Materials Science and Engineering

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Lecture - 06
Optical Microscope

Hello friends, as you were seeing me using a magnifying lens, ok. We are going to start our lectures on Optical Microscope. Of course, we choose some magnifying lens like this and how they are arranged to get a magnified image in the eyepiece ok. So, to start will I will introduce you to optical microscope first and in that, we will look at some concepts like magnification, ok.

And what is useful magnification, see we cannot keep on magnifying any image to any extent and ultimately, there has to be some useful magnification. So, we will try to understand that what do we mean by this concept of useful magnification? And that is very intimately related to the resolution ok; that whether you are able to resolve the features which you are interested in or not.

So, this useful magnification is related to resolution and then, we will see 2 other concepts which are related to microscope. These are depth of field and depth of focus,. So, all these terms we will see. We will try to analyze what do we mean by each of these term ? And; how it is useful to get a good microstructure in a sample. To start with this is your high school physics, ok. There is a thin len, thin lens equation here.

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Image formation by single lens

Thin lens equation

$$\frac{1}{f} = \frac{1}{u} + \frac{1}{v}$$

Magnification (M)

$$M = \frac{v}{u}$$

$$M = \frac{f}{u - f} = \frac{v - f}{f}$$

So, let me use explain it within a schematic here. What do we mean by v u and f, ok.

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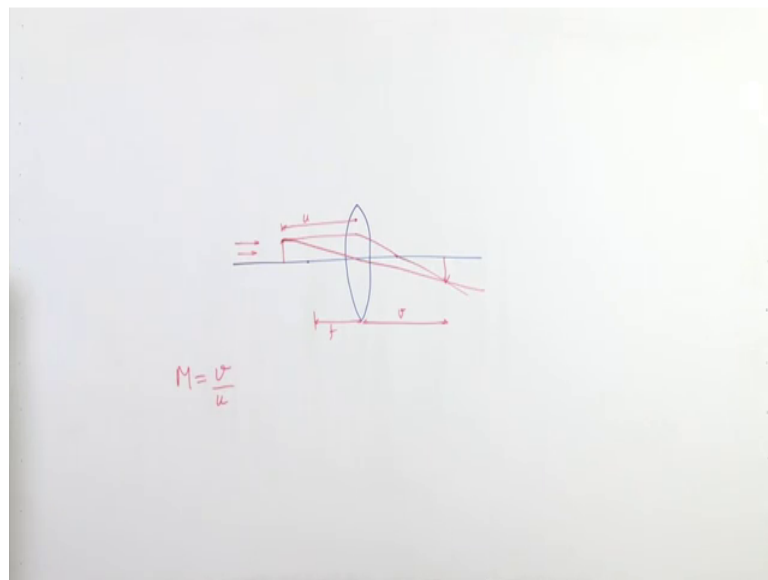

So suppose, we have an optical axis of a thin lens. Of course, I am exaggerating the convexity of this particular lens here and these are the focal point of this particular lens ok. And let us say I have, I am keeping an ob, the object here away from the focal length here, f. This is my object and suppose, we there is a parallel ray which is coming and striking this particular object.

Then, how the waves will propagate from here and interact with the lens ok. So, after interacting with the lens; it will go through the focal point here, ok. Another ray, which is going through the center of the lens ok. There, there will not be any divergence. It will go straight like this and you will see inverted image forming at this particular location ok.

And now, we can define all these values ok. So, the u is your distance from the lens of the object; v is the distance of image from the object and f is your focal length ok. And this is the thin lens equation.

Ah of course, you will have to keep in mind the sign convention which we use here. Any le any distance, we are measuring in the direction of the ray will be positive and anything which we are measuring in the direction opposite to the rays here, will be negative ok. So, for all your calculation please keep the sign convention in mind.

Your, I can also define a magnification here which is given by the distance of the image from the lens and the distance of the object from the lens. And of course, if you replace v and u with f here, you will get the other 2 relations which is shown in the slide.

This is how, this is what is used in a compound microscope or a metallurgical microscope ok. So, let me tell you the whole illumination schematic of a compound microscope which is sometime you can call as Metallurgical Microscope also. To distinguish a metallurgical microscope from a biological microscope which you might have seen in your a school.

