Natural Dyes

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Lecture No. # 33.

Now, we will go back again to natural dyes. We have still not talked about carotenoid dyes. And today's lecture is dedicated to carotenoid dye and the dyes source is delonix. You must have seen this flower all around the campus, all around your school, college and in your vicinity. And this is that famous delonix regia or gulmohar plant, which is a reddish brown in color rather orangish red in color and it is so brightly colored that one cannot escape not seeing this flower ever in the life.

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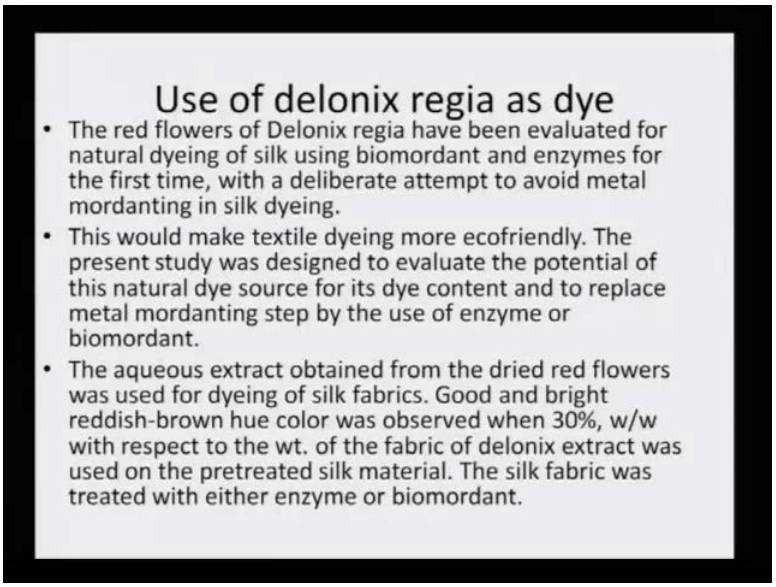

So, let us take a look at this source of carotenoid natural dye called delonix. Use of delonix regia as dye, the red flowers of delonix regia or gulmohar as what I told you, have been evaluated for natural dyeing of silk using biomordant and enzymes for the first time, with a deliberate attempt to avoid metal mordanting in silk dyeing. Because as I told you that although we need bright colors, but in order to obtain bright colors it became mandatory to use the metal mordants and the metal mordants that were used

popularly in the industry were alum, iron, copper, chromium and tin. So, we were trying to look for an alternative where this metal mordanting step can be eliminated.

Use of delonix regia as dye the red flowers of delonix regia or gulmohar as what I told you, have been evaluated for natural dyeing of silk using biomordant and enzymes for the first time, with a deliberate attempt to avoid metal mordanting in silk dyeing. Because as I told you that although we need bright colors, but in order to obtain bright colors it became mandatory to use the metal mordants and the metal mordants that were use popularly in the industry were alum, iron, copper, chromium and tin. So, we were trying to look for an alternative where this metal modanting step can be eliminated. This would make the textile dyeing more eco friendly. The present study or the discussion was designed to evaluate the potential of this natural dye source for its dye content and to replace metal mordanting step by the use of enzyme or biomordant.

The aqueous extract obtained from the dried red flowers was used for dyeing of silk fibers. Good and bright reddish-brown hue color was observed when 30 percent weight by weight with respect to the weight of the fabric of delonix of the fabric, delonix extract was used on the pretreated silk material. The silk fabric were treated with either either enzyme or biomordant. So, we did not want to use metal mordant, at the same time we wanted to show that this reddish color - hue color can reddish orangish color can be maintained and it is a very good potential dyeing falling in the category of carotenoid dyes to be used for silk and in the silk industry we were trying to popularize.

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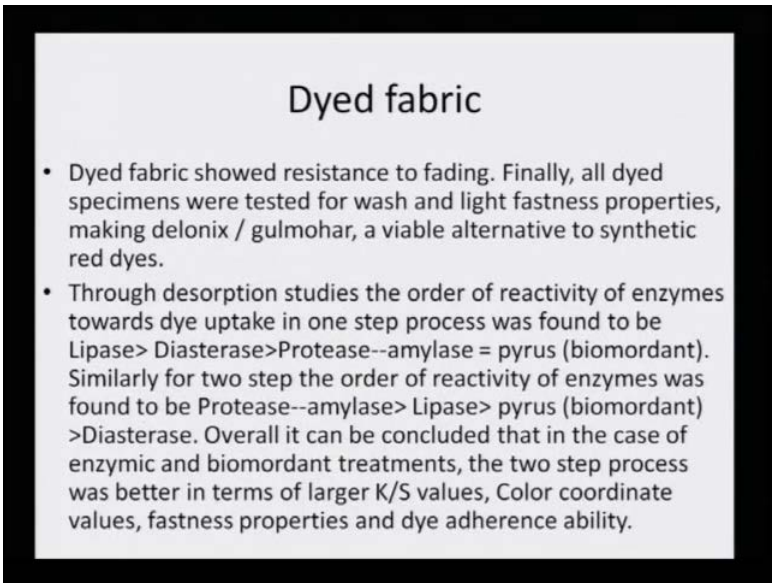

Dyed fabric showed resistance to fading. Finally, all dyed specimens were tested for wash and light fastness properties, making delonix or gulmohar, a viable alternative to synthetic red dyes. So, our soul aim was red can be use this particular dye for the preparation of red color material. Through desorption studies the order of reactivity of enzyme towards the dye uptake in one step process was found to be that lipase was best, second best was disasters and the third best was protease - amylase combination. And this was almost equal to the bio mordant that is the pyrus. Similarly, for two steps the order of reactivity of enzyme was found to be that protease - amylase combination was better than lipase which was better than pyrus that is the bio mordant, and diasterase was found to be least effective towards the silk fiber.

