Philippine Science High School - Southern Mindanao Campus

Science Fair 2023

Spinning life lines:

Utilization of Cotton Candy Machinery In Artificial Living Tissue Production

Submitted by:
Acuña L. L. , Arenas A. J., Bicbic M. J., Follante K. Y., Parreno A. J., Seblos A. G.

Brief Description

Advancements on human life development have increasingly been productive, finding ways to create sustainable and efficient solutions to past, current, and future problems using innovations such as machinery. Through machines, work can be done more efficiently in greater qualities and quantities. The products from these may range from windmills up to cotton candies and even artificial human tissues.

Through utilizing the machine used to make cotton candy, scientists were able to recreate the perfect mold to replicate capillaries. With the Rotary Jet Spinning and centripetal motion of this round machine, the replication of polymer microfibers which are used to give spaces in the capillaries inside the tissues was made possible. Afterwards heat is applied, the added sugar in the machine is heated up and broken down into glucose and fructose, releasing water yet increasing interactions between the molecules and soon solidifies. The production of artificial tissues possess a similar objective into making lab-grown meat wherein natural hydrogel is used as one of the major materials. Since natural hydrogels containing collagen and gelatin have higher water absorption capacity and greater durability, this would lead to longer lastingness and greater probability of success in a transplantation of an artificially made organ rather than synthetic hydrogels.

Within this paper, the ability of the cotton candy machine to replicate artificial tissues using natural hydrogel and sugar as they undergo the processes relating to biology, chemistry and physics is highlighted. Through the use of Rotary Jet-Spinning (RJS) machinery, current tissue production techniques, and artificial laboratory meat growth techniques, these replicas seek to discover efficient and sustainable tissue engineering. Battling the climate crisis, and the current ethics of the organ market and meat industry finds ease within laboratory living tissue replicas given the process's sustainability and ease of production.

Related Scientific Concepts

Tissues within animal bodies serve multiple purposes to create a harmonious system for the organism to function and to live. These animal tissues are categorized into the following: epithelial, connective, muscle, and nervous tissue. Each of these tissues differ in function and composition, but all remain as a group of cells with similar functions. Additionally, tissues of any kind would not be able to function if there is no constant supply of oxygen. Capillaries do such as they carry oxygenated red blood cells (RBCs) into different parts of the body. Without such a network of the circulatory system, tissues and its composing cells would cease to function.

Cotton candy and its fibers remain quite small, and with the proper machinery involved creates uniform sizes for all the fibers it produces. A single cotton candy fiber's diameter ranges from 10-100 μm (Wu et al., 2019). Close to that, most capillaries, at general maximum, have a diameter of 10 μm. Therefore, using cotton candy machinery can help attain the perfect mold for capillaries to form.

The electric candy machine by Dr. William Morrison and John C. Wharton was the first introduction of cotton candy production. As sugar is heated up in the middle, it breaks down into glucose and fructose. Such breakdown releases water from the sugar and creates increased interactions between the two forms of such sweetener. Its reduction to a syrup and the centrifugal force of the head spinning at a rate of 3,400 revolutions per minute pushes the liquid to the tiny holes and solidifies as it passes through, leaving thin cotton candy fibers throughout the container; solidification occurs quickly in which a disorganized, amorphous solid is formed (Rupp, 2016).

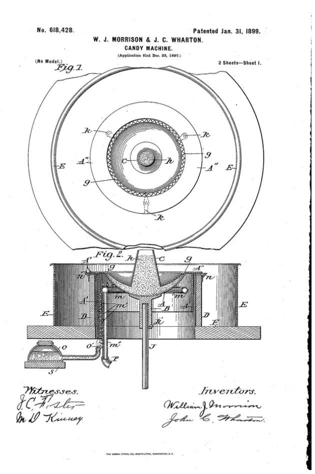

Figure 1: Electric candy machine patent made by Morrison and Wharton (1899)

For this matter, polymer microfibers are what help create the desired spaces for the capillaries to settle. Such microfibric molds can be achieved through Rotary Jet-Spinning (RJS). RJS's mechanism is quite similar to that of the electric candy machine of Morrison and Wharton. The composition of the sugar used for cotton candy production is then replaced with a polymer solution in which the same mechanism of centrifugal force pushes the liquid solution out of the spinning head and through the hole (Burrows, 2016). The solvent evaporates thus cooling down the polymer and creating the desired polymer microfibers.

