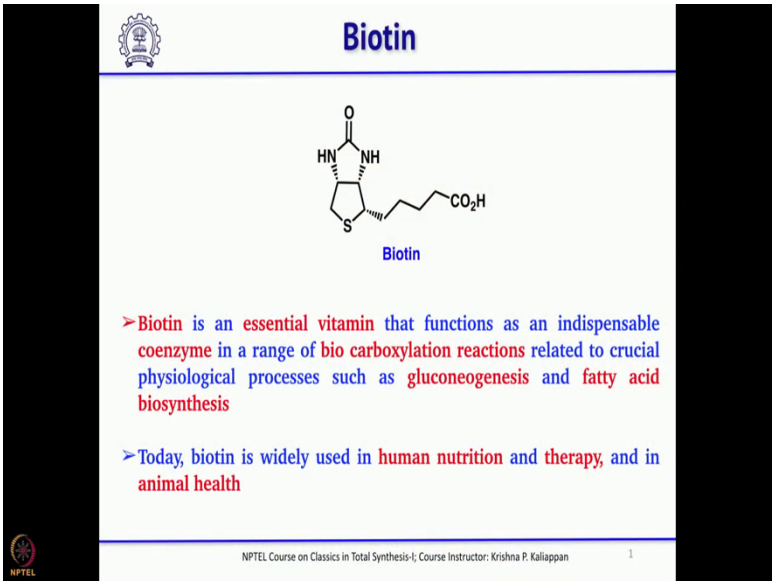
Classics in Total Synthesis-I
Prof. Krishna P Kaliappan
Department of Chemistry
Indian Institute of Technology, Bombay

Lecture - 12
Biotin and Lactacystin (i) Corey, ii) Baldwin)

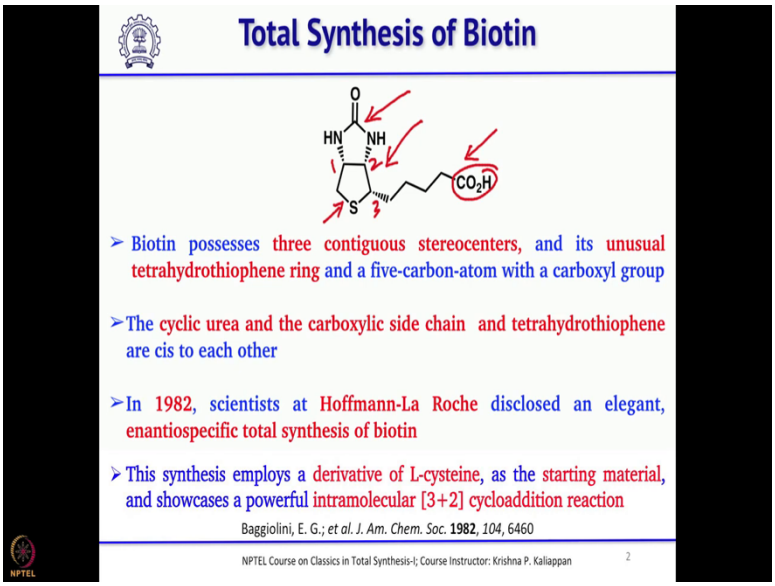
So, now, we will move to the synthesis of another important vitamin called biotin.

(Refer Slide Time: 00:26)



This biotin plays a very very important role in physiological process like gluconeogenesis and fatty acid biosynthesis. So, obviously, a lot of interest was there in early 60's, 70's, 80's to synthesize this biotin.

(Refer Slide Time: 00:49)

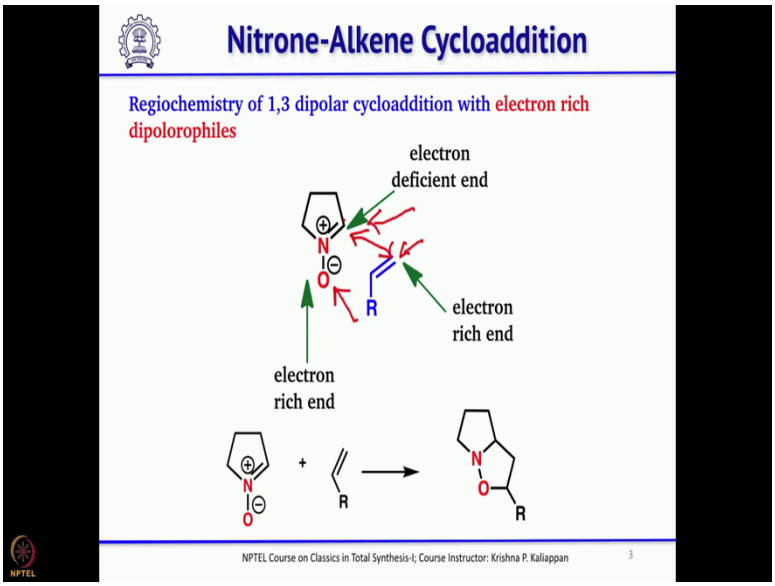


So, today we will talk about one total synthesis of biotin and when you look at this molecule immediately you can see that there are three contiguous stereocenters. How, 1, 2, 3; these three are chiral centers and they are contiguous and there is one tetrahydrothiophene ring ok. You can see completely reduced thiophene ring and there is a five-carbon side chain having carboxylic group at the terminal end.

Then you also have a cyclic urea ok, you have a cyclic urea then if you look at the whole biotin the difficult part is the carboxylic acid and these three chiral centers. How you are going to introduce these three chiral centers stereoselectively; so, that biotin can be made in naturally occurring form.

So, the first synthesis was reported by Roche group and here they used a very interesting intramolecular 3 plus 2 cycloaddition of a nitrone and an alkene. They started with naturally occurring amino acid cysteine, actually they use a dimer of cysteine cystine and they tried this intramolecular 3 plus 2 cycloaddition reaction.

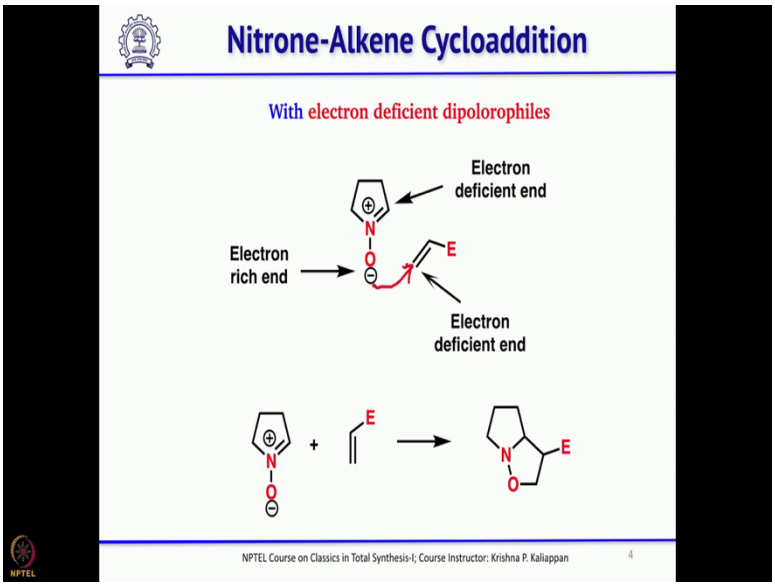
(Refer Slide Time: 02:23)



So, what is this 3 plus 2 cycloaddition between nitrone and alkene? This nitrone when you look at nitrone, nitrone is nothing but if you have an imine and if the nitrogen is oxidized ok. The oxide of nitrogen ok, normally you know we talk about N oxide, N methyl morpholine N oxide, trimethylamine N oxide. So, here nitrogen of imine if it is oxidized then that is called nitrone.

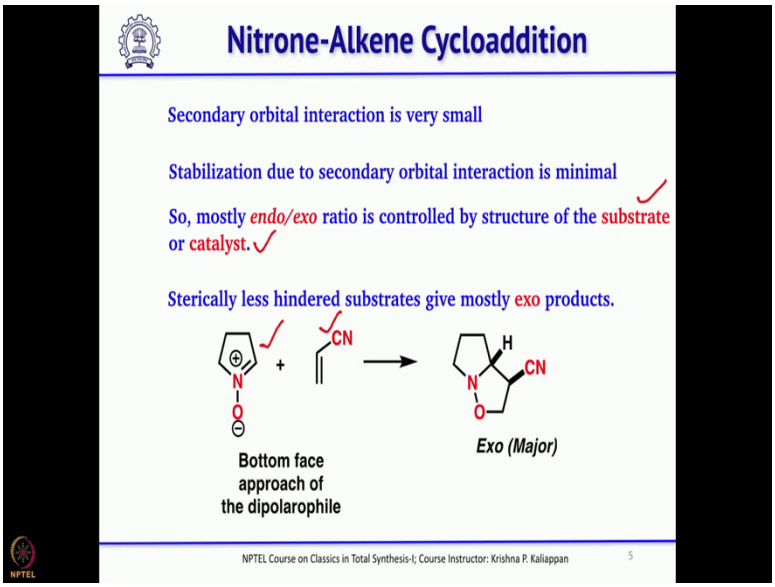
And this nitrone if you look at carefully it has one electron deficient end and one electron rich end; so, it is a distributed over three atoms ok. So, when you do a 1, 3 dipolar cycloaddition with an alkene and if the alkene is electron rich, then this is what the stereo regio chemistry of the product which you get ok. So, obviously, this is electron rich and this is electron deficient; so, you can see the bond forming between these two carbon atoms.

