Advances in Additive Manufacturing of Materials: Current status and emerging opportunities

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Lec 30 Introduction to biological system

this is the last part of my series of lectures on introduction to biological system and materials for human healthcare. Towards the end of last lecture, I have mentioned very briefly what are the different parts of a human body where synthetic biomaterial implants can be used to reconstruct, repair or regenerate damaged tissues. So having said that these biomaterials the classic definition. or textbook type of definition is that any substance that has been engineered to take a form which alone or as a part of a complex system is used to direct by control of interactions with the components of living system in the course of any therapeutic or diagnostic procedure in human or veterinary medications. we are essentially saying that these synthetic materials must have meaningful and clinically relevant interactions with the components of living system. Now this components of living system essentially means cells, protein, blood, tissue and so on.

if a material is to be used for bone related applications then you have to Consider the interactions of that materials or scaffolds with the bone cells. And if the material is to regenerate the peripheral nervous tissue or very various nervous tissue, then you have to see that whether the materials can support the neuronal functionality or biomaterials. the way this total biomaterials field has evolved over the last several decades has been shown very schematically here. what you see first generation biomaterials which are essentially bio inert materials like stainless steel materials, titanium alloys and so on.

these are what they call off the shelf materials. of the shelf materials means like these are the biomaterials are already available and this class of materials were already used for other non-biomedical applications for decades and they have shown their performance or they are time tested materials. but for different non-biomedical applications and those materials people started using for biomedical applications. this is called first generation biomaterials. Second generation biomaterials particularly the bioactive materials which has the ability to form chemical bonds with living tissues.

this ability was not there in the first generation biomaterials, so it has very minimal foreign body reactions. Foreign body reactions means whenever you have biomaterials placed in a human body so there is a tendency for the natural system to reject any artificial material and this artificial material is essentially biomaterials

natural material means that organism full organism like you know for example our human body or for example any animal model like rat model mouse model and so on. any synthetic material which was originally not part of the system organism and this has been placed inside that organism. there is a natural phenomena or natural tendency that material will not be accepted it will be rejected by the system. then slowly as the Biomaterial science has progressed, people have developed their understanding that how synthetic materials are accepted or rejected and what are the reasoning underlying mechanisms of this acceptance or rejection by the living system, then they have developed the second generation biomaterials like bioactive materials like which forms chemical bonds with living tissue.

Then you have the bioresorbable materials that is undergo control degradation and new tissue generation. Then third generation biomaterials is called functional tissue engineered constructs. Now what is tissue engineering? I think I have mentioned very briefly before and I will mention again in one or two subsequent slides on the tissue engineering. tissue engineering essentially is again an example of the interdisciplinary field of science and engineering which uses three components. One is the cells.

Second one is your materials or scaffolds and third one is a bioactive molecules or biomacromolecules like you know growth factors and different peptides and proteins and so on that can simulate. tissue engineering actually allowed people to stimulate cellular response at the molecular level which was not possible using that first generation material. this was not possible. second generation is a bioactive glasses like Larry Hench who was who pioneered this field of bioactive glasses and third one is a tissue engineering like which was founded by some of the stalwarts in the field like Professor Robert Langer at MIT. Now, what is the core of the science that you know that how synthetic biomaterials they interact with the biological system component so this cartoon essentially shows this is a eukaryotic cell the way I am essentially sketching it is cell nucleus which contains genetic materials like DNA which has been mentioned number of times in last few lectures.

Now this is the cytoskeleton or actin filaments in the cytoskeleton. Cytoskeleton has actin filaments, it has microtubules and it has intermediate filaments. all the three actin filaments, microtubules and intermediate filaments, all the three essentially constitute the cytoskeleton when an eukaryotic cell interacts with the biomaterial surface and what has been shown here, it is a single eukaryotic cell and this is your biomaterial substrate, that is the synthetic materials. which is a non-living in nature. you if you recall earlier lectures I said that proteins absorb the biomaterial surface and these surface absorb proteins essentially establishes the interactions with the integrins.

You remember the integrins which are present on the cell surfaces. these individual red dots are assemblies of adsorbed proteins and integrins or cell surface receptors. each of these red dots essentially constitute N number

of these adsorbed proteins and cell surface receptors and in biological language, they are called focal adhesion complexes. FACs focal adhesion complexes. This focal adhesion complexes are the sticking points of any biological cell on a biomaterial substrate right and the more the number of focal adhesion complexes more would be the stability and more would be the adhesion more would be the extent of adhesion on the cells on this biomaterial substrate.

