ADVANCED LEVEL BIOLOGY EXAMINATION (SET BY THE AUTHOR OF C I B TEXTBOOKS). FREE FOR EVERYONE

Item 1

A 65yr old patient experiences sudden, tearing chest pain. Dr. Gideon Alexander diagnosed the patient with an aortic dissection, where a tear in the intima (inner lining) causes blood to force its way between the layers of the vessel wall, creating a false lumen. Dr. Gideon Alexander had previously measured the aorta diameter (d) in a large population of people over 60 years of age. He also calculated the risk of an aorta wall developing a tear.

Table below shows his results.

Aorta diameter in d / cm	**Number of people in the population**	**Risk of an aorta wall developing a tear**
$d \leq 3.5$	2765	0.06
$3.5 < d \leq 4.0$	630	0.33
$4.0 < d \leq 4.5$	98	4.64
$d > 4.5$	7	380.00

Dr. Gideon Alexander briefly told the 60 year old patient that blood may push through the tears in the aorta wall. This produces a balloon-like swelling called an aneurysm and increases the aorta diameter. Aneurysms can cause the aorta to burst. This was after the patient engaged him on how aorta could have a balloon-like swelling and creating a false lumen.

Tasks with their responses

(a) Identify the three layers of the aorta and explain which layer is primarily failing in this scenario.

The layers are the intima (innermost), media (middle, elastic), and adventitia (outermost). Aortic dissection begins with a tear in the intima, allowing blood to surge into the media layer, tearing it away from the inner wall.

(b) Explain why a "false lumen" created by a dissection can lead to a drastic reduction in oxygen delivery to tissues.

The false lumen fills with blood, creating pressure that can compress the "true lumen" (where blood should flow). This reduces the effective diameter of the vessel, cutting off blood flow (ischemia) to downstream branches supplying organs like the kidneys or brain.

(c) Using all the information in the table above, what can you conclude about aorta diameter and the risk of developing an aneurysm?

Small diameters are low risk of tears so unlikely to have aneurysms. As diameter increases, risk of tears increases and risk of developing aneurysms also increase. Few people have high risk of tear. Or...Few people have high risk of aneurysms. Diameters > / above 4.5 (cm) are at high risk of tears so may have aneurysms. High risk of tear does not mean aneurysm will occur.

Item 2

Professor Gideon Alexander, a lab technician at Uganda Virus Research Institute in Entebbe is investigating the impact of a newly discovered metabolic poison on isolated liver mitochondria. Upon adding the

poison, researchers observe that oxygen consumption ceases immediately, and NADH levels in the matrix increase dramatically.

Tasks with their responses

(a) What is the consequence for ATP production?

The poison is likely inhibiting Cytochrome C Oxidase (Complex IV) or another intermediate complex like Complex I. It inhibits it by binding to the site where electrons are transferred to oxygen, preventing the reduction of oxygen to water.

(b) What is the consequence for ATP production?

Oxidative phosphorylation stops. Because oxygen is the final electron acceptor, if it is not reduced, the ETC halts, the proton gradient fails, and ATP synthase stops producing ATP.

(c) Suggest a possible solution to restore some ATP production within the cell.

The cell will have to rely solely on glycolysis for ATP production. To make this sustainable, the cell needs a method to regenerate NAD^+ from NADH without the ETC, which usually occurs via lactate fermentation in animals or alcoholic fermentation in yeast.

Item 3

A patient is brought to the emergency room after consuming Dinitrophenol (DNP), a chemical used in illegal diet pills. DNP is an "uncoupler" that makes the mitochondrial inner membrane leaky to protons (H^+). Meanwhile, researchers are investigating the role of succinate dehydrogenase in the Krebs cycle. They introduce malonate, a

compound that closely resembles the structure of succinate, into the mitochondrial preparation.

Tasks with their responses

(a) (i) Explain why DNP causes a rapid rise in body temperature and a decrease in ATP production despite an increased oxygen consumption rate.

DNP allows protons to flow back into the matrix without passing through ATP synthase. The energy released by the ETC is therefore released as **heat** instead of being used to produce ATP. Because the proton gradient is dissipated, the ETC works faster to try to restore it, consuming more oxygen.

(ii) What is the main danger to the cell's survival?

The cell fails to produce sufficient ATP for essential, active processes, leading to cell death, despite having high rates of respiration.

(iii) What is a possible clinical solution to treat this poisoning?

