Welcome to module 20, Sequence of reactions of Glycolysis- preparatory phase under Unit 3 of Paper Biochemistry and Metabolic Processes. Myself, Dr. K.K.Therisa, Dhempe College of Arts and Science, Miramar, Goa.
The outline of this module is, it comprised of sequence of reaction of glycolysis with more emphasis on preparatory phase, that is the phase I of glycolysis. The enzyme catalyzed reactions in phase one of glycolysis and importance of preparatory phase of life processes. Learning outcome at the end of this module is the student should be able to understand and illustrate the preparatory phase of glycolysis and the sequence of enzyme catalyzed reactions. The breakdown of 6 carbon glucose into two molecules of three carbon pyruvate in glycolysis that occurs in 10 steps.

The first five steps constitute the preparatory phase. The hexose chain of glucose is cleaved into two triose phosphate at the end of Preparatory phase that is fifth step in glycolysis. Steps of preparatory phase of glycolysis is like this: number one, first and foremost, the glucose that is entered into the cell through the plasma membrane will first phosphorylated at the hydroxyl group on carbon no 6. And what is formed is glucose 6 phosphate, which is then converted to fructose 6 phosphate in Step 2. Step 3 is the fructose 6 phosphate getting phosphorylated at C1 to yield Fructose 1,6 biphosphate and further step is the splitting of fructose 1/6 Biphosphate into two-3 carbon molecules of dihydroxyacetone phosphate and glyceraldehyde-3-phosphate.

And 5th step is the conversion of dihydroxyacetone phosphate through isomerization to the glyceraldehyde 3 phosphate, and thus ending the first phase of glycolysis that is the preparatory phase.
And this is an important step.
The last step is important because glyceraldehyde-3-phosphate will be entering into further steps of glycolysis. So dihydroxyacetone phosphate, which is a ketose, has to be converted to a aldose that is glyceraldehyde-3- phosphate.

Let us see step by step the sequence involved in preparatory phase of glycolysis.
Glycolysis begins with the phosphorylation of glucose molecule or hexose molecule, where the enzyme Hexokinases is playing a major role in converting glucose to glucose 6 phosphate. And later, the glucose 6 phosphate is been converted through an isomerization process into fructose 6 phosphate. And the enzyme involved is phosphohexose isomerase. During the conversion of glucose to glucose 6 phosphate, ATP molecule involved for the transfer of phosphate ie, inorganic phosphate to the glucose molecule. And in 3rd step, that is, where in the fructose 6 phosphate which is converted to fructose 1,6 biphosphate again will use one more inorganic phosphate from ATP molecule. And that's why there are two ATPs utilized in this preparatory phase of glycolysis. And the fructose 6 phosphate to fructose 1,6 biphosphate is been catalyzed by the enzyme phosphofructokinase-1.

Fructose 1,6 biphosphate is further been converted through aldol condensation into two compounds, that is glyceraldehyde three phosphate and dihydroxyacetone phosphate. These two molecules, or these two compounds, are three carbon compound, wherein glucose is a 6 carbon compound can see the structure on the other side, glucose is a 6 carbon compound and glucose 6 phosphate again is a 6C. This Carbon compound varies wherein at 6 carbon position it is been phosphorylated and then there is splitting of this

and again. Ring formation in such a way that glucose now is converted to fructose with the help of enzyme phospho hexosisomerase and later the fructose 6 phosphate again gets phosphorylated
that at carbon 6 and carbon 1 there is phosphate group attached to it and this is the one that gets split into two compound, that is glyceraldehyde three phosphate and dihydroxyacetone phosphate.

These five steps are the part of preparatory phase of glycolysis. Let us take them one by one.

