INDIAN INSTITUTE OF TECHNOLOGY ROORKEE

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NPTEL ONLINE CERTIFICATION COURSE

Biomedical Nanotechnology

Lec - 12
Nanotechnology in Organ Printing

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Hello everyone I welcome you all to the 15th lecture of this course the 15th lecture is no nano technology in organ printing. So in the previous lecture we have leant how to make artificial tissues and artificial cells in today's lecture we are going to learn how to make artificial organs using organ printing technology so in this lecture we are going to learn.

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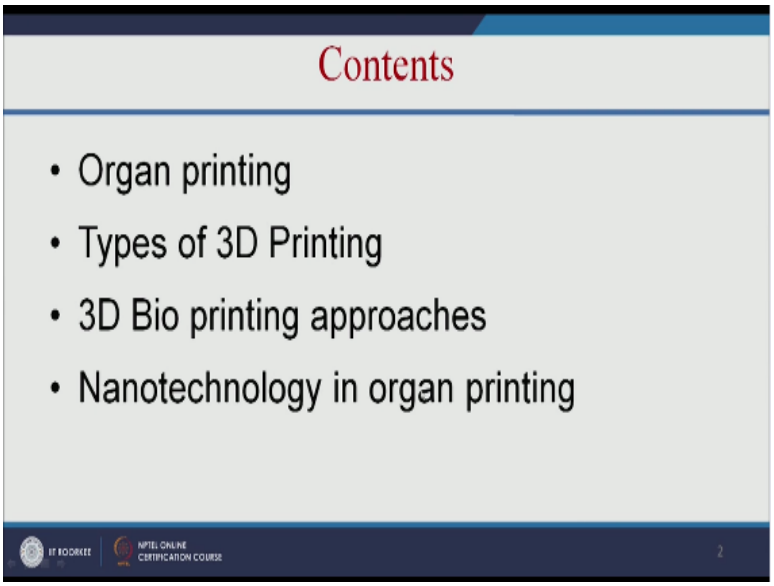

What is organ printing types of 3D printing and what are the various 3d bio printing approaches available and also we are going to learn what is the role of nano technology in organ printing.
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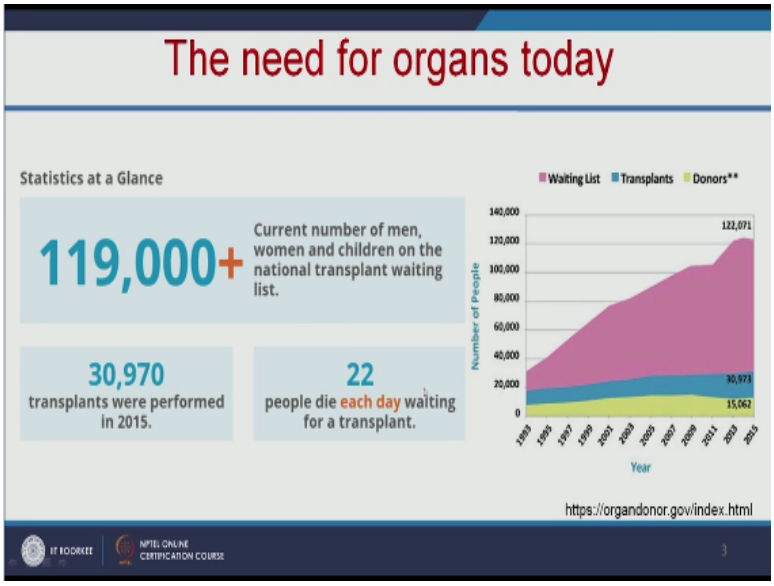

So let us see why we need organ printing so according to American organ donor website at least 1200000 people or in the waiting list for organ transplantation and each day at least 22 people loss there life due to lack of suitable organ transplantation so you can think about the whole world how many people are losing their life without suitable organ transplant.

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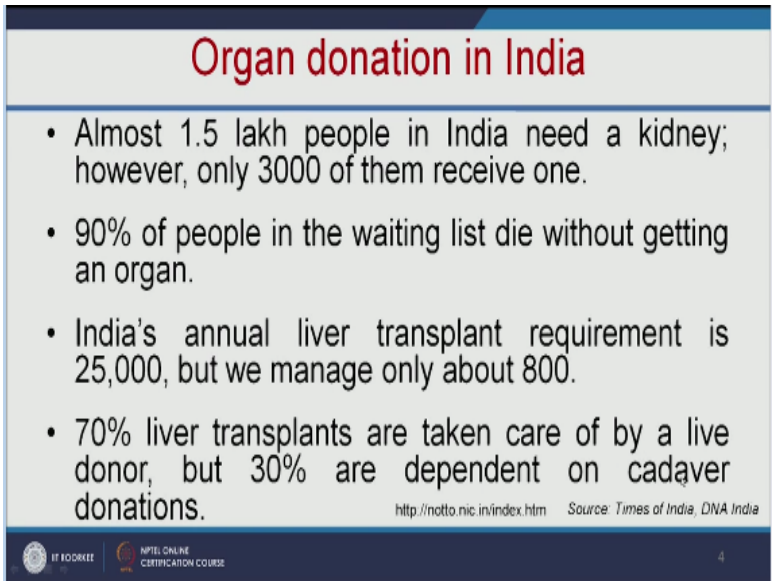

So let us see the statistics in India so in India almost 1.5 lakh people need a kidney but only 3000 of them receive it and 90% of people in waiting list die without getting an organ so Indians annual liver transparent requirement is 25000 but we manage only about 800 and 70% of liver transplants ate taken care of by live donor and reaming 30% are dependent on cadaver donation, cadaver means dead body.

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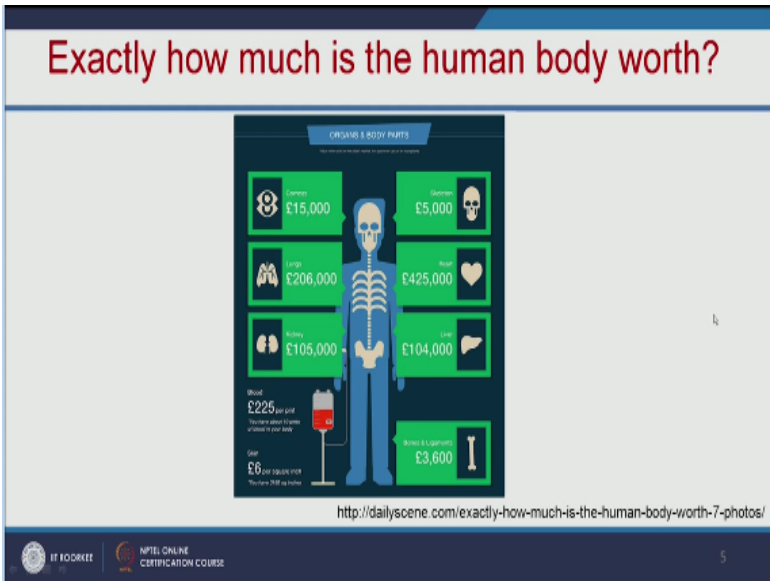

So let see exactly how much is a human body worth okay so we can see the cost of each organ okay so you should be thankful to your parents who have give this much worth property to you so to make this organ what are the approaches available so we are going to see one by one in this lecture.

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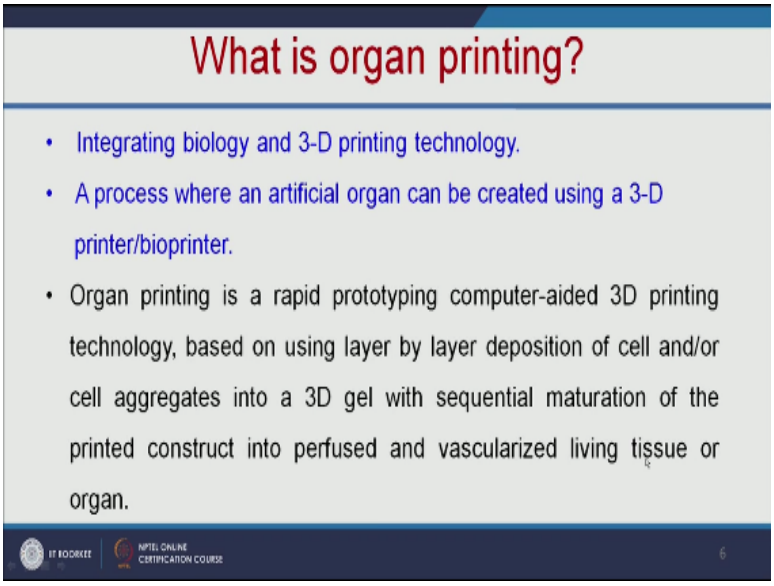

So let us see what is organ printing so it is integrating the biology and 3 D printing technology so it is a process where an artificial organ can be created using a 3D printer or bio printer so let us see the definition of organ printing so organ printing is a rapid prototyping computer aided 3D printing technology so it is based on layer by layer deposition of cell on a 3d gel with a sequential maturation of the printed construct into perfused and vascularized living tissue or organ.

So in this organ printing so we are going to print the artificial organ using the 3D printing technology and here we are going to use biological self as a ink okay and we will be printing the organ on the 3D environment and we will make the complete artificial organ.

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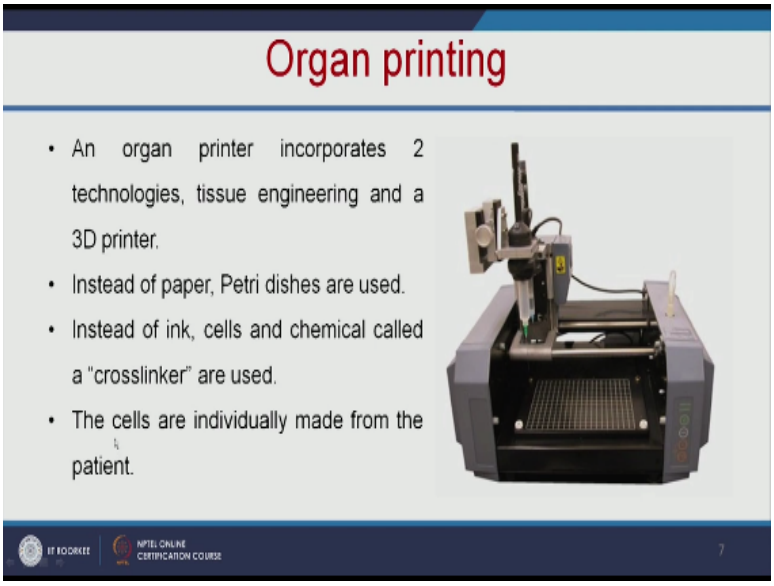

So let see what is an organ printer it is incorporates 2 technology that is tissue engineering and a 3D printer so it is similar to your normal printer but insisted of paper we will be using the Petri dishes and insisted of ink we will be using the cells and chemical called a cross linker so here we are going to take the cell from the patient's own body so there would not be any immune rejection of the organ.

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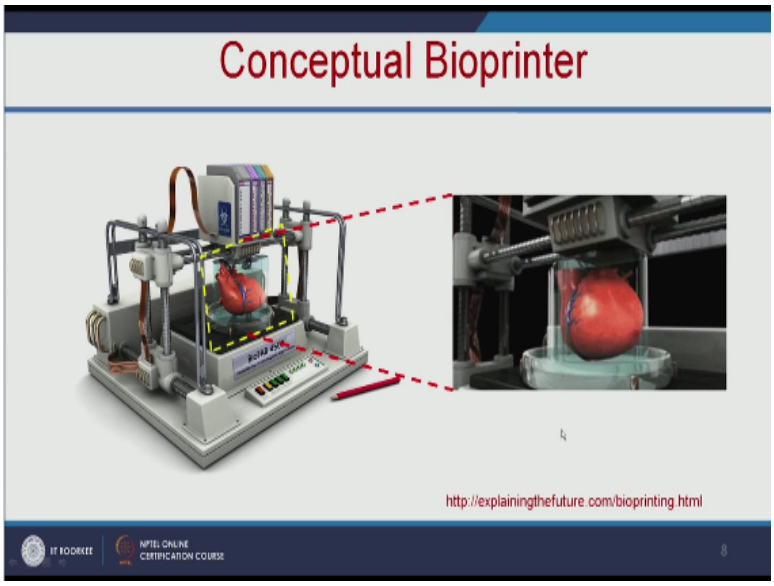

So this a conceptual bio printer in further we may get this kind of bio printer to make the customized organ for the patients need.

