Glycogen Metabolism

Suggested Reading:

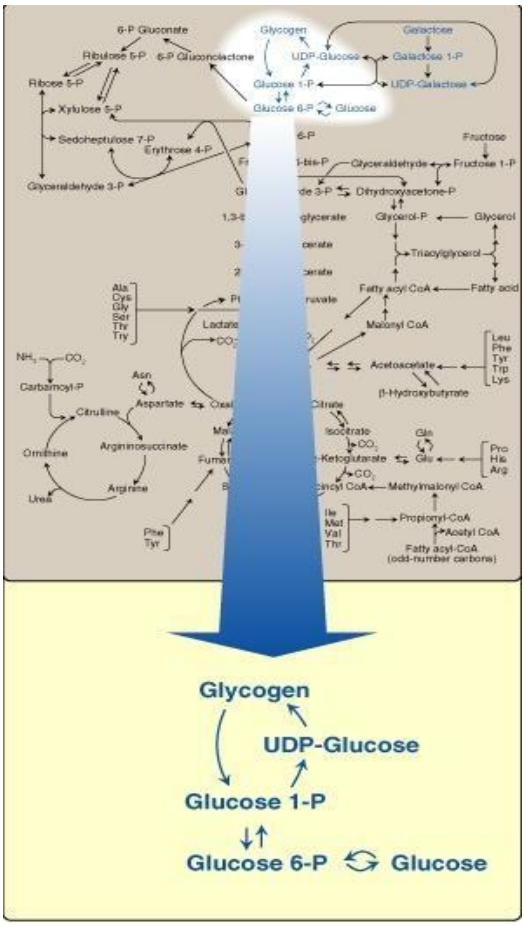
Lippincott's Illustrated reviews: Biochemistry

Sources of Blood Glucose

- Diet
  - Starch, mono and disaccharides, glucose
  - Sporadic, depend on diet,
- Gluconeogenesis
  - Sustained synthesis
  - Slow in responding to falling blood glucose level
- Glycogen
  - Storage form of glucose
  - Rapid response and mobilization.
  - Limited amount
  - Important energy source for exercising muscle.

What was mentioned in the lecture:

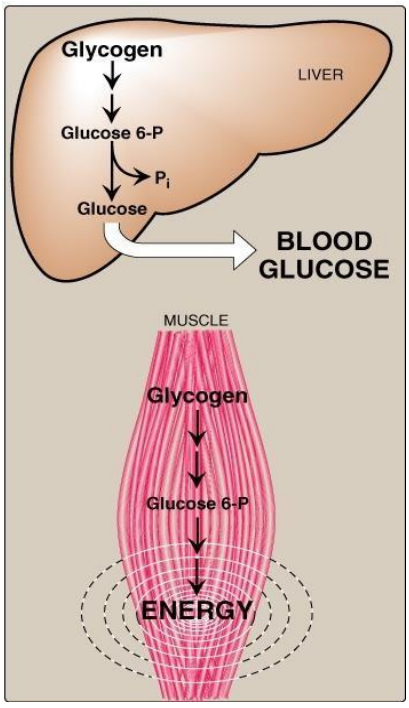
In the well-fed state (after we eat and carbohydrates are digested and absorbed), diets provides starch, monosaccharaides, disaccharides, and glucose, the problem with is that it is sporadic, you may eat 2 meals per day, or you can do not eat anything, you cannot depend only on diet because you are not eating continuously, we eat intermittently, the first solution for that is glycogen, glycogen provides a storage of glucose that is responsive and mobile, the problem with glycogen is that provides only a small amount, it is an important energy source for exercising muscle, gluconeogenesis is the source of glucose after glycogen is consumed, it provides a sustained synthesis of glucose, but it is slow in responding to low blood glucose when compared to glycogenolysis, glycogenolysis respond more quickly compared to gluconeogenesis when blood glucose level is decreased.

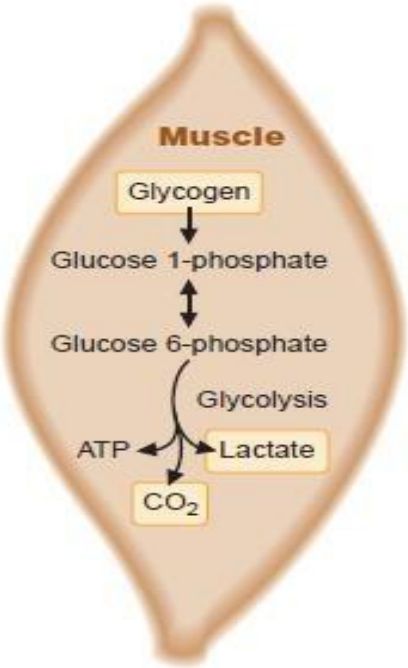


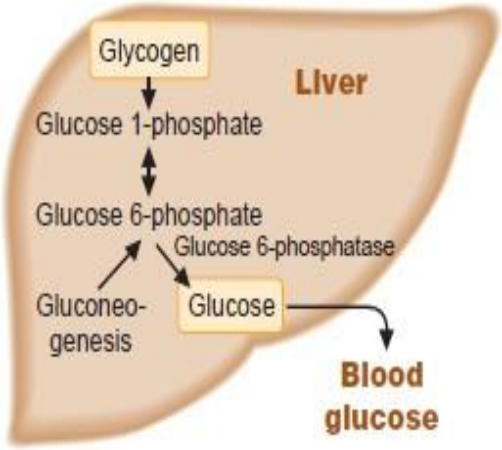
What was mentioned in the lecture:

This figure depicts the pathways of glycogen metabolism, the part of the figure highlighted in white depicts glycogen degradation (glycogenolysis), simply, glycogen conversion to glucose (glycogenolysis) proceeds as such: glycogen → glucose 1-phosphate→ glucose 6-phosphate→ glucose, while glycogen synthesis (glycogenesis) happens as follows: glucose→ glucose 6-phosphate→ glucose 1-phosphate→ UDP-glucose→ glycogen.

Degradation of glycogen produces glucose 1-phosphate which can be easily converted to glucose 6-phosphate, glucose 6-phosphate is an intermediate in glycolysis and gluconeogenesis, the pathway of glycogenesis is not the exact reverse of the pathway of glycogenolysis.

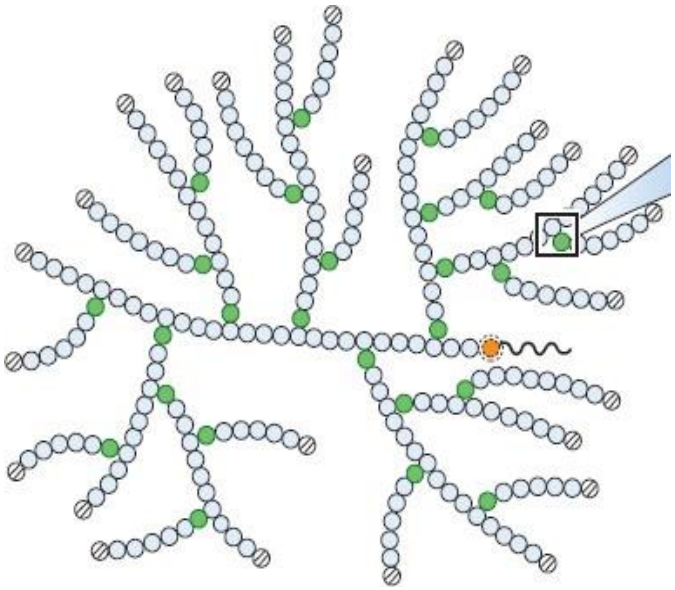






What was mentioned in the lecture:

