0:0:6.750 --> 0:0:7.470  
Evanthia Anastasiadou  
OK.

0:0:9.910 --> 0:0:10.830  
Evanthia Anastasiadou  
I think.

0:0:12.590 --> 0:0:16.710  
Evanthia Anastasiadou  
So it's recording and transcription, so I suppose that it is recording.

0:0:19.180 --> 0:0:21.180  
Evanthia Anastasiadou  
OK, so going to.

0:0:23.30 --> 0:0:23.350  
Evanthia Anastasiadou  
Today's lecture.

0:0:28.380 --> 0:0:30.980  
Evanthia Anastasiadou  
As I said, you can stop me anytime you want.

0:0:30.980 --> 0:0:40.660  
Evanthia Anastasiadou  
We can discuss it further. We can explain it further if you have any other issues that you would like to share with me, just let me know so I'll try.

0:0:44.380 --> 0:0:53.100  
Evanthia Anastasiadou  
I'll try to cover most of the the stuff for in the next hour. Then we can have a break and then we can continue.

0:0:53.900 --> 0:0:56.300  
Evanthia Anastasiadou  
So we are talking about biotechnology.

0:0:56.860 --> 0:0:59.420  
Evanthia Anastasiadou  
You already had the class on biotechnology, correct?

0:1:2.530 --> 0:1:2.850  
nickcharisis182  
Yes.

0:1:5.440 --> 0:1:9.240  
Evanthia Anastasiadou  
Briefly, what did you? You had only one so far.

0:1:14.150 --> 0:1:15.910  
Evanthia Anastasiadou  
One or two courses that you have so far.

0:1:18.420 --> 0:1:18.740  
nickcharisis182  
We had.

0:1:18.740 --> 0:1:21.500  
nickcharisis182  
I don't know if I correct correctly.

0:1:21.500 --> 0:1:23.820  
nickcharisis182  
3 lectures with Mr. Stubbows.

0:1:24.820 --> 0:1:25.420  
violetta.gk98  
Yes.

0:1:25.100 --> 0:1:25.900  
Evanthia Anastasiadou  
Oh, OK.

0:1:25.980 --> 0:1:32.980  
Evanthia Anastasiadou  
So you're OK, so you're already covered the basics. What biotechnology's about and?

0:1:34.550 --> 0:1:38.390  
Evanthia Anastasiadou  
Did he start teaching about some methodology that we are using?

0:1:41.90 --> 0:1:45.330  
Evanthia Anastasiadou  
And I have at least one of the biologists telling me what was covered.

0:1:46.590 --> 0:2:2.470  
rafailadam46  
He actually told us about micro arrays nzs and you know the sub technologies for nzs and the last time I believe he told us about some weblab stuff, mostly like Western blood northern and all that stuff.

0:2:4.360 --> 0:2:11.560  
Evanthia Anastasiadou  
Oh, my God. What about my engineer and my computer scientist? Are you still survive?

0:2:14.460 --> 0:2:15.380  
nickcharisis182  
I don't know where.

0:2:15.660 --> 0:2:16.300  
Iro Chasapi  
Trying to.

0:2:15.840 --> 0:2:20.320  
Evanthia Anastasiadou  
You trying to, OK.

0:2:17.300 --> 0:2:18.500  
nickcharisis182  
Yeah, with a bit of a view.

0:2:20.560 --> 0:2:23.360  
Evanthia Anastasiadou  
So that's heavy stuff, isn't it?

0:2:27.60 --> 0:2:29.420  
Ioannis Mystakidis  
I didn't get most of it, to be honest.

0:2:27.510 --> 0:2:27.910  
Evanthia Anastasiadou  
Yes.

0:2:31.180 --> 0:2:40.20  
Evanthia Anastasiadou  
Well, I totally understand that. I'm pretty sure that some of the biologists didn't get most of it also, so don't worry about it.

0:2:40.20 --> 0:2:55.700  
Evanthia Anastasiadou  
I hope that today's gonna be much, much lighter than that, and I hope also that it's gonna cover things that might interest you and are closer to your, let's say, biological questions.

0:2:56.710 --> 0:3:1.150  
Evanthia Anastasiadou  
However, you have to go back to the previous lectures and revise them.

0:3:1.640 --> 0:3:3.360  
Evanthia Anastasiadou  
Really. Because most likely.

0:3:5.610 --> 0:3:9.450  
Evanthia Anastasiadou  
Will be through the methodology that doctors travel already taught you.

0:3:10.50 --> 0:3:12.130  
Evanthia Anastasiadou  
So without any delays, let's start.

0:3:14.870 --> 0:3:17.270  
Evanthia Anastasiadou  
I'm gonna talk to you about genetic engineering.

0:3:18.830 --> 0:3:19.70  
Evanthia Anastasiadou  
So.

0:3:20.750 --> 0:3:23.230  
Evanthia Anastasiadou  
My engineer here will tell.

0:3:24.830 --> 0:3:28.590  
Evanthia Anastasiadou  
You even my DI people will tell you when we are talking about engineering.

0:3:30.390 --> 0:3:42.390  
Evanthia Anastasiadou  
It's the way that you are producing things. The why the way that you are creating new things and that's exactly what we are doing Internet engineering.

0:3:42.510 --> 0:3:51.190  
Evanthia Anastasiadou  
So we are using the term engineering to describe that we are using it to generate new.

0:3:51.720 --> 0:3:53.320  
Evanthia Anastasiadou  
Genetic material.

0:3:53.880 --> 0:4:0.560  
Evanthia Anastasiadou  
So basically DNA manipulation using molecular biology techniques.

0:4:1.240 --> 0:4:12.320  
Evanthia Anastasiadou  
We also, apart from genetic engineering, you are going to see it as Gen. cloning, genetic manipulation, GM, genetic editing.

0:4:12.960 --> 0:4:18.760  
Evanthia Anastasiadou  
Do not confuse the term genetic engineering gene cloning with animal cloning.

0:4:19.550 --> 0:4:21.590  
Evanthia Anastasiadou  
Animal cloning is a totally different thing.

0:4:22.530 --> 0:4:22.730  
Evanthia Anastasiadou  
K.

0:4:22.970 --> 0:4:27.490  
Evanthia Anastasiadou  
So Gene cloning is completely different than animal cloning.

0:4:29.30 --> 0:4:31.350  
Evanthia Anastasiadou  
So we are gonna talk about genetic engineering.

0:4:31.590 --> 0:4:41.270  
Evanthia Anastasiadou  
So when you're talking about genetic engineering, basically you are isolating, amplifying an individual sequence.

0:4:41.900 --> 0:4:46.820  
Evanthia Anastasiadou  
So I'm not sure how much you know about the DNA in sequences.

0:4:46.820 --> 0:5:5.620  
Evanthia Anastasiadou  
So when you say a sequence is a part of your DNA, so a specific part of your DNA, you can isolate it and you can amplify it, make it make it. A lot involves the construction of novel DNA molecules by joining DNA from different sources.

0:5:8.310 --> 0:5:11.590  
Evanthia Anastasiadou  
The product of this procedure.

0:5:11.940 --> 0:5:13.500  
Evanthia Anastasiadou  
Is called recombinant DNA.

0:5:13.500 --> 0:5:19.180  
Evanthia Anastasiadou  
You might find it also ask are DNA. R stands for recombinant.

0:5:23.60 --> 0:5:36.620  
Evanthia Anastasiadou  
So because it is biotechnology, we have to go through the methodology and starting with a clean gene, cloning basic methodology, we have to follow some events.

0:5:37.140 --> 0:5:41.900  
Evanthia Anastasiadou  
First of all you have to isolate the DNA sequence of your interest.

0:5:42.260 --> 0:5:43.460  
Evanthia Anastasiadou  
Is this a gene?

0:5:43.740 --> 0:5:46.180  
Evanthia Anastasiadou  
Is this a transport?

0:5:46.700 --> 0:5:50.780  
Evanthia Anastasiadou  
Is this a regulatory sequence like promoters?

0:5:50.900 --> 0:5:52.740  
Evanthia Anastasiadou  
So first of all you have to.

0:5:53.190 --> 0:5:57.110  
Evanthia Anastasiadou  
Late, the part of the DNA that you want to investigate further.

0:5:58.670 --> 0:6:8.870  
Evanthia Anastasiadou  
So you isolated and then you incorporated you are adding it to another piece of DNA that we call vector.

0:6:11.110 --> 0:6:17.30  
Evanthia Anastasiadou  
Vector is the carrier of the DNA that you want to investigate. So.

0:6:18.590 --> 0:6:25.550  
Evanthia Anastasiadou  
We are gonna talk about vectors in details. For the moment, I would like to keep in mind that is a small.

0:6:27.750 --> 0:6:27.950  
Evanthia Anastasiadou  
Circular.

0:6:30.230 --> 0:6:34.790  
Evanthia Anastasiadou  
Usually DNA molecule that can replicate it can multiply.

0:6:36.550 --> 0:6:40.990  
Evanthia Anastasiadou  
Tell me something. If I'm moving my cursor on the screen, can you see it or you cannot?

0:6:45.420 --> 0:6:48.180  
nickcharisis182  
If you're moving it right now, I can't see anything.

0:6:45.500 --> 0:6:46.460  
Ioannis Mystakidis  
You know.

0:6:48.420 --> 0:6:48.900  
nickcharisis182  
I can't see any.

0:6:48.530 --> 0:6:48.730  
Evanthia Anastasiadou  
OK.

0:6:51.690 --> 0:6:51.890  
Evanthia Anastasiadou  
OK.

0:6:51.930 --> 0:6:55.650  
Evanthia Anastasiadou  
So now you can see that I'm showing the 1st circle.

0:6:55.650 --> 0:6:56.930  
Evanthia Anastasiadou  
The 2nd circle correct.

0:6:58.970 --> 0:6:59.330  
Ioannis Mystakidis  
No.

0:7:1.130 --> 0:7:1.490  
Evanthia Anastasiadou  
Oh, OK.

0:7:3.30 --> 0:7:3.230  
Evanthia Anastasiadou  
OK.

0:7:3.230 --> 0:7:6.150  
Evanthia Anastasiadou  
So I'm not sure really sure how I should point.

0:7:10.150 --> 0:7:11.630  
Evanthia Anastasiadou  
Then later OK found it.

0:7:13.190 --> 0:7:13.390  
Evanthia Anastasiadou  
Later.

0:7:14.990 --> 0:7:15.150  
Evanthia Anastasiadou  
OK.

0:7:15.270 --> 0:7:16.510  
Evanthia Anastasiadou  
But now you can see it correct.

0:7:18.430 --> 0:7:18.830  
Ioannis Mystakidis  
Yes.

0:7:20.80 --> 0:7:34.240  
Evanthia Anastasiadou  
Excellent. So we said, first of all, you isolate the fragment of your DNA that is of your interest and then you are using a circular molecule of DNA, we call it vector or you can call it plasmid also.

0:7:35.790 --> 0:7:42.550  
Evanthia Anastasiadou  
This is a circular, small molecule of DNA that can multiply. It can replicate.

0:7:44.480 --> 0:7:50.960  
Evanthia Anastasiadou  
So then you merge that. These those things together and you create a recombinant plasmid.

0:7:50.960 --> 0:7:57.80  
Evanthia Anastasiadou  
This recombinant plasmid already has the background of your initial vector.

0:7:58.630 --> 0:8:2.390  
Evanthia Anastasiadou  
With insertion of the gene or the sequence of your interest.

0:8:4.70 --> 0:8:14.750  
Evanthia Anastasiadou  
Now that you have this circular molecule that can multiply what you are going to do is you are going to insert it in bacteria. We use very strange of bacteria as.

0:8:15.680 --> 0:8:20.280  
Evanthia Anastasiadou  
Strange of bacteria in the lab, the most common one is E coli.

0:8:20.920 --> 0:8:25.760  
Evanthia Anastasiadou  
So we have various techniques like electropolation or heat shocking.

0:8:25.880 --> 0:8:35.320  
Evanthia Anastasiadou  
There are various techniques that you can manage to put your circular molecule of DNA in your bacteria. When you put it in there.

0:8:35.320 --> 0:8:41.680  
Evanthia Anastasiadou  
Then you'll start multiplying it and you are start multiplying till you get enough material.

0:8:42.510 --> 0:8:44.790  
Evanthia Anastasiadou  
To move your experiments further on.

0:8:47.270 --> 0:8:52.670  
Evanthia Anastasiadou  
So first of all, isolate the fragment of your interest.

0:8:52.870 --> 0:8:55.590  
Evanthia Anastasiadou  
Insert it in a plasmid vector.

0:8:56.270 --> 0:9:3.150  
Evanthia Anastasiadou  
Put it in bacterial cells multiplied till you have enough material to work on.

0:9:6.640 --> 0:9:22.880  
Evanthia Anastasiadou  
When you are talking about your cloning factor, the plasmid that you are gonna use for your experiments, as I said, usually circular is small and has what we call multiple cloning site.

0:9:23.240 --> 0:9:29.800  
Evanthia Anastasiadou  
This is usually your choice where you're gonna insert your DNA sequence.

0:9:31.550 --> 0:9:35.910  
Evanthia Anastasiadou  
I'm gonna discuss and explain the multicloning sites in a couple of slides.

0:9:36.430 --> 0:9:37.710  
Evanthia Anastasiadou  
Now a part of your.

0:9:38.650 --> 0:9:39.850  
Evanthia Anastasiadou  
Cloning site.

0:9:41.390 --> 0:9:41.950  
Evanthia Anastasiadou  
You also have.

0:9:43.630 --> 0:9:49.310  
Evanthia Anastasiadou  
The sequence of interest and your vector is going to have another gene.

0:9:49.950 --> 0:9:58.870  
Evanthia Anastasiadou  
This gene is giving to your plasmid resistant for one antibiotic the most common one is ampicillin.

0:9:59.270 --> 0:10:8.190  
Evanthia Anastasiadou  
So you are having there a gene that the bacteria that are having this plasmid will be resistant.

0:10:9.70 --> 0:10:9.630  
Evanthia Anastasiadou  
In this specific.

0:10:10.120 --> 0:10:15.920  
Evanthia Anastasiadou  
The biotic. This is very important and I'll explain why it is important.

0:10:16.320 --> 0:10:22.480  
Evanthia Anastasiadou  
So we said that you put your plasmid with the sequence of your interest in the bacteria.

0:10:24.710 --> 0:10:29.510  
Evanthia Anastasiadou  
The bacteria will start multiplying as they will multiply.

0:10:29.550 --> 0:10:39.390  
Evanthia Anastasiadou  
They have a tendency to spit out whatever is not necessary for their survival and their.

0:10:41.870 --> 0:10:43.750  
Evanthia Anastasiadou  
And their proliferation.

0:10:44.510 --> 0:10:54.790  
Evanthia Anastasiadou  
So if you don't force them, force them to keep your plasmid after a couple of divisions, they will not have it there.

0:10:55.270 --> 0:10:58.150  
Evanthia Anastasiadou  
So that's why we are using this antibiotic.

0:10:59.710 --> 0:11:14.110  
Evanthia Anastasiadou  
Externally, putting it in the medium where we are calculating the bacteria so only the bacteria that are carrying the plasmid will be the ones that will survive.

0:11:14.510 --> 0:11:23.30  
Evanthia Anastasiadou  
Why? Because this will be the only ones that they have the resistant gene in this specific antibiotic.

0:11:23.750 --> 0:11:25.510  
Evanthia Anastasiadou  
So if you don't have it.

0:11:26.310 --> 0:11:31.430  
Evanthia Anastasiadou  
After a couple of generations, they will spit out this one because it makes them.

0:11:32.640 --> 0:11:37.720  
Evanthia Anastasiadou  
Delay so they will spit it out and you will not have anything to work with.

0:11:38.80 --> 0:11:47.560  
Evanthia Anastasiadou  
You need this antibiotic outside, so all your bacteria cells from now on will have the plasmid of your interest.

0:11:49.710 --> 0:11:52.550  
Evanthia Anastasiadou  
So we talked about the multiple cloning side.

0:11:52.750 --> 0:11:55.30  
Evanthia Anastasiadou  
We talked about the gene of your interest.

0:11:55.150 --> 0:12:2.110  
Evanthia Anastasiadou  
We talk about the resistant, the the resistant gene, we call it market, gene.

0:12:3.670 --> 0:12:7.710  
Evanthia Anastasiadou  
Also, it will have an origin.

0:12:7.910 --> 0:12:13.510  
Evanthia Anastasiadou  
You might most likely find it as Ori. This is the origin of replication.

0:12:13.790 --> 0:12:18.950  
Evanthia Anastasiadou  
That's from where the replication of your DNA will start.

