




Review

Bacterial Cellulose: From Biofabrication to Applications in Sustainable Fashion and Vegan Leather

Dheanda Absharina ¹, Mohamad Padri ^{2,*}, Csilla Veres ¹ and Csaba Vágvölgyi ^{1,*}

¹ Department of Biotechnology and Microbiology, Faculty of Science and Informatics, University of Szeged, Közép Fasor, H-6726 Szeged, Hungary; dheanda.absh@gmail.com (D.A.); gmri.csilla@gmail.com (C.V.)

² Kingdom of Saudi Arabia (DAB-KSA) Project, Development of Algal Biotechnology, Beacon Development, King Abdullah University of Science and Technology (KAUST), Thuwal 23955, Saudi Arabia

* Correspondence: mohamad.padri@kaust.edu.sa (M.P.); mucor1959@gmail.com (C.V.)

Abstract: The rising demand for sustainable materials has led to a significant focus on developing resources from renewable systems, particularly through the integration of biological processes. Bacterial cellulose (BC) has emerged as a highly promising biomaterial, gaining attention across multiple industries, such as food, pharmaceuticals, materials science, and textiles, due to its renewable, biodegradable, and eco-friendly characteristics. Within the fashion industry, bacterial cellulose (BC) biofabrication presents a groundbreaking method for producing sustainable textiles and vegan leather. This systematic review emphasizes BC's pivotal role in advancing sustainable materials, addressing challenges like low yields, strain instability, and high production costs, and exploring innovative biofabrication techniques to overcome these barriers. Current advancements aim to enhance the thickness, uniformity, and mechanical properties of BC layers by optimizing the environmental and nutritional conditions during *Komagataeibacter* cultivation and leveraging coculturing methods. Furthermore, recent innovations in synthetic biology and genetic engineering have opened new avenues for improving BC biosynthesis, making it a viable solution for the sustainable fashion industry. This review explores three core topics: (1) bacterial cellulose and its applications, (2) the biofabrication of BC for vegan leather, and (3) emerging innovations and patents utilizing bacterial cellulose as a sustainable industrial biomaterial.



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1. Introduction

The fashion industry is a major contributor to environmental pollution, particularly through the release of microplastics from synthetic textiles [1,2]. Synthetic leather production also poses significant environmental and ethical issues [3]. Typically, synthetic leather manufactured from plastic-based materials, such as polyurethane (PU) or polyvinyl chloride (PVC) [1], are composed of long chains of synthetic polymers. These materials significantly contribute to microplastic pollution by releasing microplastic particles during use, washing, and disposal. Due to their limited biodegradability, these particles persist in the environment, leading to long-term contamination [3,4]. Utilizing, washing, and disposing of these textiles, particularly synthetic leather, have caused further environmental burdens. Studies indicate that microplastics released from these synthetic materials, along with the improper disposal of fashion items, are major contributors to this environmental impact [3].

Microplastics (MPs), defined as plastic particles smaller than 5 mm [2], represent an increasingly serious global issue due to their pervasive presence in the environment

and their harmful effects on organisms. Currently, over 96% of research on MPs focuses on marine ecosystems. However, it has been confirmed that most marine MP pollution originates from land-based sources. According to various studies, both in China and internationally, microfibers from textile washing amount for approximately 35% of the microplastics detected in aquatic systems. This urgent need for greater attention to land-based sources of microplastic pollution, particularly from synthetic textiles, in efforts to mitigate their environmental impact [2–5].

Growing concerns about animal welfare and environmental sustainability have increased the demand for alternatives to animal leather [6]. This demand has driven the development of innovative materials such as vegan leather, often derived from plant-based sources and synthetic polymers [3,6]. Another option is the use of natural substances of microbial origin. One such innovative material is nanocellulose, produced by members of the *Komagataeibacter* genus [7]. Its unique structural properties, including a three-dimensional network and nanoscale fiber size, make BC highly absorbent and adaptable, with remarkable biocompatibility due to the absence of lignin and hemicellulose [1,8].

Unlike plant-derived cellulose, microbial cellulose holds significant potential to transform the fashion industry by providing a sustainable alternative to traditional textiles [3,9,10]. Its material properties are suitable for a wide range of applications, ranging from everyday clothing to advanced fashion technologies [3]. However, its widespread adoption is hindered by challenges related to scalability and cost-effectiveness in production [11]. Further research is required to optimize the production methods, enhance the material characteristics, and explore new applications. Moreover, consumer education and market acceptance will be crucial for the successful integration of BC into mainstream fashion [1,12].

