Animal Physiology
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Lecture – 34

Welcome back to the lecture series in animal physiology in NPTEL section. Today, what we will be discussing is, in the neuromuscular junction, how the energy is changed from electrical energy into mechanical energy. We talked about the neuromuscular circuit. Here, something is very interesting, what you have to appreciate first. Before I draw the whole circuit, you know how the energy transaction is taking place. What exactly is happening is that, say for example, there is signal a sensory signal, which is being sent by the sensory neuron, to this spinal cord or may be, to the higher centers of the brain. From there, through the ventral root, a mortal neuron is bringing the signal to the target organ, asking it; you move your hand like this or you walk like this, or likewise, xyz, this could be any signal.

Once the signal comes, signal is being transmitted as an electrical signal; electrical impulse; action potential; train of action potentials, ionic electricity is coming. But now, that ionic electricity eventually, the end result what we see is that, that ionic electricity leads to the contraction and the movement of muscle. How it does so? In other words, what is happening is, that ionic electricity which is being transmitted by the nervous system, is translated into some form of mechanical energy, via a chemical root.

Here, some molecules do that full transaction, what we call talk about the transaction procedure. There is transaction which is taking place; electrical impulses through a chemical root, translate into a mechanical energy, and that mechanical energy in the form of end output, what you see is the contraction of muscle. If you go back and think about what is happening in the stretch reflex art, in the stretch reflex arc, what is happening? There is stretch in your muscle; here is your muscle and there is stretch in your muscle. There are stretch sensors all over, what we call as intrafusal fiber or muscles spinal. They sense a mechanical change in them; a change in length and then, those sensors which are sitting here; they see the change, the mechanical change and they translate that mechanical energy into electrical energy; first transaction.

Then, that electrical impulse in the form of ionic waves, ionic current waves, travel all the way; from here, all the way to the spinal cord, fine. After coming to the spinal cord, then there is a computation process taking place. That computation may be supported by the higher mortal neurons, which are present in the mortal cortex of the brain, or may be, a very local computation, which is taking place at the spinal cord level, specially, in the border lines zone of the dorsal, where the information centers, and the ventral horn and through the inter neurons in between. Those mortal neurons which are sitting there, then brings back the message. All along this, it is all the electrical impulses which are getting transmitted; ionic electrical impulses.

Then, once it reaches the muscle at the neuromuscular junction, it transmits this electrical impulse to the muscle, and what the end result we see, there is a mechanical movement in the muscle, accompanied by an action potential. How it does so? Now, we will redraw the circuit and after that, there are three phases here. First phase is what I will do, I will redraw the circuit in a muscular junction, what I have just narrated you. Second thing is that, we will look at the ultra structure of the muscle, because that will help you people to appreciate, that how the ionic current electricity is translated into a mechanical force via certain chemical roots. Thirdly, we will club all these events together, and will appreciate this very wonderfully evolved phenomenon. So, lets us start with the section 5 and lecture 4.

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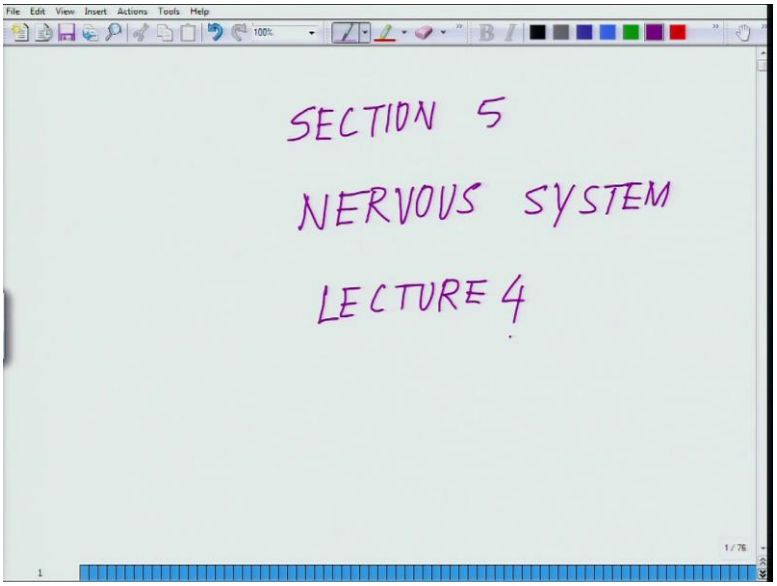

Section 5 on nervous systems, actually, it looks almost a part of the muscular system also, and lecture 4. Let us look at this circuit.

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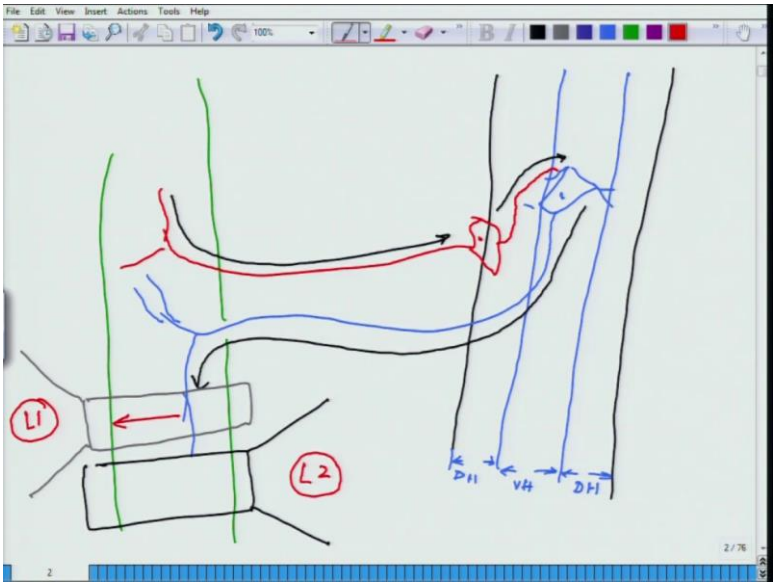

How it looks like? Here, is the circuit element; this is part of the spinal cord and here, you have the blue zone in between, is the ventral horn, VH. This is dorsal horn or the one which is carrying essential information, DH. The mortal neuron is sitting here. This is the neuromuscular junction. Here is the target tissue, which is a muscle and will, and here, is the sensory neuron, whose cell body is sitting slightly outside the dorsal horn. Here is sending path way. This is the local circuit where dorsal horn is synapsing on the ventral horn; dorsal horn neuron is synapsing on the ventral horn neuron.