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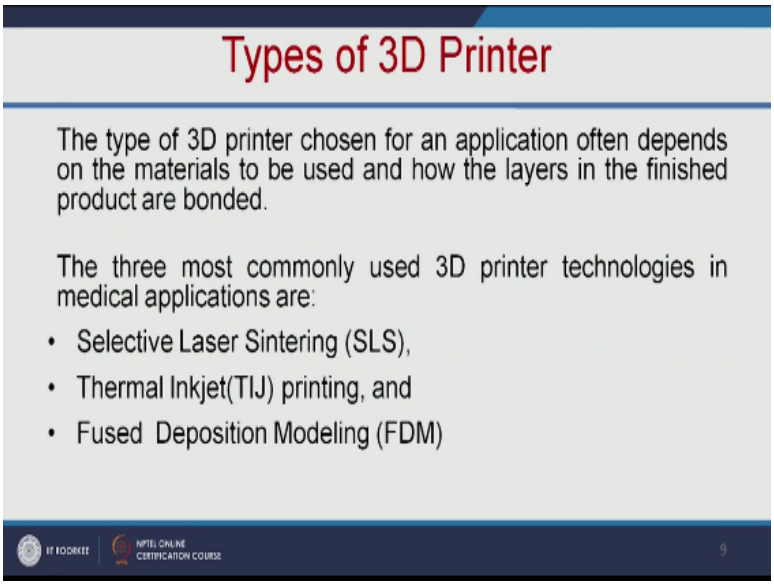

So let us see this types of 3D printer so there are 3 types first one is selective laser sintering next one is thermal inject printing the 3rd one is fused deposition modeling okay so the selection of 3D printer based on the material which you want to use and also how the layers in the finished product are bonded.

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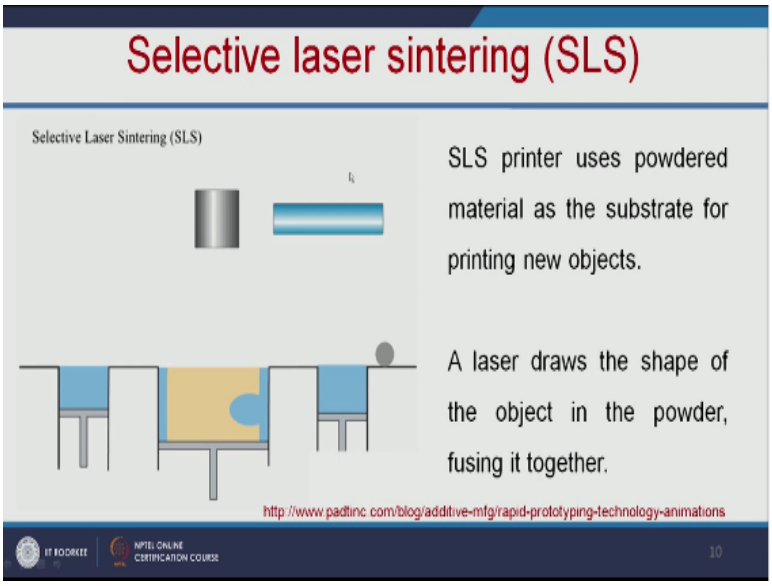

So let us see what is selective laser sintering so her SLS printer uses powder material as a substrate for printing new objects and here this laser draws the shape of the object in the powder and fusing it together.

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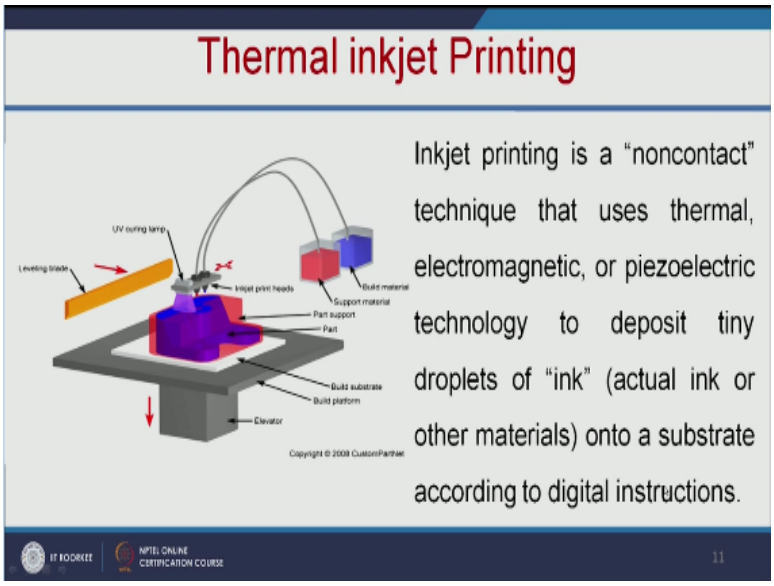

Next one is thermal inkjet printing so here inkjet printing is a noncontact techniques so that uses thermal electromagnetic or piezoelectric technology to deposit tiny drop lets of ink okay it can actual link or it can be another material onto a substrate according to the digital instructions so based on the instructions it will make the particular organ or scale whole for the organ.

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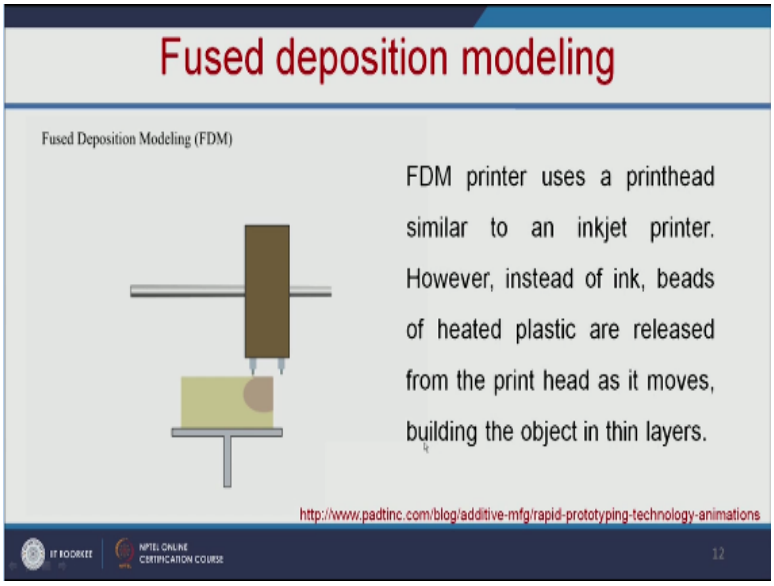

And 3rd method is FDM that is fused deposition model and here the printer uses a print head which is similar to an inkjet printer but instead of ink here the beads of heated plastic are released from the print head as it moves and it build the object in the thin layer.

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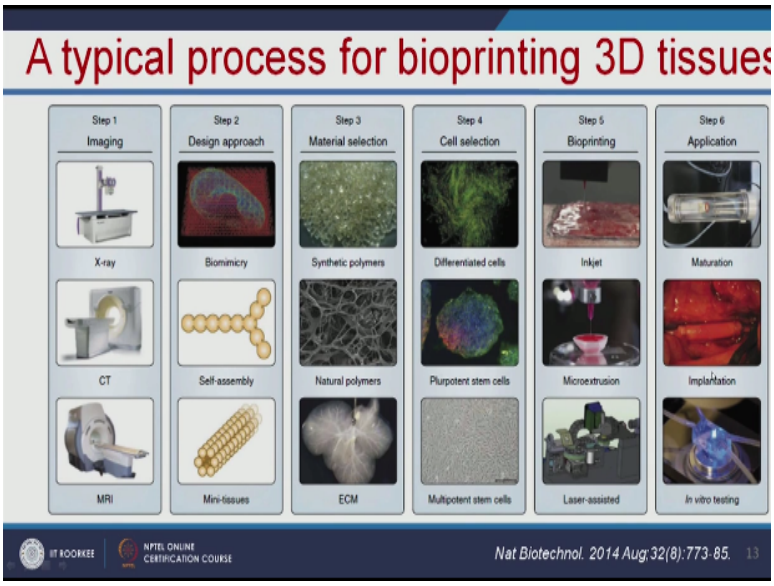

So this is a typical process for bio printing 3D tissues so it has various steps let us see one by one so the first step is imaging so if you want to make the particular organ heart or liver so you have to imaging using x ray, CT scan and MRI so that you will get that complete idea about the organ so then based on that we have to select the design approach so the design approach can be biomimicry or self assembly or many tissues and once you selected the design approach then we have to select the suitable material.

So it depends on the organ which you want to print you have to select the synthetic polymers or natural polymers and we should mimic like your extra cellular matrix then the 4th step is selection of cells the cells can be differentiated cells or it can be stem cells so you can add the cells of the scale whole and it can make the particular type of organ and once you selected this material as well as the cell then you have to bio print that particular organ using inkjet printer or microextrusion or laser assisted printing.

So this techniques I will explain later in detail so once the particular organ is printed so the printed organ will be placed in the bio reactor and it will be allowed to maturation so once the organ is matured in presence of chemical and mechanical signals and this organ will be tested in a bio reactor weather it is performing the function properly then it could be transferred to the patient.

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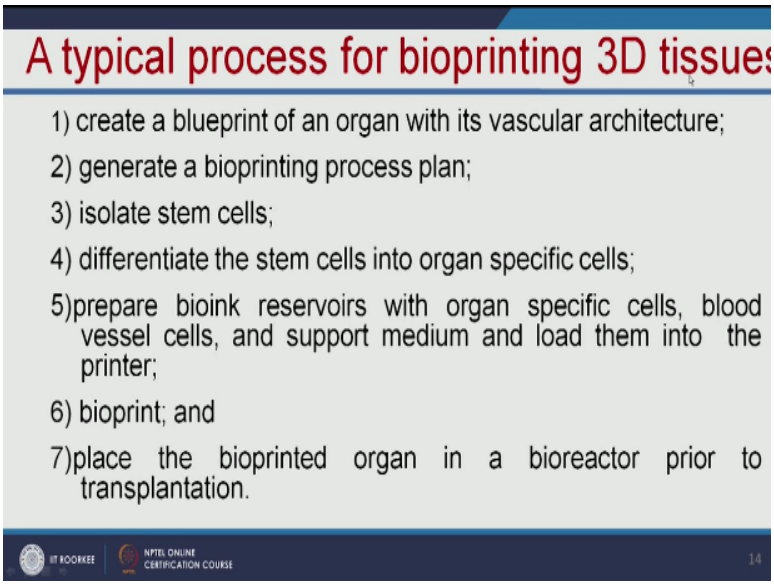

So let us see the steps in detail so as I told you earlier the first step is creating a blueprint of an organ with it us vascular architecture and seconds step is generating a bio printing process plan and 3rd one is isolate stem cells are the suitable cells and the 4th step is differentiating the stem cells into organ specific cells if you want to make the heart or liver accordingly you have differentiate the stem cells into organ specific cells and 5th point is like preparing the bio ink reservoirs with organ specific cells blood vessels and support medium and load them into the printer.

Then you print the complete organ then place the bio printed organ a bio rector prior to transplantation so these are the various steps in the bio printing of 3D tissues or organ.

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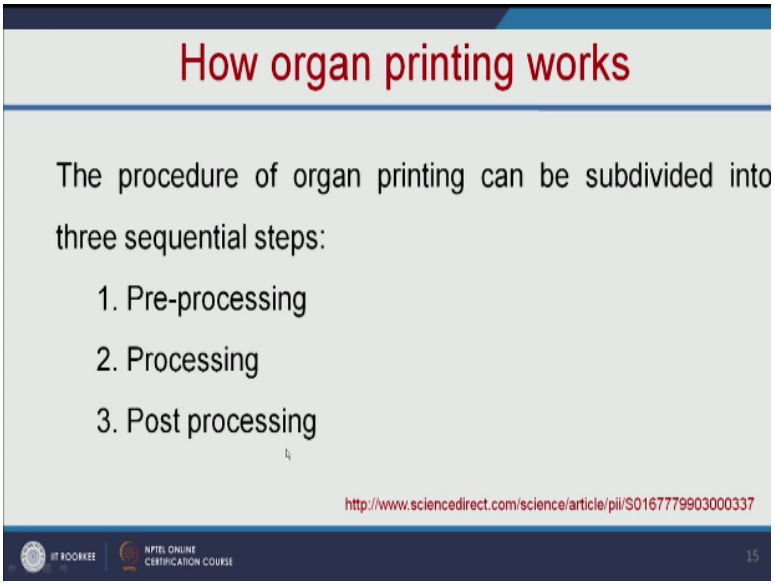

So how organ printing works the procedure of organ printing can be subdivide into 3 sequential steps the first step is pre processing next one is processing and third one is post processing so let us see the step in details.

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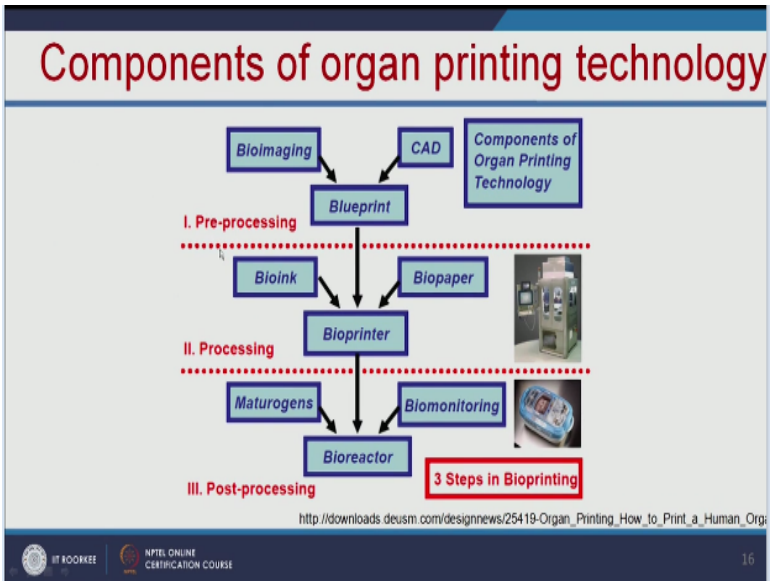

So in the pre processing the first we will be doing bio imaging of the particular organ then based on the CAD you will be making the blue print of the particular organ and in the second step is processing so you will be using these bio ink here the bio ink is your cells and your bio paper is the scale whole, so based on this bio ink and bio paper you will print the particular organ using the bio printer so the organ is printed you will be adding the maturogens.

