>> March 4th, 2015. This is actually Lecture 7 in Physiology, and it's the last one in this unit that we entitled Cell and Molecular Physiology. Tonight, we're going to look at some applications, some ramifications of some of the concepts that we've given you information about. But before we can do that, we need to finish up, if not, review what we did yesterday in lab, and that is talk about the process known as DNA replication. To replicate DNA means what?

[Inaudible Remark]

To make more of it, you bet. To duplicate it. And we made the case that cells only need to do that if and when what?

[Inaudible Remark]

If and when they're going to divide. If they never divide, there's no need to make more DNA because think about it, does the process of protein synthesis consume or change DNA in any way? No. So DNA is not harmed through the process of protein synthesis and the only occasion for its replication then would be as a prerequisite to cell division. So tonight, we're going to look at DNA replication and we'll finish up with an idea called expression or control of genetic information. So let's do what we've already introduced. Again, let's discuss the steps, the basic simple steps in DNA replication. First, we have to presume the existence of the double-helix naturally. You can't make DNA without DNA. And so the first step, separation of the helices with an enzyme called helicase. And that unravels, unzips, unwinds the DNA at one end, just as you did with your hands on the benchtop yesterday, your fingers were working as helicase. And the hydrogen bonds are easy to break, no energy required. And so the molecule unravels from one end toward the other. This uncoiling is followed by alignment of available free deoxyribose nucleotides. Free, means that they're unattached, that they're available in generous quantities. And how many different free deoxyribose nucleotides are there? Well, those carrying cytosine, those with adenine, those with guanine and those with thiamine. So here in this animation, we show these free deoxyribose nucleotides aligning. There's no direction here, that is, what guarantees their alignment is mandatory, complementary, base-pairing. C always pairing with G, A always pairing with T and these bonds, remember, are ordinary hydrogen bonds which are established at very close range between these nucleotides and the parent strand. So this occurs along both of these unraveled helices. And the next step of course is to link these nucleotides together, that is to connect sugar with phosphate and that's called strand polymerization which uses huge quantities of ATP, and those bonds then are made between the sugar of one and the phosphate of another. Not without the involvement of an enzyme--what's the enzyme that makes these bonds possible? Its name is DNA polymerase and its existence is presumed, that is it's always available. And we did say that it has a condition, it has a stipulation, it has a limitation. DNA polymerase will only work in the presence of DNA. And that guarantees that we're not just going to put these nucleotides together in a random meaningless way. It guarantees that we're going to be copying accurately the sequence of basis along each of these two strands. In short, it guarantees accuracy. And do we care about accuracy? Do we want this DNA to be a perfect copy or just sort of like it? No, we want it to be absolutely perfect. We don't want any counterfeit DNA floating around. So the DNA dependency guarantees that accuracy. So you can imagine, you can visualize, you can go to YouTube and find animations on this but in the end, as you produced yourself with the plastic models, you end up as you should end up with two identical semi-conservative double-helixes. And after all, that was the purpose--that is the goal of DNA replication. What is the destination of those now identical helices? Well obviously, they've got to be distributed into these developing daughter cells so that each cell receives all of the DNA. So this much we did and certainly looked at in close model building yesterday. So the next topic which, of course, awaits DNA replication is mitosis. And there was a tidbit of incredible information that some found hard to believe that if the three billion base pairs of the DNA found in any cell, if that DNA were stretched out, it would be how long?

>> Six feet.

>> Six feet. Now, how can you stuff six feet of DNA into a nucleus which you can't even see? So this requires some very careful, very tight packaging. And so I have this video which is just a few seconds, but a great animation. Keep in mind, nobody has actually seen this, nobody has photographed it, there aren't microscopes that are good enough, but we know that this is what's going on. So, an interesting and short little video.

>> In this animation, we'll see the remarkable way our DNA is tightly packed up so that six feet of this long molecule fits into the microscopic nucleus of every cell. The process starts when DNA is wrapped around special protein

molecules called histones. The combined loop of DNA and protein is called a nucleosome. The nucleosomes are packaged into a thread known as chromatin. This fiber is then looped and further packaged using other proteins which are not shown here.

[Pause]

The end result is that the DNA is tightly packed into the familial structures we can see through a microscope, chromosomes. Chromosomes are not always present. They form around the time cells divide when the two copies of a cell's DNA need to be separated. At other times, as we can see now after the cell has divided, our DNA is less highly organized. It is still wrapped up around the histones but not coiled in the chromosomes.

