Course on Industrial Biotechnology
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Module 08
Lecture No 37
Lactic Acid Production (Contd.)

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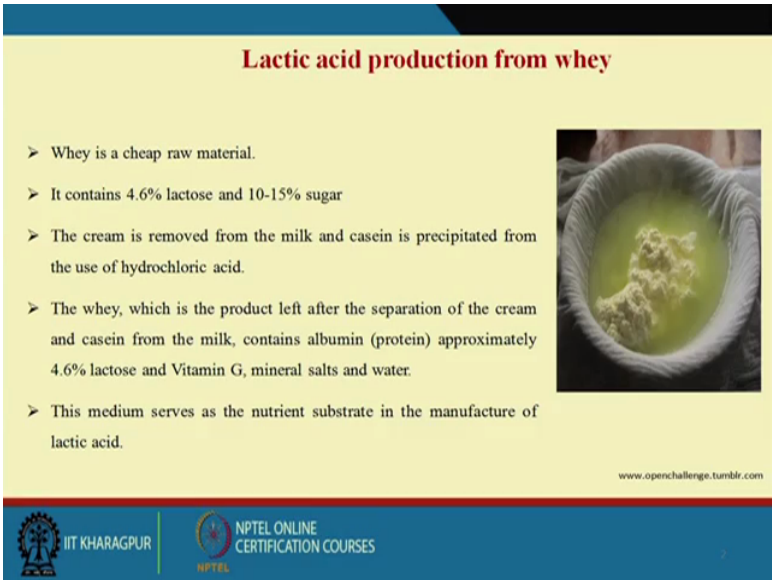

Welcome back to my goods industrial biotechnology. The so in the last lecture I was concentrating on citric acid lactic acid production, so I talk already about basic aspects of lactic acid. Now I shall show you how industry that produces lactic acid. Now so if you look at the raw material that is used suitable most suitable for lactic acid production is of whey, because whey is the by-product of the dairy industry. Because you know that when you we produce this that called as cheese you know that whey comes out.

That this contains very less amount of protein and fat but the liquid mostly contains the lactose and which is a very good raw material for the lactic acid formation. The whey why we use the whey for this fermentation process? The reason is that it is by-product of the dairy industry and second is that it is a very cheap raw material. It contains about 4.6 percent to about 7 percent of lactose then 10 to 15 percent of sugar, because the cream is removed from the milk and casein is precipitate from by from the use of hydrochloric acid or and also we sometimes we use in the house we use the calcium lactate to precipitate down the protein.

When the whey which the product left after the separation of the cream and the casein from the milk contains albumin approximately 4.6 percent of lactose, vitamin G, mineral salt and

the water, because you not that this solid material mostly it contains the milk, protein and fat and all the minerals and the vitamin G and lactose goes in the soluble forms and this medium serves as the nutrient substrate for the manufacture of lactic acid.

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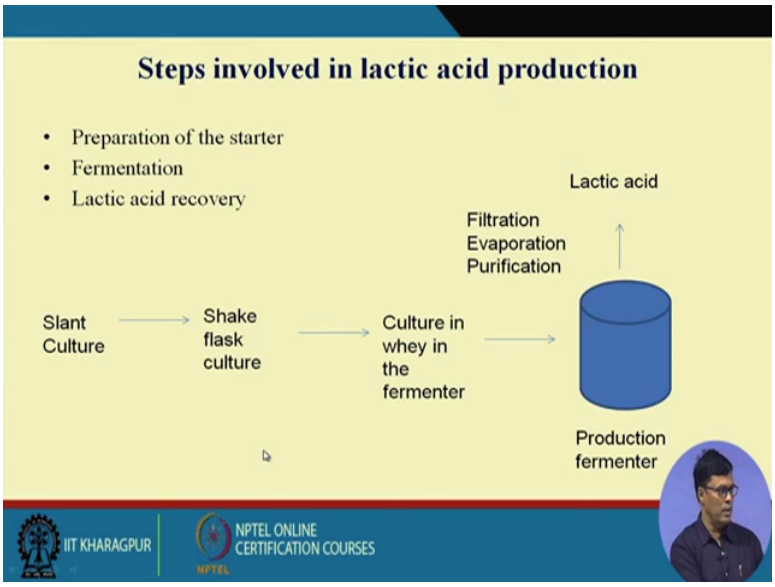

Now let me show you the steps involve in the lactic acid production, very we prepared the culture in slant then from the slant we inoculate in the media in the shake flask where in the liquid media and this is the this is the agars slant to grow the culture. Then in liquid media in the shake flask we grow this culture after that it go grow in a control fermenter and we get the cell mass and this cell mass we put it in the production fermenter which produces lactic acid but it passed through the different downstream processing like filtration, evaporation and the purification.

