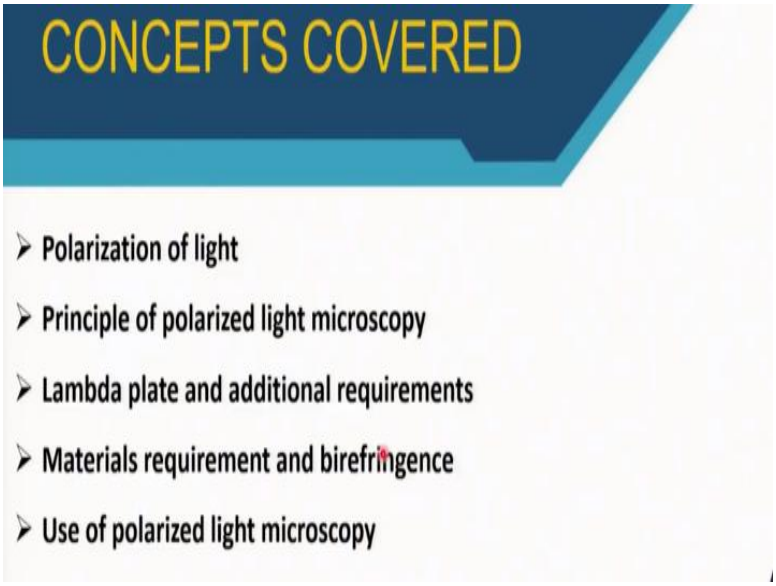
Techniques of Materials Characterization
Prof. Shibayan Roy
Materials Science Center
Indian Institute of Technology - Kharagpur

Lecture – 8
Polarized Light Microscopy

Welcome everyone to this NPTEL online certification course on Techniques of Materials Characterization and we are now in week 2. So far we have discussed various modes of optical microscopy and we discussed about bright-field mode, dark-field mode which are dark field in contrast enhancing mode and then we started discussing about another contrast enhancing mode that is phase contrast microscopy which as I mentioned that it was mostly useful for biological systems.

And now today we will be discussing about another contrast enhancing method that is polarized light microscopy. And polarized light microscopy is very useful for material scientists, geologists, and also to some extent at certain times chemists plus biologist of course.

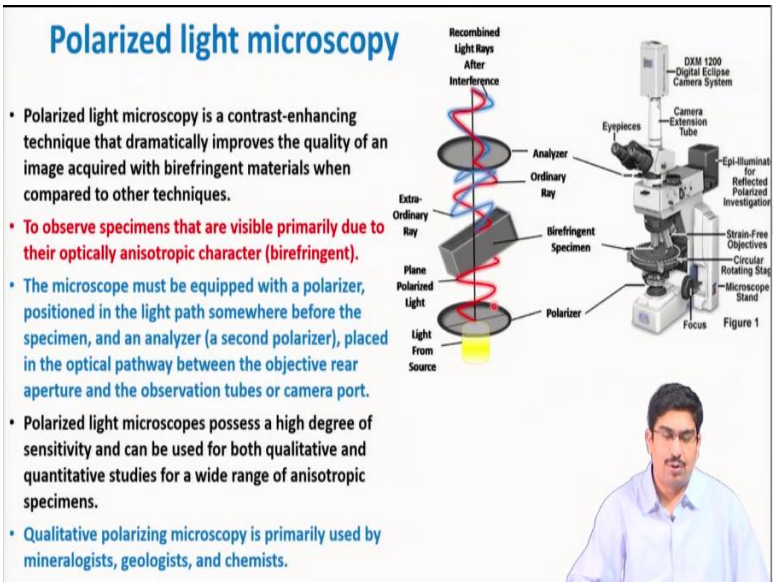
(Refer Slide Time: 01:22)



And here first we will be discussing about polarization of light and then the working principle of polarized light microscopy. And then we will discuss about the lambda plate and certain additional requirements of this polarized light microscopy, then materials requirement and a very important property of material that gives rise to the polarization

light microscopy that is the heart of it is birefringence. And finally, we will see couple of use of this polarized light microscopy.

(Refer Slide Time: 01:55)



So, let us just start with a very brief introduction about polarized light microscopy. So, as I already mentioned, polarized light microscopy is a contrast enhancing technique and it improves the quality of an image acquired for birefringent materials, mostly it is very useful for birefringent materials which are optically anisotropic material, we will learn about that birefringent material, what it is called by birefringent materials.

And for those kinds of materials just like what we have seen for phase contrast microscopy those biological specimens they were not very good for bright-field illumination. When you apply a bright-field illumination, they did not produce enhance or additional contrast, so the contrast the background was so, I would say so bright that you will not get the features indistinguishable from each other.

So similarly, in this case also for this kind of material, birefringent materials, if you take them in bright-field mode, the contrast is not very good. Contrast between different regions of a birefringent material or the contrast between if I have two different phases altogether it is not very good in a bright-field image. So, to enhance this contrast to make those features more distinguishable from each other, we try to go to polarized light microscopes at times.

