Tissue Engineering: A Myriad of Concepts

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What do the movies Star Wars, Robocop, and Kingsman have in common? All of them portray characters with bionic limbs. The movies called for events that facilitated the necessity of a new limb or new organs for a character. While this makes for a great story line, most people who require organs or prosthetic limbs do not wish for flashy replacements but rather fully-functional replicas that allows them to go through everyday life as if there was no change to their bodies.

I was in middle school when I first saw a mouse with a human ear growing on its back. It looked like something out of science fiction and stuck with me. A few years later, I saw a 3D-printed ear being grown in a petri dish; the mouse had, literally, been cut out of the equation. In only a few years, the technology had made the movement from live animal hosts to 3D-printers, thereby changing the future of the field of tissue engineering. I was captivated and announced to everyone that I wanted to be a biomedical engineer. Throughout middle school and high school, I was determined to learn as much science, technology, engineering and math as I could, taking additional courses in those subjects at local colleges, through the Center for Talented Youth, at the Cold Spring Harbor Laboratory DNA Learning Center and also online. I read articles on materials science, 3-D printing of organs and tissue engineering in research journals and science magazines. I read about the future of 3D-printers in which multiple materials could be printed into one final product with the same printer. I studied the obstacles of using stem cells versus a person's own adult cells to create organs. I researched the possibility of 3D-printing bone and cartilage instead of just soft tissue. My goal was to combine my interests in science and technology with learning more about this dynmic field so that one day, I would be able to conduct independent research in a lab that was already researching these topics. Equipped with AP science courses, laboratory skills and computer skills, my next step was to search for, and apply to labs which had summer research programs.

After being accepted to several illustrious labs, I chose to conduct research at the Garcia Center for Polymers at Engineered Interfaces at Stony Brook University which is a Materials Research and Science Engineering Center (MRSEC). There are several of these MRSECs at academic institutions across the nation. At Garcia, I created my own experiment to develop a bio-ink made with hyaluronic acid, that could be used in a 3D-printer to develop hydrogel scaffolds. These scaffolds could then be used to support the growth of cells, such as dermal fibroblast cells to create sheets of skin or dental pulp cells to create new teeth.

My programming skills were being applied in a new way, the design of a 3D product with structural support, rather than for designing an algorithm. My science and math skills were being applied in a relevant way to the creation of a new product. As I continue my education in math, science and computer science at Caltech, I have acquired a new perspective on how the work I put into those studies will be applied to whatever research I may conduct in the future.

Any high school student interested in science research should learn as much math, engineering and computer science as they can. The application of these studies to science has had a major impact and created new fields of research such as bio-informatics. Science research requires knowledge not only in biology and chemistry but also statistics, geometry and computer programming. It is important to learn as much as possible not only in school but also in other environments. Those additional educational experiences provided me with crucial perspectives and skills.

Overview of my Research:

3D-Printers have slowly become more commonplace as they become cheaper and smaller. Makerspaces and libraries have made them more accessible for the average consumer to use, normally to print something small and made only of single colored plastic. 3D-printers have become prominent in many fields, most notably, tissue engineering. My research focused on 3D-inkjet printers, which unlike most 3D-printers use a liquid ink, not a plastic. This allowed me to use the 3D-printer to create a specific shape while keeping the final product soft enough to resemble tissues and support cell growth. The bio-ink was made with a polymer called hyaluronic acid (HA) which is a key component of extracellular matrices for supporting muscle structure. I believe that using HA as a base polymer will make the printed result capable of being modified with various chemicals for the creation of different tissue types. As the bio-ink is printed, a UV light activates a photoinitiator in the bio-ink which causes what has printed to become a gel. This is what allows the 3D object to be built upwards because the lower levels are solid enough to support the next layer of ink. The main issue with using the 3D-printer was that the bio-ink must be about the viscosity of water to be compatible with the printer, however, HA makes a very thick solution. In addition, while I needed to lower the viscosity of the bio-ink, it must still be capable of creating a solid enough gel to support cell growth. First, the problem of viscosity was solved with the alteration of HA's molecular weight which is exponentially related to the viscosity of the solution it creates, as seen in the Mark-Houwink equation: $\eta=KM^a$, where