This is point number 1 for you to follow. Second point is that A biological cell initially was spherical in nature This is a biological cell, classic description of a biological cell which is a nucleus at the center. And you have this strong kind of a structures you can see. These are like integrin proteins or transmembrane proteins or cell surface receptors. And this protein adsorption has been shown by different kind of geometrical structures like square, triangle, circle, rectangle and so on which are already absorbed by the biomaterial substrate.

Now these kind of assemblies essentially constitute focal adhesion complexes. A biological cell which is spherical in nature and as you know that the stability of a sphere on a infinitely flat surface is not good unless that elastically compliant sphere will undergo some kind of shape changes. to flatten the base to have more physical interactions with that biomaterial substrate that cell would not have stable adhesion or stable interaction with that biomaterial substrate. I hope this point is very clear to you. This particular process that you know that shape morphology changes to establish or to facilitate better interaction with biological system that is possible only when this cell undergoes cytoskeletal reorganization process.

This cytoskeletal reorganization process means actin filaments must be stretched. and if it is stretched then cell also would go from more spherical shape to an oblate sphere shape And then more this extension process the more the cell surface is flattened that means more opportunity for the integrins and cell surface receptors to establish more contacts with biometrial substrate and more stability is the focal adhesion complexes more would be the cell adhesion on a biometrial substrate. at this point I would like to introduce you to implant and scaffolds. implant essentially is a 3 dimensional non-porous biomaterial component to reconstruct or repair damaged tissue. by definition it is non-porous that means it should be mechanically more strong.

It is a biomaterial so that means it should have a proven biocompatibility. What was the purpose of the implant in human body to reconstruct or repair damaged tissues like for example if certain hip bone is damaged. you know you need some component of the hip bone to be replaced then you would use the implant. This can be better substantiated by this particular example of the total hip arthroplasty and this total hip arthroplasty what happens if you see this video very carefully when the acetabulum is damaged or hip or a patient undergoes hip fracture. First thing is to put that acetabulum that means acetabular liner with that of the hip acetabular shell, then comes femoral head, so this is the femoral head part , this femoral head parts now it is interacting and then

you can see that slowly this acetabulum component first repaired by placing the acetabulum in the hip, then the femoral stem is inserted And this femoral stem is directly attached to this femoral ball head and then they interact and then they form this total hip replacement device.

there are two ways you can do, one is the cemented total hip arthroplasty and one is called uncemented total hip arthroplasty. In the cemented case, you need to use the polymethyl methacrylate bone cement. In case of uncemented you need to use this is the acetabular shell and this shell is made up of the titanium alloys or often it is a lattice structure that is been imprinted on the titanium shell outer surface for better osseointegration or this particular shell is hydroxyapatite coated. in this kind of the uncemented total hip arthoplasty here the example of the implants are essentially femoral stem which is made up of the titanium Ti6Al4V or stainless steel 316, femoral head can be cobalt chrome or can be ceramic like zirconia, toughened alumina and acetabular liner is the ultra moderate polyethylene. None of these materials exist in the human body in any form.

therefore when they are manufactured outside the human body, they are placed it inside the human body, they have a specific three dimensional shape, they are non-porous, they are dense, mechanically strong and that can serve this particular function and that is why they are called as an implant. then comes to these examples of the scaffolds. scaffolds are essentially, implants I have already covered. I am essentially describing the scaffold now. It is 3-dimensional, porous and often it is interconnected porosity, porous structure to regenerate tissues.

It can be soft tissues like urological tissues for example, it can also be used for small bone defect structures like it can also be for bone tissues which is the elastically much stiffer than that of the urological tissues now if you look at this two end of this top part of this particular slide, one side you have an implant and one side of a scaffold. One is a 3D non-porous structure implant, one is a 3D porous structure. if you ask these questions to your mind which will have better mechanical properties, when the same composition materials exist as scaffold as an implant and then same shape or size certain implant will have a better mechanical property because it is non-porous that is number 1. Now if you ask this question to yourself which particular structure or either implant or scaffold will have better biocompatibility? Scaffold will have better biocompatibility because it has a three-dimensional porous structure. It will allow cells to grow into the porous structure.

It will allow the blood and nutrient to go inside this porous structure so that cells survive, so that tissue formation is facilitated, so that tissue maturation also to take place into the 3D porous scaffolds and that was the reason these 3D porous scaffolds have attracted lot of attention right in the scientific community. And this has lead to the birth of tissue engineering which essentially requires you know scaffolds structure In a nutshell, what is the difference between implant and scaffold? An implant is synthetic biomaterial which restores the physiological function of damaged or injured tissues. A scaffold is a 3D biomaterial platform with

interconnected porosity which supports cell functionality and aids in tissue in growth. An implant provides biomechanical support. A scaffold is non-permanent and degradable in biological system.