Immediate **cooling measures** (ice baths) to lower body temperature and, if possible, administering a compound that can restore the proton gradient (though this is difficult) or supporting anaerobic metabolism.

(b) (i) Explain the effect of malonate on the respiration rate.

Malonate acts as a competitive inhibitor. It binds to the active site of succinate dehydrogenase, preventing the conversion of succinate to fumarate. This stops the Krebs cycle from functioning, reducing

the production of NADH/FADH, and slowing down the entire respiratory chain.

(ii) Based on your knowledge of enzyme kinetics, what is a possible solution to overcome this inhibition?

Increase the concentration of the substrate (succinate). If more succinate is added, the probability of the enzyme binding to the substrate rather than the inhibitor increases, thus restoring the rate of reaction.

Item 4

A 65-year-old patient presents with fatigue and breathlessness. An echocardiogram reveals that her left atrioventricular valve (mitral valve) does not close properly during ventricular systole, allowing blood to leak back into the left atrium. Dr. Gideon Alexander investigated changes in this diseased heart and changes in his heart (a healthy heart) during cardiac cycles. For each heart, he obtained a value for: The mean blood volume in a full ventricle just before the ventricle contracts (BVB). The mean ejection fraction (EF). The EF is the proportion of blood pumped out of a full ventricle in one heartbeat.

The EF is calculated using this formula:

$$\text{EF} = \frac{\text{Blood volume pumped out of a full ventricle in one heartbeat (stroke volume)}}{\text{BVB}}$$

Table below shows Dr. Gideon Alexander's results.

	Diseased heart	**Healthy heart**
Mean BVB in cm^3	100	120
Mean EF	0.45	0.58

Tasks with their responses

(a) Using the equation and the Table above, calculate the percentage change in the stroke volume of the diseased heart compared with the stroke volume of his healthy heart. (**Please attempt this task**).

(b) (i) Explain how the echocardiogram result affects the pressure in the left atrium during ventricular systole.

Normally, during ventricular systole, the mitral valve closes, and left atrial pressure is low. In this case, blood leaks back, causing a **significant increase in left atrial pressure** during ventricular systole, leading to atrial congestion.

(ii) Describe the consequence of this on cardiac output and the flow of blood into the aorta.

Cardiac output will **decrease**. Because some blood flows back into the atrium, less blood is pushed into the aorta during ventricular systole, reducing stroke volume.

(iii) Suggest a potential surgical solution to this problem.

Surgery to replace or repair the mitral valve, such as a synthetic valve replacement.

Item 5

An ECG of a patient shows that P-waves (atrial contraction) are normal, but QRS complexes (ventricular contraction) are slow and irregular. There is no coordination between the atria and the ventricles. The SAN is functioning, but the signal is not reaching the ventricles.

Tasks with their responses

(a) Identify which part of the conduction system is likely damaged.

The **Atrioventricular Node (AVN)** or the Bundle of His/Purkyne tissue.

(b) Explain why the ventricles are still contracting, even if it is slow.

The ventricles have inherent, slower pacemaker cells that can take over (idioventricular rhythm) when the main signal from the AVN fails, though this is inefficient.

(c) What is the most immediate solution to stabilize this heart rhythm?

Fitting an **artificial pacemaker** to initiate ventricular contractions.

Item 6

A 55-year-old male, smoker, with a high-fat diet, presents with severe chest pain (angina). Diagnostic imaging shows a 90% blockage in the left coronary artery caused by a fibrous plaque, and a blood clot has formed at the site of the plaque.

Tasks with their responses

(a) Describe how atherosclerosis leads to this blockage.

Damage to the endothelial lining of the artery (often due to high blood pressure/smoking) allows Low-Density Lipoprotein (LDL) cholesterol to accumulate, leading to inflammatory responses and

white blood cells forming an atheroma (accumulation of fatty, porridge-like material known as plaque composed of lipids, calcium, and cellular debris within the inner layer of artery walls). Calcium and fibrous tissue accumulate, hardening the plaque (plaque formation) and narrowing the lumen.

(b) Explain why a blood clot (thrombosis) at the site of the plaque causes a "heart attack" (myocardial infarction).

The blood clot completely blocks the coronary artery, cutting off the supply of oxygenated blood to the heart muscle. Without oxygen, heart muscle cells cannot respire aerobically, leading to the death of cells (myocardial infarction) due to lack of ATP and anaerobic respiration.

(c) Identify two possible medical interventions to resolve this issue and explain how they work.