(Refer Slide Time: 03:35)



Likewise, when you use electron deficient dipolorophiles ok, then what will happen? This is electron-rich and this is electron deficient. So, this is where the bond will form and the regio chemistry will be reversed ok.

(Refer Slide Time: 03:54)



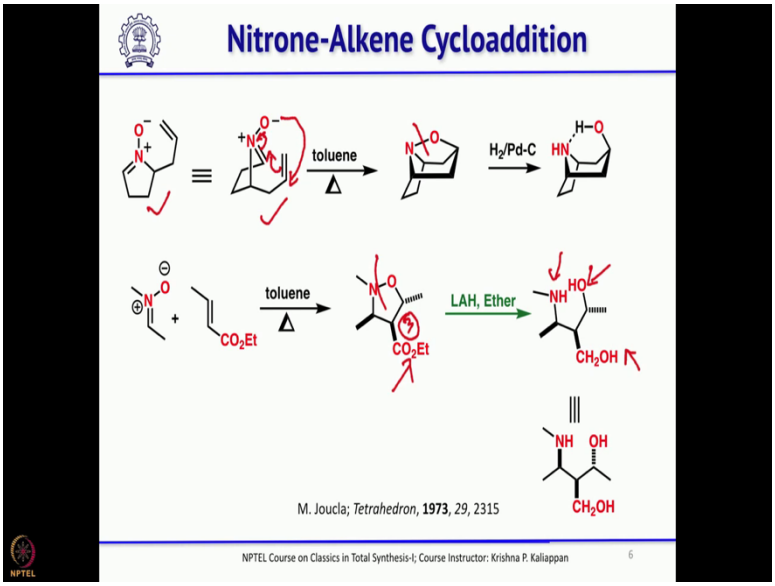
So, this nitrone-alkene cycloaddition has been successfully used to make five-membered ring, which you can call it as isoxazolidine ok. But contrary to 4 plus 2 cycloaddition that is our Diels Alder reaction. Here, the secondary orbital interaction is not there or it is very very minimal. As you know, when you talk about 4 plus 2 cycloaddition or Diels

Alder reaction, the secondary orbital interaction is important for getting endo isomer as the major product.

Since this nitrone-alkene cycloaddition that is 3 plus 2 cycloaddition does not or involve very little amount of secondary orbital interaction. You do not have to get only the endo product as the major product; in fact, you get exo product as the major product. And the formation of exo or endo is mainly controlled by your substrate or if you are using a catalyst.

Mostly it gives the exo product and if you take this cyclic nitrone and treat with dipolarophile like acrylonitrile, then you get this as the major product, this is nothing but exo product. And here this is a bottom phase approach of the dipolarophile; that means, you have the nitrone like this and your dipolarophile attacks from the bottom ok, to give this as the major product.

(Refer Slide Time: 05:21)

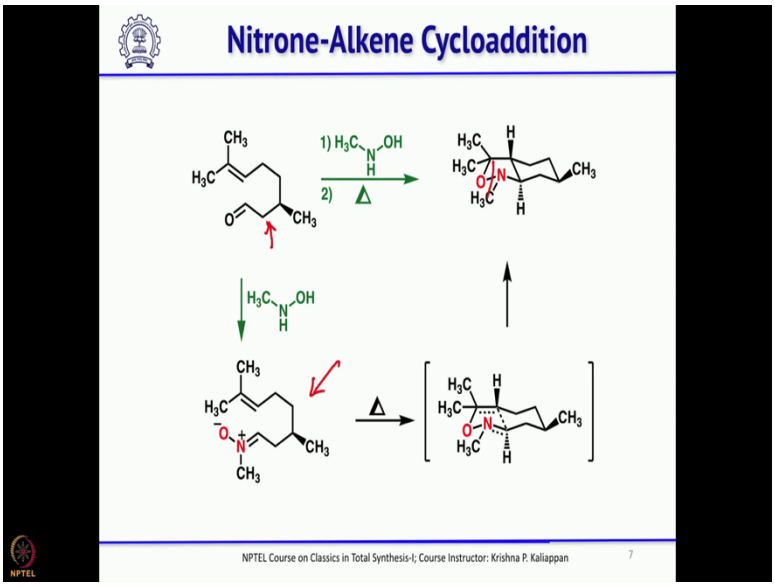


And there are several intra-molecular 1, 3 dipolor cycloaddiction between a nitrone and alkene present in the same substrate is reported for example. You can see this molecule this can be redrawn like this. Now, this O minus will attack here and this double bond will attack and the positive charge on the nitrogen will be neutralized to get this tricyclic ring ok.

And this is a isoxazolidine and this N-O bond can be easily cleaved with zinc and you can also cleave it with hydrogenation condition, you can also cleave it with LAH; so, that will give you amino alcohol. And this is another example, this is intermolecular and still you can see there are 3 chiral centers ok, 3 chiral centers one can fix during this 1, 3 dipolar cycloaddition depending on the nature of your substrate ok.

Now, again if you cleave this NO bond ok; so, that will give you this amino alcohol and when you use LAH not only the NO bond gets cleaved, but also the ester. So, you get two hydroxyl group, one primary and the other secondary in addition you get a secondary amino group ok, and this also can be rewritten like this.

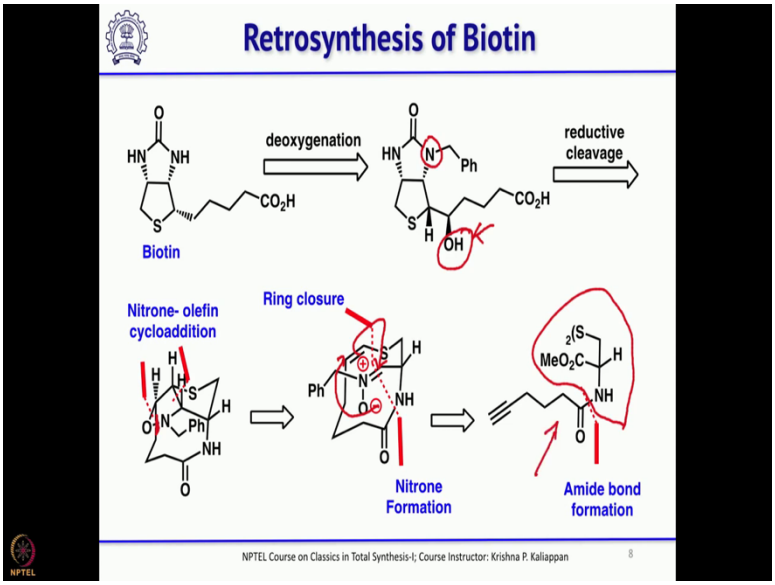
(Refer Slide Time: 06:57)



So, I also will show you another example of intramolecular nitrone alkene cycloaddition, because this is important so that you will understand the 1, 3 dipolar cycloaddition which is involved in the synthesis of biotin as the key step ok. So, this starting material is called citronellol commercially available. This on treatment with methyl hydroxylamine, it forms a nitrone and the nitrone undergoes 1, 3 dipolar cycloaddition and it goes via this nitrone.

You can see this can be drawn like this, a chair-like transition state you can write and followed by formation of this five membered ring. Again, you can cleave this you will get corresponding amino alcohol.

(Refer Slide Time: 07:43)



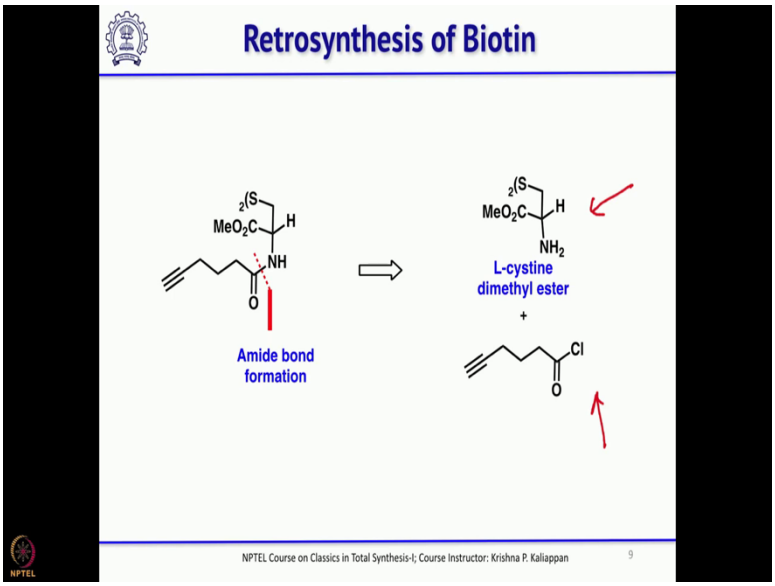
Now, let us see how this biotin was synthesized what was the retro synthesis by Roche group. So, what they did was, they introduced an extra hydroxyl group in biotin that was the first retrosynthetic. You should not call it as disconnection, but as introduction of additional functional group required for better disconnection. So, why they introduced a hydroxyl group? Because, they wanted to use a 1, 3 dipolar cycloaddition.

