**PROFESSOR:** So here's what we did. We found mutants that effect biochemistry. That's one way to make the connection between function and gene. But if we want to go the other way, we now have to do the biochemistry of genetics.

What's biochemistry about? It's purifying things in a test tube. What's genetics about? Heredity. So what do we have to do? We have to purify heredity in a test tube. But that's it. That's what we have to do.

If we're going to make that connection in that direction, all we need is to take a cell, grind it up, and purify not the enzyme that digests a sugar, but heredity. We need to get a pure tube of heredity. You can imagine that this wasn't an obvious thing to do, how you get a pure tube of heredity. The problem is you need an assay.

If you want to find an enzyme that digests a sugar, you have an acid. You purify different fraction, you make different fractions of a cell you see which fraction of the cell is able to digest the sugar. But if I make different fractions of the cell, it's not obvious how I figure out which fraction of the cell has heredity in it. And yet, that's exactly what was done, and that's today's lecture.

So purifying heredity. The discovery of the transforming principle. By the transforming principle, I don't mean an idea, like a principle. I mean a substance. This is an old medieval kind of word, a principle. It's kind of a Harry Potteresque kind of word or something like that of what is the transforming principle, the transforming substance. It's the kind of word alchemists would like to use, but it was actually what's attached to this, and it was called at the time the transforming principle, the transforming substance.

And it really is the work in 1928 of a young scientist F. Griffiths in London. Griffiths

was particularly interested in studying the bacteria pneumococcus. Why was Griffith so interested in studying the bacteria pneumococcus? Well, not so long before in 1918, there had been the terrible Spanish influenza epidemic that had killed millions of people around the world, the worst flu ever.

Millions of people died by this flew, and one of the reasons they died when they have the flu was they got pneumococcal infections. Griffiths was trying to make a vaccine against pneumococcus, a pretty good idea trying to make a vaccine against pneumococcus. Pneumococcus highly virulent stuff, but particularly if you're compromised by the influenza.

So what did he do? Well, Griffiths had a strain of pneumococcus. He didn't, by the way, infect people with it. He infected mice with it, that being considered a somewhat more ethical way to do the experiment. So he had a strain of pneumococcus that had a smooth, glistening coat. When you looked at it, it was smooth and white and glistened, the colonies that it made smooth, white, glistened. And it was virulent.

If you inject it into a mouse, the mouse got pneumococcus infection, and it died. It turns out we now know that it has a beautiful polysaccharide coat around it that provides resistance to the host's immune system, et cetera. He also had a strain of pneumococcus that had a rough coat. It was not glistening. It was kind of dull looking, and it was non-virulent.

He injected it into a mouse, mouse lives. It happens to be the case that we now know that it had a mutation in a gene that produced that coat, but that doesn't much matter. It didn't produce that polysaccharide, and therefore, was more easily fought off by the immune system.

So Griffiths does the following experiment. He takes his smooth, virulent bacteria, he injects it into a mouse, that's a mouse. And what happens to the mouse?

**AUDIENCE:** Dies.

**PROFESSOR:** Dies. Exactly. That's a dead mouse. And one of the easier assays in molecular

biology is the feet up, feet down assay. All right. Dead mouse. Now, what does he do? He takes the rough, non-virulent, he injects it into a mouse, and what happens? Lives. Right? Lives.

Then what he does, he takes the smooth, virulent and he bakes it in an autoclave. He heat kills this bacteria. Now, this heat-killed, dead, virulent bacteria when injected into a mouse, what happens to the mouse? It's alive. The mouse is fine. The bacteria was dead.

And then he does the following truly weird experiment. He takes the absolutely harmless, rough, non-virulent bacteria but alive, plus the smooth, virulent bacteria that has been killed by heat, heat-killed. The rough stuff is harmless. The smooth heat-killed stuff, harmless. Both are harmless.

