

Regeneration Biology
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W11L52_Importance of scaffolds in tissue engineering

Hello, everyone. Welcome back to another class on regenerative biology. In today's class, we will learn about the importance of scaffolding. We have seen different types of scaffolding. We have seen decellularized scaffolds, ECM, and so on. And we will try to understand how it is important, and especially having said that decellularized ECM is best many times in various biological or clinical scenarios, we have to use artificial scaffolds because they are the ones that are easily available; they are the ones that you can get in however large a quantity or to however many people, etc.

So we cannot rule out the advantages of artificial scaffolds, and we will learn what the importance of scaffolds is and how. They are defined, etc. So here is an outline: what are biomaterials, some questions we can ask, what is the need to know them, why we need to know them, the scope of biomaterials and scaffolds, the types of scaffolds used in tissue engineering, and the desirable properties of scaffolds for tissue engineering. What are the desirable qualities that a scaffold should have? And what advances in biomaterials and scaffold technology have we achieved? And what are some of the challenges that we face with this scaffold? So these are all things that we try to address.

A few books are listed here as suggested reading for understanding more about these scaffolds, the biomaterials, and tissue engineering applications: "A Review of the Past and Future Trends," "Biomaterials: An Introduction," and "Biomaterials and Tissue Engineering." The authors' names are listed here, and there are many more books available to read if you want to. Gain in-depth knowledge about the subject. You should read as many textbooks or scientific articles and review papers as possible. You should read it so that you will know adequate information about the subject.

You can obtain it through these approaches. So scaffold the concepts and the functions. By definition, it is a synthetic support material used to replace a part of a living system or a function in contact, or it can do its job by staying in contact with the living tissue. Say, for example, if you have a cardiac valve issue, the ideal thing is that you should have the exact muscular cardiac valve; many times it's not possible, so you can artificially put in a cardiac valve made of non-biological material, such as plastic or other polymers, etc., but it functions in conjunction with the heart.

The heart rejects it, or your immune system rejects it, or something. That biomaterial is so friendly that it will not create any trouble whatsoever, and it does the job just like any other normal cardiac valve. So in that scenario, you don't need the real heart valves. Because even if you could not put in a biological heart, the heart function is just the same; so in that situation, many biological materials can perform the duty without any drawbacks whatsoever. This is what we should keep in mind: it is not that if you have to use biomaterials or a regenerative approach or a fixing approach, they have to come solely from living tissue.

There are other scenarios; for example, if someone needs a... What you call a support for their bone, the center portion can be a polymer or an iron rod; that is okay as long as it doesn't give any trouble, as long as it doesn't affect that person's motility, movement, or functioning. It is okay; it is not a bad thing to have.

We cannot insist, "No, I want a bone itself inside, which should have bone marrow also, and it should produce blood inside that bone marrow." One doesn't need to. Unless it is going to be life-threatening. So we should also get this idea while learning about tissue engineering and biomaterials. So materials used for biomedical applications often fall under this biomaterial category.

And there are scaffolds and various 3D platforms used for tissue engineering, and they are used clinically or experimentally in implantable electronic devices, drug delivery systems, hybrid artificial organs, bone substitutes, ligaments, and tendon replacements, etc., so biomaterials can function in so many other parts that are outside what is mentioned here. Some of the major ones are mentioned here. The roles of engineered scaffolds include how they contribute and function; they allow cell attachment and migration because they should be friendly to the cells, enabling them to attach, migrate, deliver, and retain cells and biochemical factors, as well as facilitating the diffusion of vital cell nutrients.

These products express the minimum required qualities of a scaffold and exert certain mechanical and biological influences to modify the behavior of the cell, whether it is in a reprogrammed phase or a proliferating phase. Or in a differentiated phase, etc., it should support based on your demand. If you push a cell into a differentiation phase, the scaffold should support it; it should not stop it. So, these are all the ideas one should keep in mind when you are engineering a scaffold.

Now let us see some examples of scaffold applications, or some we have already seen. A long list is mentioned here: orthopedic tissue constructs and grafts. Neural tissue

regeneration scaffolds are used, especially for spinal cord injury; skeletal muscle regeneration scaffolds are used for joint replacements, hip replacement, or knee replacement, etc. Bone plates, especially for the patella bone or where you have lost a larger portion of the bone in an accident or some other situation, can replace joints or bone plates, etc.

and bone cement. Biomaterials are used to cement the bones, hip joint, artificial ligaments and tendons, dental implants for tooth fixation, blood vessel prostheses, such as blood capillaries made out of biomaterials, heart valves, skin repair devices, cochlear replacements, and contact lenses. Many of you would have used contact lenses, which are a good example of biomaterials because they work along with your cornea, which has, you know, refractive correction. And if you fix it, you don't need to use your glasses for reading or vision purposes, so it's a very good example of biomaterials. What I mean is the majority of the people who use glasses may have come across the applications of contact lenses. What are the desirable scaffold properties? Having said that, scaffolds are important and needed, etc.

