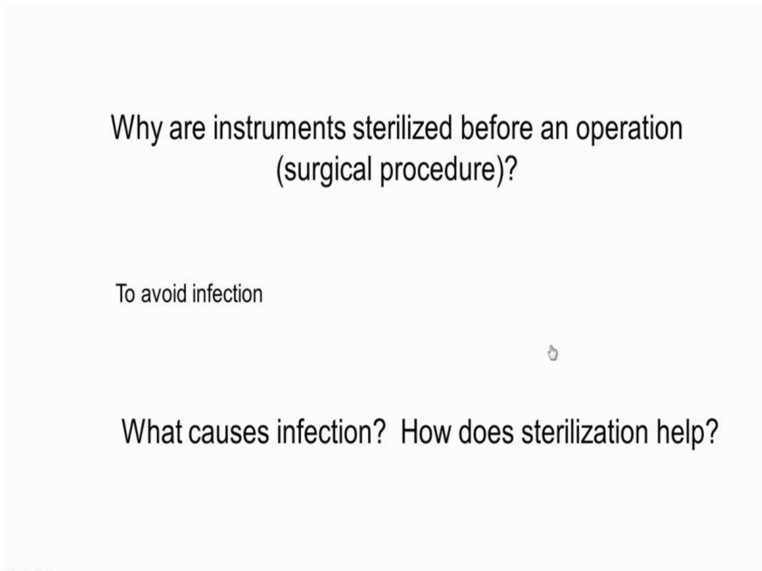
Biology for Engineers and other Non-Biologists
Professor G.K.Suraishkumar
Department of Biotechnology
Indian Institute of Technology, Madras
Lecture Number 05
Biomolecules: Lipids

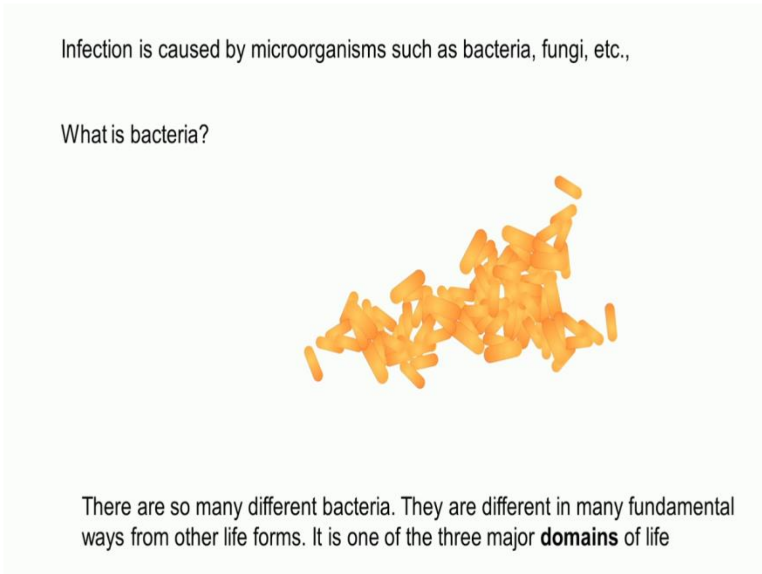
Welcome! We are discussing 'Biomolecules and their relationship to the cell structure and function'.

(Refer Slide Time: 00:44)



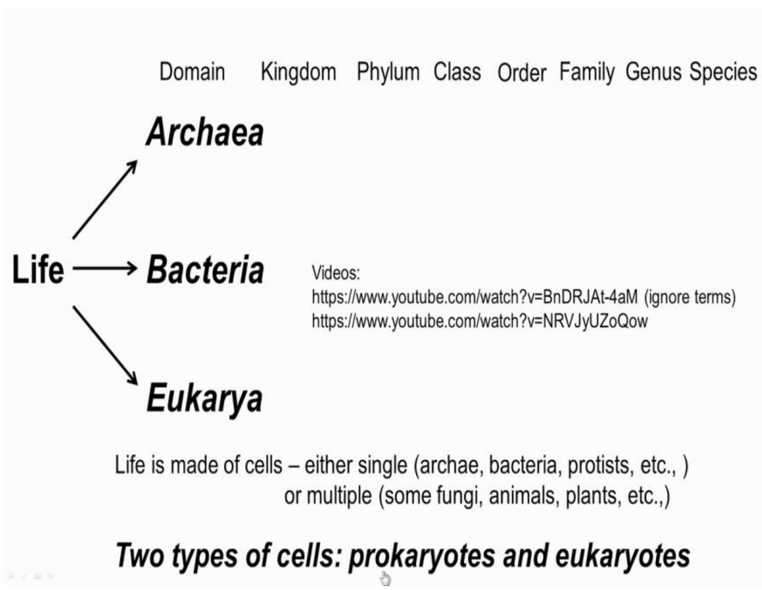
We saw in the last lecture, the micro-organisms, and to see that we had a storyline, we were looking at why instruments are sterilized before a surgical procedure and we asked a few questions and we came down to bacteria which can cause infection, which is one of the things that can cause infection.

(Refer Slide Time: 01:04)



And there are so many different bacteria in the world, in life and so large that and so fundamentally different from other varieties that we call that a domain. Life, we said had three domains; bacteria, archaea and eukarya, which are different in some fundamental ways, and these are called domains.

(Refer Slide Time: 03:03)

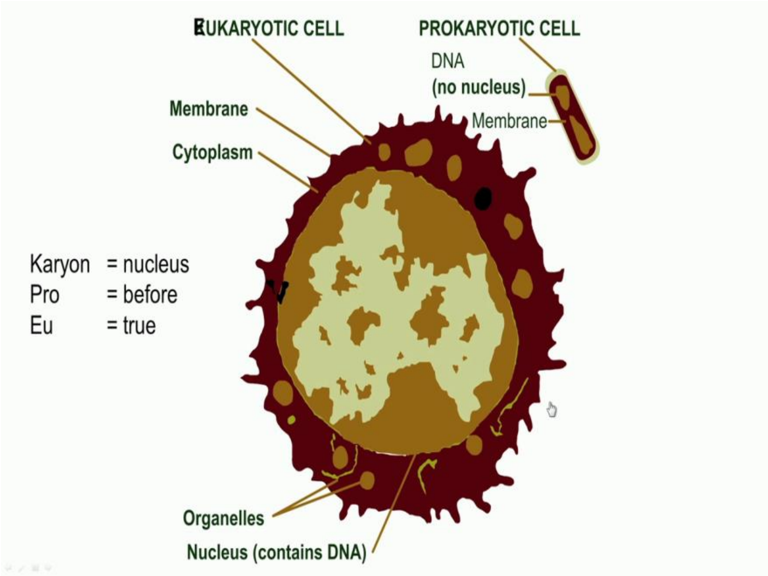


There are many kingdoms in a domain, there are many phyla in a kingdom, there are many classes in a phylum, there are many orders in a class, and there are many families in an order and there are many genuses in a family and many species in a genus. This is the taxonomy, as it is called, or the way organisms are classified so that there is no confusion. Hopefully you have gone through the videos here. We had mentioned that you could ignore some of these biological terms. They are just being, you can, you can get used to it without really knowing what they are. We will know most of the important things as a part of this course, you don't have to worry about that.

