Downstream Processing
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Lecture - 1
Introduction

Welcome to the course on bioprocess the downstream processing. For the next forty lectures, I am going to talk about the various unit operations involved in downstream processing, how do you go about designing a downstream processing equipment, what are the advantages, disadvantages of various downstream processing equipments. Downstream processing becomes very relevant in the area of bio process as well as in chemical process technology.

Downstream processing has been there over a very long period of time, especially in the area of chemical engineering where chemical engineers used to practice recovery of products using filtration, distillation, separations involving extractions and so on. But in the area of bio process technology, as you are handling enzymes or proteins or bio molecules, the complications are slightly much more, and also you need to consider the stability of these bio molecules.

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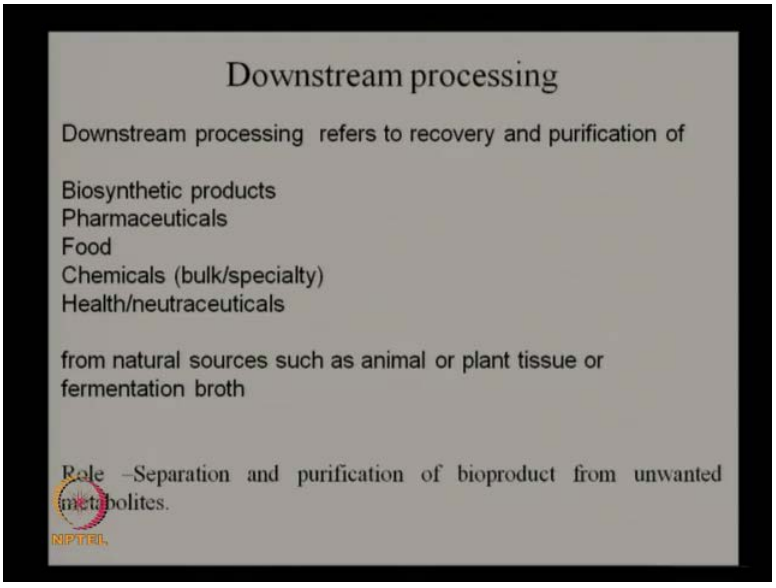

So, new techniques also came into being techniques like chromatography and techniques like membrane separations have been incorporated in the bio process technology also

actually. So, what does downstream processing means that it refers to recovery and purification of biosynthetic products, products coming out of a biological operation. It could be a pharmaceutical product, it could be related to food or neutralceuticals, it could be related to chemicals, it could be a bulk chemical or a specialty chemical, it could be products related to healthcare or medical biotechnology. So, you try to recover after you manufacture using animal or a plant tissue or a fermentation broth.

So, what is the role of downstream processing? It is meant to separation, it is meant for purification of a bio product from unwanted metabolites. So, once you carry out a fermentation or once you carry out a bio transformation, you are going to have the desired product as well as undesired waste products, side products and so many wasteful dead bio mass and so on. So, the idea is to recover and isolate the product of your interest from a large combination of unwanted products.

So, that is where the downstream processing comes. That becomes a big challenge because you would like to recover your products in a very economical way and try to recover it as much as possible. That means not lose anything in the waste, but try to recover as much as possible in to your final purification. That is the main challenge of a downstream processing actually.

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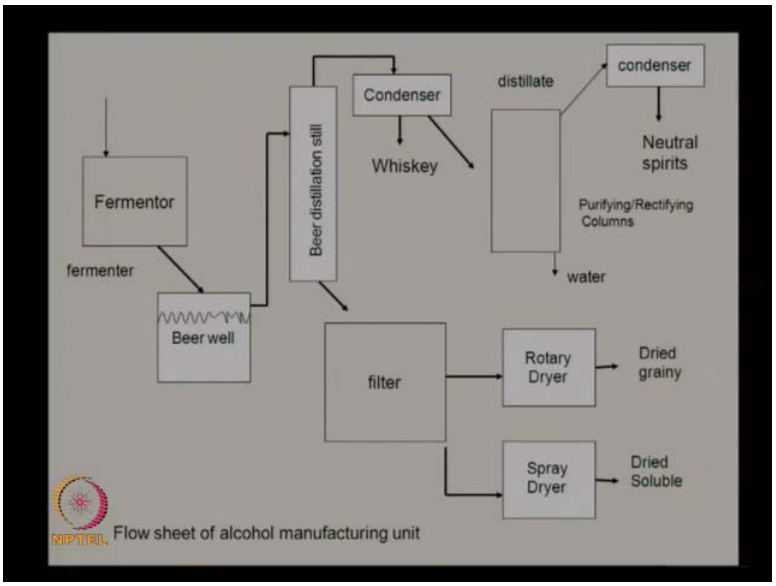

So, let us look at flow sheet, the alcohol fermentation. I mean it has been there for almost five thousand years, fermenting from sugars, fermenting from fruits, converting into

alcohol. Once they convert into alcohol, it contains a large amount of a liquid where your alcohol, the percentage of alcohol is very little. So, the idea is to recover as much of possible the alcohol from this large quantity of liquid and then concentrate it to high degree of purity. So, that is what is alcohol fermentation is all about.

So, if you look at the entire flow sheet of alcohol manufacturing, you have the fermentation taking place here. That is where the sugars are getting converted into the fermentor alcohol, but if you look at the right hand side in this flow sheet, the entire portion is the called the downstream processing. This is where you are trying to isolate your alcohol and then you are trying to purify your alcohol to a very large concentrator liquid form.

So, you can see, you have just one fermentor here, but you are going to have a very large number of unit operations on the right hand side, which help you to isolate your product as well as the purified product. Just look at these various unit operations that is called you know, you have a vessel to hold your fermentation broth. So, the alcohol concentration may be very little, 8 per cent, 9 per cent, that is all. But, ultimately you would like to get almost 90 per cent pure alcohol.

So, once you collect the fermentation broth, you may be doing the distillation because alcohol boils at lower temperature than rest of the mass. So, the distillation happens here and then the product that is coming out is condensed here. This will be mostly alcohol and of course you are going to have water. If there are any other side products, which are also going to be of lower boiling point is going to also condensed here. Now, the bottoms you filter and then you collect the solids, you dry the solids. Finally, you may get some dried grainy material; you may also get dried soluble material. So, this can be used as animal feed. It may be rich in protein. It may be very useful for animal feed.

So, once the alcohol is distilled out, which contains water and other low boilers, it is further distilled here. These are called purification and rectification columns. Finally, you get very pure alcohol on the extreme right hand side and you are going to have lot of water coming out as the bottom here. So, this particular operation of purification of alcohol from fermentation contains several distillation columns and then filters and driers.

So, the downstream in alcohol fermentation are made of filters, driers and the distillation

columns. It is quite a simple downstream operation if you consider alcohol fermentation, but if you go for other chemicals like bulk chemical or even specialty chemicals, even products related to drugs or pharmaceutical, you may have very stringent requirement for purity because a pharmaceutical product has to be extremely pure.

So, you may go into more purification steps. So, may go into steps like chromatography, different types of chromatography, you may go into separations like membranes and so on actually. This is because here it is a product, which can take, which can withstand large, high temperatures, you are resorting to distillation.

But, you are going to have products, which are like protein or which are enzymes, which might not be able to withstand the temperature. You may not be able to use distillation there. You may have to use some other downstream process. So, depending upon the type of product, which you are trying to recover, the nature of the downstream varies quite a lot actually.

So, a very simple fermentation of sugar, converting into alcohol requires several downstream. As I said mostly distillation columns, filters and driers and then we will be seeing more of these. We will also see how you go about designing these various unit operations and what are the principals involved in these unit operations. That is what we are going to look at in the course of time.

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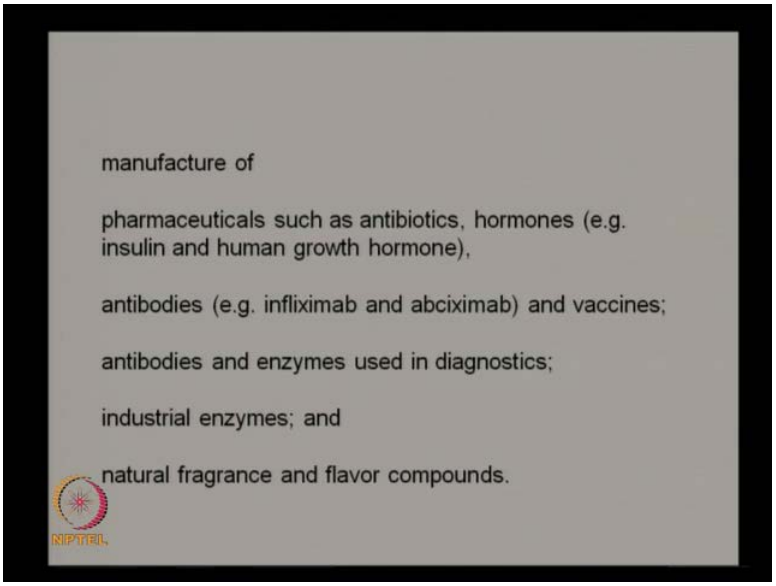

So, a downstream can help in manufacture of pharmaceutical products. All the antibiotics are made using fermentation or biotechnology routes. So, they resort to quite a lot of downstream steps. You can use it for making hormones; it could be human growth hormones or insulin. It is used in anti body manufacture, vaccine manufactures in enzymes, which are used in diagnostic kits, enzymes used in industrial applications. Even your soap for example, contains; enzyme can be used for natural fragrance, flavors, food products and so on. So, almost all biologically manufactured products require downstream steps. Hence, downstream becomes extremely important.

