Regeneration Biology
Rajesh Ramachandran
Department of Biological Sciences
IISER Mohali
Week: 12
Lecture: 61

W12L61_Future implications of regeneration in mammals

Hello, everyone. Welcome back to another session of regenerative biology. In today's topic, we will learn about the future implications of regeneration in mammals. So in today's class, our focus will be to understand what the future angles or implications of regeneration research using mammals are, and how they will bring us a step closer to human tissue regeneration, because humans are also mammals, and we use mice and other organisms as models. Adopt the strategy of regeneration in humans. So that is the overall theme of the so-called regeneration biology, or regenerative medicine, etc.

So, fortunately or unfortunately, the mammalian model that is extensively used in laboratories is mice. Of course, rats, guinea pigs, rabbits, or even monkeys are also there. The majority of the research is oriented around Mus musculus, which is normally called laboratory mice, and that is what we use for our regeneration studies. However, this animal does not have regenerative ability, so that is why it becomes a little challenging to study proper regeneration or an actual mechanistic study of regeneration in the Mus musculus model.

Therefore, we will see what future implications hold, and let us quickly overview the future research into mammalian regeneration, which holds promise of transformative medical advances, potentially enabling the repair or replacement of damaged tissues and organs, particularly those with limited or no regenerative capacity, such as the heart or brain. We do have a regenerative capacity for hair, nails, skin, etc. And also, one major organ we regenerate is the liver. So we know that very well. Other complex organs such as the brain, spinal cord, retina, etc.

Do not have or even heart do not have a proper regenerative capacity. That is becoming one of the major targets for regeneration biologists. So understanding the molecular and cellular mechanisms underlying regeneration in other animals and identifying factors that constrain regeneration in mammals can lead to the development of novel therapeutic strategies. Having said that, there are very well-regenerating models such as zebrafish and axolotls, etc. are effectively used to copy or simulate this mechanism in mammals and have yielded promising results.

What it tells us is that the regeneration mechanism or the way in which regeneration can take place does not change too much across different species. All we see is that some species, such as fish and frogs, have these mechanisms in place. Properly orchestrated manner, they are in place; whereas in mammals, say mice or humans, despite having all the options for proper regeneration and all the genes responsible for regeneration, their expression is not adequate, or their temporal regulation is not efficient. This has led to poor or lack of regenerative ability in these mammalian models, so let us see how we can revolutionize regenerative medicine. Treating currently untreatable conditions often looks forward to regenerative biology, so the ability to induce regeneration in mammals would.

Revolutionize the treatment of conditions like heart failure and neurological injuries such as stroke or spinal cord injury, where the current therapies are very limited, so overcoming the organ shortage is another approach we have to pursue in this regenerative biology, because we do not have adequate organs, whether they are cultured organs, donor organs, or cadaver organs for the heart. Organ regeneration could be achieved; it could alleviate the strain on the organ donor pool. Developing novel therapies is another approach to the immediate prospect of regenerative biology, as researchers can develop targeted therapies to stimulate it. Tissue repair and regeneration involve understanding the molecular pathways involved in regeneration. That is another approach.

Now looking into unlocking the secrets of regeneration, we will find a few interesting facts. That is understanding the blastema. We know what blastema is by now. I will not go into the details, but all we know is that blastema is a prelude to a normal regenerative response. If there is no blastema, you end up with a wound healing.

You do not end up getting a regeneration. The blastoma is specialized tissue that forms during regeneration in some animals and is a key area of research in the field of regeneration biology, studying mammalian blastema like those found in regenerating rodent digits or ear pinnae; two models used for regeneration in mice and rodents in general are either cutting the toe tip or cutting their ear pinnae, and both types of regeneration can be studied. They can provide insights into how these structures are formed, how they drive regeneration, or how they do not drive regeneration properly. By identifying therapeutic targets through the analysis of molecular signals and cellular events during regeneration, researchers can identify specific molecules and pathways that could be targeted for therapeutic intervention. So the understanding factors can also be a limiting point of regeneration; these are the mechanisms that limit regeneration in mammals, such as wound contraction.

We have seen that soon after a wound is formed, it contracts because of the cytoskeletal and other mechanical forces that come into play. Wound contraction is followed by immune responses and changes in the microenvironment; they are also active areas of research. Can we prevent wound contraction? Can we prevent unwanted immune cell migration, excessive collagen deposition, etc.? If we can, can we think of overcoming these challenges? These are all active areas of research. Modifying these factors could potentially enhance the regenerative capabilities in non-regenerative animals such as mammals.