So you could ignore the terms such as RNA, DNA and so on so forth at this stage and watch this, and watch about let's say two thirds of this video. I talked about mnemonic to remember this. Do Kualas Prefer, I said Cheese Or Fruit, I think the video says Chocolate or Fruit Generally Speaking. That's a mnemonic to remember this. And then we said that life is made up of could cells, either single cell, such as in archaea, bacteria, and some part of eukarya called protists and so on, or they could be made up of multiple cells such as fungi, animals, plants and so on. I said humans are made up of about ten power fourteen cells.

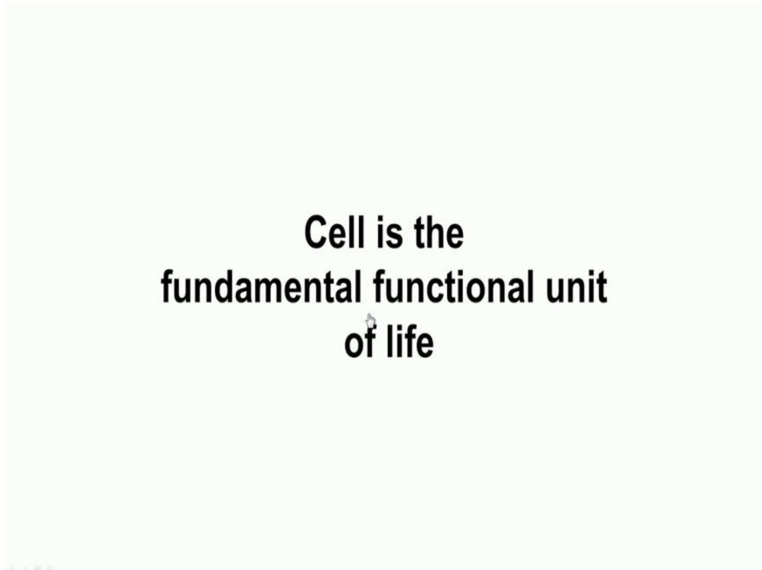
And basically, there are two types of cells, prokaryotes, before nucleus, and the ones with a true nucleus, we saw how they looked, and their sizes and so on and we also said that cell is the fundamental functional unit of life.

(Refer Slide Time: 03:09)



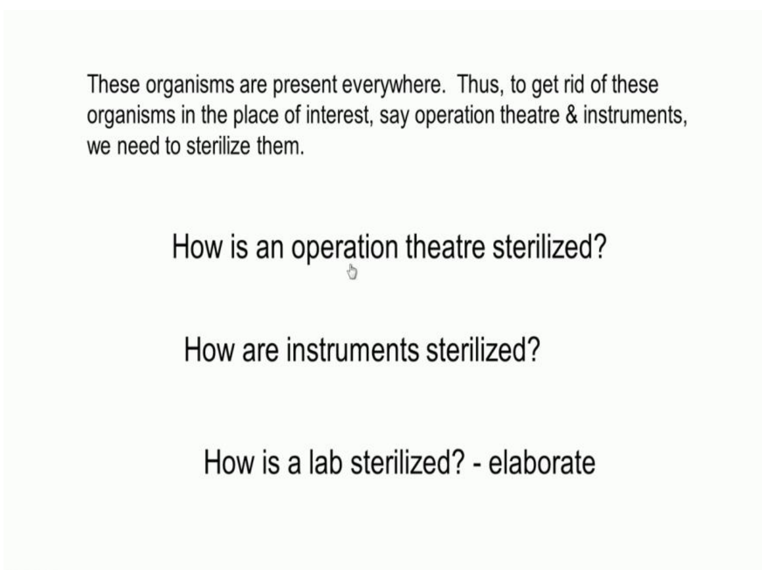
If we understand this, then we have probably understood life, and, we are nowhere close to this.

(Refer Slide Time: 03:21)



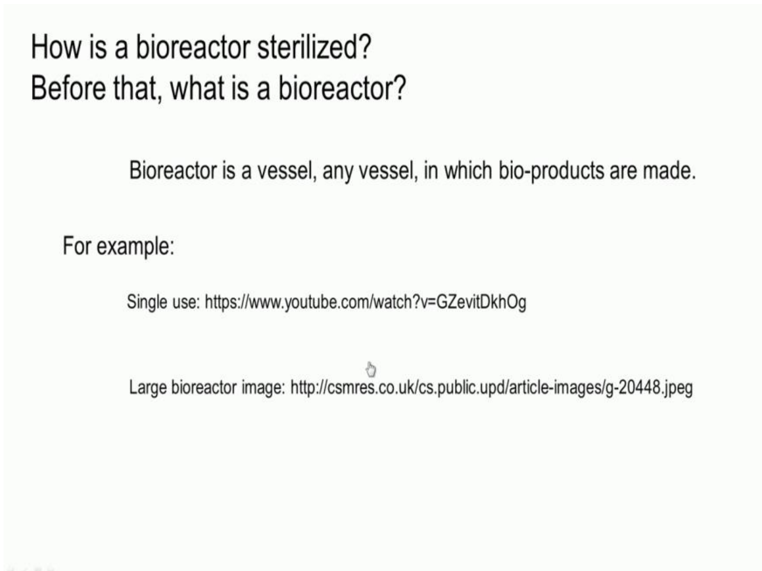
And then we talked about organisms and so on so forth and I told you how a lab is sterilized, how that can also be used to sterilize operation theatres and hospitals, and there are other means, other (mo), other more safer means of sterilizing operation theatres and instruments.

(Refer Slide Time: 03:42)



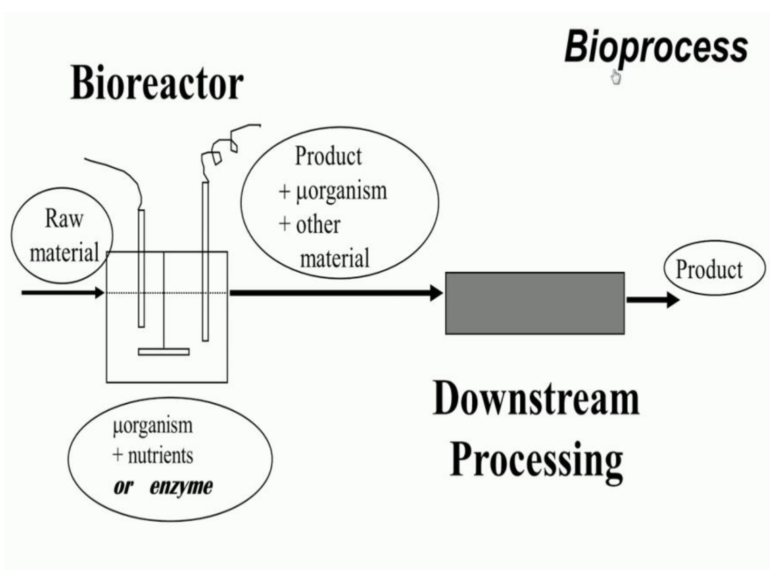
Then we talked of bioreactors; bioreactors are the production vessels. In fact, a bioreactor is a vessel, any vessel in which bioproducts are made, and there are so many bioproducts of use as we speak and there are many more potential bioproducts being developed.