Going by the arrow of information, this is the arrow of information, which is moving in the black. From here, it is reaching this; third level of computation and it is coming back like this. Today, our objective will be to look at the cross section of this muscle in detail, because this is essential to understand. Secondly, bit of further detail of this zone, these are the two zones, what we will be dealing today. One of the pertinent questions, which were asked, sometime in 1960, was this. The question was like, say for example, we have muscle like this; it is a thick muscle; or may be, some of the part of the body where are thick muscles. The neurons are coming all along this, and sending the signals. How the signal reaches all the way to the depth, because neuron is not really going all the way to the integrity details, wide, kind of cover two to three layers down. Why the

contraction? How that whole transmission taking place? If I have to draw, it will be something like this.

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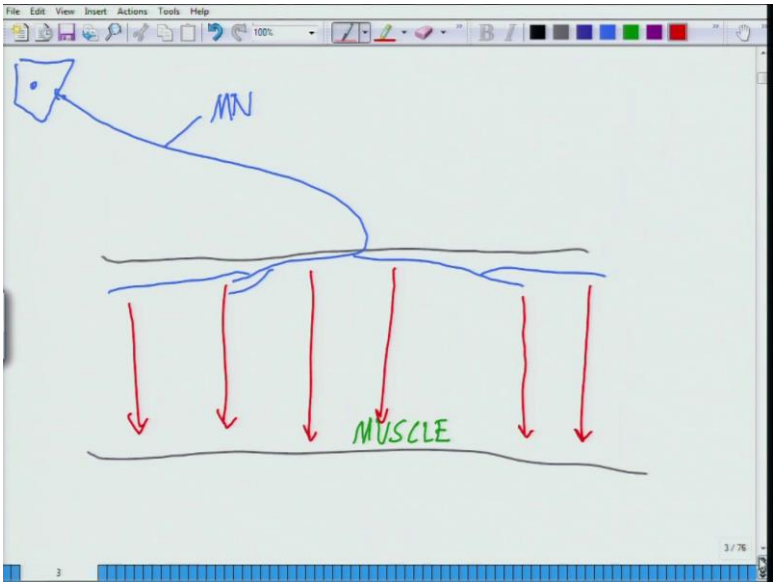

Say for example, here is a muscle. Here, is your target muscle and here, is a mortal neuron, which is coming; from the surface, it is out here for me, then your muscular junction. This is the cell body or the mortal neuron. Here, is your mortal neuron. Here is your target muscle. So, the governing question, which make people to explore this is, that how come the signal reaches all the way down, because these are very small ionic electrical current, and it has to travel all the way down. It may sound very simple, but it is actually, a very big problem. What promotes this kind of transmission? So, there it all started.

Going back to the previous slide, now, what we will do, I told you that there are two levels, what I am going to discuss. This is the level l; L1, I am putting it level 1 and this is level 2; these are the two levels. First level is the one, which I am now defining that how this signal reaches all the way down. So, this is just, I am showing like this, where the arrow is pointed towards like, towards on the left hand side.

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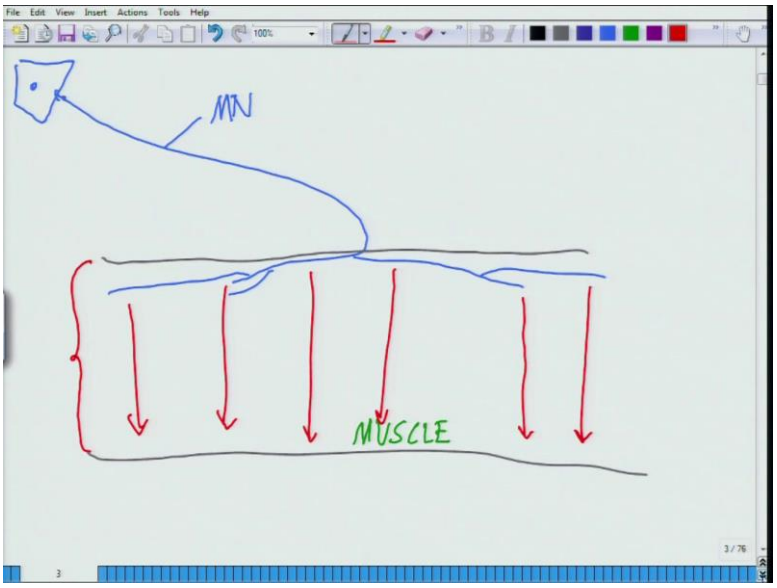

Here, how this signal is moving? In order to understand this problem, we have to look in depth to the structure of the muscle. How the muscle structure looks? If I have to draw muscle structure, before that, I am not drawing, at this stage, the ultra structure; ultra structure is much more different. The structure of the muscle is something like this.

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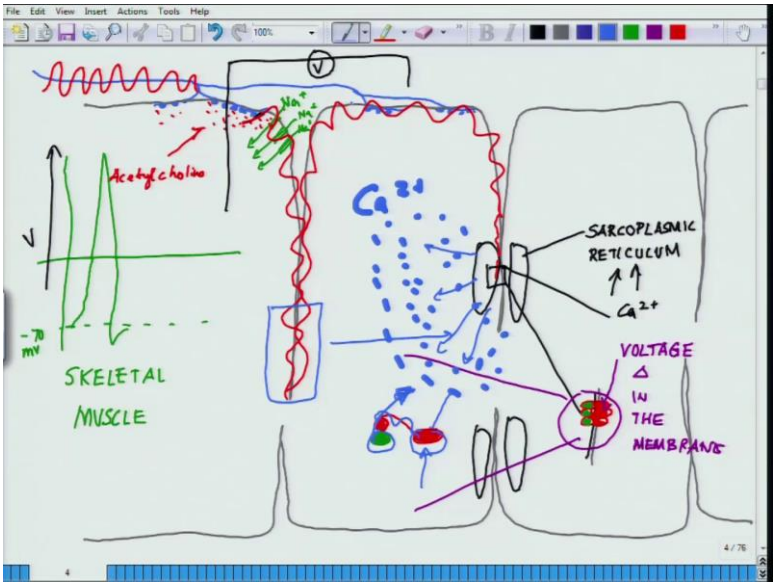

If you look at muscles' structure in the cross section, it is more like this. You see there, gorge kind of structures. Gorges are like, on two sides, if there are, if think of it, like, if you have mountains like this and there is small narrow cliff, narrow gap in between, that

is called a gorge. It is like a gorge like structures and muscles are like that. Come back to the structure of the muscle; let us move like this. Is this structural feature has a big significance, that how the muscle conducts all this pieces of information? This is your skeletal muscle.