So, pushing the boundaries of epimorphic regeneration. We know that epimorphic regeneration is the most desirable regenerative mechanism a species can have. So can we push the boundaries of epimorphic regeneration? Extending the regeneration beyond the limbs, while some mammalian tissues such as skin and hair exhibit their regenerative capabilities, the focus is on expanding regeneration to more complex tissues and organs, so this is an active area of research. This could involve stimulating the formation of blastema that somehow does not form; can we stimulate it? In other tissues and using techniques like tissue engineering to promote regeneration. So can we think of engineering regenerative tissues? So researchers are now exploring the use of biomaterials and stem cells to create scaffolds or 3D-printed organs that can be implanted to promote regeneration.

So these are also an active area that is ongoing in the field of regeneration biology. Harnessing the body's natural healing process is the most attractive prospect simply because we have discussed many times that even though you can make an organ in a Petri dish, if its blood vessels or proper angiogenesis do not intercalate with the recipient's or host's blood vascular system, the future of this organ is grim. It is a perfect organ; if the body did not nurture it, then it is less likely to be a successful organ. Hence, it becomes very important to harness the body's natural healing process. If you can allow the healing or the fixing to happen, side by side in the same organism itself, although it may be time-consuming and cumbersome, you don't need to worry about whether there will be enough oxygen supply or blood supply.

That is why the natural healing process has become more attractive. Researchers can develop synergistic therapies with these processes to enhance tissue repair and regeneration by understanding how the body naturally heals and regenerates. So, the ultimate goal of tissue regeneration biology is about natural healing. All we need is some triggering or stimulation in those animals that do not regenerate effectively. So your goal is to get into that area.

So now, as with any other topic, we have discussed many times that the ethical and societal implications are also very important. It is so important that sometimes even the research itself can come to a U-turn or it can come to a stalled state simply because you are not meeting the ethical considerations of that particular research addressing the safety concerns as regenerative therapy becomes more advanced, addressing potential safety concerns such as the risk of tumor formation or other unintended consequences. It is crucial that any approach you adopt, you have to be sure that this. A patient who received this treatment is not going to die of cancer now because of this treatment; it has happened in the past because of the gene therapy. I think it is done to treat hemophilia, the blood clotting disease.

Those two treatments done on the children ended up developing leukemia; hence, the gene therapy became a complete failure. The standing still situation happened because the approach can make them vulnerable to developing cancer, as you are dealing with cell proliferation, and whenever you see unwanted cell proliferation in your body, that is a red flag for the organism, which means that it is favoring the formation of cancer. Ensuring equitable access. This is another thing that everyone should have access to such technology. Regenerative therapy should be accessible to all individuals, regardless of socioeconomic status or geographic location.

So it shouldn't happen that the layman or financially underprivileged people are deprived. That is another serious ethical angle or societal implication that comes into the picture. Considering the long-term impacts, the long-term effects of regenerative therapy need to be carefully considered, and the research should focus on developing therapies that have a lasting positive impact on patient health and well-being. In conclusion, the future of mammalian regeneration holds immense promise for transforming healthcare. Understanding the underlying mechanisms of regeneration and developing novel therapeutic strategies make it possible to unlock the body's natural healing potential and revolutionize the treatment of a wide range of diseases.

That is why the future angle or the future implication should be considered very carefully. Let us now look at how effectively or how efficiently you can transform or modulate the so-called field of regeneration biology using other mammalian models. Do we have other mammalian models that have better or the best available regenerative capacity? So Van Beijnum, a scientist, explains that one model organism called the spiny mouse is a fascinating model for studying regeneration. So by examining the unique characteristics of this animal, the spiny mouse, which is normally seen in very dry and desert conditions, we will gain a better understanding of the possibilities for developing therapies to improve wound healing in humans and increase the likelihood of full recovery. This is the picture of the spiny mouse, which resembles an ordinary mouse except that instead of hair, it has spiny spines on its back.

Its tail also has some spines, and it has excellent regenerative capacity. Instead of Mus musculus, its specific generic name is Acomys, and it has around 20 different genera. Many of them have regenerative capacity, but not all of them. One of the commonly used ones is Acomis kempi. So many species are found and are normally seen in Africa, Brazil, etc.

So let us see a few stories of regeneration in a spiny mouse. A team of researchers at the University of Kentucky and Cincinnati Children's Hospital is delving deeper into the science behind how spiny mice can regenerate lost tissue, using what they learn to trigger regeneration in other types of mice. When other types of mice fail to do what this spiny mouse is able to do. So, advances that one may have as you keep looking into the research will one day allow us to translate the information learned from the spiny mouse for human use. Adult laboratory mice heal injuries with scar tissue, while the spiny mouse can regrow lost skin and regenerate musculoskeletal tissues, including the spinal cord.