Now, as I by now you will be able to understand why I am trying to introduce too many things to you, because you have already learned these things. You have learned the treatment of silk with enzymes and ahead specifically told that there is a dye to enzyme compatibility. So, fabric can be treated with any of the enzyme. But which dye will be more compatibly accepted by this treated fiber is what matters. And therefore, we did two things; we first did one step, that means everything was added into the same dye bath or we did step wise which mean that the first the enzyme was pretreated to the fabric and secondly then it was dyed. So, both the procedure then show different kind of effectivity and that is what you know we have tried to understand the chemistry by doing the desorption studies.

See we can do it either ways, we can do you know different types of dyeing and then evaluate the CLI CIE lab values and look at the K by S values. But at the same time we also introduce that in order to understand where the dye uptake has been better than the other one; this was the desorption studies were followed. And it showed clearly that in one step lipase was the desired enzyme and even the bio mordant showed very good result. Similarly, in the two step - the protease amylase combination was very good and that was even the same thing was observed for the pyrus bio mordant. You may recall that in pyrus perschia, copper was evaluated as the main bio mordant; so, as the main mordanting metal by atomic absorption.

So, the same bio mordant we have used in order to replace the metal mordant, because copper sulphate I had very clearly told you that it it comes under the hazardous chemical. And so we should avoid using this direct metal mordanting, but because the plant material, the bio mordant has just very minimal amount of the metal sufficient for the purpose of mordanting; that is why it is safer to handle. And another advantage is that while we are you know extracting the dye from the plant material. In this case we are talking about delonix regia flowers, the same time we can add pyrus perschia fruits into it and it will do the simultaneous mordenting. So, no need to do different steps of mordenting in the simultaneous mordenting when we are preparing the extract of the delonix. At that point of time only we can add the pyrus perschia. So that is how we try to look at the minimization of the steps. We also try to look at the overall you know processes how it can be made industrially more friendly process.

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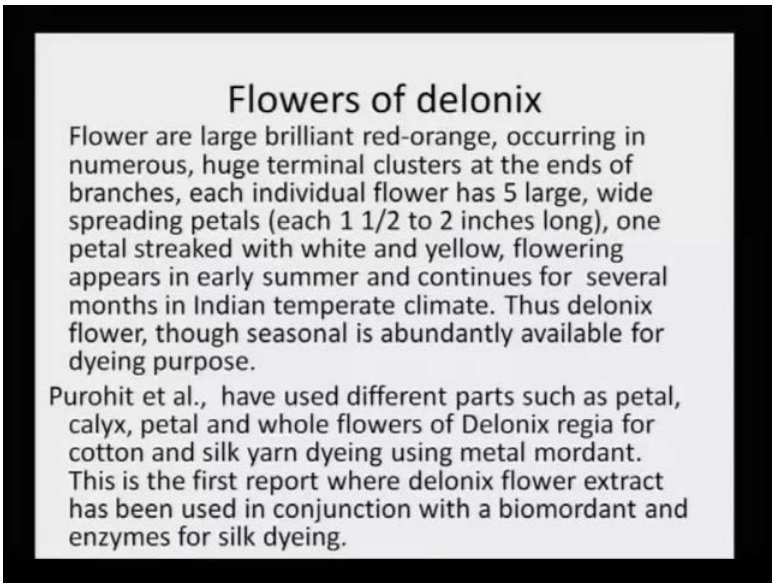

Flowers of delonix regia, let us now try to take a look at you know what is the composition of the flowers. Flowers are large brilliant red-orange occurring in numerous, huge terminal clusters at the ends of the branches, each individual flower has 5 large, wide spreading petals almost like 1 to 1, 1 and a half to 2 inches long, one petal streaked with white and yellow, flowering appears in early summer and continues for several months in Indian temperature and climate. Thus delonix flower, through seasonal is abundantly those seasonal is abundantly available for dyeing purpose. So, there is no dirt of the plant material. There is ample of flower available and the season is pro long to a very long time.

So therefore, one can make use of this rich colorant and for the industrial purpose. Purohit et al, have used different parts such as petals, calyx, petal and the whole flower of the delonix regia for dyeing cotton and silk yarn, but they have used metal mordant. What we tried to do was to minimize the step of dyeing and to popularize the extraction process, to popularize the dyeing process using enzymes and bio mordants. So, that is where we tried to bring in the innovative technology for silk dyeing with delonix regia.

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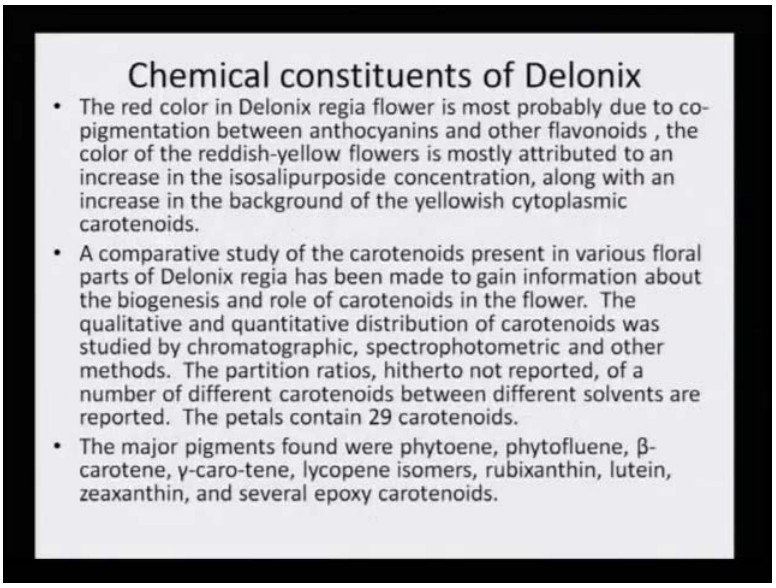

Let us try to now look at the chemical constituent of delonix. The red color in delonix regia flower or gulmohar flower is most probably due to the co-pigmentation between anthocyanin and other flavonoids. The color of the reddish-yellow flowers is mostly attributed to an increase of isosalipurposide concentration along with an increase in the background of the yellow cytoplasmic carotenoids. So, there is a major composition of carotenoids apart from some anthocyanins and flavonoids. I have told you time and again that natural dye dyes do not exist as a single dye, unlike the synthetic dye; just a while ago we were taking a look at the sulphur dye, there when we talk about sulphur dye it is just 1 molecule. But here when we talk about the extract of any flower which is a source of dye, it has anthocyanins dyes, it has flavonoids dyes, but a major component is carotenoid dyes. And therefore, we will call it as a class of carotenoid dyes. you will just take a… You will see for yourself there are large numbers of carotenoid molecules.