As I said when you do a 1, 3 dipolar cycloaddition between nitrone and alkene, you get isoxazolidine and if you cleave the NO bond you get amine and alcohol. So, that is why they put deliberately put this hydroxyl group so that this hydroxyl group along with this nitrogen you know it can form during the dipolar cycloaddition. So, then they said if you can look at this complex structure ok.

So, this is formed if you cleave this NO bond if you cleave this NO bond you will get this ok, but this isoxazolidine can be obtained from the nitrone, this nitrone and the double bond ok. Now, this O minus will attack here and the double bond will attack; so, it will form a five membered ring.

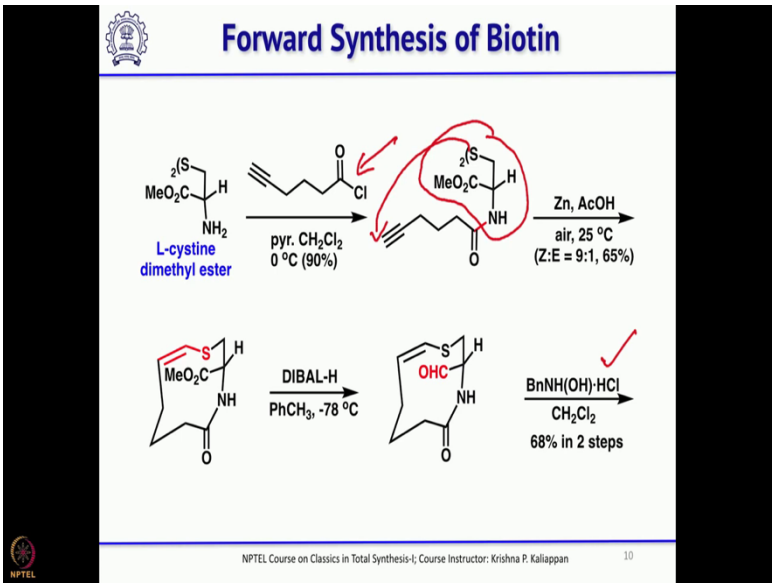
And this nitrone I leave it for some time so that you can visualize. Any of you can discuss more when we talk about the total synthesis. Now, you just see can be obtained from this particular compound and which can be made from commercially available amino acid called cystine ok. And from cystine you can isolate with this carboxylic acid.

(Refer Slide Time: 09:41)



So, basically what you are doing is, you are going to start with L-cystine methyl ester and this acid chloride which you can make it in three to four steps, and it is a known compound ok.

(Refer Slide Time: 09:55)

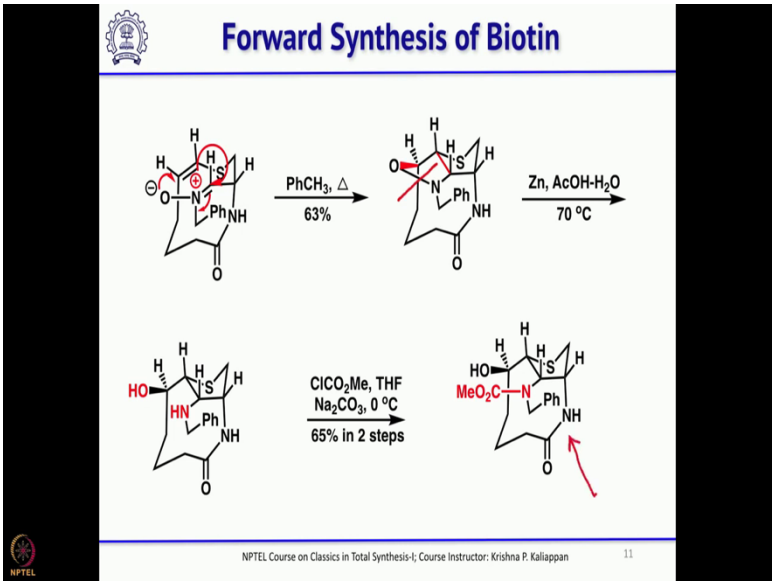


So, now let us see how the Roche group synthesized biotin; so, they started with L cystine dimethyl ester then isolated to get this compound, ok. Then the this is the dimer, you should know that this is a dimer S-S and the dimer; so, you can cleave the S-S bond with metal. So, for example, if you use zinc and acetic acid that S-S bond will cleave and

then you will get corresponding SH; the SH spontaneously will add to the triple bond, SH will add to the triple bond and you will get ene thioether ok.

Then what you do? You have to reduce the ester to aldehyde because aldehyde is required for making nitrone ok. So, you reduce the ester to aldehyde in one step with DIBAL then you treat with benzyl hydroxylamine ok. So, when you treat with benzyl hydroxylamine this aldehyde will form a nitrone ok, you can see the nitrone now.

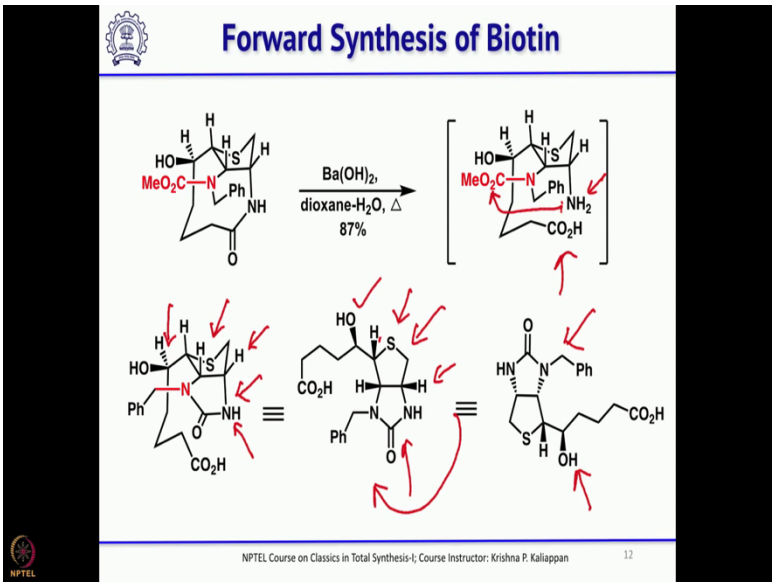
(Refer Slide Time: 11:03)



Now, once you have this nitrone this undergoes an intramolecular 1, 3 dipolar cycloaddition that will give you the corresponding isoxazolidine ok. So, I leave it for a few minutes so that you know you can understand how these five membered ring is formed. Already I put the arrows properly, so, it will be easy to understand. Nevertheless from the stereo-chemical point of view I leave it for 30 seconds so that you know you can understand.

Once you get this isoxazolidine, obviously, the next step is the cleavage of the N-O bond ok. So, you cleave it with zinc and acetic acid, you get the corresponding amino alcohol and the amine is already protected as N-benzyl and you get the free hydroxyl group. So, you have to protect the amine once again; so, that was done with chloromethyl formate in the presence of base like sodium carbonate; so, now, the amine is fully protected. Then you can hydrolyze this amide ok.

(Refer Slide Time: 12:22)



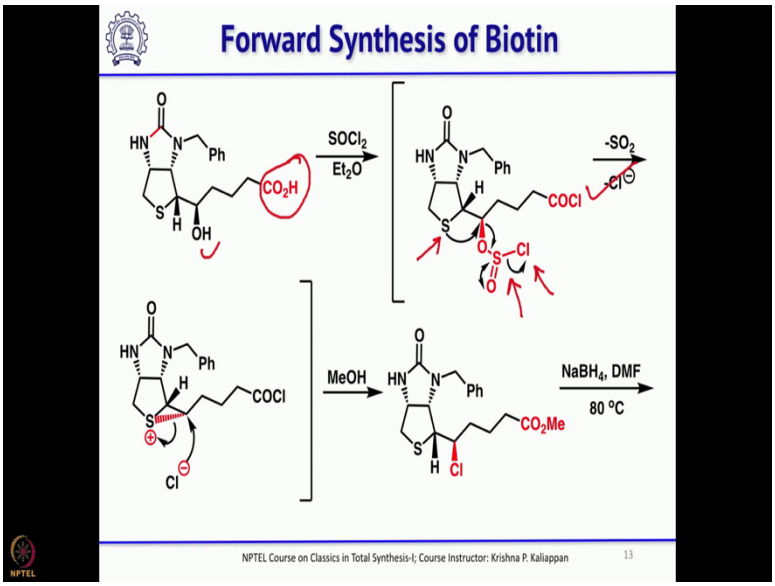
The hydrolysis of this amide was done with barium hydroxide in refluxing dioxane water to give the corresponding carboxylic acid and amine ok. The amide was cleaved. Once this amino acid is formed then what happens? The lone pair on the amine attacks this carbamate.

