Medical Biomaterials
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Lecture – 32
Natural/Biopolymers

Hello everyone welcome to the course on medical bio materials. We are going to start a new topic; it is called bio polymers or natural polymers. Bio polymers are produced by micro organisms like bacteria, fungus, animals, even plants. So, those are called bio polymers. So, they are naturally occurring. Sometimes some people use this concept of bio polymers, if the monomer is also is of biological or natural origin, but I am going to talk only about the polymer, that is of animal origin or a plant or a bacteria or fungal origin only. So, these bio polymers have several advantages and of course, they have plus because they are biological in origin. They have very good properties, such as bio compatibility many of them also degrade that is bio degradability.

So, these are some good properties, but the problems of these bio polymers are they have very very poor mechanical strength, mechanical properties. So, they cannot be used alone on their own. So, they have to be blended with natural polymers.

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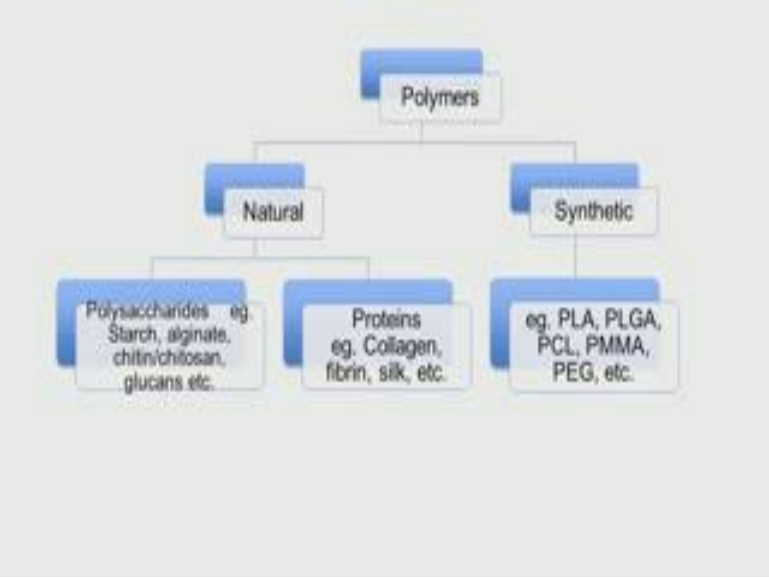

So, we will talk about these bio polymers. So, the polymers can be divided into two; the natural and the synthetic. The synthetic or I use chemical method for preparing like poly lactic acid, poly lactic glycolic acid, poly ethylene, poly propylene, poly methyl, methacrylate, poly ethylene glycol, polycaprolactum all these are polymers produced using synthetic roots either we use a catalyst or we just do, we do a condensation in the lab. Whereas if you look at the natural or the bio polymers, they are produced by animals or bacteria or fungal or even plant derived. So, these are called bio polymers, they are naturally occurring.

So, we have two big groups; the polysaccharides, the protein. The polysaccharides means we will have one of those saccharide groups like glucose or mannose connected in different architecture, different molecular weight like the starch, the alginate, the chitin, chitosan, glucans all these. They are polysaccharides; that is saccharides means sugar. Proteins they have the protein bond; that means, amide bond C double bond O N like collagen, fibrin, silk they all called protein. So, they have this amide bond into their system. So, the natural polymers or poly saccharide based or protein based; that means, these are all based through sugar, these are all proteins. So, as you can see because the body contains lot of proteins and sugars and many of these degradation cycles in the body make use of this. So, they nicely get absorbed and degraded in various cycles in the human system. That is why they are bio compatible and also they are highly bio degradable completely get eliminated from the system because they are sugar based or they are protein based.

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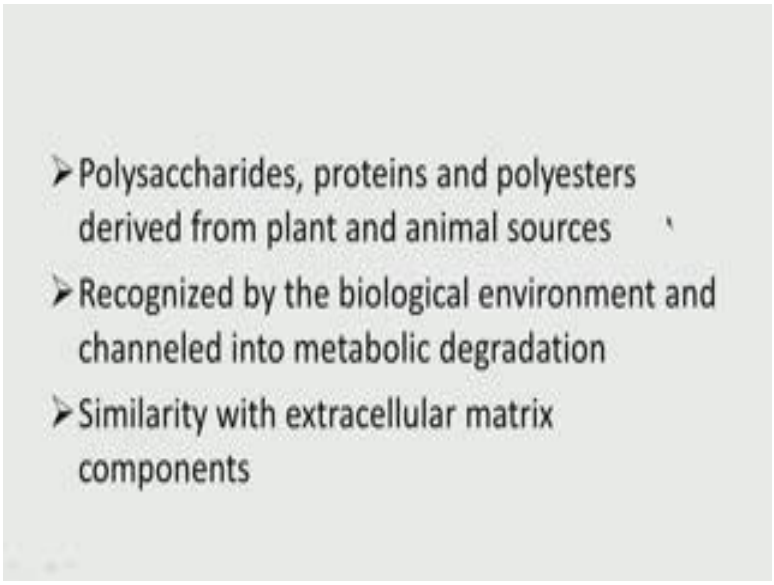

So, we are going to look at each one of them slightly in more detail and see their advantages, disadvantages and where they are used. Some of these bio polymers are not really used to as of now in practical applications, but they seem to have quite a lot of future potential. These are polysaccharides, proteins, polyesters derived from plant, animal, bacterial sources. They are recognised by the biological environment. So, like I said sugar the many of these cycles, metabolic cycles in the body either go through have sugars or protein. So, they get channelled into those degradation processes and completely get eliminated. So, there is no question of build up or accumulation of these bio polymers. They also have similar to the extra cellular matrix components. So, they will be highly hydrophilic in nature. Generally they will be highly hydrophilic in nature and many of them are also soluble in water of course, there are some which are also not soluble in water. So, we will see some of those both the classes.

