

Synthesis of Cellulose by *Acetobacter Xylinum*:
A Comparison Vegan Leather to Animal and Imitation Leather

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Abstract

Acetobacter xylinum is a Gram-negative soil bacterium that synthesizes and secretes cellulose during its metabolism of glucose (Cannon R.E, 2000). Bacterial cellulose (BC) can be made into fabric through a series of fermentation, extraction, drying, and bleaching steps. The purpose of this research is to synthesize vegan leather from *Acetobacter xylinum* in the lab and compare it to animal and imitation leather. Materials were grown in lab for a time period of 4 weeks. The product then was sanitized, dried, and bleached for the next 12 weeks. Three ottomans were made using the three different types of leather. Discussions and comparisons of the products will address product developments such as ease of construction, price, and time. In conclusion, factors to improve the vegan leather (BC) will be further addressed.

Keywords: *Acetobacter xylinum*, vegan, animal, synthetic, leather, product development

Synthesis of Cellulose by *Acetobacter Xylinum*:

A Comparison of Vegan Leather to Animal and Faux Leather

Introduction

Leather use has been documented for centuries. Although leather is widely used in many applications, the processes of leather-making release a large amount of toxic chemicals into the environment (Kanagaraj et al, 2006). In this project, we chose to work with the *Acetobacter xylinum* because it can synthesize fibers with high resilience and a unique structure which produces a biodegradable source of textile. BC is a gram-negative and rod-shaped microorganism which is also known as *Gluconacetobacter xylinus* (Mohaininet al 2014). Bacterial cellulose composes stronger cellulose fiber than plant cellulose which suggests that it may have superior characteristics to those in plants. (Mohammad et al., 2014). The most striking feature of this bacterium is its ability to polymerize glucose into cellulose (Mohainin et al 2014). Throughout the development of the BC, or vegan leather in this project, we considered the proper growth media and processing for the material.

The purpose of this study is to compare vegan leather to animal and imitation leather to promote environmental sustainability within consumers. Using vegan leather as an alternative would reduce demand for animal and/or imitation leather. The production of the animal and imitation leather utilizes harmful chemicals that leads to contamination whereas BC requires less energy and chemicals to produce. Three ottomans were made to compare the different leathers. One ottoman was made with vegan leather from *Acetobacter Xylinum*, one with traditional cow leather, and one with the original fabric for the ordered ottomans, imitation leather.

Literature Review

Leather is animal skins that have been cured, tanned, or dressed to create a stable product and to prevent decay. Most leather produced and sold in the U.S. is made from the skins of cattle and calves, but it can also be made from sheep, lambs, goats, and pigs (PETA, 2019). Before animal skins can be processed, they need to go through the process of removing the subcutaneous fatty layer and drying to reduce the action of harmful bacteria. These processes lead to different products creating varying grades and types of leather (Harri & Veldmeijer, 2014). Leather exhibits great durability and flexibility which makes it ideal for uses in clothing, accessories, furniture, and upholstery (PETA, 2019). Roughly half of all leather produced today is used to make shoes and about 25% for clothing and around 15% for upholstery (History of Leather, 2019). Modern technology has allowed for innovation in the leather industry. The development of chemicals and sophisticated processing methods have greatly improved the aesthetics, feel, and application of leather. Leather continues to be the material of choice, not just for commercial and residential furniture but also for automotive, aviation and marine applications (History of Leather, 2019). These fine properties of the leather help to maintain a large worldwide demand for such fabric which could be problematic with respect to environmental and ethical issues.