Termed rotary jet-spinning, fiber morphology, diameter, and web porosity can be controlled by varying nozzle geometry, rotation speed, and polymer solution properties. We demonstrate the utility of this technique for tissue engineering by building anisotropic arrays of biodegradable polymer fibers and seeding the constructs with neonatal rat ventricular cardiomyocytes. The myocytes used the aligned fibers to orient their contractile cytoskeleton and to self-organize into a beating, multicellular tissue that mimics the laminar, anisotropic architecture of the heart muscle. This technique may prove advantageous for building uniaxially aligned nanofiber structures for polymers which are not amenable to fabrication by electrospinning (Badrossamay, et al., 2010).

Figure 2: Polymer fibers formed through RJS machinery in Vanderbilt's Leo Bellan's lab (Vanderbilt, 2015)

Moreover, the cast of the polymer microfibers should mimic or positively interact with the host body. The use of hydrogels as the main cast material makes sense given its composition given that natural hydrogels are used. Macromolecules may be seen such as animal collagen, plants and seaweeds; which are polysaccharides and proteins (Ahmed, 2015). The variety of its natural chemical composition creates multiple reasons as to why such material is used for the "scaffolding" of this regenerative tissue structure. Compared to synthetic hydrogels, natural hydrogels have higher water absorption capacity and long service life, which both qualities enhance the likelihood of success when any organ transplantation occurs (Ahmed, 2015). Synthetic hydrogels have water absorption capacities exceeding that of what humans are capable of intaking given the most extreme heat and humidity is approximately 1L (Born, n.d.). Its water absorption capacity is at 1200 g (d-water/1g hydrogel) (Elsaeed et al., 2022) ; while natural hydrogels can be regarded as less than that given its polysaccharides which are generally insoluble in water (Omega, 2022). The natural hydrogel cast is only temporary as the tissue cells take over the cast and create a fully functioning tissue; thus using natural hydrogel to maintain hydration the cast turns into a living tissue. These points are only within the scope of human regenerative tissue composition, as overall creating artificial animal tissues would require higher water absorption capacity to ensure quality and as to not dehydrate the tissue when subjected to prolonged air exposure.

Furthermore, knowing the materials to be used is quite an important step when it comes to these kinds of research. Two scientists from different groups, Leon Bellan and Kit Parker, used different materials for their research.

Summarizing one of Bellan's interview "VU INSIDE: Cotton Candy and Artificial Blood Vessels" (2015) on their research and one of Parker's articles "Nanofiber Assembly by Rotary Jet-Spinning" (2010) on their research as well:

Leon Bellan's group has used gelatin hydrogel for their production of artificial blood vessels (Bellan et al., 2012). Meanwhile, Kit Parker's has used a variety of synthetic and naturally occurring polymers for their production of artificial muscles, potentially producing more than artificial muscles in the future (Badrossamay, et al., 2010).

Leon Bellan's group used hydrogel, a polymer, for some of the following purposes:

➢ Accurately produce artificial blood vessels, more specifically, capillaries.
➢ The material also mimics sugar when making cotton candy using a cotton candy machine.
➢ Easily control the production by changing the pH of the fibers to make them dissolve and take on the required structure.
➢ The material used with the technique allows the transportation of soluble entities via diffusion and advection, reporting improved diffusion and transportation. This extends to other applications or functions as well (Bellan, Kniazeva, et al., 2012).