Now, I told you. This is in blue, what I am showing is, wherever the neuromuscular junctions are getting formed, for example, a neuron process is happening, likewise, so on and so forth, fine. These are the zones of, these are hard zones where, you have all the neuromuscular junctions are getting formed, and these are where, that neurotransmitters are getting released, fine. In the blue button like a structure, what I am drawing now, this is where. Now, here comes an electrical impulse, fine and it reaches here. Once it reaches here, what it does? It leads to the, these are the neurotransmitters, let us say, in our situation, acetylcholine. So, acetylcholine secretion leads to the opening of iron channels out here; cat ions channel. These cat ions channels leads to the entry of the sodium inside the muscle, fine. After this, it is clear to all of you.

There are lots of cat ions, which are entering. Enter the cat ion channels, what it does, it leads to an action potential in the muscle. If the muscle was sitting like this, this leads to a rise in the, if it is sitting at minus 70 millivolt, this will lead to an action potential in the muscle like this. Now, the muscle is experiencing, if you have an electrode out here in a close approximately, somewhere out here, if you have, say for example, I place an electrode like this, with respect to the, here, and here, we have a change in voltage. So, you will be able to see a change in voltage like this.

Now, this electrical signal, which is generated by the muscle, because of the neuromuscular junction has a role to play. Now, what is happening? This electrical stimulus form the neuronal sight, now transmitted into the muscle. Now, this electrical impulse is moving on the muscles surface like this, and then, may be, it will travel like this, and likewise, it will travel.

While it is travelling, there is something very interesting happens. The membrane is getting polarized, depolarized; polarized, de polarized; polarized, depolarized; as this signal is moving all throughout, till it dampens down. But while it is doing so, it is a local phenomenon taking place at these junctures, because these are very specialized structures. So, what I will do, I will highlight this part on this section. Out here, within

the muscle, I told you there are some specific organs called sarco plasmic recticulam. They are all over, present mostly like this; out in this these are called sarco plasmic recticulam; this arc. These sarco plasmic recticulams are the store house of calcium. They have a lot of calcium in them, and it is very tightly regulated. So, what happens, when a impulse reaches here? For example, if impulse reaches here.

On the surface, as well as, it is kind of not very clear, on the surface of this sarco plasmic recticulam, whose membrane becomes continuous with the membrane of the muscle. If I have to again, look at the final details, it is something like that. If this is the membrane of the sarco plasmic recticulam, then this is the muscle membrane. They are almost on top each other. Out here, it is still not very clear, how it is like, kind of arrange; there is series of voltage sensors, sitting here and at two levels; at this and at this. Let me use different colors for you people to understand.

The one which I am now filling with green, are the second level of voltage sensors. One of them are on the sarco plasmic recticulam, side the other one, is pretty much at the border of the sarco plasmic recticulam, and the muscle membrane, likewise, which I have. Now, one of these sensors, again, it is not clear till date. One of these sensors changes a voltage change in the membrane, and as soon as it senses the voltage change, it is kind of heel joint, if I have to highlight this in much more depth, it is something like this. This one is a sensor and this one, as soon as it senses, it has a hook like structure like this, which is attached to the other sensor.

As soon as this sensor, this one which is in the red, this one senses that there is change in voltage. This sensor transmits a signal to this one, and this one rises up. What exactly happens in that process is that, this sensor which is sensing in the completely red color? Now, there is a bit of description, what we have to listen to me and process it slowly. Say for example, I showed you the intersection zone of it. Imagine, this is one sensor; this is another sensor; my two hands like this.

On your right hand side; this one; this one is the one, which is sensing the voltage. There is a change in voltage. What it does? This one is attached to the sarco plasmic reticulam because this is at the border. Once this senses, this ask it to open up like this. As soon as this one opens, from sarco plasmic recticulam, I told you, there is huge amount of calcium goes out.

What essentially happens at this stage is this. If this receptor, the one which I was showing with my right hand side, if this one gets activated, this one leads opens up a door out here; which opens up, lift the cap and then, what you see the essentially immediate out here, is a situation like this. The series of molecules which are all over the place for a very transit period of time. These molecules are ca into plus; calcium. What calcium does? Before I say what calcium does, let us talk about the nomenclature of these different receptors. Here, what you have is, now, if I will show you in terms of, in much more bigger detail, it is.

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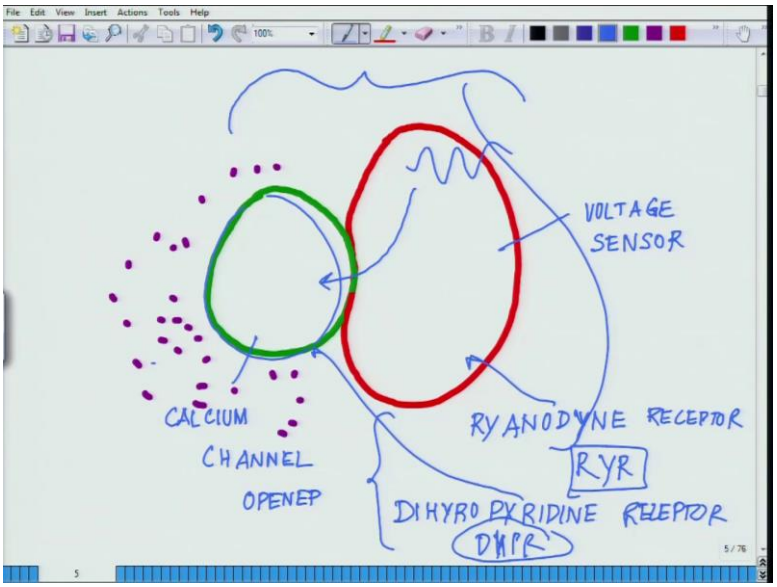

It is one which is opening up on top of the sarco plasmic recticulam. Another gate is out here, which is in close proximity with it, which is the voltage sensor. Put the names now; here is the voltage sensor and here, is calcium channel opener. This is sensing a voltage which asks the calcium channels to open up. Here is a voltage sensor and this is asking it; come on, you have to open up. As soon as this opens up, the next thing what we see, along this whole milleu of the muscle is, there is lot of calcium which has come down from the sarco plasmic recticulam. Out of this, it has been discovered that this complex consist of two different proteins, which makes this voltage sensor and the calcium channel opener. One of them is called Ryanodyne receptor; sometime in short, it is called RY receptor.