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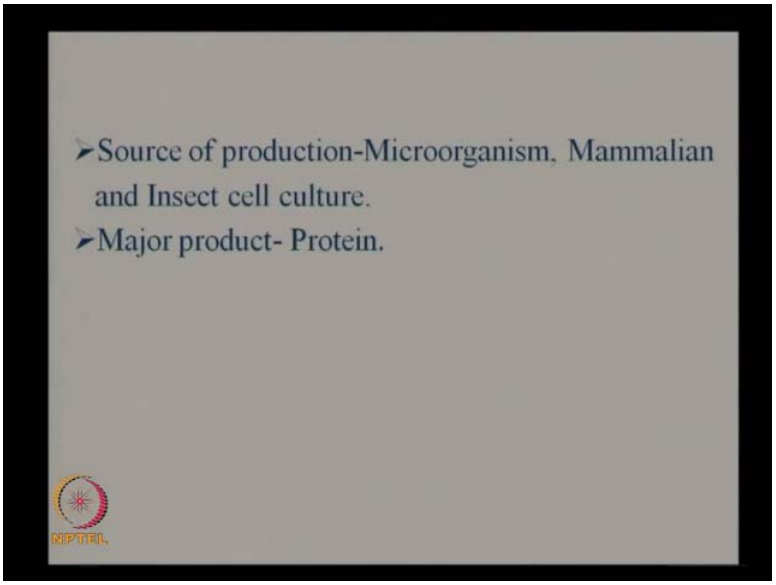

So, downstream can be used once you ferment a particular material using a microorganism or a mammalian or insect cell culture and major product is protein. But, then now days, even bulk chemicals are being manufactured using biological approaches because the techniques have become so cheap that they are becoming very competitive, when you compare it with chemical manufacturing itself.

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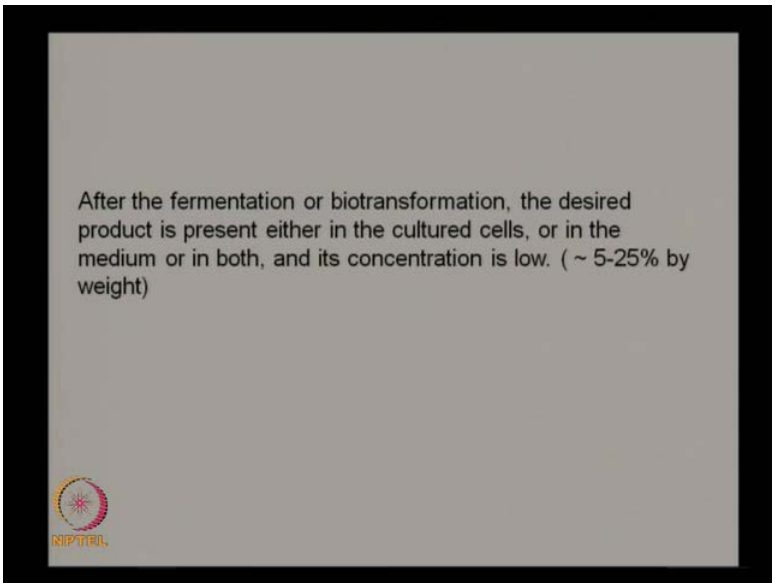

So, what happens after the fermentation or bio transformation? The desired product is either inside the cells that means microorganism or it is in the medium or in the broth. So, your product may be inside, your microorganism bacteria or fungus or it might be in the medium or the broth. Generally, the concentration is very low, 5 to 25 per cent. So, your idea is to recover, concentrate, purify to almost 100 percent. So, if it is a drug, it has to be extremely pure. The impurity levels have to be practically 0 per cent. If it is a bulk chemical, then the impurity can be even 5 per cent or 10 per cent. So, depending upon the type of application, the purity levels are decided upon actually.

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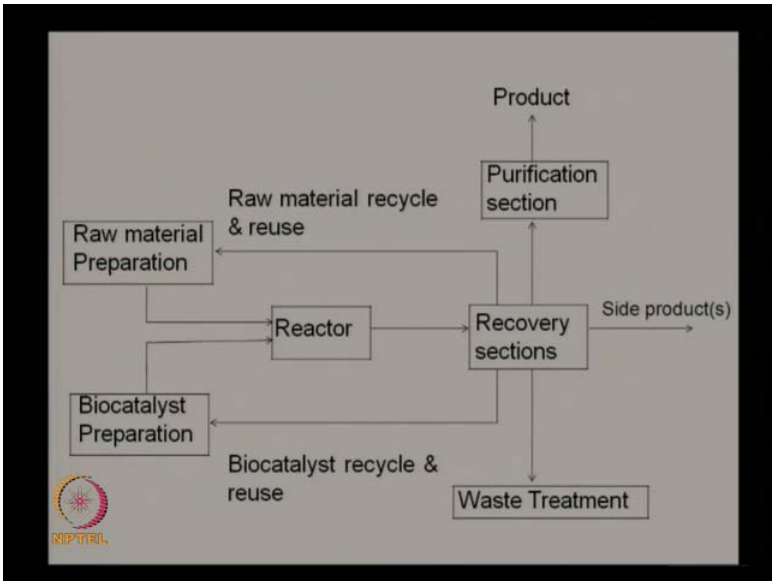

If you look at typically a bio process product manufacturing, this is called flow sheet of a manufacturing process. So, most of the bio chemical processes, will have these various units. This is the heart of a bioprocess manufacturing plant. This is called a reactor. It could be called fermentor, if you are fermenting your sugars into the desired product. It could be called a reactor if you are using bio transformation or enzyme catalyzed reaction. So, depending upon the type of a process you are doing, it may be called a fermentor or a reactor here.

This is the heart of the entire bio process. The right hand side, we call it as the downstream and the left hand side is called the upstream. This is where you are preparing your medium, preparing your microorganism and then you feed it into the reactor. Here, you are recovering your product that means you are isolating your product; you are purifying your product and making it a hundred as pure as possible. So, during the purification, you are going to get side products, unwanted products. You are going to get lot of waste material, which all goes to waste treatment plant. So, after the fermentation, you may recover the biocatalyst.

If it is an enzyme, you would like to recover the enzyme and put it back inside your reactor because enzymes are very expensive. So, you do not want to throw the enzymes out. So, you try to recover the enzyme and put it back in to the reactor. If you have unconverted raw materials, you again like to recover and put it back.

Suppose you have glucose, unconverted glucose, you try to recover the glucose and put it back because cost can be reduced. Manufacturing cost can be reduced by doing this type of recycling. This is called recycling. Once you have recycled your raw materials and your catalyst, you have your product, very dilute product, 5 per cent, 10 per cent of product. So, you are recovering your product here. Then, finally you are purifying your product and then unwanted material goes into waste treatment. Unwanted material means it could be dead biomass, it could be cell debris, it could be waste material, salts, and it could be broth and so on actually.

So, they all go into the waste treatment plant before they are completely disposed into the affluent treatment facility actually. So, a typical manufacturing plant, a bio process manufacturing plant will be looking like this. Our focus is more on this right hand side of it. This is what is called the downstream and this is what we are going to talk about in the

course of our lectures.

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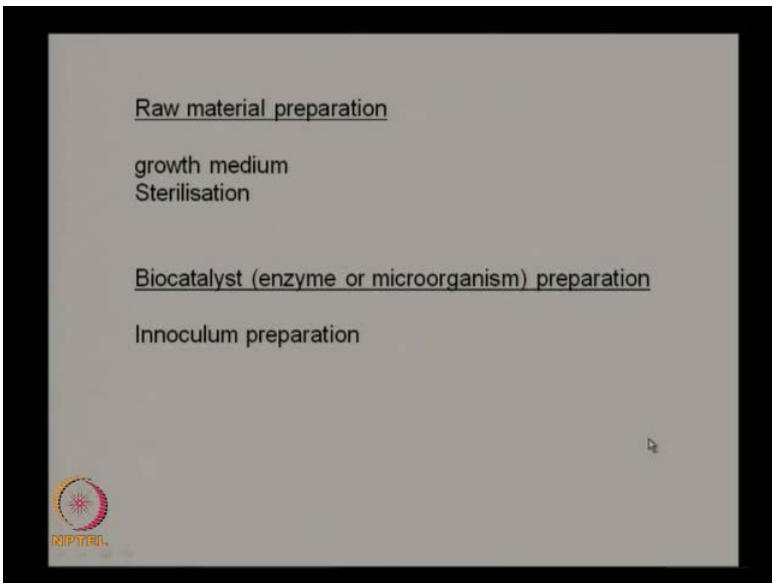

So, what is the raw material preparation? That is the left hand side, which we saw. We are preparing the growth medium. Growth medium means it may have a carbon source, nitrogen source, minerals, and micro nutrients, salts everything, which are needed for your growth of the microorganism. Then you need to sterilize all the growth media because whatever you feed into your fermentor has to be completely sterilized. So, this is what is called raw material preparation section.