We've shown the mouse can live with this injected. It can live with this injected. He injects it into the mouse, and what happens? Dead mouse. Very surprising. Not only that, when he takes the blood of the dead mouse, he can culture out of it. He can culture on a Petri plate from this dead mouse live, smooth, virulent bacteria.

How did that happen? Somehow, we didn't have any live, smooth bacteria. We had dead, smooth bacteria. We had live non-virulent bacteria. Somehow, the dead stuff transformed the live harmless stuff into virulence. It transformed it. The substance, the unknown substance that transformed it was referred to as the transforming principle or transforming stuff.

And now we have biochemistry because we have an assay. We could take the dead, virulent bacteria and break it up into fractions and see which substance, which fraction, is it a protein? Is it a carbohydrate? Is it nucleic acid? Is it something, is the transforming substance and purify heredity because we have seen the transfer of heredity to the harmless bacteria. There is an assay. The minute there's an assay, you could do biochemistry.

Now, the problem was the assay was painfully slow. You had to grind up the dead bacteria, you had to demonstrate by mixing it together, putting it into a mouse,

waiting months, and then you had to get a further sub fraction. It was just painfully slow. All this mouse stuff and all that.

And Griffiths didn't make much progress through the 1930s, but he kept going at it. And I suspect might have gotten there except for the fact that in 1941, his lab was hit by a German bomb during a blitz, and he died. And so Griffiths never saw the result of this, but he did purify fractions and all that.

But thank goodness others picked it up. He was again, Griffiths was a great guy. He worked in World War I, worked during World War II on important public health problems, and really lays this foundation, but never really purified what the final substance was because he died in a bombing.

But then you get in 1943, during World War II, folks working in New York City at Rockefeller University, then Rockefeller Institute, Avery, McCarty, and MacLeod continue this work, and they do it without the mouse. Because what they do is they grind up the virulent stuff and they sprinkle it on the living stuff, the living bacteria and played it out, and just looked for the appearance of colonies that are transformed.

Skip the mouse. Skipping the mouse makes it a lot easier, because you can do those experiments pretty quickly. Bacteria grow quickly. Same basic idea. Purify the stuff from the smooth dead stuff, grind it up, put it in different fractions, apply them, and then sprinkle them on a plate, and look for the occasional colony. And now you're just going to look for a fraction of the material that has the capability to produce some smooth colonies.

Well, they did that, and they purified it, and the purified it, and they purified it, and they purified it, and they eventually found that the particular type of molecule that they purified appeared to be DNA. But when they purified fractions containing DNA, these fractions had the ability to transform. Wow. You might immediately say, that's the transforming principle, DNA. That's the transforming substance.

What do you think the reaction was? Skepticism. First, it should be noted it's 1943.

People were busy at the time, right? We're in the middle World War II. It wasn't exactly top on people's minds, but there was enormous skepticism scientifically of those people who did follow the work. Why?

Because the one thing they knew was that DNA was truly a boring molecule. It was understood by all smart people that DNA was an incredibly boring structural molecule that had none of the fascinating diversity and richness of proteins.

Proteins could do zillions of different things. DNA, you know, it's just scaffolding. Why? What is the structure of DNA? So let's turn to the structure of DNA to see why it is that people were not impressed. Of course, when people are not impressed, you purified something and you show it transforms, what do you say to Avery, McCarty, and MacLeod?

How do you express your skepticism? You say, it's very good. You've purified this and it contains DNA, but is it absolutely totally 100% pure or is it possible that you've carried along in the fraction that you have purified some other trace quantity of a highly potent protein that is really causing heredity?

And of course, that's the problem is you can never prove that there's not a teeny smidgen of something in there. You can only show how pure it is, but you can never rule something out. So when people want to sort of dis your biochemistry experiments, it's always easy to say, it was probably something else in there too you just don't know about it. And that was what the answer was to them. But let's look at the structure of DNA.