And depending on the porous architecture, a scaffold has a weaker mechanical properties that I have already mentioned. Now, collagen. Now, why collagen is important? As I mentioned that collagen is actually organic component of the natural bone. bone has organic component and inorganic component. Inorganic component is hydroxyapatite or calcium phosphates.

and collagen is the organic component. therefore, when people started developing the scaffolds automatically their attention is more focused on collagen based scaffolds because collagen is something already present in the human organism therefore they started developing collagen but using collagen actually is kind of very very expensive because collagen cost much much more than gelatin. essentially if you take any animal tissue you extract and purify you form a collagen. But when you heat treat the materials within a narrow temperature window you get called a gelatin. gelatin is the denatured part of the collagen.

I use the denatured. this is the collagen triple helical structure. DNA has a double helical structure, collagen has a triple helical structure and then if you denature it, then it forms this kind of chain structure, then you have gelatin and gelatin you further do enzymic treatment they call collagen peptide that is also used for many biomedical applications. scaffolds are primarily made up of the hydrogel and hydrogel is an example of the self assembled biomaterials. and contains 3D hydrophilic polymeric network and characterized by presence of chemical and physical cross-linking and which can also act as a potential water reservoir. hydrogels by definition must have the ability to contain large water molecule, large amount of water molecules.

And this actually is made up of the 3D hydrophilic polymeric networks and this 3D hydrophilic polymeric networks must have the ability to be crosslinked by the physical for example, UV exposure or chemical crosslinking like for example, calcium chloride for example, if you add to the solution. Gelatin is one of the most widely used hydrogels for tissue engineering or biofabrication. This is what gelatin and these are collagen peptides you can see that amino acids is identical 18 amino acids and this is source from either skin, bone or connective tissue, how it is processed and so on that has been mentioned here. tissue engineering in particularly we need to have scaffold and then we need to have a target specific cells. For example, from nervous tissue you need to have neurons for example, for musculoskeletal you need osteoblast cells to be seeded on this particular scaffold.

This is the traditional tissue engineering approach. this is just to show in vitro models. in vitro means that is the experiments which are conducted using glassware and this resembles the smallest functional unit of in vitro.

You can do that developmental study, homeostasis, regeneration ,disease.

it is just to show co-culture. Co-culture essentially means 2 different type of cells are being cultured simultaneously in the same culture platform. to resemble the functionality of a tissue. Biofabrication, as I mentioned before biofabrication is a computer controlled layer by layer manufacturing of complex living and non-living biological products means you can use cells And you can use scaffold materials for example, gelatin or some of the other polymers like polylactic acid or polylactic polyglycolic acids which are non-living in nature. biomacromolecules, extracellular matrices and biomaterials as for the design of this particular scaffold. this is what biological biofabrication is.

And this is the last slide in this lecture just to show you remember the definition of the co-culture, they say culturing 2 different type of cells and these are bone 3D bioprinting, essentially you print the scaffolds. You culture osteoblast cells, you culture osteocytes for example and then both the cells if you culture together or if you culture chondrocytes for example, so that if it is used for osteochondral tissue, osteon is born. Chondral means cartilage, bone cartilage tissue regeneration, then you can culture it, then you can grow it, you can mature it and then that will that can be used to treat the osteochondral defects. This is just another example of the bone. For example, bone is an extremely dynamic tissue and it undergoes continuous remodeling during its whole lifetime.

that means osteoblast cells, this is the more matured form, this is maturation to osteocytes. osteocytes are the mature bone cells, osteoblasts are bone forming cells, osteoclasts are bone resorption cells. These are bone resorption cells. this is the osteocyte cells and this is the osteoblast cells. bone is made up of cells, nerve and blood tissue and you can see this is that long bone structures and if you go to the more typical structure of the bone then you have different canals and spongy bones and so on.

this kind of bone structures are also can be mimicked by 3D printed bio printed hydrogel structure. So this brings me to the end of this particular series of lectures of the introduction to biological system and with this series of lectures I am certain that you have now acquired the necessary or you have got to know or you have developed necessary understanding of the biological system for you to realize the bio fabrication and 3D printing of the 3D and 4D printing of the structures to mimic different biological tissues. Thank you