>> So that was a time-lapsed video really. The first part was animation but this last part, that's a real film so we can see in photographed mitosis but nobody has ever seen or photographed this packaging, but it is an incredible feature. And, of course, it follows DNA replication. So let's go back, that is let's talk a little bit about mitosis which is a topic you learned quite a while ago I'm sure in biology. So we're not going to get bogged down in a lot of diagramming or all the subtle steps. We're trying to look at an overview and appreciate, you know, the basic reason, the basic objective of mitosis. And so when a cell is indeed replicating its DNA, nothing can be seen or photographed, but we know that this phase called interface which literally means between phases is the time that DNA is being replicated, and that means three billion base pairs are being copied. Huge amounts of ATP are necessary and a fair amount of time. How long does it take to copy three billion base pairs? Well, I don't know, 10 to 12 hours, which is pretty impressive actually if you think about all that has to be done. So interface, an invisible phase where DNA is being replicated, and if this fails, nothing else that we mention can happen. That is none of the other phases will follow. The next thing that you can see in terms of photography or otherwise in a microscope, is that after the DNA has been packaged, we start to see as the film said, the structures which are certainly iconic and familiar, namely the chromosomes. Chromosomes appear and the nuclear membrane which is used to enclose and define the nucleus tends to disintegrate or disappear. Next is a phase called metaphase that you might recall from biology, at this point, the chromosomes line up--align at the center. This is followed by anaphase. Remember, a chromosome is actually two identical chromatids which are held together by the centromere. And these chromatids literally separate and are moved along spindle fibers. And so here's early anaphase and late anaphase. And the obvious occurrence here is that we're moving genes to opposite ends of this single cell, anaphase. After anaphase, the final phase that we care about is quite dramatic because the cell membrane gets involved now. And we think of the cell membrane as just a passive bag but we've already clarified that. Don't membranes have the capacity for phagocytosis and pinocytosis? So they're quite capable of this kind of change and they tend to furrow in, as shown here, and literally create ultimately two separate cells. So telophase will generate two cells--two identical cells which have exactly that, the genetic information that the other one has. Two genetically identical daughter cells result. So this happens. Well, how often? Do all cells divide all the time? I think we made the case that some are very active in that regard and among those are cells that make red blood cells that is myeloid tissue, bone marrow, also cells along the GI tract and skin cells. And maybe this seems like a strange question, but why are these cells dividing so often and why are others not? Well, I think about--let's think about skin. Why skin need to be replicated? Why do those cells have to be replaced so often? Because they are flying off and the same is true for the digestive tract, and the same is true for red blood cells. So the mitotic rate tends to match the death rate, in other words, the faster cells get clobbered or otherwise destroyed, certainly, that dictates that they be replaced at the same rate. And think about that, wouldn't you want that to be the case? The rate of cell death matched by the rate of cell mitosis to keep, you know, things, in let's say, balance. So as we discussed, DNA replication takes 10 to 12 hours. And what's interesting, of course, if we think about our creation, that is our own conception, all of us came from two cells, didn't we? Mom's ovum and one of dad's sperm. We all started out as a single cell called a zygote. And through nine months of gestation, there were 40 trillion cell divisions to get you to that seven pound baby that you might have been, right? Forty what? Not million, not billion, 40 trillion cell divisions. In other words, DNA had to be replicated 40 trillion times, and hopefully, perfectly. And since we all came out perfect, apparently, that's exactly what happened. But it is stunning when you think about it because aren't there opportunities and wouldn't the likelihood of mistakes be very high in something that's happening 40 trillion times? So I'm making the case that DNA replication is pretty impressive and rarely produces the kinds of problems we're about to discuss. But reality is that mistakes are made and so here we are with the definition that we introduced yesterday. Mutation is any alteration in the DNA base sequence. This doesn't care how it occurs, it doesn't necessarily occur when it occurs or where it occurs. It's just defining what a mutation is. And naturally, as we said, it's more likely to occur not when DNA is just sitting around or even when DNA is engaged in transcription or translation, because remember,