A comparative study of the carotenoids present in various floral parts of delonix regia has been made to gain information about the biogenesis and the role of carotenoids in the flower. The qualitative and quantitative distribution of carotenoid was studied by chromatographic and spectrophotometric and other analytical methods. So, it has been found by the methods that are normally used for the analysis. Chromatographic techniques help in separation of the various colorants. Spectrophotometric methods help in finding out the structural details of the molecules that have been separated out and so on and so forth. The partition ratios, hitherto not reported of a number of different

carotenoids between different solvents are reported. The petals contain about 29 carotenoids. So, do not you thing that now it is a class of carotenoid rather then I would put it under the category of anthocyanins or flavonoids. The major pigments found were phy phytoene, phytofluene, beta carotene, gama carotene, lycopene isomers, rubixanthin, lutein, zeaxanthin and several epoxy carotenoids. So, as many as 29 carotenoids have been found in delonix regia.

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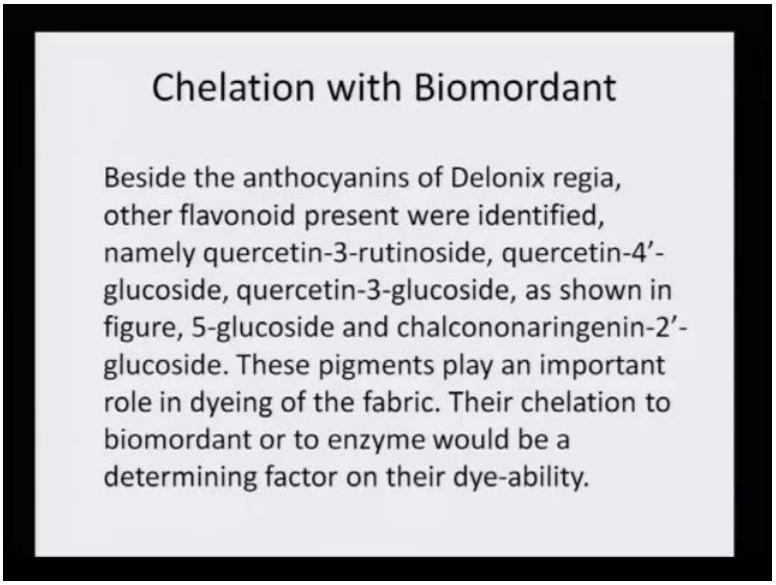

Chelation with bio mordant: Obviously, when we are talking about the structure; we are trying to look at various points at which the biomordant or the enzyme can link up. Because unless and until the structure of the these carotenoid have linking groups or in other words the you know, the attachments or the functional groups that can help or the oxzochromes that can help in binding, it will be of no use and the dye will not fall into the category of a good dye. Besides the anthocyanins of delonix regia, other flavonoid present were identified namely quercetin-3-rutinoside, quercetin-4 prime-glucoside, quercetin-3-glucoside as shown in the figure next in the next page in the next slide I will show you. 5-glucoside, chalcononaringenin- 2 prime-glucoside these pigments also play a role in dyeing of the fabric. Their chelation to biomordant or to enzyme would be determining factor of their dye ability.

But the dye adherence is probably taking through this hydroxy containing compounds, because you have learned the structure of beta carotene and you have seen that it has lot

of conjugation. And it has you know alkyl groups, but it does not have the hydroxyl group. So, similarly the flavonoids which have the hydroxyl group which are present as coke pigment along with the carotenoids are actually participating in the dyeing process. But the color attribution is coming from the carotenoids which are 29 in number.

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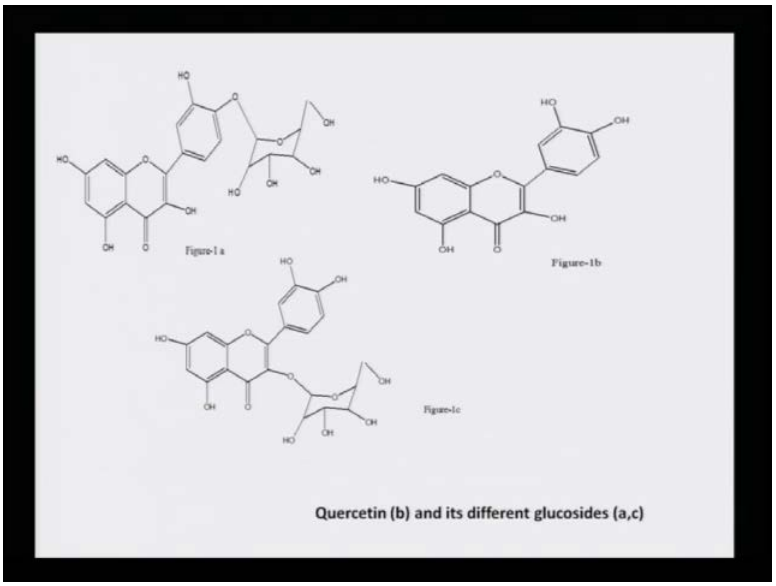

So, if you look at these three different flavonoids; you will see that all of them have ample number of, because they are all glucoside and two of them are glucosides. 1 a and 1 c and they have as many as 4 to 8 hydroxyl groups which can participate in the chelation of the biomordants, also in these are curicity molecules and their glucosides a and c, and the curicity molecule per say as 1 b. Now, when you look at these flavonoids, you can very well understand that the alpha hydroxy is the 1 which helps in the these two hydroxyl group which are present here are responsible for the chelation with the biomordant.