Then, you have a section for preparing your microorganism or the enzyme that is the biocatalyst preparation section. So, if you are preparing the microorganism, you need have an innoculum medium preparation section. You need to have the microorganism at the correct stage of growth. Then you need to sterilize the biocatalyst so that both the raw materials and the enzymes or the microorganisms are fed into the fermentor. That is the upstream section.

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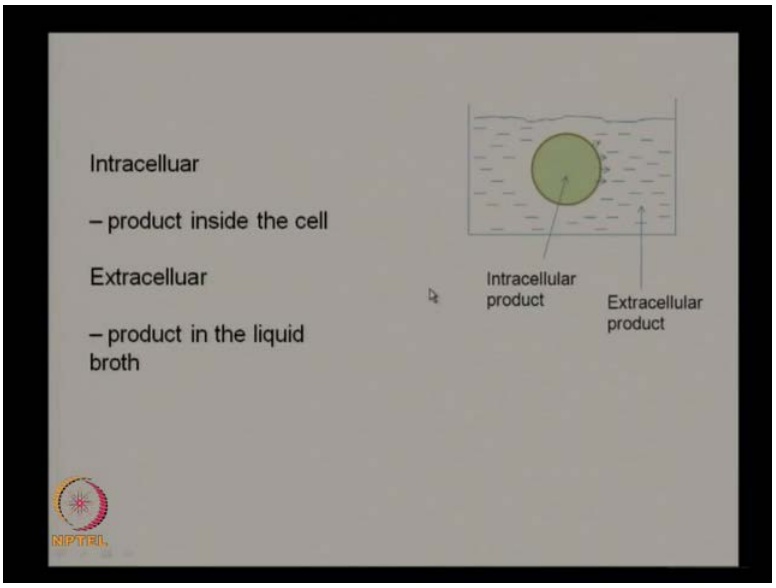

Your product could be either inside the microorganism, inside the bacteria or fungus or it could be in the medium. That means if it is outside the microorganism that is called as extracellular. If it is inside the microorganism, it is called intracellular. So, if the product is in the extracellular, life becomes very easy. All you have do is you have to remove the bacteria. Your product is here, but then if it is inside, then it is much more troublesome. You have to first recover all the microorganism or cells and then you have to break the cells and then extract your product.

So, the number of unit operations, number of process steps are more. That means you are going to take more time doing it. That means cost also increases. So, extracellular product is always desired because your product is in the medium. So, you just have to filter, take your cells and throw them out. You have your product in the liquid medium.

So, in intracellular, you have to recover your cells, break the cells and then get your product out of the cells. So, the number of steps is more. Cost is also more. There are many products, which are made intracellular; many products which are made extracellular. Some proteins are intracellular that means they grow inside as a Golgi body or they are inside. So, you need to take it out.

That means you need to break the cells and take the product out. If it is a liquid product you are interested in, then they are extracellular. You can take the liquid medium and then recover your product actually. So, ideally you would like to have extracellular

product rather than intracellular product because as I said the number of steps involved is much more in intracellular and the number of steps involved in extracellular are less.

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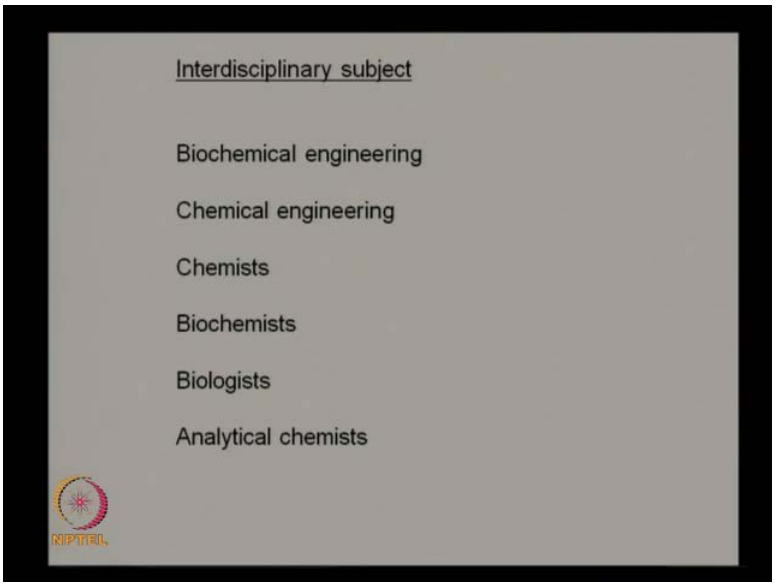

If you look at down downstream process, it is inter disciplinary area. You need expertise of biochemical engineering. You need expertise of chemical engineering also because there are several operations, which are very chemical engineering oriented. You need the support from chemist. You need support from bio chemist, biologist, and analytical chemist. So, all these people have to put in their expertise and knowledge if you want to make a good downstream process. So, it is extremely interdisciplinary and it is a multi disciplinary task.

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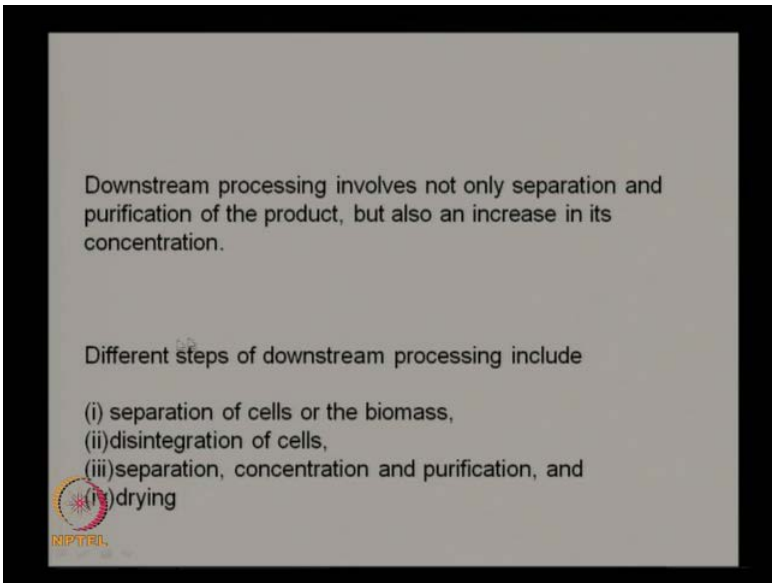

So, it not only involves separation, purification, but you are also increasing the concentration of your desired product like I talked about the alcohol fermentation after the fermentation from the fermentor, the amount of alcohol or ethanol in the broth may be 8 per cent, 9 per cent. But, finally, you want to make your concentration to almost 90 per cent. So, you are not only purifying your product. You are increasing it to very large concentration.

So, what are the different steps in the downstream process? It may have separation of cells or you sometimes call it biomass because it is a biological mass. As cells grow, they form large amount of mass. Then if it is an intracellular as I said your product is inside the cells, then you need to break the cells. That is called the disintegration of the cells. So, you need to break the cells. If it is an extracellular process or product, you do not need this particular step. You can jump from here to here. Then you need to separate your product. You need to concentrate your product. Then you need to purify your product.

So, this number of steps depends upon the type of product. If it is a pharmaceutical product, the product has to be very pure. So, you need to keep on purifying it to almost 100 per cent, but if it is a bulk chemical, you do not need to do too much purification. So, you can start from separation, concentration, little bit of purification. If it is a animal feed, again it may be lying between too much purity and too little purity. So, depending

upon the type of product, the purification steps vary. So, for each one of them, you may have several unit operations and by the time you get a product of extremely high purity, you may have several unit operations done.

Finally, if it is a solid material, you would like to dry the product so that all the moisture is removed because moisture leads to sometimes instability in the product; especially enzymes lose their activity over a period of time if there is moisture. That is called shelf life. So, if you keep your enzyme with moisture, over a period of time, enzyme may lose the activity. So, the best thing is to dry it up. So, when you dry, even solid material loses its volume.

So, storage becomes much easy. Transportation becomes much easier. So, the cost of transportation also goes down. So, drying is usually resorted to if you are interested for solid product. If it is not a solid product, then of course, if it is a liquid product, you do not need to dry the final product. So, you may stop with here. So, generally solid material is you is done with trial.