Then OMe comes out. That leads to the formation of this urea derivative cyclic urea derivative ok. This NH_2 attacks the carbamate carbonyl, OMe comes out and you get the corresponding the urea derivative cyclic urea derivative. Can you redraw this as this bicyclic compound?

You can see first the cyclic urea, yes, you have done the cyclic urea. Then, attach this tetrahydro thiophene ok. You can see both the ring junction hydrogen are beta; so, you have written beta. And next you have the five-carbon side chain, the five-carbon side chain with a hydroxyl and carboxylic acid ok. The same thing if you rotate it by 180 degree rotate it by 180 degree you will get this, rotate it exactly by 180 degree.

So, the thiophene will come down and the urea the tetrahydrothiophene will come down, the cyclic urea will go on. So, if you look at this structure, now what is missing or what you do not want? So, there are two things you do not want. One, you do not want this hydroxyl group. Two, you do not want this benzyl group. So, if you can remove the hydroxyl and benzyl group that will lead to the formation of biotin ok.

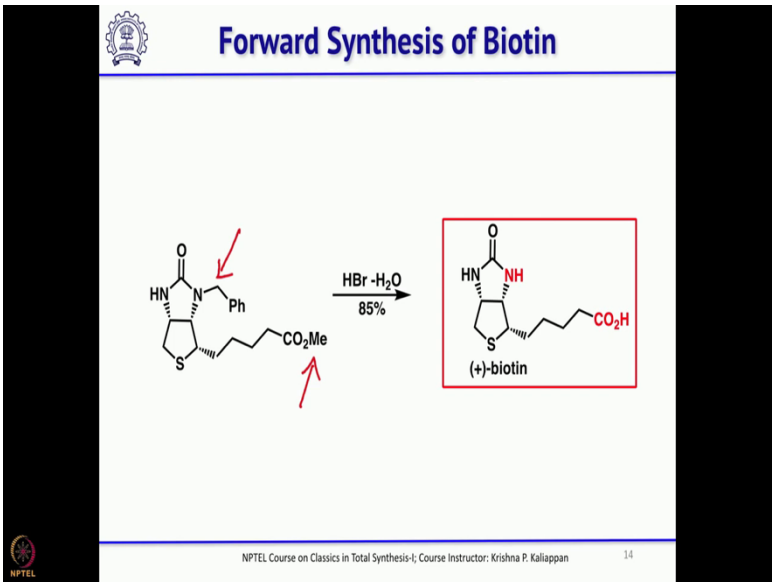
(Refer Slide Time: 14:32)



So, how it was done? You first try to remove the hydroxyl group. When you treat with thionyl chloride first the carboxylic acid will become acid chloride ok. Then that hydroxyl also will attack the thionyl chloride and it forms the half thionoester ok. So, this you have a lone pair on the sulfur of tetrahydrothiophene that will intramolecularly attack and your OSOCl will go out.

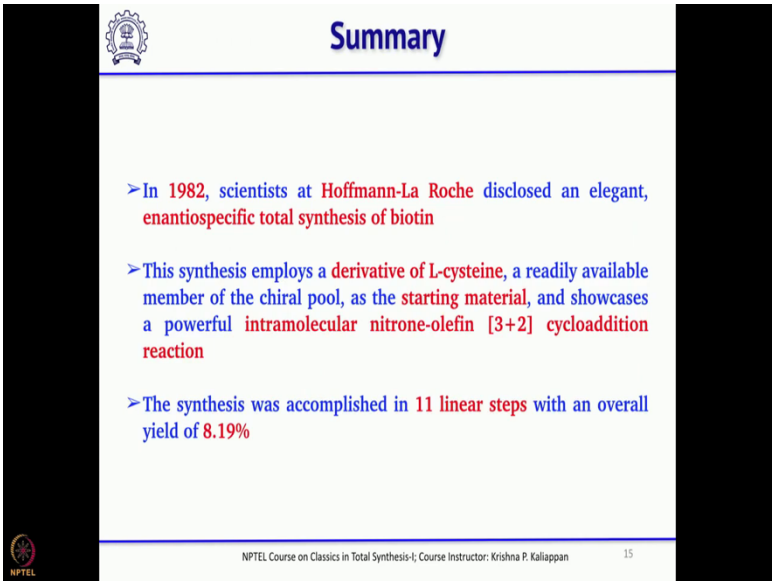
So, that will form a three membered ring with sulfur having a positive charge. Now, what will happen? The chloride which comes out, ok. So, now, if you see here this SO_2 will come out which is the neutral molecule, the chloride which comes out again it will attack. So, it is a double SN_2 reaction on that carbon; so, you get again the beta chloride ok.

(Refer Slide Time: 15:41)



Now, the chloride can be removed with sodium borohydride and HBr water not only hydrolyzes the ester, but also removes the benzyl group to give biotin ok.

(Refer Slide Time: 15:57)

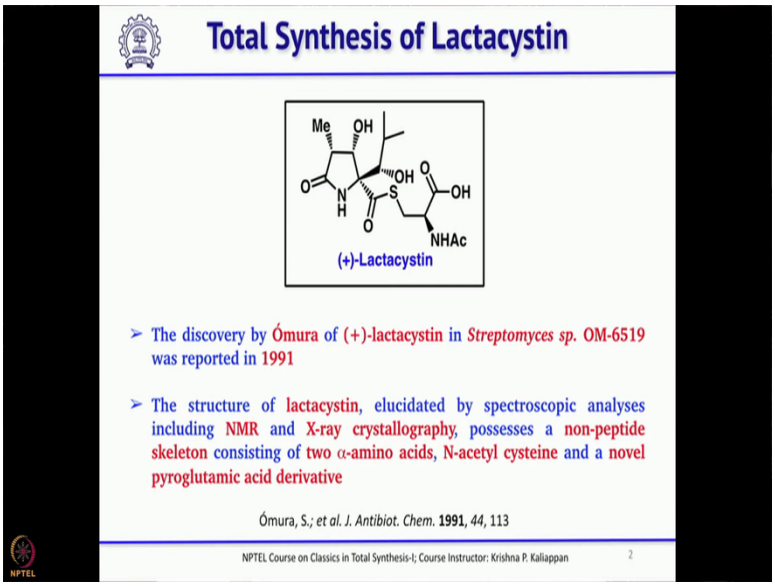


So, to summarize this in 1982, Roche group disclosed the first enantioselective total synthesis of biotin and they use the amino acid L-cystine which is a dimer of L-cysteine. And they used an intramolecular nitrone olefin cycloaddition to fix the stereochemistry of nitrogen and the hydroxyl group, the hydroxyl group as you know it has to be

removed. But, nevertheless to fix the stereochemistry of CN bond they use the intramolecular nitrone olefin cycloaddition reaction.

Overall, this synthesis was done in 11 longest linear steps and the yield was impressive of about 8 percent. Even though the molecule looks small overall yield of 8 percent is considered a decent one for such molecules ok. So, now, we will move to the synthesis of other natural products having five membered ring.

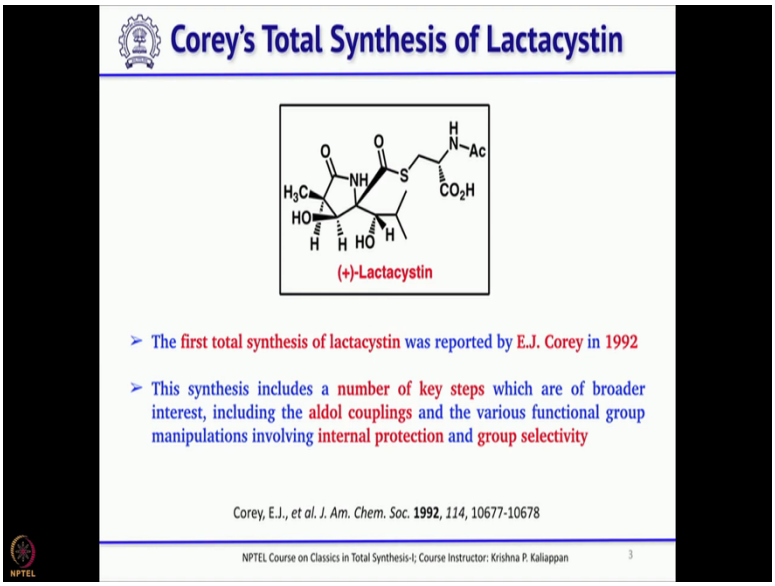
(Refer Slide Time: 16:58)



So, we have been discussing about total synthesis of penicillin type antibiotics and we will continue our discussion on one more natural product close to this. So, that natural product is called lactacystin, this was isolated in 1991 by Omura and his group. And this structure was illustrated by the various spectroscopy techniques particularly the X-ray was very helpful and if you look at this molecule closely it has two amino acids.