transcription and translation don't really alter the DNA. And so the opportunity for mistakes, the most likely occurrence is during what process? DNA replication. So if this cell is replicating more often than that cell, then clearly, these tissues are going to have a higher propensity, a greater chance for a mutation. Now, if we accept the definition of mutation and if we end up with even a slight change in DNA, will that change affect transcription? Of course, because transcription is a dumb process. We're just making RNA off of the DNA, and if DNA is bad, then so will the RNA be bad. But wait a minute, I shouldn't really use the word bad, let's just say different. So if there is a change in the DNA, that will cause some change in transcription and certainly, alter mRNA. So we'll not use the word bad. But if the RNA is altered, then so would translation be altered and then so would the protein be altered. So it would be a different protein. How different? It depends upon the type of mutation and whether that difference even matters, we'll see in a moment. Sometimes, the protein is so different, so mangled that it's called a nonsense protein, in other words, it's totally useless. But more often than not, the changes are subtle and the protein, although different, is not necessarily useless. So with that said, what are the possible effects that a mutation might have? And it's an easier question than it sounds, there are really only three possibilities and here they are. It could be no effect at all. Is it possible to change the DNA and change the protein so slightly that there is no net effect, that health and overall performance of the tissue is not affected? Absolutely. In fact, this is the most common thing. In other words, most mutations are subtle and have no real health impact to you or society or to evolution. In other words, they're just too small to matter. What's the other? The other possibility is that you have an adverse effect. That means it alters your health or your offspring or population in general. So adverse is very definitely a possibility. And the third possibility which is actually statistically the less likely of the bunch is that it might be positive. Is it possible for this new protein, whatever it is, to actually be better slightly in some subtle way than the protein that otherwise would've been made? Is that possible? Is it a possibility? Yes. And if it is, are those cells better off and is that individual better off, and will that individual perhaps live longer or procreate better? So this possibility, although statistically the least likely of the bunch, allows for the improvement--the improvement of the species through time, in other words, natural selection and the evolution of improvements that is as you know, survival of individuals who have an edge over others because of mutation that is positive. So these are the possibilities. Now, what are the types of mutations? You know where we're going with this but before we talk about the three types of gene mutations that you've already been introduced to, we have to make a clear distinction. What we dealt with yesterday in the lab are called gene mutations. But there is something called a chromosomal mutation which is actually quite different. A chromosomal mutation is typically going to occur in the course of mitosis and leads to what? Missing or extra what? Portions of chromosomes. And chromosomes by definition contains thousands of genes. So it's easy to appreciate that missing chromosomes would be, well, uniformly bad. Missing chromosomes are often devastating, meaning lethal right off the get-go. In other words, the fetus doesn't survive. The embryo doesn't develop. But there are some interesting cases of extra chromosomes which at least in theory wouldn't sound so bad. I mean, except the fact that chromosomes contain genes. So if we have missing chromosomes, well, we have missing genes and it's pretty easy to expect that to be serious. But what's the big deal about extra chromosomes? I mean, a little extra wouldn't hurt. But it basically creates confusion and chaos because we have just simply too many sets of that given chromosome, those particular genes on that chromosome. And so although missing chromosomes are almost always lethal to the development of the embryo, extra chromosomes, not always, which is not to say they're not serious. And what's the most common, familiar but yet, not lethal case of extra chromosomes? Down syndrome which is called trisomy 21. Are these infants born and do they survive and do they live for a number of years? Of course. No one is minimizing the impact that that has but the point is they're not outright lethal, at least in the case of Down syndrome. So chromosome mutations are very different and involve many more DNA alterations because we're either subtracting a great many genes that is leading them out of the process of mitosis or adding on that is bringing in more chromosomes than necessary. But our work yesterday was all about gene mutations, and you know there are three types that you played with and really considered up close. The first are substitutions. And remember, these mutations occur during replication and they don't affect the existing DNA, they affect the replicated strand. And so what is a substitution? Well, that's when you have a base that is an incorrect base substituting for a correct base along the course of DNA replication. And of the three types of mutation that you already know, the three types of gene mutation, this is the most common. And also, as you know, the least devastating that is--has the least serious impact. Here's an example that I think everybody knows or at least everyone's heard of, sickle cell disease, yes? And the protein that's involved in sickle cell disease is hemoglobin which of course carries oxygen. And hemoglobin is produced in, carried in, found in red blood cells. OK, good? So these symbols represent the amino acids that normally are part of the hemoglobin molecule. These are names of amino acids. And one of the many amino acids in the hemoglobin chain is one called glutamic acid, and the codon for that is AUG. You don't need to know any of this, I'm just building a case for what is this substitution mutation. So with normal hemoglobin, life is good. That is the hemoglobin does its job and the red blood cells are happy and healthy and these

look, well, happy and healthy. But in sickle cell disease, that codon, what was it?

>> [Simultaneous] AUG.

>> AUG changes to UUG. So how is UUG different from AUG? Well obviously, U has substituted for A, and that is of course a classic case of substitution. Well, so what? Well now, the amino acid which is brought into this position is not glutamic acid but instead valine. So the protein is different. Different by how many amino acids? One. And so it's still hemoglobin but is it the same? No. It's different in how many amino acids? One. And you might now think, well, what's the big deal? Well, the big deal is that hemoglobin still works. It still carries oxygen. These people still live. But in areas of low oxygen, that is as the red blood cells go through capillaries, the hemoglobin gets all bent up and the molecule affects a change on the cell. And the cell instead of being disc-like, becomes sickle-like, hence the name what? Sickle cell disease. Now, so what? Well these cells just don't get through capillaries very well because they don't have the pliability. They tend to jam up and so people with sickle cell disease suffer a crisis because they're not getting oxygen to some of their tissues. So it's certainly symptomatic, that is a serious enough problem. But do people with sickle cell disease live beyond childhood? Yes. Do they live long enough to engage in sexual reproduction, that's is can they develop families and so forth? Yes. So this is not outright a killer, although it may in some cases kill individuals. The gene is still with us, which does reflect back on this comment that very often, mutations are--what's the A word here? And isn't this adverse? Then why hasn't it disappeared? That is why haven't these--these people, that is people with this affliction, died off? Well, as you know, this mutation actually originated in Africa and now is throughout the world and is preponderant in African-Americans. But the interesting thing about this mutation, despite its adverse effects here, that is despite the fact that it has on RBCs, it confers resistance to malaria oddly, for reasons that I don't understand. So in Africa, it provides what? Resistance to malaria. So that which is adverse, most of the time in that setting is actually slightly positive. So we've got this sort of split personality. This mutation is adverse in some settings but actually confers a positive advantage especially in areas where malaria is prominent. So, another interesting case. There are plenty of other substitution mutations, muscular dystrophy is one that comes to mind, but substitution mutations are well-known. The other two that you know are deletions and insertions. You don't hear much about these that is I can't give you a particular disease that is caused by this or that because these are usually outright adverse, and when they happen, you're dead. In other words, there is no real problem, that is there's no way to survive this. Why is that? What does a deletion do? What is a deletion? An emission of a base, and insertion is just that. So what does that do to the reading, the grouping of these triplets in DNA? What was that word? Frameshift? And does that make each and every triplet probably different and each and every codon probably different? And what would happen to the protein? It would bear no resemblance--no resemblance to the intended protein. And if the protein is a key protein, a key enzyme, then there's just no surviving these kinds of mutations. Even though what? Even though we're deleting just one base or adding just one base, that frameshift is a real killer, literally, as you can imagine. So these are the three type of mutation. And the next question that we need to talk about is well, what causes the mutations and are there ways to avoid these kinds of changes? The causes are pretty limited. One, no cause at all. In other words, number one says, what? Spontaneous. In other words mistakes happen and spontaneous is just that no known cause. Spontaneous mutations have been around as far as we know, from the beginning of life on this planet. In other words, DNA replication has never been perfect. So spontaneous mutations have always occurred at a rate which we assume is the same now as it was in the beginning. But then we know things have changed since the beginning and we know that our world is loaded with things that weren't here in the beginning. And so these are things called environmental what? Mutagen. A mutagen is anything which ramps up or increases the background or spontaneous rate of mutation, and environmental mutagens are just that, things in the environment, which are sub-grouped into these categories, first physical. Physical factors in the world we live. And a key example here is radiation. Now, radiation can be cosmic rays coming from space which has always been the case, but are there other radiation hazards in modern society which we're quite aware of? And I'm not talking about a nuclear holocaust or even a meltdown at a nuclear plant, those are possibilities but certainly, haven't happened on a huge scale yet. But what's the day to day situation here in terms of radiation? Are we, sometimes faced with choices in that regard, we all go to have our teeth examined, right? And how do we get a good picture of your dental status? Well, they'll give you an X-ray of your mouth, or you'll have a chest X-ray actually, required, right? Required for employment some places. And the good outweighs the bad we think and certainly, we'd argue that that's the case. What's the form of radiation that all of us are exposed to everyday, healthy or not? Well, not everyday because there are clouds, but I'm talking about ultraviolet radiation. Radiation comes in many forms, so called high energy radiation and low energy radiation. High energy radiation is stuff like X-rays that will go right through your body as if it weren't there and low energy radiation is ultraviolet light which won't really penetrate very far. And therefore,