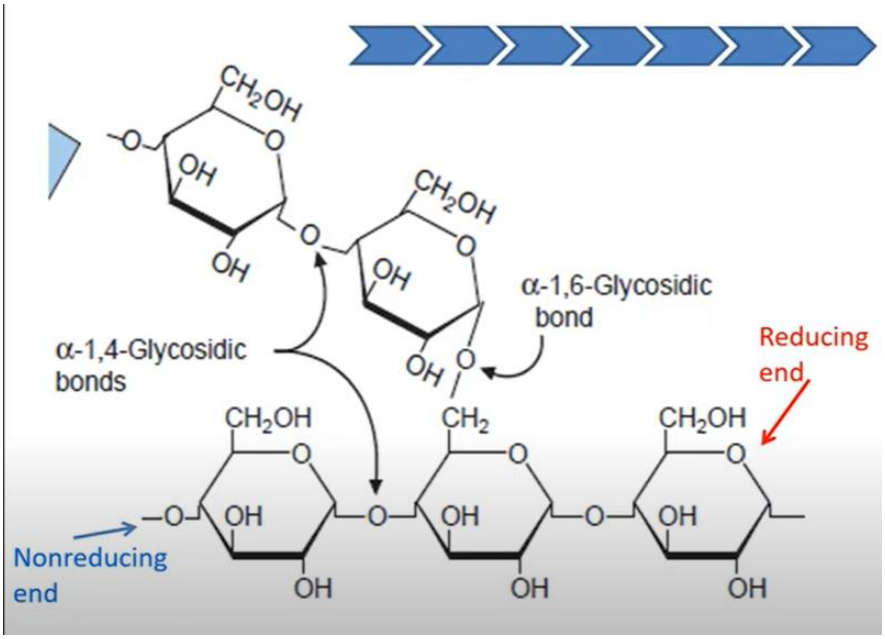
Glycogen is stored in the liver and in the muscles, the amount of glycogen in muscles (400 g) is four times the amount of glycogen in the liver (about 90 g), however, the one that releases glucose to other tissues to be used by the other tissues is the liver while muscles are selfish, they utilize glycogen for their own use, only liver's glycogen is available for other tissues.



* Extensively branched homopolysaccharide
* One molecule consists of hundreds of thousands of glucose units

What was mentioned in the lecture:

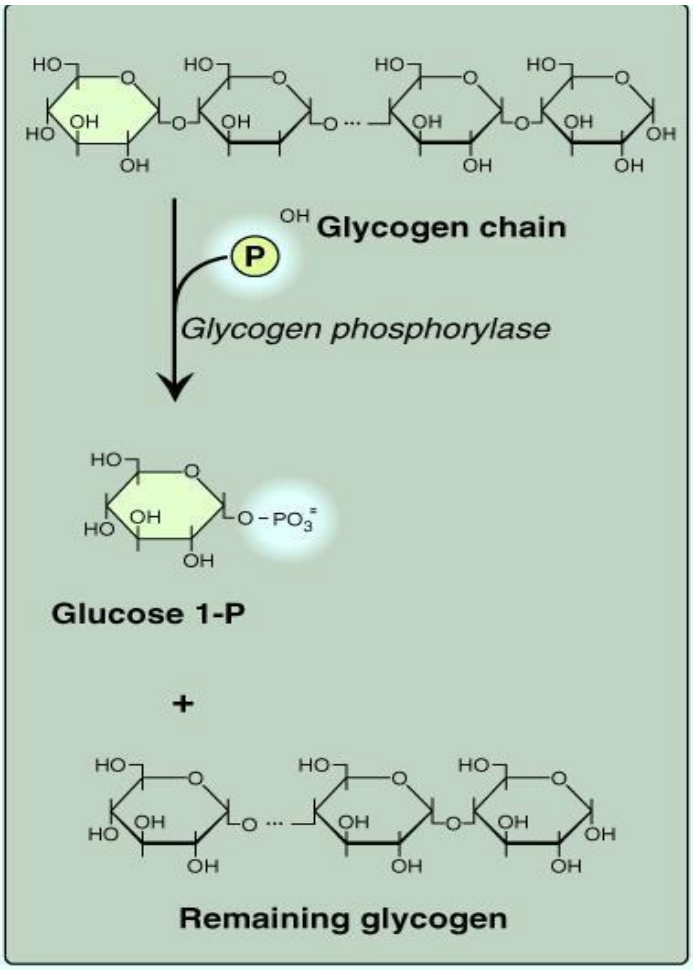
This is the structure of glycogen, it is composed of many glucose residues, it is highly branched polysaccharide, it is composed of only glucose, it is similar to amylopectin starch, the difference between then is that glycogen is more intensively branched, that's why it is called animal starch, in each chain, glucose residues are joined together through an α 1→4 glycosidic bonds, on the branching points (where a branch joins another branch), the bond is α 1→6, the glucose residues at edges have a free hydroxyl groups on carbon #4 meaning that they are nonreducing ends, but the first glucose residue that we started attaching other residues to it have a free hydroxyl group at carbon #1, meaning that glycogen has one reducing end and many nonreducing ends.



What was mentioned in the lecture:

This figure depicts the structure of glycogen, notice that the bonds between the residues in a chain is α 1→4 glycosidic bonds, while it is α 1→6 at the branching points, glycogen have many nonreducing ends and one reducing end, these ends are different, on the reducing end, the hydroxyl group of carbon number one is free, but in the nonreducing ends the hydroxyl group of carbon #4 is the one that is free.

What is the significance of branching of glycogen? The branching increases the number of nonreducing end, and the enzyme that degrades glycogen removes one glucose residue at a time, and this enzyme cleaves at nonreducing ends, therefore, many nonreducing end means many sites at which for the degradation enzyme to act, that provides rapid and efficient degradation when we need glucose, also branching makes glycogen more soluble .



Degradation of glycogen

Degradation of glycogen One glucose unit is removed at a time

From the nonreducing ends

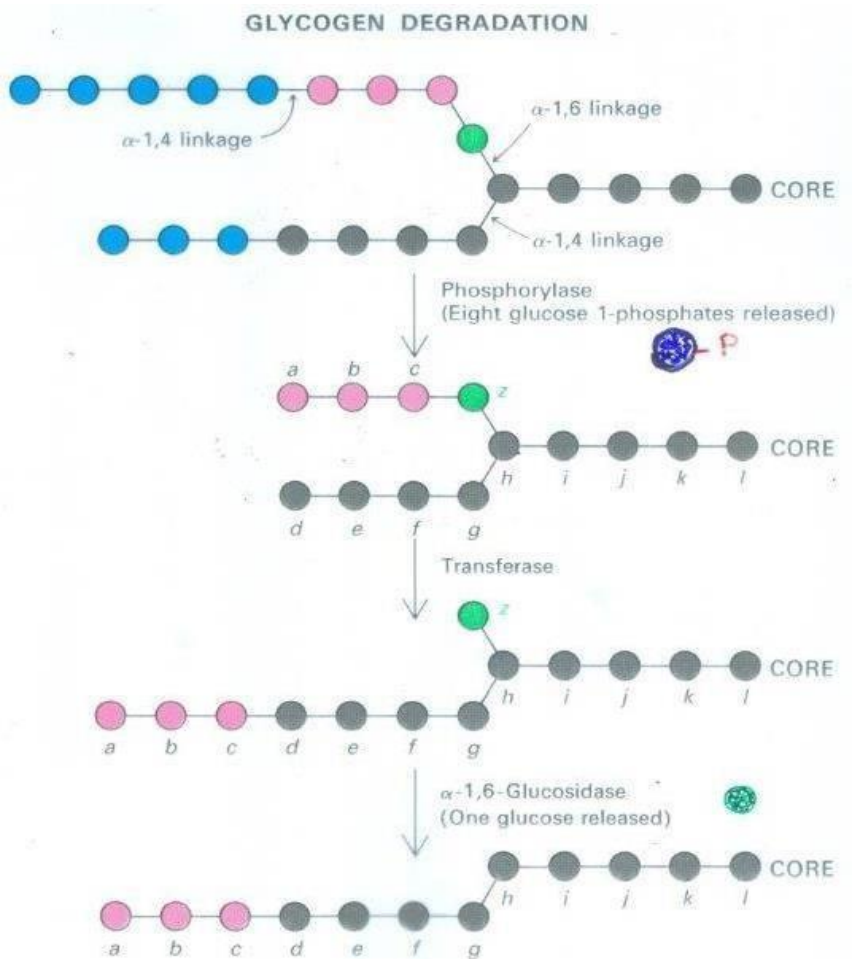
Released in the form of glucose 1-phosphate