And this polarized light microscope as I said these are mostly used for anisotropic birefringent materials and here you use some additional components. So, phase contrast microscopy also we have seen that some additional components was there like the phase plate and then for dark field you had that light stop. Light stop was basically for making the oblique illumination and so on.

So, this contrast enhancing methods, this polarized light microscopy is another contrast enhancing method and here also you need some very typical two components that must be there and those two are called polarizer, both of them are basically polarizer, both of them are exactly the same thing, just the names are a little bit different. One of them we call it a polarizer which is basically used right after the light source before the light basically reaches to the condenser lens.

And there is another very similar polarizer that is placed somewhere after the objective lens before the image forms and we call it analyzer that is what. So, if you look at here the polarizer is placed right here before the light reaches to the specimen. So, this is where the polarizer is used and before the light goes and form to the tube to the eyepiece or to the camera and from the final image the analyzer is used.

These are the two main components of any polarized light microscope. And this polarized light microscope one of the very important property of this is that out of all the techniques that is available contrast enhancing techniques or any other bright field technique also various modes that are available. This one can be used to some extent for quantitative measurements as well.

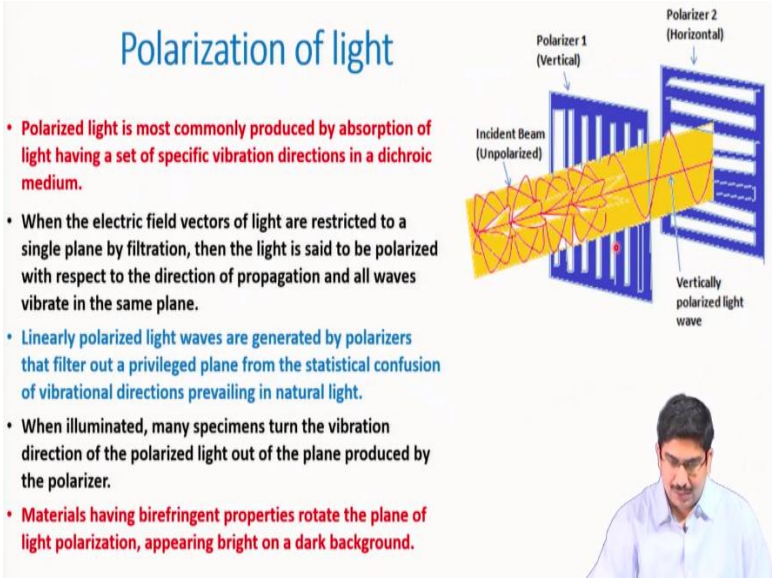
Some quantitative studies, what kind of quantitative studies I am talking about I will come when I go for a specific example. But you can do something more with these images, which is just not a visual representation of your specimen. So, all other modes is mostly like bright field of course, dark field or if you take phase contrast, all of these modes are just a mere visual representation of your specimen.

Whatever the features you have in your specimen, it just represents that. Polarized light microscopy because the way the light signals are produced here, the birefringence property, you can do something more if you are able to read that whatever image that is

produced, how it is produced, if you know a little bit about your specimen, you can extract much more data or much more information out of it.

So, this is one, I would not say it is a completely quantitative information, but it is a semi quantitative information of course. And this kind of, as I said polarizing microscopy is very regularly used by mineralogists and geologists and chemists as well other than material scientists or biological or Life Sciences, people dealing with biological samples, other than them even the geologists and mineralogists they very regularly use polarized light microscope.

(Refer Slide Time: 06:29)



Now let us talk about what is polarization of light okay. So, polarization of light is basically this most commonly produced by absorption of light with a certain specific vibration direction. So, you can imagine this polarizer or polarizing when the light is there, normal white light, sunlight, whatever, normal white light where the light has the direction. The light is an electromagnetic wave.

So, it has an electric field vector and that the electric field vector can have any random direction. The vibration direction can be of any random plane for a wide beam that is what generally we tend to see is all that lights. Now the polarizer basically acts like a gate. So, it has a very regular opening, you can imagine that it has a very regular opening and it only allows a certain plane the electric field vector.

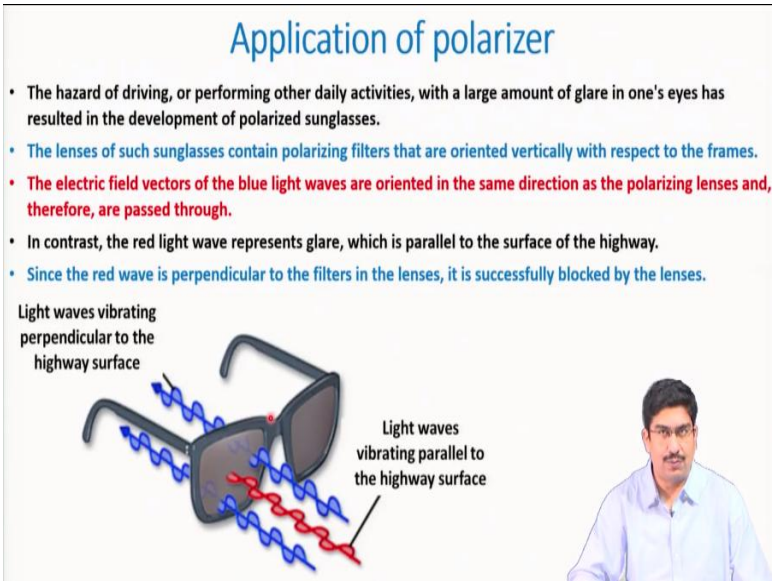
When they are aligned only along certain definite plane these are allowed on the other hand side, all others will be stopped in the side. So, you can imagine this polarizer to be just like a gate and through that gate only certain directions, certain part of the light which has their electric field vectors aligned along certain specific direction and that specific direction is again determined by this polarizer, so that will only be allowed through this.