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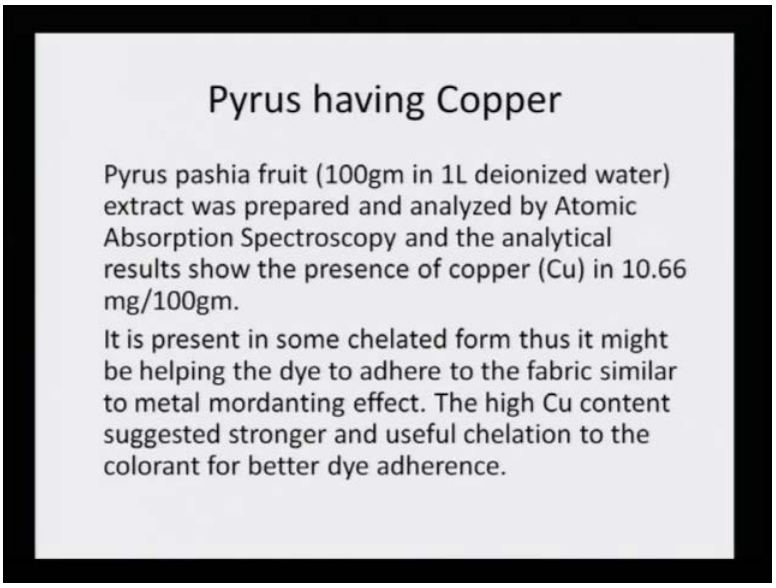

Pyrus having copper: These we have already establish that the pyrus pashia fruit when it was 100 grams in 1 liter deionized water was extracted, and it was prepared for the analysis of atomic absorption spectroscopy, and the analytical results showed the presence of copper to be in 10.66 milligram per 100 gram. So, you see this much is sufficient and where as in metal mordanting we are using in grams and the all only the required amount is being taken up by the fabric and the rest goes into the effluent. So that is what we were trying to minimize.

It is present in some chelated form thus it might be helping the dye to adhere to the fabric similar to the metal mordanting effect. The high copper content suggested stronger and useful chelation to the colorant for better dye adherence. This fact is undisputed that metal is required. Now, whether we use an external source of metal salt in larger quantity or we use a biomordant which has just for the optimal quantity of the metal is what is to be debated. And obviously, if this is doing the required functioning of creating good dye adherence, thus the purpose is solved. So, there is no need to excessively pump too much of chemical into the dye bath when it is not required. So that is what we were trying to emphasis and identification of this plant pyrus pashia and it is having copper contain was very well establish in our laboratory.

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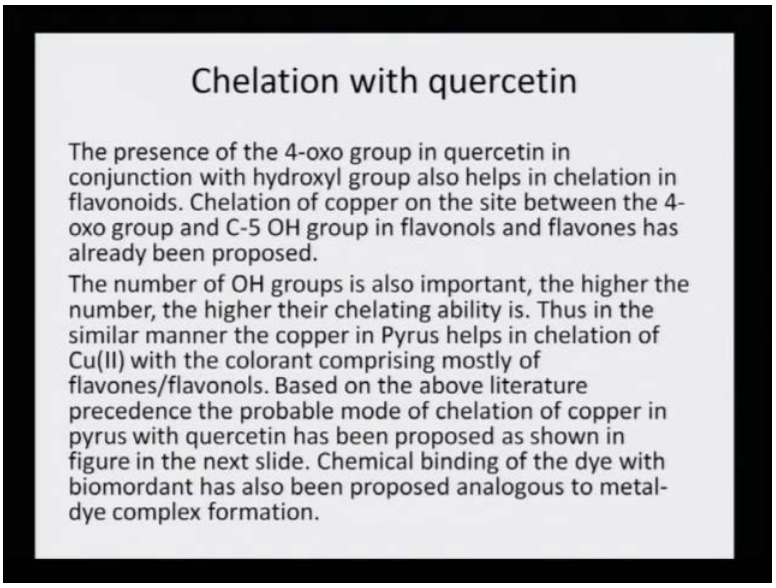

Chelation with quercetin: Now, considering the fact that the carotinoids are only imparting the color, but the actual chelation is done by the co-pigment that is the flavonoid. And quercetin is 1 flavonoid which was in abundance in that particular aqua in that delonix regia extract. The presence of 4-oxo group in quercetin in conjunction with hydroxyl group also helps in chelation in flavonoids. The chelation of copper on the side between the 4-oxo and the 5-hydroxyl group in flavonols and flavones have already been proposed; the number of OH groups is also important, the higher the number the higher their chelation ability is. Thus in the similar manner, the copper in pyrus helps in chelation of copper-2 with the colorant comprising mostly of flavones and flavonols.

Based on the above literature precedence the probable mode of chelation of copper in pyrus with quercetin has been proposed as shown in the figure in the next slide. Chemical binding of the dye with biomordant has also been proposed analogous to the metal dye complex formation. So, what we have try to draw an analogy is that, the way the metal salt actually chelates using the 2-aurtho hydroxyl groups of the flavonoids and the quercetin has ideally such a situation. The same philosophy or the same kind of chelation sites are proposed for metal for the biomordant as well.

