than half of the queens carry the gene drive allele. **FALSE**

Question 59

Which of the following best explains why genomics is considered a powerful tool across conservation biology, horticulture, and human health?

- A. It allows researchers to clone endangered species, ensuring their immediate survival.
- B. It provides rapid methods for modifying plant species to produce genetically identical varieties.
- C. It is mainly focused on studying the evolutionary history of extinct organisms.
- D. It enables precise identification of genetic variation, supporting species conservation, crop improvement, and personalised medicine. *
- E. It is only useful when working with organisms that have already had their entire genome sequenced.

Question 60



Source: ChatGPT

Why do giraffes have such long necks?

- A. They were tired of awkward conversations at ground level.
- B. To reach the Wi-Fi signal from treetops.
- C. Natural selection favoured those that could reach higher foliage. *
- D. To win prehistoric limbo competitions (they were terrible at it).
- E. Because their ancestors watched too many stretching tutorials on YouTube.

We all have an obligation to learn about our planet and to protect it from harm. NZIBO hopes you have enjoyed this exam and have learnt a little about the biological scientists working to understand and protect our wonderful planet here in New Zealand and on the global stage. We wish you well with your biology studies this year, regardless of whether you are selected for the tutorial programme. Thank you for taking the time to sit this exam.