The first step in the preparatory phase of glycolysis is the phosphorylation of glucose. You can see the six carbon compound that is a glucose with the help of a hexokinase enzyme gets converted to a glucose 6 phosphate and the phosphate ie, inorganic phosphate that is been brought here or transferred here is from ATP molecule, wherein the ATP is converted to ADP and this enzyme works only if the ATP is formed a complex with a magnesium. And then what we get, as a product, is a glucose 6 phosphate because at 6 carbon position there is a phosphate that has been attached to the molecule.

Second step is the conversion of glucose 6 phosphate to fructose 6 phosphate, which is through the isomerization process. The enzyme that is involved in catalyzing this process is the phospho hexose isomerase. It just converts the glucose 6 phosphate into a fructose 6 phosphate, wherein there is a break in a ring and then later 5 carbon ring is being formed from a 6 carbon ring. Let us see this isomerization technique using this depiction, here the glucose 6 phosphate first with the help of phosphohexoseisomerase opens up the ring of the Glucose 6 phosphate and we get this chain of glucose 6 phosphate. Then slowly a proton from this, being removed and then cis-enediol intermediate is formed.
And one more proton which is released outside will convert this into a fructose molecule at the end of Step 4, where in a 5 carbon compound is formed ie, a 6C but a 5 ring structure is being formed from a 6 ring structure and now what we have is a fructose 6 phosphate with the help of an enzyme for phosphohexoseisomerase.

And the third step of glycolysis is the phosphorylation of fructose 6 phosphate to fructose 1,6 biphosphate. Wherein the fructose 6 phosphate at Carbon 1 gets phosphorylated. Now the molecule is with two phosphate molecule, one at carbon one and another one at carbon 6 and this is been catalyzed by an enzyme for phosphofructokinase 1, an ATP is again utilized for the transfer of phosphate to carbon. Thus, a product is formed at the step three, that is fructose 1,6 Biphosphate. And this step of glycolysis is considered as a first committed step in glycolytic pathway. And the reason is before this glucose 6 phosphate or glucose may enter into cell may have any other fate, but fructose 16 biphosphate will have the fate of entering into glycolytic pathway. So also fructose 6 phosphate is 4th step involved in the process of glycolysis.

During the preparatory phase, there is the cleavage of fructose 1,6 biphosphate as pyruvate is a 3 carbon compound, the fructose 1,6 biphosphate being a 6 carbon compound has to be split into two molecules of three carbon compounds each and this is done with the aldol condensation process and catalyzed by the enzyme aldolase. What we get at this step is two different compounds. One is dihydroxyacetone phosphate and another one is glyceraldehyde 3 phosphate. Glyceraldehyde 3 phosphate is important for further steps, as this will be entering into the next phase of Glycolysis that is the payoff stage phase of glycolysis. This

cleavage takes place between Carbon 3 and carbon 4 or fructose 1,6 biphosphate, wherein the enzyme helps to split this portion in such a way that we get two different compounds of three carbon molecule in fifth step into conversion of the triose phosphate, wherein isomerization of dihydroxyacetone phosphate occurs.

Dihydroxyacetone phosphate is a ketose molecule which doesn't enter into further steps of glycolysis, so it has to be converted to glyceraldehyde 3 phosphate, and which is done with another isomerase enzyme that is triose phosphate isomerase. Triose, because it is involved with three carbon compound. And all the dihydroxyacetone phosphate is converted to glyceraldehyde three phosphate. That means from one molecule of glucose, we have got two molecules of three carbon compound that is glyceraldehyde 3 phosphate by the end of Step 5.

So to conclude, in this module, preparatory phase of glycolysis consist of First five steps of glycolysis. Which results in cleavage of the hexose chain that is a glucose molecule or 6 carbon compound and it requires an investment of two moles of ATP to activate a single glucose molecule and prepare it for its cleavage into 2 carbon compound, that is glyceraldehyde 3 phosphate.

Thus glucose molecule has been phosphorylated at carbon one and carbon 6 and then cleaved to form 2 molecules of glyceraldehyde 3 phosphate during the preparatory phase of glycolysis?

This module was prepared using the following references. Thank you.