ultraviolet light affects what cells and only what cells? Your skin. Ultraviolet light is not going to affect your heart because it's pretty dark in there and sunlight just can't penetrate. But nevertheless, radiation is a risk factor and will increase the rate of mutation.

>> Do tanning booths fit in that criteria?

>> Absolutely. Now, the people who own and operate tanning booths will tell you, "Oh, this is safe. It's been proven." Well, that's all BS. There is no safe ultraviolet radiation. Now, don't get me wrong, I'm not saying that if you go to a tanning booth, you're going to die of cancer, but certainly, your odds of getting malignant melanoma are ramped up and don't believe me, just look at the statistics. But yes, absolutely. So this is an avoidable form of mutagen. Then, there are chemicals. What are some chemicals that are in the environment, like it or not, that are known to be well, mutagens? You know, in the state of California, you can't go into a building anymore without seeing a little placard there, maybe you never noticed them but it says, "This building contains stuff that's known to cause cancer", you know. And how many of you ran in fear because of that? I mean, it's just a legal thing because so much of what surrounds us that we inhale or otherwise consume is known to cause cancer. Don't you see that at the gas pumps? All right. So what are some chemical mutagens by name? OK. I know you've probably heard of--well, maybe you've heard of benzene. Certainly, you've all heard of asbestos, right? Asbestos, an airborne material that used to be used readily and without concern to insulate pipes or plumping, especially in industrial applications. And I was telling the group here about a story back in the 1939 actually, the film The Wizard of Oz. In The Wizard of Oz, there's a scene, I don't remember it, but where there's snow, and of course, the producers didn't want to make snow on the set so they just sprayed the actors with asbestos. And they thought that was fine, it looked like snow, but did they get exposed to that mutagen? And today, if you watch TV, you'll see, you know, a commercial come up with lawyers behind and say, "Do you have mesothelioma?", right, which is basically lung cancer caused by asbestos. So they want you to come in so that they can sue somebody and make some money. But my point is we know now about asbestos, we know it's a, what is the word? Mutagen. And we're doing everything we can to get it out of buildings and out of the environment. So there are many others. And certainly, the biggest source and the most avoidable source of chemical mutagens is tobacco, yeah, and the list is too long to mention. And so obviously, is there a connection there with mutation and the consequences thereof? Certainly. Here's a thing out of a book of course and it talks about possible causes of mutation, ultraviolet light, chemicals and food, radiation and spontaneous. It also shows tobacco here but interestingly, it shows nicotine. And although we associate nicotine with tobacco and certainly, that's why people smoke, it's because of the nicotine, nicotine is very addictive but oddly, it's not a mutagen. There are plenty of others in there, so this is actually incorrect but never mind. Are there mutagens in the environment that you can avoid? Yeah. But in order to avoid them, you have to know what they are. And back in the '40s, a lot of people smoke because they thought it was healthy. So, times change. Some viruses are responsible for mutations but in the scheme of things, their contribution is minor, that is of all the mutagens that are out there, viruses may be stack up to 15 percent or so of the important sources of mutation. Now, with that said, all right. We know what mutations might do, we know the types of mutations, we know some of the causes for mutations, but what's the ramification? That is what's the fallout from a mutation? Yes, it leads to a changed protein but that's not always bad but very often, it is. So the adverse effects really depend upon the location and that means what cells are actually affected. And essentially in this category, there are two locations, somatic or not. A somatic mutation involves cells other than the gonads. So would a skin cell be a somatic cell? Yes. Would a muscle cell be a somatic cell? Yes. Every cell in your body is a somatic cell except those that produce sperm or eggs. So somatic mutations affect of course, those cells throughout your body and they are very common in all of us but the good news is what? These types of mutations are common but what not passed on to what? Next generation. So if you suffer a mutation to cells of your skin, might that bother you? Might that be serious, even lethal to you? It might be. But is it going to affect your kids? No, because you don't use skin cells for sexual reproduction. So somatic mutations, even though they're common, affect the individual but not the family, not the population, and have no long-term repercussions that way. Now, why are they lethal to you? Somatic mutations may damage what? Genes that control mitosis, and the word may means just that. They might not but if they do, think about it. A gene that controls mitosis. We said earlier that we'd like the rate of mitosis, we'd like the rate in which cells are dividing to match what?