Now if you put another polarizer which is just exactly having 90^{o}, so it is having its opening exactly 90^{o} to this polarizer then what will happen is now all the light or everything all this electric field vectors these are aligned along one direction. They will not be allowed to go through this gate. So, they will be all stopped here, right. So that is what basically the polarizer and analyzer does, we will see that.

So, polarizer analyzer when they are exactly 90^{o} to each other this is what they do, they just stop the entry of any kind of light. One of them, first one chop off everything else from without or just one set of or one direction of electric field vector vibrating in one direction only those lights are kept by one and the second one stop even that. So that is what it happens and that is what is called polarization of light in the polarized light microscope obviously as the name suggests.

We need this polarization or this polarizer at the beginning right before the specimen is illuminated by the signal, we need a polarizer. And that polarizer does exactly this, it makes the light beam to travel or the electric field vectors are all aligned along one certain plane. So, typically you can say that light is now polarized along one certain plane.

(Refer Slide Time: 09:23)



Let me give you a very typical example of a polarizer. So, this is just for fun purpose, but all of us are used to it. So, you must have heard about something called antiglare. So antiglare, these days we are using a lot of digital display and many people we are using sunglasses and all of them we are using glasses like me. So, we all have this antiglare coating. Now how does this antiglare coating work?

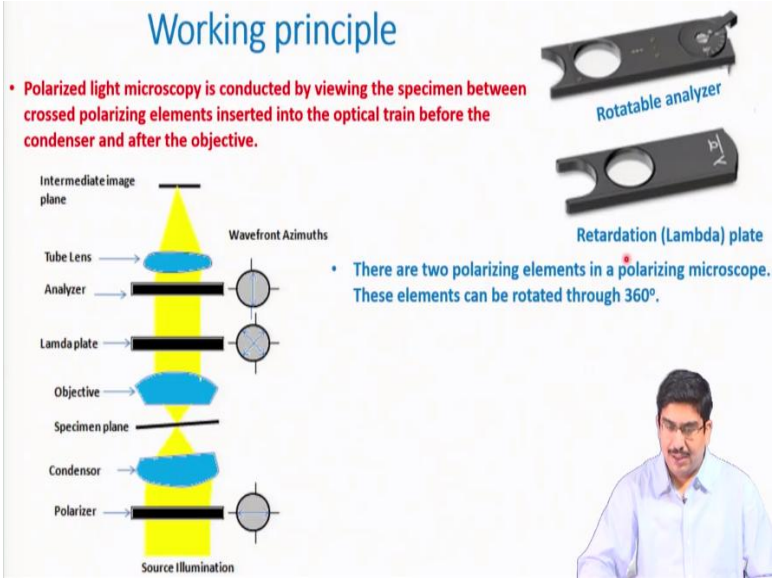
Basically, what happens is that in our glasses, we have one such coating, one such analyzer, a birefringent material is there and that coating is there. So, what does it do? It has exactly the same kind of purpose. It has these gates, but now all of these gates or all of these openings or all of this polarization is in the vertical direction. It is given in such a way that only through this vertical direction the lights can pass through.

And what happens is that generally only the blue light usually has the right kind of a plane or it is traveling in the right kind of a plane so that only the blue light is allowed through this antiglare coating or through the sunglasses. The red one typically or usually the red one has perpendicular to this direction to the blue light, this is perpendicular to the blue light and that is why in the polarizer or the antiglare coating that you have.

Polarizing coating that you have this red one is stopped. So the red light is stopped, green light only can go through right. So, this is how the polarizer basically works and the polarizer it is not only in microscope, but the polarizing or the concept of light polarization is very essential for these days for any kind of antiglare coating or that kind of making sunglasses because light rays sunlight.

When most importantly when you drive, light that is coming out of a highway surface asphalted surface that light is containing a lot of glare. So, to remove some part of the glare, this polarization coatings are very useful.

(Refer Slide Time: 11:40)



Now, let us come to the working principle of this polarization, polarized light microscopy. As I said that polarized light microscopy needs the polarizer and analyzer and this is how the polarizer basically looks like. This is written as analyzer, but as I said analyzer and polarizer basically exactly the same, both of them are same plus you have some one more additional component you need that is called lambda plate, we will come to that.