The second receptor is called Di hyro pyridine receptor; DYPR, sorry, DHPR, I think. That is how you pronounce it; these two receptors. What is the problem is that, what is

not clear is that, which one is which one? They remain as a complex like this, something like a complex like this. If this one is the Ryanodyne receptor, then this one is the hyro pyridine receptor. So, what is not clear in this complex at the molecular details yet, to mankind is, which one is sensing voltage, and which one is opening up the calcium channel? There is one which is actually opening up, or there are two situations here; some people believe that one of them is a voltage sensor, and the other one is a calcium channel opener, fine. It acts like a hinge like this, fine. There is another set of beliefs, which says that both of them are doing both the jobs together. So, there is nothing like, I will do this and I will do this; nothing of that sort; they will do it together.

This part is still, a very hotly debited topic specially, this Ryanodyne receptor and hyro pyridine receptors have been very extensively studied in the cardiac system, specially, in the cardiac excitability, because one thing, you people have to realize. These are very programmable channels or programmable structures, because it not only sends out this calcium; it has mechanism by which the sarco plasmic recticulam, pulls back the calcium. It is just like, I take a bucket of water, drop the water in a room. Immediately, within no time, within delta T time, water has flown. I have a mop and I just mop it out, back to the bucket, from the bucket where I.

So, they have very tightly regulated mechanism. It is not like that; I drop a bucket of water and then, let the water dries, happen; it does not work like that. Biology is a very well controlled system; it troughs the bucket, immediately, in no time, pretty much no time, by the time it has done its job; it is put back. Because it remains for a slightly excess movement, that will change the way our muscle twitch, the way signals are getting transmitted, that will be completely compromised. This is something, what you people have to appreciate, that it is not that simple; it is all happening at a very narrow zone of time, again within few pico second, pento second or nano second; all at the max at a micro second level. These things are all getting over. These molecular reactions are very well coordinated and very fast.

As of now, it is not really clear, out of this, Ryanodyne receptor or Di hyro pyridine receptor, which one is really doing the sensing job, and which one is really opening up the channel? This is a kind of, still hugely debated topic in the field of neuromuscular junction and muscle biology, cardiac physiology and all. Now, the next part in the question comes. There is a secretion of calcium and then, the end result, what we see that

there is muscle contraction. What calcium does? How come calcium is essential for the muscle contraction to take place? As of now, we are all being talking about, there is voltage sensing mechanism by which, there is, all of us like, at that particular movement of time, the whole muscle milleu is all, there is a, if I have to flop the calcium wave, it will look like this in this diagram.

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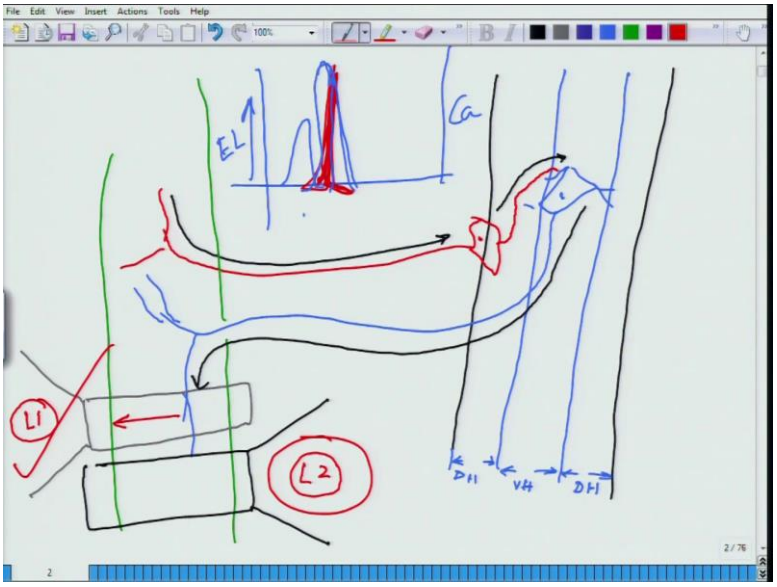

What I will see essentially is that, as soon as this signal reaches, as soon as if I have some way to plot it out here, the first signal is the neuromuscular junction signal. This is the neuromuscular junction. The next thing, very soon, I see there is a wave of calcium. If I have the y axis telling calcium, I will see a wave of calcium; a calcium wave going up and this is the electrical signal, fine. What this calcium wave does? What this calcium spike? I am showing spike, because I told you, that calcium does not remain there forever.

As soon as it is being (( )) even faster than that, it is being pulled back by the sarco plasmic recticulam. You cannot afford to have calcium sitting there and lofting around. Then what this spike actually leads to? In order to understand, how this spike has some role to play in electrical to mechanical transaction; that is what we are going to discuss in the part 2 of this, which is essentially, this part; L2. In L1, we have understood now. This is what is happening. That is how a signal, when it reaches here, through that gorge like a structure, they reach all the way down, and then the electrical signals are reaching all the way down to the muscle.

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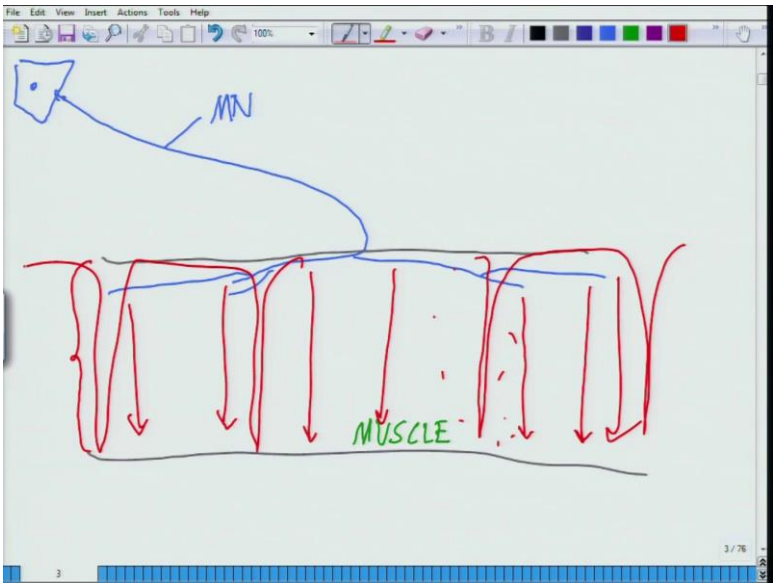

Now, we will be talking about how that is bringing about.