0:12:19.790 --> 0:12:23.750  
Evanthia Anastasiadou  
And you want that because without the knowledge of replication.

0:12:25.390 --> 0:12:46.110  
Evanthia Anastasiadou  
Multiply therefore Azure bacterial cells will multiply. If your plasmid does not multiply also, you will end up with bacteria with no plasmid, so the plasmid has to multiply to follow the multiplication of your bacterial cells.

0:12:48.750 --> 0:12:58.350  
Evanthia Anastasiadou  
So once again, when we are talking about cloning vector, you talk about a small circular DNA molecule that has an origin replication.

0:12:58.430 --> 0:13:14.110  
Evanthia Anastasiadou  
That's from where it will start replicating. It will have a resistance in most likely in one of the antibiotics that you use in the in the medium that you are growing, your bacteria will have the sequence of your interest.

0:13:14.910 --> 0:13:19.630  
Evanthia Anastasiadou  
And the multiple cloning site I said that I'm going to come back to that.

0:13:20.280 --> 0:13:21.0  
Evanthia Anastasiadou  
In a second.

0:13:23.40 --> 0:13:26.0  
Evanthia Anastasiadou  
We have various types of cloning vectors.

0:13:26.90 --> 0:13:28.490  
Evanthia Anastasiadou  
The most common one is the plasma.

0:13:30.40 --> 0:13:37.720  
Evanthia Anastasiadou  
The the plasmid is the one that you most likely are gonna use it extensively, but we have some limitations.

0:13:37.920 --> 0:13:40.200  
Evanthia Anastasiadou  
The limitations is how much?

0:13:41.880 --> 0:13:43.240  
Evanthia Anastasiadou  
The the sequence of your interest.

0:13:43.240 --> 0:13:46.280  
Evanthia Anastasiadou  
How how big is it so there is?

0:13:48.200 --> 0:13:48.600  
Evanthia Anastasiadou  
A limit?

0:13:48.640 --> 0:13:52.720  
Evanthia Anastasiadou  
How much DNA can you put in your plasmid?

0:13:53.120 --> 0:13:57.0  
Evanthia Anastasiadou  
So if you are talking about a couple of kilo bases.

0:13:57.620 --> 0:14:0.100  
Evanthia Anastasiadou  
Then plasmid is fine and you can use it.

0:14:0.380 --> 0:14:5.20  
Evanthia Anastasiadou  
But if you are talking more than that, you might have to find other.

0:14:6.560 --> 0:14:9.720  
Evanthia Anastasiadou  
Possibilities like cosmates for example.

0:14:9.800 --> 0:14:17.360  
Evanthia Anastasiadou  
But they can take up to 40K faulty thousand base pairs or phages.

0:14:19.40 --> 0:14:26.80  
Evanthia Anastasiadou  
Or yeast artificial chromosomes, bacterial artificial chromosomes, even viral vectors.

0:14:28.410 --> 0:14:33.130  
Evanthia Anastasiadou  
So for the moment, I want you to keep in mind plasmids.

0:14:33.130 --> 0:14:49.570  
Evanthia Anastasiadou  
What are the basic component of the plasmids and according to the purpose that you want to multiply your sequence and the size of your sequence, you might have apart from plasmids other sources like cosmids phages.

0:14:50.970 --> 0:14:52.530  
Evanthia Anastasiadou  
Yaks and box.

0:14:55.90 --> 0:15:1.250  
Evanthia Anastasiadou  
Provider vectors. We are going to talk about about them extensively towards the end of our today's lecture.

0:15:3.820 --> 0:15:4.580  
Evanthia Anastasiadou  
So.

0:15:6.230 --> 0:15:11.190  
Evanthia Anastasiadou  
OK, let me talk about this one first and then I'm gonna come back.

0:15:11.670 --> 0:15:13.670  
Evanthia Anastasiadou  
So restrictions and signs.

0:15:15.210 --> 0:15:19.970  
Evanthia Anastasiadou  
Remember that I was talking to you before about multiple cloning sites.

0:15:20.290 --> 0:15:31.90  
Evanthia Anastasiadou  
The multiple cloning sites is where you are gonna cut your plasmid to be able to stitch in your sequence of interest.

0:15:31.770 --> 0:15:33.170  
Evanthia Anastasiadou  
How will you cut?

0:15:33.450 --> 0:15:35.250  
Evanthia Anastasiadou  
You're gonna cut with.

0:15:37.10 --> 0:15:40.410  
Evanthia Anastasiadou  
Some chemicals that will call restriction enzymes.

0:15:40.930 --> 0:15:43.170  
Evanthia Anastasiadou  
How are you gonna stitch them together?

0:15:43.820 --> 0:15:53.860  
Evanthia Anastasiadou  
With another compound called DNA ligase, both DNA ligase and restriction enzymes are proteins.

0:15:55.410 --> 0:15:56.610  
Evanthia Anastasiadou  
Proteins with specific.

0:15:58.330 --> 0:15:58.890  
Evanthia Anastasiadou  
Properties.

0:16:0.610 --> 0:16:1.130  
Evanthia Anastasiadou  
This one.

0:16:1.130 --> 0:16:9.690  
Evanthia Anastasiadou  
The restriction enzymes can recognize a specific sequence in your DNA and cut it in a.

0:16:12.350 --> 0:16:16.310  
Evanthia Anastasiadou  
The DNA ligase will stitch together.

0:16:17.850 --> 0:16:20.770  
Evanthia Anastasiadou  
So and seal the cuttings that you have.

0:16:21.690 --> 0:16:24.770  
Evanthia Anastasiadou  
So basically, it's like your seizure.

0:16:26.850 --> 0:16:33.490  
Evanthia Anastasiadou  
And your needle you are cutting with a restriction enzymes and you are.

0:16:35.170 --> 0:16:37.410  
Evanthia Anastasiadou  
Stitching them together with adna liquids.

0:16:39.500 --> 0:16:41.260  
Evanthia Anastasiadou  
Now look what is happening.

0:16:41.580 --> 0:16:55.60  
Evanthia Anastasiadou  
Restriction enzymes are naturally you can naturally find them in a lot of prokaryotes. OK, so only found in prokaryotes like bacterial, let's say.

0:16:56.610 --> 0:16:57.490  
Evanthia Anastasiadou  
And this is.

0:16:59.370 --> 0:17:1.850  
Evanthia Anastasiadou  
This is a survival mechanism.

0:17:2.170 --> 0:17:3.450  
Evanthia Anastasiadou  
So for example.

0:17:5.130 --> 0:17:10.610  
Evanthia Anastasiadou  
In your bacteria, if your bacteria are infected by the bacterial viruses like phages.

0:17:11.530 --> 0:17:25.650  
Evanthia Anastasiadou  
They have these enzymes to cut the DNA of the viruses, so therefore the DNA will be degraded and will not be able to infect the bacteria.

0:17:26.130 --> 0:17:34.10  
Evanthia Anastasiadou  
So it's a defense mechanism that, for example, prokaryotes such as bacteria develop.

0:17:37.370 --> 0:17:41.130  
Evanthia Anastasiadou  
To to escape from infections like viral infections.

0:17:42.130 --> 0:17:47.850  
Evanthia Anastasiadou  
So this and we have many different enzymes over 100.

0:17:47.850 --> 0:17:51.370  
Evanthia Anastasiadou  
I think that nowadays is even higher than that.

0:17:51.370 --> 0:17:52.930  
Evanthia Anastasiadou  
The number is even higher than that.

0:17:53.250 --> 0:18:1.770  
Evanthia Anastasiadou  
So what is happening is your enzyme will recognize a specific sequence. As you can see here.

0:18:3.330 --> 0:18:8.50  
Evanthia Anastasiadou  
E1 will recognize a sequence in your DNA that will be.

0:18:10.810 --> 0:18:11.570  
Evanthia Anastasiadou  
GAAP C.

0:18:12.790 --> 0:18:17.630  
Evanthia Anastasiadou  
Usually these enzymes are recognizing a palindrome.

0:18:17.990 --> 0:18:23.670  
Evanthia Anastasiadou  
A palindrome means you can see you have daaja OK.

0:18:24.670 --> 0:18:31.310  
Evanthia Anastasiadou  
So this is a palindrome. So the enzymes the restriction enzymes usually realize.

0:18:32.890 --> 0:18:33.810  
Evanthia Anastasiadou  
A specific sequence.

0:18:33.810 --> 0:18:41.970  
Evanthia Anastasiadou  
So as I said, you can see E1 recognizes TAATTC bam H1 recognises GGA.

0:18:43.350 --> 0:18:49.630  
Evanthia Anastasiadou  
The CC so you can see that even one base difference can be recognized.

0:18:51.170 --> 0:18:57.690  
Evanthia Anastasiadou  
And these enzymes are gonna recognize they are gonna bind on this DNA sequence, and they are gonna break.

0:18:57.730 --> 0:19:2.930  
Evanthia Anastasiadou  
They are gonna cut it the way that they are gonna cut it is two different ways.

0:19:4.530 --> 0:19:8.890  
Evanthia Anastasiadou  
Either they will cut it in a way that you are gonna have overhangs.

0:19:9.130 --> 0:19:16.90  
Evanthia Anastasiadou  
So you can see at the end that you are having some single strand, the DNA, some.

0:19:17.790 --> 0:19:20.710  
Evanthia Anastasiadou  
They are not matching with anything else on the other strain.

0:19:23.450 --> 0:19:32.730  
Evanthia Anastasiadou  
So we are called this overhangs or it's going to be a bland end. Breaking that means you don't have overhangs.

0:19:34.410 --> 0:19:38.810  
Evanthia Anastasiadou  
Having this overhangs will help you stitch together.

0:19:40.450 --> 0:19:49.130  
Evanthia Anastasiadou  
Other overhang that you had traded before with this restriction enzymes, so you need to try to find out.

0:19:49.740 --> 0:19:51.900  
Evanthia Anastasiadou  
Which restriction enzymes they are?

0:19:51.900 --> 0:19:57.620  
Evanthia Anastasiadou  
Overhangs are matching together, or if you don't have any choice, you have to go for the bland end.

0:19:59.410 --> 0:20:13.890  
Evanthia Anastasiadou  
Ligase. So once again, when we are talking about multiple cloning sites, we are talking about a site that has many different sequences recognized by different restriction enzymes.

0:20:14.10 --> 0:20:18.330  
Evanthia Anastasiadou  
So this is giving you an option of what enzymes to use.

0:20:19.490 --> 0:20:24.770  
Evanthia Anastasiadou  
Cut and stage together your plasmic and the sequence of your interest.

0:20:26.330 --> 0:20:27.850  
Evanthia Anastasiadou  
Just to remind you for the.

0:20:30.130 --> 0:20:35.170  
Evanthia Anastasiadou  
For the isolation of and the characterization of restriction enzymes.

0:20:36.930 --> 0:20:37.370  
Evanthia Anastasiadou  
Warner Arbor.

0:20:37.650 --> 0:20:46.730  
Evanthia Anastasiadou  
Daniel Mathen and Hamilton of Smith O Smith got the Nobel Prize 80s and 80s back 1978.

0:20:47.170 --> 0:20:49.10  
Evanthia Anastasiadou  
It was a very important discovery.

0:20:50.850 --> 0:20:56.370  
Evanthia Anastasiadou  
So now that you know how we are cutting the DNA, how are we putting it together?

0:20:58.370 --> 0:21:8.730  
Evanthia Anastasiadou  
DNA ligase, as I said, is an an enzyme that seals single stranded mix between adjacent nucleotides.

0:21:10.530 --> 0:21:19.10  
Evanthia Anastasiadou  
In the double X DNA chains, so catalyse the formation of phosphodiester bonds between adjacent.

0:21:21.200 --> 0:21:25.200  
Evanthia Anastasiadou  
3 hydroxyl and five phosphate termini in the DNA.

0:21:27.410 --> 0:21:30.570  
Evanthia Anastasiadou  
Have you Start learning about DNA and the how?

0:21:32.490 --> 0:21:34.290  
Evanthia Anastasiadou  
The chemical structure of the DNA.

0:21:41.200 --> 0:21:43.360  
Evanthia Anastasiadou  
Your biology class, OK.

0:21:41.470 --> 0:21:42.30  
Vasilis Chatzitolios  
No.

0:21:43.760 --> 0:21:45.800  
Evanthia Anastasiadou  
So forget about all this.

0:21:45.800 --> 0:21:56.80  
Evanthia Anastasiadou  
Just keep in mind that DNA ligase stages, different parts of DNA together, but you had previously cut with your restriction enzymes.

0:21:58.110 --> 0:22:2.830  
Evanthia Anastasiadou  
So just to oversee once again what is happening during cloning.

0:22:2.990 --> 0:22:10.710  
Evanthia Anastasiadou  
So we said, first of all, you have to isolate the sequence of your interest might be from crimsonal DNA.

0:22:10.710 --> 0:22:21.230  
Evanthia Anastasiadou  
You have to cut the sequence of your interest, usually with restriction enzymes or amplified with apcr. Different story.

0:22:21.310 --> 0:22:25.510  
Evanthia Anastasiadou  
So cut the sequence of your interest with your restriction enzymes.

0:22:26.290 --> 0:22:28.650  
Evanthia Anastasiadou  
You also cut and open your.

0:22:29.440 --> 0:22:38.400  
Evanthia Anastasiadou  
Vector and then you stitch with restriction enzymes and then you stitch together the two parts with the DNA ligase.

0:22:39.970 --> 0:22:41.170  
Evanthia Anastasiadou  
This one will have.

0:22:42.850 --> 0:22:42.890  
Evanthia Anastasiadou  
A.

0:22:42.890 --> 0:23:2.730  
Evanthia Anastasiadou  
Marketing a gene that will make sure that your bacteria will carry your plasmid and therefore the sequence of your interest. We said that more often you use anti biotics that you include in your growing culture in your medium and then.

0:23:3.260 --> 0:23:25.900  
Evanthia Anastasiadou  
Curious. Sure that all the bacteria that will multiply from there on, they will having the plasmid and the gene of your sequence when you grow and you have enough then you can extract it or use it further. For example in vitro experiment in cell cultures or an in.

0:23:25.900 --> 0:23:29.20  
Evanthia Anastasiadou  
Vivo experiment using animal models.

0:23:29.810 --> 0:23:33.50  
Evanthia Anastasiadou  
That can be from worms to faces.

0:23:34.270 --> 0:23:35.830  
Evanthia Anastasiadou  
Mamaliy such as mice.

0:23:40.710 --> 0:23:41.110  
Evanthia Anastasiadou  
OK.

0:23:42.790 --> 0:23:43.390  
Evanthia Anastasiadou  
So.

0:23:55.460 --> 0:23:55.820  
Evanthia Anastasiadou  
OK.

0:23:55.980 --> 0:24:4.900  
Evanthia Anastasiadou  
So very often the sequence of your interest will be a gene that you would like to find out what is doing.

0:24:6.410 --> 0:24:7.50  
Evanthia Anastasiadou  
In a certain setting.

0:24:8.650 --> 0:24:10.170  
Evanthia Anastasiadou  
To be able to check.

0:24:11.970 --> 0:24:19.490  
Evanthia Anastasiadou  
The expression of your gene, let's say that your gene here is this colored bars blue, yellow and pink.

0:24:19.690 --> 0:24:21.250  
Evanthia Anastasiadou  
So this is your gene.

0:24:22.90 --> 0:24:26.650  
Evanthia Anastasiadou  
The gene by itself will not be able to provide any information.

0:24:27.540 --> 0:24:34.420  
Evanthia Anastasiadou  
Unless there you have some other sequences before and after your chain.

0:24:36.770 --> 0:24:44.450  
Evanthia Anastasiadou  
The sequences before and after your gene will make sure that your gene will produce a protein.

0:24:44.690 --> 0:24:49.50  
Evanthia Anastasiadou  
Your gene will be transcribed and translated.

0:24:50.650 --> 0:24:57.410  
Evanthia Anastasiadou  
So because only by checking your protein, most likely you'll be able to see the effect.

0:24:57.500 --> 0:25:0.180  
Evanthia Anastasiadou  
Back of your gene of interest.

0:25:1.970 --> 0:25:9.490  
Evanthia Anastasiadou  
To the immune that you have to create a transcriptional control sequence.

0:25:10.670 --> 0:25:21.70  
Evanthia Anastasiadou  
And when you talk about a transcriptional control sequence, you are talking about a gene that can be transcribed that can provide mRNA.

0:25:21.670 --> 0:25:22.950  
Evanthia Anastasiadou  
How will you do that?

0:25:23.110 --> 0:25:26.270  
Evanthia Anastasiadou  
First of all, you'll have your gene, your coding.