On the other hand, Kit Parker's group (Parker's Disease Biophysics Group, or PDBG) uses a variety of synthetic and naturally occurring polymers for some of the following:

➢ The range of polymers allows a wider range of potential applications of the machine and the products it can produce, ranging from but not limited to: ethical food (e.g., lab-grown or vegetarian meat), artificial organs, surgical implants, wound healing, and body armor (Goisman, 2022).

- ➢ The polymers are suitable for the machine they used and the objects they produced in their research (Badrossamay, et al., 2010).
- ➢ Mainly due to the technique of RJS, the capabilities of these polymers are greater than what other methods could have (e.g., self-assembly, phase separation, and electrospinning) (Badrossamay, et al., 2010).

Importantly, the polymers are spun into nanofibers due to the advantages they gain over microfibers. These advantages range from better filtration to greater surface area, tunable porosity, enhanced properties and toughness, and many others (Rogalski et al., 2017).

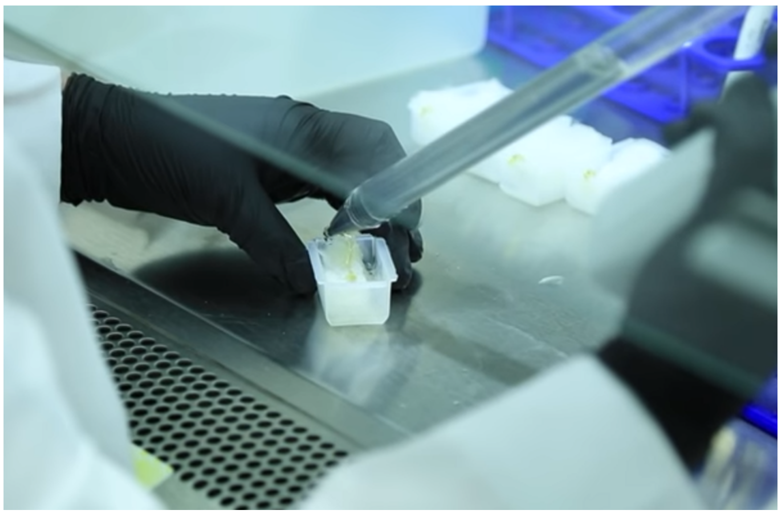

Figure 3: Hydrogel is poured on the polymer fibers to create a "cast" (Vanderbilt, 2015).

Additionally, animal tissue production can be a tool to create artificial animal tissues of any kind. Commonly within these types of tissue production, Fetal Bovine Serum (FBS) is used. It is a serum used in animal cell culture media as it allows stem cells to continue living without detecting any abnormalities or signs of death (Jochems et al., 2002). Such serum is used as well in the field of artificial meat production given its ability to sustain living stem cells and including the attributes of animal cell culture media. However, the extraction of FBS raises ethical concerns amongst researchers. A bovine fetus's blood is drawn from pregnant cows during slaughter (Jochems et al., 2002). Thus, this methodology of tissue engineering used with the current methods of growing artificial laboratory meat do not coincide with animal ethics given that it essentially is similar to buying regular meat.

Application and Outputs

Patients with terminal organ failure can be saved through solid organ transplants, which can enhance the quality of life. Organ transplants have steadily improved over the past 20 years and often provide great outcomes in children and young adults, but they are becoming increasingly difficult due to the rising number of older transplant patients who have comorbid conditions. In contrast to dialysis, renal transplantation enhances patient longevity, and irreversible disorders of the liver, heart, or lungs must be treated with life-saving transplants. The activity of solid organ transplant programs has been continuously increasing, although it still falls short of worldwide demands and varies greatly between nations. Transplanting solid organs is crucial for advanced and established healthcare systems (Grinyo, 2013).