What properties should we have? We can start from anywhere. Let us start from here. Biodegradability. Biodegradable means that once based on the location of delivery, the scaffold should get degraded after it is put in. Some good examples you can see are absorbable sutures.

You would have heard about it. Like if a C-section delivery happens, then the doctor will make a cut in the belly and also in the uterus. So then only they can access the baby; once the baby is accessed, they will put a suture inside. Also, that will be an absorbable switch, but what you put in the belly need not be an absorbable switch because it will be a removable switch: a ligature absorbable suture and non-absorbable suture ligature can be used. These days, people also use glues inside, so understand.

Whether you use glue for ligation or absorbable suture cord, they all have to be biodegradable. You don't want that glue or a suture ligature staying there for the rest of your life. That can create a lot of unwanted trouble. So biodegradability means it should stay for, say, one week or so; then it should disappear by the time the wound is healed. Then why do you need ligature present there? And then porosity; it should allow the migration of nutrients, oxygen, etc.

, so it should be porous. Porosity means it's like a sponge; you know a sponge has got lots of holes. Mechanical requirements mean it should be strong and tough, etc. You don't want a ligature that breaks when you are suturing. It is very good material: absorbable, biodegradable, but it is breaking. So you want to have good mechanical requirements,

moldability, and sterilization.

You should be able to make it into any shape and mold. Whatever shape you want for this biomaterial, it should be able to be amenable to sterilization, whether you sterilize by autoclaving or UV radiation. If it deforms, then it's not a good biomaterial for cell adhesion, proliferation, and differentiation. It should be supported. Cells should feel friendly towards this biomaterial so that it doesn't elicit an unwanted immune reaction or make the cells feel uneasy, unhappy, etc.

And transport requirements. It should allow the nutrition to pass very beautifully through this biomaterial and biofactor delivery, enabling you to deliver any growth factor or any other favorable growth-favoring molecules into the cells through this biomaterial and surface chemistry; the surface chemistry should be such that the surface should. Feel okay to that of a typical extracellular matrix, or the cells should be able to interact with this biomaterial as well as what a cell will do to its immediate extracellular matrix and biocompatibility. It should happen that it is compatible with me but not with my friends. It should not be that person-specific or something. It should be compatible with every tissue and every organ; then only will it become a good and beautiful biomaterial that can be used for making scaffolds.

Suitable properties of scaffolds include chemical properties, physical properties, and biological properties. Chemical properties should be biologically active and sterilizable. Biologically active, what it means is that it is friendly to the cells. And sterilizable means you don't want any unwanted pathogens, bacteria, fungus, or mycoplasma to grow. So you should be able to sterilize them before inoculating those scaffolds with the cells.

The physical property should be mechanically supportive if everything is fine, but the moment cells start growing, it begins to crumble because of the weight of the cells or the moment you pass the nutrients or the medium flowing through them; then they perish. You don't want those kinds of scaffolds, and the biological property should include being biocompatible and biodegradable. This has already been discussed in the previous slide, emphasizing that it should be friendly to the living cells that are growing on it. And let us look at the chemical properties. Chemical properties in detail include suitable surface chemistry and bioactivity for cell attachment.

If the cells are not able to attach to them, then that scaffold is not of good quality to facilitate binding of the biomaterial with the cell surface receptor. Every cell has different receptors. Some are used for attaching to the matrix. So the same receptor a cell should be able to use to bind to the scaffold as well. In the case of synthetic polymers, if they lack suitable surface chemistry, then their surface modification has to be performed.

Scaffold is good, tough, very friendly, but it does not welcome cells. So you can do some coating so that the cells will find a home now. So you should also keep this logic in mind. So that biomaterial should allow surface modification. That modification is now going to be helpful for the cell.

In the case of synthetic polymers, if they lack suitable surface chemistry, then their surface modification has to be performed. For example, RGD, which is arginine-glycyl-aspartic acid. It's a three-amino-acid peptide: arginine, glycine, and aspartic acid. Together, it's a small peptide inclusion for better cell attachment, so when you make the biomaterial, you include this RGD (arginine-glycine-aspartic acid) so that the biomaterial is now more friendly to the cells because it will interact with this three-amino-acid peptide. Sterilizable without any loss of properties.

So you made everything fine, but after sterilizing it with either UV, gamma radiation, or autoclaving, it lost its properties. You don't want that. So, to prevent contamination, you must sterilize the biomaterial, but it should retain its quality. Let us look into the physical property; it is a little bit of physics that comes into the picture: mechanical strength, which would withstand the shear stress generated by biological fluid flow. As I mentioned, stress and strain are represented in this graph when you apply the force and the change that is occurring in the body.