(Refer Slide Time: 04:16)



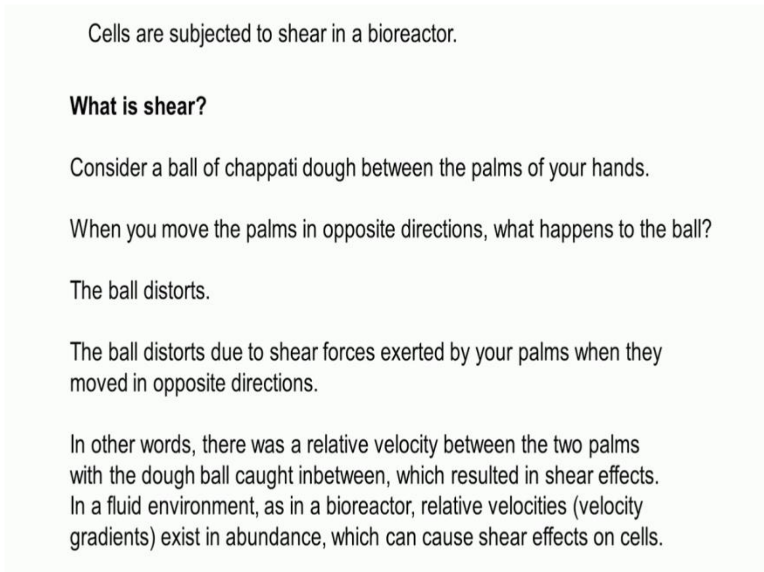
I had given you a video for a single use bioreactor and an image link for a large bioreactor, bioreactors can be very large depending on their application. And then I told you a bioprocess which is what is actually used to produce bioproducts, bioreactor is an important aspect of it.

(Refer Slide Time: 04:25)



And then, we said that cells are subjected to shear in a bioreactor. This is going to be our second story. Our first story was about infection and so on so forth. We learnt that organisms cause infection and what organisms there are, how they are classified and so on. So our second story starts with shear in the bioreactor, okay?

(Refer Slide Time: 06:28)



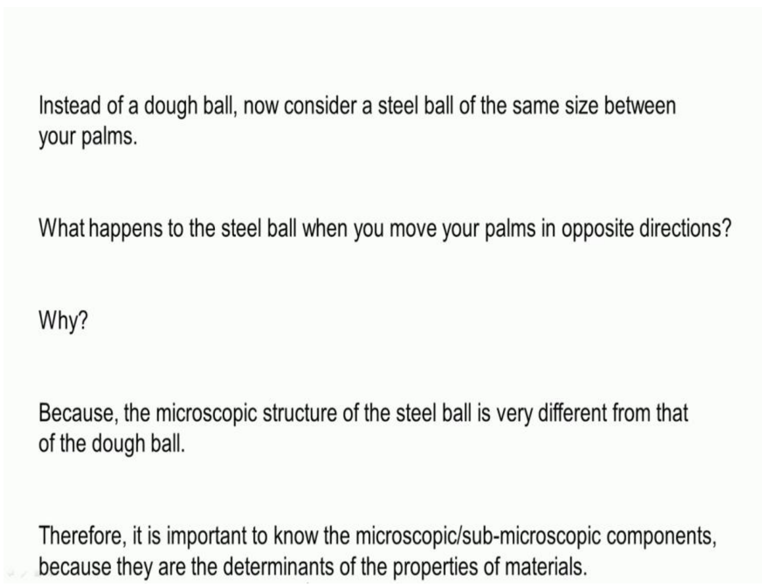
This term shear itself could be new, even to engineers who are students. If you have done, probably a degree in engineering, you would know what shear is or related fields, you would know what shear is. Let's start from the very basis, and then I'll tell you what shear is, that way we are all in the same plane. Let us consider a ball of 'chapati dough'. Okay? And place the chapati dough, puri chapati dough in between the palms of your hands, place your palms in opposite direction. I think I've kind of (step), given the various steps here. Slightly rest these palms and move the palms in opposite directions.

When you do that, what happens to the ball? The ball distorts, the chapati dough, okay, flexible, that distorts. The ball distorts due to the shear forces exerted by our palms when they're moved in opposite directions. Okay? The ball distorts when, because of, what are called shear forces that are applied on the surface. These are exerted by the palms when they are moved in opposite directions.

Okay? In other words, if you have some background in physics, you would understand this a little more easily when there is a relative velocity between the two palms with the dough ball caught in between, then it results in shear stress, or shear effects, as you can call. Right? So this is a requirement for shear to arise that there has to be a relative motion, which means one part of the dough ball in this case, the upper part needs to be experiencing a velocity that is different from the other end of the dough ball, let us say, and therefore there is a relative velocity between the two ends, and if that happens, there is a shear force that is experienced by the dough ball, okay?

In a fluid environment such as a bioreactor; bioreactor is a vessel in which bioproducts are made by cells. The vessel is typically sturd to keep the cells in suspension, it could be operated in many ways and so on, let us not get into that. So if it is sturd, there is going to be a lot of velocities, velocities in speeds in different directions, different velocities of the fluid particles in the bioreactor. And such an environment is very rife for a lot of shear forces to act. So, in a fluid environment as in a bioreactor relative velocities, or velocity gradients exist in abundance, which can (caurge), which can cause shear effects on cells.

(Refer Slide Time: 08:50)

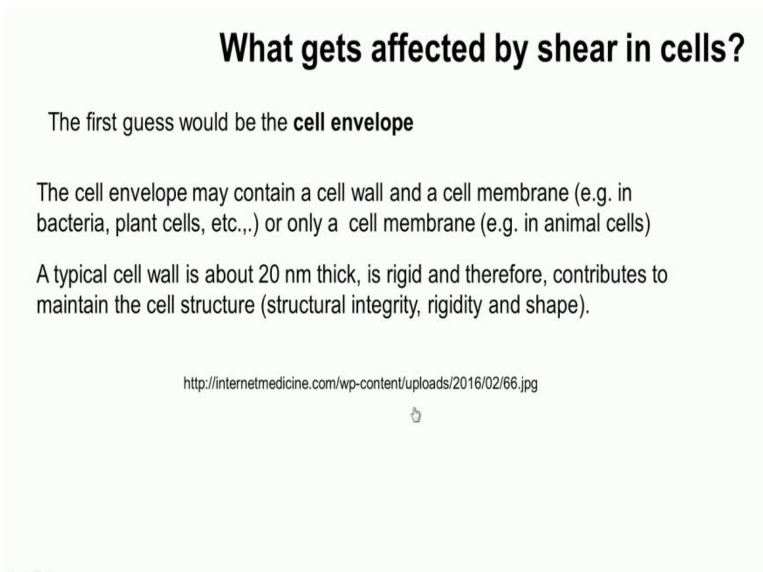


Now, instead of a dough ball, let us consider a steel ball of the same size between the palms. And what happens to the steel ball when we move the palms in the opposite directions? Pretty much nothing, right? It does not distort, in other words. Why? Because the microscopic structure of the steel ball is very different from that of the dough ball. You all know that. Dough is made up of something which we will, which you will probably be able to guess as a part of this course and steel ball is made up of steel, it is made up of steel molecules or iron plus other combination and so on and so forth.