So this is the 3 steps involve major steps, one is preparation of starter, starter means the organism that you prepare for this production and there is fermentation that what is carried out in the production fermenter and the lactic acid. So if you look at the normal this that fermentation industry whatever we have it is almost similar to that. In the normal fermentation industry also I told you that every industry we have 2 reactors, bioreactors, one for the cell mass production and another for the product production. So cell mass production we call it inoculum vessel, and production fermenter recall it PF the production fermenter.

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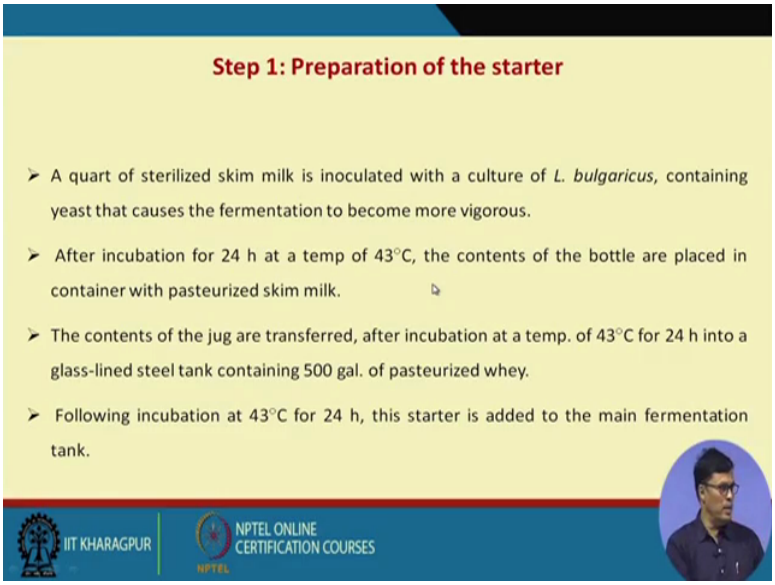

So now let us see that what is the first step that we have in the preparation of the starter, a course of sterilized skim milk is inoculated with the culture of Lactobacillus-delbrueckii containing yeast that causes the fermentation to become more vigorous. After incubation for 24 hours because you know that one thing here I want to point out that doubling time of bacteria is much less as compared to yeast cell. The even if you yeast if you add it give some kind of nutrient to the fermentation broth, so it will not affect the growth of the organism to a great extent.

I told you, you can remember that in the citric acid fermentation process we use the cane molasses as the raw material for the production of citric acid and main culprit main that contaminants that we have in the citric acid fermentation process is the yeast. Why? Because cane molasses is the best raw material for the growth of yeast cells and if you look at the doubling time of the yeast cell is much less as compared to that of fungi. So if yeast enters into the system that will not allow the fungi to grow.

So here we do not have any problems if we use this because your bacteria you look at the yeast if you bacteria doubling time is much less lactobacillus-delbrukeii and contains yeast that causes them to become vigorous. Then after incubation for 24 hours at a temperature 43 degree centigrade the contents of the bottle is placed in a container with pasteurised skim milk.

This content of the jugs are transferred after incubation 43 degree at 24 hours into a glass lined steel tank containing 500 gallon of pasteurized whey. Because you know that one thing

I want to point out that lactic acid is has a corrosive property, so special type of construction material for construction of the fermentation that the tank is to be maintained. Following incubation at 43 degree centigrade for 24 hours the starter is added to the main fermentation tank. So we first prepare the culture what we called inoculum is the inoculum and then we this inoculum we use to the production fermenter.