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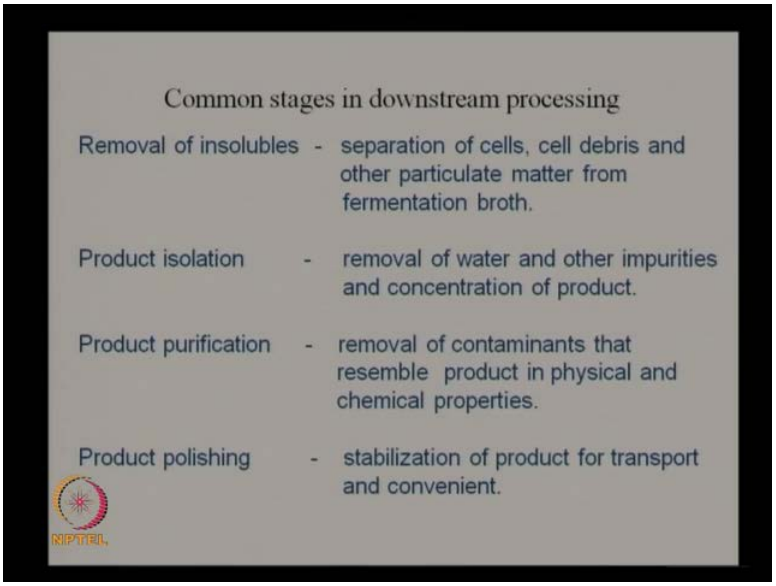

So, as I said there are the common stages in down streaming. So, you are separating the cells. You are separating the cell debris because the broken cells are called cell debris, then other particulate matter from fermentation; especially we add lot of salts in the fermentation broth. So, the salts start precipitating out. So, that is called and then they start agglomerating. That is called the particulate matter. So, all these have to be

removed. That is the very first step in downstream and that is called the removal of insolubles.

Then, the next step will be product isolation. The product concentration will be very small, 7 per cent, 8 per cent. So, you need to take out the product from this large amount of liquid. So, you may like to remove water. Water is present because most of the fermentation is done in aqueous. So, there is plenty of water. You are adding salt. So, you need water again to dissolve the salts. So, there is plenty of water in your fermentation broth, so removal of water. There are se several impurities present. So, you are removing the impurities as well. That is called the product isolation.

The next step is product purification. That means you are trying to purify the product. So, by end of product isolation, your product may be 30, 40, 50 per cent pure, but then you if you want to achieve 90 percent pure, 95 percent pure, then you need to resort to this product purification. You are removing the contaminants. There could be side products, which may be contaminants. There could be unwanted reactions, which may be reactants, which may be contaminants.

So, you need to remove all these. Then you need to bring in the physicochemical properties of those contaminants. Finally, you are doing the product polishing. That means you are stabilizing the product for transport convenience and shelf life. That is called the product polishing. You may add certain antioxidants too decrease the oxidation of your desired product over a period of time.

You may add other stabilizers so that your product does not degrade over a period of time. You may add stabilizer so that the ph does not vary when you keep it in the shelf over a long period of time. So, like that, you will be adding several stabilizers so that the product shelf life is improved and the product transportation is also improved.

So, these are the common stages in your downstream processing. Removal of insolubles; that means you are removing the solids from the liquid. Then the product isolation that means you are isolating your product from a very large quantity of liquid material concentrating it up to 50, 60 per cent, and then purifying the product almost 90 percent pure or even 99 if it is a pharmaceutical product. Finally, you are adding some stabilizers to your product so that you improve the shelf life of the product for a very long period of time. So, these are the various steps in the downstream one has actually.

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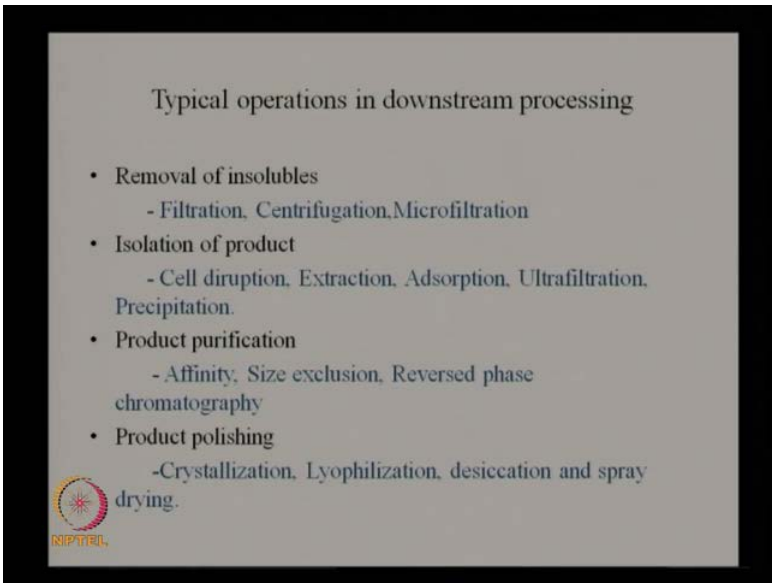

So, how do you remove insolubles? That means you are removing solid materials from liquids. So, there could be many techniques. We will spend time in the next course of the various lectures. Solids can be removed by filtration. They can be removed by centrifugation. They can be removed by membrane type of micro filtration, nano filtration, membrane filtrations and so on. So, all these techniques are meant to remove solid from the liquid.

Next is isolation of the product. It can involve cell disruption. That means if your product is inside the cell, you are breaking the cells. That is called cell disruption. The cell disruption can be a mechanical type of process. It could be an enzymatic process. It could be a thermal process. So, again we will spend the time in the next course of time weeks on this particular aspect, cell disruption. Then you are extracting your product using a solvent. You may be able to use adsorption type of technique to remove your product or you may use ultra filtration technique or you may use precipitation type of technique. All these techniques are meant for isolation of your product.

The next step is once you have isolated the product, you are purifying your product. This involves chromatography, different types of chromatography, affinity chromatography, size exclusion chromatography, reversed phased chromatography. So, you see purification is always achieved by chromatography and chromatography is a very expensive technique. So, it adds to the cost of your final product and chromatography

really purifies your product from may be 60 per cent to more than 90 per cent.

Finally, you are polishing your product. That means if it is a solid material, you can get the product in a crystal form. That means very pure product can be crystallized and you can end up with crystallized product. You can remove the water content by lyophilization or you can even use desiccation or you can even do spray drying like you are coffee powder is a spray dried product, a crystallization. Even enzymes could be crystallized. So, by advantages of crystallization is by crystallizing your enzyme, you are removing all the water present in the enzyme. So, the shelf life of the enzyme or the activity of the enzyme is retained over a very long period of time.

So, you resort to the various techniques mentioned in this slide for achieving your desired product at desired concentration and desired physical chemical properties. So, if it is a solid, bring it into a solid form. If it is a liquid, you maintain it as a liquid form and you try to stabilize it so that it does not lose its activity during transportation as well as when you keep it in the shelf.

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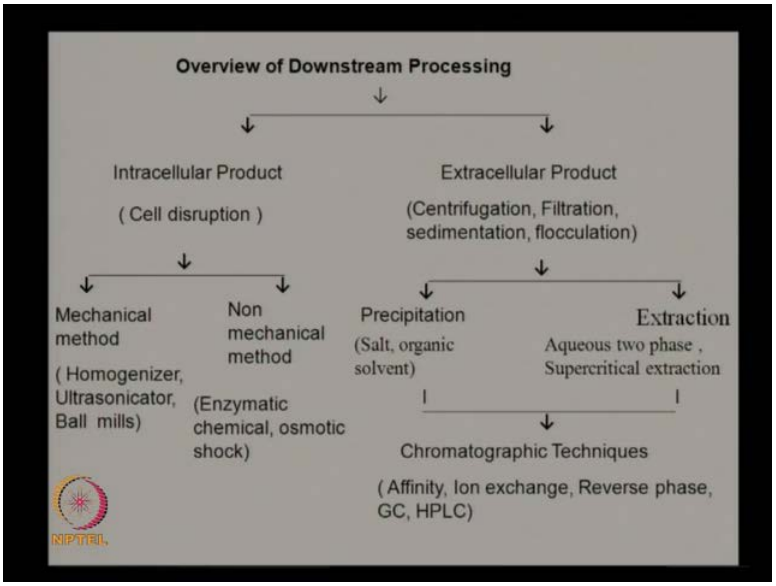

So, this is a nice slide, which sort of separates the intracellular products that means products inside the cell and the extracellular products that means when the product is in the media that is they are outside the cell. So, when you have intracellular product, of course, you have to disrupt your cell. That means you have to break the cells to get your product out, but if you are having intracellular product, you are not interested in the bio

mass or the microorganism. You can throw the microorganism out.