What was mentioned in the lecture:

Degradation of glycogen starts at the terminal nonreducing ends, by the action of an enzyme called glycogen phosphorylase, this enzyme uses phosphorolysis (--O-- + H^+ + PO_4^{-3} → --OH + --OPO_3^{-3} rather than hydrolysis through inorganic phosphate, therefore, the result is a glucose 1-phosphate molecule, in degradation, one glucose unit is removed at time from a nonreducing end.

What is the significance of using phosphorolysis instead of hydrolysis?

Because phosphorolysis produces glucose 1-phosphoate which can be easily isomerized to glucose 6-phosphate that enter glycolysis, therefore, using phosphorolysis saves one molecules on ATP, that will be used for phosphorylating glucose, in muscular tissue, the glucose 6-phosphate proceed directly into glycolysis, while in the liver, glucose 6-phosphate is dephosphorylated to glucose to be transported through the blood stream to another tissues.



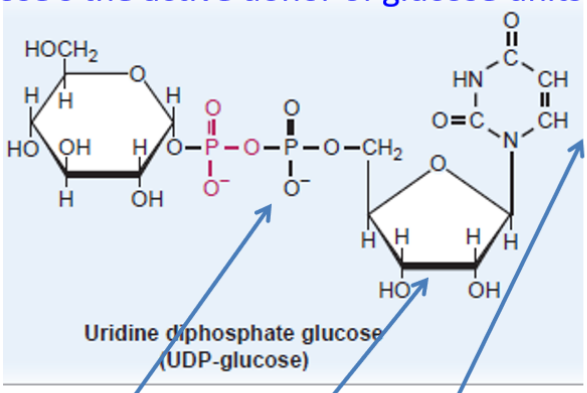
What was mentioned in the lecture:

This figure depicts glycogen degradation, glycogen phosphorylase removes one glucose residue at a time from a nonreducing end, after 7 cleavages, and we have reached a point where we are 4 residues away from the branching point, starting form this point, glycogen phosphorylase cannot cleave anymore residues (it can only cleave from long branches and not short branches), an enzyme called transferase (oligotransferase) comes and transfers three glucose residues form the branch that is being cleaved to another branch (the main stem chain), so far, one residue has remained on branching point, it is connected through an α 1→6 bond, therefore, this residue is cleaved by an hydrolase enzyme called α 1→6 glucosidase, because it is a hydrolase, then the product is glucose and not glucose 1-phosphate.

In the lab, the net result of glycogen degradation is almost one glucose molecule for each ten glucose 1-phosphate molecule, that indicates that we have almost a branching point for each 10 glucose residues.

The enzymatic activity of transferase and α 1→6 glucosidase is refered to as a debranching enzyme.

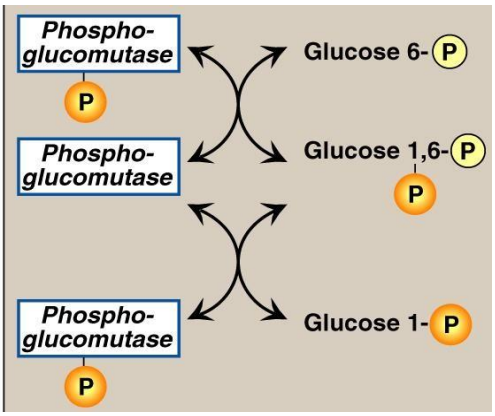
Glycogen is synthesized by adding glucose one by one
UDP-Glucose s the active donor of glucose units



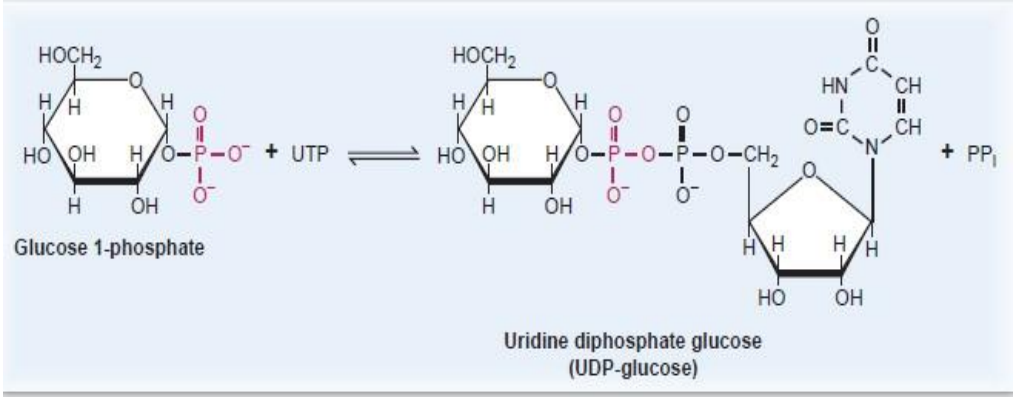
Phosphate Ribose Uracil

What was mentioned in the lecture:

On the other hand, glycogen synthesis occur by adding glucose residues one by one, UDP-glucose is the active donor of glucose units, this is structure of UDP, it is similar to ADP except that it has uracil as a nitrogenous base instead of adenine, UDP is composed of a uracil, a ribose ,and a pyrophosphate, one of them is esterified to the hydroxyl group of carbon #1 of glucose, the point behind using ADP is that it is a carrier of activated glucose units.