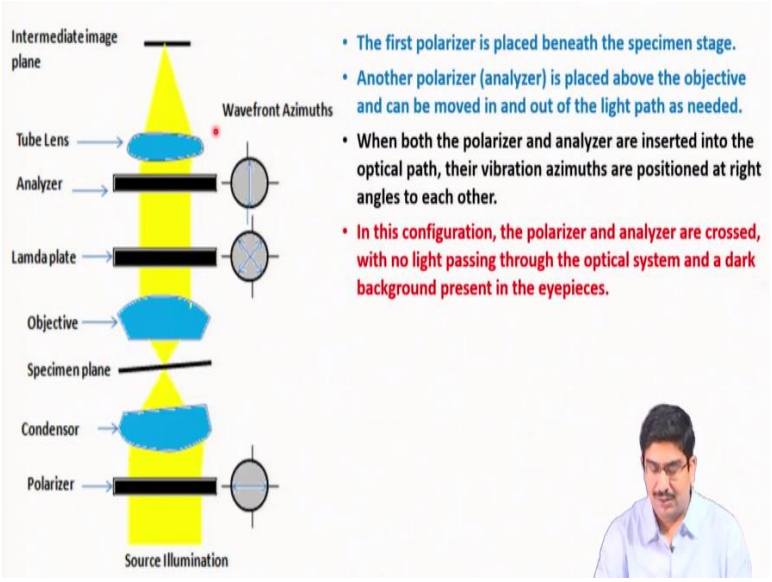
What you do is that as I said polarizer you insert the polarizer before the light passes through the condenser lenses. So, between the condenser lens and the light source you put this polarizer and that polarizes the light in one particular direction. And then you have the condenser, you have the specimen plane, you have the objective, everything remains the same. Then you usually have this lambda plate, again I will come to that.

So, you have the lambda plate here and then before these light rays or before this image is formed by the objective lens and it reaches to the tube lens, which is basically the eyepiece lens, you have the analyzer. So, analyzer is this side and polarizer is the side. So, the analyzer and polarizes in this plane, the plane in which they can polarize the light

this can be changed and this can be changed by here. You see here there is something like it can be rotated.

So, there is some material is there which is a birefringent material and that plane can be rotated and can be adjusted from here, just by rotating this you can adjust that which plane, basically you create some kind of some amount of strain and that strain creates, the strain I would say helps the light to or the plane of polarization is changed as by changing the strain here the plane of polarization can be changed in this material which is there and this can be done in 360°.

(Refer Slide Time: 13:42)

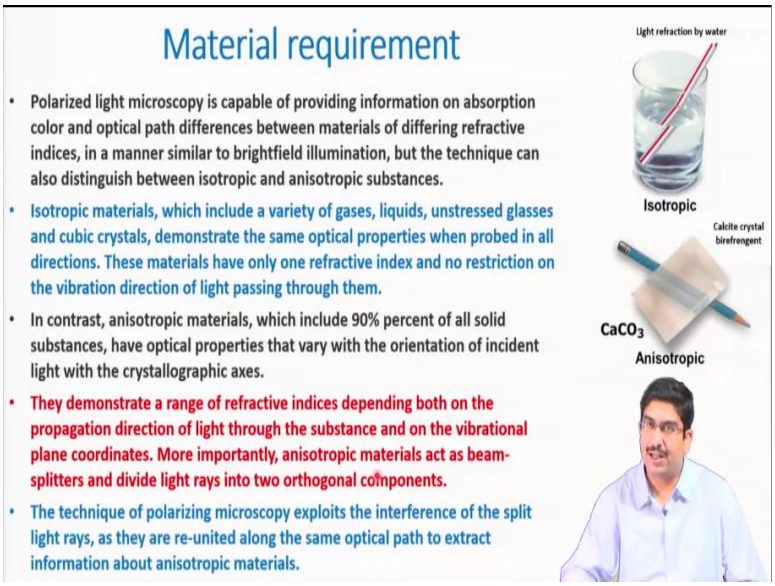


Now, what you do is that you have the polarizer here and you have the analyzer here. Most often to begin with what we do is that we keep them in a configuration known as crossed polarization, crossed or complete. And the cross polarization means that the polarizer the plane of polarization for the polarizer and the plane of polarization for the analyzer is exactly 90 degrees to each other.

That is what these arrows are suggesting. If this is the plane of polarization for polarizer, this is basically the plane of polarization for analyzer. So that means what happens is if there is no specimen in between and the light is allowed to travel freely through this condenser lens and through this objective lens, so in this side you basically will not get any light because here it is polarized in a certain way and that polarized light will be completely stopped here in analyzer.

It will not be able to travel after or through this analyzer and reach all the way to the image plane. So, it will be completely stopped. If you try to check if you look at through your tube lens in the cross polarized condition without any kind of specimen in between, you will be not able to see anything, it will be completely dark everything okay. So, this is called crossed polarized condition to begin.

(Refer Slide Time: 15:09)



Now, before we move on to how this one basically works, this actually works let us just talk about some requirement of material. What kind of material we can study in a polarized light microscope? So, this is called material requirement. So, polarized light microscope as I said basically this gives very good results for a kind of material which is known as birefringent material.

So, what is that birefringent material actually means? So, birefringent materials they have refractive index difference in that material either because of different phases or if it is a crystalline material because of different directions and so on. But it must have a difference in the refractive index in the material itself, so that is called birefringent material. And what does that birefringent property then give the difference in refractive index it does is that it will produce some differences in optical path length.