Due to the processes of leather-making, it has been categorized as one of the highly polluting industries (Kanagaraj et al, 2006). The harmful nature of the chemicals used in the tanning process can contaminate the environment (Kanagaraj et al, 2006). The high contents of dissolved, suspended organic and inorganic solids in tannery effluents give rise to potentially toxic metal salts, chromium, and metal ions and increases biological oxygen demand (BOD). BOD is the most the common method of measuring pollutant organic material in water. Low BOD content indicates good quality of water while a high level of BOD suggests polluted quality of water (Encyclopedia of Public Health, 2019). Moreover, tanning activities also emanate an unpleasant odor from the decomposition of protein waste material and the presence of sulfide, ammonia and other volatile compounds in the process (Kanagaraj et al, 2006). The salt used for preserving the hide discharges a large amount of total dissolved solids (TDS) and chlorides. There is an average

consumption of 45- 50 m² of the waste liquor and 800 kg of solid wastes per ton of rawhide discharged by the leather industry. Other polluting chemicals in the tanning industry are lime, sodium sulfide, ammonium salts, sulfuric acid, chromium salts, and vegetable tanning materials. Solid wastes generated in the leather industry pose a pollution threat in terms of BOD and TDS (Kanagaraj et al, 2006). In addition, many leather industries dispose unprocessed water onto soil and affects the soil's productivity (Kanagaraj et al, 2006). These chemicals used in the making of leather impose a significant level of contamination on the environment.

Not only does the leather industry release chemicals that can give rise to potentially toxic metal salts and increases BOD, it is also linked with the expansion of animal cruelty because the sale of leather good is directly associated with the economic success of animal factory farms (PETA, 2018). Every year, the global leather industry tans more than a billion of byproducts (skins and hides) from the meatpacking industry (PETA, 2018). During the transport of the meat manufacturing, many of these animals endure extreme crowding and confinement, deprivation, branding, tail-docking, and dehorning (PETA, 2018). Although most leathers are made from domestic animals, other exotic species are also hunted and killed specifically for their skins, including zebras, bison, kangaroos, elephants, crocodiles, alligators, ostriches, lizards, and snakes (PETA, 2018). Lastly, to support and raise awareness of animal and environmental issues, many consumers have turned to vegan and imitation leather products as an alternative for genuine leather.

Imitation leather is a common substitute choice for animal leather, it is also known as synthetic leather. Synthetic leather is more cost-effective in production and it is made with highly processed raw materials. The two most commonly researched types of synthetic leather are those produced with polyvinyl chloride (PVC) or polyurethane (Medical Design Briefs, 2015). Both are thermoplastic materials that can be shaped into a variety of forms (Medical Design Briefs, 2015). PVC and polyurethane vary in properties such as temperature, chemical compatibility, and plasticizer. One product may perform better than the other in different applications (Medical

Design Briefs, 2015). PVC may harden and become less pliable when certain fluids—concentrated acids and alkalis and polyurethane offers high elongation values like rubber and abrasion resistance that are superior to PVC. Polyurethane is naturally flexible and does not use additives to retain its flexural properties (Medical Design Briefs, 2015).

Polyurethane is composed of isocyanates, polyester polyols, and additives. The two commonly used types of isocyanates are methylene diphenyl diisocyanate (MDI) and toluene diisocyanates (TDI) (Lee, 2002). In 2000, 4.4 million tons of MDI and TDI were produced (Lee, 2002). The isocyanates need to react with polyols powered by hydroxyl groups to produce polyurethane polymers (Lee, 2002). Polyols are a group of low-digestible carbohydrates derived from the hydrogenation of their sugar or syrup source (Benefit of Polyols, 2019). In 2000, there were 850,000 tons of polyester polyols produced with the growing demand of 4-5% per year (Lee, 2002). Polyol blends include additives like catalysts, fire retardants, blowing agents, coloring agents, and fillers.