So DNA has three important components which we need to learn. A, it has a sugar called 2 prime deoxyribose. So ribose is a 5-carbon sugar. A five-part carbon sugar is a pentose. So this is a sugar, in fact a pentose, pentose of course five, pentose meaning it's a 5-carbon sugar, but it lacks a hydroxyl group.

So it's just slightly different from the 5-carbon sugar. And we draw it in this configuration where there is the 1 prime carbon here, the 2 prime carbon here, the 3 prime carbon here, the 4 prime carbon here, the 5 prime carbon here. That's very

important to know. We've got an oxygen up here. Here, we have an OH and an H. Here, we have an H and an OH. Here we have H. Here we have our OH, H, H.

But here we should have a hydroxyl off every carbon, and we don't. Only here are deoxy. That's the only difference from this being a perfectly normal ribose, deoxy at the 2 prime position. Big deal. Now, the next component of DNA that you need to know about are these nitrogenous bases. So hanging off our ribose is a base.

This base has carbons, oxygens, hydrogens, and nitrogens. And they come in four flavors, adenine, A, guanine, G, thymine, T, cytosine, C. A, T, C and G, and we'll look at their structure in just a moment. Then the next important part if we look at this conceptually, is that hanging off here, we have a triphosphate.

We have a triphosphate. So this is the monomer for making DNA, triphosphate. We have a sugar, the sugar in exactly the same place off the 1 prime carbon there has a base, off the 5 prime carbon, we have a triphosphate.

What's the triphosphate good for? Energy. We're going to do a polymerization, and that's going to contribute the energy for the polymerization, and that's pretty much it. That's the way to think about DNA. So when we do our polymerization now, we polymerize and we get base, CH2, phosphate. And then coming down this way attached here, we have our phosphate, and that attaches to the 5 prime carbon here, and onward that way.

So notice that our polymer goes from a 5-prime carbon here, 3-prime carbon here, 5-prime carbon here, 3-prime carbon there. And we go sugar, phosphate, sugar, phosphate, sugar, phosphate, sugar, phosphate, 5-prime attachment, 3-prime attachment, 5-prime attachment, 3-prime attachment. That's DNA. Pretty boring.

The same sugar, same phosphates strung together, totally boring. The only difference are these bases. And there's only four of them, and they're not very impressive. They're pretty boring, these bases. There are purines. The A and G are purines, and their ring structure looks like this. This is six-membered ring and there's a five-membered ring. There are pyrimidines, T and C, and they just have a

six-membered ring.

They've got carbons, and oxygens, and nitrogens, and hydrogens, and they don't differ really in their charges. By compared to the amino acids, positive charges, negative charges, hydrophobic groups, sulfurs that are reactive. Amino acids, that's impressive. Those 20 different side chains have wildly different chemical properties. These form measly bases, have essentially, the same chemical properties.

There's nothing very different about their chemical properties and therefore, all smart right-thinking people recognize the DNA could not be a particularly interesting molecule. It had to be largely a structural molecule of some sort.

So when Avery, McCarty, and MacLeod tell us ah, the transforming principle of DNA, nobody's impressed. But of course, it's World War II. People are busy. Lot of things going on. And not that long afterwards, not that long afterwards, another really important experiment gets done in the early 1950s, the Hershey-Chase experiment. Hershey is not the candy bar. It is Alfred Hershey and Martha Chase. Martha Chase and Alfred Hershey do a cool experiment.

People were studying something else at the time. They were studying the viruses that infect bacteria, bacterial viruses. So it turns out just like you may get a viral infection, E. coli gets viral infections too. It usually dies of them or at least often dies of viral infection, not necessarily usually, I take that back, sometimes dies of viral infections.

So that is a virus, actually, greatly magnified, glommed on to E. coli, virus E. coli. What happens is, if you mix the virus with E. coli, it gloms on, and then if you wait a little while giving it a happy medium to grow in, the E. coli some time later, half an hour later maybe, bursts open, dead, and spews out zillions of viral particles which could go on to infect new cells.