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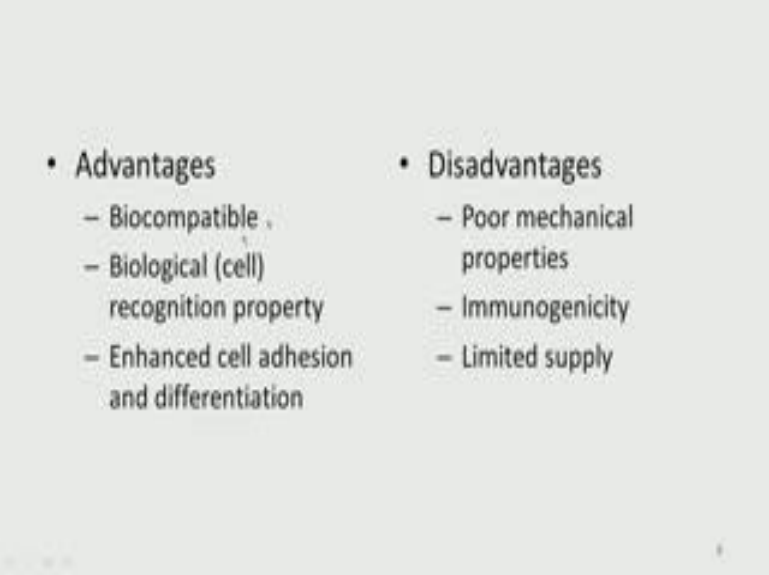

So, the advantages; they are bio plus compatible, they are recognised by the cell. So, you will have very good cell adhesion and cell differentiation. So, when I use bio polymer based scaffolds you can see very good cell addition, proliferation taking place unlike a synthetic polymers. So, sometimes what we do is we mix synthetic polymers and bio polymers to improve the cell adhesion properties. Otherwise if you are using purely synthetic polymers we have to make it very hydrophilic. So, that the cell adhesion is improved. Disadvantages of course, they have very poor mechanical properties. So, they cannot take much stress, they cannot have good flexural properties immunogenicity so; obviously, if they are derived from animal the chances are there could be some cross contamination, limited supply. We cannot have large amount for example, if you are talking at polyethylene; polyethylene is manufactured through the petrochemical crude. So, you can make tons and tons of polyethylene whereas, if I am talking about say hyaluronic acid that is a bio polymer and you can have only limited supply from bacterial source.

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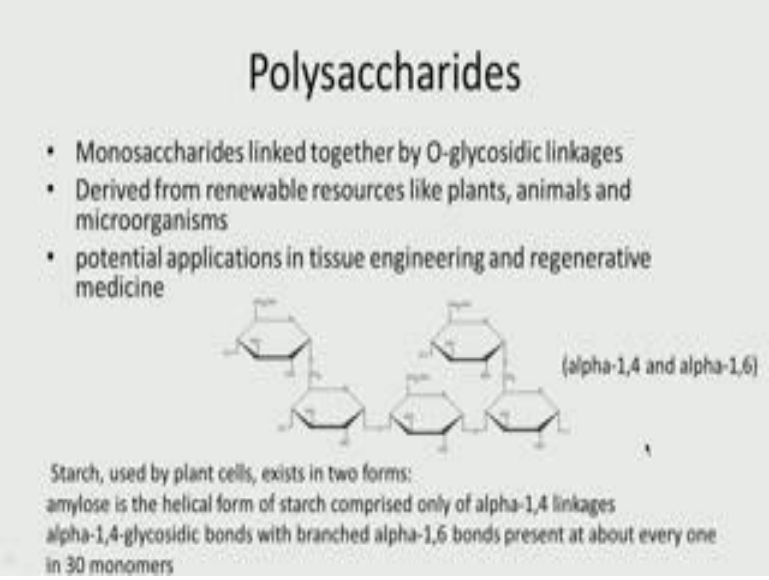

So, that is a big issue about these bio polymers, the limited supply. Let us start from each one of them, these polysaccharides are linked several mono saccharides are linked together through this glycosidic linkages. This is called a glycosidic linkage. So, we have this oxygen. So, we have a polysaccharide here I mean sorry mono saccharide here, another sugar here, another sugar they are connected so; obviously, this is called the glycosidic linkage. So, you can have alpha linkage, we can have beta linkage, we can have alpha 1, 4 depending upon how these two adjacent mono saccharides are connected. We can have alpha 1, 6 we will look at them each one of them. So, this is called the glycosidic, O glycosidic linkage and here we have one sugar, another sugar mono saccharide they are connected as you can see here. So, we can have alpha type of connection, beta type of connection, we can have 1, 4; 1, 6; 1, 3 and so on actually.

So, these are derived from renewable sources like plants, animals, micro organism. They can be used in regenerative medicine, tissue engineering. So, they form a very good scaffold. So, if you are thinking about biodegradable, scaffolds this is very good, but then of course, they will have poor mechanical strength. For example, if you look at starch, they are used by plant cells they exist in two forms before that let us look at this alpha 1, 4 and alpha 1, 6. So, look at this. So, if we have, this is called the alpha 1, 4 linkage. This is called the alpha 1, 4; in an alpha the O H is below as you can see below

these two mono saccharide link and this is called the alpha 1, 4 linkage. As you can see 1 and here this is the 4; 1, 2, 3, 4. So, this is called the alpha 1, 4; that means, two adjacent mono saccharides are connected if 1 and then you have 4 and when you say alpha the hydroxyl is below these two.

If you look at the alpha 1, 6 linkage this is called the alpha 1, 6 linkage. So, as you can see this is 1 and 5, 6; 1, 2, 3, 4, 5, 6. So, there is a connection between the first carbon, in one mono saccharide with the sixth carbon in another mono saccharide. So, we count like that. So, the oxygen becomes 0 then, we have 1, 2, 3, 4 like that. So, we have alpha 1, 4 linkage and we have the alpha 1, 6 linkage. Now if we look at starch; starch used by plants exists in two forms. One is called the amylose the helical form of starch containing only alpha 1, 4 linkage. So, it will be like this straight line with this type of linkage alpha 1, 4. You can also have another form alpha 1, 4, but after every 30 monomers we will have one alpha 1, 6 bonds; that means, alpha 1, 4 with branched alpha 1, 4 at about every one in 30 monomers. So, when we have alpha 1, 6, it is like a branching happened because here you can have alpha 1, 4 happening here right. So, alpha 1, 4 it will be like a straight line. So, it can form a helical whereas when you have once in a while alpha 1 6 you going to have this type of branching that is taking place actually.

So, you understand the concept of 1, 4; ,1 6. So, 1, 4 is; so 1, 2, 3, 4, 5, 6 and this is 1. So, this is a 1, 6 bond 1, 4 is this is 1 and 1, 2, 3, 4. So, this is the 1, 4 bond and if the oxygen is below the planes of these sugar then it is called the alpha. I will show you the beta form also later because glucons are in the beta linkage.