The most widely used thermoplastic polymeric material, polypropylene, began to be manufactured only sixty years ago (Andrady and Neal, 2009). Therefore, the sixty-year time frame would be the maximum extension of the potential observation period of thermoplastic material, but studies on the behavior of plastics in the total environment are much more recent (Thompson et al., 2009). Thermoplastics are not easily biodegradable, so they last for a long period of time. The degree of how much these polymers biodegrade depends on their chemical nature and the environment they are in (Scalenghe, 2018). Plastics are almost never used by themselves and polymers are usually mixed with additives such as carbon or silica to strengthen the matter. Thermal stabilizers, plasticizers, fire retardants, UV stabilizers, colorants, matting agents, pacifiers, or luster additives are also often added to the polymers and can raise the possibility of adverse health issues (Scalenghe, 2018). Plastic can be broken down to microplastics to be disposed of more easily. In North America, about 44,000–300,000 tons of microplastics are being

put in farmlands (Scalenghe, 2018). The increased use of plastic could impose a threat to the environment.

In addition, since polyurethanes have an unstable sensibility to sunlight, UV radiation could promote harmful reactions within the thermoplastic (Newman and Forciniti, 2001, Scalenghe, 2018). Fungus, such as *Aspergillus tubingensis* can grow on the surface of polyurethane film with biodegradation capabilities, such as surface cracking, pore formation, and loss in tensile strength (Khan et al., 2017, Scalenghe, 2018). Although synthetic fiber is free of animal cruelty, it can have a negative impact on humans and bring contaminations to the environment. Isocyanates are severe irritants to the mucous membranes of the eyes and gastrointestinal and respiratory tracts (NIOSH, 2018). Direct skin contact can also cause marked inflammation (NIOSH, 2018). Workers who are in contact with isocyanates make them subject to severe asthma attacks if they experience frequent exposure (NIOSH, 2018). There is evidence that both respiratory and dermal exposures can lead to sensitization and death has been reported from severe asthma in some sensitized subjects (NIOSH, 2018). Some common symptoms are recurring eye irritation, nasal congestion, dry or sore throat, cold-like symptoms, cough, or shortness of breath (NIOSH, 2018). Workers who are constantly exposed to isocyanates should take safety precautions as the chemicals can trigger many health hazards.

BC is a natural-like leather and it requires few resources to grow. The vegan leather industry is on the rise because there is an increasing population of vegans. The number of global consumers who identify as vegan has grown 61% from 2014 to 2017 (Global, 2018). Veganism is referred as the movement or philosophy that advocates and entails abstinence from consuming any animal product - such as meat, dairy, eggs, leather, silk, etc. - and stands against widespread animal exploitation (Ulusoy, 2015). It is a growing movement that specifically aims at reinforcing the philosophy behind social justice through extending the morality towards animals, respecting their lives, and fostering compassion between animals (Ulusoy, 2015). Sustainability

has become a key element across all industries because consumers, especially in the west, have become more aware about the practices surrounding goods they buy and have a keen interest of the origins of the goods (Choudhar, 2017). Therefore, the vegan leather industry is expected to rise with the evolution of veganism. According to Grand View Research, the vegan leather industry is expected to reach \$85 billion by 2025. With the evolving textile technology, consumers are preferring vegan fashion, which refers to adopting non-leather products (Grand View Research, 2019). It is predicted that vegan leather will serve as the most suitable substitute for animal leather.

BC is one of the most biodegradable materials available and has been traditionally used in food, cosmetics, biomedical and even electronics industries (Jang et al, 2017). While cellulose is a well-known plant product, some bacteria have gained attention as an alternative and sustainable source of cellulose with *Acetobacter xylinus* being a common BC producer. Fibers made from BC has greater strength compared to those made from cellulose because ribbons are formed at BC structure to strengthen the resulting product. On the other hand, plant cellulose only has linear formation in natural vegetation (Mohammad, 2014). BC also has excellent physical properties including mechanical stability, tensile strength, thermostability, crystallinity (i.e. ultrafine fibrous structure), purity and biocompatibility (i.e. biologically non-toxic and no immune responses (Jang et al, 2017). This organic material has self-synthesizing property and can be cultivated in any desirable garment panel shape (Chan et al., 2018). This maximizes the biological efficiency of the cultivation process because there is minimal cutting and thus less textile waste (Chan et al., 2018). Furthermore, sustainable tailor-shaped techniques like panel-shaped cultivation and contact surface-blocking cultivation have been tested and researchers found that it is possible to achieve zero textile material waste with BC (Chang et al., 2018).