The number of organ transplants conducted today exceeded 42,800 in 2022, with yearly highs for kidney, liver, heart, and lung transplants (Organ Procurement and Transplantation Network, 2023). Nevertheless, despite all of these procedures, the possibility of unanticipated failures still exists. This is where the creation of artificial blood vessels is useful because they can enhance the circulation of blood from the body to the new organ and increase the rate of regeneration of damaged cells. A significant issue, however, arises in the manufacture of these vital pathways because, first, they are difficult to manufacture due to their small stature, and second, in order for these manufactured channels to mimic real blood vessels, they need to have an inner lining of endothelial cells to control exchanges between the bloodstream and surrounding tissues (Alberts et al., 2002).

But thanks to innovative discoveries found in the field of regenerative medicine, researchers have found that rotary jet machinery, such as the ones used in making cotton candy, can be astonishingly helpful in creating artificial pathways for blood as previously discussed above. And if researchers and scientists keep building on their existing discoveries, they may be able to employ artificial blood arteries for purposes other than organ transplantation such as angioplasties or to redirect the blood flow from damaged arteries. Artificial blood arteries can also become test subjects for studying the effects of drugs or new medical devices on arteries.

With the creation of artificial blood arteries, scientists no longer have to rely on donated human or lab rat arteries to test drugs and equipment. Researchers can test the devices in vitro to determine how they interact with blood vessels, and how they affect blood flow and pressure. Additionally, they can be used to test new surgical techniques for vascular diseases. For example, doctors may want to study the effectiveness of minimally invasive procedures for treating atherosclerosis or aneurysms. By using these artificial arteries, they can stimulate the effects of these procedures on blood vessels before testing them on live patients.

Additionally, the innovation of lab-grown meat can be expounded using this methodology of tissue engineering. Similarly, the problems of inner endothelial cell lining needs to be met for such tissues to be fully functional. The ethical problem of the current system of producing lab-grown meat through the use of FBS also needs to be addressed. But, further advancements within this study of artificial tissue production can aid environmental problems related to grazing and greenhouse gas emissions through eliminating the necessity of large scale meat farms, given that this type of lab-grown meat will be mass produced. Therefore, this research on artificial tissue engineering can be a tool for climate change mitigation as livestock farming produces 15% of the current emissions — with comparable percentages to transportation (Wilde, 2022).

Bibliography

Ahmed, E. M. (2015). Hydrogel: Preparation, characterization, and applications: A review. *Journal of Advanced Research*, *6*(2), 105–121. https://doi.org/10.1016/j.jare.2013.07.006

Alberts, B., Johnson, A., Lewis, J., Raff, M., Roberts, K., & Walter, P. (2002). Blood Vessels and Endothelial Cells. *Molecular Biology of the Cell. 4th Edition*. https://www.ncbi.nlm.nih.gov/books/NBK26848/#:~:text=Endothelial%20cells%20form%20a%20single

Badrossamay, M. R., McIlwee, H. A., Goss, J. A., & Parker, K. K. (2010). Nanofiber Assembly by Rotary Jet-Spinning. *Nano Letters*, *10*(6), 2257–2261. https://doi.org/10.1021/nl101355x

Bellan, L. M., Kniazeva, T., Kim, E. S., Epshteyn, A. A., Cropek, D. M., Langer, R., & Borenstein, J. T. (2012). Fabrication of a Hybrid Microfluidic System Incorporating both Lithographically Patterned Microchannels and a 3D Fiber-Formed Microfluidic Network. *Advanced Healthcare Materials*, *1*(2), 164–167. https://doi.org/10.1002/adhm.201100052

Bellan, L. M., Pearsall, M., Cropek, D. M., & Langer, R. (2012). A 3D Interconnected Microchannel Network Formed in Gelatin by Sacrificial Shellac Microfibers. *Advanced Materials*, *24*(38), 5187–5191. https://doi.org/10.1002/adma.201200810