(1) **Stents:** A balloon catheter is inserted to push the plaque against the artery wall and a mesh tube (stent) is left to keep the artery open, restoring blood flow. (2) **Bypass surgery:** A blood vessel from another part of the body is used to create a new path around the blockage, ensuring oxygenated blood reaches the myocardium (the thick, involuntary, and striated middle muscular layer of the heart wall, situated between the inner endocardium and outer epicardium).

Item 7

Bandi nowadays a fish dealer from Hoima City imports "live" clownfish for the aquarium trade from illegal cyanide fishing dealers in Southeast Asia to rich clients living in East Africa, especially in Kampala, Uganda. However, he finds that 75% of the shipment dies within 48 hours,

despite the fish looking healthy upon arrival. Dr. Gideon, a veterinarian did inspection and suggested cyanide poisoning to the already disappointed Bandi, even though the fish were "rinsed" for ten days before shipping. Dr. Gideon collected tissue samples from the infected clownfish and unaffected fish from Tanzania.

Table below shows Dr. Gideon's metabolic analysis of clownfish gill tissues

Clownfish samples	Cytochrome C oxidase activity in % age normal	Gill cell membrane integrity in %age of damaged cells	Gill ATP concentration in millimolar (mM)	Oxygen uptake rate in $mgO_2/L/h$
Unaffected fish	100	3	5.3	9.7
Affected fish	2	71	0.6	1.1

Tasks with their responses

(a) Analyze why Bandi's imported clownfish often die 2–6 weeks after capture, even if they survive the initial 48 hours.

While lower cyanide doses don't kill the fish immediately, the poison causes long-term neurological damage, liver destruction, and intestinal damage. Cyanide inhibits oxygen consumption (1.1 $mgO_2/L/h$), leading to chronic hypoxia (oxygen starvation with only 2% Cytochrome c oxidase activity) and metabolic failure (0.6 mM).

(b) Based on your knowledge of metabolism, what specific symptoms of distress might these fish have displayed prior to dying?

Signs of distress include increased ventilation (rapid gill movement), gulping for air at the surface, erratic swimming movements (dizziness), and muscle incoordination.

(c) Using the data in the table above, propose strategies Bandi can solve his challenges. (***Attempt this task***).

Item 8

A coastal community notices that a healthy patch of coral reef, heavily frequented by fishermen targeting aquarium fish, has turned pale white and black within 48 hours of increased fishing activity. The fishermen are known to use sodium cyanide (NaCN) squirted from plastic bottles.

Mr. Alexander, an environmental scientist now working with a private environmental protection agency has suggested the ban of the import of cyanide-caught fish. However he and his team of environmentalists face a challenge: it is nearly impossible to distinguish a fish caught with nets from one caught with cyanide just by looking at it.

Tasks with their responses

(a) (i) Explain the biological mechanism by which cyanide kills coral polyps and causes "bleaching."

Cyanide (CN^-) acts as a potent inhibitor of cytochrome c oxidase, the final enzyme in the electron transport chain (ETC) in mitochondria.

By binding to this enzyme, cyanide prevents oxygen from acting as the final electron acceptor, halting aerobic respiration. Without ATP, cellular processes fail, leading to the death of the coral polyps. The "bleaching" occurs because the dying coral expels its symbiotic algae (zooxanthellae). The "black" color indicates necrotic tissue, appearing burned.

(ii) Describe why "non-target" organisms are also affected by this fishing technique.

Sodium cyanide is dissolved in water and sprayed, creating a broad-spectrum poisonous plume, not a targeted tool. The chemical disperses, affecting invertebrates, fish larvae, and microorganisms near the reef, disrupting the whole reef ecosystem.

(b) (i) Suggest a rapid, non-destructive, and reliable biological test to detect if a live fish was exposed to cyanide.

A non-invasive test can be performed by analyzing the water the fish was transported in. Fish metabolize cyanide by converting it into less toxic thiocyanate using the enzyme rhodanese, which is then excreted through urine into the shipping water. Testing this water for high levels of thiocyanate indicates previous cyanide exposure.

(ii) Why is traditional cyanide testing on fish tissue (homogenate) often inaccurate?

If fish are exposed only to a short, pulsed dose of cyanide as is typical in fishing, the fish can detoxify and excrete the poison quickly, resulting in non-detectable levels in tissue samples by the time they reach a laboratory.