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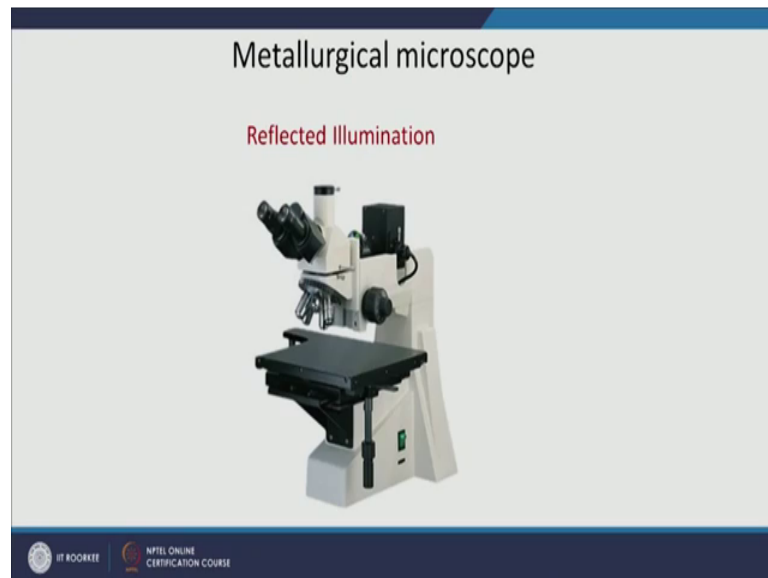

So, ok if you have done some biological experiments.

In case of metallurgical microscope, the illumination is by a reflection.

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So, the rays gets reflected from the object and then, it again go goes back to in the objective and then, you form the image. Whereas, in case of biological samples, the image transmit through the sample and then, it goes to objective and you form the image.

So, in case of metallurgical of microscope, the illumination is by reflection and I will just show you what kind of schematic is there; the ray diagram for that. This is also called a compound microscope because more than 1 lens will be used to form the image. So, first I will draw the parts of the, of a microscope. So, you have a source.

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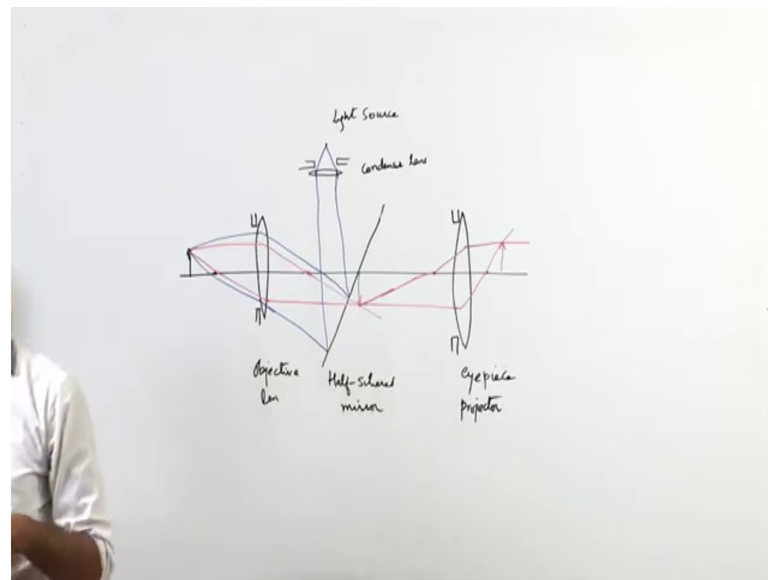

A light source. I will write on that, a light source ok. Then, you have apertures here and then, you have a what we call as a condenser lens ok, condenser lens.

From there, we will come to the other part of the microscope ok. There will be a half silvered mirror here ok; which will divert the rays coming from the condenser lens toward the objective lens ok. So, there will be an objective lens here. This is a objective lens ok.

And this is half silvered mirror. Of course, there will be some focal length for these particular lens.

Then, after this mirror again there will be another lens here which we will call as an eyepiece. This can be called as eyepiece or this, this can also be called as projector lens ok. The image can be projected on a screen or you can take this image and show it in the on your computer monitor ok.