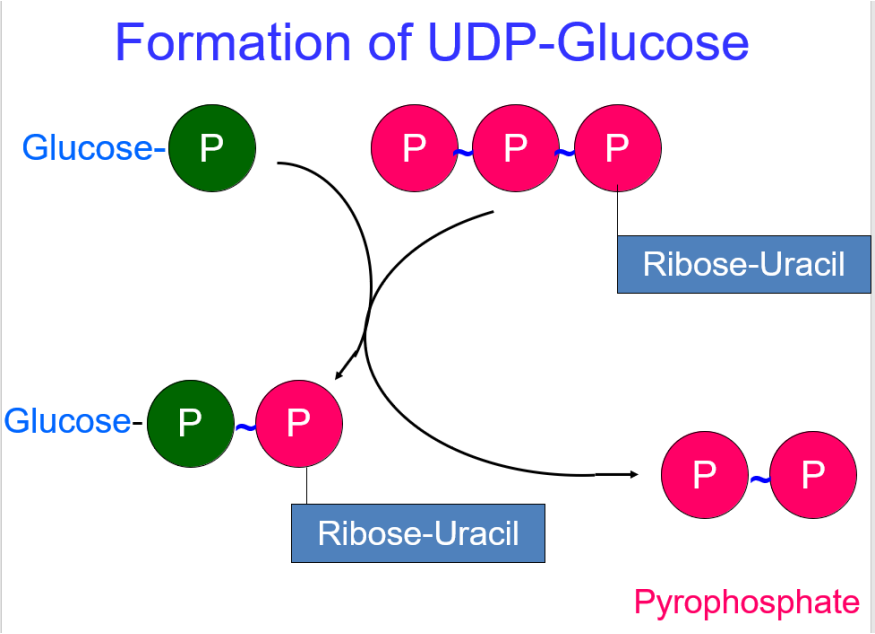


Formation of UDP-Glucose



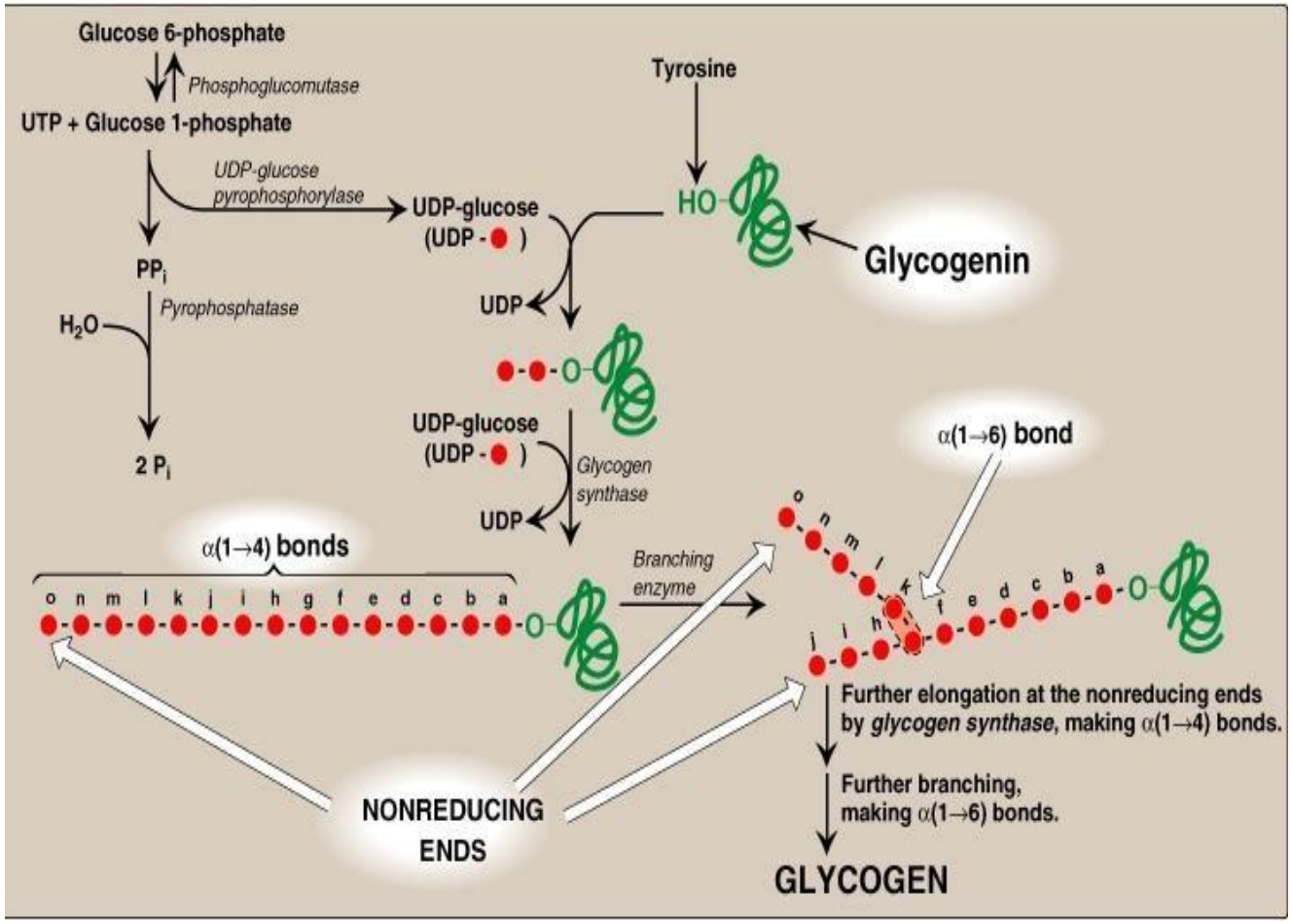
What was mentioned in the lecture:

UDP-glucose is formed as follows: glucose 1-phosphate reacts with UTP forming UDP-glucose and releasing a pyrophosphate PP_i.



What was mentioned in the lecture:

The bonds the connects the phosphate groups is the UTP molecule is high energy anhydride bonds, glucose 1-phosphate binds in the place a pyrophosphate is cleaved (in the place of the second phosphate), the bond that joins the first to the second phosphate is the one that is cleaved and replaced by a bond with glucose 1-phosphate, by this reaction, we cleaved a phosphate-phosphate anhydride bond, and made a new phosphate-phosphate anhydride bond, we cleaved a high energy bond, and generated another high energy bond, what do you expect the ΔG of this reaction to be? The ΔG of this reaction is actually very close to zero, this reaction is reversible, but if we removed one of the products, we'll shift the equilibrium in the forward direction, that's we have an enzyme called pyrophosphatase, pyrophosphate is always there, it is not regulated, whenever there is pyrophosphate, this enzyme will cleave pyrophosphatase into two phosphates, because reaction in which pyrophosphate is produced is usually reversible and we don't want to be reversed.



What was mentioned in the lecture:

this figure depicts glycogenesis and glycogenolysis, in glycogenesis, firstly, glucose is phosphorylated to glucose 6-phosphate through hexokinase, then glucose 6-phosphate is isomerized into glucose 1-phosphate by phosphoglucomutase, then glucose 1-phosphate is joined to UTP by UTP-glucose pyrophosphorylase producing UDP-glucose and pyrophosphate which is cleaved through pyrophosphatase, then UDP-glucose donates the glucose residue to a growing chain of glycogen, a reaction catalyzed by glycogen synthase, glucose is added one at a time to the growing glycogen chain, glycogen synthase cannot start at the very first glucose, meaning it cannot start glycogen synthesis by adding a glucose molecules to another one (it cannot generate a bond between glucose no1 and glucose no2), therefore, glycogen synthesis starts by a protein called glycogenin, glycogenin firstly attach four glucose residues to the side chain of a tyrosine residue in its stucture (it attaches a tetrasaccharide to itself), after that, glycogen synthase starts adding, as the glycogen chain is becoming longer, we need to form branches, a branching enzymes (functions in the opposite way of debranching enzyme) comes and make a branch, it cleaves some residues (four or five) from the main stem and transfer them as a branch.

Glycogen Storage Diseases

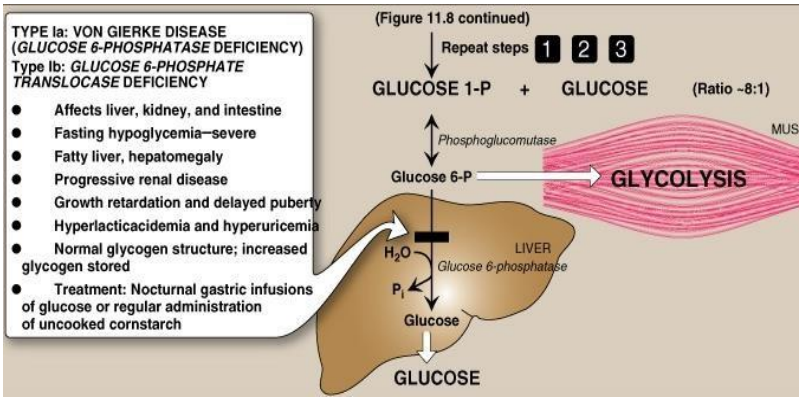
- Genetic diseases
- Defect in an enzyme required for synthesis or degradation è
- Accumulation of excessive amount of glycogen
- In one or more tissue
- Severity: FATALin Infancy..... Mild disorder

What was mentioned in the lecture:

In every metabolic pathway, if an enzyme has got a mutation making it less active or inactive, the whole pathway will be affected, causing a disease in the organism, so glycogen storage diseases are genetic diseases, they result from a defect enzyme in either synthesis or degradation, a mutation in any enzyme involved in glycogenolysis making it less active or inactive will cause accumulation of glycogen, maybe in one or a different tissues, the problems result from this disease could be very severe causing death during infancy, or they could be mild and the patient is able to cope with. We'll talk about three of these diseases.