That is your chemical signal or mechanical signal to make this artificially printed organ into work exactly like your real organ and bio monitoring they will be monitoring the organ in the bio rector weather is function is similar to the original organ okay so this step is called as post processing so let us see this steps in detail.

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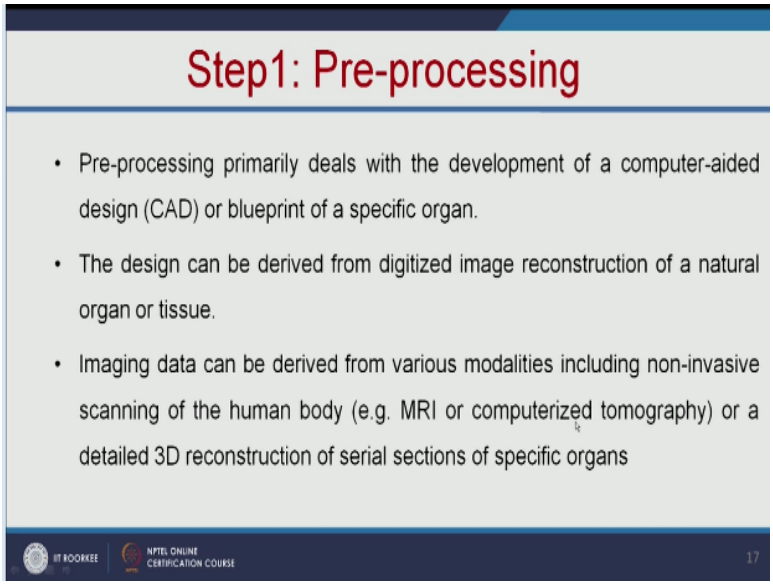

Step 1 is a pre processing so here the per processing primarily deals with the development of computer aided design that is CAD or blue print of a specific organ so here we will be making the blue print of a specific organ okay so the design can derived from digitized image reconstruction of a natural organ or tissue and the imaging data can be obtain using MRI or CT scan of the particular organ.

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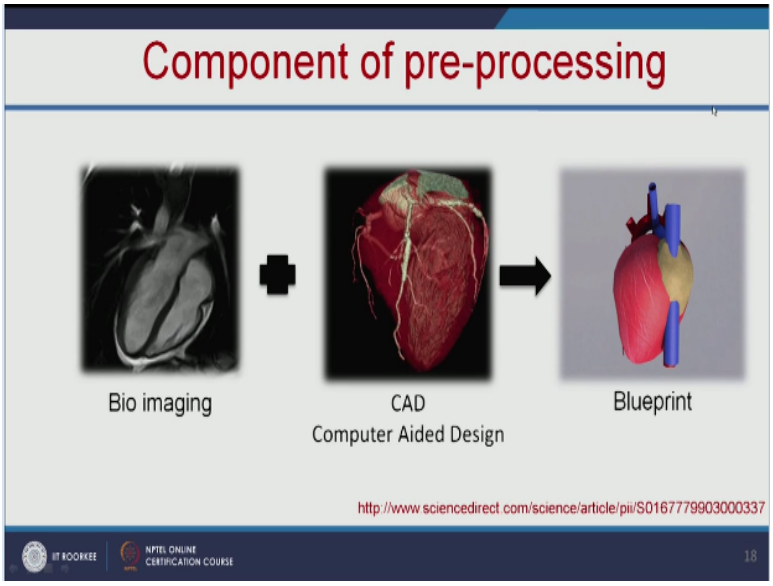

So there first step is bio imaging of the particular organ the using this computer aided design you will make the blue print of the particular organ so this step is a pre processing step.

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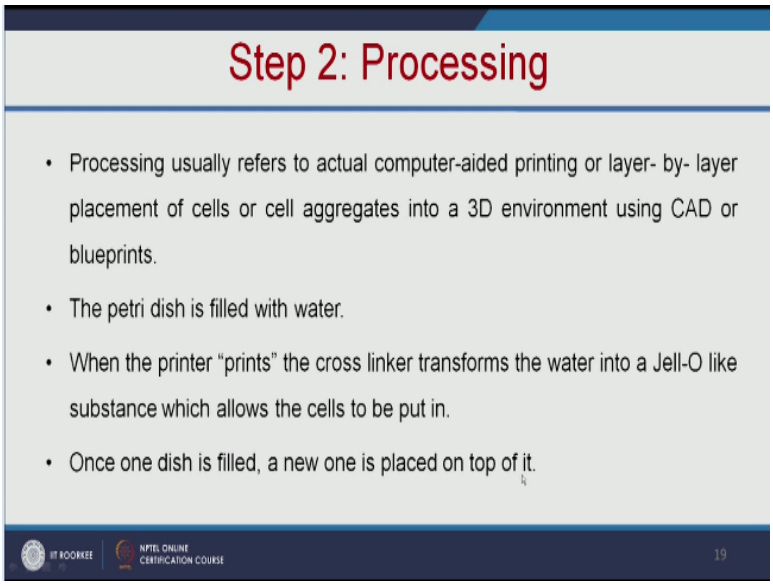

And step 2 is processing here the processing usually refers to a actual computer aided printing or layer by layer placement of cells into a 3D environment using CAD or blue prints so here you will be printing that particular organ using this bio printers and petri dish is filled with water her insisted of paper you will be using this petri dish so it will be filled with the water so when the printer prints the cross linker transform the water into jell – O like substance and which allows the cells to be put in. once one dish is foiled a new one is replaced on top of it.

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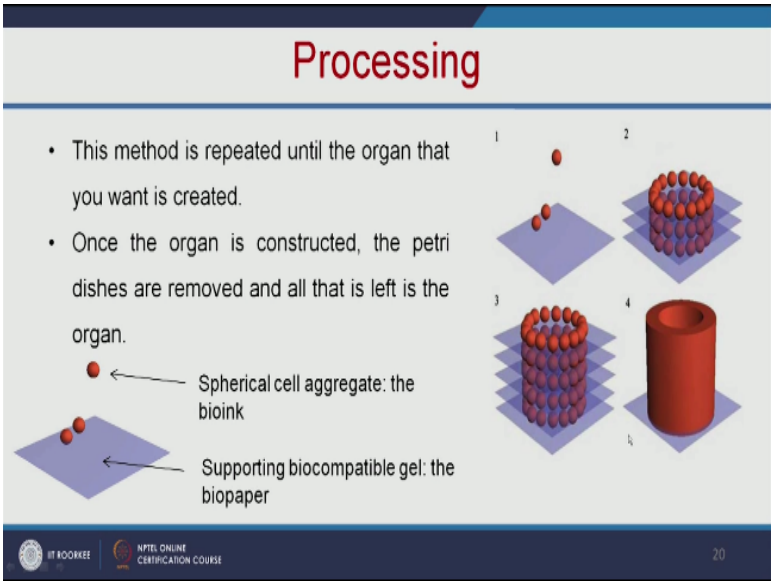

So this method is repeated until the organ that you want is created once the organ is constructed the petri dishes are removed and all that left is the organ so here the supporting biocompatible gel is the bio paper and you must cell is a bio ink the spherical cell aggregate is your bio ink so your cells will be printed on the particular bio paper okay so once the one layer is finished the you will keep the another layer of bio paper and again your print the cells so it is layer by layer assembly of the cells so once all the cells are fused together then you can remove this bio paper.

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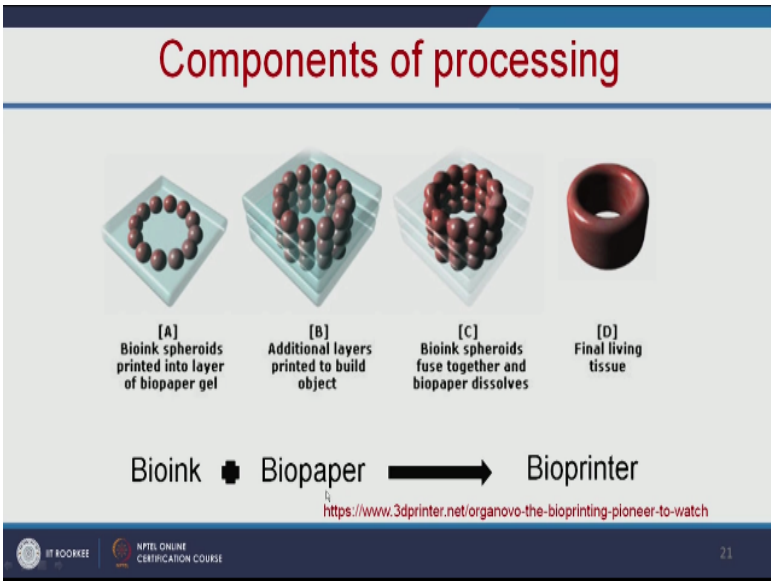

So here the bio inks spheroids are printed into layer of bio paper gel okay so the additional layers can be printed to build the object so once the cells are fused okay so you can remove this bio gel or bio paper and you will get the finial living tissue, so here the bio ink plus bio paper you can bio print the complete organ.

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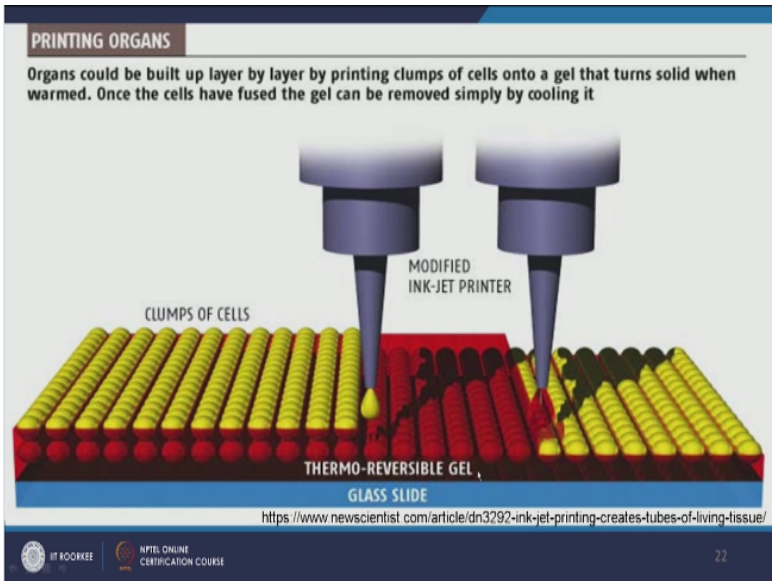

So let us see this in detail so here the organ could be build up layer by layer printing clumps of cells until gel so that turns solid when warmed and once the cells have fused the gel can be removed simply by cooling it so here you can see here this clumps of cell this yellow color one these are the clumps of cell so these are printed layer by layer and these are printed on thermo reversible gel.

So it will be in gel form when this in worm condition and once it is printed you can remove this gel by simply cooling it this is your bio paper.

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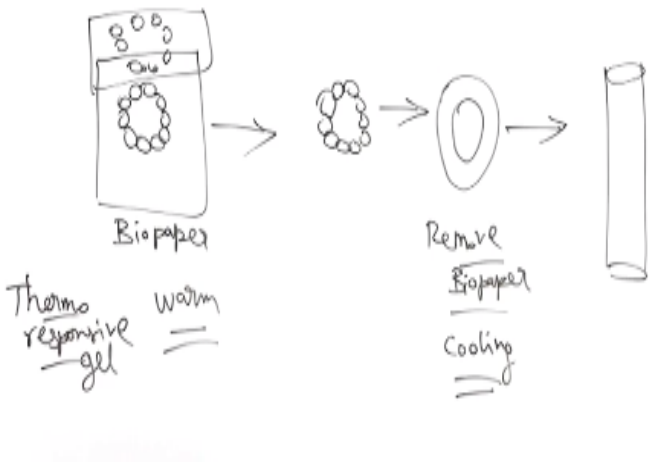

And on top of the bio paper you are going to print this cells, so once it is printed you can put the another paper on the top and you can print the another layer of cells, similarly when you printed all the cells so this cells will fuse together and once it is fused we can remove the bio paper. Make simply cooling it. See at one temperature so it will form a jelly like substance that is a it is a thermo responsive jell so when you cool it you can easily remove the bio paper and finally you will get the desired organ, so this is the overall step of organ printing, so let us see this bio printers.