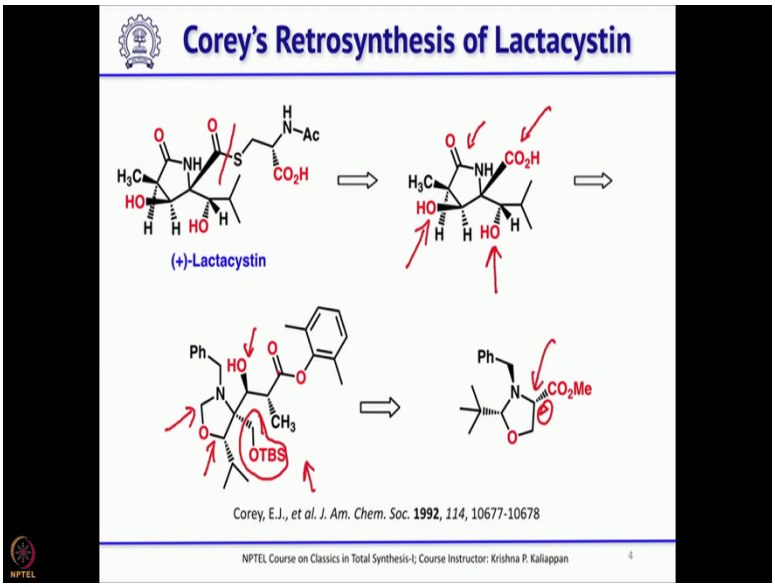
So, one, one here and then second one is here; so, two amino acids and then N acetyl cysteine is also part of this atomic acid derivative.

(Refer Slide Time: 17:50)



The first total synthesis of lactacystin was reported by E. J. Corey, I a year after it was isolated. And his synthesis involve many key reactions, which I will discuss when I talk about the retro synthesis as well as his synthesis.

(Refer Slide Time: 18:07)



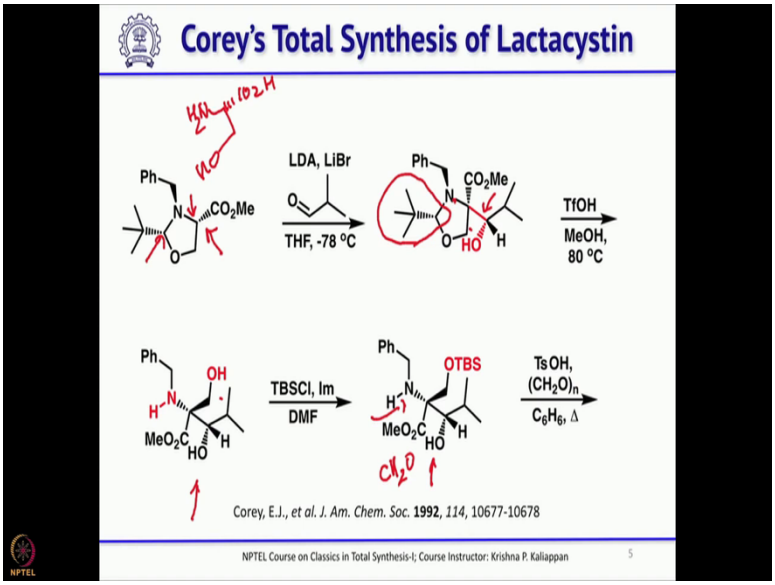
The first obvious disconnection of Corey's retrosynthesis of lactacystin was to cleave this bond ok that actually simplifies the natural product, so, into two fragments. So, now this carboxylic acid ok if you look at carefully, so, you have a carboxylic acid here and then here lactam ok. And of course, you have two hydroxyl groups here, there are four

functional groups. The carboxylic acid, a five membered lactam and two hydroxyl groups both are secondary ok.

So, if you look at this how he has made this compound from this particular compound, you can see. You have the hydroxyl group is still intact. The second hydroxyl group is still intact. And this one that is CH_2OTBS ok. So, that will form carboxylic acid that will become carboxylic acid is a later functional group transformation.

Then if you cleave this, if you remove this CH_2 then this NH can cyclize with this N can cyclize with this that will form the five membered lactam ok. Now, let us see how he made this precursor. So, this precursor he made it from here. So, just you have to generate anion and then quench with an electrophile.

(Refer Slide Time: 19:41)



And this is nothing but you know if you have to make this you can make it from another commercially available amino acid called serine ok, serine is nothing but ok; so, this is serine. So, from serine one can make this in two steps; so, that was a commercially available starting material also. You take this compound and treat with LDA; so, LDA generates anion here and quench with isobutyraldehyde.

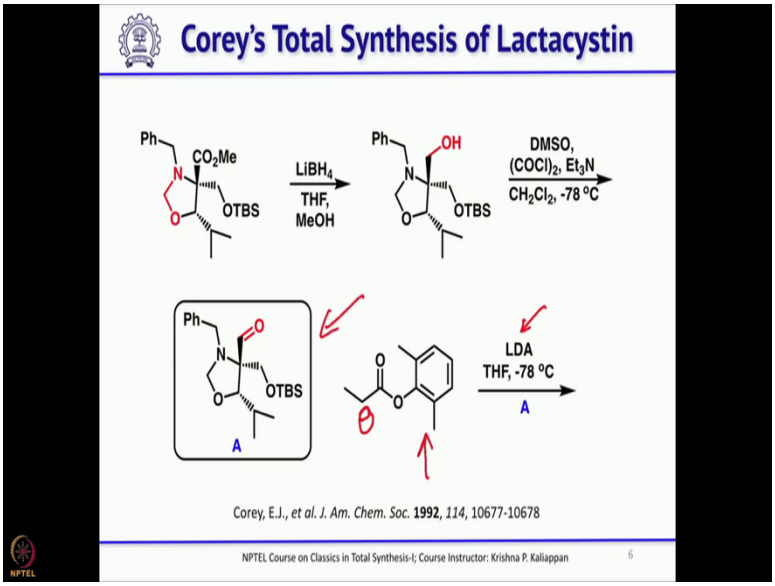
So, when you quench with isobutyraldehyde, now you can see he has introduced one more chiral center ok, already there was one chiral center here, second chiral center here.

So, he introduced the third chiral center which is very very important. Now, you treat with trifluoroacetic acid; so, the trifluoroacetic acid removes this protecting group.

So, that gives you the corresponding N benzylated amine then the CH_2OH; so, you remove that and then you get the amino alcohol. So, that was redrawn like this ok. I will leave it for few seconds; so, that you know you should be able to understand. The CH_2OH is beta; so, it is beta and then NH benzyl is there.

Take this compound and then treat with TBS chloride; so, TBDMS chloride, you can protect the primary alcohol ok. You protect the primary alcohol as TBS ether then treat with toluene sulfonic acid and formaldehyde. This is NH and this is OH ok, you can see NH OH and if these two are protected with formaldehyde again you will get a five membered ring. Is not it? Again you will get a five membered ring. So, that is what happened ok.

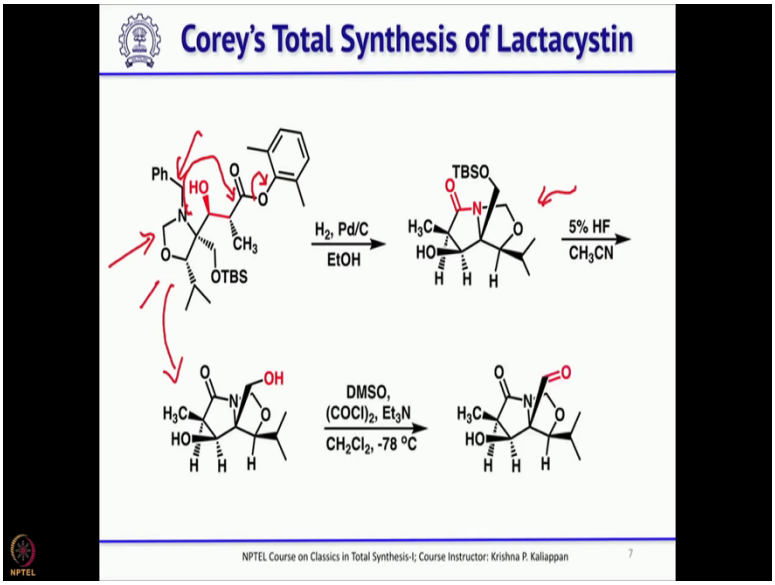
(Refer Slide Time: 21:44)



You can see you protect this amino alcohol, then the ester you could reduce with lithium borohydride to get the primary alcohol. Then, Swern oxidation will oxidize the primary alcohol to corresponding aldehyde. So, this is the key fragment which we could make in few steps starting from commercially available amino acid called serine. Then you take this two dimethylphenol 2, 6 dimethylphenol and then treat with propionic anhydride you get the corresponding ester.