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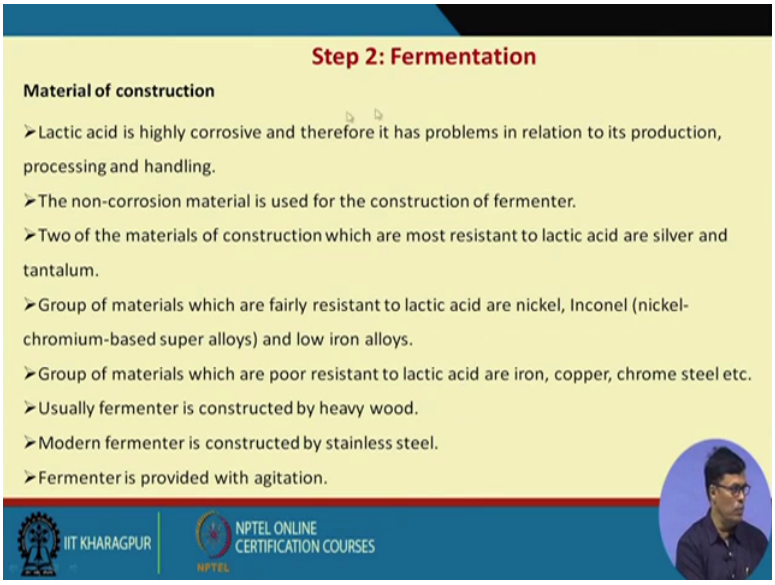

The step 2 is the material of construction, the lactic acid is highly corrosive, as I pointed out, therefore it has problems in relation to its production, processing and handling. The non-corrosive material is usually used for the construction of fermenters. The 2 of the material of construction which has mostly resistant to lactic acid are silver and tantalum, this is the 2 metals with they are quite resistant to the lactic acid, so this is usually recommended.

The group of material which are fairly resistant to lactic acid are nickel, Inconel, Inconel is nothing but nickel-chromium-based super alloy and low iron alloy. Then group materials which are poor resistant to lactic acid they are iron, copper, chrome steel etc. I mention that chromium plays important to increase the corrosive property of stainless steel but here it is not that effective. And usually the fermenter is constructed with heavy wood, because that you know I in acetic acid fermentation process I was talking about the beach wood shavings which is a soft wood with lot of pores inside, but here we are talking about the hardwood, the hardwood also like oak wood we sometimes we can use for this as a vessel, where hardwood we can use as a vessel.

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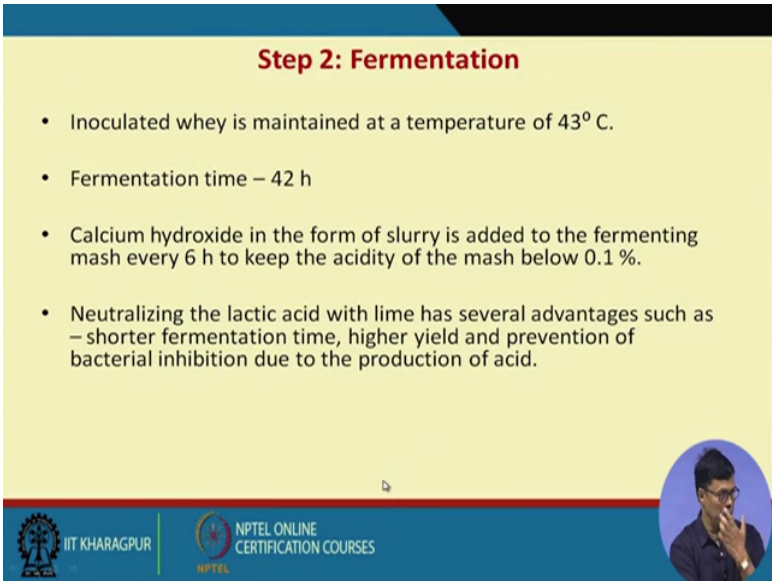

Modern fermenter is constructed with stainless steel. Fermenter is provided with the agitator, agitator is required so that you know cell can remain suspension. The inoculated whey is maintained at 43 degree centigrade. Fermentation time is 42 hours. Calcium hydroxide in the form of slurry is added to the in the fermenting mash, every 6 hours to keep the acidity much below 1 percent 0.1 percent, because if we use the high allow the acidic acid to be build up then it will hinder the growth of organism so this is to be maintained. So we add the I already mention in case of citric acid fermentation process, how calcium hydroxide is produced.