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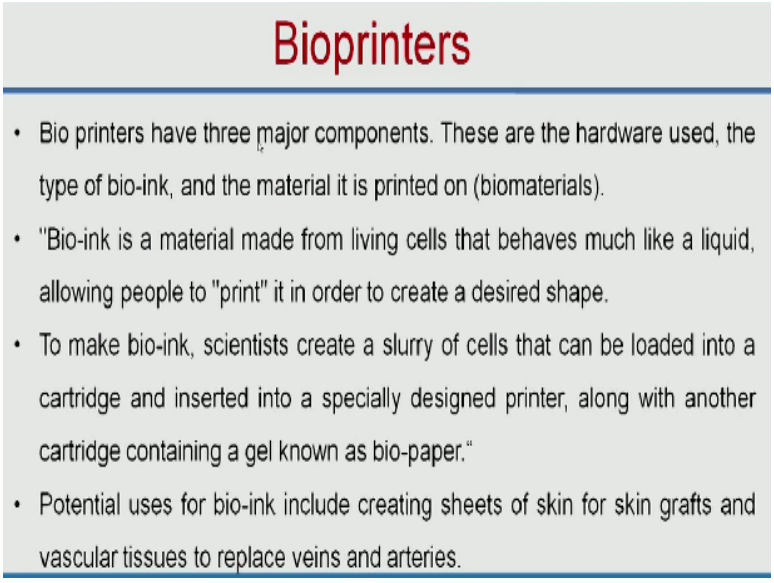

In details so the bio printers have 3 major components so the first one is hardware used the next is type of bio ink and third one is the material it is printed on, so here the bio ink is a material made from living cells that begins much like a liquid and allowing people to print in order to create a desired shape and to make the bio ink researchers create a slurry of cells okay, so that can be loaded into a cartridge.

And inserted in the specially designed bio printers and it will be printed on a gel like substance that is your bio-paper.

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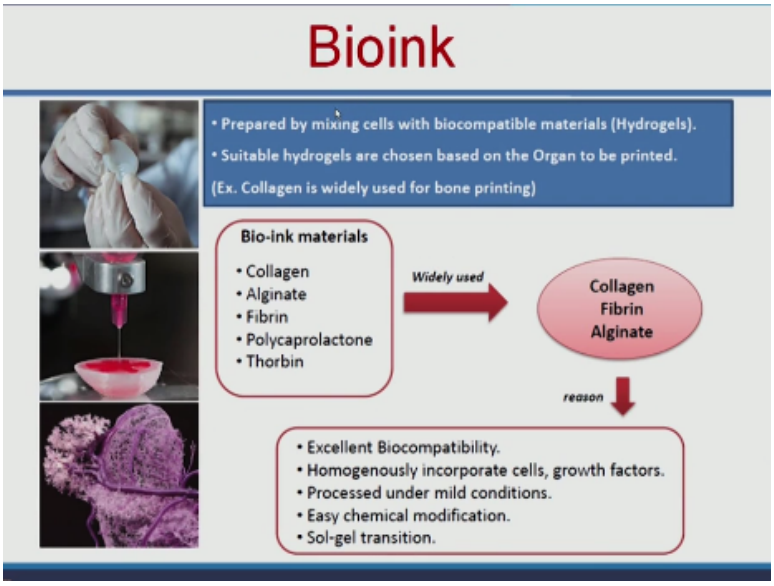

So let us see the bio ink in details, so here the bio ink as I told you earlier these are cells so these are prepared by mixing cells with bio compatible materials for example hydro gels and suitable hydro gels are selected based on the organ which you want to print and these are some of the bio ink materials we can use the collagen, alginate or fibrin so these are the widely used bio ink materials collagen, fibrin and alginate.

The reason is it has a very good bio comparability and homogenously incorporate cells and growth factors and processed under mild conditions and easy chemical modification and also sol-gel transition.

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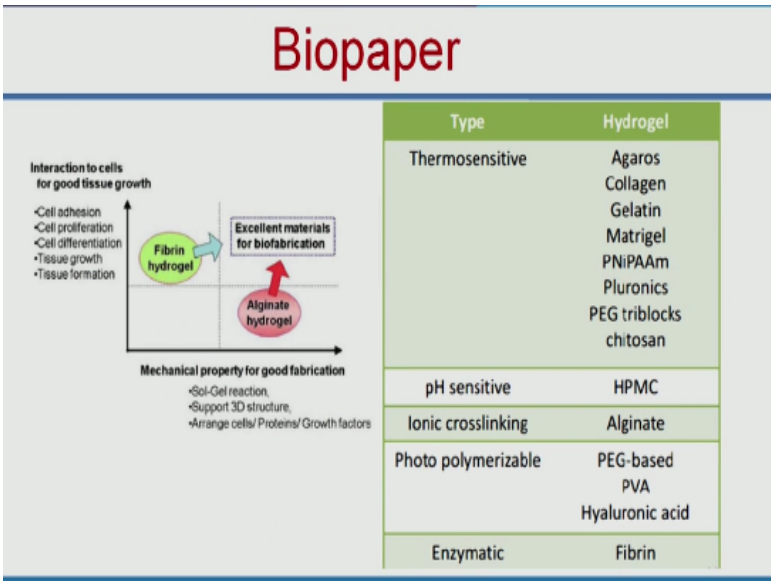

So let us see the bio paper in details so these are the various examples of bio paper we can make a thermo sensitive bio paper based on agoras, collagen and gelatin and also PH Sensitive bio paper that is on HPMC and also ionic cross linking alginate and photo polymerizable, PEG based or PVA based bio paper so out of these many bio papers fibrin hydro gel and alginate hydro gel these are very good materials for bio fabrication.

So this fibrin hydro gel have very good cell attachment property and cell purification properties and also cell differentiation and tissue growth and here in alginate hydro gel it has a very good mechanical property and it also support that 3D structure and arrange the cells and proteins and growth factors.

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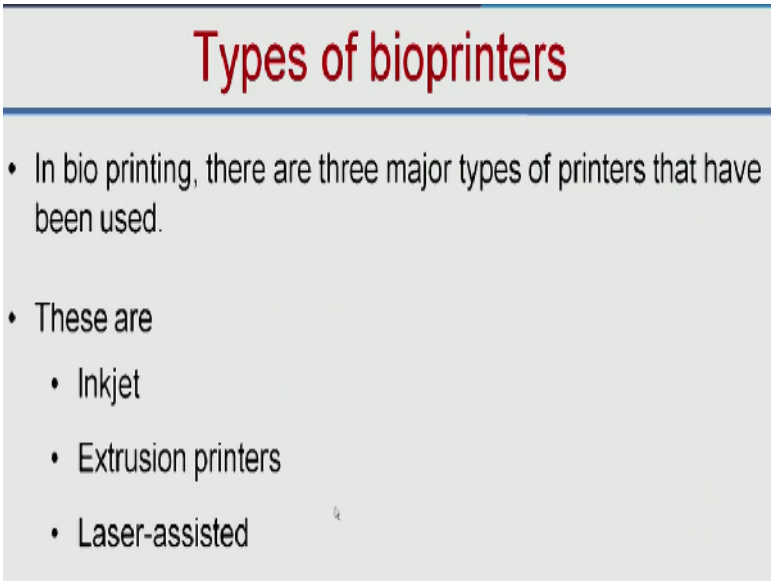

So let us see the type of bio printers, so there are again 3 types these are inkjet printers, extrusion printers and laser assisted bio printers, so let us see this in details.

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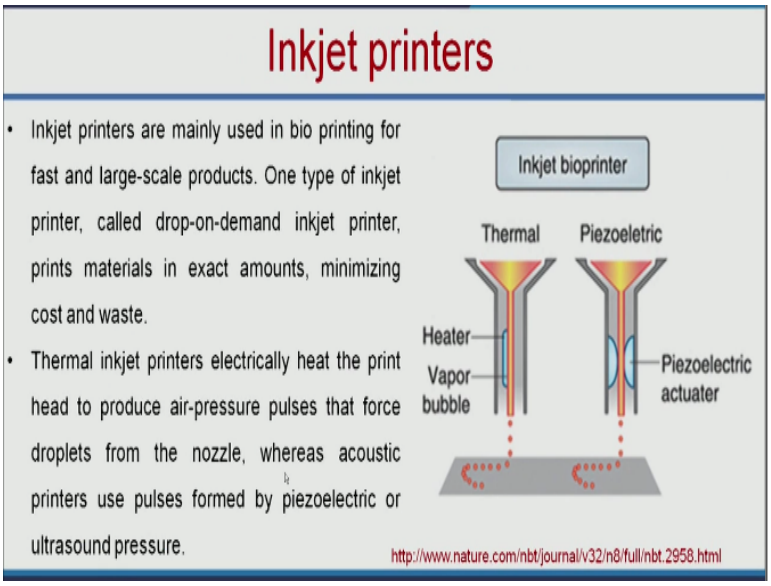

The first one is inkjet printers so these are mainly used in bio printing for fast and large scale products okay so these are type of inkjet printer called drop and demand so this printer print materials in exact amounts so it will minimize the causes with the based, so here is inkjet bio printers is divided is divided into two types thermal and piezoelectric so this thermal inkjet printers electrically heat the print head to produce air pressure pulses so that force droplets from the nozzle and here this acoustic printers uses pulses found by piezoelectric or ultrasound pressure so based on that it will deposit the cells on the particular bio paper. So let us see this next type that is extrusion printers, okay.

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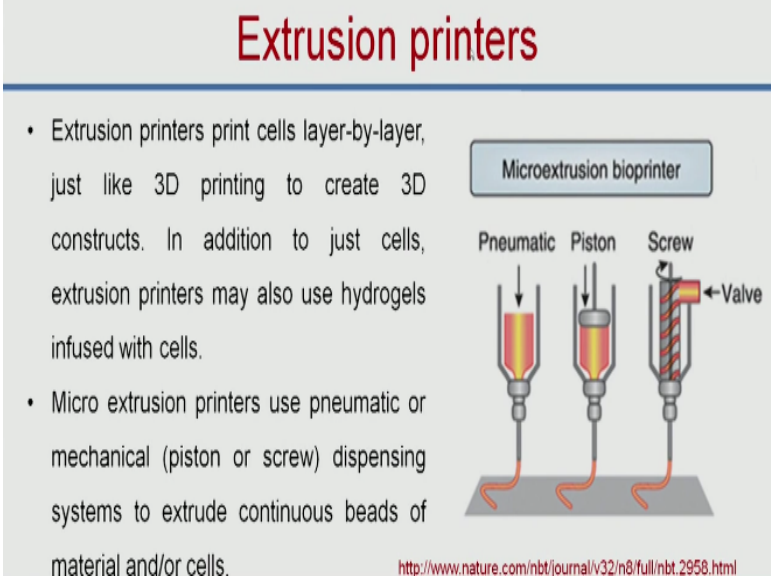

So here this extrusion printer print cells lay by layer it is also just like a 3 D printing and in addition to just cells here the extrusion printers may also use hydro gels infused with cells, okay. So here this micro extrusion printer's use pneumatic or mechanical like piston or screw dispensing system to extrude the continuous beads of the material or cells.

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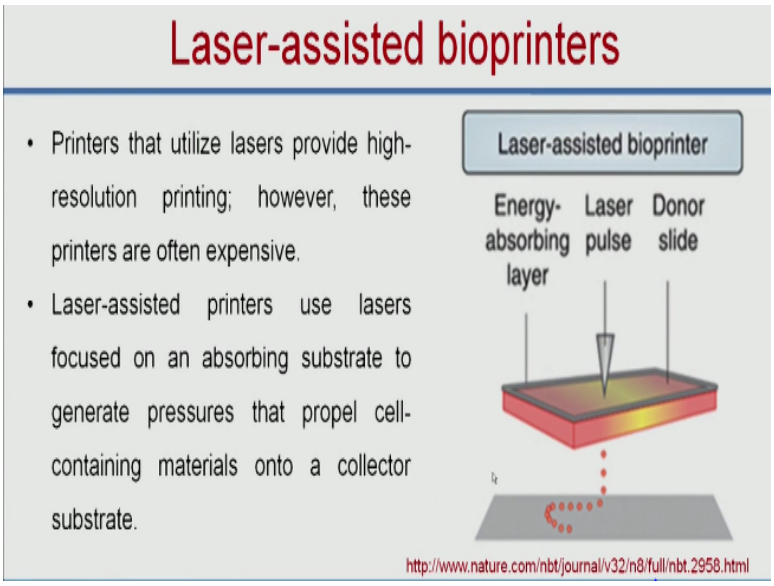

The third one is laser assisted bio printer so here the printer utilize lasers okay and the laser provides high resolution printing but this printers are little bits costly and here this lasers printers use lasers to focus on the observing substrate to generate pressure so that propose this cell containing material on to the collector substrate.

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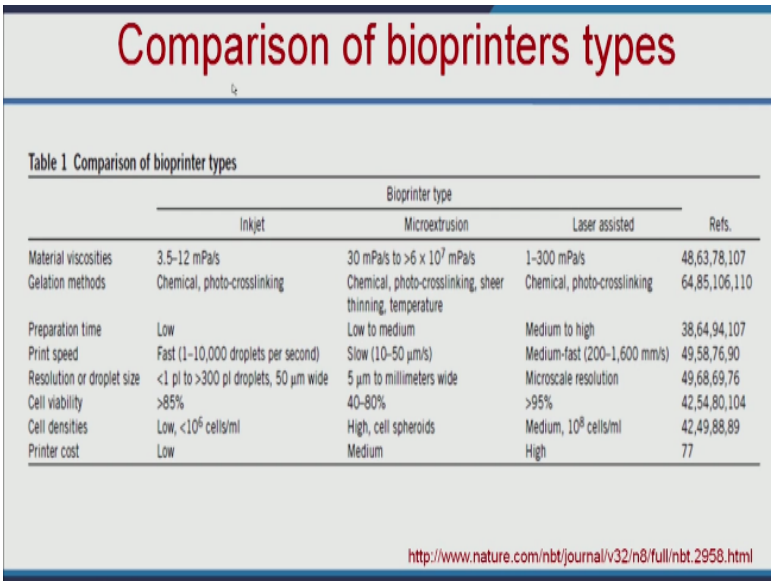

So let us see the comparison of bio printers types so depends on the organ which you want to print and also depends on the material so you have to select the suitable bio printer and each has its own advantages and disadvantage for example in case of cell viability so this laser assisted bio printer have more cell viability and in case of printing speed so the inkjet printer is very fast when compare to the other two types.

