MCD Tutorial 2 – Creatine kinase and myocardial infarction

**Take home messages**

This tutorial is wide ranging and covers many issues in molecular and cellular biology but the main themes relate to: protein structure and function; what is a cell? What does it need to stay alive? Why do cardiac muscle cells die when arteries narrow? How can this be measured quickly, cheaply and with minimal technical support in the casualty department?

You should know and understand the following concepts and will be examined based upon this. Do not worry if you do not achieve this immediately. Take your time and by the time that this term's course is complete you should have a much better appreciation of what is going on.

1. What is a myocardial infarct? – it is the death of heart muscle cells.
2. Why do the cells die? – lack of oxygen.
3. Why is there a lack of oxygen? – blockage of the cardiac arteries. (what is atherosclerosis and what kind of things are risk factors for its early development).
4. Why do cells need oxygen, how do they use it and why do cells die without it?

a. What constitutes a cell i.e. a semi-permeable membrane separating the inside from the outside?

b. There is active exclusion of some things such as Na+ ions.

c. This needs a protein pump in the membrane.

d. These are a type of enzyme called membrane ATPases.

e. They use energy in the form of adenosine triphosphate (ATP) to pump ions.

f. How is ATP generated? Via glycolysis, the Krebs Cycle and eventually oxidative phosphorylation.

g. The end point of the process requires atmospheric oxygen, hence if there is less oxygen supplied to a cell there is less ATP, pumps don’t work ion balance is lost and cells die.

1. Cell contents are released when they are dying, i.e. proteins that should be held inside against concentration gradients appear in the serum.
2. Therefore the levels of many proteins including creatine kinase (many others as well such as lactate dehydrogenase) in serum can be used as indirect indicators of cell death.
3. Where is CK normally present? – CK is present in all cells at very low levels but is at high concentrations in metabolically very active tissues including the brain, heart and skeletal muscle. Creatine phosphate is an energy store.
4. CK activity in the serum can be detected by a coupled assay (e.g. Fig. 1) leading to the generation of detectable products.



1. How then can increased CK be related specifically to the death of cardiac muscle rather than skeletal muscle and brain tissue?
2. CK is a protein made from two subunits or monomers i.e. it is a “dimer”
3. The two monomers are coded for by two different genes.
4. These generate two different monomer isoforms “B” and “M”. The two monomers have approximately the same molecular weigh (43 kDa) but differ in their pI (the pI for the B isoform is 5.2, while the pI for the M isoform is 6.7).
5. Monomers associate and bind to one another in the cytoplasm to produce active dimers (Fig 2.)
6. Thus, if both genes are expressed in a cell, three final dimers are possible, “BB”, “MM” and “MB”.
7. The Brain only expresses the B gene and hence makes only B monomers and so only the BB form can be generated.
8. Conversely the MM form is the only one made in skeletal muscle cells (useful in diagnosis of the extent of skeletal muscle damage in muscular dystrophies).The only tissue where both genes are expressed is cardiac muscle cells. They therefore make all three dimers including the hybrid BM form.
9. Thus, death of cardiac muscle fibres can be determined if the BM isoform of CK can be detected in the serum.
10. It should be stressed here that the levels of CK BM isoform in the serum are directly proportional to the amount of cell death in the heart. This is because each myocyte can be considered to be approximately of equal volume (they have equal likelihood of dying independently of their size) so, as each cell dies it releases a “quantum” of CK into the extracellular fluid and thence into the serum.
11. So, is a simple measure of CK activity in serum a sufficiently good diagnostic test for myocardial infarct”? No because the activity could be from any of three tissues. Assay of the BM form is essential.
12. In general how might you experimentally discern between different protein isoforms, what methods can be used? Electrophoresis or column chromatography perhaps? These do the job as they can separate on the basis of molecular weight or charge but are slow and require expert technical operation. Neither are ideal in a Casualty Department.
13. An immunological approach? “What other way can individual proteins be discriminated”? What do antibodies do but recognise specific proteins. It is but a short step then to thinking about the artificial manufacture of antibodies against desired proteins. The ideal test would be one that depends upon a specific antibody to the BM isoform. Unfortunately, such an antibody licensed for human diagnostic use does not yet exist. However, the reverse can be done. An antibody exists for the BB form, and by, for example, using an antibody to the BB form and then determining the residual remaining reaction, this gives the reaction for both BM and MM forms, if present.

However, this test would be given along with many others and would never be used alone to determine if and infarct had occurred but rather it is used to determine how big the infarct is and by changes in levels approximately how long ago.

1. What is the time course of CK after myocardial infarction? A typical time course for CK is illustrated (Fig. 3), together with two other markers.
2. What other markers can be used for diagnosis of myocardial damage? Serum glutamate oxaloacetate transaminase (SGOT); lactate dehydrogenase (LDH); cardiac troponin. Troponin is the calcium switch in muscle. Cardiac troponin I and troponin T are tissue specific and are not present in any other muscle. Their presence in the serum represents a specific marker for cardiac infarction (typically appearing in the serum after 48h of infarction and persisting for approximately 5 days).