0:25:28.650 --> 0:25:32.810  
Evanthia Anastasiadou  
You need to insert something in front and after.

0:25:34.410 --> 0:25:35.810  
Evanthia Anastasiadou  
Your coding sequence.

0:25:37.570 --> 0:25:42.450  
Evanthia Anastasiadou  
In front is what we called promoter element at the end.

0:25:43.90 --> 0:25:46.770  
Evanthia Anastasiadou  
Is what we call stop or poliating?

0:25:48.330 --> 0:25:49.530  
Evanthia Anastasiadou  
The promoter.

0:25:50.210 --> 0:26:7.90  
Evanthia Anastasiadou  
The promoter element is the site where your RNA polymerase. It's gonna bind had specific sequences like Tata Box, that is upstream, the starting of your gene. And this is from where?

0:26:8.770 --> 0:26:8.930  
Evanthia Anastasiadou  
Your.

0:26:10.850 --> 0:26:12.130  
Evanthia Anastasiadou  
Had to be.

0:26:14.740 --> 0:26:26.620  
Evanthia Anastasiadou  
Where your RNA polymerase is going to bind, but most often you have some other enhancer sequences that usually are located a few hundred base pairs.

0:26:28.170 --> 0:26:32.170  
Evanthia Anastasiadou  
Upstream that stimulates the expression of the genes.

0:26:34.920 --> 0:26:38.120  
Evanthia Anastasiadou  
Also might have transcription factor sequences.

0:26:38.400 --> 0:26:40.160  
Evanthia Anastasiadou  
These are sequences that.

0:26:41.730 --> 0:26:49.650  
Evanthia Anastasiadou  
Other factors might bind and these are the ones that will guide the RNA polymerase at the correct.

0:26:51.290 --> 0:27:8.690  
Evanthia Anastasiadou  
Place so we said, promoter elements, we are talking about where the promoter is going to bind enhancer sequences to enhance the transcription and also transcript transcription factor sequences that will guide.

0:27:9.140 --> 0:27:13.180  
Evanthia Anastasiadou  
The polymerase to the specific genes that needs to be transcribed.

0:27:16.760 --> 0:27:17.880  
Evanthia Anastasiadou  
Loading sequence.

0:27:21.730 --> 0:27:26.730  
Evanthia Anastasiadou  
Only 18 is at the end of your team and has many stops.

0:27:28.290 --> 0:27:32.410  
Evanthia Anastasiadou  
This is the signal where your transcription will finish.

0:27:33.340 --> 0:27:36.460  
Evanthia Anastasiadou  
So that means that this is the end of your gene.

0:27:37.20 --> 0:27:45.220  
Evanthia Anastasiadou  
That's where you have to finish. If you don't have that one, your transcription will continue and mess your results.

0:27:45.980 --> 0:27:49.820  
Evanthia Anastasiadou  
So so far we talked about cloning in general.

0:27:49.980 --> 0:27:55.980  
Evanthia Anastasiadou  
We talked about the key players in cloning and we talked about the various enzymes that are involved.

0:28:0.780 --> 0:28:2.220  
Evanthia Anastasiadou  
Do you have any questions so far?

0:28:4.770 --> 0:28:5.690  
Evanthia Anastasiadou  
Any questions?

0:28:6.190 --> 0:28:12.750  
nickcharisis182  
I'm guessing that the Poly region that you talked about is a region that consists of a, a, a basis, right?

0:28:13.910 --> 0:28:14.590  
Evanthia Anastasiadou  
Exactly.

0:28:14.290 --> 0:28:18.130  
nickcharisis182  
What? What's the point of having so many as? I mean, I'm guessing that.

0:28:17.990 --> 0:28:18.510  
Evanthia Anastasiadou  
OK.

0:28:19.900 --> 0:28:23.300  
nickcharisis182  
Having plan 2-3 dates for each to make it foolproof.

0:28:22.650 --> 0:28:28.450  
Evanthia Anastasiadou  
So does not have only, so does not have only as have some stops also.

0:28:30.10 --> 0:28:34.770  
Evanthia Anastasiadou  
You're gonna learn about them when you are gonna talk about the DNA structure.

0:28:34.970 --> 0:28:39.850  
Evanthia Anastasiadou  
So there are some codons that are making it to stop.

0:28:41.650 --> 0:28:51.770  
Evanthia Anastasiadou  
But as you're gonna learn again when you're gonna learn about mRNA, polya is an addition at the very end of your mRNA.

0:28:53.140 --> 0:28:54.300  
Evanthia Anastasiadou  
RNA is very fragile.

0:28:54.300 --> 0:28:56.540  
Evanthia Anastasiadou  
It's very no fragile.

0:28:56.980 --> 0:29:0.340  
Evanthia Anastasiadou  
It's very prone to degrading.

0:29:1.140 --> 0:29:12.820  
Evanthia Anastasiadou  
We have all these enzymes that we call the RNA CS that if you have an RNA fragment there will start degrading it OK.

0:29:12.900 --> 0:29:18.260  
Evanthia Anastasiadou  
So you need some protection. You need to protect your RNA from.

0:29:19.50 --> 0:29:21.290  
Evanthia Anastasiadou  
The RNA seats that will break it down.

0:29:21.610 --> 0:29:25.530  
Evanthia Anastasiadou  
One of the protection is to put additional sequences.

0:29:26.260 --> 0:29:27.460  
Evanthia Anastasiadou  
But they are not important.

0:29:27.460 --> 0:29:39.220  
Evanthia Anastasiadou  
They are not part of your gene, but they are there to protect the rest of the sequence. So the polyaids are there. Yep, go ahead.

0:29:39.440 --> 0:29:43.240  
nickcharisis182  
So the idea that we use that as armor, that's like kind of farmer that.

0:29:45.650 --> 0:29:47.730  
nickcharisis182  
Oh oh, you got the polymerase.

0:29:47.730 --> 0:29:54.930  
nickcharisis182  
How you call it so that they would this sequence would be degraded instead of the army that we actually do need.

0:29:54.930 --> 0:29:57.490  
nickcharisis182  
That's the idea, OK?

0:29:56.30 --> 0:29:58.150  
Evanthia Anastasiadou  
Exactly. So yeah, exactly.

0:29:58.150 --> 0:29:59.270  
Evanthia Anastasiadou  
It's for protection.

0:29:59.550 --> 0:30:6.110  
Evanthia Anastasiadou  
So at this end, at the polio here you have many stop codons that will stop.

0:30:7.650 --> 0:30:10.250  
Evanthia Anastasiadou  
Your the making of the protein later on.

0:30:10.770 --> 0:30:12.810  
Evanthia Anastasiadou  
But so they will give the signal.

0:30:12.810 --> 0:30:30.410  
Evanthia Anastasiadou  
That's where you should stop creating, but in the case before we are adding more protection so till it will reach the point to make the protein it will be protected from the enzymes that are gonna break it down.

0:30:31.760 --> 0:30:31.800  
Evanthia Anastasiadou  
K.

0:30:32.240 --> 0:30:33.160  
Evanthia Anastasiadou  
It's more or less.

0:30:33.200 --> 0:30:37.400  
Evanthia Anastasiadou  
I don't know if you heard about Telomers in.

0:30:39.10 --> 0:30:54.930  
Evanthia Anastasiadou  
Chromosomes. So in our so in our chromosomes we are talking now about DNA, not RNA in our chromosomes. At the very end we have some repetitive sequences that we call them telomeres.

0:30:40.540 --> 0:30:41.60  
nickcharisis182  
What happened?

0:30:56.770 --> 0:31:4.410  
Evanthia Anastasiadou  
These telomeres, when we are starting our life, this repetitions are many and long. But as we go along.

0:31:5.430 --> 0:31:8.70  
Evanthia Anastasiadou  
They are destroyed.

0:31:9.610 --> 0:31:15.610  
Evanthia Anastasiadou  
With every division as you are growing and yourselves are multiplying and more.

0:31:17.250 --> 0:31:21.650  
Evanthia Anastasiadou  
Mistakes are gonna be included in your DNA and in your genome.

0:31:21.890 --> 0:31:26.970  
Evanthia Anastasiadou  
So the ends will start getting shorter and shorter.

0:31:27.130 --> 0:31:33.890  
Evanthia Anastasiadou  
The repetitions will start being less and less and that's why when you get older your DNA.

0:31:35.100 --> 0:31:40.940  
Evanthia Anastasiadou  
Your DNA are not protected anymore and that's why you accumulate more diseases.

0:31:40.940 --> 0:31:54.140  
Evanthia Anastasiadou  
You are more prone to diseases than when you are earlier because your DNA now is more accessible to outside factors that can cause problems.

0:31:55.890 --> 0:31:56.210  
Evanthia Anastasiadou  
OK.

0:31:56.290 --> 0:32:8.250  
Evanthia Anastasiadou  
So it's a common mechanism in the cell both for RNA and DNA, but at the very end you have some repetitions of sequences that you don't really need them, but they are just there to protect you.

0:32:9.590 --> 0:32:9.630  
nickcharisis182  
K.

0:32:9.710 --> 0:32:14.230  
nickcharisis182  
So this makes the other question why only in the end?

0:32:14.270 --> 0:32:15.910  
nickcharisis182  
Why not have them at both sides?

0:32:16.310 --> 0:32:17.590  
nickcharisis182  
I mean, you can't be too safe.

0:32:17.710 --> 0:32:23.550  
Evanthia Anastasiadou  
Ah, OK, you might have them both sides for chromosomes.

0:32:23.830 --> 0:32:31.590  
Evanthia Anastasiadou  
The telomeres are in both sides. For here we have other protected mechanism to cover this side.

0:32:32.350 --> 0:32:35.750  
nickcharisis182  
We do have protection, but it's not a fully secured, it's something else.

0:32:33.90 --> 0:32:33.130  
Evanthia Anastasiadou  
OK.

0:32:36.370 --> 0:32:36.850  
Evanthia Anastasiadou  
OK.

0:32:37.410 --> 0:32:44.730  
Evanthia Anastasiadou  
So, but it's a different mechanism and I don't want to start, we call it cap.

0:32:41.50 --> 0:32:41.370  
nickcharisis182  
Thank you.

0:32:45.530 --> 0:32:46.370  
nickcharisis182  
Yeah. Thank you.

0:32:45.650 --> 0:32:49.970  
Evanthia Anastasiadou  
So there is a cap that you are going to put on this side also, OK.

0:32:50.570 --> 0:32:51.10  
nickcharisis182  
Thank you.

0:32:51.530 --> 0:32:53.770  
Evanthia Anastasiadou  
So you do have for both sides.

0:32:56.20 --> 0:32:57.340  
Evanthia Anastasiadou  
So do you have any other?

0:32:57.460 --> 0:32:59.340  
Evanthia Anastasiadou  
Any other questions my students?

0:33:2.770 --> 0:33:3.850  
Evanthia Anastasiadou  
Any other questions?

0:33:6.600 --> 0:33:20.240  
Evanthia Anastasiadou  
OK. Can we have like a 10 minutes break now? And when you'll come back, I will continue with a different types of transgenic animals that we are creating.

0:33:22.470 --> 0:33:23.270  
Evanthia Anastasiadou  
Is that OK?

0:33:24.920 --> 0:33:25.560  
Despoina Voulgari  
Yeah. Thanks.

0:33:27.430 --> 0:33:29.430  
Evanthia Anastasiadou  
Minutes not more than 10 minutes.

0:33:29.710 --> 0:33:31.910  
Evanthia Anastasiadou  
Just go and refresh yourself.

0:33:32.630 --> 0:33:36.470  
Evanthia Anastasiadou  
Coffee, restroom and back in 10 minutes, OK.

0:33:39.680 --> 0:33:40.280  
Evanthia Anastasiadou  
You then?

0:48:58.530 --> 0:48:59.290  
Evanthia Anastasiadou  
OK. Do you?

0:48:59.290 --> 0:49:0.650  
Evanthia Anastasiadou  
Are you back?

0:49:1.90 --> 0:49:2.210  
Evanthia Anastasiadou  
Is everyone here?

0:49:2.450 --> 0:49:3.490  
Evanthia Anastasiadou  
Shall we continue?

0:49:10.550 --> 0:49:13.590  
Evanthia Anastasiadou  
Apart from your ghost, anyone else here with me?

0:49:14.220 --> 0:49:14.700  
Ξανθίππη Λούκα  
Yes.

0:49:15.210 --> 0:49:15.690  
nickcharisis182  
Yeah, yeah.

0:49:17.120 --> 0:49:17.480  
Evanthia Anastasiadou  
OK.

0:49:17.480 --> 0:49:18.240  
Evanthia Anastasiadou  
Thank you.

0:49:18.800 --> 0:49:20.520  
Evanthia Anastasiadou  
So you haven't fallen asleep as yet.

0:49:24.380 --> 0:49:24.700  
Evanthia Anastasiadou  
OK.

0:49:25.460 --> 0:49:30.900  
Evanthia Anastasiadou  
So what about my biologists? Are you bored to death? Anything.

0:49:30.900 --> 0:49:32.140  
Evanthia Anastasiadou  
You are learning more.

0:49:32.380 --> 0:49:33.100  
Evanthia Anastasiadou  
Nothing at all.

0:49:33.100 --> 0:49:34.180  
Evanthia Anastasiadou  
Nothing new for you.

0:49:41.680 --> 0:49:46.320  
Evanthia Anastasiadou  
Let's see if you are gonna learn anything new in the next few slides.

0:49:47.850 --> 0:49:47.930  
Evanthia Anastasiadou  
So.

0:49:49.490 --> 0:49:52.490  
Evanthia Anastasiadou  
Why do we really want to manipulate DNA?

0:49:54.730 --> 0:50:7.290  
Evanthia Anastasiadou  
We were saying that you might have some sequences of interest, might be genes, or might be regulatory elements, might be tasked, might be whatever, and you want to.

0:50:8.970 --> 0:50:9.970  
Evanthia Anastasiadou  
Investigate them further.

0:50:12.600 --> 0:50:15.0  
Evanthia Anastasiadou  
So we said, how will you investigate them?

0:50:16.570 --> 0:50:22.570  
Evanthia Anastasiadou  
You're you're gonna start most likely by using them in prokaryotics like bacteria.

0:50:24.370 --> 0:50:26.450  
Evanthia Anastasiadou  
And you'll see if they have any effects.

0:50:28.50 --> 0:50:31.770  
Evanthia Anastasiadou  
Eventually you want to move to an animal model.

0:50:33.450 --> 0:50:43.450  
Evanthia Anastasiadou  
And the people the biologists already know that according to what you want to study, you might choose a different type of model.

0:50:44.450 --> 0:50:48.970  
Evanthia Anastasiadou  
For example, aging and longevity, you are gonna.

0:50:50.570 --> 0:50:54.610  
Evanthia Anastasiadou  
Use C elegans or cardiovascular diseases.

0:50:56.330 --> 0:51:2.970  
Evanthia Anastasiadou  
And the developing of a heart and the vessels etcetera, you might use zebrafish.

0:51:5.330 --> 0:51:6.890  
Evanthia Anastasiadou  
Now for most.

0:51:8.850 --> 0:51:9.650  
Evanthia Anastasiadou  
For most.

0:51:11.490 --> 0:51:13.690  
Evanthia Anastasiadou  
Diseases. Drugs. Research.

0:51:13.690 --> 0:51:15.850  
Evanthia Anastasiadou  
You will eventually use mice.

0:51:22.600 --> 0:51:25.600  
Evanthia Anastasiadou  
It's not a rhetorical question. I'm waiting for your answer.

0:51:27.860 --> 0:51:29.300  
nickcharisis182  
Could you repeat it? You cut off.

0:51:31.90 --> 0:51:31.370  
Evanthia Anastasiadou  
OK.

0:51:31.370 --> 0:51:32.570  
Evanthia Anastasiadou  
I'm sorry about that.

0:51:32.690 --> 0:51:36.690  
Evanthia Anastasiadou  
Let me close my let me close.

0:51:40.890 --> 0:51:41.930  
Evanthia Anastasiadou  
Let me close.

0:51:43.690 --> 0:51:44.450  
Evanthia Anastasiadou  
The video.

0:51:46.660 --> 0:51:47.100  
Evanthia Anastasiadou  
OK.

0:51:55.670 --> 0:51:57.150  
Evanthia Anastasiadou  
You let me close that one.

0:51:58.690 --> 0:51:59.450  
Evanthia Anastasiadou  
Close the camera.

0:52:2.230 --> 0:52:4.30  
Evanthia Anastasiadou  
Are there a lot of breaks?

0:52:7.100 --> 0:52:8.20  
nickcharisis182  
Right now, no.