So in case of printer cast so this inkjet printer is low and this micro extrusion is medium and the laser assisted printer is very high, so you have to select the bio printer according to the type of organ which you want to print.

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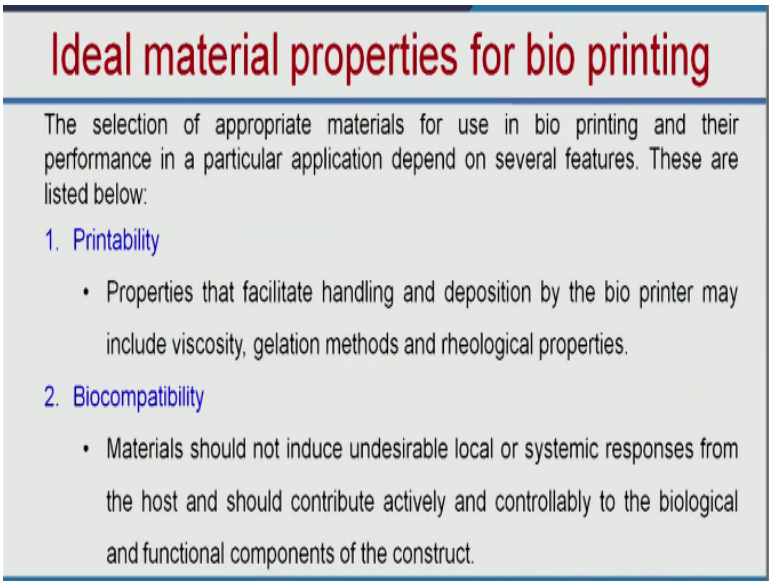

And let us see the ideal material properties for bio printing, so the selection of appropriate materials for using bio printing and they performance in a particular application depends on several futures, the first one is printability that is the properties that facilitate handling and deposition by bio printer and it may include the viscosity gelation methods and rheological properties.

The next one is bio compatibility, the printed organ should be compatible to the biological system so here the material should not induce undesirable local or systematic responses and it should also contribute actively and controllably to the biological and functional components of the construct.

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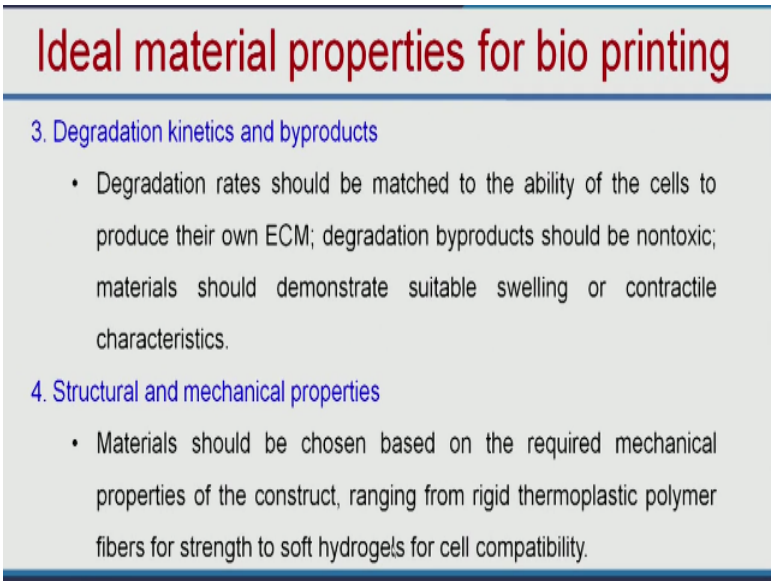

Next one is degradation kinetics and by products, so here the degradation rates should be matched to the ability of the cells to produce their own ECM, so when the scaffold degrade so before that scaffold degrade the particular cell should make its own extra cellular matrix and the degradation by products should be non toxic okay. That means it to be bio degradable, so once the scaffold is degrading it should not induce any toxic or immune response in the body.

Fourth one is structural and mechanical properties, here the material should be selected based on the required mechanical properties of the construct so it can range from rigid thermo plastic polymer, fibers for strength to soft hydro gels for cell compatibility, so depends on the organ which one going to print you have to select the particular material.

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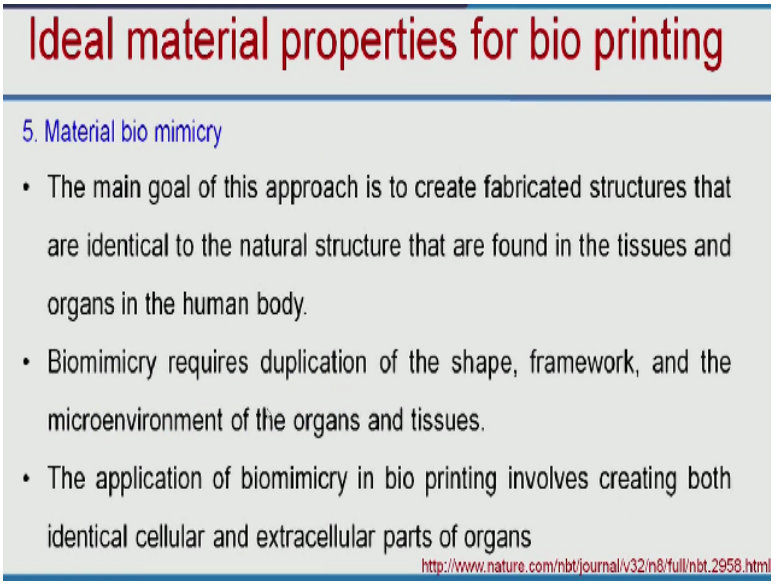

And fifth one is important that is the material bio mimicry so here the bio mimicry requires the duplication of the shape frame work and the micro environment of the particular organ and tissues and here the application of bio mimicry and bio printing involves creating both identical cellular as well as extra cellular parts of the organ, so in this bio mimicry approach we have to create the same environment, okay. So that is the important thing. So let us see the step 3 that is your post processing.

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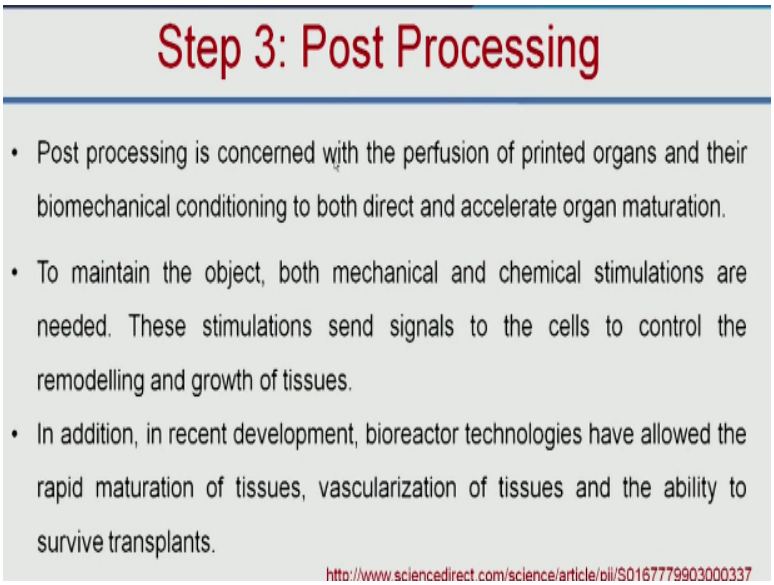

So the post processing is concerned with the perfusion of printed organs and their biomechanical conditioning to both direct and accelerate the organ maturation, so to maintain the organ both mechanical and chemical simulations are needed okay. So this stimulation sends signal to the cells to control the remodeling and growth of tissues, in addition in reason development bioreactor technology have allowed the rapid maturation of tissues, vascularization of tissues and the ability to survive the transplants.

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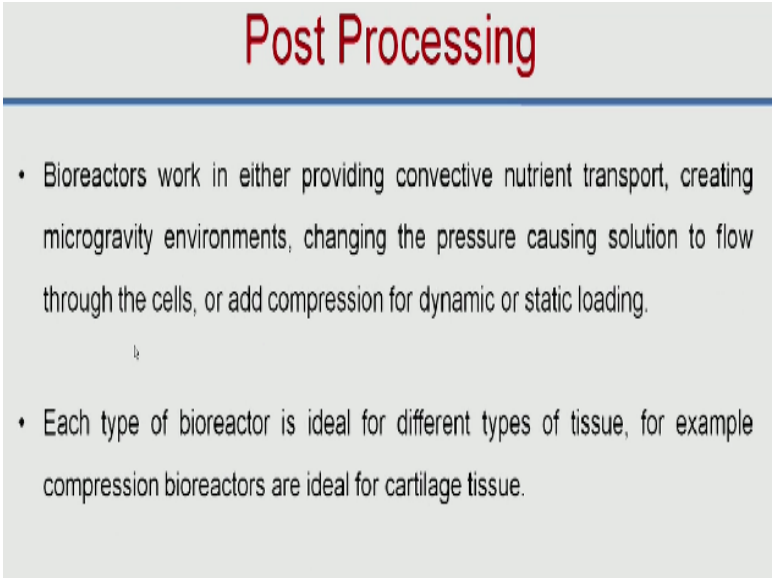

So here this bio reactors work in either providing nutrient transport or it can create the micro gravity environments, so each type of bio reactors ideal for different types of tissue for example compression bio reactors are ideal for cartilage tissue.

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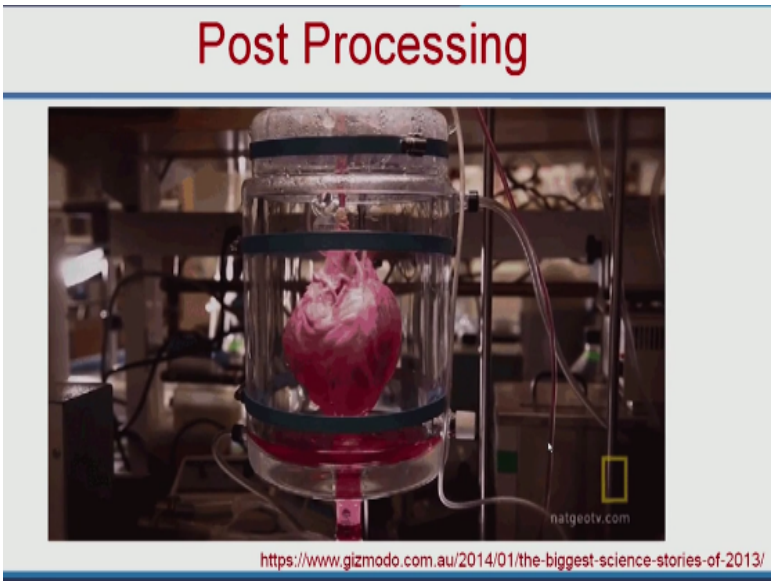

So this is your example of post processing, okay so here this artificial printed heat is growing in a bio reactor.

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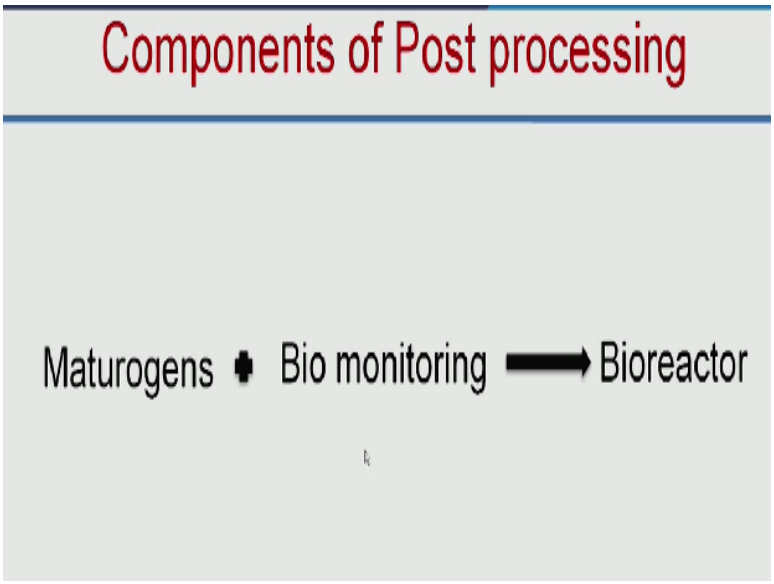

So these are the components of post processing so the first one is maturogens next one is bio monitoring okay so in the bio reactor we are going to keep the artificially printed organs so as I told you earlier this maturogens means chemical signals or mechanical signals so that will provide the real enamel to this artificial organs okay and in bio monitoring we will be monitoring the metabolism of the artificial printed organs. Whether it is performing like your original organ.