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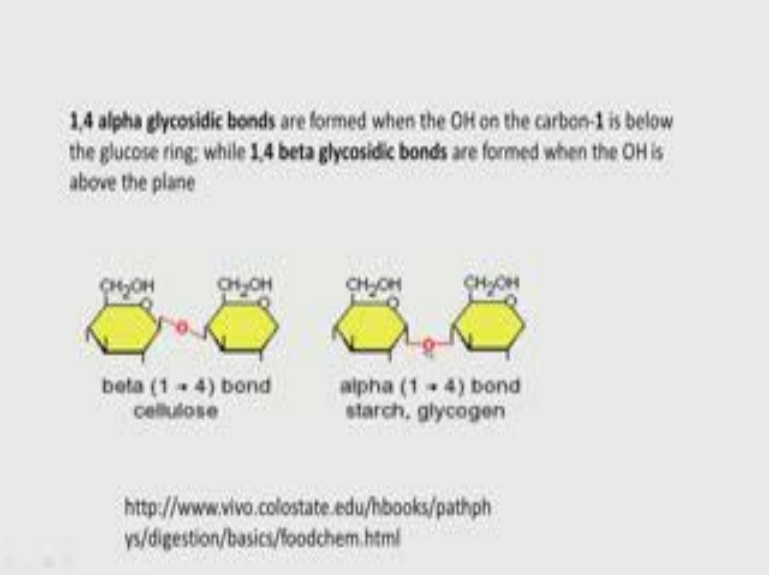

So, once more see here in the alpha the oxygen is below these mono saccharides like starch. glycogen and so on. Whereas in beta when the O H is above the plane. So, you can see this O H is above like cellulose, glucon, all these are beta linkage. So, the O H is above the plane whereas, here the O H is below the plane. So, the glycosidic bond happening below the plane whereas, here the glycosidic bond is above the plane. So, this is again 1, 4; 1, 2, 3, 4. So, this is one. 1, this is again 1; 1, 2, 3, 4. So, this is a 1, 4 beta linkage. This is 1, 4 alpha linkage we can see the difference.

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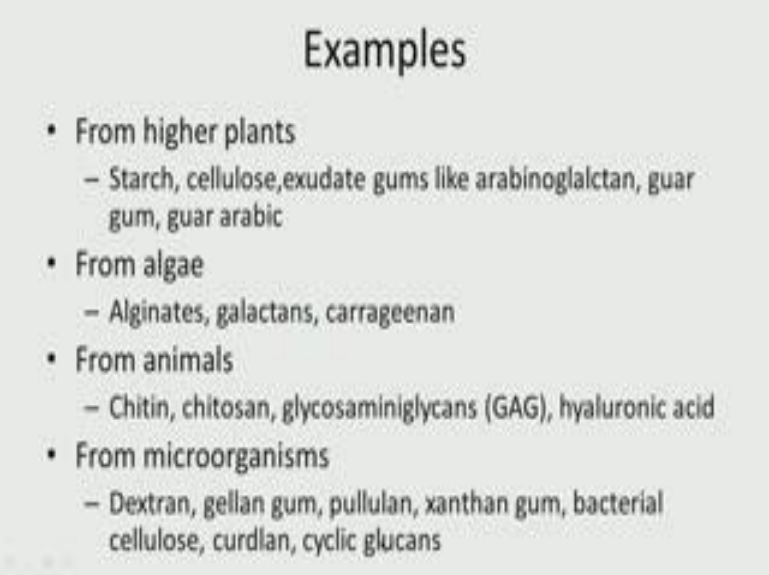

Now, you know how to nomenclature it. This is taken this picture was taken from this reference. So, starch, cellulose, gums like arabinogalactan, guar gum, guar arabic these are from plants. From algae you have alginates, galactans, carrageenan; from animals we have chitin, chitosan, glycosaminiglycans, hyaluronic acid. From micro organisms we have dextran, gellan gum, pullulan, xanthan gum, bacterial cellulose, curdlan, cyclic glucans, linear glucans and so on actually.

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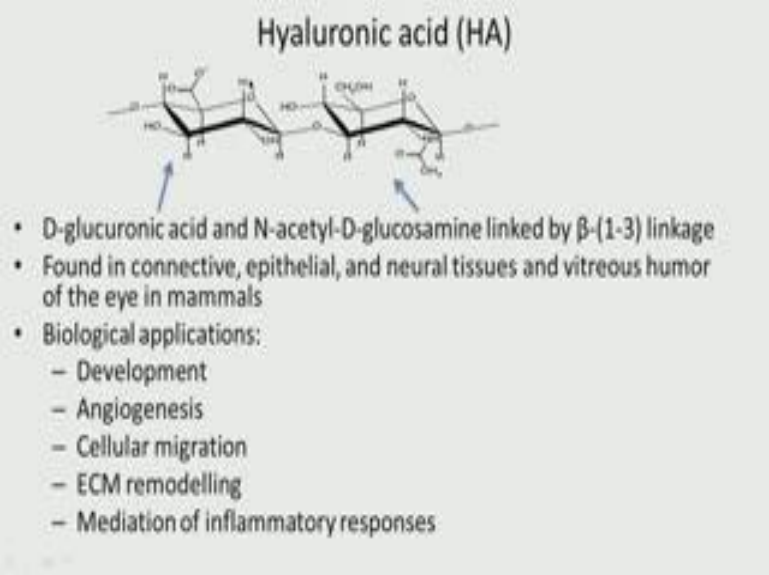

So, let us look at hyaluronic acid, this is obtained from cell bacteria. So, hyaluronic acid is a D glucuronic acid. This is called the D glucuronic acid, so you can see the acid D glucuronic acid with N acetyl, D glucosamine. So, this is the acetyl group C H 3 C O is called the acetyl group, this is the amine group. So, N acetyl D glucosamine linked by beta; 1, 3 linkage, this is 1; 1, 2, 3. So, you understand 1, 3 linkage. So, hyaluronic acid is made up of glucuronic acid, D glucuronic acid and N acetyl D glucosamine linked by beta 1, 3. So, this plus hyaluronic is interestingly found in connective tissues, epithelial, vitreous humor of eye in mammals and so on actually. So, hyaluronic acid is very very important especially in the connective tissues, it gives you some sort of a lubrication. They are also found in the eyes, so the biological applications development, angiogenesis, cellular migration, extracellular matrix remodelling, mediation of inflammatory responses.

So, hyaluronic acid is found in the human body and it is got lot of requirements. So, hyaluronic acid is also used widely as a bio material, but the production generally by bacteria is very very low. So, it is made up of these type of glucuronic acid here and acetyl D glucosamine on this side.