0:52:8.380 --> 0:52:10.180  
nickcharisis182  
But they're kind of infrequent.

0:52:11.520 --> 0:52:13.0  
Despoina Voulgari  
Yeah, I think every now and then.

0:52:11.810 --> 0:52:12.170  
Evanthia Anastasiadou  
OK.

0:52:14.920 --> 0:52:19.600  
Evanthia Anastasiadou  
OK, but it's not creating like a huge problem for you, is it?

0:52:20.370 --> 0:52:20.730  
Despoina Voulgari  
No, no.

0:52:20.470 --> 0:52:23.510  
nickcharisis182  
No, I think that now you're you're slightly better.

0:52:24.880 --> 0:52:26.200  
Evanthia Anastasiadou  
OK, OK.

0:52:26.200 --> 0:52:27.40  
Evanthia Anastasiadou  
So let's see.

0:52:27.200 --> 0:52:35.520  
Evanthia Anastasiadou  
So I was asking you, I I was telling you that always our research starts with procario.

0:52:35.520 --> 0:52:51.960  
Evanthia Anastasiadou  
It's like bacteria, but eventually we are moving to animal models and according to what you want to study, you are going to use different animal models. I told you that for aging and longevity you are going to use a worm called C elegans.

0:52:52.770 --> 0:52:53.210  
Evanthia Anastasiadou  
For.

0:52:55.10 --> 0:52:56.90  
Evanthia Anastasiadou  
Cardiovascular. Cardiovascular.

0:52:56.620 --> 0:52:58.140  
Evanthia Anastasiadou  
Development and disease.

0:52:58.420 --> 0:52:59.420  
Evanthia Anastasiadou  
Most likely you are.

0:52:59.420 --> 0:53:1.740  
Evanthia Anastasiadou  
You gonna use a small face.

0:53:2.60 --> 0:53:3.460  
Evanthia Anastasiadou  
That is quite.

0:53:6.930 --> 0:53:9.90  
Evanthia Anastasiadou  
Small and you can culture it in your.

0:53:11.170 --> 0:53:22.930  
Evanthia Anastasiadou  
In the lab and you can see through it, we call it zebrafish, but in one way or the other, in the end of the day, most likely when you're talking about.

0:53:25.10 --> 0:53:26.210  
Evanthia Anastasiadou  
Human genes.

0:53:27.380 --> 0:53:36.620  
Evanthia Anastasiadou  
Disease, human drugs and treatments. You are gonna use as an intermediate amount a mouse model.

0:53:36.980 --> 0:53:43.140  
Evanthia Anastasiadou  
Why do you think we are using mice as models for human diseases and for?

0:53:44.730 --> 0:53:45.410  
Evanthia Anastasiadou  
Drug treatments.

0:53:51.470 --> 0:53:57.350  
nickcharisis182  
No, I'm guessing, but I think it's a good compromise between availability complexity.

0:53:59.10 --> 0:54:1.370  
nickcharisis182  
Expectability I don't know.

0:54:1.410 --> 0:54:5.530  
nickcharisis182  
It's it's not that bad shape.

0:54:3.800 --> 0:54:4.0  
Evanthia Anastasiadou  
Mm hmm.

0:54:5.770 --> 0:54:6.490  
nickcharisis182  
Very politely.

0:54:8.210 --> 0:54:8.770  
nickcharisis182  
It's not that.

0:54:10.370 --> 0:54:14.130  
nickcharisis182  
Offensive test on mice. But testing on monkeys or humans?

0:54:14.290 --> 0:54:15.730  
nickcharisis182  
It's kind of, you know.

0:54:18.710 --> 0:54:21.710  
nickcharisis182  
Payback for the color and the medieval ladies? Something like that.

0:54:23.820 --> 0:54:25.460  
Evanthia Anastasiadou  
What about my biologist? Come on.

0:54:24.730 --> 0:54:30.650  
nickcharisis182  
But my my first part of the question is what I do believe it's a good compromise?

0:54:31.970 --> 0:54:33.50  
Evanthia Anastasiadou  
Good, good, excellent.

0:54:32.170 --> 0:54:32.210  
nickcharisis182  
And.

0:54:33.50 --> 0:54:34.210  
Evanthia Anastasiadou  
That's a very good answer.

0:54:35.170 --> 0:54:36.290  
Evanthia Anastasiadou  
I want a biologist.

0:54:45.350 --> 0:54:45.750  
Evanthia Anastasiadou  
On them.

0:54:45.750 --> 0:54:46.630  
Evanthia Anastasiadou  
Be shy.

0:54:47.310 --> 0:54:49.710  
Vasilis Chatzitolios  
Can I say I'm a biologist, but I can guess.

0:54:51.60 --> 0:55:0.220  
Vasilis Chatzitolios  
Well, maybe it's because they they are mammalians, so probably they're structuring their systems more similar to.

0:54:51.540 --> 0:54:52.300  
Evanthia Anastasiadou  
Oh yes.

0:55:1.770 --> 0:55:9.50  
Vasilis Chatzitolios  
The human ones than the fish that you said the worms and also.

0:55:10.300 --> 0:55:15.100  
Vasilis Chatzitolios  
So the really small first psyche so.

0:55:16.690 --> 0:55:23.10  
Vasilis Chatzitolios  
It's really easy to watch them grow and die and give birth grow.

0:55:23.650 --> 0:55:25.490  
Vasilis Chatzitolios  
It takes weeks maybe.

0:55:25.490 --> 0:55:28.810  
Vasilis Chatzitolios  
I don't know, but yeah, I think it's like this.

0:55:28.380 --> 0:55:30.500  
Evanthia Anastasiadou  
Excellent, excellent, excellent.

0:55:32.50 --> 0:55:35.250  
Evanthia Anastasiadou  
Biologists, where are you?

0:55:42.340 --> 0:55:42.900  
Evanthia Anastasiadou  
Where?

0:55:42.440 --> 0:55:47.360  
Ξανθίππη Λούκα  
I I was going to say the same because it's in Amalia and it's more close to humans.

0:55:49.100 --> 0:55:50.380  
Evanthia Anastasiadou  
But OK.

0:55:50.380 --> 0:56:2.100  
Evanthia Anastasiadou  
So basically it's because they are closer to humans. You will be surprised to know that more 95% of our sequences are.

0:56:3.690 --> 0:56:13.810  
Evanthia Anastasiadou  
Same or very similar with mice. Most of our molecular functions are the same. Our organ systems are very similar and.

0:56:15.450 --> 0:56:17.170  
Evanthia Anastasiadou  
Apart from the DNA gene.

0:56:18.950 --> 0:56:31.310  
Evanthia Anastasiadou  
Is your logical and anatomical similarities. Also, they are small, so they can grow in smaller places in cages that we have in animal houses.

0:56:32.850 --> 0:56:34.370  
Evanthia Anastasiadou  
They multiply fast.

0:56:36.10 --> 0:56:39.770  
Evanthia Anastasiadou  
It's a mouse can deliver like 8.

0:56:41.890 --> 0:56:45.370  
Evanthia Anastasiadou  
10 new mice litters offsprings as we call them.

0:56:45.610 --> 0:56:50.410  
Evanthia Anastasiadou  
So the you can produce a lot of them. They have a quick cycle.

0:56:51.280 --> 0:56:53.640  
Evanthia Anastasiadou  
So they grow.

0:56:54.0 --> 0:56:55.400  
Evanthia Anastasiadou  
I mean, imagine that.

0:56:55.400 --> 0:57:1.880  
Evanthia Anastasiadou  
Their maximum life is 2-3 years, so by year by month by the 1st.

0:57:3.970 --> 0:57:4.370  
Evanthia Anastasiadou  
Month.

0:57:6.130 --> 0:57:19.250  
Evanthia Anastasiadou  
They are already start breeding so in two months time they are grown-ups and ready to start breeding so you can see that you can produce many mice in a small space relatively cheap.

0:57:19.930 --> 0:57:23.130  
Evanthia Anastasiadou  
They are resembling to humans and actually.

0:57:23.810 --> 0:57:28.370  
Evanthia Anastasiadou  
They through because you are doing inbreeding.

0:57:29.50 --> 0:57:32.610  
Evanthia Anastasiadou  
Most of them are almost identical.

0:57:32.890 --> 0:57:35.170  
Evanthia Anastasiadou  
So you have a good control sample.

0:57:37.110 --> 0:58:1.90  
Evanthia Anastasiadou  
So we through because they are such a good model, we developed quite a few different ways of using them and inserting in there all sort of different sequences and we have various ways of developing mouse models. The first one that I'm going to talk about is the trans.

0:58:1.550 --> 0:58:2.510  
Evanthia Anastasiadou  
Mouse model.

0:58:3.450 --> 0:58:3.770  
Evanthia Anastasiadou  
And.

0:58:5.370 --> 0:58:7.970  
Evanthia Anastasiadou  
Main well in the literature you might find.

0:58:8.550 --> 0:58:30.470  
Evanthia Anastasiadou  
Transgenic the term transgenic used a bit differently. So from where I come, and actually it's the lab where the transgenic mice started, and our boss Martin Evans got the Nobel Prize for that when we called. When we say transgenic mice.

0:58:32.10 --> 0:58:36.170  
Evanthia Anastasiadou  
We mean these sort of mice, let's say in more detail.

0:58:36.170 --> 0:58:44.250  
Evanthia Anastasiadou  
So remember before that I was telling you that when this is a transcriptional unit, so you have your gene.

0:58:44.740 --> 0:58:59.580  
Evanthia Anastasiadou  
Recording sequence. You have your regulatory elements upstream, your promoter that will regulate when and how much your gene is gonna be made. And at the end you have the stop sequences to.

0:59:2.550 --> 0:59:7.470  
Evanthia Anastasiadou  
To determine where your gene is ending and Poly a sequence is for protection.

0:59:8.30 --> 0:59:15.350  
Evanthia Anastasiadou  
So this is your transcription unit and this is that is part of your plasmid.

0:59:16.890 --> 0:59:23.10  
Evanthia Anastasiadou  
The one that you created and the one that you are going to insert in your mice.

0:59:24.920 --> 0:59:25.760  
Evanthia Anastasiadou  
Sharing the mice.

0:59:27.570 --> 0:59:30.690  
Evanthia Anastasiadou  
So you have a mouse that you.

0:59:33.930 --> 0:59:38.10  
Evanthia Anastasiadou  
That you get the fertilized eggs.

0:59:39.690 --> 0:59:46.250  
Evanthia Anastasiadou  
So you have fertilized eggs and you know when you're talking about eggs, these are your ova.

0:59:48.280 --> 0:59:51.640  
Evanthia Anastasiadou  
You insert your DNA in there.

0:59:53.330 --> 0:59:57.730  
Evanthia Anastasiadou  
As well as in the rest of your in their DNA.

0:59:57.890 --> 1:0:17.130  
Evanthia Anastasiadou  
So you insert the gene of your interest in the nucleus in their fertilized eggs, and what you would expect is that the gene of your interest will get incorporated in the genome of the mouse.

1:0:19.350 --> 1:0:20.150  
Evanthia Anastasiadou  
It will start.

1:0:21.690 --> 1:0:29.130  
Evanthia Anastasiadou  
Multiplying, transcribing, translating, and you're gonna see the effects in the whole animal.

1:0:31.860 --> 1:0:51.740  
Evanthia Anastasiadou  
So once again you created the chain of your interest. You have all the regulatory elements before and after your dream, so make sure that it will be produced, and then you insert it in the DNA in your mice.

1:0:53.290 --> 1:0:53.330  
Evanthia Anastasiadou  
So.

1:0:54.970 --> 1:1:1.90  
Evanthia Anastasiadou  
When it will be inserted then it will be multiplied and eventually.

1:1:1.690 --> 1:1:9.210  
Evanthia Anastasiadou  
Some of the cells will derive from this one and will have the characteristics of the gene that you want to study.

1:1:11.240 --> 1:1:21.200  
Evanthia Anastasiadou  
So once again, it's the gene of your interest that was gonna be transcribed in mRNA and translated into proteins.

1:1:21.640 --> 1:1:23.280  
Evanthia Anastasiadou  
The effect of the proteins.

1:1:23.280 --> 1:1:28.920  
Evanthia Anastasiadou  
You're gonna observe them in the cell of the mouse that you will develop.

1:1:31.590 --> 1:1:33.670  
Evanthia Anastasiadou  
OK, this is a straightforward.

1:1:35.210 --> 1:1:35.770  
Evanthia Anastasiadou  
Way.

1:1:37.490 --> 1:1:40.290  
Evanthia Anastasiadou  
But has quite a few disadvantages.

1:1:40.650 --> 1:1:45.690  
Evanthia Anastasiadou  
So advantages, it's relatively quick and easy to clone.

1:1:47.370 --> 1:1:47.930  
Evanthia Anastasiadou  
Disadvantages.

1:1:49.570 --> 1:1:50.650  
Evanthia Anastasiadou  
It's not well controlled.

1:1:51.210 --> 1:1:53.970  
Evanthia Anastasiadou  
You might have a false observations.

1:1:55.610 --> 1:2:2.130  
Evanthia Anastasiadou  
Why? Because your Gen. of interest, your sequence of interest, will runly.

1:2:2.690 --> 1:2:14.650  
Evanthia Anastasiadou  
Be inserted in the DNA of your animal so you cannot control where exactly it's gonna be in inserted. It's gonna be spontaneous.

1:2:16.210 --> 1:2:22.530  
Evanthia Anastasiadou  
Random sites and we have some hot spots where you usually have a lot of insertions.

1:2:22.650 --> 1:2:27.330  
Evanthia Anastasiadou  
We have other ones that are more or less unaccessible.

1:2:29.90 --> 1:2:32.570  
Evanthia Anastasiadou  
And not only you have random insertions.

1:2:34.750 --> 1:2:44.270  
Evanthia Anastasiadou  
But so you cannot control the position of integration, but also you don't know how many of your sequences are gonna get inserted in there.

1:2:44.950 --> 1:2:45.590  
Evanthia Anastasiadou  
They might be.

1:2:45.590 --> 1:2:46.870  
Evanthia Anastasiadou  
One they might be.

1:2:46.870 --> 1:2:48.510  
Evanthia Anastasiadou  
Two, they might be 10.

1:2:48.990 --> 1:2:50.550  
Evanthia Anastasiadou  
That means that.

1:2:52.90 --> 1:3:4.650  
Evanthia Anastasiadou  
The effect that you see it's not because of your gene, it's because you inserted multiple copies of this gene, or even because you are disturbing a sequence.

1:3:6.150 --> 1:3:7.390  
Evanthia Anastasiadou  
In the mouse genome.

1:3:7.910 --> 1:3:25.670  
Evanthia Anastasiadou  
So inside of adding instead of having one characteristic that you're studying in your cells, because of the gene that you are inserting you it might be because of the genes that you are disturbing by this insertion.

1:3:26.630 --> 1:3:28.390  
Evanthia Anastasiadou  
Do you understand what I'm trying to say?

1:3:35.940 --> 1:3:42.580  
Evanthia Anastasiadou  
Because you are having random insertions in the genome, you don't know where these insertions are gonna be.

1:3:44.130 --> 1:3:48.170  
Evanthia Anastasiadou  
They might be disturbing a gene that already exists there.

1:3:48.970 --> 1:3:58.290  
Evanthia Anastasiadou  
So instead of saying the effect of your gene that you are putting in there, you see the effect of the gene that you have destroyed.

1:3:59.410 --> 1:4:5.490  
Evanthia Anastasiadou  
So you have to be really careful with the outcome of your transgenic mice.

1:4:9.210 --> 1:4:20.210  
Evanthia Anastasiadou  
Another, more elegant way to develop mice, genetically modified, is what we call knocking and knockout.

1:4:21.250 --> 1:4:29.250  
Evanthia Anastasiadou  
So no, keen is when you are putting a certain sequence at the specific point in the mouse genome.

1:4:30.810 --> 1:4:37.770  
Evanthia Anastasiadou  
No cow is when you are taking out a specific sequence of the mouse genome.

1:4:38.730 --> 1:4:54.570  
Evanthia Anastasiadou  
This is much more elegant because you know exactly what is the locus, the DNA locus that you are affecting so you don't have random disruptions, random integrations.

1:4:54.770 --> 1:5:3.570  
Evanthia Anastasiadou  
So it's a very well organized at a very specific site that you are either inserting or.

1:5:4.370 --> 1:5:11.130  
Evanthia Anastasiadou  
Deleting sequences and this is happening through homologous recombination.

1:5:12.120 --> 1:5:16.560  
Evanthia Anastasiadou  
So you are using big arms of DNA that are the same.