And that will finally cause this polarized light microscopy is the contrast generation and of course this contrast generation can be also we can tweak it and we can produce a color out of it using the lambda plate. So, that also we will come to come to that. But

basically, you need in your material is a difference in refractive index. Then only this polarized light microscopy works.

So, of course what you need is an anisotropic material. Isotropic material we all know, couple of isotropic material like gases, liquids, water is the best example and unstressed glasses and cubic crystals. Out of various crystalline materials if you have cubic crystals basically because of the symmetry the cubic crystals are more symmetric, so the cubic crystals every direction if you consider just like 0, 0, 1 direction, every direction the refractive index is the same.

Cubic crystals have this unique four-fold axes and then two-fold axes, four three-fold axes and so on. So, that is why they are very much isotropic. They do not have any refractive index difference. And best example as I said the best example is something like water and if you look at through this water, then this straw will look perfect. All if you look here that means if you are looking now through air.

Again, the straw will be completely looks the same. There will not be any difference between this like this straw which is here, this will look completely complete, it is perfect. So, there will not be any difference. Of course, there will be a difference between this air and glass because of their refractive index difference. But if you look at material something like calcite, calcium carbonate.

And if you look at this pencil here and if you look at from different directions, you will be able to see two different images of this and that is basically because it has a diffractive index difference in different directions. And that kind of material is called optically anisotropic materials and they are mostly like 90% of all solid materials are anisotropic.

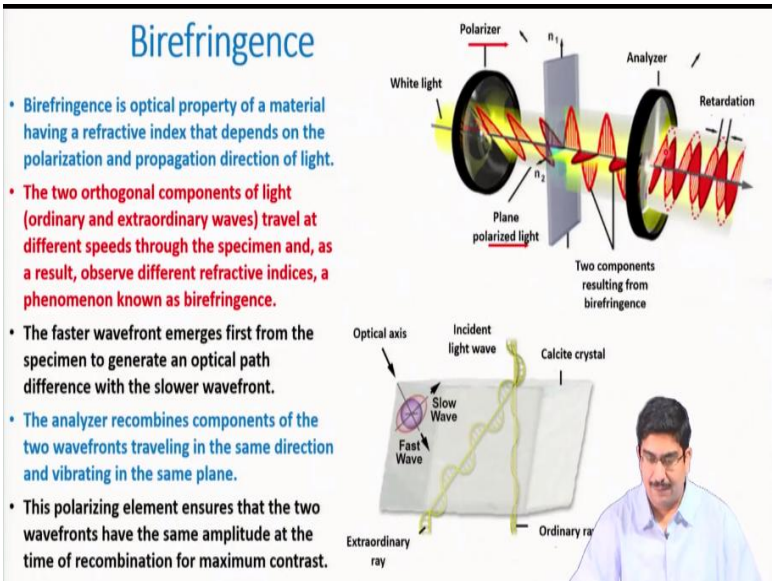
Particularly the complex crystal structures hexagonal or even more complex crystal structure they are all having anisotropy. That means they have their optical properties or refractive index particularly that is sensitive to the orientation of the incident light okay with the crystallographic axis. So that kind of material is called birefringent material or anisotropic material.

Now, what does this do this refractive index then depends on the propagation direction of the light. So at which plane light is entering, depending on that the refractive index will change. And this is very severe for obviously you can understand for plane polarized light. So, if you have a polarized light, now that polarized light everything all the lights are having the same direction, all of them having the vibrational directions in the same plane.

If that encounters the birefringent material, then obviously the refractive index will be different than which plane the polarized lights are or if you change the plane of this polarization then that will experience different kinds of refractive index within the same material. And what it will do is that we will just now next we will see that. That basically these birefringent materials, this polarized light what they will do on anisotropic material?

They will divide this light into two orthogonal components because of this change in refractive index. And finally, what we do is that these two different rays which are produced by this birefringent material or anisotropic material basically recombine them and in that way we generate a contrast.

(Refer Slide Time: 20:10)



So, now let us look at this birefringence property and how that we are getting this contrast and what we are meaning here. So, if you have this white light here you have the polarizer in the beginning and polarizer will give you this light which are polarized along

certain direction. Next it will encounter birefringent material here and what this birefringent material will do?

As I said because of the difference of refractive index in different directions it will create two components. From the same ray it will create two components and that is what the birefringence is all about. Basically, you have one single incident light here and then the light has two orthogonal components, one is ordinary ray, one is extraordinary ray. I am not going into this optics and all what is extraordinary, what is ordinary.

Just believe that the light rays when they travel through this birefringent material, they will always split up with these two different light rays, one is ordinary rays and two different orthogonal components. So, these components, light components are exactly orthogonal to each other, exactly 90^{o} to each other. This like if a polarized light enters here one single, they will always be divided into two light rays which are 90^{o} to each other their vibration direction.

The plane of vibration will be exactly 90 degrees to each other. And they will have this ordinary rays and extraordinary rays. What they will do is that they will always have two different speeds. Their speed will vary within this and that is again we understand this in case of a refractive phase contrast microscopy. Because of the change in refractive index, these two rays will always have, because they are exactly 90^{o} to each other.