So, you use filtration technique, you can use the centrifugation technique or even sedimentation. Sedimentation is nothing but settling technique. So, if the solids are very heavy, they settle down. So, the liquid on the top rises and you can take your liquid because your product is in the liquid flocculation technique. If your solid is not settling down that means it is not very heavy and the solid is floating on top of your liquid. You can collect the solid by flocculation and remove the top portion. So, the remaining liquid will be containing your desired product.

So, two majors areas, one is intracellular products and the other is the extracellular product. So, both has different types of techniques once you have disrupted your cell. The methods could be either mechanical based methods or it could be non mechanical based methods. Mechanical based methods contain homogenizers, ultrasonicators or ball mills or non mechanical method could be an enzyme.

You can use an enzyme to break the cells or you can use a chemical to break the cells or you can use osmotic shock to break the cells. So, by breaking the cells, you are releasing your product that is trapped inside the cells to outside. Once you have done that, you can go the right hand side for the product isolation and the product purification.

So, product isolation will contain things like precipitations, extractions. So, the precipitation may contain using salt, using organic solvent. Extraction can be you can use a solvent like an acetone or chloroform or ethyl acetate or you can use even water or you can use techniques like supercritical type of extraction here. So, this is the product isolation side of it.

Then the purification part of it is using different types of chromatography like I mentioned before the affinity chromatography, the ion exchange chromatography, reverse phase or gas chromatography or even liquid phase chromatography can be resorted to and you can purify your product actually. So, intracellular requires cell disruption. Once you have disrupted the cell, you can go to this side of it and then use the recovery, product recovery techniques and product purifying techniques.

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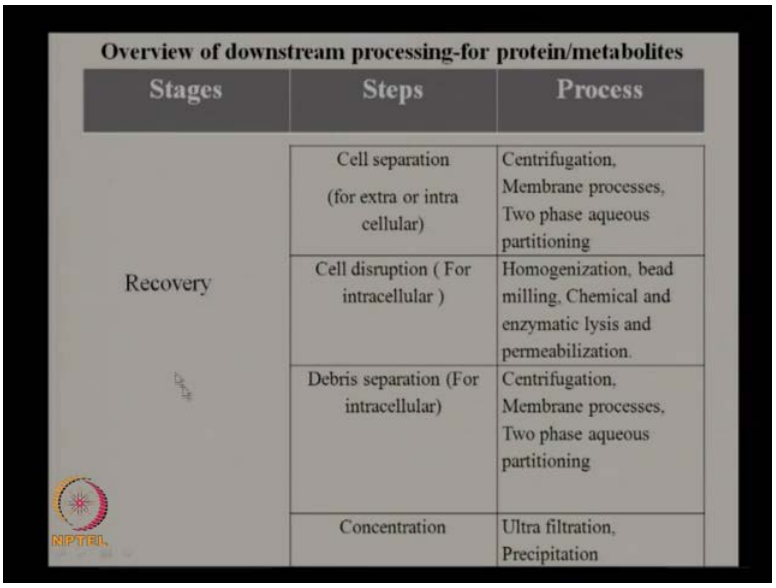

So, if you are talking about recovery whether it is protein or whether it is a metabolite, so you are talking about cell separation. That means you collect you cells first and if your cell is your contains your desired product, your cell is disrupted using various techniques I talked about homogenization, bead milling, chemical techniques, enzymatic techniques lysing techniques.

Then once you have broken the cells and extracted the product from the cells, you can remove that cell debris that means the broken cells using the centrifugation membrane process, two phase aqueous partitioning technique and so on. Then finally, you are concentrating your product using different types of precipitations and filtrations.

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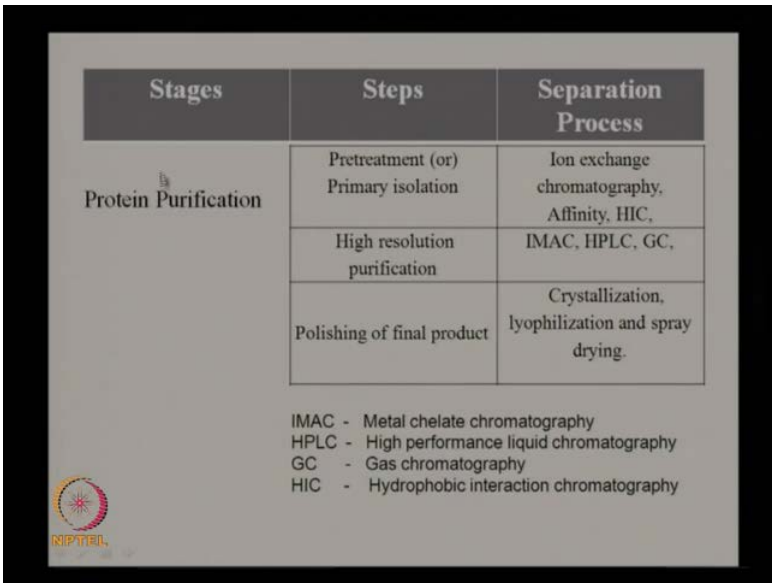

Then, you go into protein purification using ion exchange methods, chromatography techniques, and then finally, the polishing methods where you are using crystallizers, lyophilizes and drying method.

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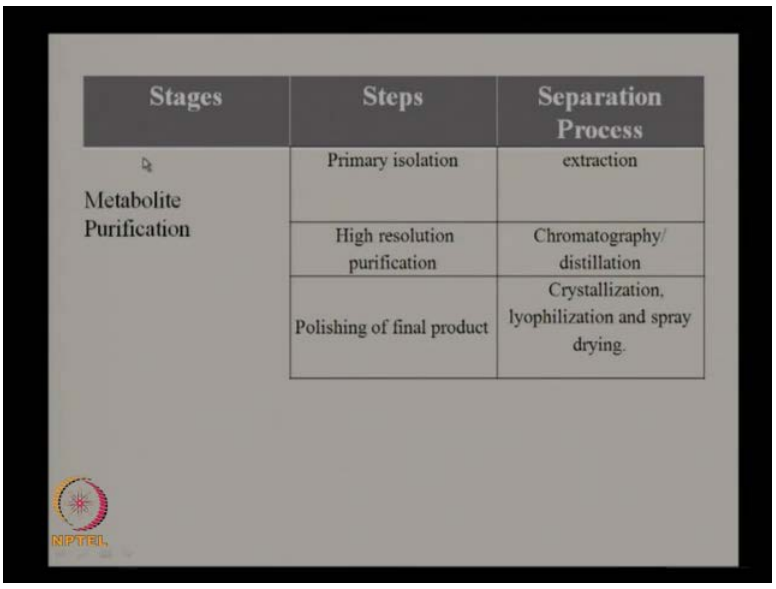

If it is a metabolite instead of a protein, then you can use even distillation type of techniques because a metabolite or a organic chemical can withstand higher temperature unlike a protein. Protein will not be able to withstand very high temperature. So, most of the protein separation technique will involve extraction or involve precipitation, whereas

if it is a metabolite, we can use distillation type of method.

Distillation is very simple method and it produces very pure product depending upon its vapor pressure or depending upon its boiling point with respect to rest of the material. So, distillation is a very easy technique to resort to if the material can withstand very high temperature. Then later on, whether it is a protein or whether it is a metabolite, you may resort to crystallization or you may resort to spray drying or lyophilization depending on the nature of the product, of the desired product.

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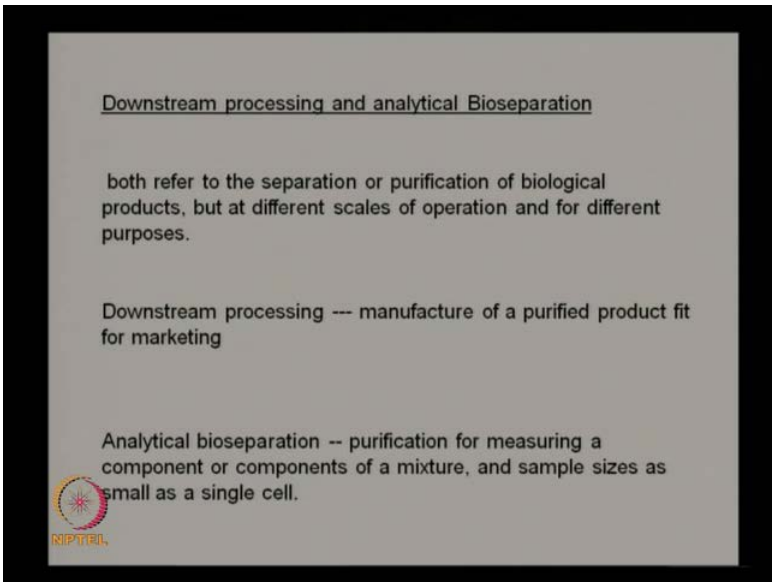

Just like downstream processing, we also have techniques called analytical bio separation. So, in analytical bio separation, just like downstream processing, you are purifying a product. You are separating a product. You are purifying a biological product. That means a biological product could involve DNA or it could be involving a protein or it could involve amino acid and so on. But, the only difference between downstream processing and analytical bio separation is the scale of operation.