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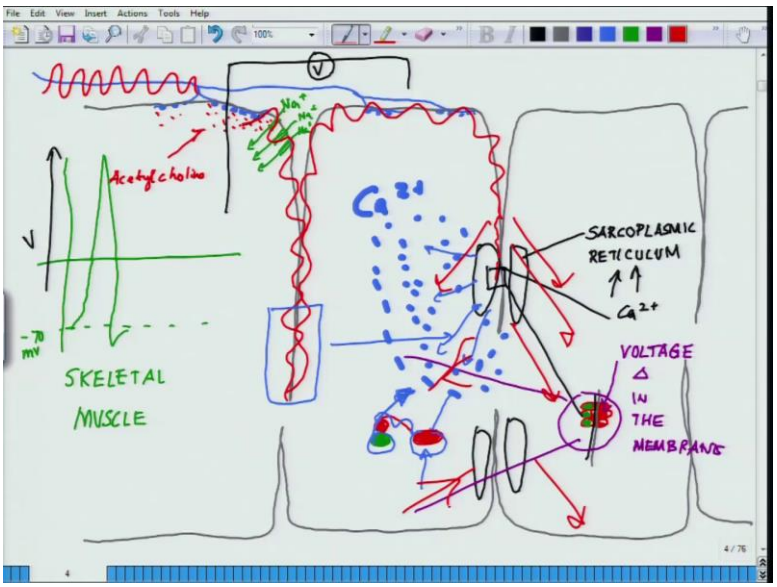

This is how it is happening. It is reaching all the way; the muscle is receiving all the electrical signal, all throughout like this. Now, we will talk about the structure of the muscle.

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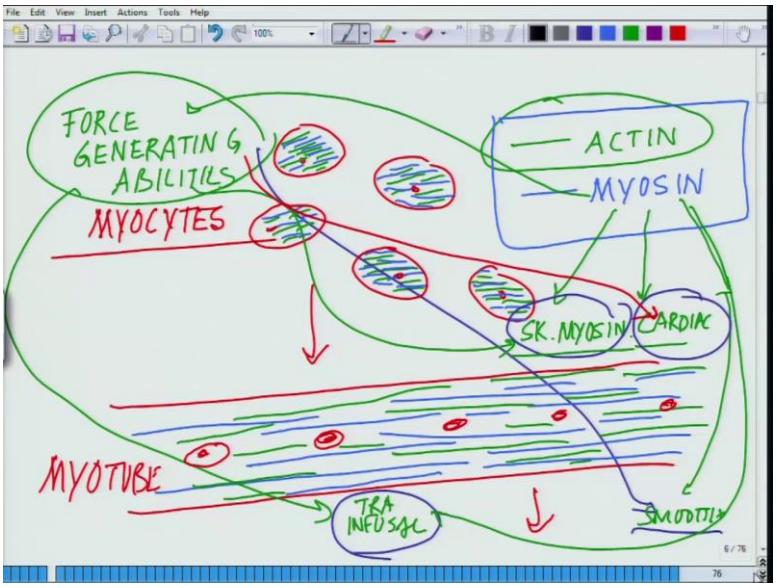

As of now, what we have learnt about is, you have individual muscle cells, I have already discussed this part. These muscle cells are coming close to each other and forming the myotubes; these are the nucleus; these are the nucleus; these are the nucleus; these are the nucleus. These are the myocytes and these are the myotubes. What I essentially did not mention? Eventually it forms myofiber cells and everything. But I essentially did not mention at that point of time is that, these fibers, these individual muscle cells have a crisscross of two different kinds of proteins, I will come to that, what are those. All of them are like this; they are aligned like this in individual cells, at different orientation, like this. When the muscle is attaching and forming a myotube muscle cells, these proteins align themselves like this.

Let me tell you this is not Ryanodyne, we will come to that. These are the protein fibers which are constituting the muscle. What are these proteins? There are two proteins which are making this muscle structure. Here, the green one I represent as actin filament, and the blue one I represent as myosin filament. Here, I wish to highlight something. This myosins acting fairly is well conserved; this myosin filaments have different variations, different variants. There is skeletal myosin, is different as compared to myosin of cardiac muscle; they are different. Myosin of smooth muscles is different. Myosin of intrafusel fibers is different.

So, there are different variants of myosin. What does that mean? That means, in terms of functionality, they have different force generating abilities and that essentially, means

who is generating ability. Now, if you look at it, intra fusel fiber needs different kind of force, whereas, skeletal myosin needs different kind of force; cardiac muscle needs different kind of force; smooth muscle needs again, another different kind of force. So, how this is being regulated? This is being regulated by these different kinds of myosin fiber. But is it random? If during 1960, there is enormous amount of research which was done, it was figured out that this is not random; this is very well organized. How this is getting organized? Well, I have to do; these are the pictures what you see in the books.

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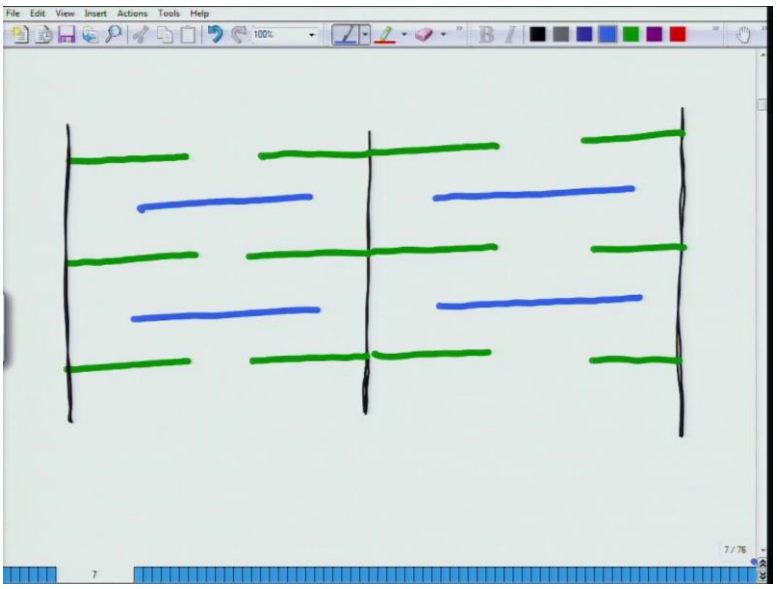

They are kind of, tough to figure out. I will draw a simple picture for you people which will clarify your doubts, that how the cross section of the muscle cells. This is what we are doing, we are going through a cross section of a myotube or myofiber specially, let us think of simple in a unit as myotubes.