1:5:18.640 --> 1:5:30.160  
Evanthia Anastasiadou  
And you are trying to change whatever lies in between, either by inserting your sequences or getting rid of sequences that are already there.

1:5:30.880 --> 1:5:35.80  
Evanthia Anastasiadou  
So we call that knocking when you are inserting a new sequence.

1:5:35.480 --> 1:5:39.960  
Evanthia Anastasiadou  
No code when you are deleting a sequence that already exists.

1:5:41.280 --> 1:5:45.520  
Evanthia Anastasiadou  
And this is happening through homologous recombination.

1:5:49.370 --> 1:6:6.490  
Evanthia Anastasiadou  
So talking about knockings and knockouts here, you cannot just inject your DNA because by by injecting, your DNA would randomly integrate. So in this case you are not starting.

1:6:8.40 --> 1:6:18.320  
Evanthia Anastasiadou  
You are starting with some cells that we call them embryonic stem cells. Embryonic stem cells are the cells that are gonna give rise.

1:6:18.920 --> 1:6:25.800  
Evanthia Anastasiadou  
To all the different cell population in an Organism so embryonic stem cells.

1:6:27.390 --> 1:6:29.470  
Evanthia Anastasiadou  
Are the ones that you are gonna use.

1:6:31.40 --> 1:6:49.520  
Evanthia Anastasiadou  
These are the ones that you are gonna manipulate through homologous recombination and their genomic material. The material of these cells is the one that you are gonna manipulate and create a knocking or a knockout.

1:6:51.240 --> 1:6:57.960  
Evanthia Anastasiadou  
Now, after manipulating these cells then you are gonna grow them.

1:6:59.420 --> 1:7:9.460  
Evanthia Anastasiadou  
You are gonna insect them in the blastocyst when we are talking about the blastocyst is when you are having.

1:7:11.40 --> 1:7:29.960  
Evanthia Anastasiadou  
Fertilized egg that will start multiplying then after some divisions you have the formation of the blastasys you are gonna inject your is ESL synth there and these are gonna give rise to a population that will be all identical.

1:7:30.600 --> 1:7:36.80  
Evanthia Anastasiadou  
And all well organized with the genomic manipulation that you had already.

1:7:37.640 --> 1:7:38.160  
Evanthia Anastasiadou  
Performed.

1:7:40.820 --> 1:7:43.900  
Evanthia Anastasiadou  
So talking about embryonic stem cells.

1:7:46.720 --> 1:7:49.600  
Evanthia Anastasiadou  
Are so how do we produce them?

1:7:49.760 --> 1:8:0.440  
Evanthia Anastasiadou  
So you, as I said, you take an ova and a sperm an OVA and the sperm gets together to make a fertilized.

1:8:2.880 --> 1:8:9.920  
Evanthia Anastasiadou  
Egg this will start multiplying and as it multiplies from 1 cell you have many cells.

1:8:11.640 --> 1:8:18.400  
Evanthia Anastasiadou  
At day five to six, after the egg is fertilized, you have the formation of the blastocyst.

1:8:19.460 --> 1:8:24.580  
Evanthia Anastasiadou  
The blastocysts look like a cyst. That inside has an inner mass.

1:8:24.780 --> 1:8:27.980  
Evanthia Anastasiadou  
This inner mass is the one, as you can see here.

1:8:29.520 --> 1:8:40.960  
Evanthia Anastasiadou  
It's this inner mass that are the cells that you can connect, cultivate, and it's the one that they are going to give you the embryonic stem cells.

1:8:42.0 --> 1:8:47.40  
Evanthia Anastasiadou  
I have a video here if you want to see how we are sucking the cells.

1:8:49.120 --> 1:8:52.320  
Evanthia Anastasiadou  
So why are we working with embryonic stem cells?

1:8:52.680 --> 1:8:55.880  
Evanthia Anastasiadou  
Embryonic stem cells are primarily.

1:8:58.880 --> 1:8:59.520  
Evanthia Anastasiadou  
Sorry.

1:9:4.730 --> 1:9:5.690  
Evanthia Anastasiadou  
Our primary.

1:9:6.10 --> 1:9:9.730  
Evanthia Anastasiadou  
Our primary and differentiated cells.

1:9:11.280 --> 1:9:19.520  
Evanthia Anastasiadou  
So our cells with no specific tissue characteristics which have the ability under specific.

1:9:21.280 --> 1:9:22.80  
Evanthia Anastasiadou  
Conditions.

1:9:24.500 --> 1:9:29.460  
Evanthia Anastasiadou  
To differentiate into the different types of cells in your body.

1:9:32.200 --> 1:9:40.960  
Evanthia Anastasiadou  
So basically you have a stem cell and under specific conditions it can become any other cell in your body.

1:9:43.90 --> 1:9:51.650  
Evanthia Anastasiadou  
The main characteristics of an embryonic stem cells, the main properties of an embryonic stem cells, is first of all, that it can multiply.

1:9:51.810 --> 1:10:8.250  
Evanthia Anastasiadou  
It can proliferate and create similar and differentiated cells, but also under certain conditions, can produce any other cell type in your body like neurosense.

1:10:10.850 --> 1:10:12.490  
Evanthia Anastasiadou  
So we say that.

1:10:14.40 --> 1:10:15.240  
Evanthia Anastasiadou  
They are totally potent.

1:10:17.40 --> 1:10:17.920  
Evanthia Anastasiadou  
Or omnipotent.

1:10:19.600 --> 1:10:39.0  
Evanthia Anastasiadou  
So the potent stem cells can give rise to any of the 220 different cell types found in an embryo, as well as extra embryonic cells like your placenta, so 30 total.

1:10:40.840 --> 1:10:41.400  
Evanthia Anastasiadou  
All cells.

1:10:42.320 --> 1:10:45.960  
Evanthia Anastasiadou  
So this embryonic stem cell can give.

1:10:47.520 --> 1:10:50.480  
Evanthia Anastasiadou  
Generation to any other cell, any other cell.

1:10:50.480 --> 1:10:51.280  
Evanthia Anastasiadou  
Type in your body.

1:10:57.110 --> 1:10:57.550  
Evanthia Anastasiadou  
OK.

1:11:0.210 --> 1:11:8.570  
Evanthia Anastasiadou  
So as I was talking earlier about lockings and knockouts, So what is happening is that as I said, you manipulate.

1:11:12.450 --> 1:11:13.210  
Evanthia Anastasiadou  
Give me a second.

1:11:17.740 --> 1:11:20.140  
Evanthia Anastasiadou  
You manipulate yourselves.

1:11:23.670 --> 1:11:28.590  
Evanthia Anastasiadou  
In the cell state, embryonic stem cell states.

1:11:30.320 --> 1:11:35.40  
Evanthia Anastasiadou  
You select for the ones that are already. You inserted the gene of your interest.

1:11:35.400 --> 1:11:42.640  
Evanthia Anastasiadou  
You take this this ESL and you put them in the blastocyst of a pregnant mouse.

1:11:44.460 --> 1:11:51.300  
Evanthia Anastasiadou  
So this one is gonna grow and what is gonna happen is.

1:11:52.840 --> 1:11:55.960  
Evanthia Anastasiadou  
At the first stage you're gonna get what we call Chimera.

1:11:57.640 --> 1:11:57.720  
Evanthia Anastasiadou  
So.

1:12:1.370 --> 1:12:2.490  
Evanthia Anastasiadou  
What is happening here?

1:12:2.490 --> 1:12:11.90  
Evanthia Anastasiadou  
You can see the blastas and you are inserting your dreams of your you're inserting the ESL with a gene of your interest.

1:12:12.640 --> 1:12:27.600  
Evanthia Anastasiadou  
Usually to be able to identify if our experiment worked or not, we are using cells with from different background that are usually linked with the color of the mice for example.

1:12:29.280 --> 1:12:31.920  
Evanthia Anastasiadou  
You are using the blastocyst of the brown mouse.

1:12:33.290 --> 1:12:37.290  
Evanthia Anastasiadou  
And you're inserting es cells of black mice.

1:12:39.160 --> 1:12:50.600  
Evanthia Anastasiadou  
What is gonna happen? You are gonna have a mixed population of cells with brown background and cells with black background.

1:12:52.280 --> 1:12:53.80  
Evanthia Anastasiadou  
These are es cells.

1:12:53.480 --> 1:13:1.360  
Evanthia Anastasiadou  
And remember that I told you that the ES LS are giving rise to all different types of cells.

1:13:1.920 --> 1:13:5.120  
Evanthia Anastasiadou  
So when you're gonna get the mouse, eventually.

1:13:6.120 --> 1:13:9.120  
Evanthia Anastasiadou  
Some parts of the mouse will be black.

1:13:9.760 --> 1:13:14.560  
Evanthia Anastasiadou  
That means derive from the cells that you manipulated.

1:13:16.120 --> 1:13:16.800  
Evanthia Anastasiadou  
Or will be brown.

1:13:17.280 --> 1:13:21.960  
Evanthia Anastasiadou  
This means the original unmanipulated cell.

1:13:25.910 --> 1:13:35.830  
Evanthia Anastasiadou  
Having to extend of the cells that you manipulated and the unmanipulated cells, we call them chimeras.

1:13:37.600 --> 1:13:41.560  
Evanthia Anastasiadou  
So this is the first step of knocking knockout.

1:13:42.360 --> 1:13:55.280  
Evanthia Anastasiadou  
Then you put your kindness together and after a couple of breathings, you have pure populations that can be either black or brown.

1:13:56.840 --> 1:14:2.360  
Evanthia Anastasiadou  
And that's when you can start investigating in a very nice and elegant way.

1:14:2.440 --> 1:14:8.440  
Evanthia Anastasiadou  
The effect of your gene advantages and disadvantages of knockings and knockouts.

1:14:8.960 --> 1:14:10.640  
Evanthia Anastasiadou  
It's a more elegant design.

1:14:11.200 --> 1:14:17.320  
Evanthia Anastasiadou  
It's well control and tone DEA and you have endogenous regulatory elements.

1:14:18.780 --> 1:14:19.740  
Evanthia Anastasiadou  
Disadvantages.

1:14:22.690 --> 1:14:26.330  
Evanthia Anastasiadou  
Even for the cloning, it takes you like months and months.

1:14:27.880 --> 1:14:32.360  
Evanthia Anastasiadou  
Time consuming. You need many more mice and many more cycles of breeding.

1:14:33.200 --> 1:14:45.840  
Evanthia Anastasiadou  
And you always might face embryonic lethality. That means that the gene that you are putting in there is killing your embryos, and you will never manage to get an A live mouse.

1:14:48.100 --> 1:14:54.740  
Evanthia Anastasiadou  
Now you can use conditional expression.

1:14:55.100 --> 1:15:11.540  
Evanthia Anastasiadou  
So we are putting one more element in there, so be able to regulate when your team will be on or when your team will be off for that purpose. Initially we use site specific recombination systems.

1:15:13.80 --> 1:15:18.120  
Evanthia Anastasiadou  
That we got such a A systems are flipped.

1:15:19.370 --> 1:15:20.690  
Evanthia Anastasiadou  
And Crane looks big.

1:15:21.50 --> 1:15:29.850  
Evanthia Anastasiadou  
The Flipper Party originates from Sakharam. Isis cerebral spy is from P1 bacterial phase.

1:15:32.690 --> 1:15:38.810  
Evanthia Anastasiadou  
With these systems you can control better the expression of your genes.

1:15:39.890 --> 1:15:52.250  
Evanthia Anastasiadou  
Since then, we developed many other tools, but the principle is the same like the zinc finger tile effector nuclease, CRISPR CAS 9.

1:15:54.450 --> 1:16:4.130  
Evanthia Anastasiadou  
The principal is usually the same as the original one, with the site specific recombination systems, so most of them what they required.

1:16:6.890 --> 1:16:12.410  
Evanthia Anastasiadou  
Consequences. It's your target sequences and an enzyme.

1:16:13.10 --> 1:16:24.890  
Evanthia Anastasiadou  
A recombinase. This enzyme is the one that will realize your target sites, and it's going to cause early combination between them, for example.

1:16:26.40 --> 1:16:42.320  
Evanthia Anastasiadou  
Here in the locks P you have the locks P that are your target sites and then your recombines will bring the sides together and cut out everything that lies in between.

1:16:44.50 --> 1:16:49.570  
Evanthia Anastasiadou  
So you can use them in various different ways, but the principle is always the same.

1:16:51.290 --> 1:16:58.930  
Evanthia Anastasiadou  
You need some target sides in your DNA and a recombinase that will recognize these sides.

1:16:59.890 --> 1:17:4.490  
Evanthia Anastasiadou  
Rearrange them and put them together in a different way.

1:17:7.650 --> 1:17:14.930  
Evanthia Anastasiadou  
You can also have the zinc finger nuclease, CRISPR CAS 9.

1:17:16.610 --> 1:17:21.970  
Evanthia Anastasiadou  
The last one is the one that is more popular nowadays and extensively used.

1:17:22.690 --> 1:17:29.130  
Evanthia Anastasiadou  
I'm not going to go through details, but these are the ones that we are currently using.

1:17:29.880 --> 1:17:37.640  
Evanthia Anastasiadou  
For genome editing and actually nowadays you can even do the genome editing in C2.

1:17:37.960 --> 1:17:42.760  
Evanthia Anastasiadou  
That means in C2 means at the specific place of your interest.

1:17:44.330 --> 1:17:44.570  
Evanthia Anastasiadou  
Without.

1:17:46.290 --> 1:17:50.90  
Evanthia Anastasiadou  
Having to go through different cell manipulation in advance.

1:17:53.80 --> 1:17:53.400  
Evanthia Anastasiadou  
OK.

1:17:53.400 --> 1:18:6.200  
Evanthia Anastasiadou  
So the crisper is the most widely used method so far, and once again you have your the targets just trying to simplify things.

1:18:6.280 --> 1:18:17.200  
Evanthia Anastasiadou  
You have the targets and you have the enzyme that will go and recognize where it has to recombine and stitch together.

1:18:22.450 --> 1:18:22.610  
Evanthia Anastasiadou  
OK.

1:18:22.610 --> 1:18:27.890  
Evanthia Anastasiadou  
We have many applications of crisper past nine genome editing.

1:18:27.930 --> 1:18:30.650  
Evanthia Anastasiadou  
I would say it's the most popular nowadays.

1:18:32.660 --> 1:18:33.820  
Evanthia Anastasiadou  
And just keep this one.

1:18:36.230 --> 1:18:39.270  
Evanthia Anastasiadou  
So when we are talking about applications.

1:18:40.810 --> 1:18:46.290  
Evanthia Anastasiadou  
Advances in genome editing, so cutting and pasting our genome.

1:18:48.90 --> 1:18:48.690  
Evanthia Anastasiadou  
Using it for.

1:18:50.650 --> 1:18:55.10  
Evanthia Anastasiadou  
Training sessions, translocations, even single base editing.

1:18:57.310 --> 1:19:10.910  
Evanthia Anastasiadou  
You can use it for gym regulation like transcriptional regulators, CRISPR interference, CRISPR activations, or even regarding epidemic editing.

1:19:13.400 --> 1:19:25.120  
Evanthia Anastasiadou  
You can also use it for dynamic control of CAS 9 function, chemical induction or optogenetics, and we can even use it to target RNA.

1:19:30.180 --> 1:19:37.180  
Evanthia Anastasiadou  
For the moment, although the Tino made the thing with a crisper cast, 9 is improving fast.

1:19:37.500 --> 1:19:57.580  
Evanthia Anastasiadou  
Still, we are having important issues. For example, during cell modification, the percentage of cell modified varies and if only a few cells are modified. If you have a low yield of modified cells, this is a barrier.

1:19:58.370 --> 1:20:1.10  
Evanthia Anastasiadou  
You will not achieve what you are aiming to.

1:20:2.340 --> 1:20:7.260  
Evanthia Anastasiadou  
Enough cell must be modified to achieve a phenotypic effect.

1:20:9.810 --> 1:20:16.170  
Evanthia Anastasiadou  
Precision is key and the serious concern in off target editing.

1:20:16.930 --> 1:20:21.530  
Evanthia Anastasiadou  
So what do we mean about when we are talking about off target editing?

1:20:21.770 --> 1:20:25.810  
Evanthia Anastasiadou  
So you want specifically to manipulate this site?

1:20:27.370 --> 1:20:35.930  
Evanthia Anastasiadou  
But it's not guaranteed that you're going to manipulate only this site. You have off target effects like.

1:20:37.610 --> 1:20:39.170  
Evanthia Anastasiadou  
Other positions will be edited.

1:20:40.670 --> 1:20:42.110  
Evanthia Anastasiadou  
It was not in your plan.