These cultivation techniques allow BC to significantly reduce waste, energy, time, and cost during production. Therefore, BC is the more environmentally sustainable choice of material compared to synthetic and animal leather.

In this project, the product development process was referenced from May-Plumlee & Little, 1999 and Davis & Sanders, 2014 (May-Plumlee, & Little, 1998). The product development process provides a guideline of bringing new products to the market. The planning stages we used were *Marketing, Merchandising, Design and Development, and Production*. For the *Marketing* stage, we conducted a literary search to investigate the consumers for vegan leather and we decided our target audience to be vegans. For the *Merchandising* stage, the choice and sizing of the home interior piece was planned, and we decided to use a gender-neutral color, white, interior piece to avoid any gender/color biases. The gender-neutral ottomans were made from traditional leather, imitation leather and bacterial cellulose vegan leather to enhance the understanding of differences among the materials. For the *Design and Development* stage, we decided on the aesthetic of the ottomans and we chose the color white for all three ottomans. The BC material for the ottomans was brewed and processed in our lab. Ottoman covers were modified to accommodate the different materials. In the *Production* stage, we grew the material, constructed, and inspected the prototypes and made sure that the construction of each ottoman was consistent to one another.

Methodology

To synthesize the BC in lab we used *Acetobacter xylinum*. First, the pure bacterial cellulose strain was cultivated in standard Hestrin Schramm media with mannitol to promote bacterial growth. All media were sterilized prior transferring the bacteria to make sure the absence of any other containments. For the next three weeks, the bacteria were transferred into the containers with the media to grow at 32 degrees Celsius. The mats were disinfected by immersing in a 1 % NaOH soak for 24 hours at room temperature. Next, the mats were further

disinfected in a 4% glycerol, 1% Germaben solution at room temperature for 24 hours to guarantee the absence of live cultures. Then, the mats were bleached at room temperature with a 10% bleach and deionized water solution for 24 hours. After bleaching, it was treated again with the glycerol and Germaben soak. Finally, all materials were rinsed and dried in an incubator at 32 degrees Celsius. During the bacterial cellulose's drying process, we added weights on top of the mats to keep them flat as they were drying. Figure 1. shows bacterial cellulose being dried in the incubator. Before the drying process, the color was whiter compared to the dried product.



Figure 1. Bacterial Cellulose mat.

The mats also had to be stretched daily by hand in the horizontal and vertical direction. They were also layered with towels in between them to obtain minimal curling for the mats to stay flat. Curling on the mats would cause the material to shrink and fold on top of each other making the material smaller and thicker on the side, which we want to avoid.

We bought three ottomans to conduct a comparison of the three types of leather. We removed the ottoman covers and used flat pattern techniques to create modified patterns for each of the different materials used as the ottoman covers. For the bacterial cellulose mat, the ottoman

patterns were modified in order to fit the ottoman. The ottoman cover consisted of seven pattern pieces for each ottomans, one top panel, two identical short panels, and a total of four panels for the long side panel. Due to the shrinkage of the bacterial cellulose, the patterns had to be modified the most. The ottoman top panel had to be 1” smaller than the imitation and the real leather. Additionally, since the mats from the bacterial cellulose were grown in smaller containers, we had to split the long panel in half for the mats to be of the correct size. For consistency, we had to split the long panel into two pieces for the real animal and imitation leather as well. Before cutting the material, we had to ensure that the pattern pieces would fit the ottomans. The pattern pieces were first cut out of muslin, a lightweight cotton cloth, used for making test garments before cutting it out of the final material and sewn together to test the fit. After producing and altering the patterns, we then proceeded with the cutting and construction of the ottoman covers. Products were developed with a heavyweight thread and a polyester thread for topstitching. All materials were sewn with a leather sewing needle on a domestic machine. The traditional animal leather, and the bacterial cellulose leather had the stitch length set to 5, while the imitation leather had a stitch length of 4 when sewed together. However, the tension varied from all the products. Noted earlier about the modification on the bacterial cellulose top panel cover, the bacterial cellulose mat had to be hand-sewn to finish for the cover to fit on the ottoman. Only the bacterial cellulose leather was brushed with tragacanth gum. The covers were attached by being glued to the wood base of the ottoman with wood and/or leather and suede glue.