Let me see what is the color coating I am using, I think actin is in green, fine. Keep actin as green, take a bigger marker, fine. Now, actins are arranged; these are the green actins. Now, we are looking at the real cross section of a muscle. Now, the other side, we have like this, like this, like this, like this, like this, like this, like this, fine. Now, we have another component which is a blue component, which is all myosin, right, fine, the myosin component which is in blue, fine.

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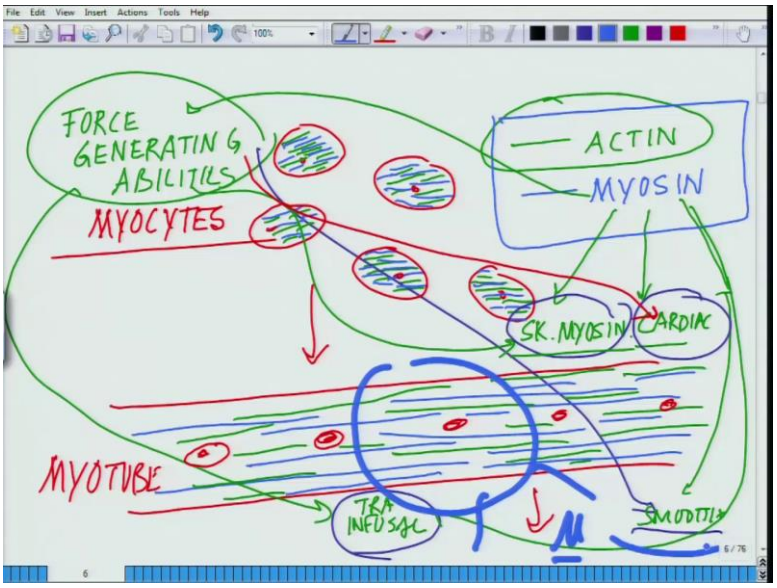

What I was trying to draw here, you see all these random patches out here, what you see here; they are not actually random. Now, I am looking through the microscopic structure what you are seeing now. Here, we have the myosin filaments.

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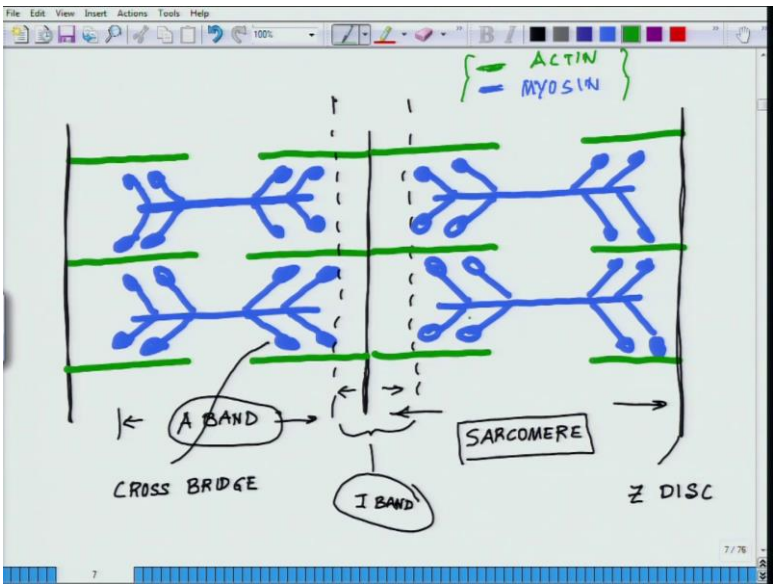

The myosin filaments are not that simple as you think. These filaments have a head like structure like this; will come to the ultra structural details of those, likewise, likewise. They have this kind of a structure, like this, like this. These are the myosin heads. We will talk about what those head means, likewise, likewise, likewise. Then you have like

this, like this, like this, like this. These are the structures which you will see in text book; a bit complex structure. That is why I am just taking that additional step to ensure, that you people understand the whole bio mechanics involved here.

This is how the basic structure looks like. Within this structure, this small, this unit is called a sarcomere. This unit is called a sarcomere. Be very carefull, because that is why, I am drawing it in real time, so that, you people understand this. This is called z disc and these are the terminologies, which I cannot help; you just have to, people have to kind of remember all, appreciated. This zone is called the cross bridge zone. On top of this, this particular part, this part which is another microscopic this means, say for example, I draw a line like this; draw a line like this; where you only see another microscope the actin bands, you have not seeing the, once second. This is called, from here to here, is called I band; this is lighter band. Then from here to here, what is being called the A band; I band, A band, and let me give color coding. This is your myosin and this is your actin, fine.

So, we finished all the color coding. This is very essential. Now, what I will do, I will come back to this drawing. Before I come back to this drawing, I will give an idea of the ultra structure, because I do not want to make it very crowded. Let us talk about the ultra structure. What are color codes, again? Myosin, let us talk about the myosin.

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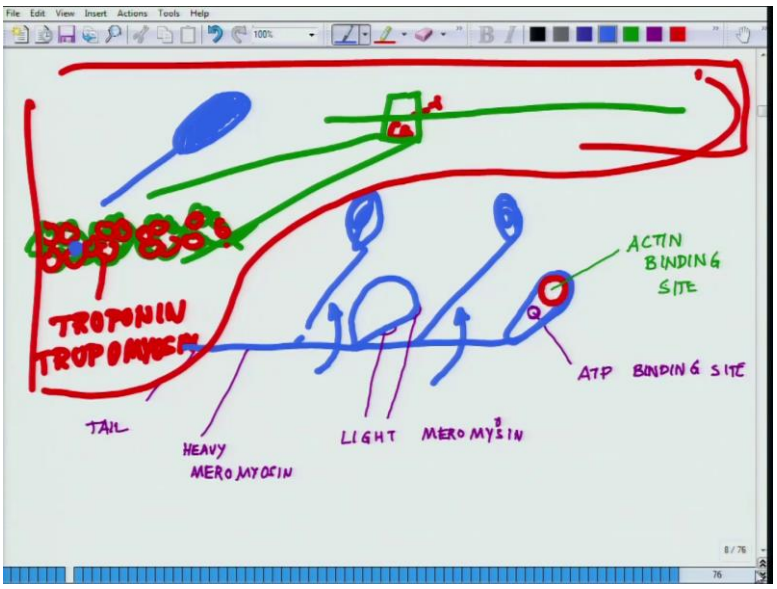