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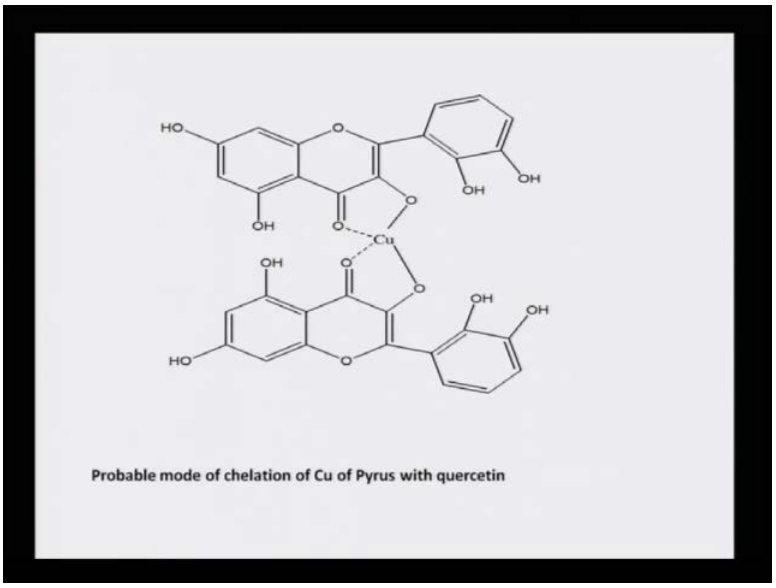

Now, you here you will see there are two quercetin molecules where the copper is nicely sitting in between, there are two coordinate bonds and two covalent bonds with the oxygen. So, it is actually participating with the carbonyl and 1-hydroxyl of the two groups, where copper can form two coordinate bonds and two covalent bonds. And these free hydroxyl groups actually can have hydrogen bonding to keep the molecules in tag, and therefore, the molecule can be nicely made to adhere on the fabric.

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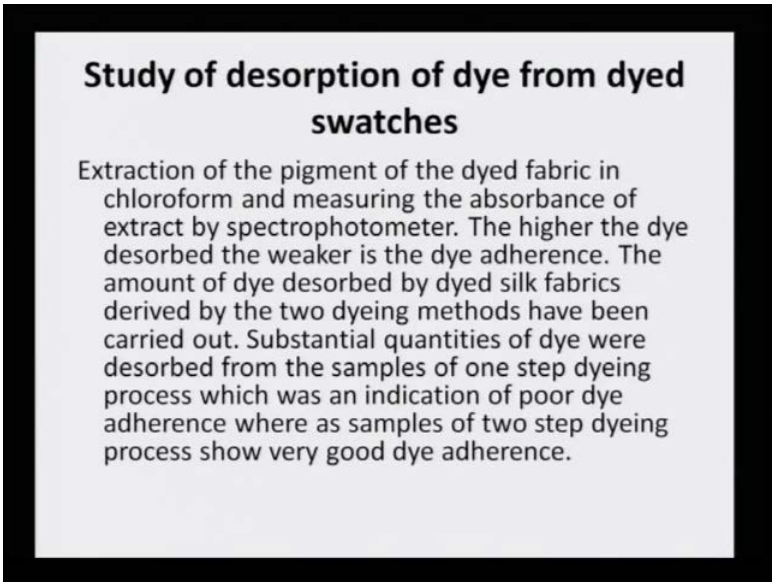

Study of the desorption of the dye from the dyed swatches: It was technique which we thought would be give us an inside about the dye ability or the dye up take. So, extraction of the pigment of the dyed fabric in chloroform and measuring the absorbance of the extract by spectrophotometer; the higher the dye desorbed the weaker is the dye adherence; the amount of dye desorbed by dyed silk fabric derived by the two dyeing methods have been carried out. Substantial quantities of dye were desorbed from the samples of the one step dyeing process, which was an indication the of poor dye adherence where as samples of the two step dyeing process show very good dye adherence.

Now, you see it also gives a deep inside into the dye adherence, although superficially the C I CIE lab values could be give us some kind of a desorption. But when it comes to actually measuring, the dye that can be desorbed from the dyed fabric, and then trying to take a look, whether the one step process that means pumping in everything, that is the biomordant and the dye in the same dye bath or doing it step wise that is first treating the fabric with the biomordant, and then subsequently treating it with the dye extract. These two processes when they are separated we call it two step processes, when everything is put together or the simultaneous mordanting method then it is called one step process. It showed that substantial quantity of dye was desorb.

Now, dye dissolving faster or more is an indication that dye has not adhered properly. So, we were trying to make a correlation between dye adherences and desorption and that gave an idea; that the two step process is far better and for delonix regia than when we are using biomordant or enzyme as compare to the one step process. But for industry and for industrial, you know, betterment - process betterment, technology up gradation one can suggest the one step process also.

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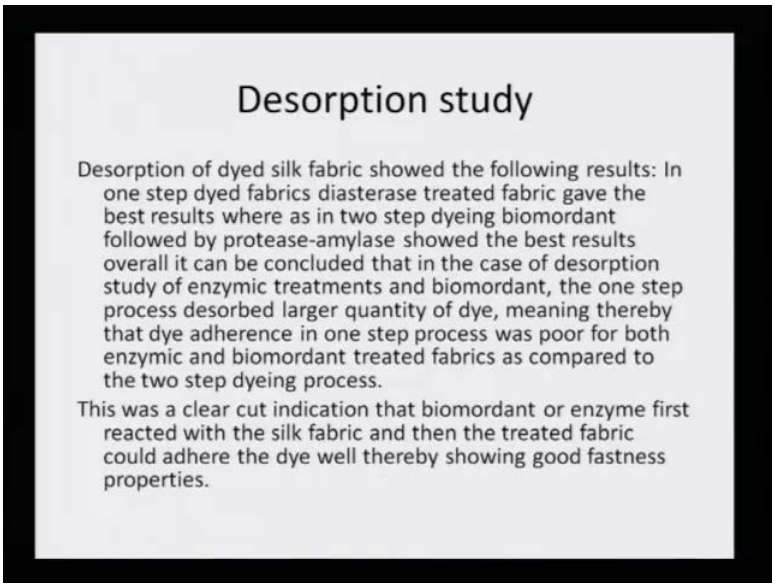

Desorption of dyed silk fabric showed the following result: In one step dyed fabrics, disasters treated fabric gave the best result, where as in two step dyeing biomordant followed by protease amylase showed the best result. Overall it can be concluded that in the case of desorption study of enzymatic treatments and biomordant, the one step process desorbed larger quantities of dye meaning thereby the dye adherence. In one step process was poor for both enzymic and biomordant treated fabric as compared to two step dyeing process. So, two things came into notice; one thing that which is the best enzyme suited. So, for one step process it is disasters, in the two step process it is the biomordant followed by protease and amylase. So, this is the first conclusion.