How does it do that? How does it instruct E. coli to make viral particles? It must be bringing information. It's having progeny. It is passing on heredity too. It has some transforming information. Where is the transforming principle in the little virus? It

gloms on to the cell somehow gives something into the cell, and poof, 20 minutes later, half an hour later, lots of viruses. Where's the information carried?

Now, this was a much simpler system. This system, you're asking what's in the bacterial virus. There's not a lot in a bacterial virus. It's not like a cell that have zillions of things. The bacterial virus is a pretty simple particle.

The bacteria virus consists of protein coat, proteins are on the outside, DNA on the inside. That's it. You don't have a lot to work with, a limited number of proteins, DNA in the middle.

These things just as an aside were thought to kind of like eat bacteria. Because they were thought to eat bacteria in a way by at least the early things, they're called bacteriophage, bacteriophage. The word phage means to eat. So you may hear me talking about bacteriophage, meaning eaters of bacteria.

Indeed, actually there was some nutty ideas in the 1920s and 1930s when bacteriophage were first discovered that the way to cure a bacterial infection was to drink a lot of bacteriophage. They would kill the bacteria. It's a thought. People actually tried these things. Anyway, it turns out not to be such a good idea.

So Hershey and Chase decided we're going to figure out which is it? Is it the DNA or is it the protein? How do you find out? Yeah?

**AUDIENCE:** [INAUDIBLE].

**PROFESSOR:** Put in only protein, and see what happens. So take the bacteriophage, purify it from protein verses DNA. I've got a pure component of the protein, I sprinkle it on, nothing happens. I take the DNA, I sprinkle it on, nothing happens. Neither works. Why is that?

**AUDIENCE:** [INAUDIBLE].

**PROFESSOR:** The shape, those little feet in the shape were critical for the pathogenicity. So when we grind up the virus, it doesn't work anymore. It's a great idea. If it worked, bingo, we'd have it, and that should be the first experiment we do because it's so easy. But

it turned out not to work. Yes?

**AUDIENCE:** [INAUDIBLE].

**PROFESSOR:** Put a chemical marker on the protein, put a chemical marker on the DNA, and see which one goes into the cell. What chemical marker? How are we going to attach a chemical marker to the protein without messing it up? We can't mess up the protein, right? It still got to function. How do we get a chemical marker on it?

**AUDIENCE:** [INAUDIBLE].

**PROFESSOR:** Sorry?

**AUDIENCE:** [INAUDIBLE].

**PROFESSOR:** So what chemical do you want me to put in? Well, how am I going to tell whether, DNA's got phosphorus. How am I going to follow the phosphorus?

**AUDIENCE:** Radioactive tag.

**PROFESSOR:** Radioactive tag. Bingo. What if I used radioactive tags, and I made a radioactively-labeled virus. How can I radioactively label the DNA?

**AUDIENCE:** [INAUDIBLE].

**PROFESSOR:** Sorry?

**AUDIENCE:** [INAUDIBLE]

**PROFESSOR:** A radioactive base. I could do that. What else could I do? Yup?

**AUDIENCE:** Phosphorus.

**PROFESSOR:** Phosphorus. Phosphorus has the nice property that phosphorus is in my DNA, but it's not an proteins. So what do I use? Phosphorus-32, P-32. So I use P-32, and how do I manage to chemically create a virus that has P-32 in it?

**AUDIENCE:** [INAUDIBLE].

**PROFESSOR:** Just throw it in the solution with P-32, and the virus will take care that itself, right? So simply grow virus for a while in the presence of P-32. Let's do that. So grow virus in a test tube with bacteria. Here's my bacteria. Here's my virus I've put in there, and let me put in P-32, and what I'll get is P-32-labeled labeled virus.

How do I label my protein? Someone said it already. What elements can we find that's in proteins but not in DNA?