Born, S. (n.d.). *Hydration - What You Need to Know*. Hammer Nutrition. Retrieved May 1, 2023, from https://hammernutrition.com/blogs/advanced-knowledge/hydration-what-you-need-to-know#:~:text=At%20the%20most%2C%20you%20can

Burrows, L. (2016, October 7). *A new spin on nanofibers Techniques offer better, tunable production of materials for bulletproof vests, cellular scaffolding*. Www.google.com; Harvard School of Engineering. https://seas.harvard.edu/news/2016/10/new-spin-nanofibers#:~:text=Rotary%20Jet-Spinning%20(RJS)%20works%20likes%20a%20cotton%20candy

Elsaeed, S. M., Zaki, E. G., Abdelhafes, A., & Al-Hussaini, A. S. (2022). Response Surface Method Based Modeling and Optimization of CMC-g Terpolymer Interpenetrating Network/Bentonite Superabsorbent Composite for Enhancing Water Retention. *ACS Omega*, *7*(10), 8219–8228. https://doi.org/10.1021/acsomega.1c03194

Goisman, M. (2022, March 15). *Making steak from vegetable protein*. Seas.harvard.edu; Harvard School of Engineering. https://seas.harvard.edu/news/2022/03/making-steak-vegetable-protein

Grinyo, J. M. (2013). Why Is Organ Transplantation Clinically Important? *Cold Spring Harbor Perspectives in Medicine*, *3*(6), a014985–a014985. https://doi.org/10.1101/cshperspect.a014985

Jochems, C. E. A., van der Valk, J. B. F., Stafleu, F. R., & Baumans, V. (2002). The use of fetal bovine serum: ethical or scientific problem? *Alternatives to Laboratory Animals : ATLA*, *30*(2), 219–227. https://doi.org/10.1177/026119290203000208

Morrison, W., & Wharton, J. (1899). *Candy Machine*. https://patents.google.com/patent/US816114

Organ Procurement and Transplantation Network. (2023, January 10). *2022 organ transplants again set annual records; organ donation from deceased donors continues 12-year record-setting trend - OPTN*. Optn.transplant.hrsa.gov; U.S. Department of Health and Human Services. https://optn.transplant.hrsa.gov/news/2022-organ-transplants-again-set-annual-records-organ-donation-from-deceased-donors-continues-12-year-record-setting-trend/#:~:text=In%20brief%3A

Rogalski, J. J., Bastiaansen, C. W. M., & Peijs, T. (2017). Rotary jet spinning review – a potential high yield future for polymer nanofibers. *Nanocomposites*, *3*(4), 97–121. https://doi.org/10.1080/20550324.2017.1393919

Rupp, R. (2016, July 15). *The Sticky-Sweet Story of Cotton Candy*. Culture. https://www.google.com/url?q=https://www.nationalgeographic.com/culture/article/the-sticky-sweet-history-of-cotton-candy&sa=D&source=docs&ust=1682949730948418&usg=AOvVaw31TLLSgonU5kTqKqcxf4oO

Vanderbilt University. (2015, March 27). *VU INSIDE: Cotton Candy and Artificial Blood Vessels*. Www.youtube.com; Vanderbilt University. https://www.youtube.com/watch?v=Yf3JIaFxZVQ

Wilde, W. (2022, October 30). *Fact check: How bad is eating meat for the climate? – DW – 10/30/2022*. Dw.com. https://www.dw.com/en/fact-check-is-eating-meat-bad-for-the-environment/a-63595148#:~:text=It%20can%20be%20concluded%20that

Wu, Y., Ye, K., Liu, Z., Wang, B., Yan, C., Wang, Z., Lin, C.-T., Jiang, N., & Yu, J. (2019). Cotton Candy-Templated Fabrication of Three-Dimensional Ceramic Pathway within Polymer Composite for Enhanced Thermal Conductivity. *ACS Applied Materials & Interfaces*, *11*(47), 44700–44707. https://doi.org/10.1021/acsami.9b15758