The second conclusion was that the dye desorbed was much more as quantity in quantity wise in the one step process, whereas in the two step process the dye desorption was less. So therefore, it was a clear cut indication that biomordant and enzyme first react with the silk fabric, and then if they are treated would adhere the dye well thereby showing good fastness property. So, it was a clear cut indication that step wise that mordanting either with biomordant or enzyme first, and then dyeing with the extract will definitely give good dye adherence and will be reflected in the fastness property. Because for every dye there is one ultimate goal and that is that the dye should have good fastness property. Otherwise, if it does not have good life fastness, wash fastness, rubbing fastness, it will not be taken up by the industry and it will not be categories as a good dye for the industrial process.

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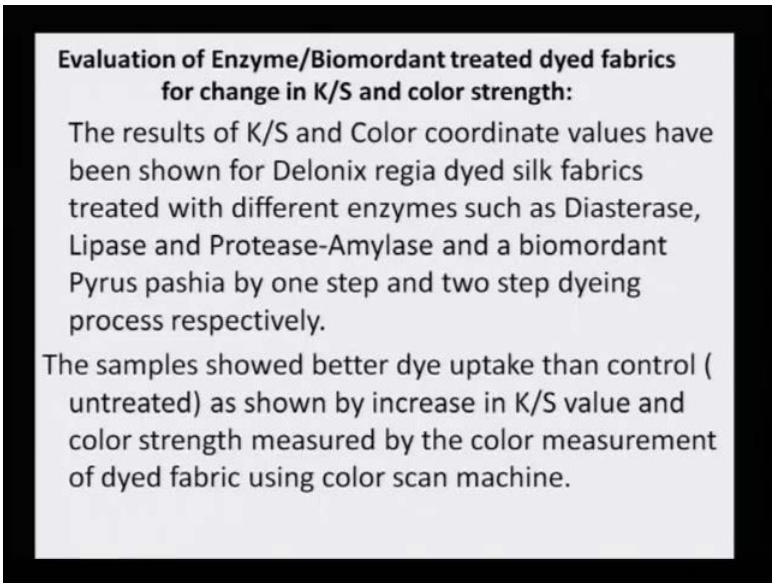

Evaluation of enzyme or biomordant treated dyed fabric in change for change in K by S and color strength: The results of K by S and color coordinate values have been shown for delonix regia dyed silk fabric treated with different enzymes such as disasters, lipase and protease-amylase combination along with biomordant such as pyrus pashia by one step and two step process respectively. So, for all of them for all the three enzymes that is lipase, disasters and protease-amylase combination as well as for biomordant both these processes were one step and two step dyeing were carried out. So, now I suppose it is very clear that what is meant by one step dyeing and what is meant by two step dyeing. The sample showed better dye uptake than control that is untreated as shown in by increase in K by S value and color strength measured by the color measurement of the dyeing fabric using color scan machine. So, it was very clear from the dyed fabric that there was very good dye up take.

Suppose if a control was not taken, how would we compare, whether the dye up take was better or worst. So, a control is always done, a control is a fabric which does not have any pretreatment or any mordanting, it is just a scward fabric which is dipped into the dye solution. So, then one can see whether there is any role of the enzyme which is being played or the biomordant and one can compare the CIE lab values and the K K by S values. And this was done by the help of color scan machine which we have already learned in great detail.

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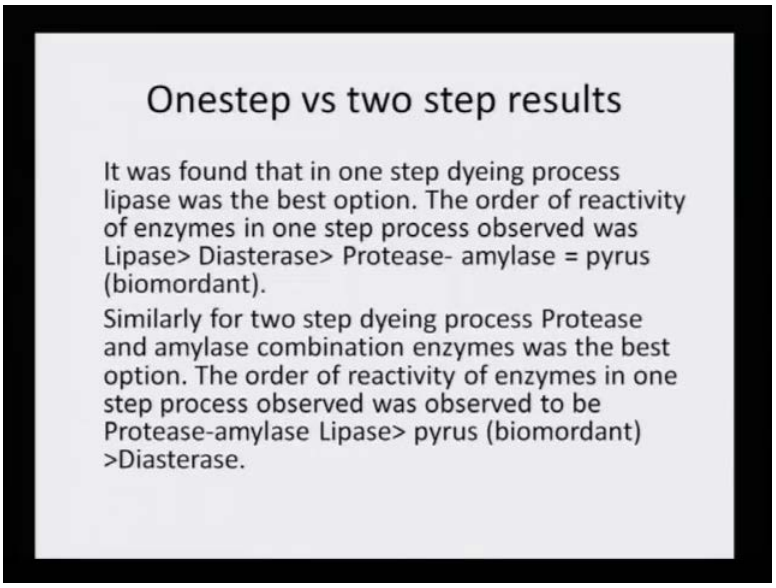

The one step process verses two step results: You see it is important to understand the chemistry and in order to minimize the step can we have a sacrifice on the dye adherence or should we have a sacrifice on the dye adherence, no. Why, because the ultimate goal for any dyeing is good dye adherence, good fastness property and that should not be sacrificed. So, it was found that in one step dyeing process; lipase was the best option. The order of reactivity of enzyme in one step process observed was that lipase is greater than dye disasters which is better than protease-amylase combination and protease-amylase combination was almost equal to the results obtain by the pyrus partia biomordant.