Glycogen Storage Diseases (examples)

- I Glucose-6-phosphatase (von Gierk's) disease
  - Liver, kidney and intestine.
  - Severe fasting hypoglycemia
  - Hepatomegaly fatty liver.
  - Normal glycogen structure.
  - Progressive renal disease.
  - Growth retardation.



What was mentioned in the lecture:

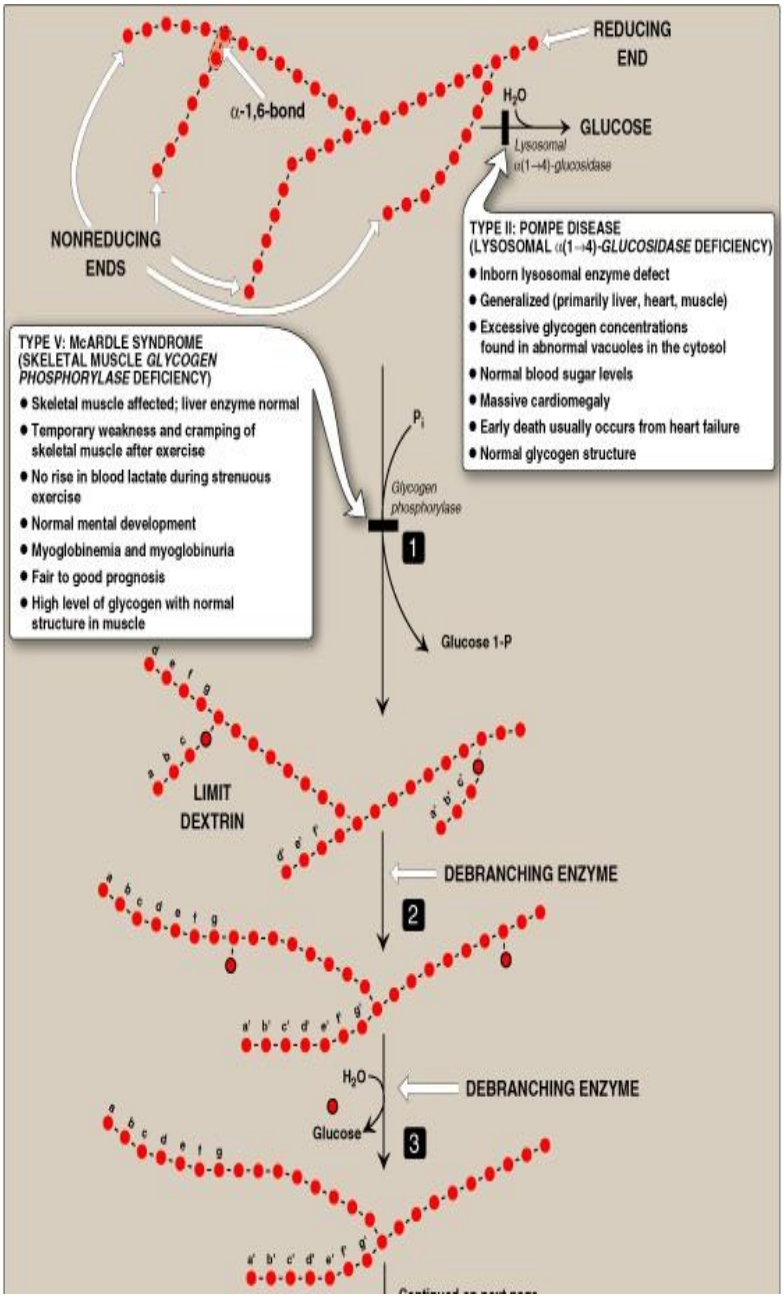
Glycogen storage disease I results from a mutation that affects glucose 6-phosphatase of gluconeogenesis, this disease is called Von Gierk's disease, it affects the liver, the kidneys and small intestine, it causes severe fasting hypoglycemia, because both gluconeogenesis and glycogenolysis requires this enzyme, it also causes a hepatomegaly (enlarged) fatty liver, in this disease, the structure of glycogen is normal if examined under the electron microscope, it causes progressive renal disease

(because glycogen is stored also in the kidney actually), and growth retardation, it is characterized mainly by severe fasting hypoglycemia.

Glycogen Storage Diseases (examples)

- V Muscle glycogen phosphorylase (McArdle syndrome)
  - Only muscle is affected;
  - Weakness and cramping of muscle after exercise
  - no increase in [lactate] during exercise

this disease is glycogen storage disease type 5, it affect the muscles by a mutation in muscular isoform of muscle glycogen phosphorylase, it is called McArdle disease or McArdle syndrome, it affect only muscles, its symptoms include weakness and cramping of the muscles after exercise with no increases in blood lactate during exercise, this disease do not cause fast hypoglycemia, because muscles only store glycogen for their own use.



Glycogen Storage Diseases (examples)

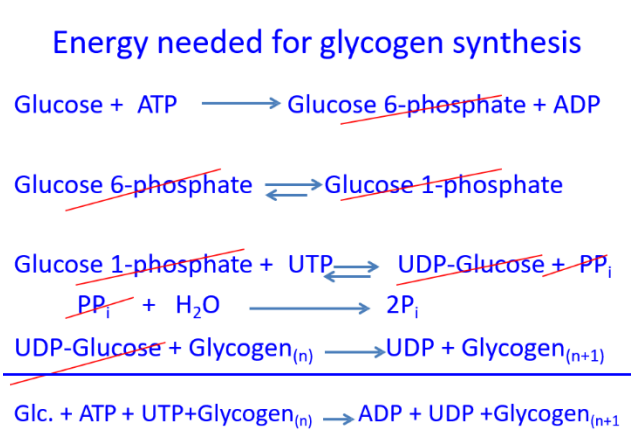
- II Lysosomes α (1→4) glucosidase → POMP Disease
- Degradation of glycogen in the lysosomes
- ≈ 3% of glycogen is degraded in the lysosomes
- Affects liver, heart and muscle
- Excessive glycogen in abnormal vacuoles in the lysosomes
- Massive cardiomegaly
- Normal blood sugar, normal glycogen structure
- Early death from heart failure.

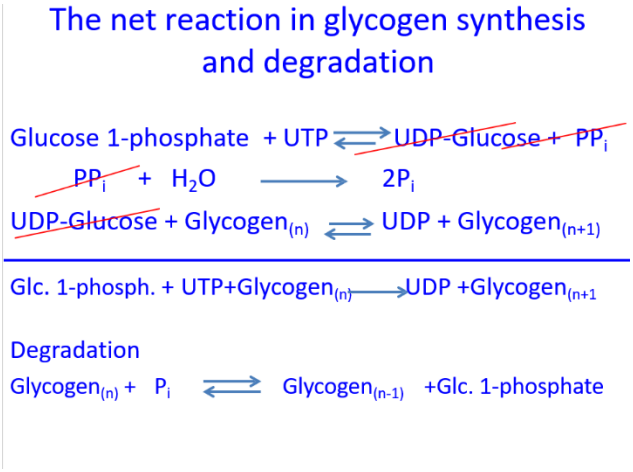
What was mentioned in the lecture:

the third type of glycogen storage disease is glycogen storage disease type II, in which a mutation affects the lysosomal isoform of α 1→4 glucosidase,

it is called POMP disease, a small part of glycogen is actually degraded in the lysosomes 3%, it affect the heart, the liver and the muscle, muscles degrade glycogen using α 1→4 glucosidase rather than glycogen phosphorylase, it causes massive cardiomegaly but normal blood sugar and normal glycogen structure, usually causes early death from heart failure.