So it can regenerate complex tissues; that is the beauty, not just about regenerating some epithelium, skin, or intestine, but that we also have the ability. Making a complex tissue regenerate is more important than ever. One of the pioneer scientists is Ashley Seifert, and his research group has pioneered the use of spiny mice and other animal models to understand how complex tissue can regenerate, bridging regenerative biology and medicine. So the knowledge from spiny mice is going to revolutionize the field of regeneration biology. Let us see the science behind spiny mouse regeneration.

A study from Seifert's group focused on the cellular response to injury during tissue healing. So the study showed that how ERK signaling works. Acts as a key switch in balancing the healing response, and they have demonstrated that increasing and maintaining this type of signaling means more than usual, and not just more than usual sustained signaling; ERK signaling in the laboratory mice could trigger a regenerative response instead of scarring. So the message is that in spiny mice, ERK signaling is active and persistent throughout the regenerative response, whereas in laboratory mice, ERK signaling triggers but then disappears; hence, it is not regenerating but healing the wound with scar formation. Also, how specific immune cells respond to injury and directed tissue regeneration deserves proper attention to understand the involvement or to explore the involvement of specific immune cell types.

The scientists looked at a specific type of immune cell known as macrophages. We have discussed many times that there are two types of macrophages: M1 and M2 macrophages. M1 is pro-inflammatory; M2 is anti-inflammatory. During fin regeneration class, we discussed that very effectively and elaborately, so the macrophages, which play a crucial role in regulating the inflammatory response, can be pro-inflammatory or anti-inflammatory, depending upon the context. The macrophage you are discussing and the inflammatory response defend injured tissue against pathogens while simultaneously promoting tissue repair in both the spiny mouse and the lab mouse.

Let us further examine the macrophage phenotype between these two models of healing, namely Mus musculus and Acomys, specifically tissue regeneration and scar formation, as they differ. Tissue regeneration in spiny mice, scar tissue formation in Mus musculus, which is the laboratory mouse. While it is unclear to what extent macrophages ultimately control different healing trajectories, the observed subtle and distinct macrophage signatures aligned specifically with regeneration. It's just like people say; there is a saying that winners don't do different things, they do things differently. In that way, the macrophage is present in both.

But the gene expression, the factors that are expressed in the spiny mouse, is different from that of the laboratory mouse. Using identical injuries in both rodents means.

.. Mice, Mus musculus, and spiny mice. The researchers studied signals released by macrophages that communicated with nearby cells, telling them to rebuild all missing tissues. How does the building occur? During those processes, they also noted the cell characteristics or phenotypes and how the cells respond to the signals released by the macrophages. The study showed that the spiny mice macrophages release distinct proteins partially responsible for reforming specialized tissues at the injury site and protecting the cells from stress. That means it is helping the cell not to die, not to succumb to stress and injury, and also to make different types of cells because they release some unique factors. Overall, these experiments point toward the macrophages establishing a tissue microenvironment conducive to regeneration.

Researchers examined and compared the macrophages from the bone marrow of both species. Then they used RNA sequencing to identify the proteins that these cells secrete. RNA is a molecule that carries genetic instructions from DNA to make proteins; RNA is the intermediate. Researchers found that the macrophages from bone marrow responded differently on their own to a protein called interferon gamma (IFN gamma), which is a pro-inflammatory cytokine, and M1 macrophages can secrete it as well. A protein produced by cells in response to infection and lipopolysaccharide, a molecule found in bacterial lipopolysaccharide (LPS), stimulates the immune system; because of this stimulation, plenty of interferon gamma is produced at or is present at the site of injury.

In response to inflammatory cues, macrophages in the spiny mouse release unique communicating proteins that promote the growth of new blood vessels, lymphatic vessels, anti-inflammatory activities, and tissue rebuilding, which positively contribute to regeneration. The same molecule is also present in mice, but with interferon gamma, the spiny mouse undergoes these steps that form new blood vessels and other tissue rebuilding activities. To answer more scientific questions about the spiny mouse, researchers have found a specific type of protein called vascular endothelial growth factor C. The normal one is VEGF; if we say vascular endothelial growth factor, this is VEGFC, which is uniquely secreted by spiny mouse macrophages during regeneration. It also plays a multifunctional role, encouraging the growth of new blood vessels and lymphatic vessels.