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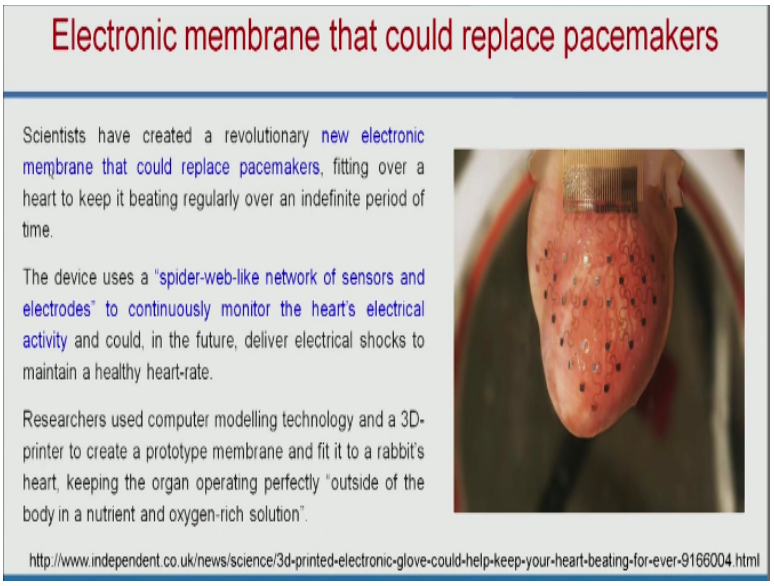

So here the scientist have created a new electronic membrane so that could replace pace makers and they made a membrane using like a spider wed like network of sensors and electrodes, so which will continuously monitor the heart's electrical activity and in future it could deliver electrical shocks to maintain a healthy heart rate so here researchers used a computer modeling technology and printed this prototype membrane.

And they fit into rabbit hearts and they check it whether it is working properly in a rabbit heart, okay. And in this rabbit heart outside the body in a nutrient and oxygen rich solution it is working perfectly like a real heart, so in future it could be useful for human application also.

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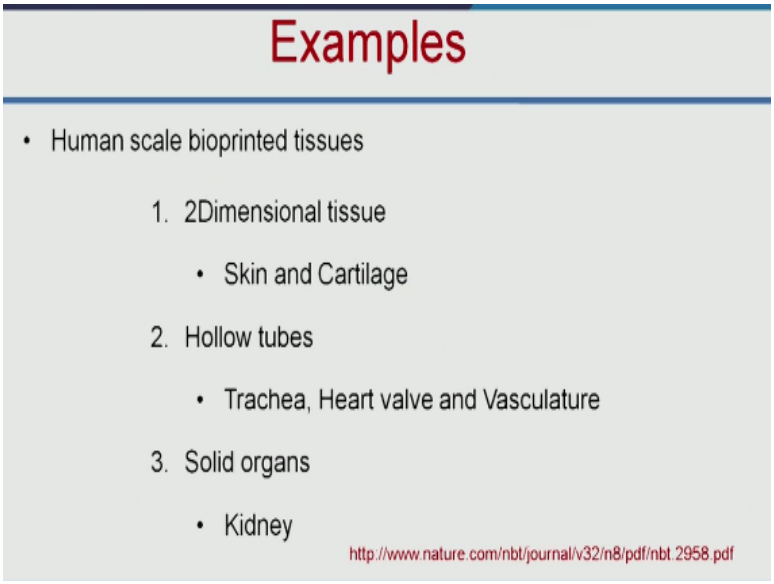

So let us see some of the other human scale bio printed tissue, the first one is two dimensional tissues.

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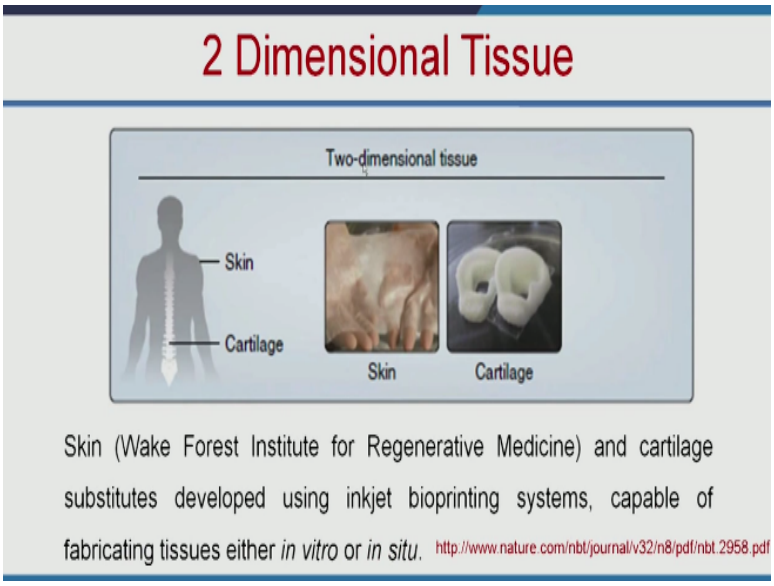

So like a skin and cartilage so this was already printed using inkjet bio printing systems okay by Wake forest institute for regenerative medicine and similarly they also printed this heart valve and trachea and using micro extrusion bio printed technique so similarly they have also printed solid organs like kidney using laser bio printing as well as micro extrusion bio printing technology.

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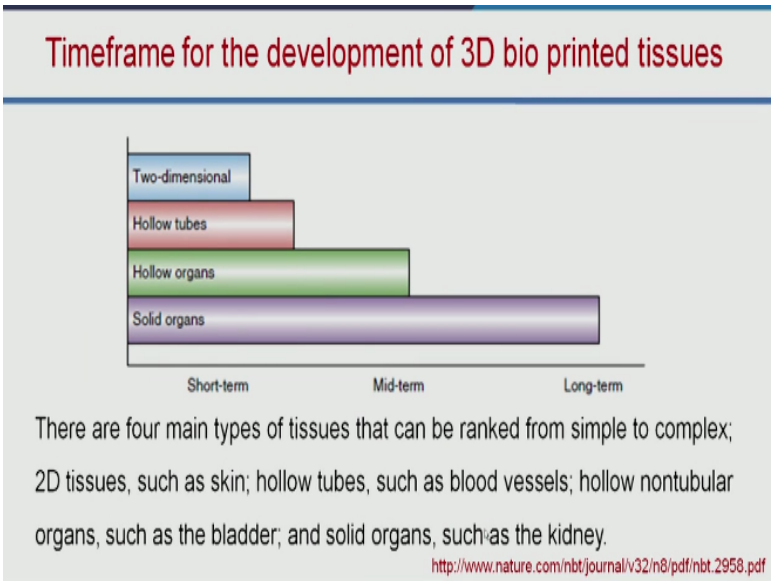

And here the time frame for the development of 3D bio printed tissues depends on the organ which you want to print, so in case of 2 dimensional it requires very les time and in case of solid organs it need more time.

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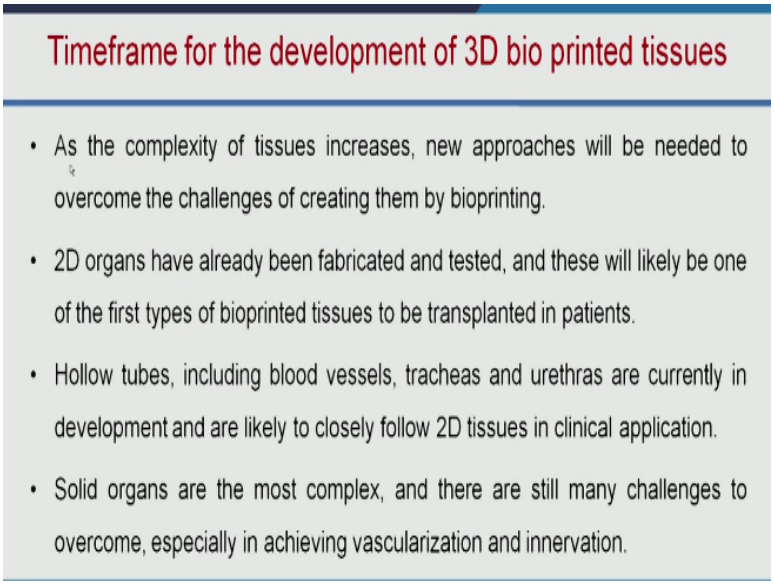

So as the complexity of the tissues increases new approaches will be needed to overcome the challenges of creating them by bio printing, so here the 2D organs have already been fabricated and tested, and these will be likely to be one of the first prototypes and which could be transplanted in the patients. And next one is hollow tubes including blood vessels, so scientist are already developed this hollow tubes and it is in the clinical trial okay. And the third one is solid organs are the most complex, and there are still many challenges to overcome, especially in achieving vasualization and innervations.

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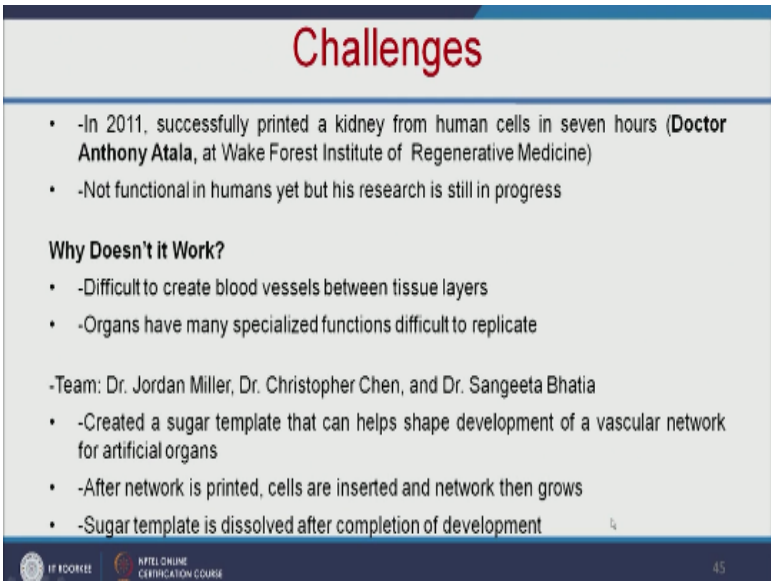

So let us see the what are the challengers? So these groups have artificial kidney in seven hours but it will not function in human. So why does not work? Because it is difficult to create blood vessels between tissue layers and organs have many specialized functions difficult to replicate. So these things came up with some new solutions to overcome these problems. So they created a sugar template that can help shape development of a vascular network for artificial organs. And after network is printed, cells are interested and network then grows okay. So here that sugar template is dissolved after completion of development.

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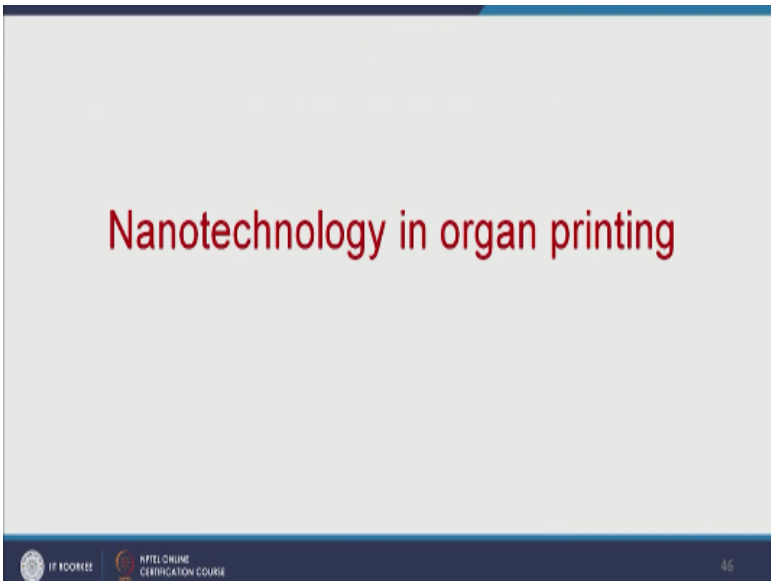

So let us see the role of nanotechnology in organ printing.

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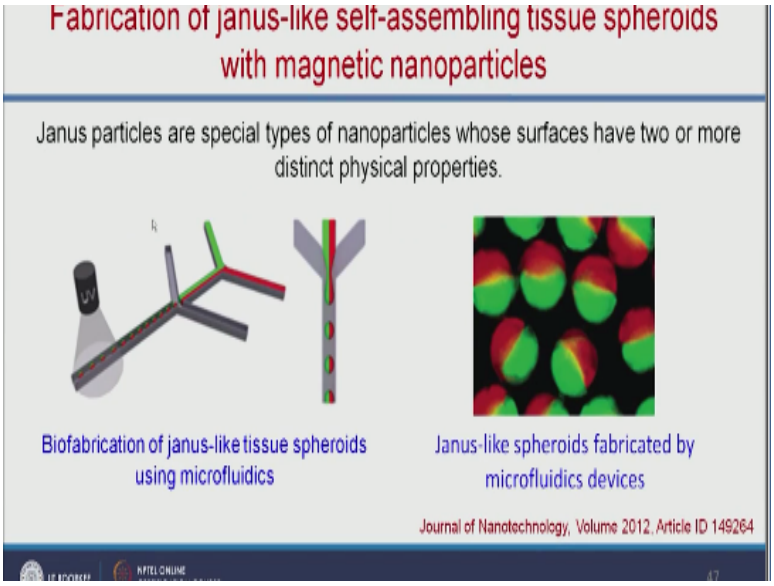

So here we can bio fabrication of the Janus like tissue spheroids using microfluidicus. So a Janus particle means these are special types of nanoparticles whose surface have two or more distinct physical properties. So in this case a Janus like spheroids so spheroids are the cancer cells and it is also having the magnetic properties.