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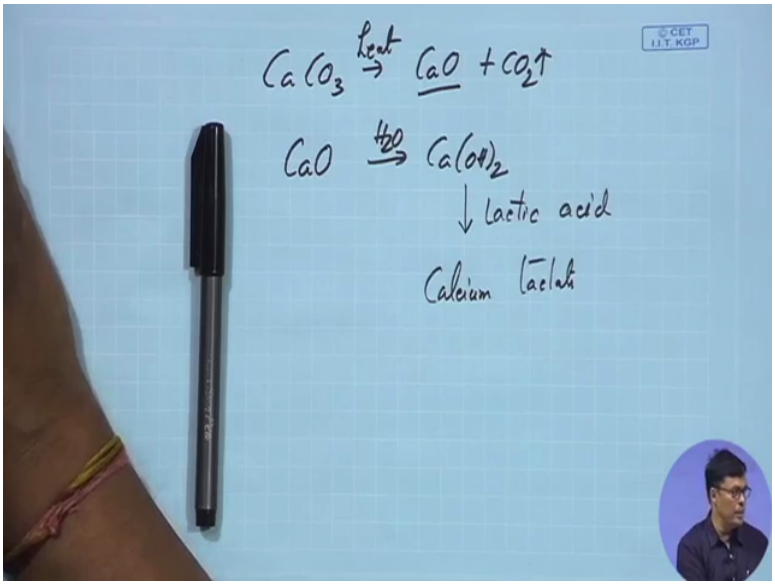

I told you that calcium carbonate which is in the present in the rock this when you heat it produces calcium oxide and carbon dioxide, so carbon dioxide we goes out. Then this

calcium hydroxide calcium oxide in presence of water you may slurry you will get calcium hydroxide. Then this calcium hydroxide you have to add, that you have to add and when you add the calcium hydroxide when in presence of lactic acid it produce calcium lactate. So neutralising the acetic lactic acid with lime have several advantages such as the shorter fermentation time, higher yield, prevention of bacterial inhibition due to the production of acids, so these are the reasons why we use the lime.

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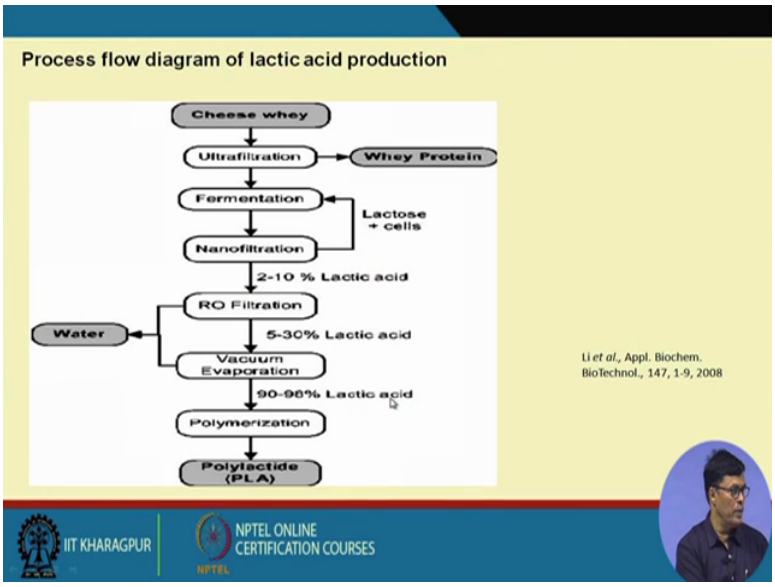

Now this is the flow diagram of the process, this flow diagram is like this that we take cheese whey we can take whey from other source also. Then to the ultrafiltration, whey proteins we take it out maybe you heat it then protein will precipitated out, then you can take out the whey protein through the ultrafiltration. Then in the fermenter we inoculate the Lactobacillus lactic acid bacteria here. Then after production we pass through the Nano-filter we get 2 to 10 percent of lactic acid.

Then RO filtration, the reverse osmosis that we carried out to get the higher concentration of lactic acid, then vacuum evaporation we get 90 to 98 percent lactic acid. And this lactic acid we can go for the polymerisation reaction to get the poly-lactic acid. So we what you called the poly-lactide, because the I told you that poly-lactic acid has the tremendous potential in the pharmaceutical industries.