And the microscopic structure of steel is very different from that of the dough ball, and with the shear forces that one is able to exert, just by moving the palms in opposite directions, nothing happens to the steel ball. Therefore, it is important to know the microscopic or the sub-microscopic components because they are the determinants of the various properties of materials, right? This you would all be familiar with, different backgrounds, you would all be familiar with this concept that it is a microscopic structure, or a sub-microscopic structure and components and their properties that determine the properties of materials as a whole.

Now let us ask the question. What gets affected by shear in the cells? Let us say we know nothing about cells, we have been talking of various terms, I am sure, even in the other lectures by Dr. Madhulika Dixit, she would have mentioned a lot of terms we will clarify all the required ones, in this, as a part of this course at different points in time. You do not have to worry about that.

(Refer Slide Time: 11:30)

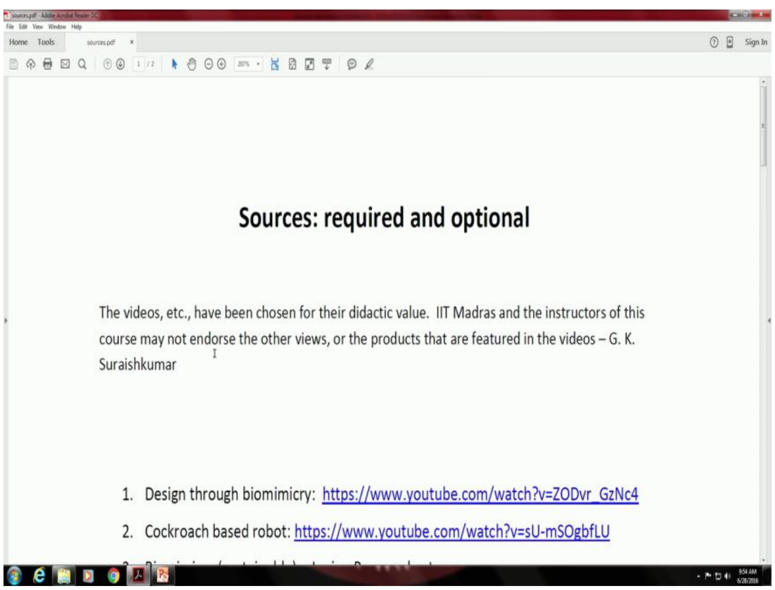


So what gets affected by shear in cells? The first guess would be the envelope that covers the cells, right? It is, it is a very rational guess. So what does the cell envelope contain?

The cell envelope may contain what is called a cell wall and a cell membrane. Remember these two are different things. One is called a cell wall, the other one is called a cell membrane, and both of them envelope the cell, right? The cell wall and the cell membrane both are found in some bacteria in plant cells and so on, or the cell wall could be completely absent, okay? Only the cell membrane distinguishes the cell from its surroundings, right? And that is the case with all our cells, all animal cells do not have a cell wall, they just have a cell membrane and that is what is exposed to the environment.

Typical cell wall is about twenty nanometers thick, okay? You can imagine the size, meter, then centimeter is ten power minus two meter, millimeter is ten power minus three, micro is ten power minus six, nano is ten power minus nine, okay? This twenty nanometers thick, is rigid and therefore contributes to maintain the cell structure. So just integrity, rigidity, shape and so on and so forth, that is typically what a cell wall does. You could look at this particular link here; by the way I think this is the right time to show you the various links. I did mention a file, a pdf file that would have these links that can be clicked, I think control and a click would take you there. Let me show you that file. I think it is the right time to show you that. Right?

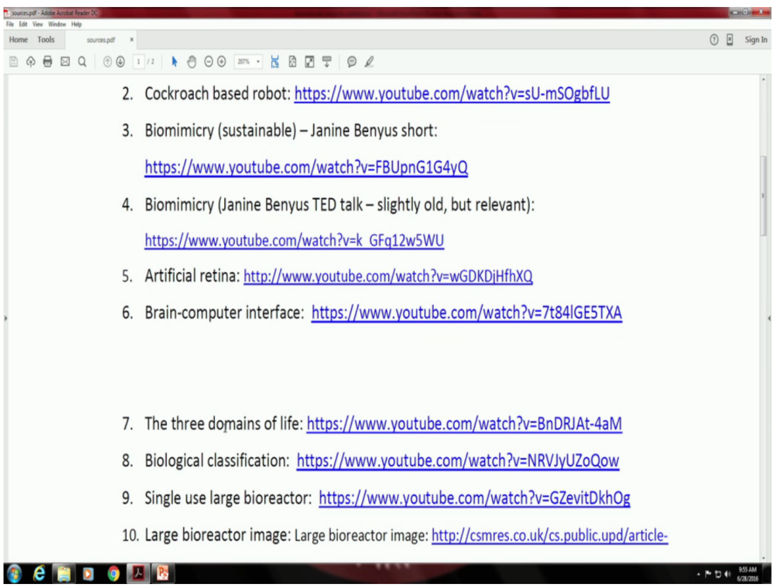
(Refer Slide Time: 12:43)



I this would be available as a part of your resources, I think it is available right below or right above I think there (were) there is a link, a button on the right hand side of the video, when you play them. And if you click that, it will take you to this file. So these are sources, many of which are required, and some of which are optional. I will point out which are optional. If it is, if I do not say it is optional it is required, okay, required to appreciate the course better. As I mentioned, we cannot play these videos here, therefore we have given you the links so that you can go and take a look at them. The videos have been chosen for their didactic value, for their, for their value to teach.