Item 9

A lab is investigating the impact of a newly discovered metabolic poison on isolated liver mitochondria. Upon adding the poison, researchers observe that oxygen consumption ceases immediately, and NADH levels in the matrix increase dramatically.

Tasks with their responses

(a) Which specific part of the electron transport chain (ETC) is likely being inhibited, and why?

The poison is likely inhibiting **Cytochrome C Oxidase (Complex IV)** or another intermediate complex (like Complex I/rotenone). It inhibits it by binding to the site where electrons are transferred to oxygen, preventing the reduction of oxygen to water.

(b) What is the consequence for ATP production?

Oxidative phosphorylation stops. Because oxygen is the final electron acceptor, if it is not reduced, the ETC halts, the proton gradient fails, and ATP synthase stops producing ATP.

(c) Suggest a possible solution to restore some ATP production within the cell.

The cell will have to rely solely on **glycolysis** for ATP production. To make this sustainable, the cell needs a method to regenerate NAD^+ from NADH without the ETC, which usually occurs via lactate fermentation in animals or alcoholic fermentation in yeast.

Item 10

Professor Gideon Alexander, a researcher adds an increasing concentration of Ammonium Chloride (NH_4Cl) to a culture of

Azotobacter growing on N_2 gas. Initially, nitrogenase activity is high. As NH_4Cl concentration increases from 0.1 mM to 1.0 mM, the nitrogenase activity drops rapidly, despite there still being N_2 present. Nitrogen-fixing bacteria such as *Azotobacter chroococcum* use the enzyme nitrogenase to produce ammonia from nitrogen gas in the air.

Professor Gideon Alexander investigated the effect of an increase in the concentration of ammonium chloride on the activity of nitrogenase in this bacterium. He prepared several liquid medium cultures of the bacterium. Each liquid culture had the same volume. He grew each culture in a different concentration of ammonium chloride.

In each culture:

• he recorded the nitrogenase activity in arbitrary units

• he removed the bacteria and then recorded the concentration of ammonium chloride remaining in each liquid medium.

Table below shows Professor Gideon Alexander's results.

Concentration of ammonium chloride in µg cm^{-3}	**Nitrogenase activity in arbitrary units**	**Concentration of ammonium chloride remaining in liquid medium in µg cm^{-3}**
0	45	0
20	30	0
40	17	0
60	7	0
80	0	6
100	0	14
120	0	20

Tasks with their responses

(a) Explain why the nitrogenase activity decreases when the NH_4Cl concentration increases.

Nitrogenase converts N_2 to Ammonia (NH_3) which becomes NH_4^+ in cells. High levels of NH_4^+ provide a ready-made source of nitrogen, making energy-intensive nitrogen fixation unnecessary. The NH_4^+causes a short-term, reversible inhibition of nitrogenase, sometimes referred to as the "switch-off" effect, which is a form of negative feedback regulation to conserve ATP.

(b) Identify the covalent modification that occurs on the enzyme to inhibit it.

The iron (Fe) protein subunit of the nitrogenase complex is modified by ADP-ribosylation (reversibly inactivated).

(c) Describe one possible complication of this inhibition on the bacterial population.

If ammonium levels fluctuate (e.g., in soil), the constant switching off and on of nitrogenase consumes energy (ATP) for repairing or modifying the enzyme, which can reduce overall bacterial growth efficiency.

(d) Apart from temperature and pH, give two variables Professor Gideon Alexander would have controlled when preparing the liquid medium cultures. ***Please attempt this task***.

Item 11

A group of senior six students were culturing *Azotobacter* for an experiment on nitrogen fixation. They notice that in the presence of low oxygen (anaerobic conditions), the inhibition of nitrogenase by NH_4Cl is severe. In moderate oxygen conditions, the nitrogenase is less inhibited by the same amount of NH_4Cl.

Tasks with their responses

(a) Explain why low oxygen levels make the nitrogenase more sensitive to NH_4Cl inhibition.

Under low oxygen (microaerophilic conditions), the respiration rate of the cell is low, leading to a smaller pool of available energy (ATP). The presence of NH_4Cl leads to rapid "switch-off".

(b) Why does moderate oxygen reduce the inhibition?

Higher oxygen availability allows for a higher respiration rate, providing a large supply of ATP. This allows the cell to maintain high metabolic turnover and potentially reverse the covalent modification (ADP-ribosylation) of nitrogenase more rapidly.

(c) What is the risk of providing *too much* oxygen to the culture?

Nitrogenase is irreversibly inactivated by high oxygen concentrations.

Item 12