1:20:43.650 --> 1:20:49.650  
Evanthia Anastasiadou  
If the dreams are there, the knows, then those targeting are modified of target editing.

1:20:49.650 --> 1:20:56.370  
Evanthia Anastasiadou  
The potential for serious adverse events exist, including cancer.

1:21:0.260 --> 1:21:28.780  
Evanthia Anastasiadou  
Applications, so we said Ding anything gene therapy. But not only these ones. You can also use it for microbia microbial fermentation, like antibiotic production, vaccines like hepatitis B, mammalian proteins to produce insulin intact through clotting factors. You can also use it for transgenic plants and animals. Examples in.

1:21:29.460 --> 1:21:30.140  
Evanthia Anastasiadou  
Disease resist.

1:21:31.60 --> 1:21:40.100  
Evanthia Anastasiadou  
Improve product quality, production of pharmaceuticals and for environmental biotechnology.

1:21:46.470 --> 1:21:47.390  
Evanthia Anastasiadou  
Are you still with me?

1:21:51.680 --> 1:21:52.80  
Despoina Voulgari  
Yes.

1:21:53.330 --> 1:21:54.890  
Evanthia Anastasiadou  
That OK.

1:21:55.370 --> 1:21:58.290  
Evanthia Anastasiadou  
Because now I'm gonna change gears a bit and.

1:22:0.90 --> 1:22:4.890  
Evanthia Anastasiadou  
I I'm still talking about gene regulation and gene manipulation.

1:22:4.100 --> 1:22:5.500  
nickcharisis182  
Yeah, I'm sorry. You cut off again.

1:22:6.690 --> 1:22:18.570  
Evanthia Anastasiadou  
OK, I'm I'm still talking about gene regulation and gene manipulation, but instead of playing with ADNA now, we are playing with the RNA.

1:22:19.210 --> 1:22:23.930  
Evanthia Anastasiadou  
So they are quite a few different RNA modules.

1:22:24.650 --> 1:22:28.610  
Evanthia Anastasiadou  
That we can use to regulate the gene expression.

1:22:29.370 --> 1:22:42.290  
Evanthia Anastasiadou  
For example, we have two small single strand RNA, we call them micro RNA and Myrna and SI RN.

1:22:44.490 --> 1:22:46.850  
Evanthia Anastasiadou  
These ones they combine.

1:22:48.50 --> 1:22:58.650  
Evanthia Anastasiadou  
Excuse me, they combined to your mRNA and regulate inhibit actually the gene expression through.

1:23:0.930 --> 1:23:2.370  
Evanthia Anastasiadou  
Personal mechanism.

1:23:4.520 --> 1:23:17.800  
Evanthia Anastasiadou  
So we are in AI either MI or SI is an important that I gave a Nobel Prize in 2006.

1:23:19.370 --> 1:23:23.130  
Evanthia Anastasiadou  
For RNA interference in silen by double stranded RNA.

1:23:27.440 --> 1:23:30.120  
Evanthia Anastasiadou  
So I'm gonna talk very briefly about.

1:23:31.690 --> 1:23:36.210  
Evanthia Anastasiadou  
Micro RNA and SI RNA short interference RNA.

1:23:36.210 --> 1:23:48.570  
Evanthia Anastasiadou  
Both of them are post transcriptional mechanisms for inhibition of gene expression by small single stranded Rna's.

1:23:50.330 --> 1:23:54.410  
Evanthia Anastasiadou  
Initially it was discovered in a worm. See elegance.

1:23:56.90 --> 1:23:56.690  
Evanthia Anastasiadou  
Micro RNA.

1:23:58.20 --> 1:24:8.300  
Evanthia Anastasiadou  
Denise inhibits inexpressions or blocks your gene expression by blocking the translation of the mRNA.

1:24:9.850 --> 1:24:29.410  
Evanthia Anastasiadou  
Just to remind you briefly from the DNA, through transcription information of the mRNA and from mRNA through translation, you have the production of proteins. Now both micro RNA and short interference RNA.

1:24:29.970 --> 1:24:32.650  
Evanthia Anastasiadou  
Are blocking the production.

1:24:34.530 --> 1:24:35.330  
Evanthia Anastasiadou  
Binding to the.

1:24:37.330 --> 1:24:50.50  
Evanthia Anastasiadou  
To humans express about 500 Micron as so we are actually expressing them and it's a mechanism for fine tuning gene regulation.

1:24:51.810 --> 1:24:53.970  
Evanthia Anastasiadou  
Some plans express over.

1:24:56.450 --> 1:25:6.130  
Evanthia Anastasiadou  
Very nice, because a single microtna combined to more than one target mrnas, not only.

1:25:8.290 --> 1:25:8.530  
Evanthia Anastasiadou  
OK.

1:25:8.530 --> 1:25:10.650  
Evanthia Anastasiadou  
Forget about it, it is estimated.

1:25:13.630 --> 1:25:17.590  
Evanthia Anastasiadou  
Jeans may be regulated by MICROARRN as.

1:25:19.130 --> 1:25:23.570  
Evanthia Anastasiadou  
No sin as sort interfering RNAs inhib.

1:25:23.650 --> 1:25:24.10  
Evanthia Anastasiadou  
Keeping it.

1:25:25.770 --> 1:25:35.930  
Evanthia Anastasiadou  
Specifically targeting a complementary mRNA for the graduation. The mechanism of gene silencing by ESSA.

1:25:37.750 --> 1:25:44.630  
Evanthia Anastasiadou  
As RNA interference RNA I and is an important research tool.

1:25:46.170 --> 1:25:54.570  
Evanthia Anastasiadou  
Rnai is thought to play a natural role in protection of cells from RNA viruses and retro transposons.

1:25:54.690 --> 1:26:4.850  
Evanthia Anastasiadou  
So when you have RNA virus, you have SIRN as that will bind on the RNA of the viruses and therefore protect your cells from infection.

1:26:6.420 --> 1:26:18.260  
Evanthia Anastasiadou  
So once again, both of them are small single strand Rna's. They are binding to the mRNA. This one is stopping for for.

1:26:20.490 --> 1:26:20.970  
Evanthia Anastasiadou  
Protein for.

1:26:24.140 --> 1:26:25.700  
Evanthia Anastasiadou  
Your RNA is degraded.

1:26:30.80 --> 1:26:30.600  
Evanthia Anastasiadou  
OK.

1:26:32.630 --> 1:26:33.870  
Evanthia Anastasiadou  
Just give me one second.

1:26:33.910 --> 1:26:36.590  
Evanthia Anastasiadou  
I'm gonna pass. This one's really fast.

1:26:38.740 --> 1:26:43.620  
Evanthia Anastasiadou  
Actually, I'm gonna open the parenthesis here and I'm gonna talk about animal.

1:26:45.370 --> 1:26:57.330  
Evanthia Anastasiadou  
So when you're talking about animal coloning is completely different from gene cloning. OK, when you are talking about gene cloning, we are talking about cutting and.

1:26:59.190 --> 1:27:0.190  
Evanthia Anastasiadou  
Different sequences.

1:27:1.850 --> 1:27:5.490  
Evanthia Anastasiadou  
About animal cloning is a total different process.

1:27:7.650 --> 1:27:15.330  
Evanthia Anastasiadou  
Most likely you will be familiar with Dolly the sheep that was created.

1:27:17.280 --> 1:27:17.800  
Evanthia Anastasiadou  
England.

1:27:20.90 --> 1:27:20.970  
Evanthia Anastasiadou  
Many decades back.

1:27:22.570 --> 1:27:30.490  
Evanthia Anastasiadou  
How was this formed? OK, so once again, I said that gene cloning is completely different.

1:27:32.130 --> 1:27:36.250  
Evanthia Anastasiadou  
So when you are talking about animal cloning, you are not playing with the DNA.

1:27:37.930 --> 1:27:43.810  
Evanthia Anastasiadou  
What you are doing is you are getting the whole DNA, the whole nucleus.

1:27:44.50 --> 1:27:45.10  
Evanthia Anastasiadou  
So you know that.

1:27:45.770 --> 1:27:46.490  
Evanthia Anastasiadou  
Your DNA is.

1:27:48.170 --> 1:27:51.50  
Evanthia Anastasiadou  
Nicely protected in your new nucleus inside your cell.

1:27:52.0 --> 1:27:58.0  
Evanthia Anastasiadou  
So when you are talking about animal cloning, what is happening is you get a, you get an egg.

1:27:58.0 --> 1:28:2.440  
Evanthia Anastasiadou  
The egg most likely is gonna form an embryo.

1:28:3.240 --> 1:28:4.440  
Evanthia Anastasiadou  
What you are doing is you.

1:28:11.570 --> 1:28:12.610  
Evanthia Anastasiadou  
Inserting.

1:28:14.410 --> 1:28:19.10  
Evanthia Anastasiadou  
Those from another cell, from the a donor cell.

1:28:20.50 --> 1:28:20.970  
Evanthia Anastasiadou  
So you have.

1:28:22.570 --> 1:28:30.50  
Evanthia Anastasiadou  
Your excel you remove the nucleus OK and you insert the nucleus from another cell.

1:28:32.700 --> 1:28:42.740  
Evanthia Anastasiadou  
Start, multiply foster mother and you expect to have a new baby that will have the genetic.

1:28:44.530 --> 1:28:46.50  
Evanthia Anastasiadou  
Donor cell.

1:28:48.450 --> 1:28:48.730  
Evanthia Anastasiadou  
The.

1:28:51.50 --> 1:28:53.410  
Evanthia Anastasiadou  
Real that you took from from this.

1:28:55.280 --> 1:28:57.840  
Evanthia Anastasiadou  
So for why is this important?

1:28:58.360 --> 1:29:7.560  
Evanthia Anastasiadou  
Because for example, this has to be an excel, but the donor cell can be any type of cell, can be a skin cell.

1:29:8.160 --> 1:29:20.320  
Evanthia Anastasiadou  
So you can have a cell from your skin. You can take the nucleus from the skin cell and put it back on an egg cell. So in theory.

1:29:21.130 --> 1:29:22.290  
Evanthia Anastasiadou  
You are going to generate.

1:29:23.890 --> 1:29:24.850  
Evanthia Anastasiadou  
An Organism?

1:29:25.740 --> 1:29:28.140  
Evanthia Anastasiadou  
Will have the DNA from.

1:29:30.730 --> 1:29:31.890  
Evanthia Anastasiadou  
You understand that?

1:29:34.440 --> 1:29:42.480  
Despoina Voulgari  
Sorry for the interruptions, but I think there are many interruptions in your speech. In recent previous slide.

1:29:44.940 --> 1:29:45.700  
Evanthia Anastasiadou  
OK.

1:29:45.860 --> 1:29:47.180  
Evanthia Anastasiadou  
Let's go again.

1:29:47.300 --> 1:29:55.940  
Evanthia Anastasiadou  
So I'm talking about animal cloning, OK? And I'm saying that animal cloning is completely different than.

1:29:58.390 --> 1:30:8.390  
Evanthia Anastasiadou  
And in cloning, cloning and it's clicking different because here you are not cutting and pasting parts of your DNA.

1:30:15.240 --> 1:30:16.40  
Evanthia Anastasiadou  
Type of cell.

1:30:19.850 --> 1:30:20.810  
Evanthia Anastasiadou  
Hey, let me see.

1:30:21.290 --> 1:30:22.770  
Evanthia Anastasiadou  
I might have some issues.

1:30:24.290 --> 1:30:24.930  
Evanthia Anastasiadou  
Just give me one second.

1:30:27.330 --> 1:30:28.170  
Evanthia Anastasiadou  
What is happening?

1:30:36.90 --> 1:30:36.530  
Evanthia Anastasiadou  
OK.

1:30:45.550 --> 1:30:46.870  
Evanthia Anastasiadou  
I think that we are.

1:30:53.520 --> 1:30:54.520  
Evanthia Anastasiadou  
Glad we are fine.

1:30:56.90 --> 1:30:56.490  
Evanthia Anastasiadou  
OK so.

1:31:4.740 --> 1:31:10.660  
Vasilis Chatzitolios  
But I don't know if the same for the other search have big poses like 5-6 seconds delays.

1:31:13.530 --> 1:31:14.610  
Vasilis Chatzitolios  
But I don't share anything.

1:31:14.500 --> 1:31:14.860  
Evanthia Anastasiadou  
OK.

1:31:17.240 --> 1:31:18.80  
Evanthia Anastasiadou  
OK.

1:31:21.610 --> 1:31:22.730  
Evanthia Anastasiadou  
Can you hear me now?

1:31:26.490 --> 1:31:28.50  
Vasilis Chatzitolios  
Yes, I can hear you.

1:31:28.290 --> 1:31:33.610  
Vasilis Chatzitolios  
I could hear you before as well, but at some point there is a big gap in your speech.

1:31:35.290 --> 1:31:36.850  
Evanthia Anastasiadou  
OK, OK.

1:31:36.850 --> 1:31:38.290  
Evanthia Anastasiadou  
I'm sorry about that.

1:31:39.850 --> 1:31:45.210  
Evanthia Anastasiadou  
Hopefully we are recording it also, so hopefully it is recorded.

1:31:52.930 --> 1:31:56.250  
Evanthia Anastasiadou  
This is due to my battery that is running low.

1:32:6.830 --> 1:32:6.990  
Evanthia Anastasiadou  
OK.

1:32:32.360 --> 1:32:33.120  
Evanthia Anastasiadou  
OK.

1:32:35.20 --> 1:32:35.180  
Evanthia Anastasiadou  
OK.

1:32:52.780 --> 1:32:53.860  
Evanthia Anastasiadou  
OK so.

1:32:56.520 --> 1:32:57.760  
Evanthia Anastasiadou  
I will continue.

1:32:59.490 --> 1:33:2.250  
Evanthia Anastasiadou  
Most likely my computer.

1:33:4.210 --> 1:33:5.370  
Evanthia Anastasiadou  
Is in like 10.

1:33:7.290 --> 1:33:12.890  
Evanthia Anastasiadou  
Will continue as long as my battery is allowing me.

1:33:20.580 --> 1:33:24.460  
Evanthia Anastasiadou  
Yeah, because I don't have the charger with me and.

1:33:28.970 --> 1:33:30.10  
Evanthia Anastasiadou  
Oh, oh, let's see.

1:33:30.10 --> 1:33:31.530  
Evanthia Anastasiadou  
Let me think, OK.

1:33:33.250 --> 1:33:35.570  
Evanthia Anastasiadou  
Can can you give me 5 minutes so.

1:33:37.870 --> 1:33:38.790  
Evanthia Anastasiadou  
Position and.

1:33:40.490 --> 1:33:44.50  
Evanthia Anastasiadou  
The charger. OK, just 5 minutes.

1:33:44.850 --> 1:33:45.410  
Evanthia Anastasiadou  
I'm moving.

1:33:46.930 --> 1:33:47.690  
Evanthia Anastasiadou  
And I'll be back with you.

1:36:29.810 --> 1:36:30.170  
Evanthia Anastasiadou  
Hey.

1:36:31.980 --> 1:36:33.140  
Evanthia Anastasiadou  
It's hope that it will be.

1:36:41.800 --> 1:36:42.160  
Evanthia Anastasiadou  
OK.

1:36:51.670 --> 1:36:53.70  
Evanthia Anastasiadou  
Do you hear me better now?

1:36:54.790 --> 1:36:56.590  
Evanthia Anastasiadou  
Or do you still have caps?

1:36:58.860 --> 1:37:4.980  
nickcharisis182  
Quality is not as good, but it seems to be more so to be more persistent so.

1:37:1.420 --> 1:37:2.60  
Evanthia Anastasiadou  
Same same same.

1:37:6.730 --> 1:37:7.610  
nickcharisis182  
There's this trade off.

1:37:6.750 --> 1:37:7.550  
Evanthia Anastasiadou  
OK.

1:37:10.420 --> 1:37:20.940  
Evanthia Anastasiadou  
I'm sorry about that. If you if there are big gaps, just please interrupt me and let me know so I can explain it again, OK.

1:37:23.10 --> 1:37:23.730  
Evanthia Anastasiadou  
OK.

1:37:23.770 --> 1:37:30.890  
Evanthia Anastasiadou  
So I was talking about animal cloning and I was saying that animal cloning is completely different than gene.

1:37:37.610 --> 1:37:38.890  
Evanthia Anastasiadou  
And be a name manipulation.

1:37:42.850 --> 1:37:45.850  
nickcharisis182  
Yeah, sorry. They got the gaps are back.

1:37:43.20 --> 1:37:43.700  
Evanthia Anastasiadou  
We talked about.

1:37:45.450 --> 1:37:45.570  
Evanthia Anastasiadou  
The.

1:38:4.280 --> 1:38:6.0  
Evanthia Anastasiadou  
Have I lost you completely?