So, now let's see, I will start drawing the rays which are coming from the light source ok. So, it will be diverging from the light source. So, this condenser lens will be, this light

source will be at the focus focal point of this particular condenser lens. So, after this, it will go parallel ok; something like this ok. And then, this will be put on the objective lens and from there whatever object is placed here ok.

Suppose some object is placed here. These rays will fall on that. there will be some diversion towards the object here; after falling on the object my rays will now gets reflected toward the object to back.

So, it will go again, towards the objective. From this and after going from here, it will be parallel to the optic axis here. And this will be going through the focal point and it will make another image here, which will be intermediatary image ok; a intermediate image will be there.

And from here, again, I will also have to draw focal point for this particular lens. So, this ray you will go like this. This one is going like this and after passing through the lens it will become parallel to the optic axis and this will go through the focal point to give you a magnified image here.

So, this is the whole schematic for a compound microscope, which is used in this kind of geometry reflected microscope for metallurgical application.

Now, we want to understand as you can see here, if I keep on putting lenses here ok; for each lens there will be some magnification ok. And I can keep on adding the magnification, if I keep on adding the lenses here and that way I can go to any magnification I want. But we cannot do that because with position of each my lens the image ditoriates and that can be very nicely shown by this particular slide here ok.

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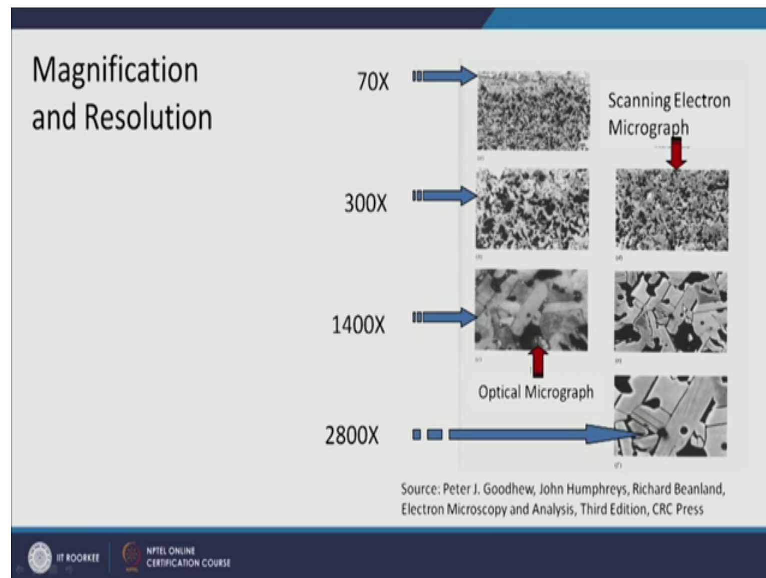

Where this 3 images are taken from an Optical Microscope and the same from the same material another 3 image were taken from a another type of microscope which is called the Scanning Electron Microscope ok. And you can see that up to 300X, if you see both the images they have almost similar features; the sharpness of the image is same. The clarity of the image is same ok.

But, if you come to 1400X here ok; now, you can see that the features, which are shown in this optical micrograph are little bit blurred. They are not very clear ok. Whereas, if you see the scanning electron micrograph here, the image same image is very sharp ok. All the features are very sharp.

You can distinguish between the 2 different features or you can be, you will be able to see the sharp boundary between these 2 features here; where in this case, if the boundary is not very sharp.

So, you are not able to resolve it ok. So, in this compound microscope as you can see, I have magnified this particular image of this particular object here, which is an intermediate image. Again, I can put another lens here and I can magnify the image further and I can keep on putting the lenses here and I can keep on magnifying the image. But, there is a limit to that I cannot keep on doing that because if you keep on putting lenses here, the resolution of the image will decrease with the magnification in case of optical microscope, ok.

The reason and to understand this resolution that what do we mean by resolution? ok. I will come to this next slide here.