Note: actually most tissues can store a small amount of glycogen for their own use,



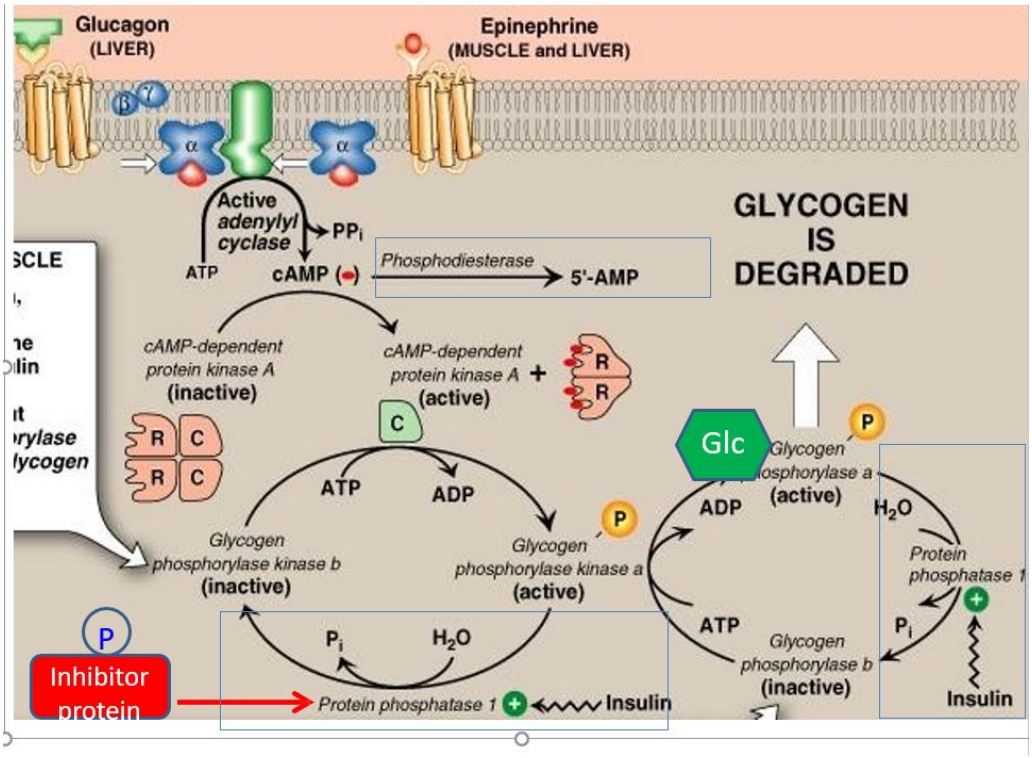


What was mentioned in the lecture:

This slide reviews glycogen from an energy point of view, regarding the synthesis, phosphorylation of glucose require 1 ATP molecules, isomerization of glucose doesn't require any energy, and joining the glucose to an UDP and then joining it to glycogen requires conversion of one UTP to UDP, therefore to add 1 glucose molecule to a glycogen tree, that requires 2 equivalents of ATP, but it requires 1 ATP equivalent is you started from glucose 1-phosphate.

Notice that we do produce or utilize ATP in degradation of glycogen, therefore, if glycogenolysis and glycogenesis happened at the same time, we'll not gain any thing but loss of energy, therefore, they shouldn't occur at the same time in the same tissue, and should be regulated.

Notice that the degradation reaction is reversible, while the synthesis reaction in irreversible because of the usage of UTP as a glucose donor, that makes glycogen synthesis irreversible and exergonic.



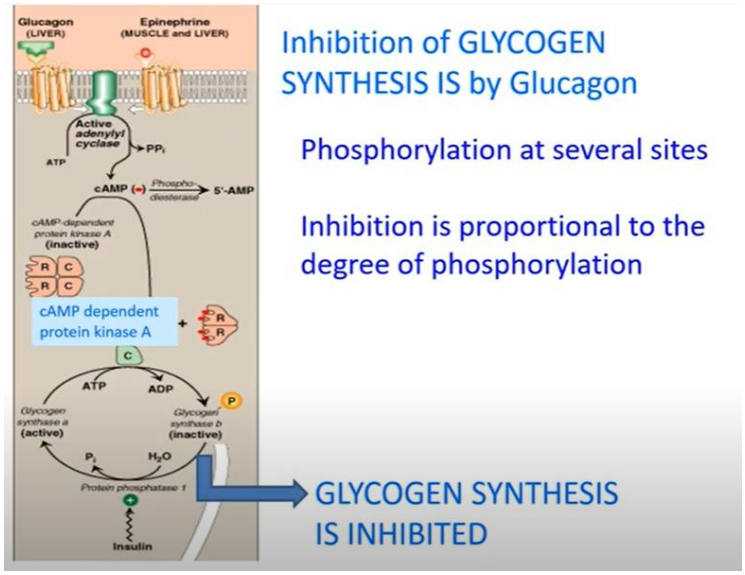
What was mentioned in the lecture:

Regulation of glycogenesis and glycogenesis occur through several mechanisms, firstly, one of which is hormonal and occur through blood glucose, when blood glucose level is low, glycogen degradation should be activated, how can the liver “know” that the blood glucose level is low? By hormonal message through glucagon, glucagon is secreted by the pancreas, it tells the liver the blood glucose is low, therefore you must start making glucose available, when glucagon’s blood level is elevated, it binds to a G-protein coupled receptor composed of 7 helices on the surface of hepatic cells, one glucagon binds to its receptor, a G protein binds to the receptor, actually, many G proteins binds one after another to the glucagon receptor, causing activation of G proteins by replacing GDP with GTPA and disassociation of G_{α}, which goes in turn and activate adenylyl cyclase, activation od adenylyl cyclase allows it to convert ATP to cyclic AMP, cAMP (the second messenger) in turn activates the cAMP-dependent protein kinase (protein kinase A), protein kinase A is a tetramer of two catalytic subunits and two regulatory subunits, upon binding to cAMP, the two regulatory subunits disassociate from the catalytic subunits causing activation, protein kinase A adds phosphate groups to many proteins (broad specificity), including a protein known as glycogen phosphorylase kinase, glycogen phosphorylase kinase is converted to the active form upon phosphorylation, then, it phosphorylates glycogen phosphorylase B, converting it from the inactive form to the active form (glycogen phosphorylase A), the net effect of glucagon is stimulation of glycogen degradation. Why do we have these too many steps? To allow us to amplify the signal, each step produces more of the product, therefore, few molecules of glucagon binding to the receptors will produce a very large number of active glycogen phosphorylase enzyme.

Regulation always must be easily done and easily reversible (terminated), it is easily done because it depends on the conversion of ATP to cAMP, and ATP is abundant is the cell, cAMP is a regulatory molecule should no other function but activation of protein kinase A, this activation can be terminated through hydrolysis of cAMP, in cAMP (cyclic adenosine monophosphate) structure , the phosphate group

is esterified to both carbon #5 and carbon #3 (the phosphate group of cyclic AMP is forming two ester bonds) it is called 3'-5' cAMP, so it has a diester structure, hydrolysis of one of the ester bonds forms AMP and the enzyme that catalyzes this reaction terminating the regulatory effect of cAMP is called phosphodiesterase, phosphodiesterase hydrolyzes the ester bond on carbon #3 producing 5' AMP which is the regular AMP, then it enters the ATP/AMP pool.