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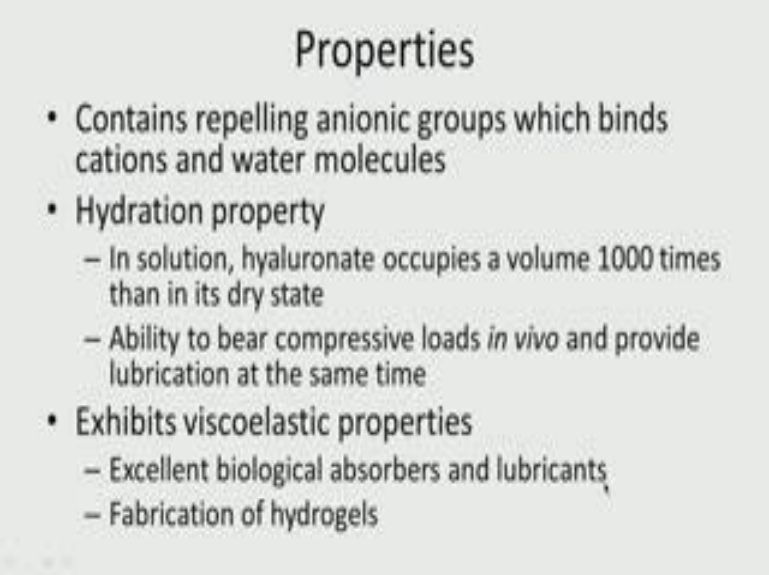

So, what are the properties it contains repelling anionic groups. So, as you can see here. So, we have the anionic group because of the acid there. So, it contains repelling anionic groups which binds cations. So, if there any cations like calcium 2 plus or magnesium sodium and water molecules also. So, it is got very good hydration property. So, in solution hyaluronate occupies a volume of thousand times than it is dry state. So, it expands quite a lot in the presence of water because of the anionic groups. So its ability to bear compressive loads in vivo, so it provides lubrication. So, it expands quite a lot almost 1000 times water in the solution form and also its got very good compressive loads it is also very good lubricant. It also exhibits viscoelastic properties. So, it is used biological absorbers, lubricants especially if there are joints where the lubrication has gone down because of (Refer Time: 15:04) process or age, hyaluronic acid injection is a very good example; Fabrication of hydrogels because it can occupy 1000 times, because it observes so much of water.

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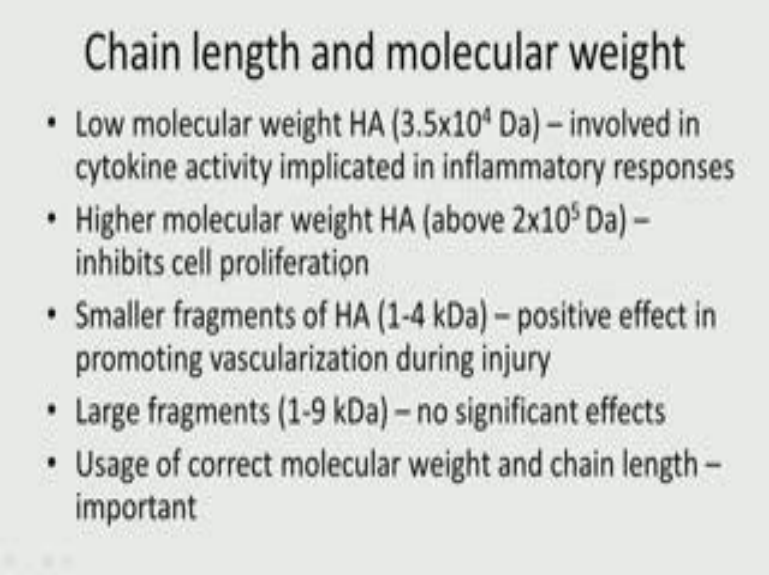

So, the chain length and molecular weight are very very important in determining their properties. So, low molecular weight H A it is involved in cytokine activity implicated in inflammatory responses; that means, low molecular weight you are talking in terms of 10 power 4 Dalton whereas, if you take higher molecular weight like 10 power 5 Dalton it inhibits cell proliferation you can see, there is lot of difference low molecular weight and high molecular weight inhibits cell proliferation. Whereas, smaller fragments like 1 to 4 they have positive effect in promoting vascularisation during injury. So, it helps after an injury the vascularisation.

So, generally one is looking at very very low molecular weight fragments like this or like this. So, large fragments they are no significant effect. So, use of correct molecular weight and chain length are very very important. So, as you can see low molecular weight involved in cytokine activity for inflammatory response, higher molecular weight inhibits cell proliferation very very small fragments and they help in vascularisation during injury larger fragments have no use. So, one is to select or design or synthesise the correct molecular weight hyaluronic acid.

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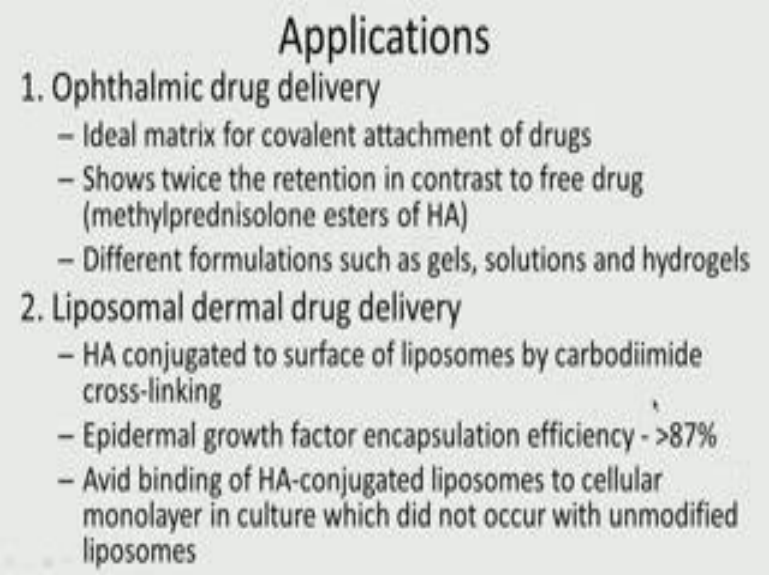

Applications; it is used in ophthalmic drug delivery because it absorbs lot of moisture. So, it can be used in ocular region, ideal matrix for covalent attachment of drugs because it is got a C double bond O negative charge shows twice the retention in contrast to free drugs. So, when we put with H A, it retains more than different formulation such as gels, solutions, hydrogels all these can be prepared with the hyaluronic acid. Liposomal dermal drug delivery so in hyaluronic acid conjugated to surface of liposomes through cross linking. So, we can encapsulate almost 80 to 90 percent of the drug.

So, it also helps in H A conjugated liposomes cellular monolayer in culture whereas such things do not happen when we do not have the H A at all.