1:38:8.330 --> 1:38:10.370  
nickcharisis182  
I know. But you were cutting off before.

1:38:12.580 --> 1:38:12.980  
Evanthia Anastasiadou  
OK.

1:38:15.820 --> 1:38:16.460  
Evanthia Anastasiadou  
Let's see how.

1:38:18.890 --> 1:38:20.170  
Evanthia Anastasiadou  
The beginning, did we?

1:38:23.350 --> 1:38:23.670  
nickcharisis182  
Correct.

1:38:25.200 --> 1:38:29.880  
Evanthia Anastasiadou  
You didn't have serious issues in the beginning, did we?

1:38:31.50 --> 1:38:33.770  
nickcharisis182  
Not for the cat, not not for the time I can think of.

1:38:35.480 --> 1:38:38.520  
Evanthia Anastasiadou  
Becoming more and more disturbing, correct.

1:38:44.340 --> 1:38:49.580  
nickcharisis182  
Yeah, I'm not going to like troubleshoot it with you, but are you running anything else today or the background?

1:38:49.580 --> 1:38:50.900  
nickcharisis182  
Maybe it's that it's not.

1:38:50.940 --> 1:38:51.380  
nickcharisis182  
It may not be.

1:38:51.380 --> 1:38:53.60  
nickcharisis182  
The connection might be the.

1:38:54.610 --> 1:38:56.610  
nickcharisis182  
The usage of your PC resources.

1:39:0.60 --> 1:39:0.100  
Evanthia Anastasiadou  
A.

1:39:0.940 --> 1:39:4.300  
nickcharisis182  
No, I I don't know if that's the case, but it might be.

1:39:5.440 --> 1:39:6.160  
Evanthia Anastasiadou  
OK.

1:39:9.660 --> 1:39:10.340  
Evanthia Anastasiadou  
OK.

1:39:13.450 --> 1:39:13.850  
Evanthia Anastasiadou  
OK.

1:39:13.850 --> 1:39:15.330  
Evanthia Anastasiadou  
Let's see, let's try.

1:39:17.370 --> 1:39:18.90  
Evanthia Anastasiadou  
To continue.

1:39:19.810 --> 1:39:23.90  
Evanthia Anastasiadou  
And you let me know if we have any issues.

1:39:23.90 --> 1:39:24.90  
Evanthia Anastasiadou  
Just repeat.

1:39:26.770 --> 1:39:28.850  
Evanthia Anastasiadou  
You don't like animal cloning? Definitely.

1:39:30.410 --> 1:39:37.570  
Evanthia Anastasiadou  
So, OK, going back to animal cloning, I said that it's a a completely different procedure than.

1:39:39.250 --> 1:39:39.570  
Evanthia Anastasiadou  
Dean cloning.

1:39:40.310 --> 1:39:45.950  
Evanthia Anastasiadou  
And the main difference is I I described previously that in the gym cloning.

1:39:48.10 --> 1:39:52.650  
Evanthia Anastasiadou  
Basing different bits and pieces of the DNA.

1:39:53.170 --> 1:39:56.690  
Evanthia Anastasiadou  
Here you're taking the DNA as a whole.

1:39:58.330 --> 1:40:0.850  
Evanthia Anastasiadou  
The DNA in the cell is in the nucleus.

1:40:1.330 --> 1:40:3.850  
Evanthia Anastasiadou  
So you are taking the whole nucleus.

1:40:4.570 --> 1:40:10.690  
Evanthia Anastasiadou  
You have a donor cell and the recipient cell. The recipient cell is always an egg.

1:40:11.600 --> 1:40:12.240  
Evanthia Anastasiadou  
It's like.

1:40:14.190 --> 1:40:15.310  
Evanthia Anastasiadou  
Multiplied.

1:40:17.200 --> 1:40:18.0  
Evanthia Anastasiadou  
And eventually.

1:40:20.310 --> 1:40:29.870  
Evanthia Anastasiadou  
The donors, their hand can be any cell can be like a cell for skin. A cell from your skin.

1:40:29.990 --> 1:40:36.310  
Evanthia Anastasiadou  
So you are taking some cells from your skin. You are taking the nucleus, you are putting the nucleus.

1:40:39.470 --> 1:40:43.870  
Evanthia Anastasiadou  
And then eventually this will multiply to make an.

1:40:48.130 --> 1:40:56.930  
Evanthia Anastasiadou  
So you are cloning organisms and this is the theory that we might even be able to clone ourselves.

1:40:58.490 --> 1:41:1.90  
Evanthia Anastasiadou  
Why? Because you don't need to start from the beginning.

1:41:1.370 --> 1:41:8.970  
Evanthia Anastasiadou  
You don't need to cut and paste different bits and pieces of the DNA. All you need to do is take your whole DNA.

1:41:11.360 --> 1:41:14.720  
Evanthia Anastasiadou  
Nucleus of any kind of cell.

1:41:16.290 --> 1:41:21.290  
Evanthia Anastasiadou  
And transfer it in a cell that you have delete the nucleus.

1:41:23.980 --> 1:41:24.820  
Evanthia Anastasiadou  
Then this will.

1:41:26.370 --> 1:41:27.90  
Evanthia Anastasiadou  
Start multiplying.

1:41:27.490 --> 1:41:28.530  
Evanthia Anastasiadou  
Make an embryo.

1:41:28.930 --> 1:41:31.930  
Evanthia Anastasiadou  
Put it in a foster mother and the foster mother.

1:41:31.930 --> 1:41:33.570  
Evanthia Anastasiadou  
That's where the embryo is gonna.

1:41:35.290 --> 1:41:35.850  
Evanthia Anastasiadou  
Be form.

1:41:37.490 --> 1:41:39.330  
Evanthia Anastasiadou  
And eventually keep an Organism.

1:41:40.10 --> 1:41:42.530  
Evanthia Anastasiadou  
That's how Dolly was created.

1:41:44.770 --> 1:41:48.650  
Evanthia Anastasiadou  
So talking about Dolly once again so.

1:41:50.450 --> 1:41:53.90  
Evanthia Anastasiadou  
The egg that we take from 1:00.

1:41:54.890 --> 1:41:56.130  
Evanthia Anastasiadou  
From 1 sheep.

1:41:56.980 --> 1:42:6.420  
Evanthia Anastasiadou  
You take the nucleus out so you have the egg, but with no DNA in there, and then you take the nucleus from another cell.

1:42:7.970 --> 1:42:9.410  
Evanthia Anastasiadou  
Might be a skin cell might be.

1:42:9.410 --> 1:42:17.570  
Evanthia Anastasiadou  
A muscle cell might be a neural cell might be whatever type of cell, so you're putting the nucleus in this empty cell.

1:42:17.570 --> 1:42:25.970  
Evanthia Anastasiadou  
It will start multiplying and when you have a critical mass, you put it in Foster mother in a surrogate mother.

1:42:26.720 --> 1:42:29.760  
Evanthia Anastasiadou  
And eventually what you expect is that.

1:42:31.890 --> 1:42:35.90  
Evanthia Anastasiadou  
These will give rise of a new animal.

1:42:36.730 --> 1:42:37.930  
Evanthia Anastasiadou  
OK this.

1:42:39.890 --> 1:42:48.50  
Evanthia Anastasiadou  
Easy as I am presenting them to be able to get an alive animal you have to do this experiment many times.

1:42:49.610 --> 1:42:50.170  
Evanthia Anastasiadou  
They injected.

1:42:51.890 --> 1:42:52.690  
Evanthia Anastasiadou  
Nuclei extracted from.

1:42:54.450 --> 1:42:55.650  
Evanthia Anastasiadou  
From the sheep.

1:42:57.850 --> 1:43:1.50  
Evanthia Anastasiadou  
Into 300 empty eggs.

1:43:2.770 --> 1:43:4.170  
Evanthia Anastasiadou  
So you can say that you.

1:43:6.50 --> 1:43:9.690  
Evanthia Anastasiadou  
Procedure again and again from these 300.

1:43:11.250 --> 1:43:12.770  
Evanthia Anastasiadou  
Attempts they create.

1:43:14.450 --> 1:43:15.650  
Evanthia Anastasiadou  
More than 30 embryos.

1:43:17.410 --> 1:43:29.210  
Evanthia Anastasiadou  
From their embryos, only 5 developed into a lamp and only one lamp survive. So as you can see, it's not a a very efficient procedure.

1:43:29.770 --> 1:43:35.930  
Evanthia Anastasiadou  
So you have to do this nuclear transfer many times to be able to have.

1:43:36.860 --> 1:43:38.940  
Evanthia Anastasiadou  
In a live Organism at the end.

1:43:40.660 --> 1:43:48.380  
Evanthia Anastasiadou  
And that's why we are saying that we cannot use it in humans yet. We have quite a few steps to develop.

1:43:49.20 --> 1:43:50.660  
Evanthia Anastasiadou  
This is not a safe procedure.

1:43:54.400 --> 1:43:58.200  
Evanthia Anastasiadou  
There are many examples. We've done it for many animals so far.

1:43:58.480 --> 1:44:3.760  
Evanthia Anastasiadou  
There was a huge trend, sometimes back to generate cats and dogs.

1:44:6.330 --> 1:44:7.90  
Evanthia Anastasiadou  
So you could.

1:44:8.890 --> 1:44:16.930  
Evanthia Anastasiadou  
Be seeing cat and get some cells and try to recreate a new version of your disease cat.

1:44:19.470 --> 1:44:19.870  
Evanthia Anastasiadou  
We're.

1:44:21.520 --> 1:44:22.640  
Evanthia Anastasiadou  
We tried like.

1:44:24.210 --> 1:44:27.970  
Evanthia Anastasiadou  
Shapes my scarlet goats, pigs, cats and rabbits, etcetera.

1:44:30.980 --> 1:44:37.180  
Evanthia Anastasiadou  
We we went tide and you might have heard that in the news again and again we are.

1:44:37.180 --> 1:44:40.500  
Evanthia Anastasiadou  
They are trying even to recreate.

1:44:42.50 --> 1:44:54.570  
Evanthia Anastasiadou  
Spaces like the Tasmanian tiger or the woolly mammoth for the woolly mammoth. They are using a surrogate mother, an elephant, of course. None of these had worked so far.

1:44:56.900 --> 1:44:59.300  
Evanthia Anastasiadou  
Talking about animal cloning again.

1:45:0.850 --> 1:45:1.90  
Evanthia Anastasiadou  
Expensive.

1:45:2.770 --> 1:45:17.490  
Evanthia Anastasiadou  
It's highly inefficient with a low success rate. A cloned animal stand to have more compromised immune function and higher age of infection, tumor growth and other disorders.

1:45:17.610 --> 1:45:19.450  
Evanthia Anastasiadou  
So they have a tendency to like to.

1:45:21.950 --> 1:45:29.150  
Evanthia Anastasiadou  
Location the overall efficacy of cloning is usually between 0 and 3%.

1:45:29.240 --> 1:45:29.520  
Evanthia Anastasiadou  
And.

1:45:40.50 --> 1:45:42.50  
Evanthia Anastasiadou  
The efficacy is really, really low.

1:45:45.240 --> 1:45:48.280  
Evanthia Anastasiadou  
OK. We also have some other.

1:45:49.830 --> 1:45:50.550  
Evanthia Anastasiadou  
Mouse models.

1:45:50.670 --> 1:45:52.190  
Evanthia Anastasiadou  
Some other animal.

1:45:54.160 --> 1:45:56.200  
Evanthia Anastasiadou  
According to what we want to study.

1:45:57.750 --> 1:46:11.310  
Evanthia Anastasiadou  
For this models, so I already talked about transcendence. I talked about knocking to knockouts, we talked about Inc 2, genome editing like with the CRISPR and we.

1:46:15.150 --> 1:46:22.30  
Evanthia Anastasiadou  
Now you can also have mouse bone marrow models, transplantation models.

1:46:22.510 --> 1:46:29.270  
Evanthia Anastasiadou  
These are the ones that you are using exactly as you are doing with humans.

1:46:29.830 --> 1:46:34.710  
Evanthia Anastasiadou  
So be able to do a bone marrow transplantation.

1:46:34.950 --> 1:46:39.910  
Evanthia Anastasiadou  
You are having your donor mice and your recipient mice.

1:46:40.710 --> 1:46:46.790  
Evanthia Anastasiadou  
So the donor mice are the ones that they are going to give you the bone marrow cells.

1:46:48.260 --> 1:46:49.500  
Evanthia Anastasiadou  
You are gonna take them.

1:46:49.940 --> 1:47:4.260  
Evanthia Anastasiadou  
You're gonna manipulate them very often with viral vectors, and after the manipulation you are gonna put them back to the recipient. Marks the recipient marks.

1:47:7.450 --> 1:47:13.330  
Evanthia Anastasiadou  
There with Proteation or with chemicals to create their own cells.

1:47:13.450 --> 1:47:15.930  
Evanthia Anastasiadou  
So the ones that you are putting in.

1:47:17.470 --> 1:47:23.230  
Evanthia Anastasiadou  
They will go and form and multiply and replace the original ones.

1:47:23.830 --> 1:47:29.190  
Evanthia Anastasiadou  
So talking about bone marrow, you have the donor, you have the recipient.

1:47:29.430 --> 1:47:30.590  
Evanthia Anastasiadou  
Just give me a second.

1:47:36.320 --> 1:47:47.80  
Evanthia Anastasiadou  
So talking about bongo transplantation, you have the donor, you have the recipient. The donor is the one that is giving the signs. In our case, the bone marrow cells.

1:47:48.930 --> 1:47:53.250  
Evanthia Anastasiadou  
Arizard and manipulated through viral vectors.

1:47:54.790 --> 1:47:59.190  
Evanthia Anastasiadou  
And after the manipulation you are putting them back to a recipient mouse.

1:47:59.190 --> 1:48:12.630  
Evanthia Anastasiadou  
This recipient mouse you need to kill the bone marrow transplants. The bone marrow cells in advance. So the ones that you are putting in, they will go find a place.

1:48:14.310 --> 1:48:21.710  
Evanthia Anastasiadou  
Home and multiply and then you will follow these animals to see how they will do if they will develop disease.

1:48:22.740 --> 1:48:23.220  
Evanthia Anastasiadou  
Etcetera.

1:48:25.600 --> 1:48:30.720  
Evanthia Anastasiadou  
I think of it last model that I will present you today is the mouse xenograph.

1:48:32.470 --> 1:48:34.310  
Evanthia Anastasiadou  
What do we mean about xenoprophs?

1:48:34.790 --> 1:48:41.230  
Evanthia Anastasiadou  
Xenografts is when you are using tissues, cells, organs from a.

1:48:43.550 --> 1:48:53.30  
Evanthia Anastasiadou  
So in this case, what we are doing is in the xenografts we are using human cells and put them in mice.

1:48:54.630 --> 1:48:59.30  
Evanthia Anastasiadou  
In this, human cells can be like from human tumors.

1:49:0.870 --> 1:49:7.150  
Evanthia Anastasiadou  
See how they will develop and if a specific drug will inhibit its growth or not.

1:49:7.550 --> 1:49:13.470  
Evanthia Anastasiadou  
So human cells that are isolated, for example from humors.

1:49:14.250 --> 1:49:24.970  
Evanthia Anastasiadou  
Are gonna be injected either in specific places or randomly in mice and see how they will develop various tumors.

1:49:28.130 --> 1:49:34.130  
Evanthia Anastasiadou  
You're inside millions of sex and you follow your mice for a period of time.

1:49:35.670 --> 1:49:42.950  
Evanthia Anastasiadou  
Now, because the uter cells are human and not mouse cells.

1:49:44.900 --> 1:49:48.820  
Evanthia Anastasiadou  
You would expect that there will be rejected from the animal.

1:49:50.390 --> 1:49:54.790  
Evanthia Anastasiadou  
So cannot cross species so.

1:49:56.550 --> 1:50:7.670  
Evanthia Anastasiadou  
The animal will realize that the human cells are foreign organisms and system will be activated and they will gonna fight and eliminate the human sense.

1:50:9.270 --> 1:50:12.390  
Evanthia Anastasiadou  
The only way that you can make this mice.

1:50:14.110 --> 1:50:17.430  
Evanthia Anastasiadou  
Not to kill the human cells is if you.

1:50:19.670 --> 1:50:26.830  
Evanthia Anastasiadou  
Own system. So the mice that you are using in xenografts we call them scape mice.

1:50:28.910 --> 1:50:29.910  
Evanthia Anastasiadou  
Severe comb.

1:50:31.590 --> 1:50:31.750  
Evanthia Anastasiadou  
Ined.