η is the intrinsic viscosity of a material, M is the molecular weight, and a and K are constants dependent on the polymer-solvent system. Commercially available HA is normally found at over 1 milliDalton, which is similar to what's commonly found in the body. However, this generated an ink over a hundred times too viscous to be compatible for use with the printer. I was put in contact with a lab at New York University which was producing HA at only 20 kiloDalton. I found that this HA made a solution that was a compatible viscosity for the printer. Secondly, I faced the issue of keeping the gel strong enough so that it would gelate quickly enough for another layer to be printed on top of it. I ran tests initially without the 3D-printer to see if the gel would set and altered the chemical makeup of the bio-ink. Once I had a working sample of bio-ink, I contacted the 3D-printer company, ChemCubed, which allowed me to use their labs and printers to test my samples of bio-ink. The amount of time it took for the gels to set sufficiently ranged from 2 minutes under UV light to over an hour. This remains the biggest source of error in my research and I have done additional research to address the issue in my future work. The final test was to make sure that the gels were safe for use with cells. After the gels had been solidified, they were prepared for cell culture by swelling them in media and then plated with dermal fibroblast cells. I found that while all molecular weights of HA were nontoxic to cells, only the high molecular weight cells were conducive to cell adhesion. So while none of the samples would actively kill the cells that came in contact with them, the low molecular weight samples did not allow cells to adhere to and grow on the gels. While this is not the outcome I was hoping to have, the low molecular weight, non-adhesive gels still have applications in the medical world. The high molecular weight samples are applicable for the recreation of a tissue or organ.

Real World Applications:

After hearing about some lectures, I discovered that my nontoxic, non-adhesive gels had applications in spinal surgery. When any surgery is done that requires healing near the spinal cord, there is the threat that scar tissue could fuse with the spinal cord and potentially cause paralysis. My gels could be applied between the spinal cord and the surrounding tissue to prevent adhesion during healing while remaining nontoxic to the body. This way the body could heal with the two areas separated and there would be no chance of fusing. After the body had healed, the gels would deteriorate and leave the body unchanged.

The adhesive gels on the other hand, have the potential to become scaffolds and eventually tissues. In the future, the goal is for the gels to be seeded with a person's own stem cells or adult cells to develop a specific tissue or organ the person needs. I tested dermal fibroblasts which could develop the gel into a piece of skin. This could be used with burn victims who need skin grafts. Instead of taking a skin from another part of the patient's body, only a few healthy cells would be needed for growth on a gel in the desired size and shape. The healthy cells from the human would be plated on a specially made gel, and a short period of time later, the patient would be give new skin in place of the burn. This would reduce the amount of surgeries the patient needed to go through and the number of scars they would accumulate. It would also reduce the amount of time they need to stay in a hospital. Eventually, it will be possible to create organs instead of just tissues. I believe as 3D printer technology advances, researchers will be able to print different types of ink into the same gel which could develop into one organ with multiple cell types growing in it. If this becomes possible it will help the hundreds of people who are waiting for organ donations. It will also prevent these patients that need organ donations from needing to take drugs to suppress their immune system and eliminates the possibility of rejection from the patient's body because the organ will be made with their own cells.

Future Plans:

For future research, I would like to explore the unique scientific capabilities of the 3D-printer in depth. I'd like to expand the types of cell that are compatible with the gels by changing the porosity and topography of the gels. For example, dental pulp stem cells grow well in cylindrical spaces. I started to design samples with an open-source 3D modeling software called Blender; each gel contains thousands of tiny cylinders which would support cell growth. I hope to make the gels easily alterable with chemicals to increase the range of their usage. Long term, this would allow one base gel to be changed into many different gels and a wider array of treatments could be addressed.

I plan on continuing research in tissue engineering in the future. I believe the possible improvement in many people's lives will be realized within the next few decades and the creation of 3D-printed tissues will completely change medicine and healthcare. I hope to be a major contributor to that change.