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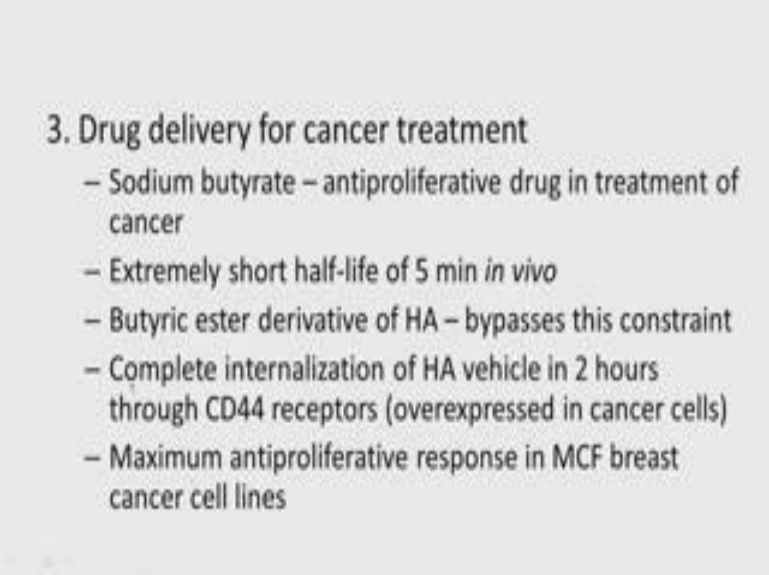

Drug delivery for cancer treatments, there are some examples of drug delivery for cancer treatment; sodium butyrate. For example, it is an antiproliferative drug in the treatment of cancer, it is got a very short life time 5 minutes in vivo, but butyric ester derive H A and when we use this together this bypasses this particular constraint. So, it goes even after 2 hours internally. So, H A gets completely internalized through C D 44 receptors. So, we get very good anti proliferative response in MCF breast cancers cell lines also.

So, the drug alone, it is a very good anti proliferative drug, but it is got very short half life 5 minutes, but when we use butyric ester derivative H A and use that as a drug delivery vehicle for the this particular drug, we can extend it to almost 2 hours and we get very good anti proliferative activity and this has been tested. Of course, they have still not come into commercial, but these are some experimental studies.

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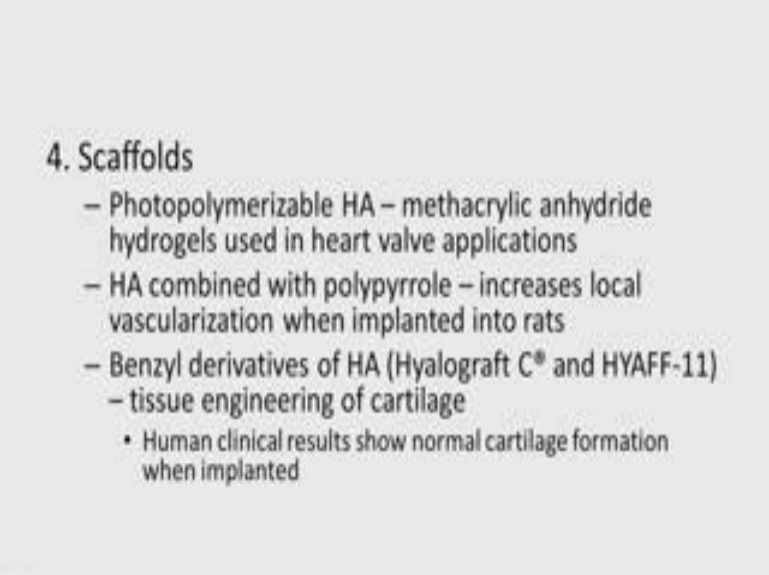

Scaffolds; so, if I want to design scaffolds photo polymerizable H A like a methacrylic anhydride hydrogel used in heart valve applications. H A combined with polypyrrole increases local vascularisation when implanted into rats. The benzyl derivatives of H A, there is a commercial product they can be used for tissue engineering of cartilage, human clinical trials are in progress for the benzyl derivatives of H A.

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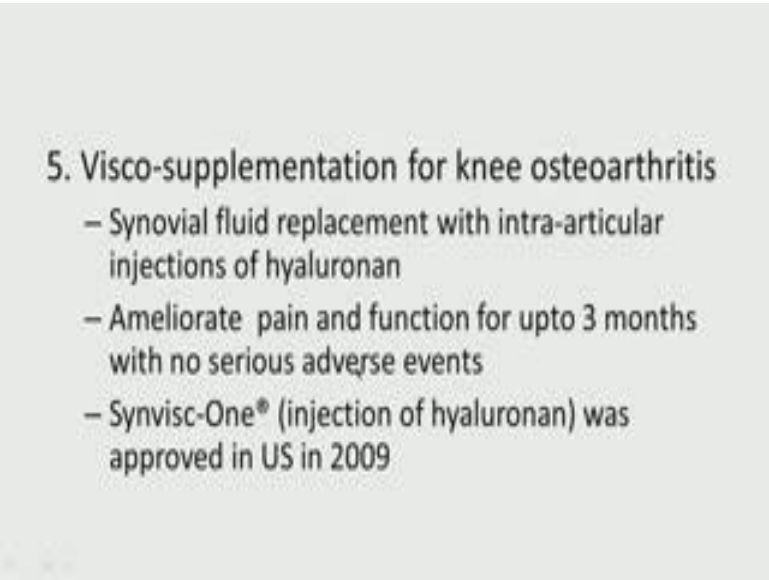

Visco supplementation for knee osteoarthritis, like I said when the lubricant between in the knee joints get depleted because of age or arthritic problems then H A could be injected as a supplementation. So, synovial fluid replacement with intra articular injections of hyaluronan, it can be, it has been tested even up to 3 months. There is a commercial product called syncisc one injection of hyaluronan this was approved in US FDA in the year 2009. So, it is a supplementation for arthritis, osteoarthritis especially when the synovial fluid gets exhausted because of arthritic problem or old age.

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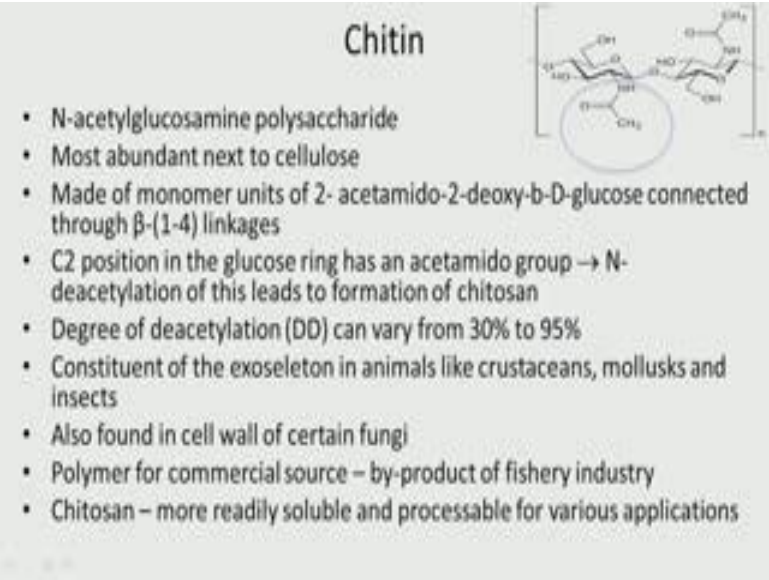