>> The death rate.

>> The death rate. Why do we want this balance? Well, if we don't keep up with the death rate, then our tissues are going to become crippled or, otherwise, handicapped. But what's the other side? If we manufacture more cells than are

dying, then it might not sound bad but that is a serious problem because it leads to an overgrowth, an overgrowth of tissue. And what's the name for that?

>> [Simultaneous] A tumor.

>> A tumor. So cells that control mitosis--I should say genes that control mitosis couldn't be more important because that rate of mitosis really determines whether or not your cells are replaced at an appropriate rate to avoid the consequences of tumor growth. The good news here is that we don't have a single gene that controls the rate of mitosis. There are many, perhaps hundreds. We don't really know. And those genes that do control mitosis fall into one or two camps. Those that are called tumor-suppressing genes are just that. What's it sound like? A tumor-suppressing gene. Well, that must be a gene that suppresses or restrains the rate of mitosis, in other words, keeps it from getting out of hand. The opposite of a tumor-suppressing gene is called a proto-oncogene which actually codes for proteins that stimulate mitosis. So there are many genes in both of these camps. And together, they maintain this balance, this rate of mitosis which matches the rate of cell death. So if these genes are mutated, then that balance is upset. And as a result, we're going to have what? Uncontrolled rapid cell mitosis which produces something called a tumor often referred to as a cancer or a cancerous tumor. Luckily, it's not just a single mutation. As far as we know, the trigger to cancer requires what? Damage to both tumor-suppressing genes and proto-oncogenes. And no one knows exactly the combination or even the number that trips or otherwise triggers this rapid cell mitosis. But there is a point at which that trigger is pulled and cancerous tumors develop. Tumors come in a couple of forms. And I guess you know, those are called benign, right? And what's the other kind? M word? Malignant. Benign tumors are those that tend to self-limiting, that is they reach a certain size and typically don't get any larger or if they do, they're still rather confined and have a low propensity for entering the lymphatic system or the circulatory system. The other kind, what's the opposite of benign? The other one was malignant. Malignant tumors develop rapidly or at least in a very prodigious manner and they tend to scatter, that is the cells move into the lymphatic system or into the circulatory system where they go where? Who knows. Everywhere. So malignant tumors are much more sinister because, of course, they spread, whereas a benign tumor tends to be rather, self-limiting. Either way, the interesting thing about a tumor is that there's nothing categorically poisonous to metabolism about a tumor because basically, what a tumor is is just a bunch of cells doing what? Dividing, dividing, dividing. Now, how could that be harmful? Just a bunch of cells dividing and dividing and dividing. Well, the reason that's harmful is that we get something that is sizable at some point. And what are these cells competing with? Those cells are competing with healthy tissue for what? Three things, they're competing with healthy tissue for oxygen, competing with healthy tissue for nutrients, glucose and competing with healthy tissue for space, S-P-A-C-E. Now, in the early stages, there's plenty of oxygen and plenty of nutrients for everybody to be happy, that is the tumor is happy, healthy cells are happy, but what's the problem ultimately? Tumors get so big that they compete with the healthy tissue for that final thing, what's that? Space. Now, in some sites, space is not an issue. You can have a tumor growing in your abdomen and there's plenty of room for expansion and therefore, that doesn't really represent a problem. But what about the cranial cavity? A lot of available space in there? No. So a tumor there is going to be obviously pressing on healthy tissue and killing it simply because it is expanding and putting pressure on there. And the same is true in other sites, like the liver and so forth. So ultimately, a tumor will reach a size which will compete with and ultimately destroy healthy tissue, making its presence known. With that all said, what are the therapies, what are the strategies for dealing with tumors benign or otherwise?

[Inaudible Remark]

There's a bunch of them but what's the first and most logical approach? Well, not--cut it out. Somebody said that right. Because if you cut it out, it's now in the dumpster, right? Now, that works just fine. It really does, provided what?