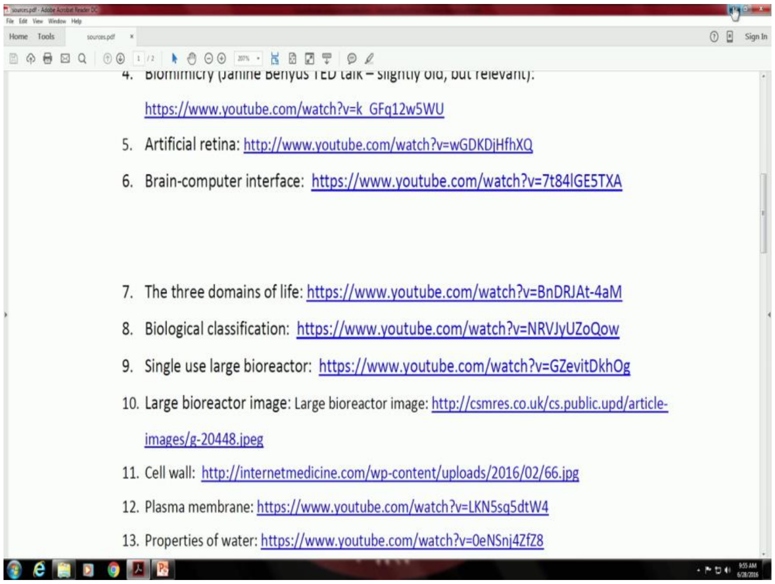
IIT Madras and the instructors of this course, both Madhulika and I, may not endorse the other views that are expressed in the video or the products that are featured in the videos. Some of these very nice videos have been made by companies to sell their products. But we do not endorse the products.

(Refer Slide Time: 13:12)



So we have seen all these videos designed through mimicry; I did not mention the second one, but please go through it, it’s a very nice short video from the science magazine about a cockroach base robot, this is in the introductory lecture, and these were the various biomimicry lectures and then the artificial retina lecture, brain-computer interface lecture, and then the three domains of life, biological classification, single use bioreactor; these three were videos and the large bioreactor, this is an image, and this cell wall number eleven is what we are talking about right now. Okay?

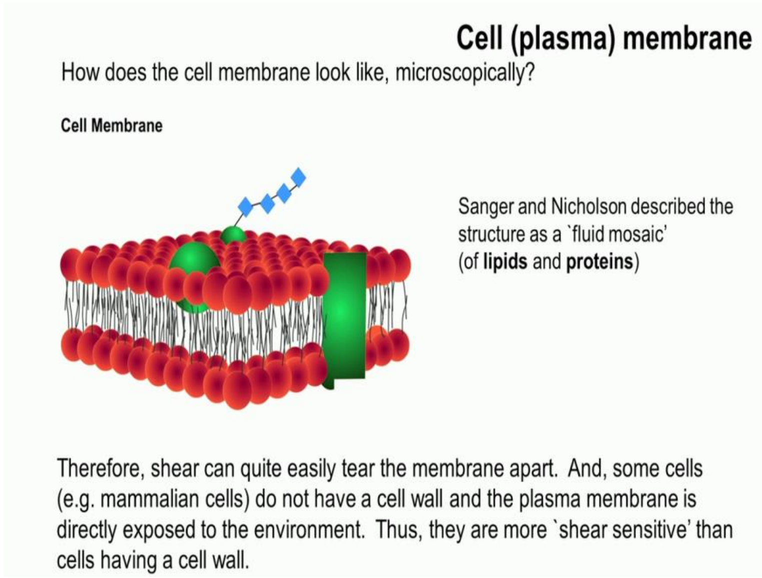
(Refer Slide Time: 13:33)



If you click on this, you will get an image that you could see. Let us get back. As a cell wall is predominantly made up of two kinds of molecules called 'peptidoglycan' and 'teichoic acids', okay?

This is the terms and this going to float this around do not worry about the terms as of now. If it is important enough, we will go and see what it is; if it is not, just hear the terms so that if you hear the terms later, you will at least vaguely remember, and you know, I have kind of heard this somewhere. That is good enough. And if it is important enough, then you would be repeatedly exposed to it and you can pick it up.

(Refer Slide Time: 17:16)



The plasma membrane; the earlier we talked of the cell membrane, the, oh sorry, the cell wall, now let us talk of the cell membrane or the plasma membrane. This is where it is very interesting. How does the cell membrane look like, in a microscopic cells. It looks something like this, you know, this is a two-dimensional structure, this is shown as some sort of a, you know rectangular, a cuboidal kind of a structure. This is a cell, right, you take a small part of the cell, it look like this, and if you extend it in all directions and assume that it is covering some sort of let us say, a spherical shape or an oval shape, then you can understand how this fits in into the structure of the cell, right?

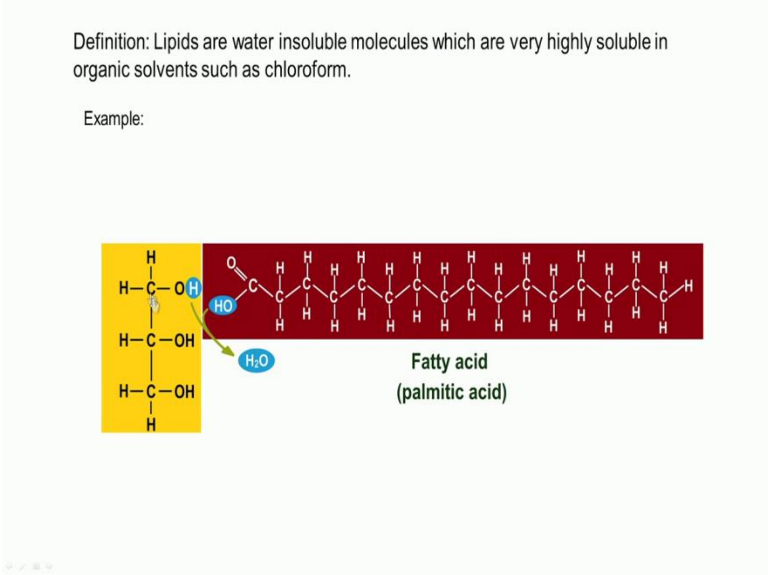
So you have the cell membrane consisting of two layers as you could see; the first layer is this circle here and some of these, the, lines here, the second layer is circle here and the other lines in the opposite direction, right? So in other words, it has a double layer here. It so happens that the nature of this is very different from the (na), the nature of the ball is very different from the nature of these lines here, we will come to that. In addition there are a few other things, there are these green things that I have shown, and there are these blue things that I have shown, we will get to that. The green things are actually proteins, we will see what proteins are, although we have heard some terms so many times, we will see what those proteins are. And these are carbohydrates which stick onto it, we will see what they are. Okay?

For now, let us focus on this double layer here, which has, if you take one molecule that comprises a double layer, it has this round head and a (())(16:03). Okay? So Sanger and Nicholson described the structure of a cell membrane as some kind of a ‘fluid mosaic’; mosaic is just these things put in together, so this is a mosaic of, what are called ‘lipids’. In fact, these molecules you know that we have been describing so much here, these are called lipids and there are proteins, and there could be other things also. So Sanger and Nicholson described this structure as a fluid mosaic of lipids which are these and proteins which are seen here. That is good enough for now, okay, that is what a cell membrane is.

Therefore shear, you know, can easily, can quite easily tear the membrane apart because they are all floating around, okay, you provide enough surface force, the shear is going to tear apart these lipids. And some cells such as, as I mentioned, the mammalian cells are cells, do not have a cell wall and the plasma membrane is directly exposed to the environment. And therefore they are much more shear sensitive than when compared to the cells that have a cell wall, okay, this is common sense. When you use these cells in the bioreactor, mammalian cells are used in the bioreactor for production of high value products such as monoclonal antibodies which are used for cancer therapy and so on.