1:50:33.390 --> 1:50:36.510  
Evanthia Anastasiadou  
And these mice, they're immune system is compromised.

1:50:36.510 --> 1:50:41.910  
Evanthia Anastasiadou  
Their immune system is not working so well so they you can see.

1:50:43.990 --> 1:50:48.510  
Evanthia Anastasiadou  
You will not realize that these cells are foreign cells.

1:50:48.790 --> 1:50:52.110  
Evanthia Anastasiadou  
They are gonna grow in the mouse and they will gonna.

1:50:54.800 --> 1:50:54.920  
Evanthia Anastasiadou  
Umm.

1:50:56.710 --> 1:51:0.350  
Evanthia Anastasiadou  
Maybe two more to study.

1:51:2.350 --> 1:51:4.70  
Evanthia Anastasiadou  
They forget about this.

1:51:5.670 --> 1:51:5.830  
Evanthia Anastasiadou  
Ones.

1:51:7.430 --> 1:51:13.950  
Evanthia Anastasiadou  
The advantages and disadvantages mouse in a bus is a a model that you can generate quickly.

1:51:14.550 --> 1:51:15.750  
Evanthia Anastasiadou  
It's humanized.

1:51:15.750 --> 1:51:20.990  
Evanthia Anastasiadou  
So the background of the cells is human, so it's closer to reality.

1:51:23.170 --> 1:51:37.450  
Evanthia Anastasiadou  
And most likely you are talking about a known genetic event, meaning that the cells that you are getting from the tumor you already know the genetic alteration that is causing the tumor.

1:51:39.590 --> 1:51:42.310  
Evanthia Anastasiadou  
Disadvantages against can be anywhere.

1:51:45.150 --> 1:51:48.110  
Evanthia Anastasiadou  
Controlled and you are using.

1:51:49.790 --> 1:52:2.390  
Evanthia Anastasiadou  
Anomised immune system so you don't know what is going to happen in real life when you have an Organism will with a full normal immune system.

1:52:4.970 --> 1:52:5.530  
Evanthia Anastasiadou  
OK.

1:52:10.190 --> 1:52:11.70  
Evanthia Anastasiadou  
Mouse models.

1:52:12.630 --> 1:52:23.670  
Evanthia Anastasiadou  
It's very important because we started with the genetic engineering. OK, how you can cut and paste bits and pieces of your DNA.

1:52:26.490 --> 1:52:27.650  
Evanthia Anastasiadou  
I did animal.

1:52:30.520 --> 1:52:32.880  
Evanthia Anastasiadou  
Like transgenic knockouts, etcetera.

1:52:35.560 --> 1:52:39.840  
Evanthia Anastasiadou  
But you are gonna use for to check for drugs.

1:52:41.750 --> 1:52:47.910  
Evanthia Anastasiadou  
Gene based drugs and the body drugs regenerated medicine gene therapy.

1:52:49.810 --> 1:52:56.90  
Evanthia Anastasiadou  
So usually when you're studying for therapy.

1:52:57.710 --> 1:53:6.310  
Evanthia Anastasiadou  
It's a long and painful procedure and has various steps. Always. The first step is basic research.

1:53:6.550 --> 1:53:9.990  
Evanthia Anastasiadou  
That's from where everything is starting exploring.

1:53:10.390 --> 1:53:11.910  
Evanthia Anastasiadou  
Perform your basic research.

1:53:11.910 --> 1:53:17.110  
Evanthia Anastasiadou  
This will take years and years, and this is a very expensive procedure.

1:53:19.40 --> 1:53:21.600  
Evanthia Anastasiadou  
If you are lucky enough and you have this.

1:53:23.750 --> 1:53:31.590  
Evanthia Anastasiadou  
Then you are gonna move to the clinical test. The clinical tests are having.

1:53:33.190 --> 1:53:39.830  
Evanthia Anastasiadou  
3 stages 1-2 and three. So the clinic is when you will gonna.

1:53:42.430 --> 1:53:48.310  
Evanthia Anastasiadou  
Check the results of your drug and patients, but before rating that step.

1:53:49.40 --> 1:53:56.80  
Evanthia Anastasiadou  
You always have the preclinical step. The preclinical step is your mice.

1:53:56.560 --> 1:53:58.480  
Evanthia Anastasiadou  
You are using your mice.

1:53:58.720 --> 1:54:13.840  
Evanthia Anastasiadou  
You have already generated a mouse model for this specific disease and you are using in huge numbers to check if your compound is causing any effect. If you pass the mouse.

1:54:14.630 --> 1:54:18.710  
Evanthia Anastasiadou  
State the preclinical tests. Then you are moving to the clinical tests.

1:54:19.110 --> 1:54:20.870  
Evanthia Anastasiadou  
The clinical tests are divided.

1:54:21.450 --> 1:54:23.50  
Evanthia Anastasiadou  
In three different stages.

1:54:23.530 --> 1:54:29.730  
Evanthia Anastasiadou  
The first one is just to check if your drug is causing any.

1:54:32.220 --> 1:54:39.580  
Evanthia Anastasiadou  
Or not. So the first one. You don't even check how good your drug is working.

1:54:39.580 --> 1:54:42.580  
Evanthia Anastasiadou  
You are just checking that it's not killing your patient.

1:54:42.580 --> 1:54:55.420  
Evanthia Anastasiadou  
Basically, if you get the approval and the side effects are not serious, then you are moving to phase two in the phase two you have a small group.

1:54:56.990 --> 1:55:4.350  
Evanthia Anastasiadou  
Selected patients and you are gonna check them and see if your your drug will improve their state.

1:55:6.310 --> 1:55:9.390  
Evanthia Anastasiadou  
Finally, your final step.

1:55:10.950 --> 1:55:15.190  
Evanthia Anastasiadou  
And that's where 95% of all the drugs fail.

1:55:15.430 --> 1:55:24.590  
Evanthia Anastasiadou  
You're talking about the huge population of patients that you're gonna use in clinical trials and see what is the outcome of your drug.

1:55:26.310 --> 1:55:31.30  
Evanthia Anastasiadou  
If you pass all this, then you are safe to move to the market.

1:55:34.160 --> 1:55:36.160  
Evanthia Anastasiadou  
OK so.

1:55:37.670 --> 1:55:37.990  
Evanthia Anastasiadou  
I think.

1:55:39.630 --> 1:55:41.150  
Evanthia Anastasiadou  
That. That's where I'm gonna stop.

1:55:43.830 --> 1:55:53.70  
Evanthia Anastasiadou  
So we talked about various different types of gene manipulation and mouse models.

1:55:56.190 --> 1:56:0.390  
Evanthia Anastasiadou  
I wanted also to talk about cell therapies, but.

1:56:3.910 --> 1:56:5.30  
Evanthia Anastasiadou  
It's not gonna happen today.

1:56:6.590 --> 1:56:8.750  
Evanthia Anastasiadou  
So I'm gonna stop here.

1:56:10.390 --> 1:56:17.670  
Evanthia Anastasiadou  
And we'll see how the biology lectures will go. And maybe if we'll have enough time, I can squeeze some information there.

1:56:19.390 --> 1:56:23.470  
Evanthia Anastasiadou  
So for the moment, I want you to keep in mind that today we talked about.

1:56:25.110 --> 1:56:32.150  
Evanthia Anastasiadou  
Gene manipulation. We talk about gene cloning and animal cloning.

1:56:32.590 --> 1:56:36.30  
Evanthia Anastasiadou  
We talked about the different mouse models that you can use.

1:56:37.970 --> 1:56:40.730  
Evanthia Anastasiadou  
And I think that's where I'm gonna stop.

1:56:40.730 --> 1:56:43.50  
Evanthia Anastasiadou  
Do you have any questions?

1:56:48.230 --> 1:56:51.470  
nickcharisis182  
Hey, are you going to upload those these slides from the class?

1:56:55.0 --> 1:56:57.480  
Evanthia Anastasiadou  
Load them because.

1:56:56.330 --> 1:56:59.10  
nickcharisis182  
But it yeah, you kind of get sorry.

1:57:0.660 --> 1:57:4.660  
Evanthia Anastasiadou  
Yeah. So I'll see how I can upload them.

1:57:7.190 --> 1:57:7.350  
Evanthia Anastasiadou  
OK.

1:57:7.350 --> 1:57:8.70  
Evanthia Anastasiadou  
Just give me one second.

1:57:16.190 --> 1:57:19.430  
Evanthia Anastasiadou  
I'll see how I can upload them because.

1:57:20.990 --> 1:57:26.30  
Evanthia Anastasiadou  
These presentations are really big and I have to sort out how we should do the.

1:57:27.830 --> 1:57:29.790  
Evanthia Anastasiadou  
The recording also I don't know.

1:57:29.790 --> 1:57:32.550  
Evanthia Anastasiadou  
Do you have? Do you think that you have access to the recording?

1:57:35.300 --> 1:57:35.340  
nickcharisis182  
I.

1:57:35.340 --> 1:57:37.540  
nickcharisis182  
I really don't know how to how to works.

1:57:35.680 --> 1:57:36.40  
Evanthia Anastasiadou  
Looks like.

1:57:37.740 --> 1:57:39.340  
Evanthia Anastasiadou  
Yeah. Yeah, yeah, yeah, yeah. OK.

1:57:39.420 --> 1:57:50.340  
Evanthia Anastasiadou  
So these are issues that we have to sort out together because as I said, I haven't used it for teams, but I'm sure that there will be ways.

1:57:50.620 --> 1:57:54.580  
Evanthia Anastasiadou  
So definitely I'm gonna upload them.

1:57:56.150 --> 1:58:7.30  
Evanthia Anastasiadou  
And I'm gonna share the the the recordings with you also, but just give me some time to sort out how and where I should do it. OK.

1:58:8.160 --> 1:58:8.520  
nickcharisis182  
OK.

1:58:8.520 --> 1:58:9.0  
nickcharisis182  
Thank you.

1:58:10.310 --> 1:58:11.430  
Evanthia Anastasiadou  
Any other questions?

1:58:11.750 --> 1:58:15.270  
Vasilis Chatzitolios  
The Professor of machine learning uploads them on YouTube.

1:58:12.870 --> 1:58:13.30  
stella mertzani  
Yeah.

1:58:15.270 --> 1:58:18.30  
Vasilis Chatzitolios  
I don't know in a private link.

1:58:17.890 --> 1:58:18.290  
Evanthia Anastasiadou  
Oh.

1:58:19.680 --> 1:58:20.880  
Evanthia Anastasiadou  
OK.

1:58:20.920 --> 1:58:21.880  
Evanthia Anastasiadou  
That's good.

1:58:22.40 --> 1:58:27.560  
Evanthia Anastasiadou  
I'm letting you think I'm gonna. I'm gonna explore that one. Also, I love that.

1:58:22.490 --> 1:58:23.890  
Vasilis Chatzitolios  
Sends Azel in so.

1:58:29.70 --> 1:58:29.110  
Evanthia Anastasiadou  
OK.

1:58:31.660 --> 1:58:32.780  
Evanthia Anastasiadou  
Any other suggestions?

1:58:34.790 --> 1:58:36.190  
stella mertzani  
It's not a suggestion.

1:58:36.190 --> 1:58:37.750  
stella mertzani  
It's more like a question.

1:58:38.390 --> 1:58:41.470  
stella mertzani  
Do you recommend any books for the?

1:58:43.700 --> 1:58:44.460  
stella mertzani  
Lesson.

1:58:47.40 --> 1:58:47.760  
Evanthia Anastasiadou  
You know what?

1:58:47.880 --> 1:58:52.400  
Evanthia Anastasiadou  
There are many. There are like hundreds books up there out there.

1:58:52.560 --> 1:58:54.520  
Evanthia Anastasiadou  
But the problem is that.

1:58:58.70 --> 1:59:15.830  
Evanthia Anastasiadou  
I mean like nothing is tailor made. So my suggestion is just stick on the lectures, stick on the PowerPoints if you need any specific information, more information apart from the PowerPoints, just contact your teacher's directly.

1:59:18.860 --> 1:59:25.660  
Evanthia Anastasiadou  
In the air of chat GP, I'm like we rarely use, I think textbooks anymore.

1:59:30.930 --> 1:59:35.170  
Evanthia Anastasiadou  
So trying so try not to make your life too complicated.

1:59:31.20 --> 1:59:31.460  
stella mertzani  
Thank you.

1:59:35.170 --> 1:59:42.890  
Evanthia Anastasiadou  
I mean, I'll talk to doctor struggle for this and we will try to make a list of books that you can.

1:59:47.590 --> 1:59:52.710  
Evanthia Anastasiadou  
Check, but I think that you're gonna be completely lost if you start checking other textbooks.

1:59:52.710 --> 1:59:55.630  
Evanthia Anastasiadou  
Also, just stick on the PowerPoint presentations.

2:0:0.30 --> 2:0:0.830  
Evanthia Anastasiadou  
Anything else?

2:0:4.530 --> 2:0:6.970  
Evanthia Anastasiadou  
So my students Thursday, we will be together again.

2:0:8.510 --> 2:0:18.310  
Evanthia Anastasiadou  
We said 9:00, if that's possible. I'm gonna send you the link through teams again, and let's hope that.

2:0:20.190 --> 2:0:22.350  
Evanthia Anastasiadou  
We will have a more smooth journey.

2:0:24.110 --> 2:0:26.350  
Evanthia Anastasiadou  
On Thursday, with not many interruptions.

2:0:28.140 --> 2:0:30.860  
Evanthia Anastasiadou  
If you need anything at all, yes, of course.

2:0:28.280 --> 2:0:29.160  
Vasilis Chatzitolios  
There's something.

2:0:31.550 --> 2:0:37.710  
Vasilis Chatzitolios  
On Tuesdays, the lessons are gonna be on 10:00 or 12:00.

2:0:43.30 --> 2:0:46.270  
Evanthia Anastasiadou  
They're gonna do three lessons together.

2:0:46.790 --> 2:0:50.870  
Evanthia Anastasiadou  
No, sorry. Yes, Tuesdays, huh?

2:0:55.560 --> 2:0:56.400  
Evanthia Anastasiadou  
Just give me a second.

2:1:4.600 --> 2:1:5.480  
Evanthia Anastasiadou  
OK.

2:1:8.80 --> 2:1:17.200  
Evanthia Anastasiadou  
No, Tuesdays I'm not gonna teach you again on Tuesdays. So I think that you are gonna go back to your normal time.

2:1:18.860 --> 2:1:26.20  
Evanthia Anastasiadou  
So this is the only Tuesday that I shared with you, so I would expect that you're gonna go back to your normal time.

2:1:19.650 --> 2:1:20.210  
Vasilis Chatzitolios  
Thank you.

2:1:28.330 --> 2:1:28.690  
Vasilis Chatzitolios  
Thank you.

2:1:29.780 --> 2:1:36.60  
Evanthia Anastasiadou  
We are gonna have like 31st of October 7th of November 14th of November.

2:1:36.500 --> 2:1:44.300  
Evanthia Anastasiadou  
We are gonna be together. So on Thursdays, as I said, if that's OK with you, let's start at 9:00.

2:1:50.450 --> 2:1:56.210  
Evanthia Anastasiadou  
I'll send you links for Thursdays and if you have any questions, you can always e-mail.

2:1:57.750 --> 2:1:58.150  
Evanthia Anastasiadou  
Me. OK.

2:2:4.960 --> 2:2:5.800  
Evanthia Anastasiadou  
Thank you very much.

2:2:7.350 --> 2:2:8.270  
Evanthia Anastasiadou  
Any other questions?

2:2:13.590 --> 2:2:14.190  
Evanthia Anastasiadou  
Thank you guys.

2:2:14.190 --> 2:2:17.270  
Evanthia Anastasiadou  
It was a pleasure meeting you and I wish you.

2:2:18.830 --> 2:2:22.590  
Evanthia Anastasiadou  
A nice and fruitful journey in this master course.

2:2:25.690 --> 2:2:26.90  
Despoina Voulgari  
Thank you.

2:2:26.530 --> 2:2:27.50  
Yioryos  
Thank you.

2:2:27.50 --> 2:2:27.570  
stella mertzani  
Thank you.

2:2:27.650 --> 2:2:28.210  
mariadeftereou  
Thank you.

2:2:27.720 --> 2:2:28.40  
Ξανθίππη Λούκα  
Thank you.

2:2:29.520 --> 2:2:30.280  
Evanthia Anastasiadou  
Thank you.

2:2:30.270 --> 2:2:30.990  
dinakostandina1234  
Thank you.

2:2:30.280 --> 2:2:32.160  
Evanthia Anastasiadou  
Bye bye bye.

2:2:33.710 --> 2:2:33.830  
nickcharisis182  
Bye.