In downstream processing, you are manufacturing and purifying a product in very large scale. It is meant for manufacturing and marketing, whereas in analytical bio separation, you are purifying a product, a component from a mixture of components, where the quantity may be very small. It is meant for analytical use. You are interested in a particular bio molecule; you want to study the characteristics of the bio molecule in very large small scale in your lab. So, the quantities are very small unlike a downstream

processing. So, in a downstream processing, you are manufacturing a product.

So, the scale of operation is very large, whereas in analytical bio separation, you use similar separating techniques. But, the idea is you are isolating a small product from a very large quantity of material and you are going to study its properties in the lab. You are going to do certain experiments with that particular bio molecule. So, the scale of operation is very different. That is the difference between an analytical bio separation and a downstream processing.

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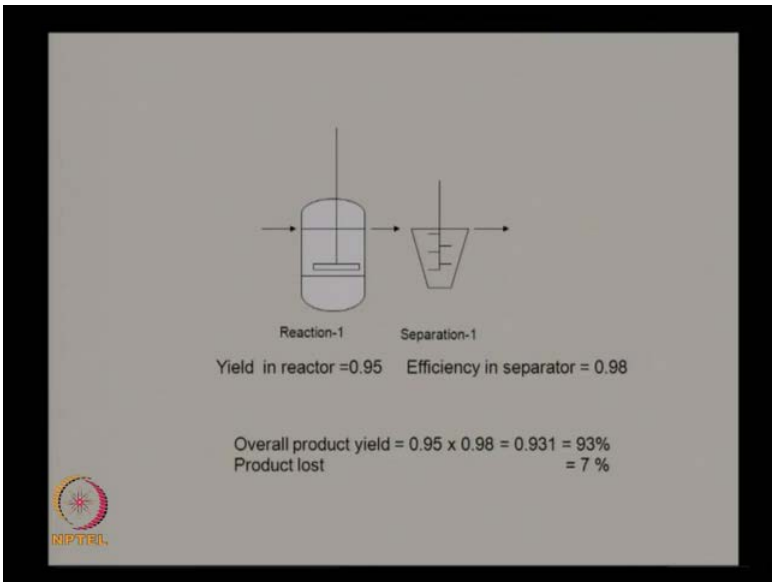

Let us look at the various issues that are involved in downstream unit operation. Now, let us consider. Imagine you have a reaction taking place in a reactor. It could be a bio reactor. It could be a fermentor. Then you are doing a separation, some type of separation, any type of separation you are resorting to and you are getting some product out here.

Imagine the ray yield of the reaction is 95 per cent that means 0.95 and the efficiency of the separation is 0.98. That means a pre efficiency of the separation process is 98 per cent. So, the overall product yield you are going to get is a multiplication of this number with this number. So, when you multiply these two numbers, you are getting 93 percent. That means you are losing the product, 7 percent of the product when you move from here to here. Some product is lost here and some product is lost here during these two steps.

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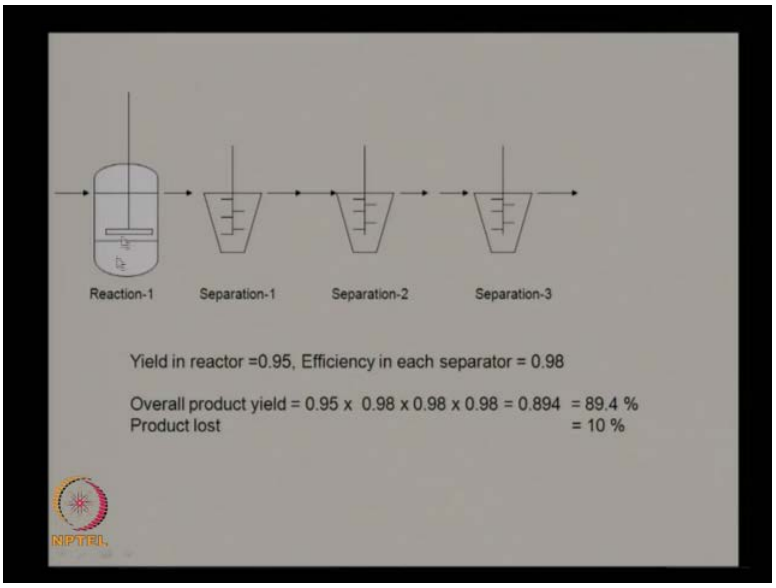

Now, let us go further imagine I have a fermentor or a bio reactor here and I am using three different separation steps. I am doing some separation in separator one. Then I am doing some other separation in separator two and then another separation in separation three. Each one of the separators has certain efficiency. It cannot be 100 per cent. It can be 98, 95, 90 and so on. It will be never 100 percent efficient.

So, you are always going to have some little bit of loss. If the yield in the reactor is 95 percent and efficiency of separation in each of these separators is 0.98, 0.98, 0.98, so over all product yield if I do, I multiply all these numbers. I multiply 0.95 and then I multiply 0.98, 0.98, 0.98. So, if I multiply all these terms together, I get 0.894. That means about 89.54 per cent.

So, about 10 per cent of the product is lost when I move from here to here. Did you notice that? So, when I have three separators and one reactor. Each one has certain, either the yield or efficiency. Although each of the separator efficiency is 98 per cent and the yield in the reactor is 95 per cent, when I multiply all of them, I end up only with 89 percent. That means I am losing 10 percent product as I move from here to here.

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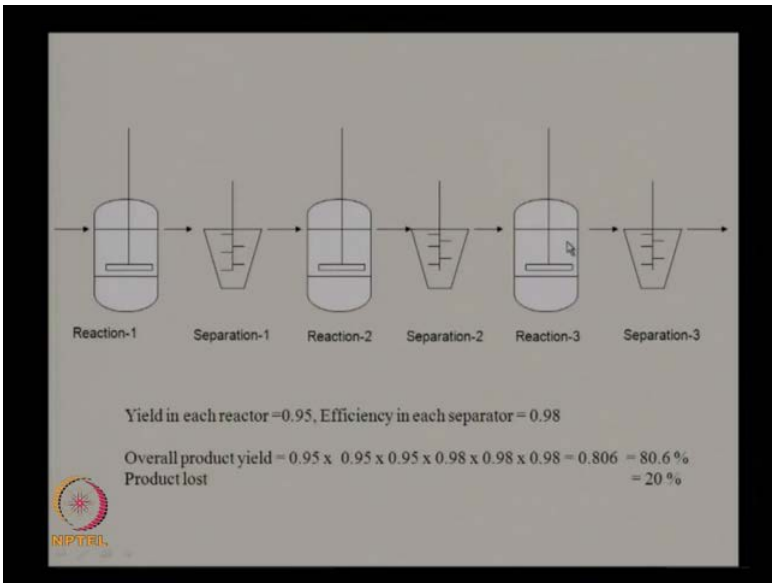

Let us slightly make it more complicated. I have one reactor. I have one separator. I have another reactor. I have another separator. Then I have third reactor and third separator and yield in each reactor is 95 percent and yield in each separator that means efficiency in each separator is 98 percent. So, if I want to calculate the overall efficiency of the entire train of reactor separator, I will just multiply 0.95 three times, 0.98 three times. So, what do I get? I get overall efficiency as 80 per cent. That means I am losing about 20 per cent of the product when I move from here to here.

So, just by putting three reactors and three separators and although the efficiency in each separator is very high, 0.98 and yield in each reactor is very high, 0.95, I am losing 20 per cent of the product. That means my overall production yield is only 80 per cent. If I keep reducing this lost, I will be able to increase this particular term. That means I will be able to manufacture more of the desired product that means which adds to the overall sales. So, this is a very important concept.

One needs to understand that efficiency in each separation operation has to be extremely high and yield in each reactor has to be extremely high. Even if they are very high, if you have a train of separators, if you multiply the efficiency in each of the separator, you may end up having an overall yield for the entire train to be much less, which you could not have imagined.

That means the amount of product lost could be pretty high. Although when I look at

each of the separator, I may think each of the separator is performing extremely well because each of the separator has 98 per cent separation efficiency. Each of the reactor has 95 percent yield, but when I multiply all of them, I will end up only with 80 per cent and which leads to 20 per cent of loss of overall product when I move from this place to this place.

So, the efficiency of the entire process flow sheet is only 80 per cent. That means we are losing lot of material, which is lost, which cannot be manufactured and sold. So, one needs to consider when one is designing a downstream operation that efficiency of the downstream operation has to be as high as possible. If it is lower, it is going to add up to several lower downstream operations and the overall down efficiency of the entire downstream process could be extremely low.