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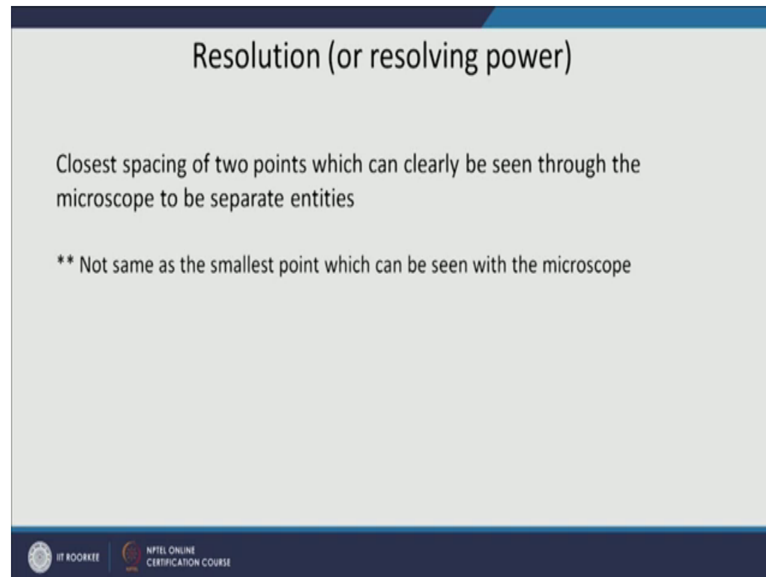

The resolution is basically the closest spaced spacing between two points which can you can clearly see through a microscope as separate entities. You must be remembering when you jo any friend who actually wears these kind of spectacles ok. We usually joke. We will take his specs out and we will put two fingers and we will ask him, [FL] how many fingers are there ok. And you will be joke whether he will be able to say 2 or 1 or whatever.

So, what we are trying to test for that particular friend that whether he is able to resolve these 2 fingers or not without his a spectacles ok. So, what we do in these fun activities? We are defining resolution ok. So, that is what is here also that whether I am able to see 2 entities which are there in the sample, features which are there in the sample. Whether I am able to see them as 2 different features or 2 different entities or not ok?

So, it is not you can should not confuse it with the that it is the smallest point which can be seen with a microscope. A smallest point can be even smaller than the resolution ok. A smallest point you can see is much smaller than the resolution. What we are trying to say is these 2 small points. How closer they can be? And we were still here, will be able to say that there are 2 different features or 2 different entities ok. They should not be seen as 2 ones 1 single uni unit or single entity.

To clarify this idea more and why this resolution problem comes in microscope or any optical, optical system this is the concept.

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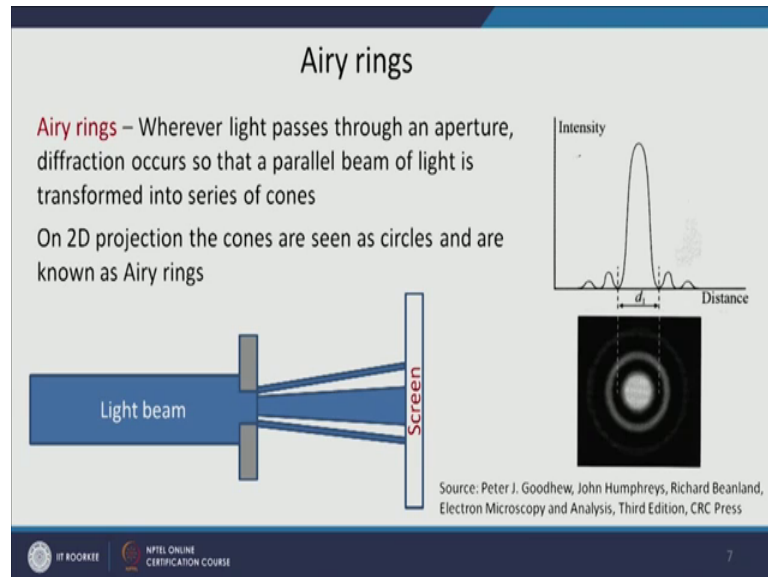

What is, what we know as Airy rings ok. So, The Airy rings means when a parallel beam of light, when it passes through any contraction or any location where it is kind of constricted, it is allowed to go through a aperture ok; then, it gets diffracted ok. And during this diffraction as you can see in this particular slide, you get 1 central spot here; a bright spot. Then, you get another ring here with another brightness. Then you get the third ring here and so on.