That compound upon treatment with LDA will generate anion will generate anion then quench with this aldehyde ok, it is an aldol reaction basically aldol reaction.

(Refer Slide Time: 22:43)



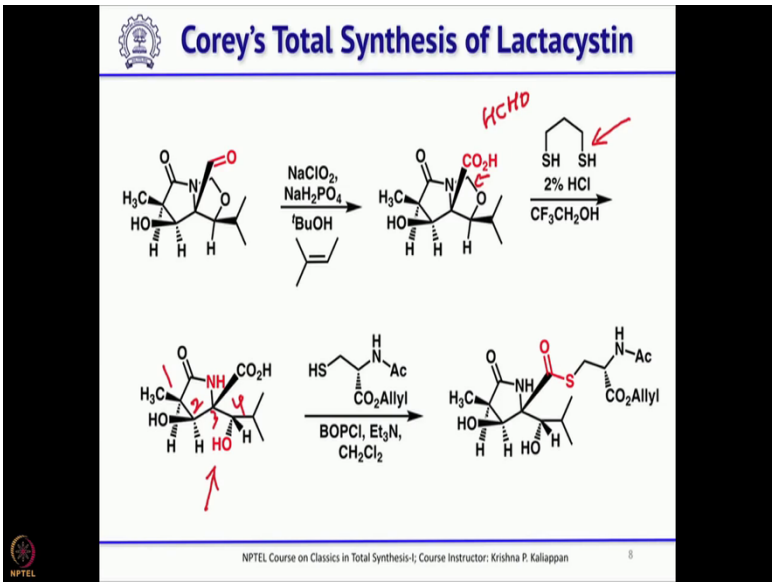
And you get this aldol ok. Now, from here how he goes to this one carefully observe ok. We have the aldol. Next you have to remove this CH_2. You have to remove this CH_2. At the same time, you also have to remove the N benzyl. So, if first if you do hydrogenolysis what will happen?

The benzyl group will be cleaved; the benzyl group will be cleaved. So, that automatically once it cleaves, it will attack this carbonyl group and you have a good leaving group. 2, 6 dimethyl phenol, phenol is a good leaving group ok. And that will give this intermediate ok. Is it easy to visualize?

Because I have to I have rotated 180 degree, so that is why I am just leaving it for some time. From the natural product side you know you have to write like this. So, that is why I have rotated 180 degree yeah. I leave it for some time ok. This portion has come to the right side ok that is why everything is exactly opposite, ok.

Now, TBS was removed with HF then Swern oxidation gives the corresponding aldehyde ok, Swern oxidation gives the corresponding aldehyde.

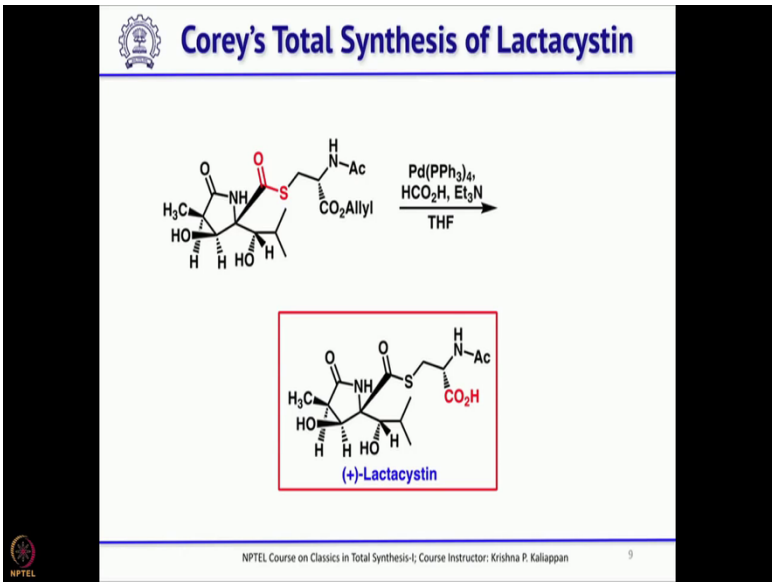
(Refer Slide Time: 24:23)



Next step is oxidation of the aldehyde to corresponding carboxylic acid following Pinnick's protocol. Then treat with 1, 3 propane dithiol. What will happen when you treat with the 1, 3 propane dithiol? 1, 3 propane dithiol in the presence of HCl, this CH_2 that is formaldehyde, is not it, that CH_2 when it is cleaved under acidic condition it is formaldehyde; that formaldehyde will be protected with this 1, 3 dithiol and leaving out your amino alcohol.

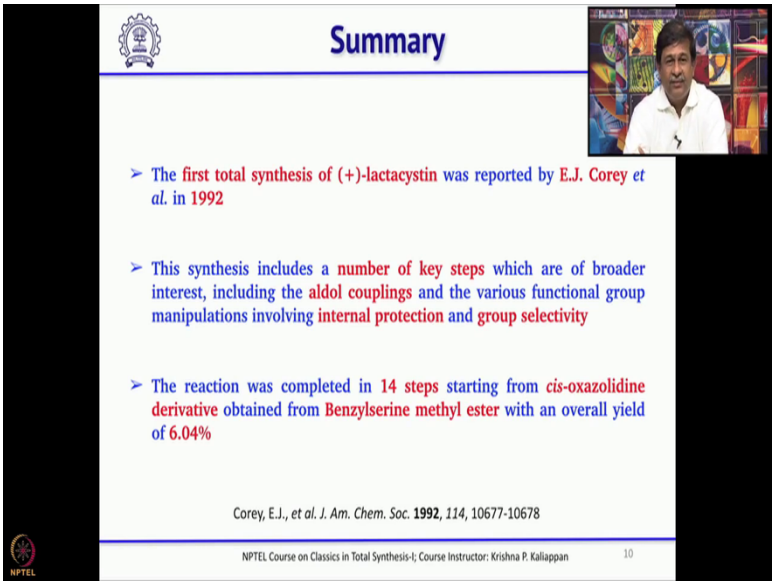
Basically, you are cleaving that protecting group with 1,3 propane dithiol with HCl ok. So, now, as I said there are four chiral centers in lactacystin, 1, 2, 4 all four are done ok. Now, you have to attach the thiol side chain so that you can do from cystine ok. The cystine N is protected as acetate and then the carboxylic acid is protected as a allyl ester ok. So, now, you couple this with carboxylic acid, so, you get the corresponding thio ester ok. So, what is left? You have to remove the allyl group ok.

(Refer Slide Time: 25:56)



So, how do you remove allyl group? Palladium catalyst; so, if you treat with tetrakispalladium in the presence of formic acid; so, you get lactacystin. So, basically this was a very elegant total synthesis starting from commercially available amino acid called serine. See there are two amino acids he used, one serine another one is cysteine ok, other chiral centers he used based on these two amino acids.

(Refer Slide Time: 26:25)

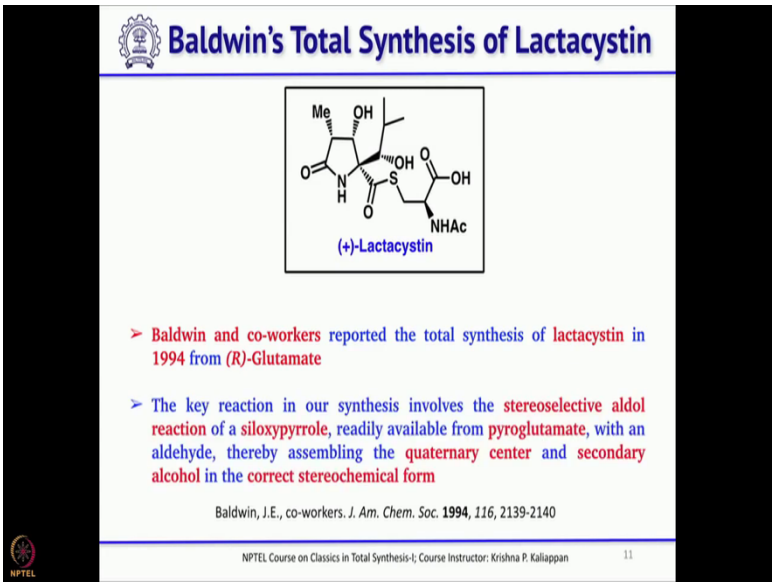


So, this was the first total synthesis reported by E. J. Corey and the key reactions are aldol reaction mainly aldol reaction and then two types of protections he carried out. So,

that was important while carrying out the next aldol reaction. Overall, this synthesis was accomplished in 14 steps and he has to use few protecting groups.