[Inaudible Remark]

All right, if you get all the cells. And if you get them before those cells have--what's the M word? Metastasized. Maybe you haven't heard that, but metastasis means to spread. So you've watched enough doctor shows on TV and doctor comes out and says to the family, "Well, we think"--what's the phrase? "We think we got it all", right? Very reassuring. We think we got it--and that's really all here she can say as a physician or as a surgeon because do they know they got it all? And if they left two cells behind or one cell behind, are you cured? No. You just got some time there. Time for what? Time for this to reoccur. And whether that's a year or 5 years or 10 years, it's not a cure but it does buy some

time.

[Inaudible Remark]

Is it possible to cure cancer with surgery? The answer is yes. Absolutely. It happens all the time. And what's a form of cancer which is more amenable to that approach? That is more successful with regard to surgery? Skin cancer. Why is that? Well first of all, you notice it sooner, right? And second of all, you can cut out huge pieces of the skin and then saw it all together and you know you got it all because, you know, the tiny little thing and you took out six inches of skin or something. So yeah, that's possible. No one can guarantee but are cures known in that or other forms of cancer? Sure. But the key as always is what?

>> Early detection.

>> Early detection. So cancer doesn't always get cured with surgery. So what's the next approach? Actually, there are a couple of approaches and you know both of them, radiation and chemotherapy. These are very different modalities that is they're not the same at all. And there is some irony, and certainly some risk associated with both. Radiation therapy. Why is that ironic? Let's say you have prostate cancer. And you don't want to have surgery or otherwise didn't notice in time. Can we irradiate the prostate? The prostate is a gland below the bladder. And then can we pass radiation into that site, let's say, here's the prostate. Sure, that's not a problem. And what's that radiation designed to do? The interesting thing is it's designed to mutate those cells again. And why is that ironic? Or what caused this cancer was a mutation. So, we're going to mutate it again. And what's the hope? We're going to mutate it so much that's unable to do the one the one thing that it's doing, and what's that?

[Inaudible Remark]

And is that possible? Does it work? Sure, it does. But what's the risk of radiation? Where is that prostate? What's in front of prostate? What's behind the prostate? You're going to hit the prostate about on its way in, it's going to through the bladder. On its way out, it's going to go through the rectum. So you're not only irradiating the cancer in the prostate but at the same time, you're irradiating the bladder and the rectum on the other side. Is that going to diminish the size of the tumor? Yeah, but what risks, what side effects, what gamble are we playing there? This irradiation very often causes mutations in healthy tissue. And therefore, it sets the stage for a what later?

[Inaudible Remark]

All right? So, you roll the dice with irradiation. What's the other thing?

>> Chemo.

>> Chemotherapy. Chemotherapy is just that. It's chemicals that you consume, either orally or given intravenously. Do these chemicals that you know off or that we have at our disposal right now, do these chemicals have a way of seeking out, going door to door and finding only those cells that are cancerous? Do they have that ability to distinguish healthy cells from cancerous cells? No, they target mechanisms that are common to all of them whether that's protein synthesis or cell replication. So if we have a cancer agent, a chemotherapeutic agent that blocks DNA replication, would that stop mitosis? Yes. Would that slow the advance of a tumor? Yeah. Would that buy some time and make your life extended? Sure. But are these agents able to discriminate between cancer cells and non-cancer cells? So what are some of the notoriously unpleasant side effects of chemotherapy? Hairs falling out, what else? Vomiting, nausea and RBC counts' very low. Why are the skin, bone marrow and the digestive tract sure fire targets for this cancer chemotherapy which is suppose to be doing you good.

[Inaudible Remark]

They have a high mitotic rate. Any tissue that has a high mitotic rate is going to be vulnerable to those agents. So, it's not a perfect science. That is we don't have anything that can selectively target cancer cells because our cancer cells more like or more unlike healthy cells. They're really more alike. And so, until we can identify how they're unlike and