So, so, 1 single light beam is divided into 1 beam a central spot; then, another ring and then, another ring ok. And the intensity is also plotted here ok. So, you can see the central spot is the most intense. Then there is a second ring, intensity is much lower than the third ring and so on. And between 2 maxima, there is always going to be a minima here and this is what is shown here in the schematic, a light beam going through opening here and it gets diffracted ok.

So, you have a central spot here and the 2, the ring which you get is because of this. Because this is a section you see it as a as a 2 dimensional object, but it will be actually will be a ring. So, when it goes through this opening it will become a cone; central beam and a cone, 1 first cone, second cone and so on and which you on the screen will see is a centrally spot first ring and second ring and so on ok.

So, this parallel beam of light, when it goes through any opening it; it has this formation of Airy ring. Now you can understand from this light source, we had were first opening here and aperture a lens is there.

So, there must be a formation of Airy ring here. Then, after reflection it goes through another lens here. There will be another set of aperture, I have not drawn here ok; you can draw here a set of apertures. Again, it is going through an opening ok. Again, there will be formation of Airy ring. You come to the eyepiece, again, there will be some aperture here also ok.

Again there is an opening again, that. So, this particular source has undergone this kind of airy ring formation at 3 places and it each place, the beam is dividing into multiple beams ok. And why it will bring down the resolution? We will just see in this next slide ok.

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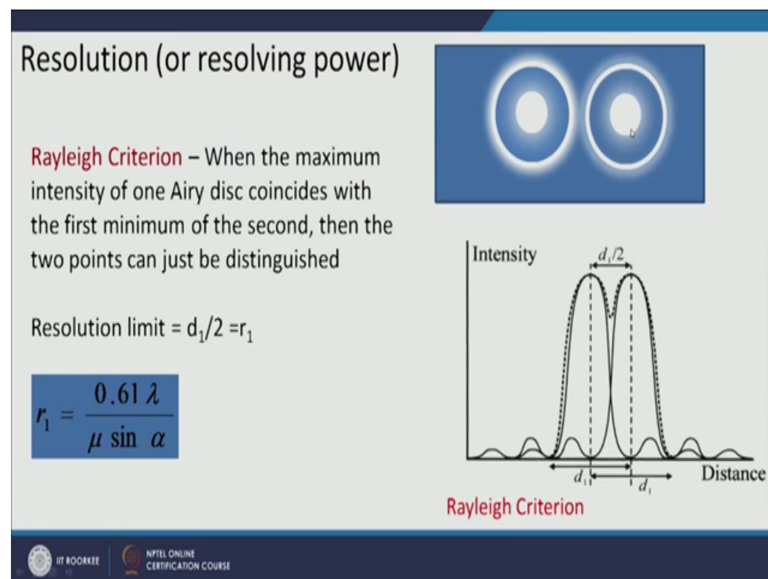

And that there are this 2 by 2 sources ok. 2 sources means there are 2 features which are emitting light or a light rays ok. So, how close we can see them to a separate ok. So, just you see this animation that I am bringing these 2 sources close to each other and how much closer I can bring them together that is what we want to see ok.

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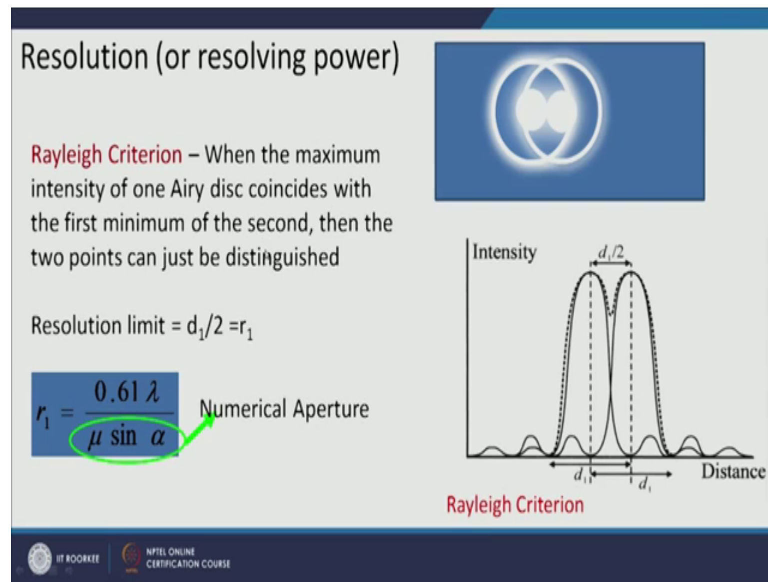