When similar study was done with two step dyeing process; protease-amylase combination enzyme was found to be best option, the order of reactivity of enzyme in the two step process was found to be protease amylase was better than lipase which was better than pyrus and the least was disasters. So, you see that reactivity completely differs, and reactivity differs, because the chemistry of the linking is differing. I showed you how christine played a role in the copper biomordanting and the copper was coming from the pyrus perschia which was in 10.66 milligram per 100 gram of the material.

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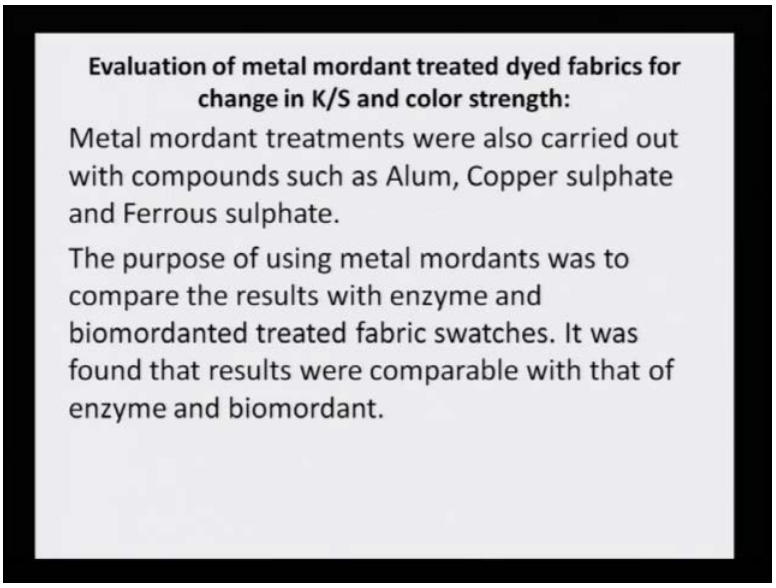

When we make an evaluation of the metal mordant treated with dyed fabric. I mean we also carried out it simultaneously in order to check, compare, just the way we compare one step, two step, different enzymes, we also tried to take a look at different metal mordents and the metal mordents that were used were alum, copper sulphate and ferrous sulphate. The purpose of using metal mordant was to compare the results with enzyme and biomordanted treated fabric swatches. It was found that results were comparable with that of enzyme and biomordant. So, you see their whole purpose of this exercise by using alum, copper sulphate and ferrous, we tried to show, that yes, for dilonix ragia one can use metal mordant, but is it really necessary to use the biomordant? Is it really necessary to use the metal mordant? No, biomordant can do the needful. So, therefore, the metal mordanting was found to be as comparable in terms of its lab values, in terms of its K by S values, as compared to the values obtained by the enzymic treatment or the biomordant treatment.

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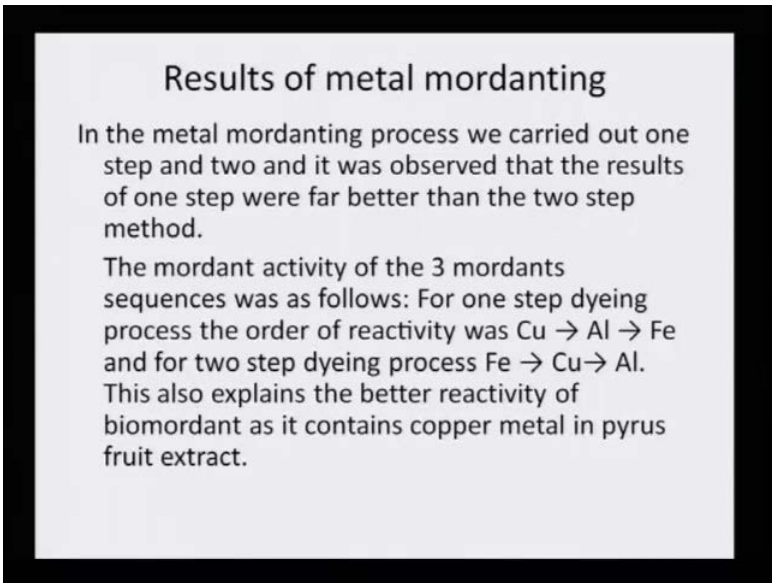

Even than take a look at the results of the metal mordantor; what was the kind of sequence that one comes across? In metal mordanting process, we carried out one step and two step, it was observed that the result of the one step were far better than two step. The mordant activity of the 3 mordants sequence was as follow; for one step dyeing process the order of reactivity was the copper better than alum which was better than iron and for two step dyeing process iron was better than copper which was better than alum. This also explains the better reactivity of the biomordant as it contains copper metal in pyrus fruit extract.

So, it also explain to us many hidden facts, because by making a comparative data study of the metal salt mordanting and the pyrus biomordanting, we could make an analogous study and find the analogy how it may be reacting; why copper was found to be better, why in in both the cases, because copper is common to them and why pyrus showed such good results was explained by the fact that copper salt mordanting shows very good results. So, so on and so forth, so one comes to a conclusion unless and until we make a comparative analysis, unless and until we have you know a goal to compare with, we will not know whether it is better, equal or worse. So, in order to understand any science very logically, it is important to make comparative data.