Stress is the change that occurs if you apply stress onto an elastic body; then its body deforms and becomes longer. Stress and strain have a connection until it reaches the elastic limit. You also know that a rubber band can be stretched up to a certain limit, and it will not go exponentially long. After that, it will not stretch anymore.

And if you stretch any more, it will break. So, from the elastic limit to the break point, there is a slight change in the shape. Once you continue to apply strain, the change in shape will not occur. And after that, there will be a breakpoint where it will lose the so-called elastic property. Different types of forces are applied to the scaffold to test whether it is good or not: tensile force, which is a force tending to tear it apart, like when you take a paper and try to tear it; that is the tensile strength. Then there is compressive strength, which is the force that squeezes an object's surface together to cause its mass to bulge.

When you're pressing something, you take something and try to press it to see how much force it can take. And then, shear. Shearing forces are unaligned forces pushing one part of the body in one direction and another part of the body in the opposite direction. It's different from tensile stress. This one, shear stress is like twisting something.

That is sheer stress. It's not just a uniform. You take a cloth and twist it. That is shear stress. And then comes the torsional stress. Torsional stress is the winding force on an object.

That means... One side of the object is pushed in one direction, and the other side is pushed in the opposite direction, creating a torque force when applied. It should be able to withstand this; you don't want the body or the scaffold to keep breaking when any of these forces are applied. Terminology: stress is an applied force or a system of forces that tends to strain or deform the body. Stress is the force; strain is the change in the body. Strain is a change in the dimension of the body under a load, like an elastic one-inch rubber band.

If you pull it into a ten-inch rubber band, ten inches is the new shape, and it became ten inches long because of the force you applied, and that force is the stress. Changing the dimension, and once you leave the stress, it will come back to the original dimension; it may be the ratio of lengths, areas, or volumes; thus, it is dimensionless. The strain is dimensionless; it gives the extent to which a body is distorted when it is subjected to a deforming force when it is under stress, and the load weight is the weight. Divided by the force applied, that is the load which you describe in physical terminology, and the elastic limit is the stress that can be applied to an elastic body without causing permanent deformation, the stress point at which a material will no longer return to its original shape when subjected to a higher stress; once it reaches the elastic limit, it will not come back. You may have seen that once you put an elastic around a box for one year, and you try to take out that elastic, it will not return to its small shape.

It will remain for longer. So brittle materials tend to break shortly after their elastic limit, while ductile materials deform under stress beyond their elastic limit. If a good rubber band is there, even if it has reached its elastic limit, it will not break. It will stay there, but it may not return to its original shape. So, like a one-inch rubber band, I pulled it to ten inches multiple times.

Now it will not come back to one inch every time. It may now stay at 1.5 inches because of multiple pullings. Breakpoint is a point of discontinuity, a change, or a cessation. That means it is not going to come back to its original shape at all.

It broke into two pieces. Yield point is a term used for the point in the stress-strain curve at which the curve levels off and plastic deformation begins to occur. So this is the yield point at which we should understand the stress-strain curve; the curve levels off, and they just weigh normally. So this is all some terminology you should keep in mind. Yield stress, the yield strength, or the yield point of the material is defined in engineering

material science as the stress at which a material begins to deform plastically. So this is the yield stress, the terminology that is used to explain the ability of biomaterials to deform plastically.

Prior to the yield point, the material deforms elastically and returns to its original shape when the applied stress is removed. So this is a good property for a biomaterial to possess. Once the yield point is passed, some fraction of the deformation will be permanent and non-reversible, like I gave in the example of the rubber band; if you stretch a one-inch rubber band multiple times, after multiple stresses, it will no longer come back to one inch once you remove the force; it will stay at 1.5 or 2 inches. Young's modulus, the slope of the elastic portion of the stress-strain curve, is a quantity often used to assess a material's stiffness.

So then comes the mechanical property, the ultimate strength. That is the maximum value of load-bearing at which it may become permanently deformed. That is what the ultimate strength and tensile strength are, the load bearing up to which a scaffold could be elongated. Prior to breaking, it is tested for several fibrous materials, and then compressive strength is the degree of compression tested for a porous scaffold. If it has pores, think about a sponge; you are pressing it, and you don't want to reach a stage where all the pores are closed. If it allows the closing of the pores, then there is a chance that when the fluid force comes strongly, the pores may get clogged.

So even while pressing, the pores should not reach a stage where they completely close. And also suitable tailor properties such as pore size, percentage of porosity, etc. It also has to be taken into consideration. Now, if you look into the biological properties, biocompatibility, we have discussed it; that is basically the ability of a material to elicit an appropriate biological response in a specific application by not producing a toxic, injurious, or immunological response in living tissue. If any of this happens, say you get inflammation because the cells are interacting, because the biomaterial is somehow attracting immune cells, and you get inflamed, and that is bad for that tissue and maybe for the rest of the organ.