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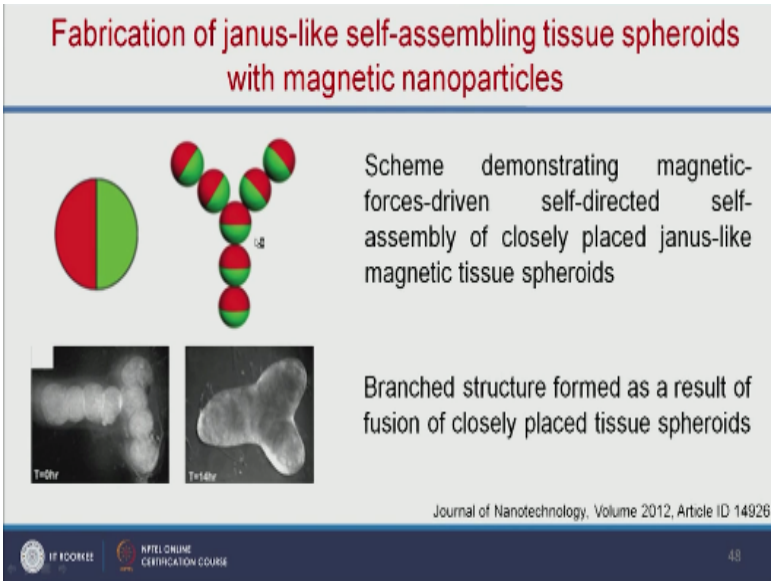

So using these magnetic properties we assemble these using the external magnetic fields. So once it should assemble in a particular shape then these cells will choose together and formed a particular organ or a tissue. So we can see here zero hours these organ tissues spheroids are assembled using magnetic field. And the fourteenth hour it will form a branched structure as the result of fusion of closely placed tissue spheroids.

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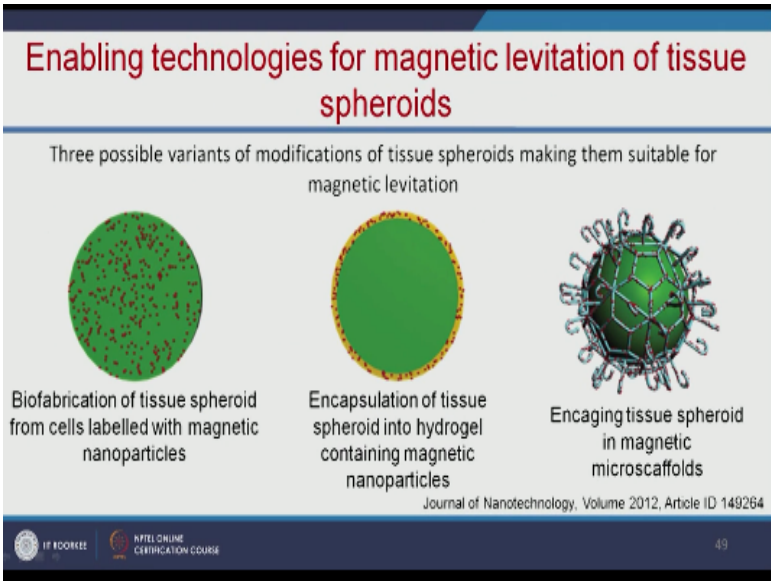

And let us see how we can make some technologies for magnetic levitation of tissue spheroids. So first approach is we can have the cells labeled with magnetic particles and a sign approach is we can have the tissue spheroid into hydrogel containing magnetic nanopatrticles. And third approach is encaging the tissue spheroid in magnetic micro scaffolds.

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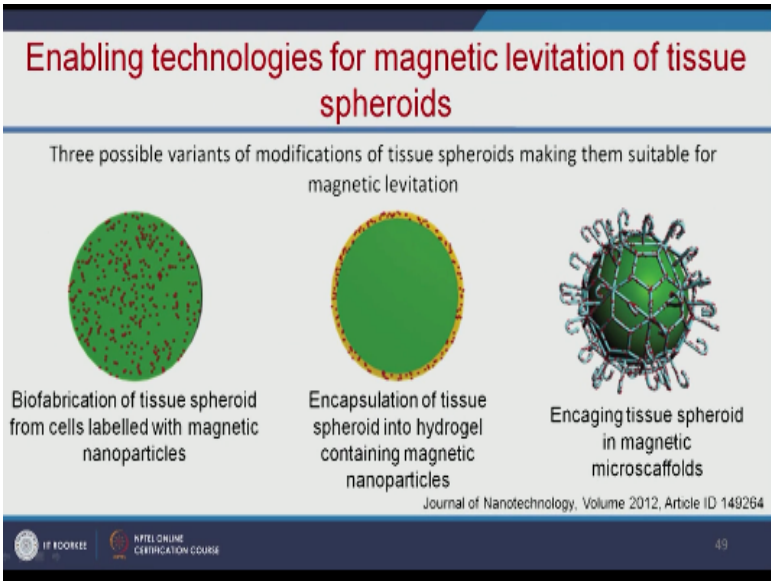

We can make this lock and hook like structures and w can make this kind of lockyballs okay. So this lockyballs are inter lockable micro scaffolds for encaging the tissue spheroid. So using this approach we can make this kind of shapes and in this we can load the tissue spheroids. We can see here this is the section of lockyballs it is having magnetic nanoparticles inside these the vectorism nano particles. And this one is lockyball with magnetic surfaces and the third one is lockyball with the functionalized nano particles on the surface.

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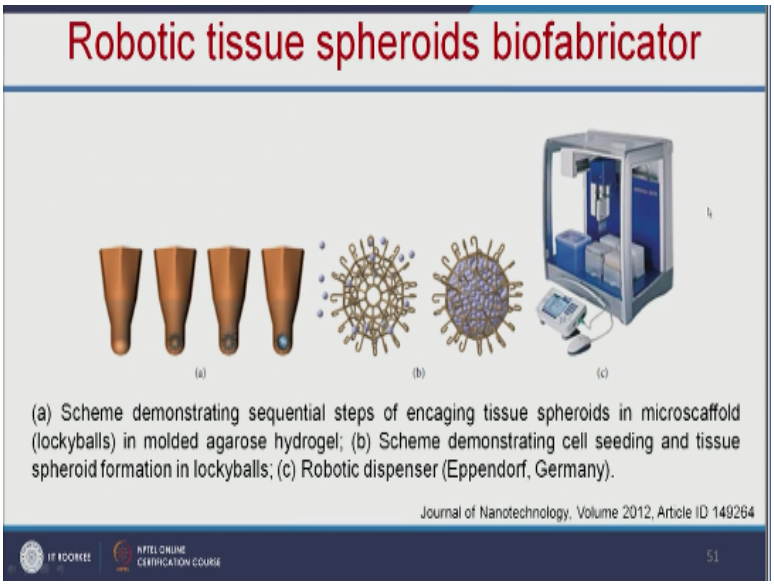

So this is the robotic tissue spheroids biofabricator okay. so using this make the micro scaffold that is the lockyballs in a molded agarose hydrogel. And in that we can see the cells and we can make the tissue spheroid formation in the lockyballs.

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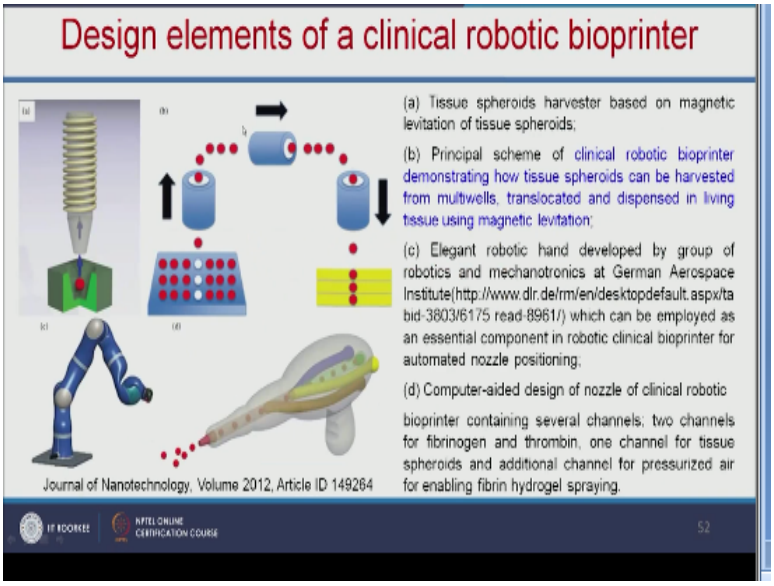

So let us see the design elements of clinical robotic bio printers so beg on this lockyballs we can make clinical robotic bio printer. So here this one is your tissue spheroid harvester. It is based on magnetic levitation of tissue spheroids. And here we can use that and clinical robotic bio printer. And this tissue spheroid are harvest from the multi bells okay. And it can be translocated and dispends into the living tissue using magnetic levitation.

So this is a computer aided design of in organ of physical robotic bio printer. So it contains several channels so two channels for fibrinogen and throbbing, and one channel for tissue spheroids okay. And additional channel for pressurized air for enabling fibrin hydrogel spraying.

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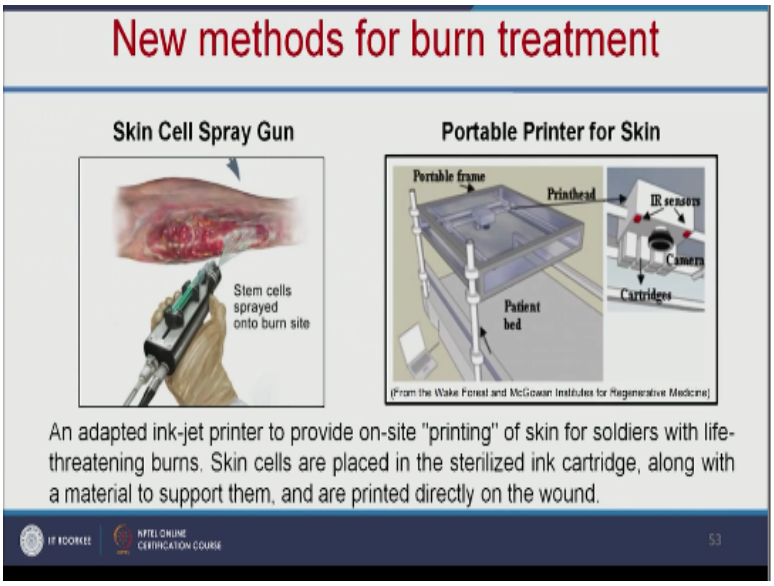

So similarly we can make the portable printer for skin okay. So it is an adapted ink-jet-printer, so provide on-site printing of skin for soldiers with life threatening burn. Skin cells are placed in the sterilized ink cartridge, along with a material to support them, and are printed directly on the wound. So here we can print the particular cells okay on the wound side directly.

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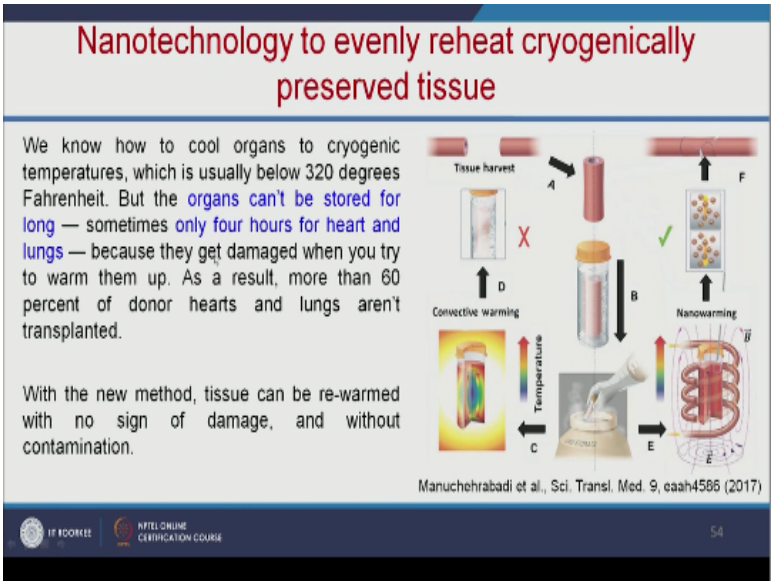

So here the nano technology not only for the printing the artificial organs it can also use to preserve the tissue. So usually the organ will be stored at 320 degrees Fahrenheit. So but the organs cannot be stored for long. So for example, we can store the heart, lungs for four hours because they get damage when you try to warm them up okay so due to this more than 60 % donor heart and lungs are not transplanted. So with this new method the tissue can be re- warmed with no sign of damage, and without any contamination.