So, quite a lot of applications of hyaluronic acid, but the production is still a big issue. So, still being used in very small amounts commercially, next comes chitin. Chitin as you can see it is got a acetyl group, N acetyl group that is called a chitin. It is a N acetyl glucosamine polysaccharide. So, we have the polysaccharide here. So, we have the acetyl group attached to the nitrogen amine. It is most abundant next to cellulose made of monomers of units of 2 acetamido, 2 deoxy. So, we have acetyl here the glucose connected through here we have the beta linkage. Alpha means it would have been down; this is beta linkages 1, 4 as you can see the 1 and 1, 2, 4; 1, 4 linkage. So, we have the C 2 position when we have the amine that is replaced here by we have the acetyl group.

So, if I remove this acetyl that is called deacetylation that leads to formation of chitosan. Chitosan also has very good important properties. So, when we remove this acetyl and when we put H here that is called plus deacetylation. Chitosan is much more hydrophilic then chitin. So, degree of deacetylation generally can vary between 30 to 95 degrees, it is called DD. This is constituent of exoseleton in animals like crustaceans, mollusks and many insects also found in cell wall of certain fungi. This polymer for commercial source by product of a fishery industry also. Chitin is found quite lot from fishery fish scales. Chitosan is more readily soluble because we are removing the acetyl group and putting H. So, it becomes more hydrophilic unlike chitin, chitosan is more soluble.

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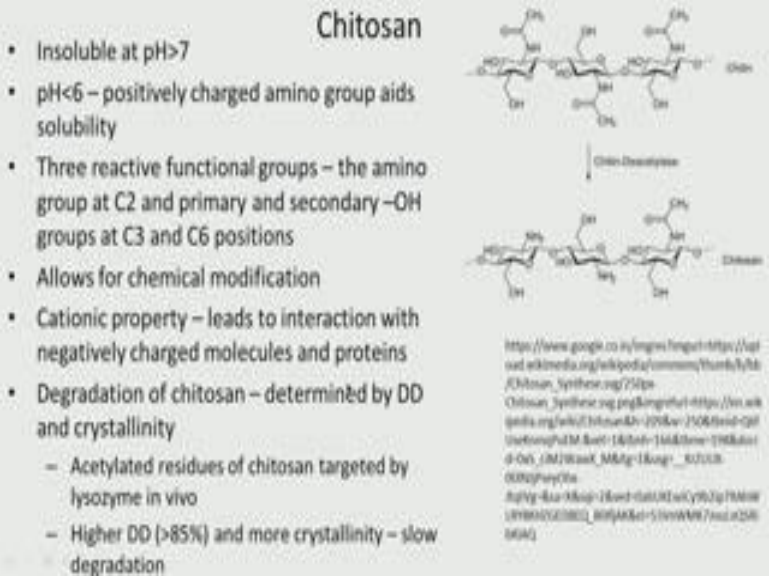

So, chitosan how do you make? We remove the acetyl group. So, we have deacetylation enzyme. So, it forms N H 2 here. So, with the degree of plus deacetylation could vary 30 to 90 percent. So, we can increase the solubility further and further by deacetylasing more and more. So, it is insoluble at p H greater than 7, p H less than 6 positively charged amino groups. So, they aid solubility. So, if you look at the functional groups here we have the amino group at C 2 position; primary and secondary alcohols. This is primary alcohol this is the secondary alcohol groups at C 3 and C 6 positions. So, all these help in functionalising these polymers. So, I can functionalize this polymer either at this position, this position and this position. So, it is highly functionalizable. So, there

are lot of papers if on chitosans. So, if you go to Pub Med and look at applications of chitosan you will see lot of modifications to this because of so many different functional groups.

So, chemical modification; cationic property leads to interaction with negatively charged molecules because. So, we can have plus charge. So, negatively charged molecules, negatively charged proteins start interacting with them, degradation of chitosan determined by degree of plus deacetylasation crystallinity. So, it can form many crystalline surface the acetylated residues of chitosan targeted by lysozyme that particular enzyme. So, higher the degree of plus deacetylasation more crystallinity slow degradation. So, if I have very high deacetylated material chitosan then it forms a very good crystal structure and so the degradation is also very very slow. Whereas if the crystal structure is less; that means, if you have less deacetylasation then it can degrade much faster. This particular picture is taken from this particular reference.

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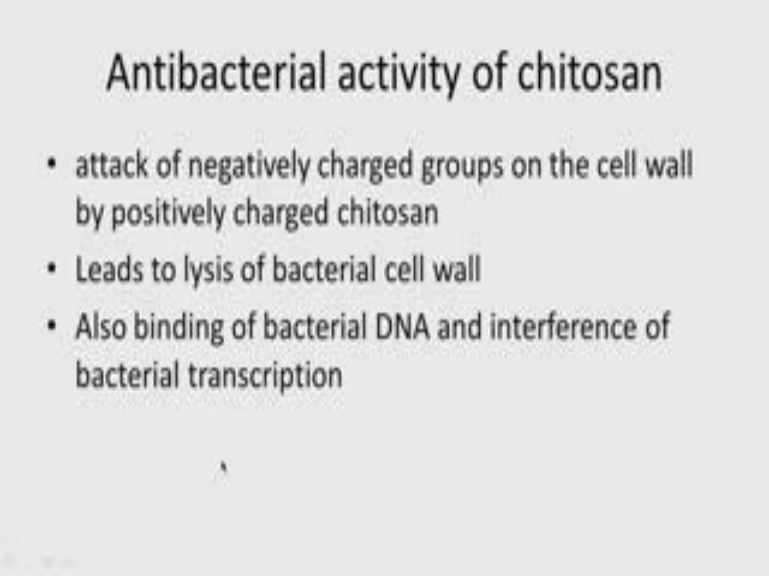

So, antibacterial activity of chitosan, it attacks negatively charged groups on the cell wall by positively charged chitosan. So, as you can see we have positively charged and plus. So, they attack the negatively charged groups on the cell wall and so cell wall breakage lysis of bacterial cell happens actually. So, they chitosan exhibits anti bacterial property,

they also bind to bacterial DNA and interferes with bacterial transcription.

So, these are the antibacterial activity of chitosan. It is used as a dietary supplement it lowers low density cholesterol.