While I was drawing the myosin, it looks like; I was drawing it like this. I told you that there is myosin head here. What I will do now, I will draw the, it is like this. This head has the ability to move like this. It can take a shape like this, essentially, it can move like this. This actin, sorry, this myosin head has a zone which is called, be aware with me; it is a complex drawing. This is called actin binding site. There is just underneath it, there is something called a small zone out here. This is called ATP binding site; the energy molecule, fine. There are some other nitty gritty details, which are not really essential for you people, these are called light mero myosin; you do not need to know this, but since, if you come across in the book, do not get confused.

Out here, you have the heavy mero mysin. These are different variants of myosin, sorry, myosin, This is the tail of the myosin, which is dangling out there. Now, what we will do, we will draw the actin simultaneously, that will help you to appreciate, how the actins look like. Here is the actin filament which is in green; yes, actin is in green, fine. Actin has something called, this actin is something like, if you have to take a cross section view, it will be look like this. It will be more like a spiral kind of a structure of different proteins, likewise. It looks like this, the way I am drawing it, different proteins.

If I represent the proteins like, sorry, like this, different proteins sitting out there. Now, if you look at this structure, the way it looks like is, within the actin, you have something called a troponin, where proteins out here, which are called, we can represent them, I cannot. Troponin and tropo myosin and underneath that, you have the calcium binding site. This is very important, because I told you that this calcium leads to the mechanical energy transaction; from electrical to mechanical energy transaction.

Calcium binding site is somewhere out here. These are kind of, so imagine, this is a calcium binding site; on top of that is that troponin. What exactly happens in that situation? Let us go back to the basic drawing again. What exactly is happening is this; whenever, look at this drawing, whenever, under a normal condition, the contact between, under normal condition, they are not in touch, fine. There is no touch. Look at this arrow; there is a gap out here. Physically, they are not in touch; they are close to each other, but they are not really bounded by anything. They are loose and they can move like this; there is not a problem.

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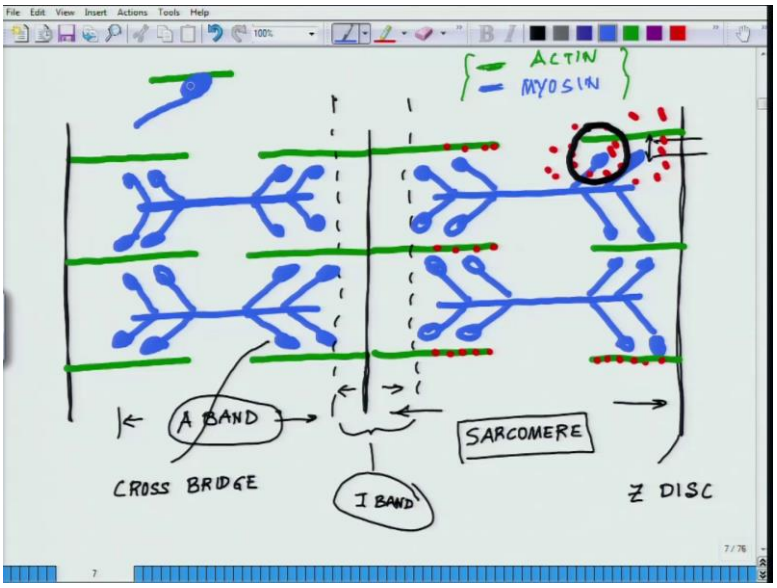

But as soon as the situation comes like this, calcium comes like, now, this is the part what you have to understand. Now, there is calcium in flux, because of; the calcium comes and binds to the actins. This is the calcium binding in red, I am showing. As soon as the calcium binds there, it removes the blockage. The blockage is being created by; the binding between actin and myosin cannot take place, because it is the troponin and tropo myosin which blocks that. Say for example, if my cell phone is here, and if this is the myosin; myosin filament.

Then, here is the actin which is like, this hand; which is actin. Actin has this troponin, tropo myosin and all this, is this cell phone. So, it is like this, fine. Now, the calcium comes. As soon as the calcium comes, what happens? My mouth is in calcium now. In other words, what happens? The calcium comes and removes this blockage like this, exactly, something like that happens. At the molecular level, the calcium comes and it removes the troponin, tropomyosin blockage. As soon as that blockage is removed, the next thing what happens at this zone, if you look at in depth at this zone, what is happening at this zone? Now, the myosin head is in direct contact with the; here is the actin and here is the mysosin head, is in direct contact with actin. Now, if you go back to the next thing.

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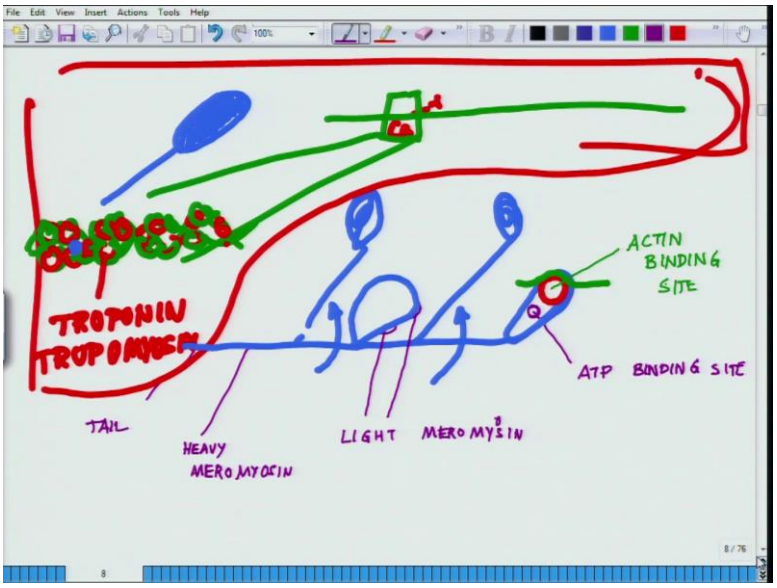

Here, it has ATP; actin binding site. This actin binding site is now active. This is in touch with the myosin, fine. Now, at this stage, there is an ATP molecule which binds here. This ATP molecule helps to give it up, one second.