Results

Constructing the traditional animal leather was difficult. The material was a nonwoven material, so the fabric could not be pinned together because it would create holes in the fabric. The material was hard to control, even just sewing a straight stitch because it would shift, making one side longer than the other. There were many problems with the stitch length of the material as well as the tension. We set the stitch length to a five, however, due to its tough interior and stitching over two layers, the stitch would come out short and several seams had to be reconstructed. The tension had to be tested multiple times so the stitch would not come loose. Constructing the side panels of the leather was relatively simple once the appropriate tension was determined; attaching the top panel to the side panels was more delicate because the corners of the top panel were curved. Once everything was attached, topstitching had to be done parallel to the straight stitching on the top of the ottoman cover. Topstitching is created to be seen on the outside of the material either for design or practical purposes. In our case, the topstitching served as the ottoman's final touches to produce a more polished product. Figure 2. shows the final product result of the traditional animal leather ottoman cover.



Figure 2. Traditional animal leather ottoman cover.

It has a smooth surface and it carries that distinguishing leather smell with it. The estimated price for the traditional animal leather would be \$100.00, without labor costs.

As for the construction of the imitation leather, it was not as thick as the traditional leather. Sewing the side panels together was easier than the traditional animal leather. The stitch length was set to four and overall had a better and consistent appearance with its stitches compared to the animal leather. However, sewing the topstitching on the imitation leather was the hardest to sew. We had kept the stitch length of five with the topstitching. The topstitching on the imitation leather was inconsistent in size and overall was difficult to sew through. Puckering also occurred which could have been caused by an uneven ply feeding and excessive machine thread tension. With the uneven ply feeding, the fabric fed into the seam at a different rate than the other plies and the excessive machine thread tension was caused by the thread trying to return to its original length which most likely explains the inconsistency of stitch length. Figure 3. shows the final product result of the imitation leather ottoman cover.



Figure 3. Imitation leather ottoman cover.

Pictured above is the imitation leather, notice there are relatively no differences between Figures 2 and 3. The colors are close to identical, but the textures are different. The imitation leather has

no recognizable odor to it. The estimated price of the imitation leather ottoman would be \$29.99, without labor costs.

Comparing all three covers, the bacterial cellulose was the easiest to sew but the most challenging to assemble on. The material was stiff and paper-like, making it the easiest one to maintain a straight stitch because it did not shift when sewn through. Connecting the side panels together was straightforward and was the easiest to sew through. The top panel of the material was curved and shorter on the sides thus leading us to manipulate the seams on the shorter length side. During the attachment of the top panel to the side panels, it was difficult to manipulate because of its inflexibility and how rigid and stiff it was. Most of the ottoman cover got stitched on with the machine except for the corners. Before the corners got stitched on, we decided to try and fit it onto the ottoman. It was extremely strenuous to get on and because we had to shorten the top panel, it did not line up exactly in the corners of the ottoman. We then decided it would be best to finish the corners of the cover by hand-stitching it with the polyester thread. This step was immensely laborious to hand sew. During hand-sewing, an all-purpose sewing needle was used, and a slipstitch was being applied; this had made it especially difficult to puncture through the bacterial cellulose and thread through the material. Figure 4. shows the final product result of the vegan leather out of bacterial cellulose ottoman cover.