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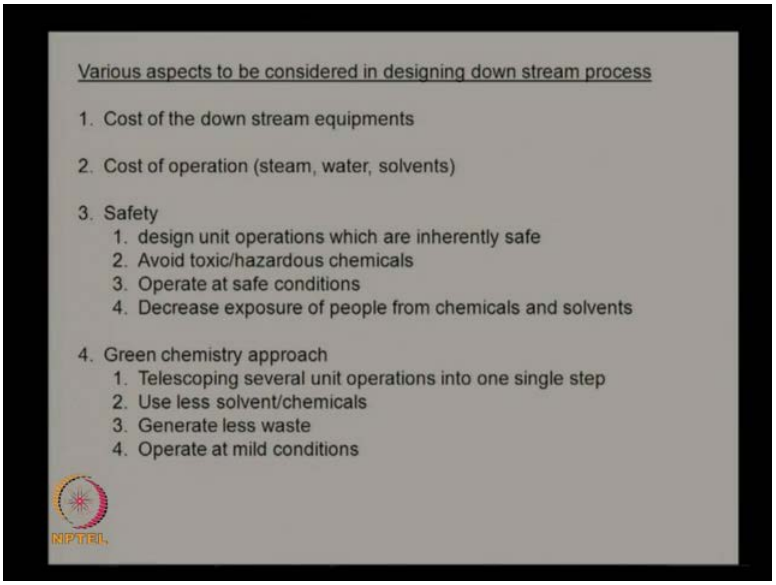

There are various aspects one needs to consider while designing downstream process. The very first thing is what is the cost of the equipments, which we plan to purchase? If I am purchasing a filter, how expensive it is. If I am purchasing a centrifuge, how expensive it is because that is going to add to the capital cost of the equipment. Should I buy a centrifuge or should I buy a filter? Can I do the same job using a centrifuge, which I can do with a filter? If the filter is cheaper than centrifuge, then isn't it better to go for a filter rather than a centrifuge? So, one needs to consider various options with respect to the cost so that the overall cost of the plant is low.

The next step is cost of operation, how much water I am going to use, how much steam I am going to use, what are the various solvents I am going to use because each of them costs money in running the plant. Each of them adds to the operating cost, manufacturing cost of my product. So, higher the manufacturing cost, higher is the selling price of my final product. So, I would like to keep the use of the steam, use of water, use of solvent, use of other utilities as much as low as possible so that the overall operating cost is low.

Next is safety. The units I am designing, do they have inherently safe operating conditions or they are going to be unsafe? Am I using toxic chemicals? Am I using hazardous chemicals? Are the conditions I am using like the temperature, pressure, are they safe conditions or are they unsafe conditions? Am I exposing my people to these chemicals and solvents? All these aspects need to be considered when you are designing downstream equipment. They all come under the aspect of safety.

The next one is something called the green chemical chemistry approach. Can I combine two or three downstream into single one so that I do not have to perform some downstream, and then go to another vessel, perform another downstream. Can I do two things at the same time? That is called telescoping.

Can I use less solvent? Can I use less chemicals so that I can use I can produce less waste so that the environment is not affected. Can I generate less waste? Can I do things at milder condition? Do I have to use very higher pressure or I can do at very low pressures or ambient conditions, ambient temperatures. So, all these aspects need to be considered when I am designing downstream equipment because they all add up to a very safe and green chemistry based downstream reactor or operation design.

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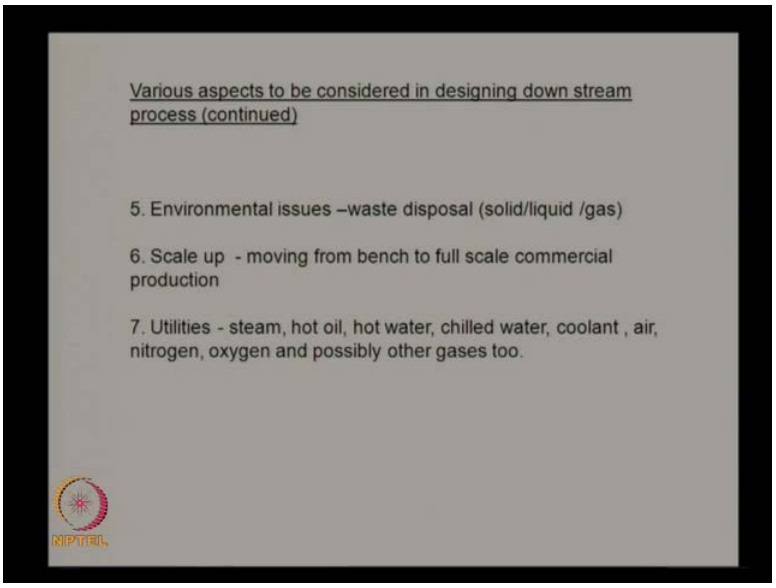

To continue further, what type of waste I am producing after I do the downstream purification or downstream isolation; when I am isolating a product, whatever is left behind is a waste. Does the waste contain solid? Does the waste contain liquid? Is the waste toxic? Can I just dispose it in environment or do I need to do any treatment? Does the waste produce gases? All these aspects need to be considered. So, when I am talking about waste, it is solid, it is liquid, it is gas. All these three need to be thought of. Am I producing toxic, obnoxious gases during my downstream operations? So, these environmental issues need to be considered when you are designing a downstream.

Next is scale up. When we develop a downstream process in the lab, we are doing it at 100 milliliters scale, but when we go for a manufacturing facility, we are talking about in terms of 1000, 10,000 liters scale. So, that is a big large increase in volume. Will I be able to carry out the same efficient process as I did in my lab at 100 milliliter scale? So, that is called the scale up; moving from bench to full scale commercial production. So, when you move from lab to large scale 1000 or 10,000 liter scale, do things happen same way as happened in a small scale? So, you need to consider those aspects as well.

Finally, the utilities, what type of utilities are required for performing these operations. Do I require steam? Do I require hot oil? Do I require hot water, chilled water coolant? Do I require nitrogen gas or oxygen gas? So, all these need to be considered. So, if I am cooling something, I will require cold water or chilled water. If I am heating something, I

will require steam. If I heat at very high temperature, I will require hot oil? If I want inert condition, I will require nitrogen or carbon dioxide. If I want to do an oxidation, I will require oxygen. So, you need to depending up on the type of downstream you are doing, you need to de decide what type of utilities are required and utilities add to overall operating cost. So, how efficiently can I do so that my overall operating cost is always low? So, you need to consider from that angle as well the use of utilities, minimize the use of utilities so that your operating cost is less. So, all these aspects need to be considered when you are doing a design of downstream unit.

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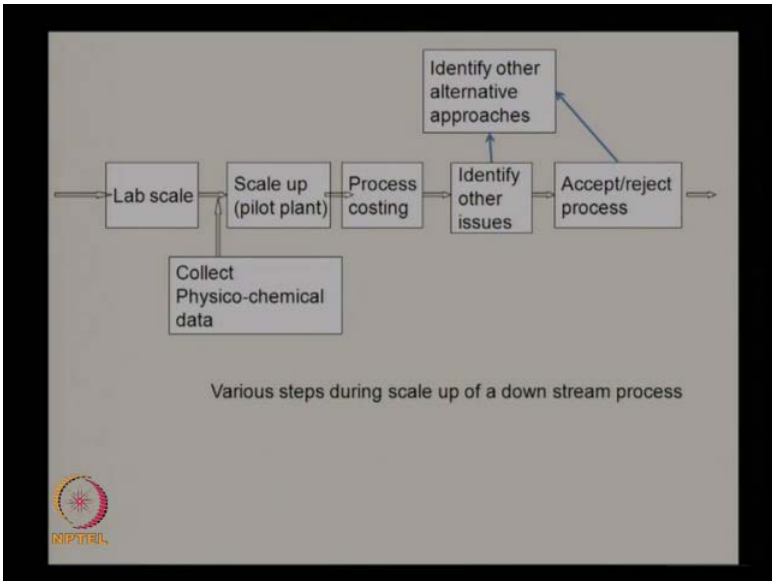

So, I talked about scale up. When we move from a lab scale, which is say about 100 milliliter scale, when you move to 1000 or 10,000 liter scale that manufacturing stage, it does not happen in one go. You need to consider several aspects when you are doing a scale up. So, this particular slide show you what are the various steps involved in scale up.

So, this is your first step. This is lab scale process. You are developing a process in the laboratory scale. Their overall quantity may be only 100 ml. You are modifying conditions, checking on ph, checking on temperature, checking on carbon source, nitrogen source, checking on quantity of various fluids so that you get a very optimum process in the lab scale.

Then, you move to pilot scale. So, from 100 milliliters scale, you do not directly go to

1000 or 10,000 liter. You may go to 10 liters or 100 liters scale. So, that is called a pilot scale. So, at that time, you will require a lot of data. You will require physicochemical properties, you will require density of the fluids, you will require surface tension, boiling point, vaporizing point, all these physicochemical properties for various fluids you are using are required. So, you may be collecting it from literature or you may be collecting it from your own lab because if you want to design something, you need all the physicochemical data here.