**AUDIENCE:** [INAUDIBLE]

**PROFESSOR:** Sorry?

**AUDIENCE:** Sulfur.

**PROFESSOR:** Sulfur. Sulfur. S-35 is a radioactive isotope of sulfur, and if I grow it, I can S-35 label the proteins in my virus. Nice. This radioactive labeling trick is very cool. So I take it, I take some P-32-labeled virus where these P-32 was only in the DNA. I got some S-35-labeled label virus where the S-35 is in the protein. I could mix them together, now do my experiment, wait 20 minutes and, or even wait last 10, 15 minutes, and see which element has gone into the cell.

How do I do that? See I've got my cells here, and I've got the viruses attached to them, and they've injected something in here. They've either injected a protein or they've injected DNA. What was injected? I need to carefully go in there and remove the virus and look at just what's in the cell. I have to now separate the virus glommed onto the outside of the cell from the cell.

So do I use micro manipulator tweezers to pull off the virus?

**AUDIENCE:** [INAUDIBLE].

**PROFESSOR:** Well, if I denature, I might crack open the cell. Centrifuge it. If I centrifuge it, the whole thing will spin down. I need to kind of knock the viruses off the cell, physically. I just got to agitate it so I get them off the cell.

With enough kind of hydrodynamic agitation, the viruses fall off. So a device was created that was able to just perfectly knock the viruses off. It's referred to as the Waring kitchen blender.

It turns on your kitchen blender is perfect for this. Take the viruses, add it to the bacteria, sit for a little bit, put it in your kitchen blender, press puree. And let's say on puree setting, the viruses fall off, and now you can spin it in a centrifuge, the bacteria are denser, they come down. The viruses are lighter, they stay in the supernatant, and you can take the supernatant and the pellet at the bottom over your radioactivity counter and see which one is in the bacteria.

These were referred to as the famous Waring blender experiments. They really are, actually. So you put this in the Waring blender, you knock off the viruses, you spin it down, and what happens is after you've done it, there's a pellet here of the bacteria that are spun down in the centrifuge. The virus particles are still up here. We take this pellet over to our counter, and which element do we find in great abundance, S-35 or P-32?

**AUDIENCE:** P-32.

**PROFESSOR:** P-32. The DNA is what's going in. Bingo. Nice experiment. Now if you were churlish, couldn't you say, yeah, look it's mostly the DNA, but there's a little smidgen of protein maybe, that came along too. Do you think they found absolutely zero S-35 in there? No, because they don't perfectly knock the virus off. Some of it kind of sticks. There's 1% S-35.

And if you're being really churlish about this you would say I still don't believe you. But now you have it from two different directions. You have it from the pneumococcus, this bacteria experiment from Avery, McCarty, and MacLeod Hershey and Chase coming from two different systems.

They're giving you the same answer. It's pretty clear. It's in the air. People know DNA is the stuff. They're believing it now. DNA is the stuff. But how does it work? How can this dumb molecule possibly be the transforming principle?

Well, smart, young people want to know. So an erstwhile ornithologist, that is a college kid from the University of Indiana who particularly liked bird watching got very enamored by this problem, actually based on some fabulous faculty at the University of Indiana. He got really intrigued by how could DNA possibly do this.

But he recognized he didn't know any chemistry. He decided to go to Cambridge, England to the Medical Research Council lab, the MRC lab in Cambridge, England where he teamed up with someone who did a lot of talking and very few experiments. A physicist who had worked for the British admiralty during World War II on classified things and had somehow gotten interested in biology.

And because he had this kid, this recently graduated college kid, and you had this 35-year-old physicist who nobody was quite sure what to make of, they kind of hung out with each other in the same office. And they didn't really do many experiments, but boy did they do a lot of talking, and thinking, and looking at all the data that were out there.