Komagataeibacter is a well-studied microorganism for nanocellulose production, and optimizing its growth conditions can significantly improve the yield and efficiency [7,10,11]. Previous studies have examined various nutrients and environmental factors that influence the growth of *K. xylinus* [1,6,7,13], but comprehensive industrial-scale studies remain limited. Understanding the growth dynamics of this microorganism is essential to optimizing production processes [13,14]. Contamination during production has been a notable challenge, particularly in the nata de coco industry, where most operations in Southeast Asia are home-based and rely on conventional sterility techniques [13,15,16]. However, the use of cocultures with other microbes may provide natural nutrient supplementation, promoting the growth of *K. xylinus* and yielding thicker cellulose layers than monocultures [15,17]. This approach leverages microbial interactions to optimize production processes and tailor the characteristics of the cellulose sheets [11,14,15]. Bacterial cellulose has emerged as a promising biopolymer with significant potential in the textile industry, particularly as a sustainable alternative to leather [1,11]. While biofabrication is becoming a key technique for enhancing the yield and producing durable, high-quality textile materials [12], the industry currently faces numerous challenges in its transition toward more sustainable practices, including the need to improve scalability and production consistency [7,13].

BC produced by *K. xylinus* is a highly pure, renewable, and biodegradable material with excellent mechanical properties, including high tensile strength, flexibility, and moisture retention [12,15,18]. Unlike conventional leather, which relies on animal hides and chemical-intensive processing [3,6], BC is synthesized through a natural fermentation process, offering a low-impact and environmentally responsible option for bio-leather production [6,7]. This process involves cultivating cellulose-producing bacteria, which form a dense network of nanofibers in a gel-like structure [15,19]. This material can be processed to mimic the appearance and texture of traditional leather while offering enhanced sustainability benefits [3,12]. BC can be grown using minimal resources, such as

agricultural waste, and its production does not require harmful chemicals or synthetic polymers [19,20].

Challenges remain in maintaining the homogeneity and yield of BC, particularly in industries such as fashion, where high-quality cellulose layers are essential. The thickness and durability of the cellulose layer directly impacts the quality of bio-leather produced [6,15]. Researchers have conducted numerous experiments in recent decades, concerning optimizing nutrient sources, acclimatization techniques, and bacterial propagation [1,6,7,11,18]. Recent studies have also explored by utilizing cocultures with yeast, microalgae, and other bacteria to improve fermentation processes [17,21,22]. Advances in genetic engineering are also being applied to create unique biomaterials with tailored properties, as performed by several of BC manufacture [18,21].

In this review highlights the potential for alternative methods by tailoring BC-based biofabrication as an efficient alternative resource that reduces the environmental footprint on the fashion industry, particularly on bio-leather production. Within the context of sustainable fashion, BC offers a promising alternative to conventional or synthetic leather, providing both environmental and material advantages.

2. Bacterial Cellulose and Its Application

Bacterium aceti was first identified by A.J. Brown in 1886 during his experiment on acetic fermentation. Due to its distinctive fermentation yield by cellulose formation, he classified *B. aceti* into *B. xylinum* (*A. xylinum*) [8]. Later, Yamada and colleagues in 2012 recognized the combination phenotypic characteristic from former genera, which were *Acetobacter*, *Gluconobacter*, *Asaia*, *Swaminathania*, *Saccharibacter*, *Neoasaia*, *Granulibacter*, *Tanticharoenia*, *Ameyamaea*, and *Neokomagataea*. They proposed a new genus, *Komagateibacter* (gen. nov.), in honor of Dr. Kazuo Komagata, a Professor at the University of Tokyo, Bunkyo-ku, Tokyo, Japan, who made immense contributions to bacterial systematics, particularly of acetic acid bacteria [23]. In 1990, Brown and colleagues [19] successfully isolated and sequenced a gene involved in cellulose biosynthesis from *A. xylinum* [8,19]. This strain is widely recognized as a model organism for cellulose synthesis due to its highly crystalline structure and remarkable mechanical stability [19,24]. Moreover, the insertion of cellulose synthase genes into diverse organisms has emerged as a pioneering technique and a hallmark step in modifying cellulose formation in bacteria for tailored applications. This advancement underpins the large-scale industrial production of bacterial cellulose (BC), significantly enhancing its potential for the development of high-value, functional materials across various fields. The key industrial applications of BC are illustrated in Figure 1.

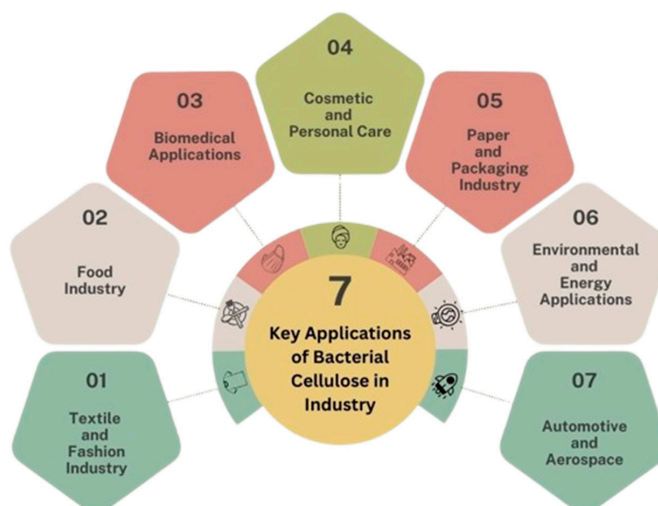


Figure 1. Application of bacterial cellulose (BC) in industry.