And when you, when these cells are used in the bioreactor for production of these, they (ha), they are directly exposed to the various stresses because of the velocity gradients in the bioreactor and those shear stresses can break the cells apart. Okay? So what are these? Let us look at the nature of this molecule, it is an important class of molecules. Definition is very big, okay? And that is the best that we can get, and let us start with the biggest definition in biology pretty much.

(Refer Slide Time: 19:55)

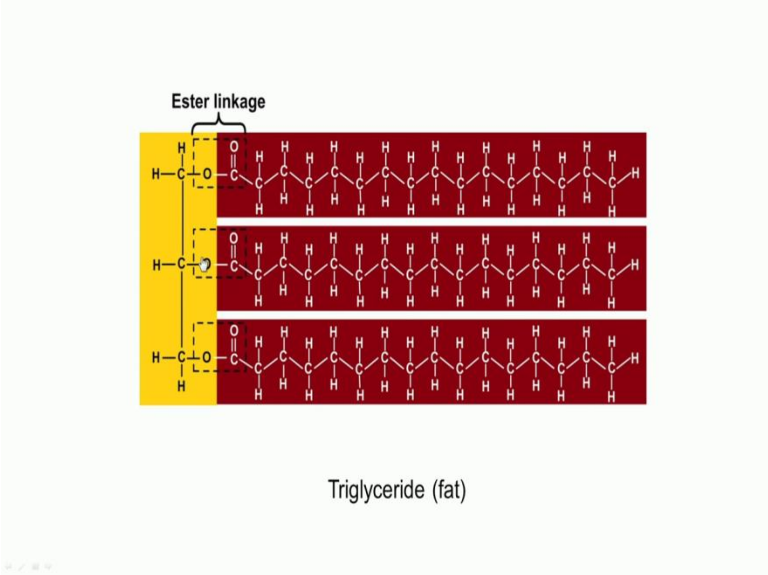


'Lipids' are water insoluble molecules which are very highly soluble in organic solvents such as chloroform, okay? I know it is vague, but let us stick to it. That is what a lipid is. And that is one of the four major classes of basic biomolecules that are present in life. Okay?

So for example, you all, okay let us take this molecule here, okay? This is a three carbon molecule, okay, if you know a bit of chemistry you would know this is glycerol, the one that is shown in yellow or some shade of yellow there, is glycerol. It has three OH groups here, three carbons here, and to one of these OH groups, a long chain carbon molecule with a COOH gets attached, okay? When this gets attached, the H2O gets out and the C joins with this O, so you have a CO, on the other side this O, on the other side, and this bond going onto this O to provide an example of a lipid, okay?

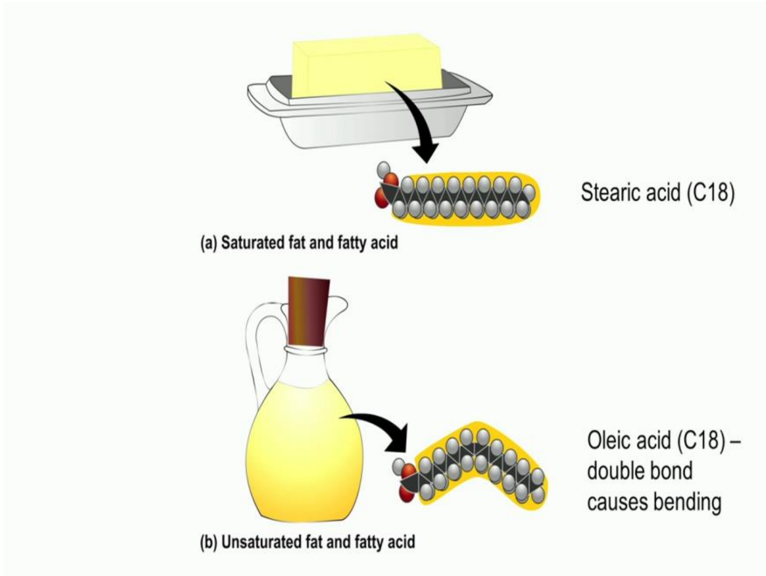
So you have a fatty acid, which is hydrophobic, and you have glycerol, which is hydrophilic and attached, you could in principle, get a lipid. So this has I think eighteen, one two three four five six seven eight nine ten eleven twelve thirteen fourteen fifteen sixteen, sixteen palmitic acid, C16 is the palmitic acid and all these bonds in between these carbon molecules are all single bonds. Therefore this is saturated fatty acid. Saturated fatty acid with glycerol is an example of a lipid. Now, instead of one fatty acid attached to one of this OH, what was initially OH of the glycerol.

(Refer Slide Time: 20:50)



If you have three fatty acids attached to the three oxygens of the glycerol through ester linkages, as they are called. Then you get fat. Right? All the fat that that causes a lot of social difficulty is nothing but this, okay, it is a triglyceride, you have three fatty acids attached to a glycerol molecule and that is your fat. Fat is a lipid. So fat causes whatever difficulties, and all that is a lipid. It is called a triglyceride, okay because you have you have glycerol, you have a glyceride bonds being formed here for three fatty acids and you have a triglyceride there.

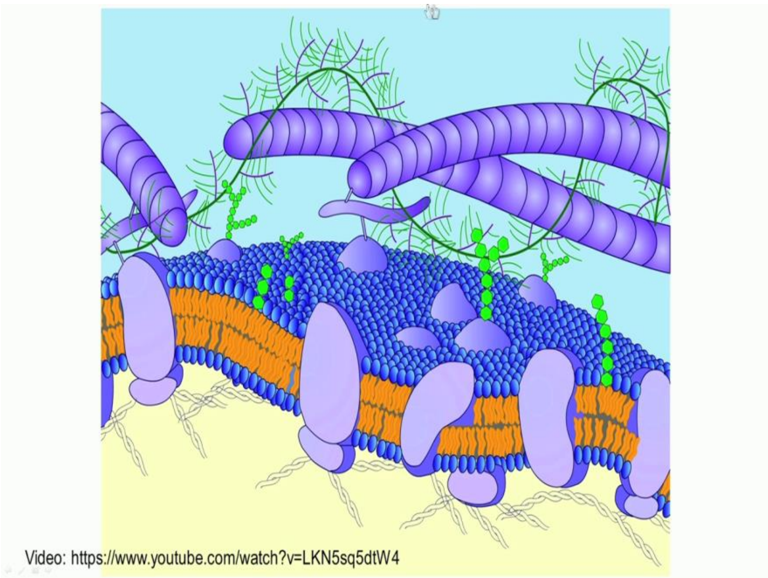
(Refer Slide Time: 21:52)



C18 stearic acid is nothing but butter, okay, it is saturated; if it is saturated you get butter. And C18, with a single double bond gives you oil.