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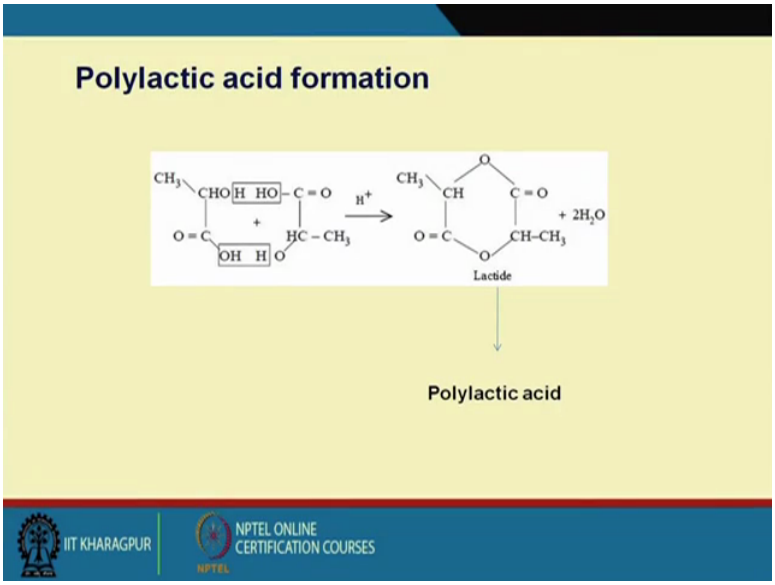

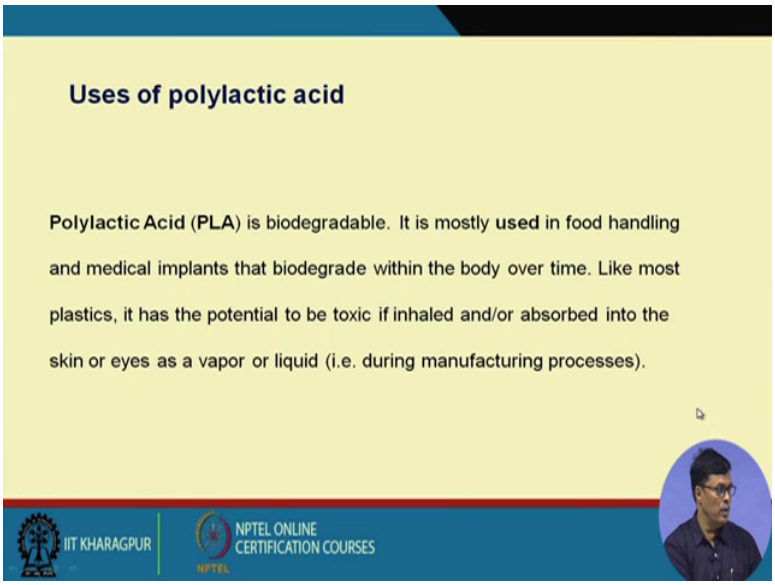

Now I want to show you the reactions how poly-lactic formation is there, this is the lactic acid if you look at. 2 lactic acid when formed it is form the lactide and this lactide actually help for the formation of the poly-lactic acid. Now if you look at the poly-lactic acid is biodegradable, why we use the polylactic acid? The reason is that it is biodegradable and it is mostly used in food handling and medicinal medical implants that biodegradable within the body over time. That this is very important medical implants that biodegrade within the body over time.

So this is why it is used for the medicinal purpose or for particular in case of surgical purpose is largely used. Like most of the plastics it has the potential to be toxic if inhaled or absorbed and absorbed into the skin, eyes as a vapour or liquid. During manufacturing process

obviously if this polymer equally affected to the human health it is no good, but when it is solidified it does not have bad effect on the human health it is usually biodegradable.

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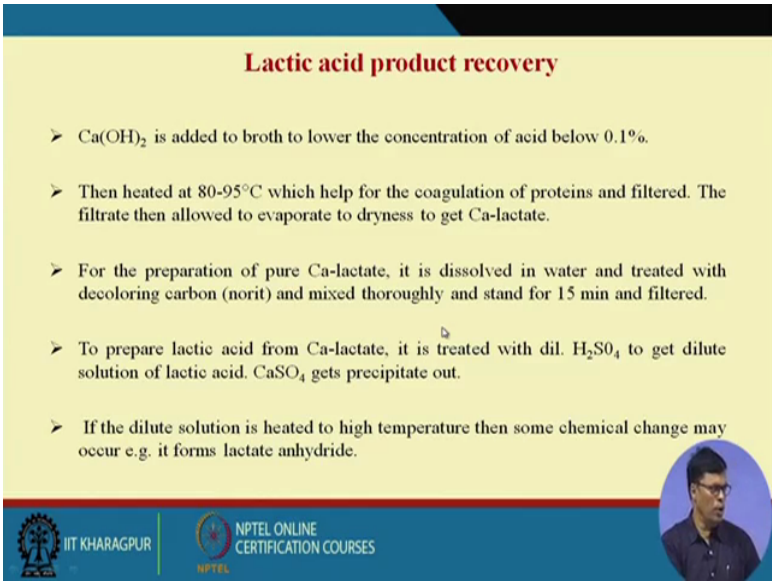