And you don't want to have any toxins present in these biomaterials. So that is why biocompatibility comes into the picture. Biodegradable means the rate of scaffold degradation is equal to the rate of tissue formation. Because when a tissue is formed, that tissue also secretes its ECM.

No tissue needs to be told, okay, secrete ECM. That is their normal behavior. And when that is produced, you don't want this biomaterial again. So it should coincide. If one gram of ECM is produced, one gram of degradation of the biomaterial should occur so that

there will be equilibrium between them. So if you look at the scaffolds, different qualities have to be taken into account; those are the polymers used in ocular implants, drug delivery devices, skin, cartilages, several polymers, ceramics, and bioscaffolds.

What they use is for bone replacements, heart valves, dental implants, etc.; semiconductor materials are used for implantable microelectrodes, biosensors, etc. Metals are used for dental implants, orthopedic screws, fixation, and so on. So they are all coming under scaffold, but they have different chemical properties.

So, some examples of scaffold materials are listed here. The polymer type is given here. Silicone rubber. The materials are Dacron, polymethylacrylate, polyurethanes, hydrogels, cellulose, collagen, silk fibroin, chitosan, gelatin, alginate, etc. They are examples of polymers. And they are used for applications in catheters, tubing, vascular grafts, intraocular lenses, bone cement, catheters, and pacemaker lids.

You can read the list; many are there, and metals include stainless steel, titanium, and alumina. They are used as metal examples and are mainly used for orthopedic devices, stents, orthopedic and dental devices, etc. Ceramics include bioactive glass, mainly silica. Hydroxyapatite, beta TCP, and volastonite are all used for several orthopedic and dental devices, many bone tissues, and various solid applications, which are very rugged and demanding. Synthetic polymers also come into the picture; they are more controllable from the compositional and material processing viewpoint, and scaffold architecture is widely recognized as an important parameter when designing a scaffold.

We have to take good care of the scaffolds' architecture; otherwise, it will not pass all the biological, physical, and chemical requirements. They may not be recognized by the cells due to the absence of biological signals, but you have to make sure that these scaffolds are not. Anti or against any biological signal, they should act neutrally so they don't favor or oppose; sometimes, for a given tissue, they should favor, like we mentioned that RGD (arginine-glycine-aspartic acid) should allow the attachment of cells to them, so you have to include that tripeptide. Many natural polymers are used.

Natural materials are readily recognized by cells. Interaction between cells and biological ECM is a catalyst to many critical functions in tissues. So natural polymers are quite friendly for this kind of biological reactions. So the classification of polymeric biomaterials and scaffolds broadly includes natural polymers and synthetic polymers. Natural polymers include silk fibroin, chitosan, starch, gelatin, alginate, cellulose, and others.

Synthetic polymers include polyglycolic acid (PGA), polylactic acid (PLA), poly...

Caprolactone, polyvinyl alcohol, and polymethyl methacrylate. Some of the examples we discussed in the very beginning of this scaffold.

I told you that we would see more in detail about them. So you may go back and revisit different morphology photos we had shown, and some of them have come in that situation. For advances in scaffold technology, we need to know cell matrices for 3D growth and tissue reconstruction. So, for any shape of tissue you want to create, you must have a suitable scaffold; we don't have a universal scaffold. Or one, like a Swiss Army knife, you have one solution for all. It is not like that because every tissue has a different density, different functionality, different morphology, etc.

Everything is a different form for different tissues. You don't want your kidney to be as solid as a bone, nor do you want your bone to be as liquid as a kidney or liver. So both are useless, so we should understand the material suitability, which is also decided based on that. Biosensors, biomimetic, and smart devices should have unique properties; like I mentioned about the heart valves, they should not cause any blood clotting or any inflammatory response in the heart tissue.

Because of this valve being implanted. So these things have to be taken care of. Controlled drug delivery and targeted delivery. When you are administering a drug to the animal, we don't want this scaffold to block the entry of the drug or the exit of any biologically degraded material, even while making this tissue. You don't want this scaffold to block it.

This all comes under biocompatibility. Bio-hybrid organs and cell immunoisolation. So we want to be sure that the scaffold, while present in a living organism, is non-existent. So, that is the so-called bio-hybrid organ. When you make it, it will last forever. So what are some of the challenges that we face? To more closely replicate complex tissue architecture and arrangement in vitro.

That is one of the challenges. To better understand the extracellular and intracellular modulators of cell functions. That is another challenge. To develop novel materials and processing techniques that are compatible with biological interfaces. And to find better strategies for immune acceptance. So these are all some of the challenges that are faced by scaffold manufacturing or synthesis time.

We will learn more about regeneration biology and tissue engineering in the next class. Thank you.