So, this is what is the animation ok. So, you can see that my this bright spot is almost coinciding this, with this bright spot here ok. If I bring any for any closer than this particular position; then, I will start looking it as a 1 single entity ok. And at this position, what is happening is the maxima of this particular feature or a spot is coinciding with the minima of this particular spot and that is what is plotted here in terms of intensity ok.

For first one, this is what is the maxima ok. For the second beam, this is the maximum and it minima of that is intra is coinciding with the maxima of the first one. Similarly here, the maximum of the second one is coinciding with the minima of the first one.

So, this is the closest I can bring them together, if I go any further than that ok. Then I won't be able to see these 2 features as separate entities and that is my minimum distance given by d 1 by 2; d 1 is your diameter of this particular central spot ok. So, this is equal to d 1 by 2 ok. That is what we are saying is it is my resolution limit or r 1 ok. And which is given in terms of wavelength of light or and I will write down, what do we mean by all these parameters here, in this particular equation.

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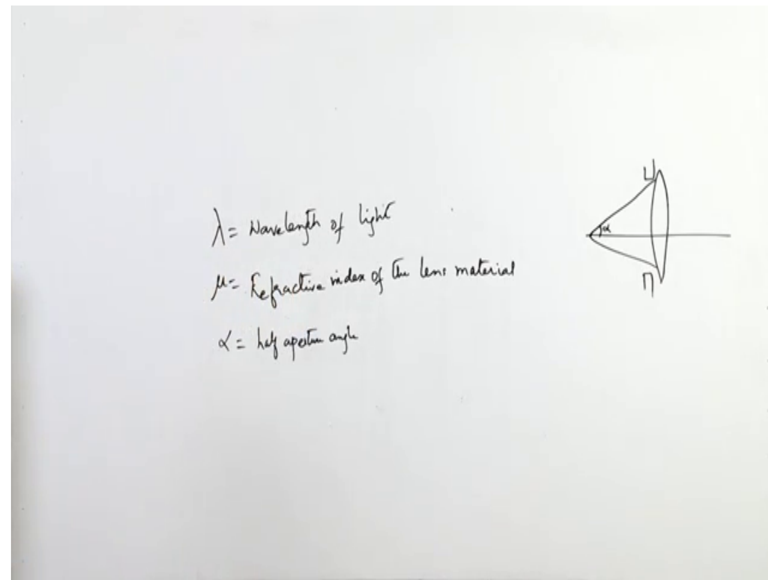

So lambda is wavelength of the light we are we using; mu is refractive index of the lens material and alpha is the half aperture angle ok. I will just show a schematic for that also to show you what do we mean by that ok.

So, this is my optic axis and this is my lens. So, the and suppose this is my aperture ok. So, a light beam going like this just next to the aperture; what is the maximum angle it can make ok, half of that. So, alpha will be this angle, half aperture angle here.

So, this is my resolution can be expressed in terms of the wavelength which you use for a particular microscope. What is the refractive index of the lens and what is your half aperture angle? Ok. Refractive index of the lens you can also understand ok, if you wear this glasses or some of your friends wear glasses ok.

So, when they go to any optical shop and ask to make a new glasses for him ok. He will ask that we you want a normal kind of lenses or you will want this higher refractive index lenses ok. And what they do is when you have this higher refractive index lenses, your if you if your friends the power is very high. For example, minus 3, minus 4, minus 5. Then, the thickness of the lens is going to be very high; if you are not using a higher refractive index glasses.

But nowadays, what we get in the market these are all higher refractive index glasses of course, made of polymers ok. And what they do is they bring down the thickness of the

lens. So, same power you can get with a very thin lens and that is what is here also that I can use a higher refractive index material to have better [noise resolution.

So, in this case the resolution will be given by this ok. And mu sin alpha which is shown here is also called as numerical aperture of the that particular lens.