So then question comes how we recovered the lactic acid? Calcium hydroxide is added to the broth to lower the acidity below 0.1 percent and then heated to 80 to 95 degrees centigrade with the help of coagulation protein, coagulation of protein and filtered. The filtrate than allowed to evaporate to dryness to get the calcium lactate and so initially we our intention is to produce the calcium lactate and this lactate pure calcium lactate it is dissolved in water and treated with the de-colouring carbon and mix thoroughly and stand for 15 minutes and filtered, because it may give some kind of colour to the product that is why we use the activated carbon just to activate carbon.

I mentioned before also it has some kind of bleaching property, so if we use the colour can be removed you have and then we prepared the lactic acid, calcium lactate we treated with H2SO4 to get the dilute dilution solution of lactic acid and calcium sulphate get precipitated out we can separated out like you know. In case of citric acid also we hydrolyse the calcium citrate by H2SO4 we separate the calcium sulphate in the form of gypsum filter.

In the gypsum filter we separate this calcium sulphate to H20 that is called gypsum which is a very good raw material for the cement industry. And then the dilution the dilute solution is heated to high temperature then some chemical changes may occur and it from the calcium anhydride. So if we use the very high temperature it will again and again you will get the different product that like to which is not desirable.

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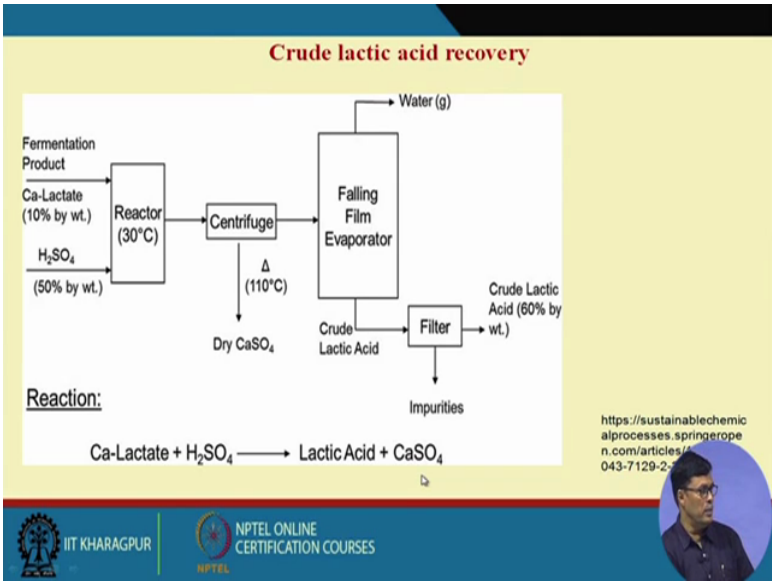

Now let us see that how actually lactic acid is produced, this is the fermentation product after that you will get this and you get the calcium lactate about 10 percent weight by volume and when it goes to the reactor where you push this H2SO4 and heat it for 30 degree centigrade. Now and then we can in previous case I mentioned we heat it to 80 to 90 degree centigrade and then we centrifuged it at the high-temperature we get calcium sulphate and then we evaporate it so that concentration of citric lactic acid increases. We filter it, impurities will go out lactic acid we get 60 percent by weight this is available in the liquid form. This lactic acid the reaction is like this calcium lactate plus H2SO4 gives lactic acid and calcium sulphate.