And that's pretty much what James Watson and Francis Crick were doing. They knew this problem was important. And Jim and Francis would talk every day about this stuff, and they will talk to people down the hall who really knew about the chemistry of nucleic acids. And they went down to London to Maurice Wilkins' lab where crystals were being made of DNA. And Rosalind Franklin, who was a fantastic scientist and had managed to make crystals of DNA, showed Crick and Watson her crystals of DNA.

Francis Crick being a physicist was very good at understanding crystallography and how crystal structures and x-ray diffraction patterns related to each other. And Francis knew immediately this thing had to be a helix. He could tell it was a helix. And they went back, and based on Rosalind Franklin's x-ray diffraction patterns, went and made a model, a model for the structure of DNA.

You all know the model. You've seen the double helix forever. It's a cultural icon, but that's how this came about.

And *The Double Helix, the Structure of DNA,* April of 1953 is published. The double helix has two strands running an anti-parallel directions, 5 prime to 3 prime, 5 prime to 3 prime anti-parallel directions, and it has a perfect base pairing between purines and pyrimidines.

If you have a T, and I'm just going to draw this very quickly. You can look in your book for getting it just right. You have two hydrogen bonds. That's T and A, and if you have a C, you have three hydrogen bonds that perfectly hold it to the G, et cetera.

So notice C and G fit perfectly together to make three hydrogen bonds. A and T fit perfectly together to make two hydrogen bonds, and that was the key was to recognize that when you stick them together in that way, you get exactly the same width. They fit perfectly. You couldn't match an A with a G, an A with a C, you could only match the A with a T. That was it. Brilliant. Beautiful.

Now, you guys should read Crick's book *The Double Helix* in which he tells the stories because it's just a fascinating, fascinating business. He'll tell, or others will tell actually, the story. So you know what this means by the way? This means that the amount of A should equal the amount of T. And the amount of G should equal the amount of C.

There should be a ratio, a one-to-one ratio that the A to T ratio should be one to one. And the G to C ratio should be one to one. Although any organism might have more As than Ts and Gs than Cs, the ratio of these guys should be one and these guys should be one. This actually was discovered by a chemist at Columbia called Chargaff. These were called Chargaff's rules.

Chargaff was a very distinguished chemist who came up with Chargaff's rules with the As equals the Ts, and the Gs equals the Cs, and he didn't know what to make of it. By the way, Chargaff actually visited Cambridge, England while Crick and Watson were there before their discovery, and he had lunch with them. And he related that Crick and Watson seemed like Bozos to him, because they couldn't even keep straight the exact structure of the four bases. They always had to keep looking it up.

They hadn't memorized the structures the four bases, and Chargaff was such a brilliant chemist, he, of course, knew this instantly, et cetera, et cetera. And he said, these guys are never going to get anywhere because they really don't even understand the structure of the bases, haven't memorized it. When Crick and Watson's discovery turned out to be the single most important biological discovery of the 20th century, Erwin Chargaff who lived a very long life was sort of bitter because he's kind of worked it out in a way with the ratio and never figured out what it meant.

And he said one of the bitterest, cuttingest comments I've have ever heard from a scientist which is referring to Crick and Watson as still not being impressed even after they won their Nobel Prize for this. He said that two such pygmies should cast such giant shadows only shows how late in the day it is. Anyway, he was not happy to have missed this point.

Crick and Watson were very happy to have figured this out. They, when they figured this out in February of 1953, what do you do in England when you make a big discovery?

**AUDIENCE:** You have tea.

**PROFESSOR:** No, you don't have tea. You go to the pub. They went to the pub. They ran down to the Eagle Pub, they brought everybody drinks, and they told everybody at the Eagle Pub, we've discovered the secret of life. The people at the Eagle Pub had no idea what they were talking about, but were happy to have a round of drinks, and there you go.

They immediately raced to write this up in *Nature.* It appears in *Nature* in April of 1953, and it's a one-page paper. And get it on the web and read it. It's the single best one page that has been written in biology in the 20th century.