Once you have done that, you need to look at the cost. How costly it is? Is it okay? Can I do this whole job? Is it economical or should I follow some other technique? So, this is where you do a costing here. Once you have done a costing, you know how expensive this particular step is. Then you identify other issues. There could be several other issues which I talked about. There could be safety issues. There could be very large increase in cost issues. There could be green chemistry issues and so on actually.

So, if you are not very happy with that, you may think about alternative approaches. That means how can I perform this aspect using some other technique, some other downstream purification techniques. So, that is the time you say should I accept or reject. If I accept, I may go further, do further scale up, go the manufacturing plant and do it in a very large scale.

But, if I decide to reject, what do I do? I then think about some other operations. So, initially I might have thought of doing the purification using distillation, but I am not very happy because of safety. So, I may go and think of filtration or membrane type of filtration. I might have initially thought of a centrifugation. I find centrifugation not very good.

So, I may go into membrane filtration. So, you may change from one downstream to another downstream because the cost may be very high or the safety issues may be very problematic or you may be using unwanted toxic chemicals, waste issues may be too much for you to handle. So, you may go into some other type of downstream process actually.

So, these are the various steps in scale up when you move from the lab scale right up to the manufacturing scale, so it is almost like an iterative process sometimes. You may go all this way might have spend about six to twelve months and you find out there are

issues and you may come back again and start all over again. So, sometimes you may lose out time because of this particular aspect of accept of or reject.

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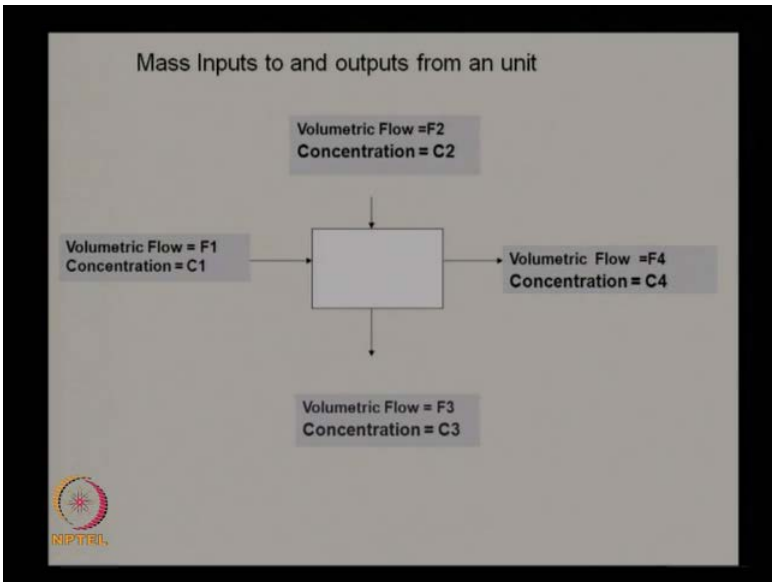

Let us look at something called the most important thing that is called the mass balance. Imagine a unit. It may have two inputs. Liquid is coming in at certain volume and at certain concentration of the product, two liquids are going out at certain volume at and some other concentration.

So, this is liquid one coming in at volume flow rate 1. The volumetric flow rate could be say liters per hour or meter cube per hour and so on. Similarly, the concentration can be gram mole per liter or grams per liter micro molds per liter and so on actually. Then another liquid may be coming at some other flow rate, some other concentration. Two streams may be going out at some other flow rate and some other concentration. I am just showing two inputs and two outputs. You can have multiple inputs and multiple outputs depending upon the type of downstream.

For example, you may be having a extraction. In extraction, you have your liquid. You are adding a solvent. So, you have two liquids input and then you are extracting using a solvent. So, your product may be in the solvent. So, that could be a liquid and the other one could be the bottoms. So, this is a typical extraction type of unit. So, if you want to look at the mass balance at steady state, whatever comes in has to go out at steady state.

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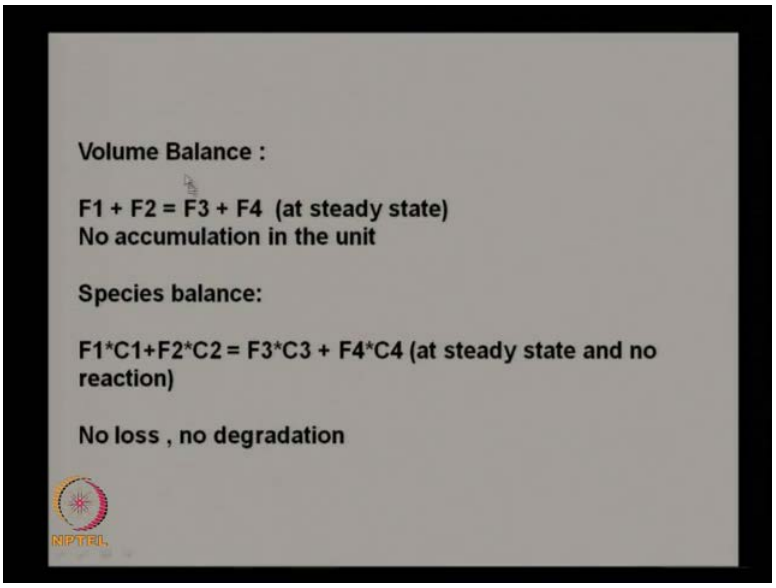

So, if you are looking at the volume balance flow rates, you have a flow rate 1 plus flow rate 2 is equal to flow rate 3 plus flow rate 4. That is at steady state because there is no accumulation in the unit, but if there is an unsteady state that means you are starting the unit at the beginning of the time, there will be some accumulation. But, once it has reached its steady state, whatever comes in that is the two flows inside will be equal to what ever going out. So, F1 plus F2 is equal to F3 plus F4. So, this is a very important equation.

So, if you know F1, F2 though in one stream quantity, you can calculate other stream quantity. That is called the volume balance. Similarly, you can do a species balance that means you know that F1 contains C1 concentration, F2 contains C2 concentration and F3 contains C3 concentration F4 contains C4 concentration. So, if you are doing a species balance, F1 C1 is the quantity of the species coming in, in the stream one and F2 C2 is the quantity of the species coming in, in the stream two.

So, this should be equal to the quantity of the species going out. That is on the right hand side that is F3 into C3 that is quantity of species going out through stream three and F4 C4 is the quantity of species going out in stream four. So, these two on the left should match exactly with these two on the right that is at steady state and no reaction. Please remember, no reaction, if there is reaction, there is going to be change.

So, if you are adding a chemical and the chemical is getting reacted, so it is going into

some other product. So, it would not, this will not match, but if there is no reaction, then F1 whatever is coming in for the species should be equal to whatever is going out at steady state.

So, these two equations are very important if you are going to do a mass balance for species as well as mass balance for the volume. So, the most important point is there is no reaction; there is no loss and no degradation. So, you need to consider those aspects very clearly actually. Similarly, just like we did mass balance, you also have something called an energy balance or you also have something called heat balance.

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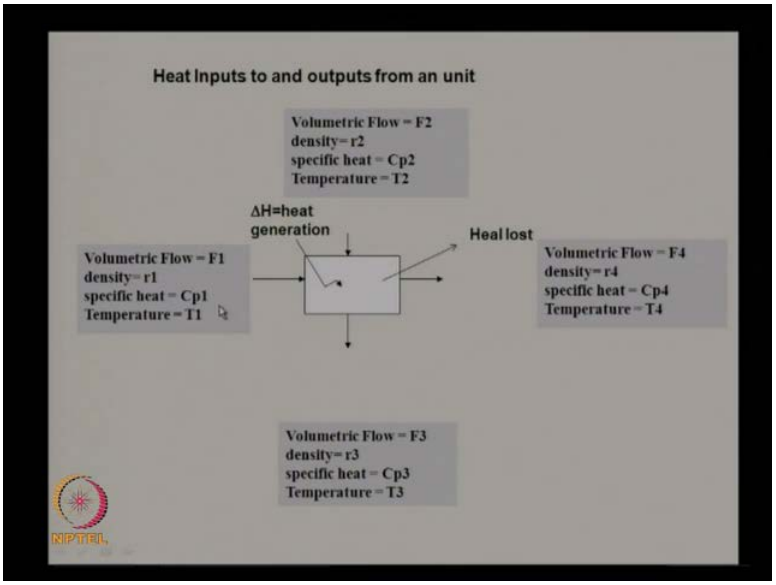

In heat balance, what happens is whatever heat that is coming in and there are heats going out. There are heat generated because of reaction or you are putting in some heat and there are going to be heat loss to the surroundings. So, there heat balance is slightly complicated. You are going to have many streams for heat balance. I think we will talk about this in next class.