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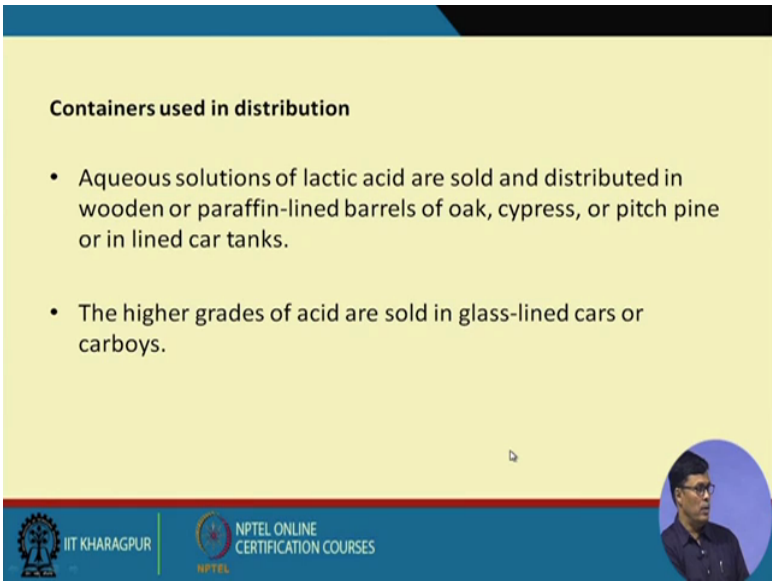

Now containers used in distribution, now question comes since lactic acid has highly corrosive properties, so distribution in the containers also is very important. So aqueous solution of lactic acid is sold and distributed in wooden or paraffin lined barrels of oak, cypress and peach pine or in lined car tanks. The higher grade acid are solid in glass-lined in cars or carboys. So this is some glass lined materials also can be used for that for transferring this material then it will not affect the material of construction to a great extent. Otherwise we have to do otherwise the material will be affected with the help of lactic acid this is very important.

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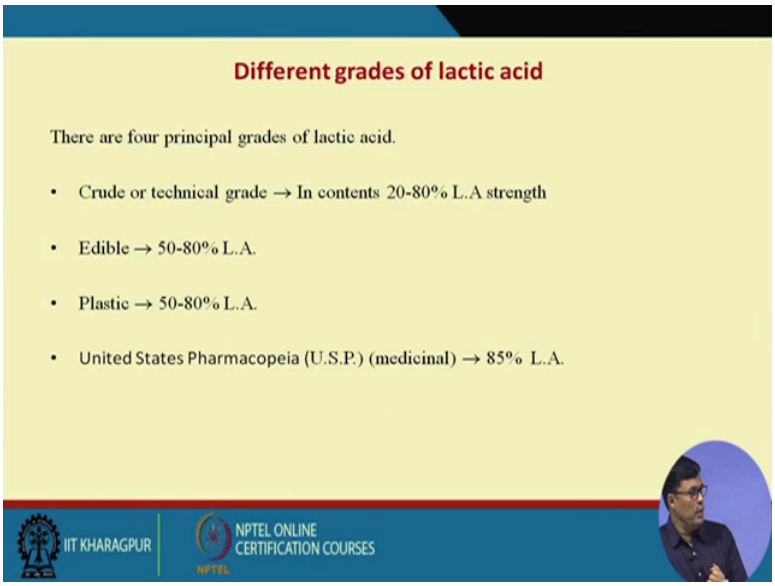

Now let me tell you that, what are the different types of lactic acid usually available in the market? Because that is also very important. Then lactic acid can be available in the different forms, one is the crude or technical grade lactic acid which is 20 to 80 percent, this is largely used there is a lactic acid strength is there. Edible, that is 50 to 80 percent that we have lactic acid. Plastic we require 50 to 80 percent lactic acid. Now United States Pharmacopeia that is UPS medicinal grade that is 85 percent. So different percentage of lactic acid that is used for different purposes that we have.

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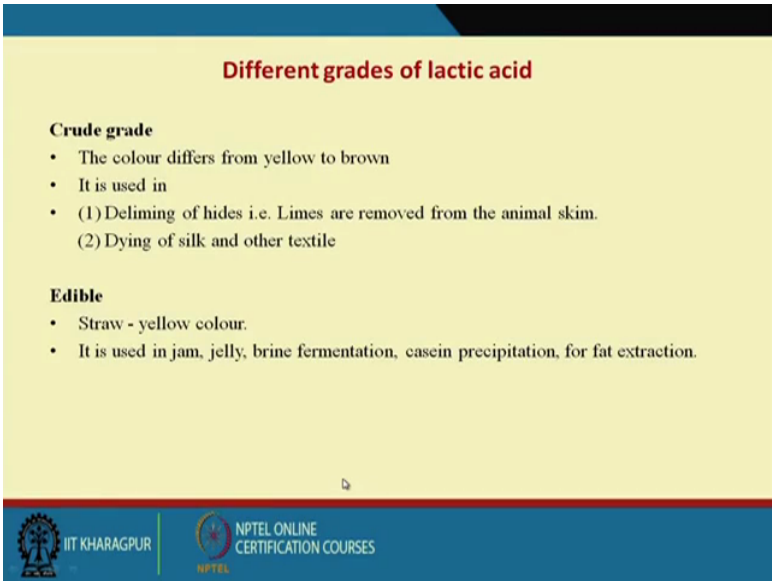