So, this criteria a was given by ray lord Rayleigh and that is why it is called a royl Rayleigh Criterion ok. And it is explained in the statement here that what do we mean by that and that defines my resolution.

So, now, you can understand that when my light goes through 2 different apertures and in each aperture there is going to be formation of this Airy ring and these Airy rings from different features are going to interact with each other.

So, it will going to reduce the clarity of the image and I won't be able to see my all the features very clearly ok; because all the airy rings will interact with each other, with each other and bring down the clarity of the image.

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To get best resolution

λ ↓
α ↑
μ ↑

$$r_1 = \frac{0.61\lambda}{\mu \sin \alpha}$$

Example
- 400 nm – green light
- sinα approaching 1 - by using as large an aperture possible
- μ (1.7) can be increased by using high refractive index medium

Resolution of human eye – 0.2 mm

Now, to get best resolution ok, how I can improve resolution of a microscope, ok? What I have to do is? I have to bring down my lambda as you can see my objective is to reduce r 1 ok. So, r 1 I can reduce, if I bring the wavelength of the light I am using or any electromagnetic radiation you can say because in electron microscope ok, we use

electrons to do that and the wavelength there is very small ok. So, my resolution is very high in case of electron microscope.

In optical microscope, we have to use visible light only and for example, we can use a green light which has a 400 nanometer wavelength ok. I can again, increase the resolution or reduce the r 1 by increasing the angle alpha. So, I can open up the aperture to get maximum resolution or I can use a higher index glass or glass material ok. So, that will also will have better, will give you better resolution ok.

So, you can have sin alpha almost approaching 1- by using as las large and aperture possible mu maximum. Now I think what type of material, we have in optical microscope; it gives mu of 1.7 ok. So, these are all higher refractive index medium that you can use ok. Just for your information resolution of a human eyes only 0.2 mm ok.

So, this you can keep in mind and you can use this value to find out that what can be the maximum useful magnification which you can have ok; if your resolution is given by all these numbers. You have just put all numbers like 400 nanometer here.

Sin alpha approaching almost 1; mu 1.7 and see how much is the resolution and that resolution has to be a kind of you the final resolution of your human eyes 0.2 and this is your resolution you which you can get from the microscope. So, the diff the their ratio will give you the magnification.

Now, after knowing the resolution there are some 2 more terms which can be of interest to any microscopist is, what is the depth of field. So, let me again draw a schematic here to show you what do we mean by depth of field.

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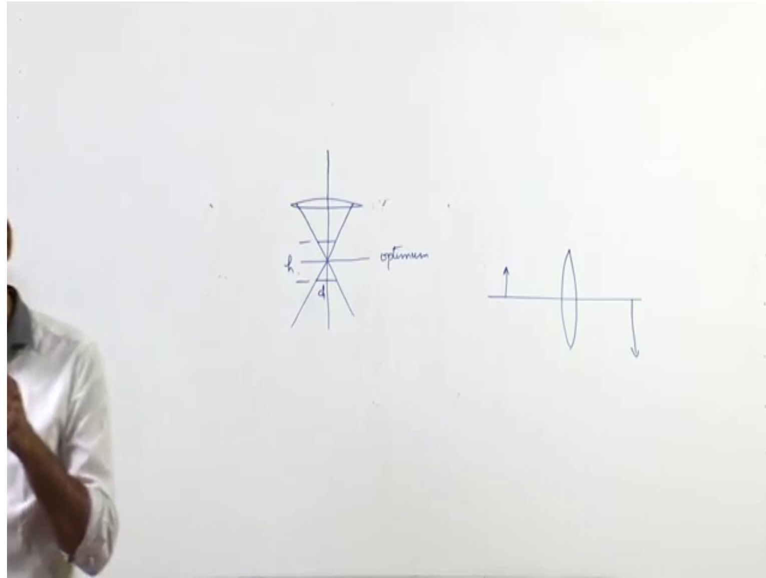

So, this is my objective lens and this is my optic axis and my rays are coming like this from here. They are intersecting at the focus there ok. So, from there, I can tell you that this is my optimum focal distance or optimum focus, I will get or best image I will get, if I have object here ok. So, this is where I will have optimum focus or optimum image.