So, they will travel in two completely different direction and as I said anisotropic material, birefringent material, they have different refractive index. The refractive index is very sensitive to the orientation of this which crystallographic direction the light is entering here, which crystallographic direction they are entering depending on that you will have this ordinary ray and extraordinary ray they will split up and then they will experience two completely different refractive index.

So, their speed will also vary, that means they will have an optical path difference which will be seen as a change in their phase. So, they will have a phase shift as well here. So, finally they will have phase shift. So, this polarized ray is entering here, it is encountering now the specimen birefringent specimen, it is divided into two different

rays of 90^{o} to each other and plus they will have this ordinary and extraordinary ray, they will have a phase shift between them.

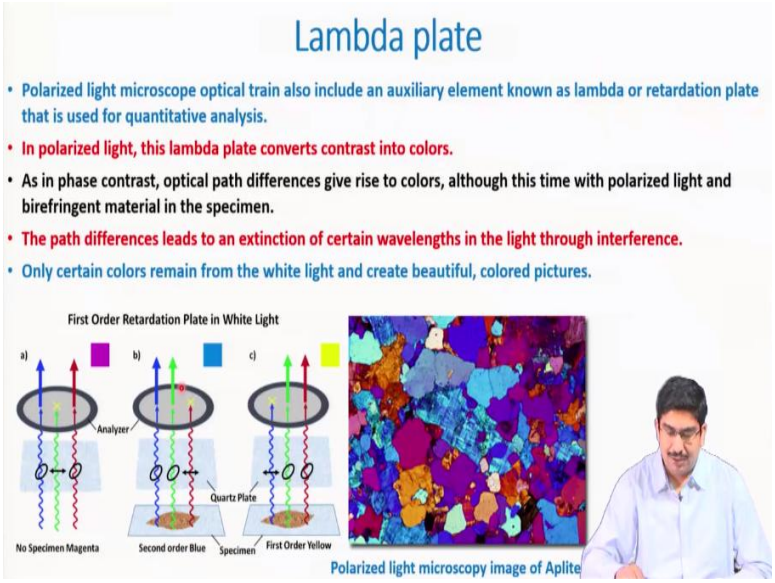
Now, after they passes through the analyzer and remember these two are kept in a cross polarized condition, but you can also keep them a slightly uncrossed condition that is also possible. I am not going into that because that will increase the complexity. But you can keep them a slightly instead of 90^{o} you can change this angle also depending on your purpose and whatever gives you the best contrast.

So, this analyzer now what it will do is that it will now allow only one ray from out of these two. So, there will be one ray, so there will be an interference that will happen. This will recombine, this analyzer will recombine these two arrays, these two components. Extraordinary and ordinary rays, these two components will be combined, interference will happen, they have a phase difference and that is why finally you will get a contrast.

Again, the same kind of a contrast that was there in case of phase contrast. So, same thing will happen here and you will get a different contrast. So, that means in the different places within the same birefringent material, they will have completely different phase relationship after this polarized light passes through this birefringent material.

They will have completely different phase relationship these rays and of course in the analyzer when they are recombined, they will generate completely different amplitude. I mean as we already discussed that our human eye is sensitive to amplitude. So, they will finally generate different amplitudes here.

(Refer Slide Time: 24:35)



Now what the lambda plate that is how finally the image will form and what is lambda. Now the lambda plate basically is responsible for color. So, this beautiful color that you are seeing here, this is how the polarized light microscopy works. This is for a mineral called apatite which is an igneous rock basically. And this polarized light microscopy basically gives you this beautiful color.

And here these are grains and the color comes because of the orientation as I said. So, if you have a crystalline material polycrystalline material, which has these different grains, every grain will work like birefringent material and it will give you this different kind of contrast or a different amplitude for the rays after recombination. What you can do is that you can use a lambda plate and that lambda plate after recombination.

Now what you have is that you can change this amplitude difference into a corresponding color difference which is a real color basically. The difference you can do a coloring, you can use any software basically you can take a bright-field image or dark-field image and produce a color. Depending on this amplitude, you can just calibrate that amplitude to the corresponding color and you can do it.

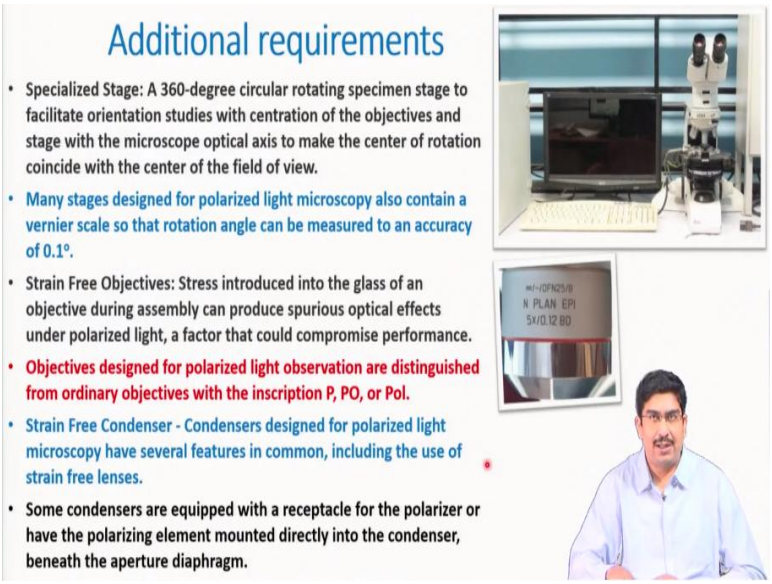
But here the color is a real color which is given by this lambda plate and how it happens is that basically because of the optical path difference as I already told you that optical path difference is there if you have a birefringent material. Different parts of this birefringent material will produce your different lights, lights with different optical path difference, which will finally generate the amplitude difference.