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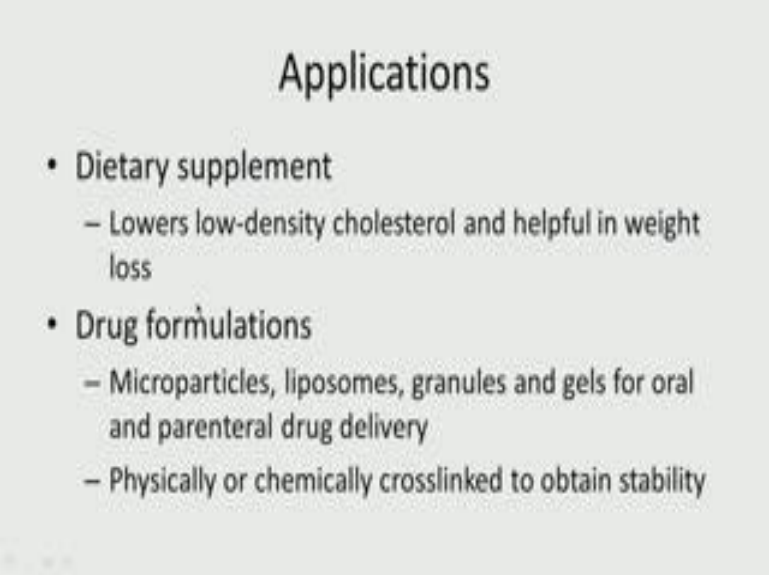

So, low density cholesterol is involved in many issues cardio vascular issues and so it helps in the weight loss. So, it is used in drug formulation as I said the chitosan could be modified quite a lot it is got three functional, different functional groups; the primary O H, the secondary O H, and the N H 2. So, many drug formulations are made micro particles, liposomes, granules, gels for oral and parenteral drug delivery. It can be physically or chemically cross linked; we can cross link this to improve their stability of this chitosan.

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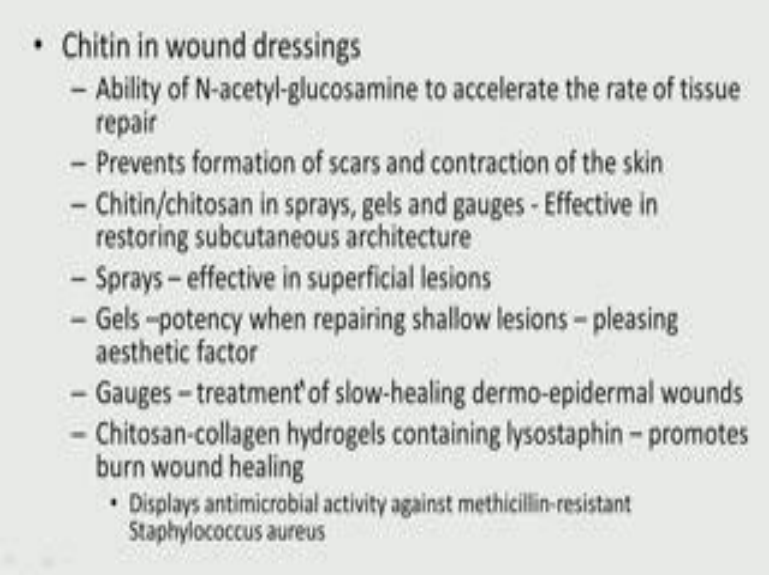

So, chitin is also used in wound dressing, the ability of the N acetyl glucosamine to accelerate the rate of tissue repair. What is this N acetyl? Like I said this is the N acetyl glucosamine.

So, the ability of N acetyl glucosamine to accelerate the rate of tissue repair; prevents formation of scars and contraction the skin. This chitin chitosan in sprays, gels and gauges; effective in restoring subcutaneous architecture, sprays effective in superficial lesions, gels repairing shallow lesions, pleasing aesthetics. So, as you can see many of these advantages on surface. So, we are not bothered about mechanical strength or mechanical stability. They are used as sprays, scars, tissue repair, aesthetics factors, slow healing dermo, epidermal wounds, hydrogels, burn wound healing.

So, all these are surface application because chitosan chitin they have a very good functional groups. So, they are also very good in wound healing applications, burn wound healing. And because they also have the antimicrobial properties, we can use them as such and they can target especially MRSA; that is methicillin resistant Staphylococcus aureus strains also or we can impregnate anti antibiotics. So, that it gives you an added antibiotic activity. It is also used in ocular drug delivery vehicle for topically applied vancomycin.

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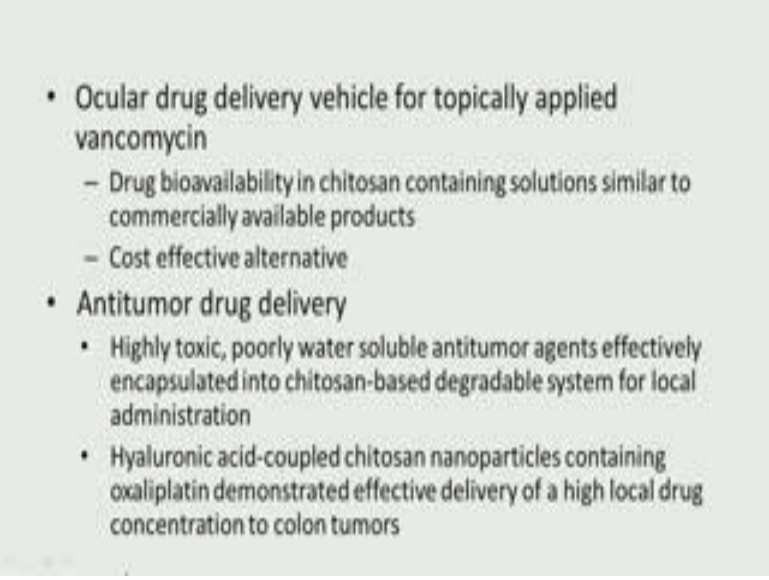

So, we can combine it with vancomycin for delivery especially in the high region. So, the bioavailability when you have chitosan as a drug delivery improves also it is very very cost effective. It is been even tested as drug delivery, antitumor applications. Highly toxic poorly water soluble antitumor drugs when you encapsulate with chitosan based degradable system. So, it is very good drug delivery and degrades at the location where it has to deliver the drug. So, it can take care of highly toxic drugs. So, hyaluronic acid another bio material, bio polymer which I talked about earlier coupled with chitosan nanoparticles containing anticancer drug like oxaliplatin it goes very good local drug delivery concentration in colon tumour.

So, as you can see, they can form very good drug delivery and at the point of a discharge they degrade. So, that is the advantage of this actually.