Like I mentioned, the future of the newly formed tissues is only that much important or that much successful. As the blood vessel or lymphatic system is formed properly. Otherwise, the future of the newly formed tissues is grim. Through specific antibody blocking of the same VEGF-C protein in the ears of spiny mice, which are used as a model, you cut the ear pinna and look at whether it is regenerating. Researchers noticed notable changes in the formation of blood vessels, lymphatic vessels, and cell division.

So they found that if you block it, you don't get proper regeneration. So VEGF-C is needed. This led to a decrease in the new hair follicle formation, and the blocking of VEGF-C led to a decrease in new hair follicle formation and an increase in inflammation, ultimately disrupting tissue regeneration. So, if you block one factor, VEGFC released by the macrophage, you can enhance the regenerative ability in the spiny mouse. Additionally, we speculate that the researchers believe that the enhancement of the secretion of VEGFC and other factors by macrophages during fibrotic wound healing may be contributing to a regenerative outcome in tissue healing.

However, further experiments are required to confirm this idea. This is a smoking gun. You can prove this point, but one has to delve deeper to understand the exact mechanism. The study suggests that the macrophages in specific tissues throughout the body help direct and regulate the cellular repair programs.

So you may wonder how one molecule can play a role. We should understand that the wedge of C is a master player, provided there is its receptor and those cells respond in a different manner. It feels like I am giving a lecture now. Maybe you are listening. Maybe your pet animal is also listening.

You will understand what I am saying. But your pet animal will listen to it as if someone is speaking. It won't understand as clearly as a human being can understand. It simply means that the ability to respond depends on which cell responds. It is responding to that signal. Importantly, the results of this study show that the type of macrophage matters and suggest that altering what one type of macrophage secretes could alter how tissues repair themselves.

Of course, macrophages are important. What protein it is secreting is important, but it also matters which tissues are responding to that secreted protein from this macrophage. Another spiny mouse regeneration scenario we can look at. When comparing the tissue healing in laboratory mice and spiny mice, it was observed that adult laboratory mice generally heal injuries with scar formation. That is an injury. It is common in both, but in one species, wound healing is normally reflected by scar formation; in another, my spiny mouse, regeneration is scarless.

Spiny mice are known for their unique ability to regrow skin, especially to restore lost skin, and to restore functions of the severed spinal cord. The same scientist we were talking about found that previous studies show that repair of damaged heart tissue can effectively maintain heart function. It can regenerate even damaged heart tissues. The extracellular signal-regulated kinase ERK, which we discussed earlier, is a major cellular response to injury during tissue healing. ERK-mediated signaling acts as a key controller of the healing process in spiny mice.

The same group of researchers observed that ERK activity responded to tissue damage in both rodent species. ERK activity decreased in lab mice as the initial wound healing phase and tissue inflammation resolved. So, like I told you, ERK signaling is present in every species, and it is turned on in lab mice as well as in spiny mice. But the ERK signaling in Mus musculus, normal mice, is turned off. When the wound healing phase and the tissue inflammation phase are completed.

In contrast, the ERK signaling was maintained in a diverse array of cells at high levels in the spinal mass as new tissue was generated. Remember, wound healing is fast, and regeneration is slow. So these mechanisms that is helping for regeneration should be there for a prolonged time. As part of the same study, the researchers applied ERK activators to the ear of the lab mice, such as ear pinna cutting, which normally doesn't regenerate in lab mice, but they applied ERK signaling activators and achieved the same regenerative healing they observed in spiny mice. All they did was prolong the ERK signaling activity, and they could achieve a better regenerative response.

Researchers found that sustained high ERK activity is critical for regeneration. From this message, we can learn that just like VEGFC, if you block it, you don't get it. In the same way, sustained ERK signaling can favor the regenerative response in lab mice's ear pinna. That partially explains why some closely related species, like spiny mice and lab mice, may have very different cellular responses to the same injury.

It is quite surprising. In this spiny mice regeneration research, researchers asked whether poor regenerators, namely laboratory mice and even humans, are not laboratory animals, but our interest in regeneration research is to help humans. Have they completely lost their ability to regenerate or do they still possess their latent regenerative traits? This is a million-dollar question. If the latter hypothesis holds true, that means we still have this ability to regenerate; it would have enormous implications for regenerative medicine, implying that a regenerative response can still be triggered in the cells and tissues of poorly regenerating adult mammals and ultimately can benefit humans. So we should understand that this regenerative ability can be modulated across species to achieve the ultimate or most desirable regenerative capacity. So we will study regeneration biology with a different topic in the next class. Thank you.