Figure 4. Vegan leather ottoman cover made from bacterial cellulose.

Illustrated above, there is a noticeable color difference between the traditional animal leather, the imitation leather, and the bacterial cellulose leather. There is a slight discoloration in the bacterial cellulose leather as it appears slightly darker compared to the other covers. Also, notice that the bacterial leather cover is not smooth compared to the traditional animal leather and the imitation leather.



Figure 5. Real, imitation, and vegan leather ottomans.

Illustrated above are the three-ottoman leather covers together. From the picture, notice the difference in colors on all three of the ottomans. The traditional animal leather, bottom left side, is whiter than the other two ottomans. Next to the traditional animal leather is the imitation leather, it has a more off-white color to it and on top, the bacterial cellulose ottoman has a creamier color to it.

Initially, the pricing of the bacterial cellulose ottoman is estimated approximately double the price of the traditional leather ottoman and eight times that of the imitation leather. The bacterial cellulose is estimated to be \$200.00, without labor costs. Initially, labor costs for all three materials would be around the same price range. Comparing all three ottomans, the traditional animal leather had the smooth surface with some texture to it. It was also the thickest

material out the three but was also the most flexible to work with and manipulate. It did carry that distinguishable leather smell to it while the imitation and the bacterial cellulose had no identifying smells. Lastly, the traditional animal leather had the whitest appearance. For the imitation leather, the surface was smooth with no texture to it. It was a thin material that was also flexible, but not as flexible as the traditional animal leather. The color of the leather was a more off-white color when put next to the traditional animal leather. Finally, the bacterial cellulose ottoman had the roughest texture out of the three ottomans combined. The material was the thinnest out of the three but it was also the stiffest. Additionally, the color of the ottoman was more of a tan or a cream.

Conclusion

From the literature review, the process of making animal-leather releases many toxic chemicals which pollutes the environment. Other negative impacts of the leather industry include higher BOD content, high amounts of TDS, and soil and air contamination. Although imitation leather is free of animal products, the production causes health hazards among workers. In addition, since imitation leather is a type of thermoplastic, it is not easily biodegraded and contributes to a large amount of waste. Due to the rise of the vegan industry, BC has become more easily accessible to consumers. Different cultivating techniques have been developed to reduce energy, time, and cost of production in BC.

The material BC was successfully synthesized in lab with minimal health hazards and environmental pollution. We constructed three ottoman covers from vegan, animal, and imitation leather and compared them based on cost, ease of construction, and overall appearance. For the cost the prototype development, the BC ottoman is twice the price of the traditional leather ottoman and eight times that of the imitation leather. We expected the cost of imitation leather to be lowest but did not anticipate the cost of BC to be exceedingly greater than the cost of animal leather. We want to reduce the cost of BC by testing different alternatives to growing the

bacterial cellulose. Some possibilities could be the use of symbiotic culture of bacteria and yeast from Kombucha, mushroom or pineapple cellulose. The animal leather was the most difficult to construct because the material was thick, and it had to be sewn over two layers. The topstitching on the imitation leather was inconsistent in size and made it the most challenging to sew through. The stiffness of BC made it the easiest to sew because it was easy to maintain a straight stitch. The time of construction was approximately the same for all three ottomans. Moreover, improvements on appearance need to be made by getting rid of its wrinkle and attain a whiter color for the BC. The animal leather was the whitest, followed by the imitation leather and lastly the vegan leather. The animal leather also had the smoothest and softest texture among the three. In the future, we want to expand on development of prototypes in home furnishings, accessories, and clothing to test BC in a wider range of applications. A prototype evaluation will be added in the future to determine the market share of vegan leather versus the other leathers. Lastly, we hope that this project will increase the popularity and usage of bacterial cellulose as an alternative to animal leather.

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