Now, we have just seen that I have, I am going to have some resolution possible means I can bring 2 objects closer up to a distance of d 1 and still I will be see able to see them as 2 separate entities ok. So, from optimum focus, if I go up to any distance which is d here ok.

I should be able to not see any difference between the image ok. So, if I my image plane or my object plane varies between these 2 limits, I won’t be see able to see any difference in the image; because it is still in the my resolution limit. and that distance I am giving by h ok. So, that is what is shown here, which is given by here 1.22 lambda upon mu sin alpha and tan alpha. ok.

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Depth of field

The range of positions for the object for which our eye can detect no change in the sharpness of the image is known as the *depth of field*

$$h = \frac{1.22\,\lambda}{\mu \sin\alpha \tan\alpha}$$

So, that is my depth of field. Of course, it is different dependent on alpha here as you can see ok.

So, if I have a smaller aperture, I will have better depth of field. We can do calculations based on this in assignment. Then, you will be able to understand it better that if I reduce the alpha., Then I will have a better resolution or better depth of field. As you can see is if I reduce alpha sin alpha and tan alpha both values will be lower ok. So, the h will be higher ok.

So, if I reduce the apertures ok, angle; then, my depth of field will be higher. But if you remember, what we discussed in the resolution my resolution will be poor. My depth of field will be higher; my resolution will be poor. I don't know if you have used this SLR cameras or maybe some of your friends have this SLR camera ok.

You use this idea of aperture to increase or decrease the depth of field ok. Suppose, this you cannot do in a norm normal my normal cameras, but SLR camera you because you do manual adjustment. So, if you reduce the aperture ok, if you have a SLR camera; if your friend has sna have an SLR camera, you can ask him that what you do ok?

So, if you want to have image such that I am also in the focus and something which is in front of me maybe and let's say 10 meters in front of me that guys also in focus. What he

will do? He will reduce the aperture means he will reduce this angle alpha and then, both the subject will be in focus.

But he wants to have a kind of bring a artistry into this thing; sometime you see these images where a flower is in the front and you want to take a photograph of somebody who is behind that and you want to keep this out of focus and bring that subject into focus. Then, you increase the aperture. Now you can focus only at 1 point either this flower or you can have that subject in focus ok.

So, these apertures actually you play with in SLR camera to change the depth of field and that is what is here ok; your depth of field will change if I change the aperture angle ok. And you can play with that of course, in optical microscope the depth of field is very small and that is why you will understand that why we have to do polishing, when we go to next lectures that why we do polishing because in optical microscope the depth of field is very small.

And from that another concept which comes is called Depth of focus ok. The depth of focus is basically as depth of field is for the objective lens; the depth of focus is for the eyepiece and you can see that it is a magnification.

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Depth of focus

The range of positions at which the image can be viewed without appearing out of focus, for a fixed position of the object

$$dv = -M^2 h$$

Change in distance of image formation

Magnification

Depth of field

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Square of magnification times, the depth of field. So, it is very high. So, if this is in micron and suppose, I am using a magnification of let's say 100. So, it is 100 into 100

one 10000 times of the depth of field of micron length ok. So, it will be something in mm or maybe centimeter ok.

So, depth of focus means see when you see all this optical that this is my object; this is my lens and some image is forming at this point. From this, we understand that I have to be exactly at this position to see the image because this is where the image is forming. If I go anywhere in this side or on this side, I should not be able to see the image ok. It defines a very clear plane on which the image is forming.

So, for a person if he suppose, he is seeing a an image in a microscope; he has to be all the time at this particular location to see the image and that is a you are putting a great restriction on him just to have it every point suppose, he is watching that image for 10 minutes. For 10 minutes he has to be at that particular position to watch this, the image ok. But that is not true.

Actually, you have leeway here that you can vary between this two and still you will be able to see the image because you have some depth of focus here. And which is some order of magnitude times the depth of field ok. So, it gives you some relaxation that I can be little bit changing my position and I still will be able to see the image. So, that is what is depth of focus ok. So, with this I think we have covered all our, terms, which are related to microscope ok.

So, thank you here., We will now see the how we actually prepare sample for looking at the microstructure and that is what we call as metallography.

Thank you.