Butter, saturated because all the bonds between carbon atoms are single bonds, and here one bond is a double bond, which actually introduces a kink in the structure and you have a fatty acid with one (un) one bond unsaturation, and then you get oil, okay? You know how useful these products are, and this is the molecular nature of these products. C18 is butter, C18 with a double bond is oil, unsaturated oil and so on, okay? All these saturated unsaturated terms that are used by doctors, actually refer to the nature of the bonds between the carbon atoms.

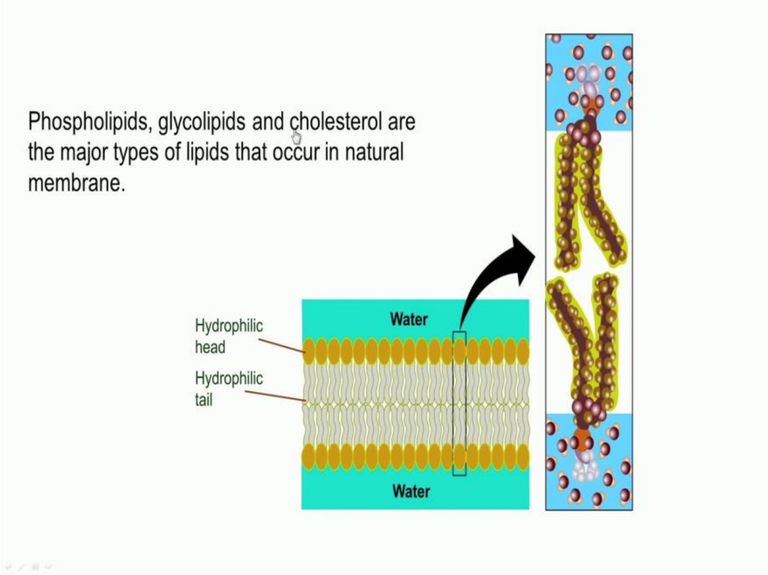
(Refer Slide Time: 22:35)



So this is the three dimensional view, let us just focus on some parts of it; there are many parts to it which let us not worry about. So this is the double layer plasma membrane as I said, it covers the cell, this is a part of the plasma membrane, assume that it is extending in various directions, and you see how it can cover this.

So this plasma membrane has a double layer of lipids with a lot of proteins sticking around a fluid mosaic of lipids and proteins, and then there are various other things which we will not worry about now. Okay? If you are interested, you could see this video, by clicking I think it is video number twelve, the item number twelve here, plasma membrane, you could see this, and that will improve your appreciation of the plasma membrane.

(Refer Slide Time: 24:47)

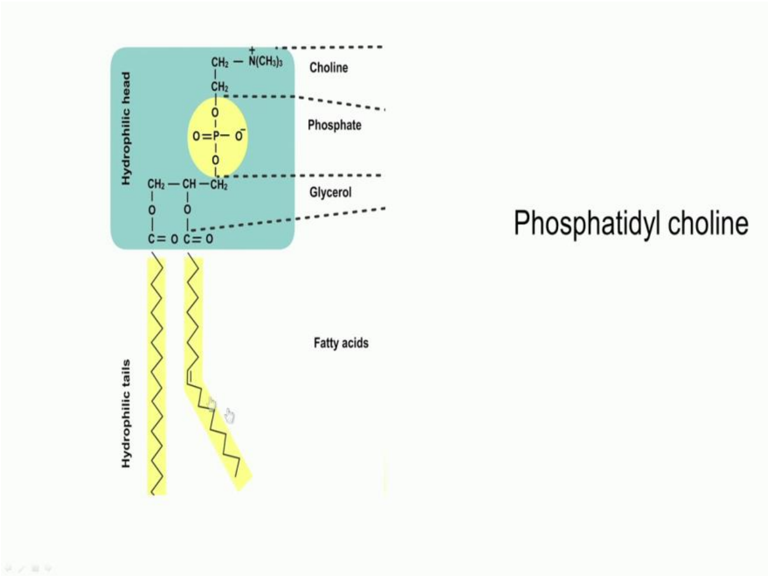


So let us go a little deeper, let us try to understand this a little better. So I am taking a part of the plasma membrane, a two dimensional view. So this is the bi-layer of lipids, you have the hydrophilic heads here, hydrophilic just means water loving, and therefore when it is exposed to water, it will tend to orient itself towards water, and you have these hydrophobic tails; in other words, this is a part, this could be a part of the glycerol molecule and this could be the various fatty acids, okay? Typically there are two fatty acids here and this is what is been blown up here, right, these are the fatty acids, this is probably saturated, this is probably unsaturated, and you have some other molecules here which are hydrophilic, they like water, and these are hydrophobic molecules, and this is a structure of one of this pair blown up, okay? Nothing else.

Phospholipids, glycolipids and cholesterol are the major types of lipids that occur in a natural membrane, okay? In other words, we we saw the triglyceride, right, the glycerol molecule being converted into triglycerides. That was an example. In the phospholipids this is different as we will see later. There is another set of molecules called glycolipids where this part is different and cholesterol which is completely different molecule, we will see that in a little while, okay? Cholesterol, is a very common, commonly known molecule, it was supposed to cause a lot of difficulty. People have the, American Heart Association has changed its view on cholesterol now, but it is popular anyway.

So you, it it will be nice to see what it is all about, okay?

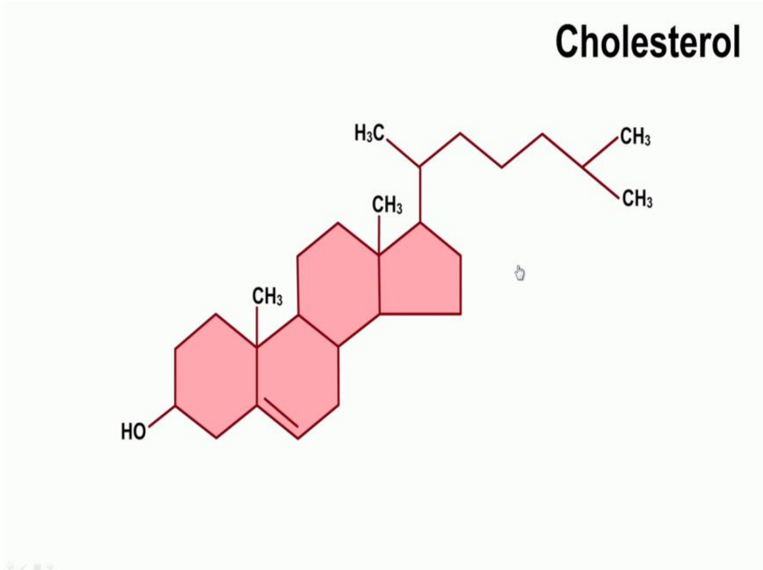
(Refer Slide Time: 25:50)



So this is a phospholipid, particularly phosphatidyl choline. These are the fatty acid chains, the hydro, hydrophilic, hydrophobic, it should be hydrophobic here; hydrophobic tails that are here and this is the glycerol molecule as you can see here, the three carbon chain here and two of those O molecules are attached to the fatty acid.