And of course, what did they say? The title is kind of unassuming, "A Structure for the Salt of Deoxyribonucleic Acid," nothing too exciting. But Crick and Watson realized something. What do they realize? Crick and Watson realized why is this a

big deal? Why is this double helix so important?

Well, the implication of the double helix is that if I have a double helix and those strands were to separate, each would be a template for a double helix, heredity. How do you pass information to two daughter cells? You got a double helix. It's redundant. If you know the A is on one strand, you know the Ts on the other. Unzip it, copy, voila. I now have two copies of heredity.

What's a mutation? Occasionally get it wrong. Bingo. They saw it. They knew. Now, they didn't have time to prove this. I mean, who has time to prove this. This is such an exciting discovery secret of life. Drinks for everybody, they write it up, but in the last paragraph, they certainly don't want anybody to think that they missed the point.

And they write the coyest sentence in molecular biology. They write, "It has not escaped our notice that this structure offers an explanation for heredity and mutation. We'll address this in another paper." Cute. Very cute. They put down their marker, they knew what it meant, but they got the thing off to *Nature* very quickly.

It was a hot topic. They were competing with other people. You'll read about the competition with Linus Pauling and other things like that. It had not escaped their notice that this pretty much explains heredity.

Now, of course, are you convinced that it explains heredity? It's a nice model, but don't we require proof? We do require proof. It's an ex post facto model, we need proof, It's a pretty good ex post facto model, but we need proof. So the last step, which I'll touch on very briefly, was proof of what's called semi-conservative replication. Meaning that each strand is used for the other.

And I might run two minutes over. We'll see. I'll try to keep it within time. Out at Caltech, two graduate students, Frank Stahl and Matt Meselson hear of this. Matt Meselson by the way is still working in Harvard Square. He's at Harvard. He's a wonderful guy. Matt is there. You could ask Matt about this, young graduate student at Caltech in the early '50s.

Obviously, this model looks like it must be right. How do you prove it? Well, Matt and

Frank, Meselson and Stahl came up with a cool experiment to prove it. Meselson and Stahl, they take bacteria, here's my DNA in there, they want to show that each strand, each, when we make a new generation of bacteria, the new DNA has one old strand and one new strand. That each old strand is being used as a template for a new strand.

How are we going to tell? We gotta label it somehow. We got a label the old strand different than the new strand. How do we possibly label and they're the same chemical composition? What are we going to do?

**AUDIENCE:** Radioactivity.

**PROFESSOR:** Radioactivity or some kind of isotope. Well, they used an isotope. What they did, super cool, they grew this up, but not in normal nitrogen, but in N-15, not, by the way, radioactive, but different weight. They grew it up in N-15. They added bacterias that had grown up, and all of their DNA had lots of N-15 in it on both strands. They then pour in a lot of medium that has N-14. Tons, they swamp it with N-14 so what's this strand going to be? N-14.

So what can you tell me about the difference between this DNA and that DNA?

**AUDIENCE:** One's going to be lighter.

**PROFESSOR:** One is going to be lighter. How do you measure how much lighter it is? They came up, they invented the technique for this purpose of centrifuging in a salt gradient. They put in the right amounts of salt, cesium salt, and they centrifuge it so hard that there's a gradient of densities, denser here, lighter here. And they find that this DNA and this DNA centrifuge to different places. There's a difference in their density.

The new DNA is half old, N-15, half new, N-14. It has the intermediate density. If I grow another generation, I'm going to see some N-15 14s, and I'm also going to see some N-14, 14s. And that's what they found. They invented this technique it's isopycnic centrifugation and Matt Meselson and Frank Stahl provided an experimental prediction of the beautiful Crick-Watson model that was fair to say the

secret of life.

Anyway, these are the foundations. Notice now, we've taken genetics and used it to do biochemistry. We've taken biochemistry and used it to understand genetics. And so finally, we are making the bridge of molecular biology. Next time.