And you convert them to amplitude difference and then you finally get the image. But if you use this lambda plate, now what does it do is that it is converting this amplitude difference into certain wavelength. So, this path difference will lead to an extinction of certain wavelength in the light through interference. So, this optical path differences which will be there between the extraordinary and ordinary rays.

And the phase shift is there, optical path differences there, after interference this will produce a ray with certain kind of wavelength, certain particular wavelength. That means if you have a certain particular wavelength, that means that wavelength if it falls within the visible spectrum, it will generate a color. It will generate a particular color through this. So, for example let us say you have this analyzer here, no specimen basically okay.

So, if you have this, now what it will do is that through this analyzer certain in the visible light, only certain part of the visible light will be able to produce or the light that will be produced from this analyzer will have certain wavelength only and that certain wavelength will give you certain type of color here, which is a real color that is what the lambda plate does.

(Refer Slide Time: 27:39)



You need to have certain additional requirements for this polarized light microscope of course. One of them is that you can have a 360 degrees rotating stage here. Generally, we tend to have that and that is because you like to rotate your specimen and brings

different orientation, if you have a polycrystalline material you will try to bring different regions with different grains and different orientations.

So that you finally could change the colors and finally could generate different images. That is first thing that is why you tend to have a nice rotated stage when you are using the polarized light microscope. And generally, these stages all they are having a vernier on this, so you can know exactly how much you are rotating this. So, this is very important for quantitative analysis.

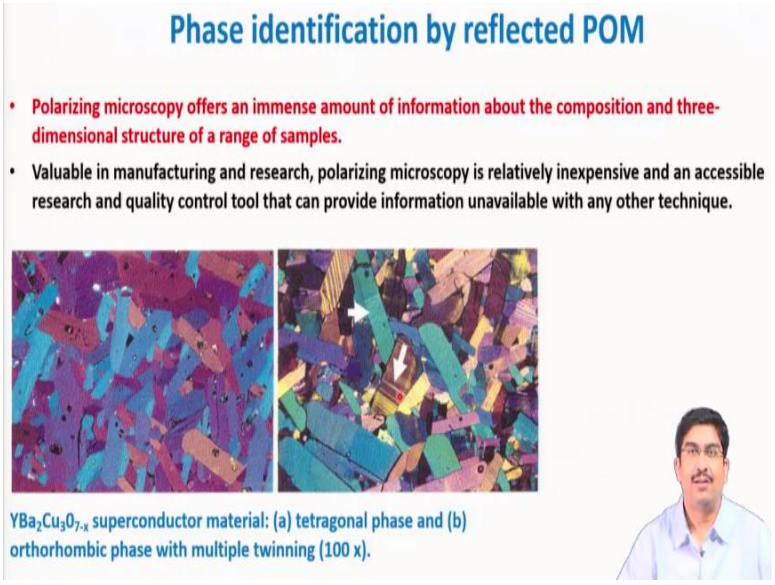
You need to know how much rotation you are giving to your specimen on the stage itself so that you know that possibly which directions you are possibly because you know the crystallography all the directions are related by certain angles. So, by rotating this you are rotating the specimen, you are bringing some other directions. So, you can possibly very carefully if you calibrate this you can know that exactly how much you rotate the specimen and which directions you are now bringing there.

Also, you need to have a strain free objective because if you have some stresses on this glass after this objective lenses are prepared, same thing forgoes for strain the condenser. So, condenser lens and objective lenses if they have stresses, then of course the polarized light that is produced will be having a completely different plane altogether.

The polarizer which is working there you may have set it for something, but ultimately what the polarizer light when it encounters this objective lens of condenser lens it will have completely a different polarization. So, the cross-polarization condition may or may not work. So, in order to reduce that you tend to have a strain free objective as much as possible. And generally, the objectives that is designated for polarized light is a slightly different type of objectives.

And if you have this polarized objective meant for, I mean completely stress free polarized if you want to do polarized light microscopy on your machine, then generally you are provided with a different type of objective lens altogether which cost you some money. And you will generally tend to have this kind of some sign written over it like P, PO, Pol and something, so from there you can make out that objective lens is meant for doing polarized light microscopy.