The third molecule of O is attached to a phosphate group, 'PO4-'group, and then there is a choline group that is on top of that, okay? This is called phosphatidyl choline. As you can see this is a hydrophilic end and this is a hydrophobic tail, so this fits into the various needs of a lipid. And this is an important constituent of natural membranes.

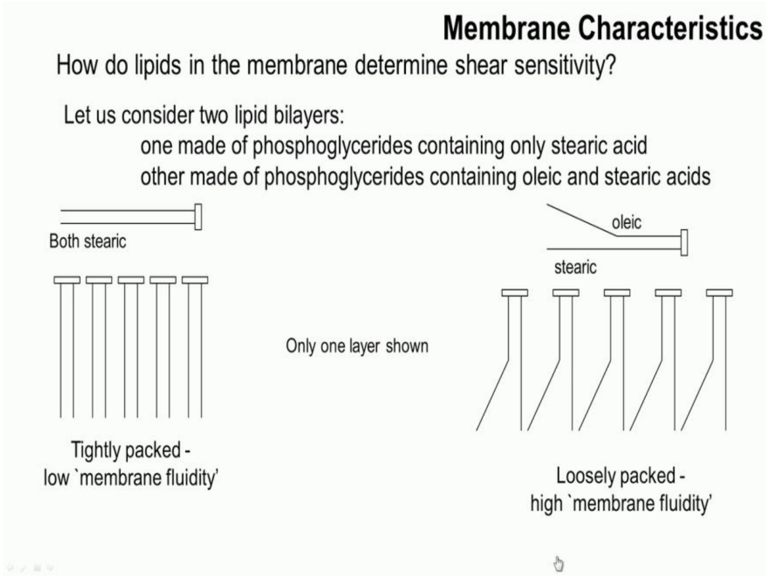
(Refer Slide Time: 26:15)



This is cholesterol, right? You see all this here, all this is the hydrophobic part, and this OH is the hydrophilic part, okay, and the cholesterol is also a lipid that doctors thought caused so many problems earlier.

Okay. How do lipids in the membrane determine shear sensitivity? How, how sensitive the shear is, okay? There are various types of lipids. So depending on the types of lipids that are present, the shear sensitivity should vary, okay, because the properties of the constituents determine the overall properties of the whole. Right? So let us see how that is and let us see an application of that towards our needs.

(Refer Slide Time: 29:03)

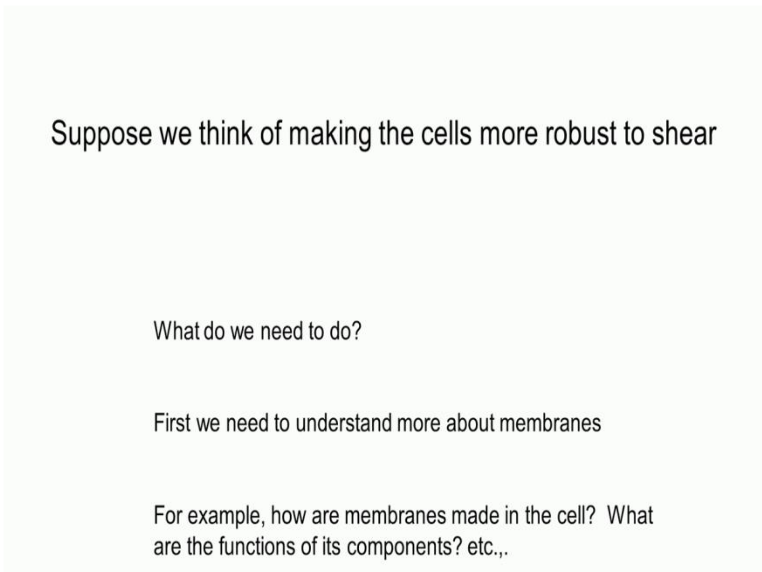


For that, let us consider two lipid bi-layers; the first bi-layer is made up of phosphoglycerides containing only stearic acid, okay, C18. C18 saturated; and the other is made up of phosphoglycerides containing oleic and stearic acids. Oleic, as you know has one unsaturation, stearic is, has no unsaturation. Because of that, if both are stearic, okay, this, this is the hydrophobic tail, hydrophilic head; if both are saturated, such as stearic acid, the structure is going to be something like this. Whereas, if it is one oleic and one stearic, the stearic is going to be straight, the oleic is going to have a kink, and therefore it is going to occupy more space compared to stearic here, okay?

And therefore, if we tightly pack lipids containing both stearic acids, it is going to pack something like this, okay? These going to be tightly packed, I am just showing one layer for clarity, you can imagine the other layer on the other side; whereas if we have both stearic and oleic present on the phosphoglycerides, then it is going to pack like this. The area that is the space that is occupied by each of this is going to be higher, therefore the packing itself is going to be much less dense than the one with both stearic acids, and therefore the ‘membrane fluidity’ is going to be high due to the loose packing, whereas here the membrane fluidity is going to be low due to the tight packing.

So it is quite easy to see that the tightly packed thing is going to resist shear much more than the loosely packed thing. A small amount of, smaller amount of shear force can tear this apart compared to this, okay? So that is very simply how the structure of the cell membrane determines its shear sensitivity, and the structure of the lipids or or the constituents the nature of the constituents determine the structure of the membrane and the structure determines its property. For example, 'membrane fluidity'; loosely packed structure will be more shear sensitive. Suppose we think of making the cells more robust to shear, okay, shear is our story here. Suppose we think of making the cells more robust to shear, what do we do?

(Refer Slide Time: 30:06)



We need to understand, of course more about membranes and how are membranes made in the cell and so on and so forth, what are the functions of the various components, and then somehow make the cell to make to generate the kind of lipids that we need to have a more densely packed membrane, and therefore more shear sensitive cell lines, or mammalian cells, may be, okay, if you are going to use them in the bioreactor, right? So this is one way of thinking about it. Fine, I think we will stop here for this lecture, we will continue further when we meet next. Very briefly, in this lecture, our story was about, was about shear. We saw what shear was, let me quickly go back to what we saw.

We saw what shear was; I told you in some detail what shear was and that it can cause damage to cells, especially in an environment such as a bioreactor environment which is full of shear, and the microscopic or the sub-micoscopic components need to be known because they determine the properties of materials, and then we ask the question what gets affected by shear in cells and we came up with our first guess, a cell envelope which consists of a cell wall and a cell membrane. We very briefly looked at the cell wall and then we looked at the cell membrane in some detail. We saw that it was a fluid mosaic of lipids and proteins and we saw what are lipids, which I said was one of the four major classes of biomolecules and they are defined as, lipids are defined as water insoluble molecules which are very highly soluble in organic solvents such as chloroform.

And then we saw a few examples, our fats are actually lipids, and then butter is a lipid, oil is a lipid, and then we looked at some of the details of phospholipids, and cholesterol and there is another type, glycolipids and all these three are commonly found in natural cell membranes. Then we saw how the membrane characteristics determine membrane shear sensitivity, and we also looked at a case where we could possibly modify the lipids to modify the shear sensitivity. Let us meet again later and when we meet again, we will continue with another story. See you then.