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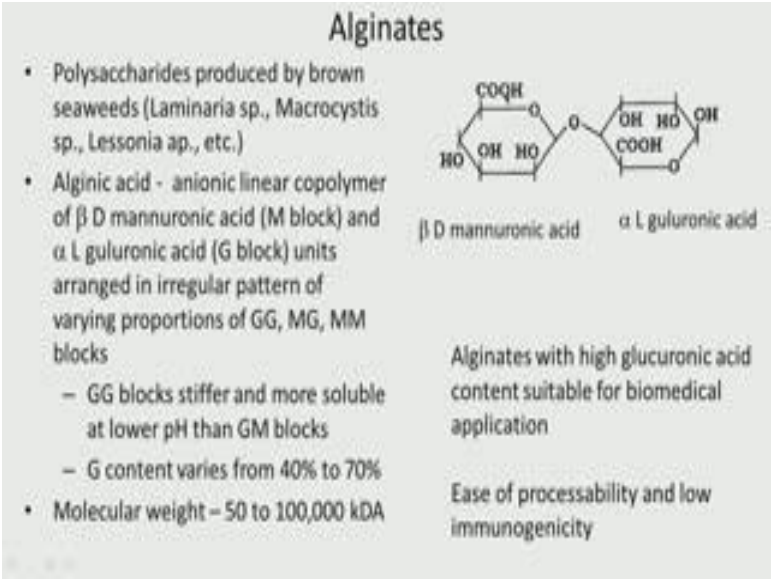

Now, look at alginates; these are connected by beta D mannauronic acid and alpha L guluronic acid. So, alginates with high glucuronic acid, they are suitable for biomedical applications because they are very easy to process and low. So, they are very good if I have very large amount of this. So, they are combination of beta D mannuronic acid and alpha L guluronic. So, these are polysaccharides produced by seaweeds different types of seaweeds. Alginic acid this is an ionic linear copolymer, this is anionic because we can form C O O minus because the H can come out. So, it is got; it is called M block, this is got a G block, G is the guluronic acid, the M block is mannauronic acid. So, they are arranged G G, M G, M M different combinations of it actually. So, the G G is stiff more soluble at lower p H than G M, G G is stiff more soluble. That is why it suitable for a biomedical because it is stiffer. G content varies from 40 to 70 percent, the molecular weight we are talking in terms of 50 to 100, 000 Dalton.

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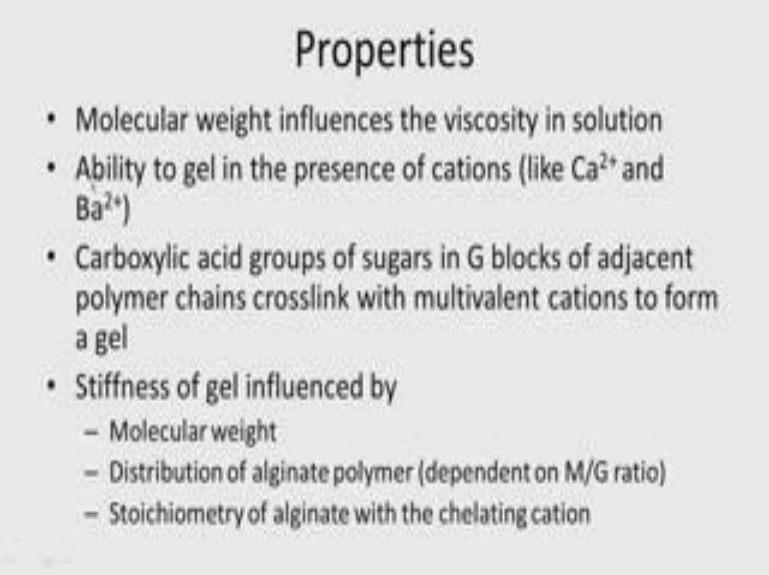

Molecular weight influences the viscosity in solution. So, depending upon the molecular weight viscosity varies, higher the molecule weight it becomes more viscous. So, they can gel in the presence of cationic like calcium 2 plus and barium 2 plus. Why is that? Because we have C O O minus, so they can nicely take in different cations. The carboxylic acid groups of, sugar in G block of adjacent polymer chains cross linked with these cations and forms the gel. So, the stiffness of the gel depends on the molecular weight, distribution of alginate polymer; that means, depending upon the M by G ratio, stoichiometry of alginate with a chelating cation. So, that is how the stiffness looks like actually.

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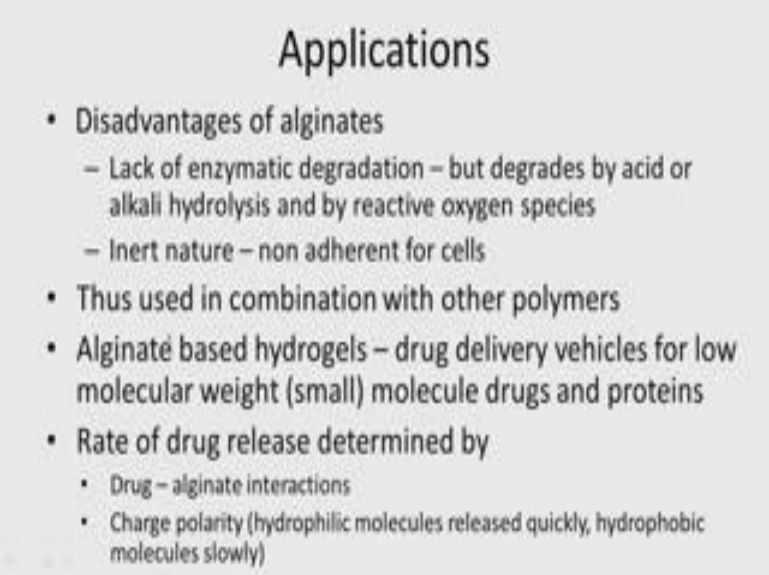

So, that what are the disadvantages? Lack of enzymatic degradation, there were no enzymes which can degrade this particular material that is one thing, but we can use acid or alkaline hydrolysis and then reactive oxygen species. So, it is got inert nature. So, the cells do not attach to that. They have to be used in combination with other polymers. So, alginate based hydrogels can be used for drug delivery vehicles for low molecular weight drugs and proteins. So, the trade of drug release depends on the drug alginate interaction, charge polarity hydrophilic molecules released quickly, hydrophobic molecules get released very very slowly. So, that is the difference between these actually.

So, they have some disadvantages because they are inert cells do not get attached to that. So, we need to use with other biopolymer or even a synthetic hydrophilic polymers. So, we will continue further on these biopolymers in the next class also.

Thank you very much