Now if you look at the what are the different grades that we have that uses for different purposes? As for example crude grade, this is coloured varies from differ from yellow to brown and it is use in deliming hides. Limes are removed that animal skin which is particularly I can tell in the tannery industry is largely used for that. And then dying the silk and other textile that is a good grade of lactic acid is largely used.

Then edible usually this is the yellow colour and that is use in the jam, jelly and brine fermentation, casein precipitation and for fat extraction. So that different purpose it is used for edible when you use I can give the example of calcium lactate I was telling the largely we use in the house for precipitating milk, protein and fat, because we use for the civil purpose.

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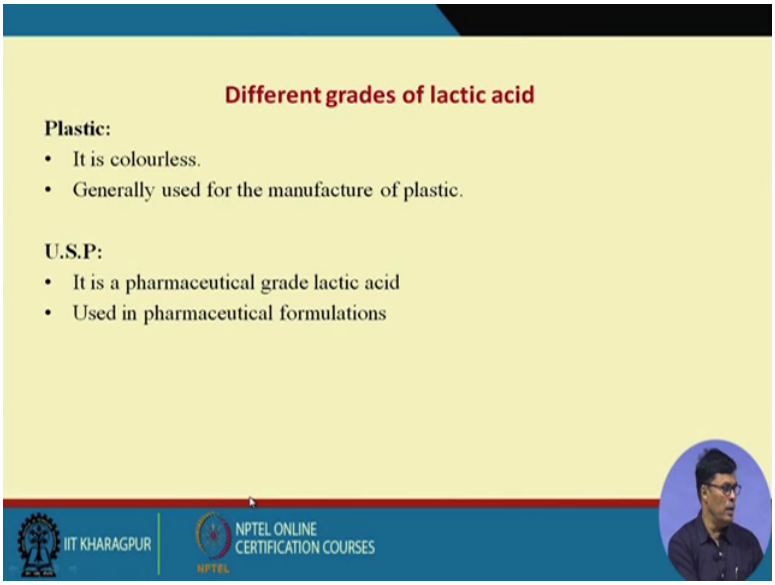

Then we have other different grades, we have plastic grades it is colourless and generally used for the manufacture of plastic. Because I told you that lactic acid has huge market potentiality in the pharmaceutical industry and this plastic is has lot of use in the in this industry. And UPS is the pharmaceutical grade of lactic acid and use in the pharmaceutical formulation purpose.

So this is all about the lactic acid fermentation process. Now here I want to summarise like this that lactic acid is produced by using 2 different type of bacteria in the industry largely used one is Lactobacillus-delbrueckii and other is Lactobacillus-bulgaricus. Then here temperature is little bit high usually 43 degree centigrade or higher temperature that is used for this fermentation process. And raw materials contains we find that whey which is the by-product of the dairy industry it is very find suitable for this fermentation process. And it has been observed amino acid present in the yeast extract and malt extract they find very suitable for the growth of the yeast cell growth of the lactic acid bacteria.

Then when you come to the fermentation process that after during the fermentation process we maintain the acidity by using calcium hydroxide or calcium carbonate and since we are adding the calcium hydroxide, the calcium hydroxide will react with lactic acid and form calcium lactate. And then after this requires about 42 to 46 hours and after fermentation your inoculum fermentation for the cell mass production is require 24 hours and then after the production fermenter the material we treated with we concentrated that we separated out the calcium lactate and then we hydrolysed in presence of H_2SO_4 we get the lactic acid and calcium sulphate.

Calcium sulphate precipitated out with the help of filtration. Other cases also we do some kind of filtration process just to separate out lot of suspended material in the fermentation broth including the bacterial suspension. So this is all about the lactic acid bacteria, thank you very much.