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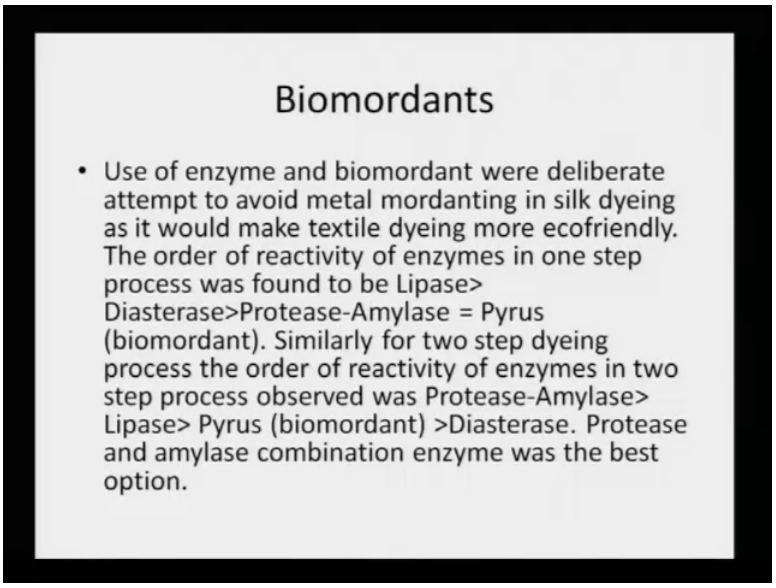

Biomordants use of enzyme and biomordant were deliberate attempt to avoid metal mordanting in silk dyeing as it would make the textile dyeing more eco friendly. The order of reactivity of enzyme in one step process was found to be lipase was better than diasterase which was better than protease-amylase and this was almost equivalent to pyrus perschia the biomordant. Similarly, for two step dyeing process, the order of reactivity of dyeing and enzymes in the two step process was observed to be protease-amylase was better than lipase which was better than pyrus perschia biomordant and which was far better than diasterase.

So, thus we can conclude that protease-amylase combination of enzyme was best option in the two step process. Now, why we were using enzyme and biomordant is of very important issue, because we have been talking about metal mordanting being extremely harmful and non eco friendly, and it was this particular fact that we wanted to come back, and that is why we wanted to replace the metal mordant by using either enzyme or bio mordant, because if both show similar color affectivity than the purpose would be solved and rightly so, the color strength affectivity was evaluating.

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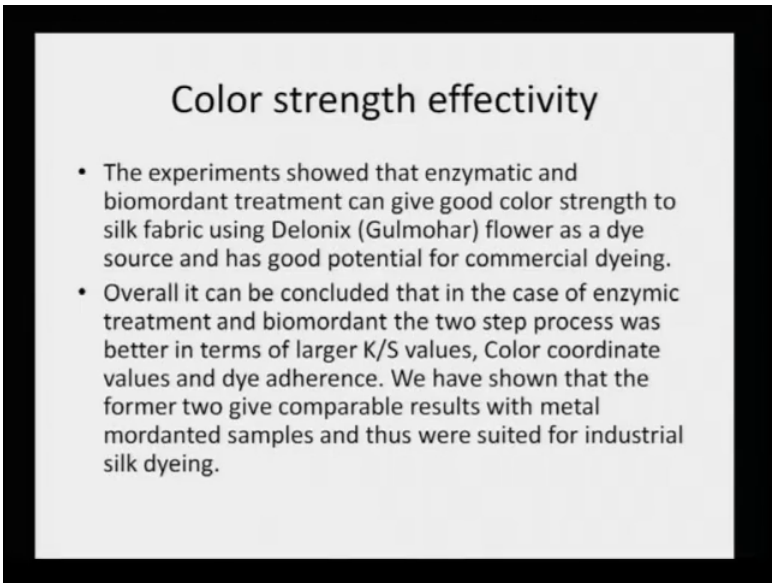

The experiment showed that enzymatic and biomordant treatment can give very good color strength to the silk fabric using delonix regia that is gulmohar flowers as a dye source, and has good potential for commercial dyeing. And thus it was establish that the color strength affectivity did not better or did not become less as compare to the metal mordanted silk dyeing fabric. So therefore, we can say that it was a (( )) with the metal mordanting procedure. Overall it can be concluded that in the case of enzymatic treatment and biomordant the two step process was better in terms of larger K by S values, color coordinate values and dye adherence. We have shown that the former two give comparable results with metal mordanted samples and thus were suited for industrial silk dyeing. So, this we have shown that delonix ragia the abundantly available flower can be used for silk dyeing in conjunction with enzyme or we can even use biomordant like pyrus perschia and it can give the same result as what metal mordantanted fabric would have given.

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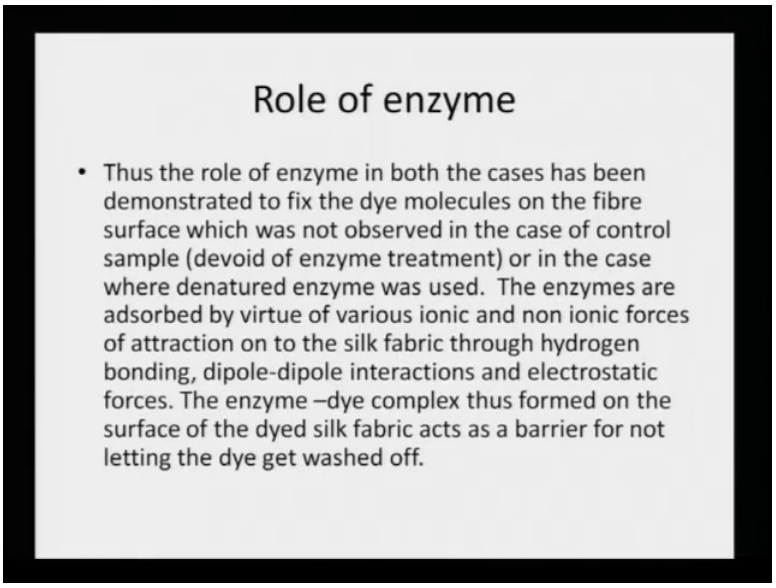

Thus there is a very important role of this enzyme. Thus the role of enzyme in both the cases have been demonstrated to fix the dye molecules on the fabric surface which was not observed in the case of control sample, which is devoid of enzyme treatment or in the case of where it is denatured enzyme was used. The enzymes are absorbed by virtue of various ionic and non ionic forces of attraction on to the silk fabric through hydrogen bonding, dipole-dipole interaction and electrostatic forces. The enzyme dye complex thus formed on the surface of the dyed silk fabric acts as a barrier for not letting the dye get washed off. So that is how it entangles the dye molecule and cages it in the dye enzyme and fabric complex.