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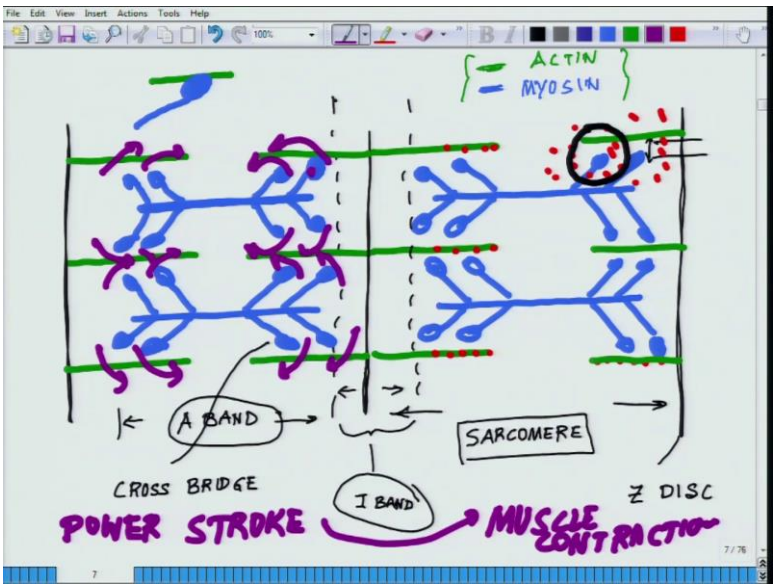

This ATP molecule comes back as soon as this is; now, think of it is like this one or bigger. Now, this is in contact and there is an ATP molecule, which comes and binds here. Let me represent ATP with some other color so that, the confusion is not there. This ATP molecule leads to a motion like this. This motion, imagine now, this motion is

taking place in all of them like this. Essentially, these all can move to other direction; these to other direction; and these ones to these direction.

All of a sudden, it gives something called a higher stroke; this is called a power stroke. This power stroke is nothing, but your; which leads to muscle contraction. Now, if you think of it, what exactly happened in this whole process, I told you that, initially, there are these nerves; let me go back to the slides. That will help you to appreciate, now what happened; first slide.

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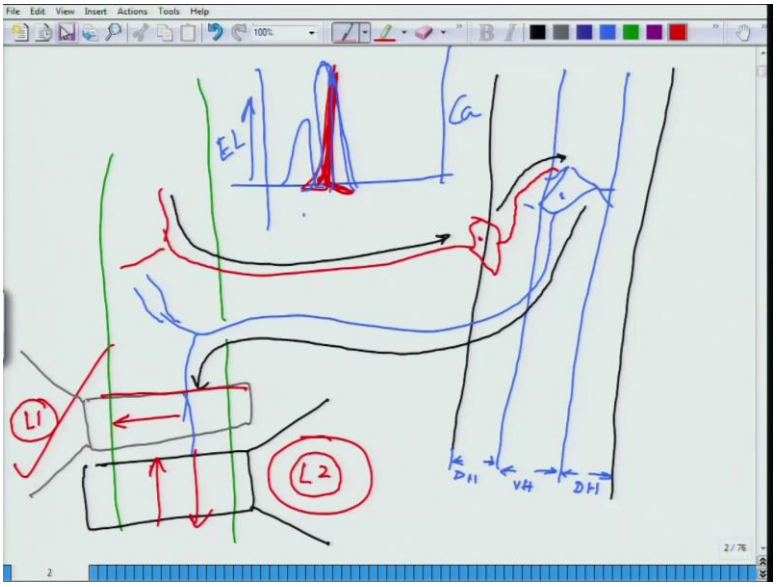

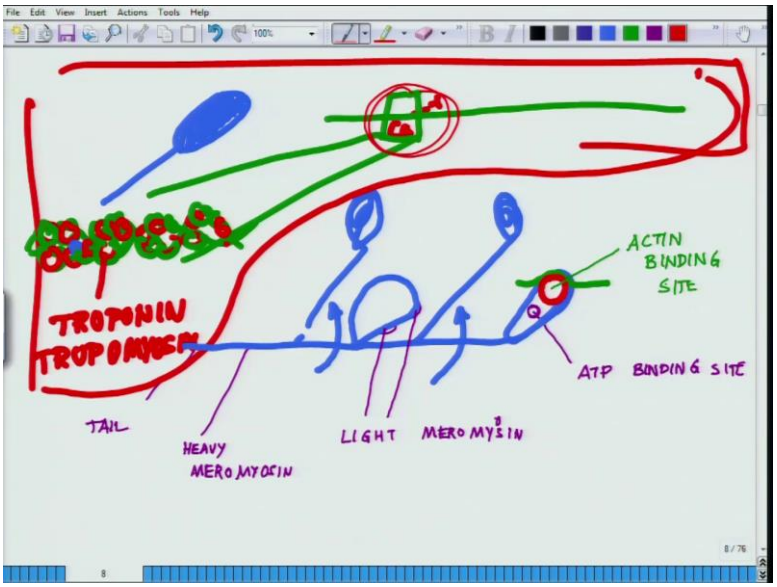

Here, the electrical impulse came and this is transmitted through the Ryanodyne and sarco plasmic recticulam; the Ryanodyne and dy hyro py ridine receptors. Then there is a rise in calcium and I showed you that calcium spike, which took place. That calcium spike leads to a motion in the muscle. That was essentially done, because this calcium came, because of the rise in calcium, this calcium which clot, grows up here. You see this calcium; this calcium essentially went here, and bind to the actin molecules. After binding to them, as already I showed you, it has an actin calcium binding domain out here, within the actin molecule.

It binds to the actin molecules and removes the blockage between the actin and the myosin; if this is the actin; this is myosin; there is a blockage, because of some of the troponin tropomyosin. As soon as calcium comes here, it binds it and this get, and it has a power stroke using ATP. That is how, the muscle contraction takes place. This is essentially, is an electrical to mechanical energy transaction process, which is very fundamental, all across the living system. This is what I wish you people to appreciate. So, I will close in here. I hope you people have understood this concept, if you go through any picture online or book, now everything will make sense to you people.

Thank you.