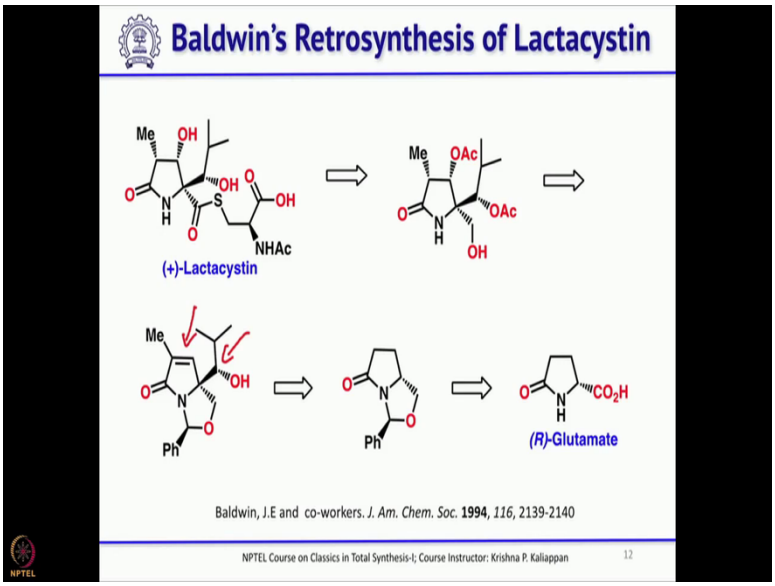
See when you use once protection you are adding two reactions; if you use more protecting groups accordingly you have to multiply it by 2, because one step is required for introduction and one step is required for removal. So, that is how the number of steps has increased to 14 and with the overall yield of 6 percent.

(Refer Slide Time: 27:15)



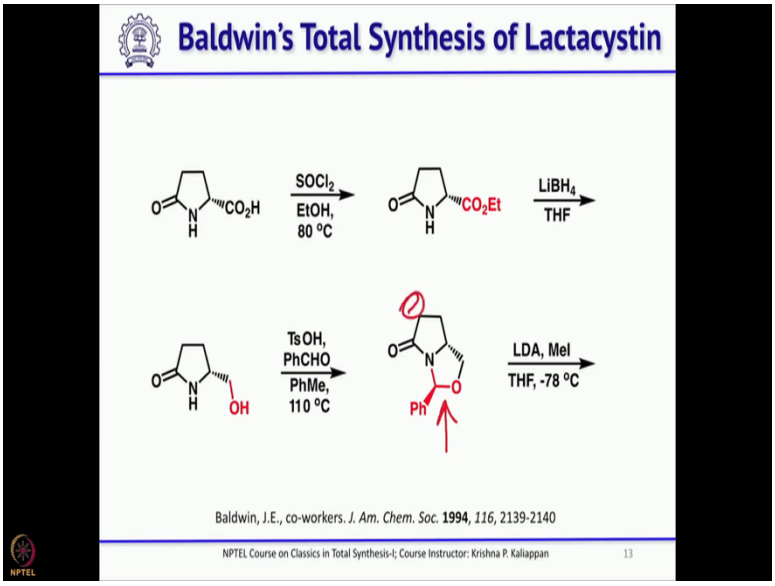
Then the second total synthesis was reported by Baldwin in 1994, two years after E. J. Corey reported the total synthesis. And here again he started with another commercially available amino acid glutamate and let us see how he made this.

(Refer Slide Time: 27:33)



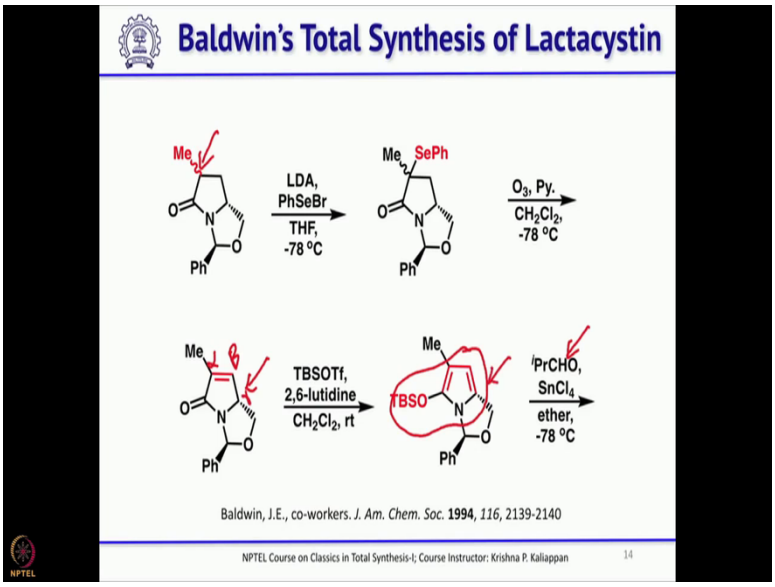
His retrosynthesis again as you know the cleavage of CO CS bond is the first thing. Then he wanted to introduce the hydroxyl group here using a dihydroxylation method if you have a double bond then you can do dihydroxylation and then selectively cleave one of the hydroxyl group ok. So, he wanted to do that and then this chiral center he wanted to introduce using aldol reaction ok. So, that aldol reaction will give this compound and this can be obtained from amino acid, it is a glutamic acid ok.

(Refer Slide Time: 28:21)



So, now let us see how he started and then how he got the synthesis of lactacystin. So, glutamic acid, pyroglutamic acid and treatment with thionyl chloride, ethanol you convert that into ester ethyl ester. And ethyl ester can be selectively reduced in the presence of lactam using lithium borohydride to get the corresponding primary alcohol. This upon treatment with paratoluene sulfonic acid and benzaldehyde you get the bicyclic ring and one more chiral center is fixed ok.

(Refer Slide Time: 29:00)



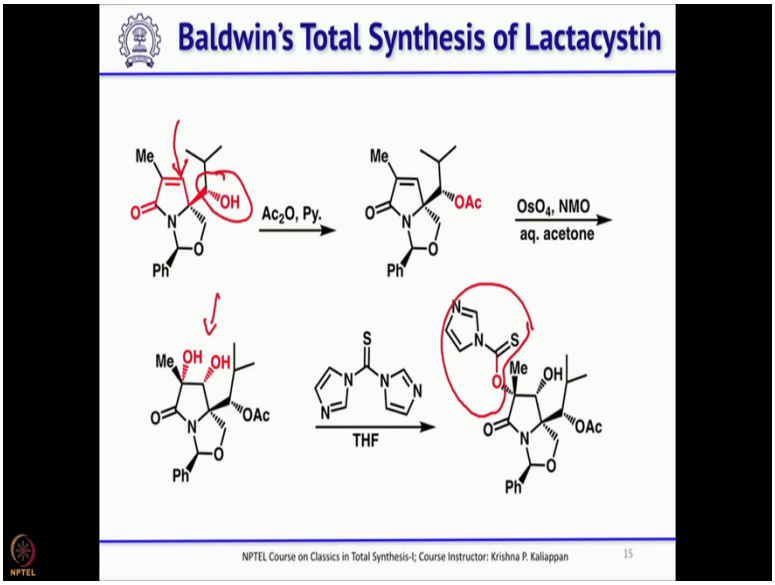
With this one can generate anion here and quench with methyl iodide to get the methyl group. Then you introduce a phenylselenyl group. Introduce a phenylselenyl group here by treating with LDA and quenching with phenylselenyl bromide followed by treatment with ozonolysis. You introduce or you oxidize the phenylselenyl group to phenylselenyl oxide followed by elimination to get that double bond.

And as I mentioned once you have the double bond, the next step is the dihedral oxidation. So, the dihydroxylation before doing the dihydroxylation he wanted to do an aldol at this carbon. So, at this carbon means you know alpha, beta, gamma, carbon because if he does dihydroxylation at the double bond then the gamma position it will be difficult to acylate or do aldol reaction.

So, what he did? He wanted to do the aldol reaction first. So, he treated with the TBS triplet and then 2, 6 -lutidine. So, that form the dienolate. The dienolate was quenched to form the corresponding TBS ether; so, this is almost now we can see it is like pyrrole

ring. Is not it? The pyrrole, now, if you treat with isobutyraldehyde in the presence of Lewis acid like tin tetrachloride, the aldol reaction takes place at gamma carbon ok.