specifically wreck those mechanisms, we're left to what? Radiation and chemotherapy. So, here's an interesting bit of trivia, number of cancer diagnosis, 1.4 million in 2005. Now, it's up to 1.7. So it's a fair statement--a tragic statement to say that your life will be touched by cancer, if not personally, then certainly your family. Here's a stunning statistic, probability of developing some type of cancer over one's lifetime. For men, one in two, for women, one in three. And what are the top contenders for cancer in men, it's prostate cancer. In women, it's breast cancer and then behind that, colon cancer, breast cancer and so forth. Why does cancer occur later in life? Why not at age 10 or 20? Well, certainly there are cancers that occur early but remember, cancer is due to M word--mutation. Not just one mutation but a series of mutations that occur over time. So, are we all ticking time bombs, you know, I might have got a mutation today that I have no idea about but it just adds to those that I've had before. And then one day you wake up with just enough to trigger this cancer growth there. So, cancer, of course, is associated with aging simply because of our exposure to these mutagens. Can you reduce your risk? Sure, you can reduce your risk by avoiding to the best of your ability these things. And the more we identify these mutagens, the more intelligent our behavior might be. Are people smoking less than they use to? Sure. And why, because people are aware and accept the obvious conclusion that the mutagens in cigarette smoke are indeed carcinogenic. Now remember, there's another location and that's in the sex cells. Where are the sex cells or what are the gonads anyway? Those are the testis and the ovaries. What are they doing that's special or otherwise different? Well, they're making sperm or eggs. Now, if we mutate those, you're using those for what if you're using them at all? You're using that for sexual reproduction. So if you mutate your cells that are making sperm, will those mutations be utilized unwittingly, unknowingly to conceive a child, and will that child then be born with that mutation? Yeah. Are they going to be dead on the spot? Not necessarily. Because remember, every gene that you have, you have two copies of, right? You got a copy of that gene from your dad and a copy of that gene from your mom, so that mitigates some of the impact. But are there individuals? Are we all inheritors of mutations that our mother or father may have passed us unknowingly? Yes. And so, does that give us some risk factor for cancer? Sure. So, are we more aware now of that and do we use that to screen or otherwise choose a course of action? I'm thinking of Angelina Jolie, right? Famous movie star. What she do recently? Had both of her breasts cut off. You missed that story? No, I'm serious. And that's because her family, her sisters, her mother died of breast cancer. And she was tested for the genes that are now known to be associated with that event. And she was positive on both counts. She knew it's just a matter of time for her, right? So, what did she do? She's very proactive, cut the breast off. Will that extend her life? Undoubtedly. So, that's a case of surgery before what? We don't want to wait until the tumor is there. Let's cut off these cells that are likely to be a future problem in that regard. So, sexual mutations are less common but when they happen, they're going to be passed to offspring. And that may or may not produce immediate birth defects or an increased incidence of cancer. Why are sexual mutations less common? Well, because frankly is the gonads are pretty well protected certainly from ultraviolet light, right? So, not likely to be affected except with systemic radiation exposure or chemotherapy. All right, final topic for the night. Actually, a complete change of focus but an intriguing question really. The title of this is Regulation of Gene Expression. So let's accept this number, how many genes in a homosapien genome, that is genes in your cells, let's say, 20,000. OK, 20,000 genes. Do all cells of your body have the same 20,000 genes? The answer is yes. Really? So that means, a skin cell has the same genes as found in a brain cells. Is that right? Absolutely true. They all have the same number of genes. Are they all using or do they need to use all 20,000? You might say, I don't know. You know, but the answer is decidedly no. So this chart is kind of interesting, accepting the notion that there are may be 20,000 genes. How many does the brain use on an ordinary day or the rest of these organs? Interestingly, the brain needs and uses about 3000 of those on a daily basis whereas what? Bone uses only what? 904. Now, why is that? Well, the whole premise, the whole design of the human body is that cells will specialize. So let's be really facetious if not silly here. Does every cell in your body have the gene to make the pigment known as melanin? Melanin is a brown pigment, right? Do all of our cells have that gene? Do all of cells use that gene? Do all of our cells make melanin? No. And the reason is obvious. Do we need a brown heart? Being funny there. But we don't need a brown heart. We don't need a brown colored ovary or something. What cells do make and do depend on the production of melanin, skin cells. And what is the function of melanin in that location? To screen, to block ultraviolet light. So, never mind that example. Do all cells of your body have the gene to make insulin? Yeah, they do. Do all cells in your body make insulin? No. Why not? Well, the simple answer is they don't need to because the whole strategy of the human body is you do this, we'll do that. In other words, you specialize in that, we'll specialize in that. So what cells specialize in produce melanin? What cells specialize in make insulin? Those at the pancreas. Do we need the heart to make insulin? You get my point. So, these numbers are based upon the activities that this organs conduct and the numbers are greater for those tissues that have a more broad, more specialized, more serious of functions. So, what's my point is, we all have 20,000 genes. Are they all turned on all of time in every cell? No. Some are turned on. Some are turned off. Which of these on the chart here apparently have more of those 20,000 turned on? It seems like that. Which have very few turned on? So it

correlates obviously with the complexity of the organ in question. And why should this even be important? Do we want every cell in your body to make insulin? No. Do we want every cell in your body to make melanin? No. So, if we're going to control whether gene is turned on or off, where we're going to do that? What do I even mean? Genes are on the DNA, right? DNA, genes. And they control the production of first of all, what, mRNA. And the name of that process that leads to that is called transcription, right? And after that, we have the next phase which leads to protein synthesis. And that's called translation. So, if we're going to control the expression of genes, that is if we're going to determine what genes are turned on or not, where would we want to control that? Would we want to control it by blocking or affecting translation or blocking and affecting transcription? You might say, "Well, I don't know the difference." But consider this to be in assembly line that's making something, in this case, protein. Do you want to start or stop that at the beginning or start and stop it near the end?

>> At the beginning.

>> At the beginning. Why does it make more economical sense to control it at the beginning? Well, you're not going to waste all this time and energy to produce something that you don't need or otherwise don't want. So, where that is--at what stage is gene expression controlled. It's at the transcription stage. Because if we block transcription, we stop it right at the get go. And the case for this is right here. Why do we even want to do this? We want to control protein synthesis for the sake of two things. What are we saving, what are we mindful, what's the advantage of controlling gene expression is that it saves what? It saves energy and it enables cells to specialize. So without the ability to control gene expression, all cells would make all proteins all the time. And what would that be? You would be a pile of 70,000 identical cells. And it would be an ugly mess over here, science fiction. So, what is the advantage of controlling gene expression is that it enables cells to specialize. It enables the skin cells to make melanin and other's not. But incidentally, that fact--what was the fact? All cells have what? All cells have all the genes and so can theoretically the heart produce insulin. Does the heart have the gene to make insulin? It does. Why doesn't it? Because it's not its thing, right? But does it have the gene? Yeah. And because it does, that's an intriguing idea because what if the cells that normally make insulin, the beta cells of the pancreas, what if they're not producing it as in type 1 diabetes? Does the heart have that gene? And could we turn that on? And if we did, would we've solved the problem? You bet. So, the fact that all cells have all the genes, at least, makes it possible for these kinds of therapies. So let's finish this off. We've said that we control gene expression right here at the transcription level. So, what we're about to explain in simplest term as we can is what's called gene induction and gene repression. What's it means to induce something?