(Refer Slide Time: 30:21)



Now, I will give you two examples, very good examples of this polarized light microscopy in reflection mode. By the way, you can use polarized light microscopy both in reflection and in transmission mode, it does not matter. By now, you have understood that all of this contrast enhancing modes work fine with either reflection or it depends reflection and transmission is just the way you illuminate them, just the way you have the microscopy configuration.

These are two different microscopy configurations which you can use for various modes, all modes are possible. This is one nice example where you can use this polarized light microscopy for phase identification. For example, if you have this, this is a yttrium barium copper oxide superconductor material and this is a tetragonal phase, which has certain kinds of crystallography.

As I already told you that for crystalline material, this entire polarized light microscopy the concept is related to the crystallographic directions, crystallographic planes, direction because the refractive index, they are anisotropic material refractive index changes as per the directions in the material. So, this is very sensitive out of all the different modes, which are possible, we will discuss about another mode just after this class which is an extension of polarized light microscopy.

So, digital interference, differential interference contrast. So, this interference contrast in polarized microscopy, these are the two modes which are sensitive to crystallography

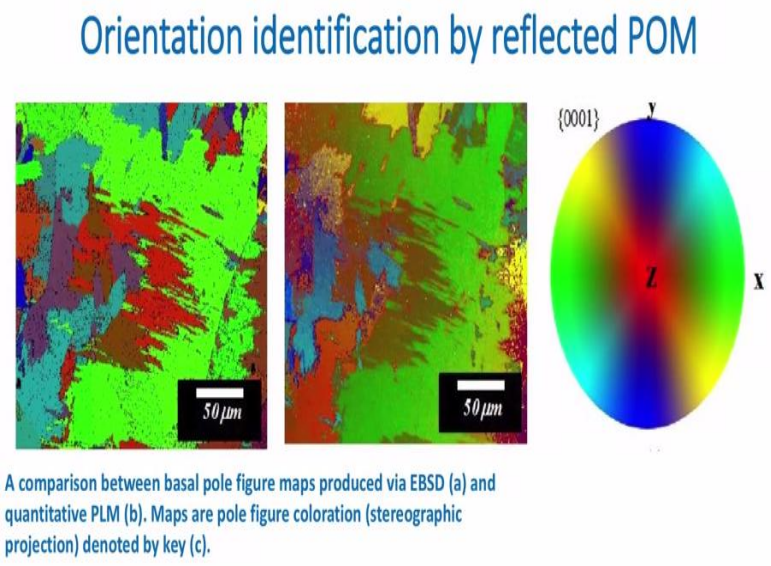
because the way images are forming here polarized light that is sensitive to refractive index change. So, when you have a different phase, I mean you have a phase transformation from one phase tetragonal phase it is transforming to an orthorhombic phase, of course you have a completely different kind of refractive index.

I mean the change in refractive index was the dependence or refractive index on the orientation completely changes. So, definitely if you keep your polarizer analyzer everything you take images for a tetragonal phase and you take an image from orthorhombic phase, you will get completely different colors altogether, because again the change and you have a lambda plate there.

So, the entire material or entire prints or entire mechanism of image formation in polarized light changes if you change the phase, if you change the crystallography. And even better if you look at this region, this is basically twinning. A twinning means a slightly different change in orientation just like you have a change in orientation between this grain and this grain and same thing within the grain also you have this change just because of twinning.

So, this is giving you a very nice sample and only under polarized light you are getting this kind of sensitivity, this kind of twinning you can identify. You cannot do that if you use bright-field images. So, that is one very good area for polarized light microscopy to be applied or people in applying in study phase transformation.

(Refer Slide Time: 33:20)



A comparison between basal pole figure maps produced via EBSD (a) and quantitative PLM (b). Maps are pole figure coloration (stereographic projection) denoted by key (c).

Another one as I said it is a semi-quantitative method, you cannot really get a complete idea about the orientation, but you can try to get some amount of idea regarding your orientation if you nicely calibrate this image formation, the way the images are forming and if you can identify at which angle you are basically rotating a specimen and so on. Then you can calibrate these colors or the calibrate the signal that you are getting with the orientation in the material.

So, this is again shown two examples. One is a EBSD electron backscatter diffraction, which is a typical way of finding orientation because there the signal generation the way it works completely depends on the orientation or crystallography and you can measure that that is a quantitative data. But you can get a same kind of an information from or at least not up to that level but lower level.

You can get information about the orientation of various features that is producing or that is there in the microscope or in the material you can get that orientation related information by taking this polarized light microscopy image. You can basically here what you can get is that what is the orientation of certain crystallographic direction in this with respect to the external frame of reference.

So that kind of studies you can also do using a polarized light microscopy out of all different kind of microscopy techniques that you have. So not only polarized light microscope can give you nice, bright, colorful images which you can also enjoy. So, I wish please go to internet, try to check polarized light microscopy and you will get fascinated microscopy images developed by chemist and developed by minerologists, geologists and so on.

But other than that, you can also try to use polarized light microscopy for phase transformation, orientation determination and so on. It is a very nice utilization of this out of all different modes of optical microscopy. This is a very nice utilization of one at least this polarized light microscopy. So, with this we are stopping here and we will continue in the next class. We will continue with another technique that is called differential interference contrast. Thank you.