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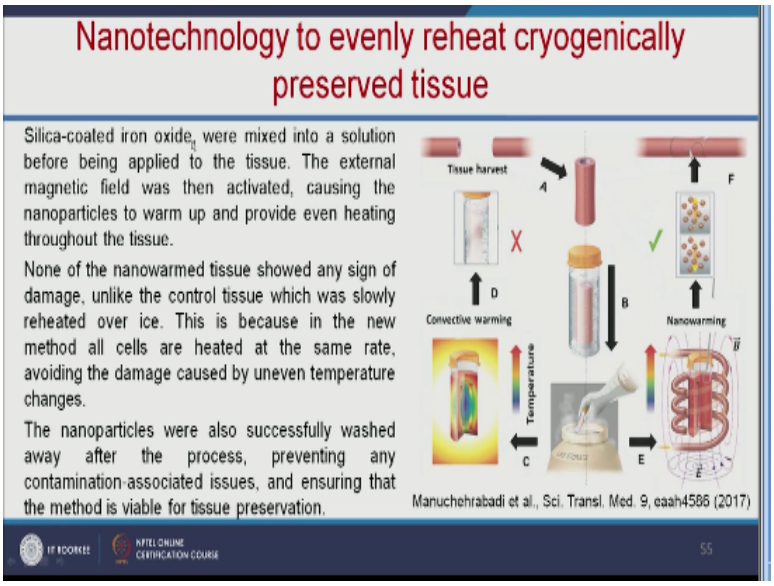

So here the silica-coated iron oxide okay, so this will be applied to the tissue and the external magnetic field was then activated, so it will causes the nanoparticles to warm up will provide even heating throughout the tissue. So here none of the nanowarmed tissue showed any sign of damage okay, unlike the control tissue which was slowly reheated over ice. And this nanoparticle could be washed away doing the process and it will prevent the any contamination associated tissues and ensuring that the method is viable for tissue preservation.

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So let us see the another approach okay, that is organ decellularization approach. So usually of printing the complete organ we can use this approach and we can decellularization the organ from the donor. So let us see this approach in detail.

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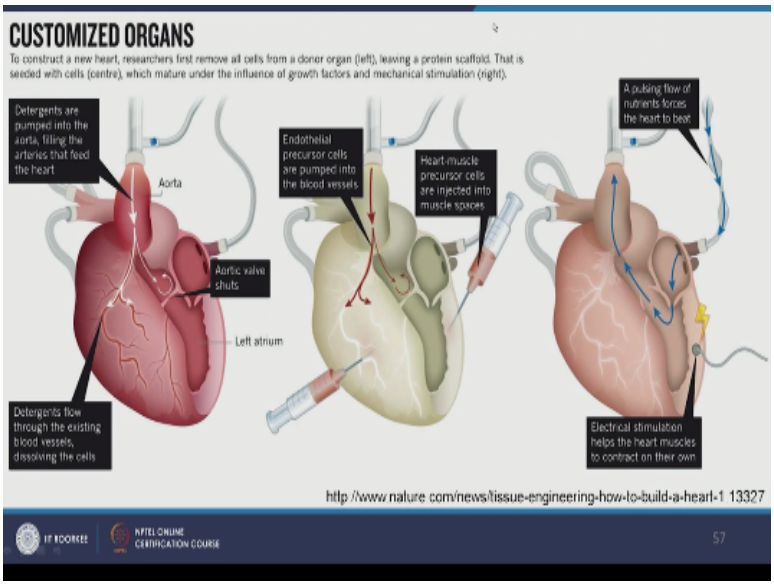

So we can have the customized approach, so the first one is for example if you want to construct a new heart. So we can remove all the cells from the donor heart and it will leave the only protein scalp hole. Then we can inject the cells and grow on the top of this heart okay. And you can give the grow factors and the mechanical simulation and we can make the customized organs.

So the first step is detergents or pumping into the aerator and filling the aerators that fed the heart okay. And these detergents flow through this blood vessels and dissolve all the cells. Then the cells are removed so it will left with only the particular scalp hole. Then we can precaution cells can be pumping to the blood vessels. And heart muscles precaution cells can injected into the muscle spaces.

So what the patient cells are gone on the scalp hole and this can be transfer to the bio reactor. So in the bio reactor a pulsing flow of nutrient will force the heart to beat and we can also give the electrical simulation and which will helps the heart muscles to contract on their own. And in the bio reactor it will work like a exact heart. And once it is monitored in the bio reactor and we can transplant to the patient.

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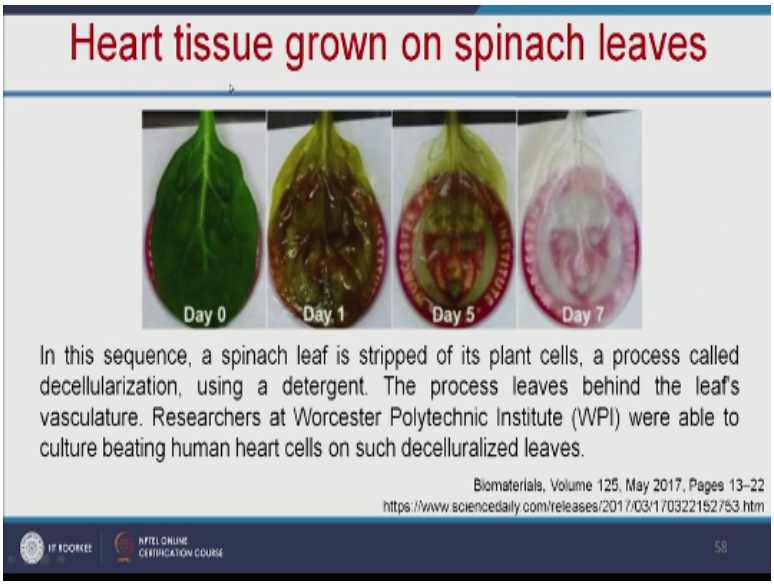

So this the reason this which their grown this heart tissue on this spinach leaf. And they also follow the same approach so here this spinach leaf is stripped of its plant cells through a process called decellularization using detergent. And this process leaves behind the leafs vasculature. And here the cells were culture on the top of these decellularize leaves. Consider at day 0 it is a spinach leaves and they removed this cells from this spinach leaf and the day 7 decellularized and its left with the only vasculature. So the scientist are used these as a scalp hole and they grow these beating human heart cells on the decelluarized leaves.

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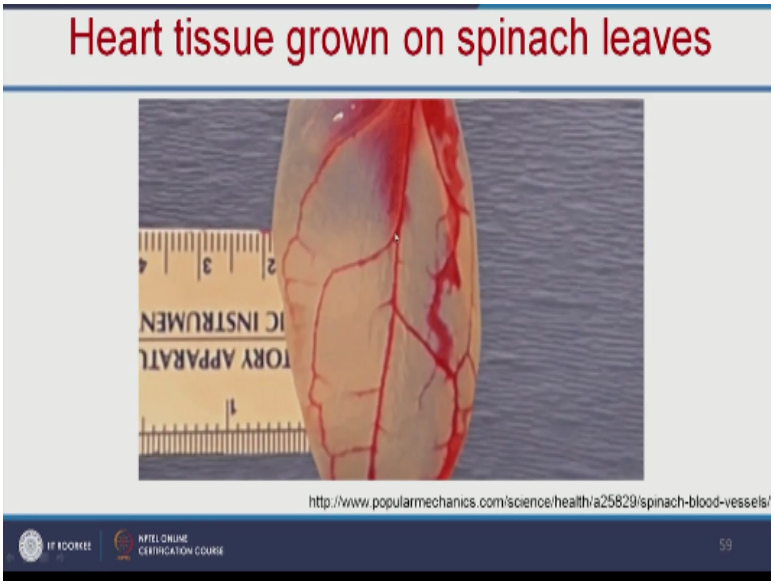

So here we can see in this animation so these frankly vasculature it is act like your blood vessels so growing the heart cells on these leaf scaffold.

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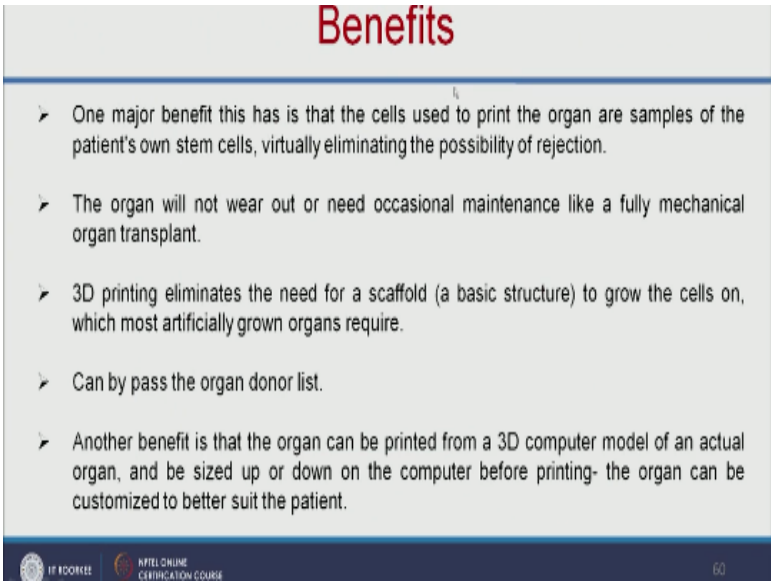

So let us see the some of this benefits of the organ printing, so one of the major benefits we are going to take the patient is own stem cells, so that will remove the possibility of hemo rejection. And also here this organ will not wear out or need any occasional maintenance like your fully mechanical organ transplant. And also these 3D printing eliminates the need for this scaffold and we can bypass the organ donor list and an another benefit is using this computed aided design we can make the customized organ which will suit the particular patient.

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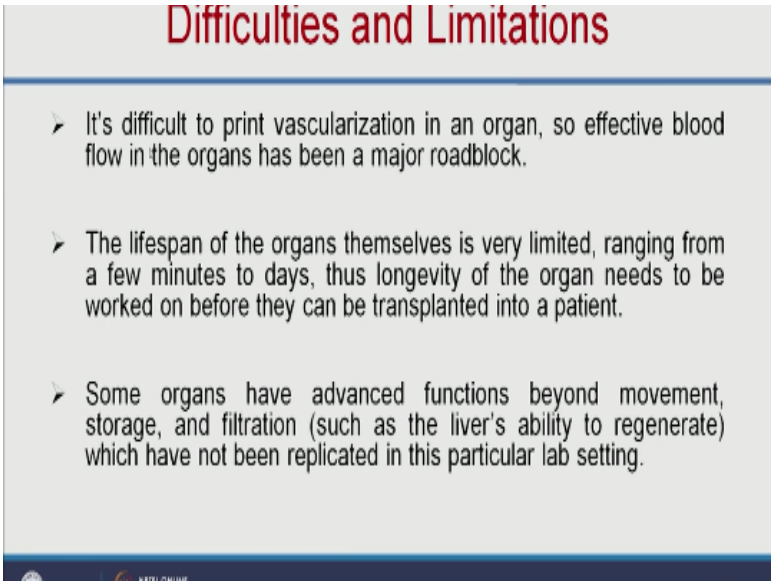

So let us see some of the difficulties and limitation in this organ technology. So the main thing is it is difficult to print vascularization in an organ, so effective blood flow in the organ has been a major roadblock. And again the lifespan of the organs is very limited. So it can range from a minutes to days. So thus the longevity of the organ needs to be worked on before they can be transplanted into a patient. And some organs have advanced functions beyond movement, storage and filtration. For example the liver is ability to regenerate. So which have not been replicated in this lab sitting organ?

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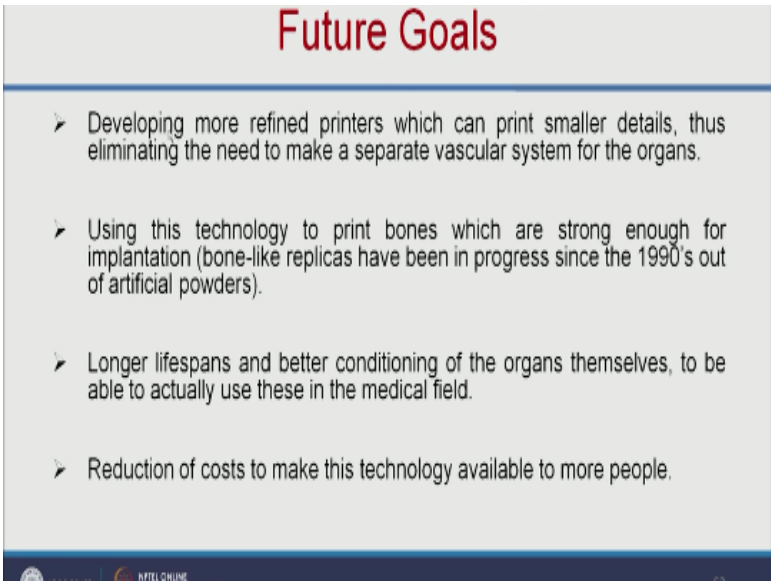

So these are the future goals, the first one is developing more refined printers which so can print smaller details. So these eliminating the need to make a separate vascular system for the organs. And also it can increasing the lifespan of the artificial printed organs okay. And also it can release costs of this technology it can made available to more people.

So as summary of this lecture, so in this lecture we have learnt what is organ printing? And what are the types of 3D printing and we also learnt various 3D bio printing approaches okay. And also we learnt what organ decellularation is and also learnt the nano technology organ printing. So I will end my lecture here I thank you all for listen this lecture. And I will see you in the next lecture.

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