[Inaudible Remark]

What's it mean to repress something? Repress, turn off. Induce, turn, on. So, what are the mechanisms, the known mechanisms for gene induction? Gene induction basically involves something called an activated transcription factor which attaches to locations upstream of a gene that is on the DNA that control that gene. And dictate and determine the attachment of this enzyme or in a polymerase too. So, this is a very primitive diagram but I think it makes some sense. Here is the gene shown us a double helix that's making an enzyme called enzyme X, right? If we transcribe that gene, we're going to make what? RNA. And once we have transcription then translation is automatic. That is once we have mRNA, we're going to make the protein, like it or not. So clearly, controlling this step which we said is transcription will determine whether and when that protein is produced. But what determines whether and when that gene is in fact transcribed is an area usually adjacent to the gene called, what's the word here, promoter region. The promoter region has two sites on it. One which accommodates this enzyme called RNA polymerase II. The other which accommodates unknown, that is largely unknown, so-called transcription factors. This fit into this promoter region. And when they're both present, when the activated transcription factor is there, RNA polymerase will attach and what process will occur? Transcription. And will the protein be made? Yes, for as long as what? For as long as that activated transcription factor is there. So, as an analogy, this activated transcription factor is like a finger which is flipping the switch and allowing this to take place. And the protein will be made as long as what's present? As long as the activated transcription factor is there. What does that even mean? An activated transcription factor is activated. That is, it's a molecule which is activated you see by what? Extracellular or intracellular signals which I know is vague. So let's try to make it real with an example. Do all cells of your body have the ability to make the enzyme amylase? Yes, they have that gene. What cells of your body do make amylase? Those in your salivary glands and also those in your pancreas. What induces them apparently is an activated transcription factor which turns out is linked to the availability of starch. So when we dealt with amylase in our enzyme lab, you spit in a cup. And some tables, the reaction was very quick. Others, it was kind of

slow. And I made the comic remark, "Well, that's powerful spit." But it wasn't quite that farfetched. Do people have more or less amylase? My salivary glands would be making more amylase than yours? And what's the reason? Well apparently, my gene might be, what's the I word?

>> [Simultaneous] Induced.

>> Induced. And yours might not be because of the ability of a what? Extracellular signal, in this case, the presence of starch. So, just as a continuation of that thought, are people who have very low starch in their diet. Not in this country but OK, are there societies where their food is notoriously low in starch? And the best example that I know of is the actual Alaskan natives because they're not growing corn, right? How do they live, those guys? They kill seals and they eat blubber and basically, fat and protein is their whole deal. It's Atkins to the max. All right. So, do they have a lot of carbohydrates in their diet? Do they have the gene to make amylase? Yes. Do they make amylase? No. Why, because the gene is not induced, it's what? Repressed. And what's the advantage of it being repressed? Think of it logically. Of course, it's simple to say, well, there's no point in making amylase if you're not consuming what? Starch. And that's the simple fact because it enables as we've said earlier, what's the physiological significance of all these? It allows for energy what? Energy efficiency, not wasting. Because does it take energy to make a protein? And not only does it take energy to make protein, it takes raw materials like amino acids. So anyway, what is gene repression? Well, obviously, it's the opposite of gene induction. In other words, the promoter site is blocked or otherwise not active, not because there is an RNA polymerase. RNA polymerase is there but it can't attached because there is no, what is it? Activated transcription factor. And therefore, what gets shut down? What process is stopped? Transcription. And is that a good thing? Why is no transcription a good thing? Well, what's the point--yeah, making a protein for which there is no purpose and for which there is no substrate is an example. So, there are plenty of other examples. And one that I'll finish with is familiar enough. There are some people who rarely consume alcohol. And when they do, they usually get tipsy with six ounce beer. Am I making this up?

[Inaudible Remark]

Where in the other hand, you have guys that are knocking down, you know, three six packs and they're still standing, all right? And what's the deal there? Well, you see they have a higher tolerance. But really what they have is more of an enzyme called alcohol dehydrogenase. The liver makes that enzyme. It called alcohol dehydrogenase. And it breaks down and metabolizes alcohol. So why do some people have very little alcohol dehydrogenase?

>> Because they don't drink.

>> They don't drink. Therefore, do they have that gene that could make that enzyme? Sure. Why aren't they cracking out that enzyme? They don't drink. What's the point of making an enzyme, all right? So, how do you get that kind of tolerance? Well, start drinking.

[Laughter]

No, I didn't actually say that, did I? Yeah, I'll meet you over at Schooner's [assumed spelling] and we'll talk about it more. We're done.

[Inaudible Discussion]