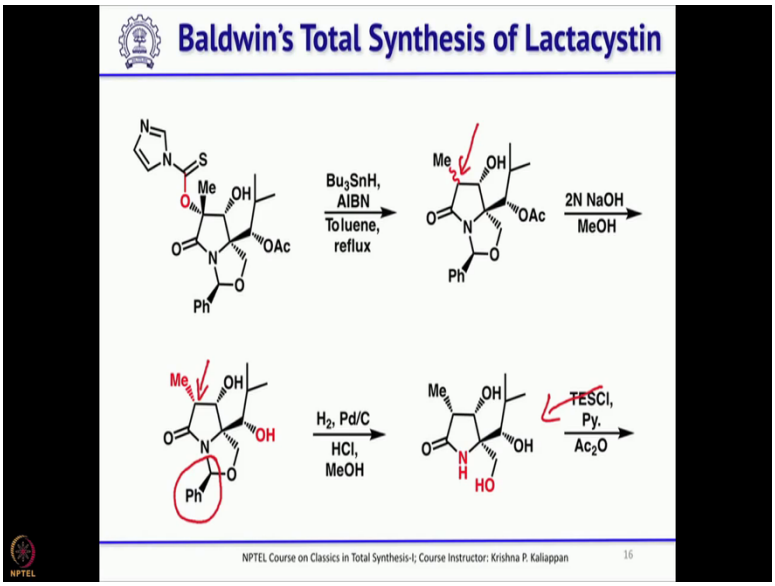
(Refer Slide Time: 30:24)



So, that is how we can see this stereo center was fixed. So, now, you need one more hydroxyl group at this beta carbon. Again, the free hydroxyl was protected followed by osmium tetroxide treatment, he got the diol ok. Now, you have a tertiary alcohol and secondary alcohol.

The secondary alcohol should be intact but tertiary alcohol should not be. So, what we did cleverly? He treated with the CDI that is carbonyldiimidazole; so, that reacted with tertiary alcohol to form the corresponding thio derivative. So, now this type of derivatives are known to undergo deoxygenation. If you take this compound and treat with tributyltin hydride in the presence of AIBN ok; so, that will undergo deoxygenation ok.

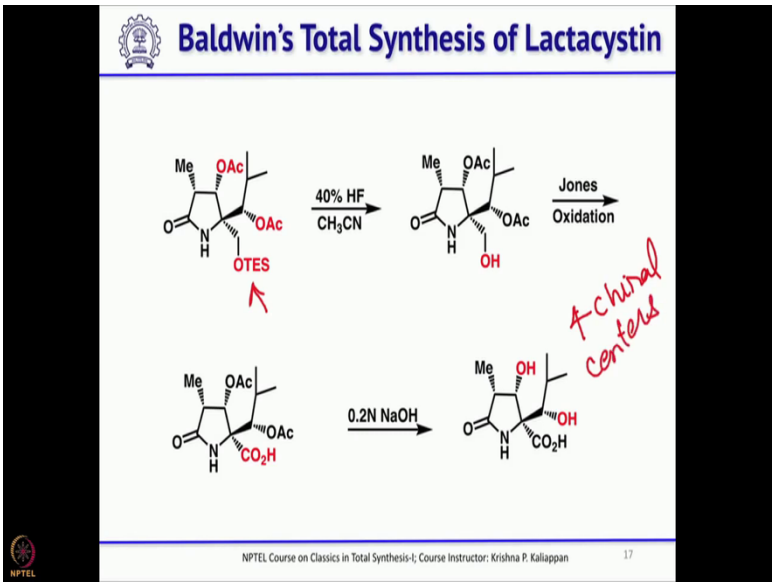
(Refer Slide Time: 31:14)



So, only problem is this methyl group ok, that chiral center is not one isomer ok, nevertheless you treat with sodium hydroxide. So, sodium hydroxide will hydrolyze the acetate as well as it will epimerize this ok, sodium hydroxide will hydrolyze the acetate as well as it will epimerize the carbon adjacent to the carbonyl group, done.

Then this protecting group can be easily cleaved by hydrogenolysis. So, you get a free amino alcohol ok. You have come up to very very key intermediate now. How many chiral centers are fixed? Four chiral centers are fixed starting with one chiral center of pyroglutamate. Then protect the primary alcohol as TES ether and secondary alcohol as acetate.

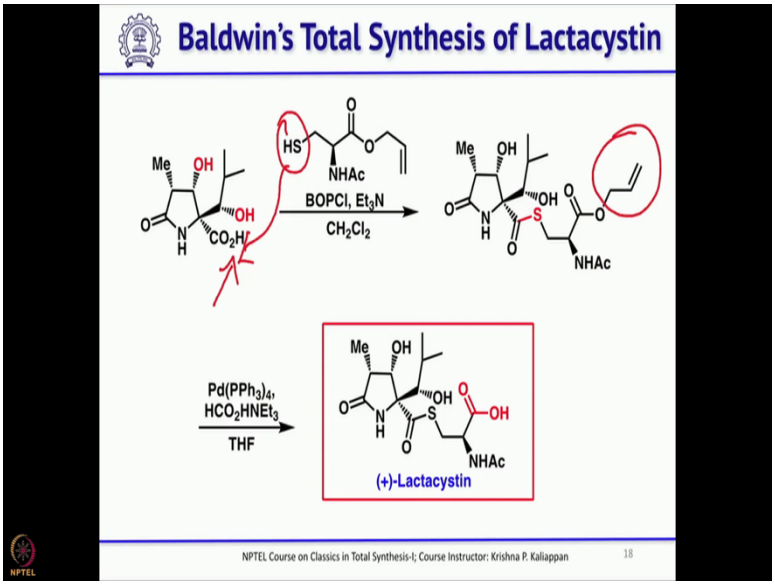
(Refer Slide Time: 32:13)



Then remove the TES with 40 percent HF to get the primary alcohol ok. The primary alcohol what you want in lactacystin is carboxylic acid. Is not it? So, in one pot you can do with Jones oxidation without touching the acetate, without touching the chiral center. The primary alcohol can be oxidized to carboxylic acid in 15 to 30 minutes ok.

Once you have the carboxylic acid, then the acetate group can be cleaved with sodium hydroxide solution. Now, you can see all the four chiral centers. All the four chiral centers are fixed. Is not it?

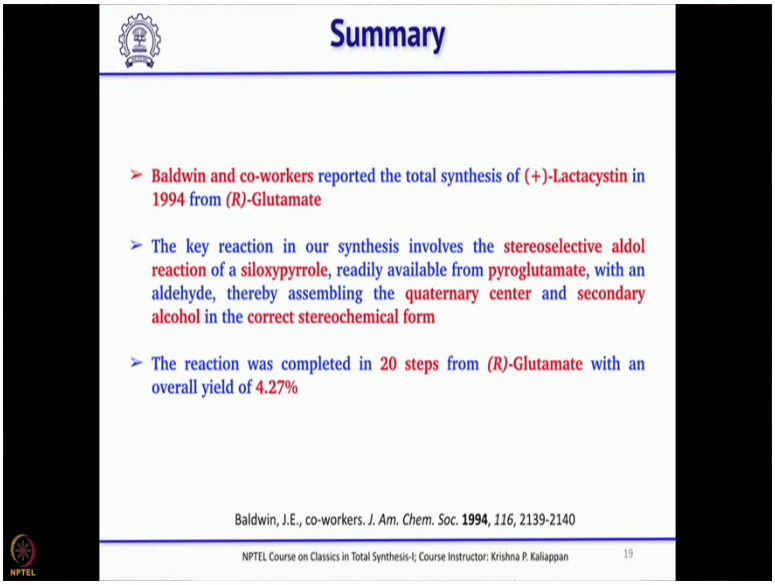
(Refer Slide Time: 32:58)



So, what he what he has to do? He has to couple this carboxylic acid with the thiol and the thiol should be derived from cysteamine. So, he took cysteamine that NH was protected as acetate and then carboxylic acid was protected as alanine ester. Then this thiol can undergo coupling with carboxylic acid.

Yes, it went and it formed the corresponding thio ester and what is required for the total synthesis of Lactacystin is to remove the allyl group. So, the allyl group can be easily removed with tetrakis triphenyl phosphine palladium and the formic acid and triethylamine lead to lactacystin ok.

(Refer Slide Time: 33:47)



So, this is one of the simple and straightforward total synthesis of lactacystin and starting from pyroglutamate and it was also reported three years after isolation of Lactacystin. And the key reactions involved in the synthesis of lactacystin by Baldwin, a stereo selective aldol reaction. If you look at the siloxy pyrrole, the aldol reaction took place at the gamma position ok, the gamma position that is our first key reaction where a quaternary center was incorporated with the chiral center.

Then the this side aldol reaction and finally, attachment of the cystine. Overall this sequence took about 20 steps slightly longer than what Corey has reported. Nevertheless the starting material is commercially available that makes huge difference and it is not expensive. And the overall yield compared to Corey's, Corey's was 6 percent and this is about 4.27 percent ok.

So, with this we complete the total synthesis of five membered you know antibiotics. So, we will move to the next natural products having six membered ring as the core structure ok.

Thank you.