How about the phosphorylated proteins? How can their phosphate groups be cleaved? They are hydrolyzed protein phosphatases, those of glycogen phosphorylase kinase and glycogen phosphorylase can be removed by protein phosphatase, those phosphatases are activated by insulin, this makes sense, we don't want to degrade glycogen if we have a lot of sugar in the blood, when we eat, blood sugar increases and so is insulin/glucagon ratio, high glucose means high insulin, that's why insulin inhibits glycogen degradation, by stimulating phosphatases to hydrolyze the phosphate groups of phosphorylase kinase and glycogen phosphorylase, phosphatases are also inhibited by glucagon, through an inhibitor protein, this inhibitor protein when phosphorylated, it goes and bind protein phosphatase inhibiting it, that's why phosphatases under dual regulation, to make sure that the regulation is strict.

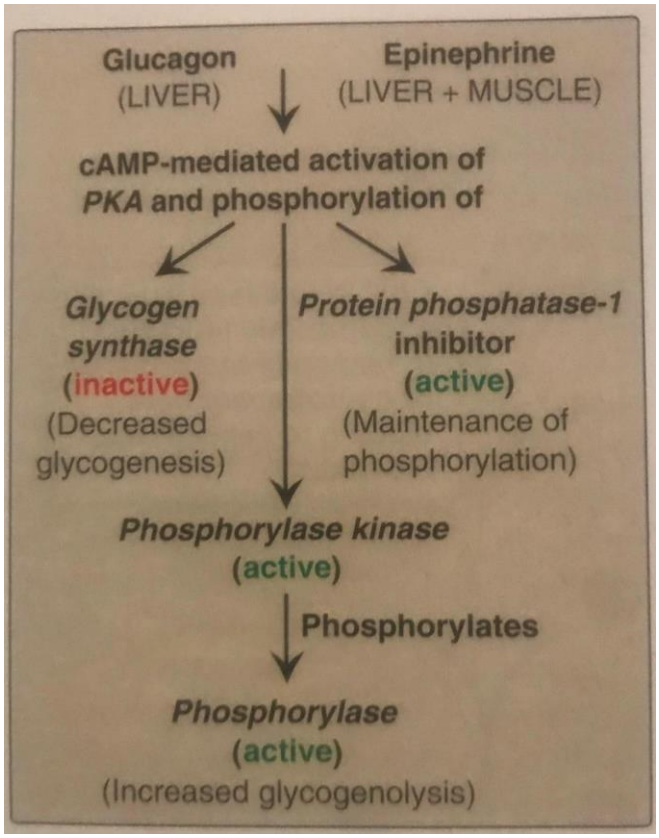


What was mentioned in the lecture:

Glucagon -as it promotes glycogen degradation- it inhibits glycogen synthesis by a similar mechanism: binding of glucagon to a receptor → G protein activation→ adenylyl cyclase activation → cAMP production → protein kinase A activation, then -this time- phosphorylation of glycogen synthase A, converting it to glycogen synthase B that is inactive, inhibiting glycogen synthesis, the same mechanism that activates degradation (covalent modification by phosphorylation) inhibits synthesis. That inhibits synthesis and degradation to happen at the same time.

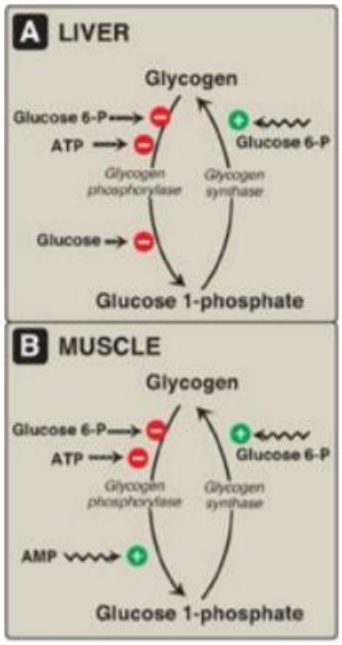
Remember that always phosphorylation of any enzymes involved in glucose metabolism makes glucose more available.

One enzyme can be phosphorylated at several serine and threonine residues (many phosphate can be added to a protein at a time), these amino acids contain an -OH group is their side chains, to which a phosphate group can be covalently esterified, inhibition is proportional to the degree of phosphorylation, more phosphate groups to the protein attached means more inhibition.



What was mentioned in the lecture:

This figure summarizes the action of glucagon and epinephrine in the liver and muscles, they stimulated degradation but inhibit synthesis, and they function by a similar mechanism.



Allosteric Regulation
Rapid response to cell's needs

Available substate and ATP

synthesis

↓↓Glucose and ↓ATP è

Glycogenolysis

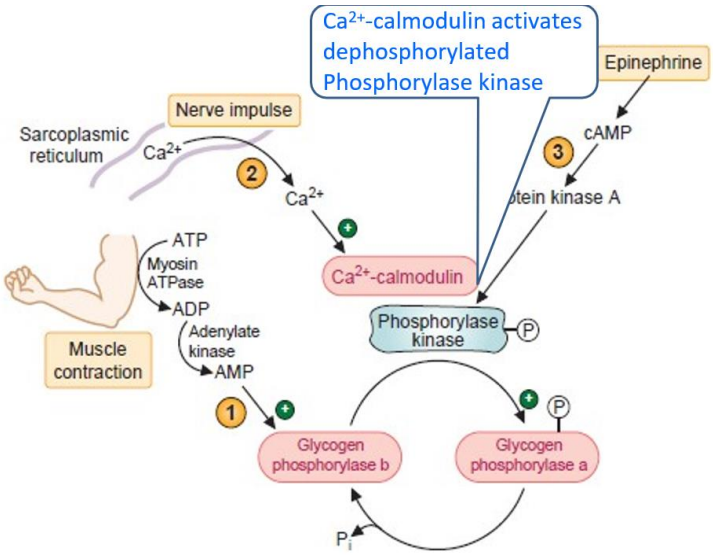
What was mentioned in the lecture:

Glycogenesis and glycogenolysis are also regulated allosterically, not only by covalent modification (phosphorylation), regulation by covalent modification is somewhat slow (it takes from minutes to hours), because it depends on hormones that must be synthesized, released into the circulation, etc. therefore, allosteric regulation is necessary, because it is instant, it is much faster than hormonal regulation, allosteric regulation is carried using small molecules called allosteric modifiers, which bind the enzyme activating or inhibiting it, allosteric regulation provides a rapid response to cell needs (available substrates and ATP), liver's glycogen phosphorylase is inhibited by ATP and glucose 6-phosphate, the idea is that glycogen degradation causes accumulation of phosphorylated sugars (actually glucose 1-phosphate is in equilibrium with glucose 6-phosphate), that will trap all inorganic phosphates making them unavailable for oxidative phosphorylation, that's why it makes sense that high levels phosphorylated sugars inhibit glycogen phosphorylase, also high levels of ATP will inhibit glycogen degradation because if you have a lot of ATP you'll not need to degrade glycogen, liver's glycogen phosphorylase is also inhibited by glucose.

On the other hand, glucose 6-phosphate that inhibits glycogen degradation always promotes glycogen synthesis allosterically.

In muscles, the case is similar, glycogen degradation is inhibited by ATP and glucose 6-phosphate, with the later being an activator for glycogen synthesis, but in muscle, glycogen degradation is also promoted by AMP and Ca^{+2}. The significance of calcium being an activator is that calcium is also the signal that induced muscle contraction, and contraction also requires ATP (it requires energy, it needs glucose), therefore, it make sense that calcium induce contraction and the requirements of contraction.

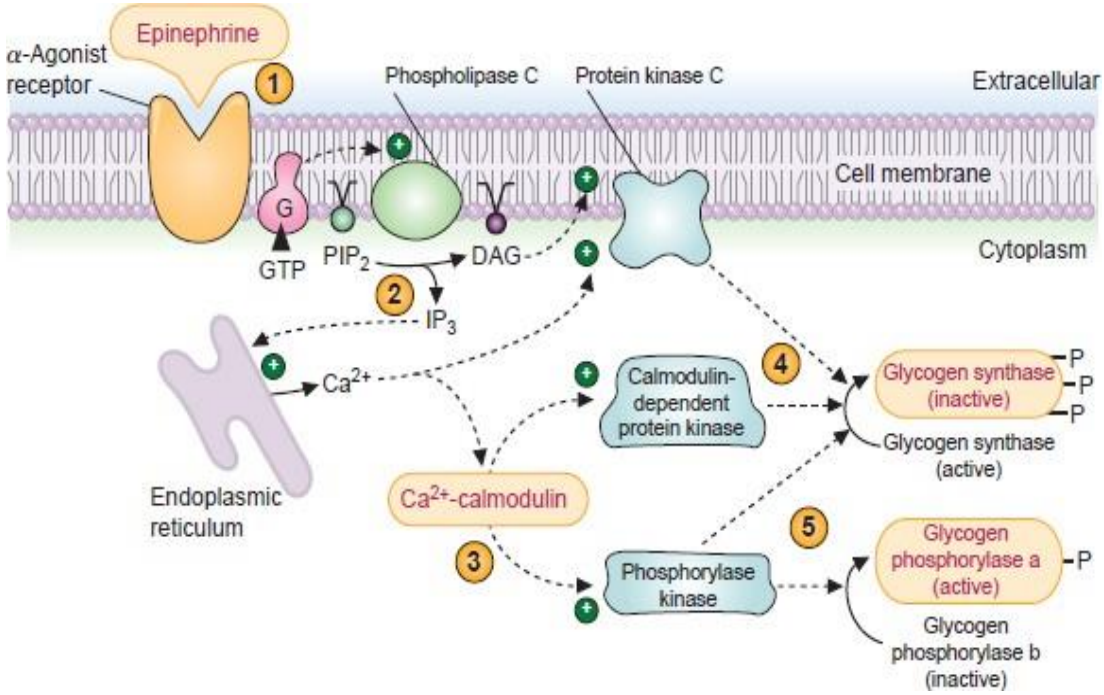
We've said previously that AMP is produced in muscles when an ADP molecule donates a phosphate group to another ADP forming AMP and ATP (a reaction catalyzed by an enzyme known as adenylate kinase), AMP indicated that the energy level in the cell is very low, therefore, it makes sense that AMP is an activator for glycogen phosphorylase.



What was mentioned in the lecture:

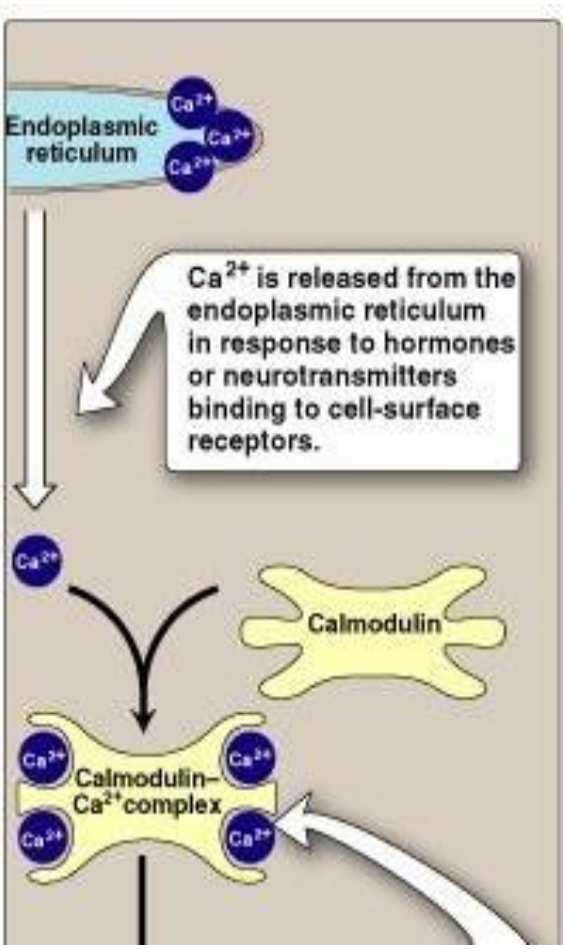
A nerve impulse stimulates calcium release form intracellular storages in muscle cells, that induce muscle contraction through interactions between actin and myosin, muscle contraction causes production of ADP which is then converted to AMP by adenylate kinase, AMP activates glycogen phosphorylase, calcium do not act on glycogen phosphorylase directly, rather, by binding to calcium binding proteins, it activates a protein known as calmodulin , one of the most abundant calcium binding proteins, Ca^{+2} -calmodulin complex binds glycogen phosphorylase kinase activating it, which in turn phosphorylates glycogen phosphorylase causing its phosphorylation, epinephrine also promotes glycogen degradation through activation of protein kinase A (cAMP-dependent).

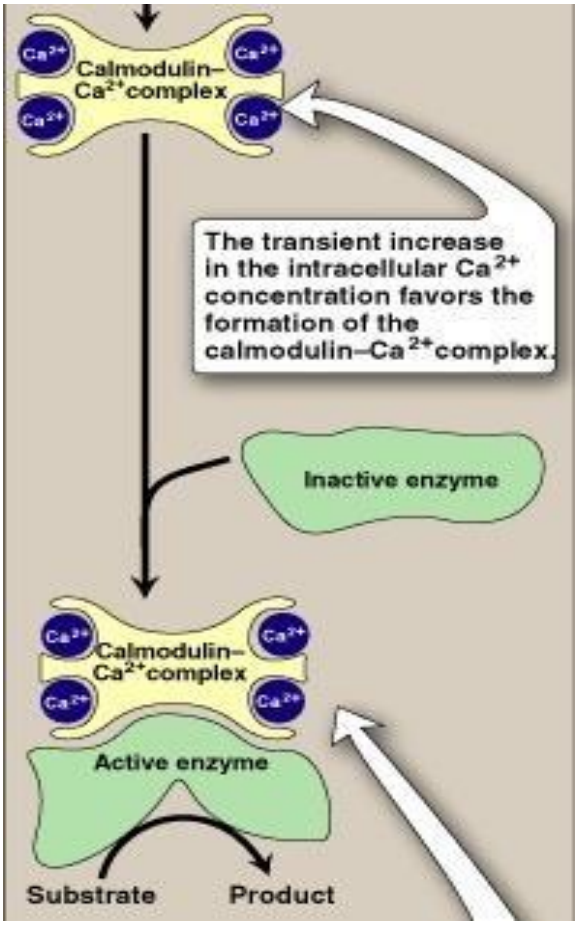
Calcium Activation of liver phosphorylase Kinase



What was mentioned in the lecture:

Activation of liver's phosphorylase kinase can occur through the action of hormones, such as epinephrine and glucagon, epinephrine binds to an α adrenergic receptors in liver cells, causing activation of phospholipase C, phospholipase C catalyzes hydrolysis of ester bonds of phospholipids, specifically one type of phospholipids called phosphatidyl inositol bisphosphate (PIP_2), resulting in a molecule called inositol triphosphate, which binds to calcium channel in the membranes of smooth endoplasmic reticulum causing increasing concentration of calcium in the cytosol, activating calmodulin and then glycogen phosphorylase, therefore, glycogen degradation is activated by calcium in the liver also, but calcium release here is triggered by hormones, unlike muscles in which calcium release is activated by synapse.





What was mentioned in the lecture:

Regarding calmodulin, calmodulin is a small protein that can bind four calcium ions at saturation, binding of calcium to calmodulin is reversible, Ca^{+2}- calmodulin complex can act on many enzymes, one of them is phosphorylase kinase.