2.1. Textile and Fashion Industry

Bacterial cellulose (BC) is gaining significant attention in the fashion industry as a potential alternative for conventional synthetic fibers, such as polyester and nylon, which are harmful to the environment [25]. At the same time, nanocellulose represents a step towards more sustainable and eco-conscious materials, offering extensive innovation possibilities [26].

Pioneering vegan leather companies have utilized raw materials derived from fungi as the basis for their products. Fungal bio-leathers represent an emerging class of sustainable materials produced from mycelium, the root-like network of filaments that constitutes the vegetative structure of fungi [3]. The production of fungal bio-leathers involves cultivating mycelium under controlled environmental conditions, including temperature and humidity, which influence its growth and quality [27]. Mycelium's rapid and efficient growth process, coupled with its adaptability to various substrates, allows for scalable production methods that utilize agricultural waste, contributing to circular economy models [3,27]. Companies at the forefront of this innovation, such as MycoWorks [28], Reishi [29], MycoTech [30], and Bolt Threads [31], have pioneered the commercialization of mycelium leather, creating products such as shoes, handbags, wallets, and watch straps.

Through microbial fermentation processes, nanocellulose can be synthesized, harvested, and processed into a leather-like material [1,10,26,32]. This method requires fewer resources than traditional leather tanning or synthetic leather manufacturing [10,32]. Microbial synthesis of nanocellulose fibers, primarily by bacteria such as *K. xylinus*, which generates a dense and highly pure cellulose structure [10,24]. Due to its remarkable mechanical properties, BC is receiving significant attention in the fashion and material science sectors for its potential to replace traditional leather and synthetic polymers in various applications [10,26,32]. Several innovative companies are leading the charge in utilizing BC to manufacture sustainable and functional fashion products. Modern Synthesis [33], based in the UK, is pioneering the use of BC to develop next-generation bio-fabrics for footwear and apparel. This company focuses on cultivating BC composite materials and yarn-reinforced bacterial cellulose hybrid materials focusing on fashion and textile applications. Nanolose [34], an Australian biotech company, is also at the forefront of BC innovation with its creation of Nullarbor™ Tree-Free lyocell fiber, which is spun into yarn, with potential use in garments. Another notable company, ScobyTec [35] from Germany, specializes in producing Scoby leather—a 100% biodegradable and vegan leather alternative made from BC. Although no official cooperation was announced, ScobyTech showcased several sample products using their material. Similarly, the Hungarian-Estonian start-up Smobyta [36] is developing NanoTwine™, a nanocellulose manufacturing platform to produce BC in a high-quality, cost-competitive way to displace animal and fossil fuel-based materials in leather-like and cosmetic applications. Figure 2 depicts the starting material of bacterial cellulose (BC) and the leather-like material produced from it.



Figure 2. Wet BC pellicle (left side) and BC-based vegan leather (right side). Photo Credits: Smoby, Hungary [36].

2.2. Food Industry

One notable food industry project is InnoCell, [37], which emerged from the “From Peel to Peel” initiative. Originally a bachelor’s thesis exploring microbial cellulose (MC) as a sustainable packaging alternative [14,37], InnoCell now aims to increase the value of food production byproducts. It promotes the development of circular food and material systems, emphasizing upcycling and the transformation of food waste into valuable food and non-food products [14]. InnoCell’s current products include edible films and coatings, as well as biodegradable food packaging.

Bandyopadhyay et al. [38] investigated the cross-linking of BC and guar gum (GG)-based hydrogel films, modified with polyvinyl pyrrolidone–carboxymethyl cellulose (PVP-CMC), designed for fresh berry packaging. The films were evaluated for various properties including their mechanical strength, barrier capability, bioadhesion, color, gloss, rheological behavior, surface hydrophobicity, permeability, water solubility, biodegradability, and effectiveness in extending the shelf life of berries. Their findings demonstrated that incorporating GG into the PVP-CMC-BC films enhanced their elasticity, load-bearing capacity, and barrier properties. These biofilms also exhibited superior hydrophobic qualities compared to the other films tested. Importantly, all biofilms showed 80% biodegradability within 28 days in vermicompost, positioning them as a promising biodegradable alternative to traditional packaging materials.

Cazón et al. [39] tested biodegradable films made from bacterial cellulose, glycerol, and polyvinyl alcohol (PVOH) for their mechanical properties, water vapor permeability, and UV-protective characteristics. The results showed that the composite films exhibited high elongation (up to 49.89%) and good rupture resistance (13.78 MPa). The films showed a UV transmittance as low as 5.59%, 2.4%, and 0.57% in the UV-A, UV-B, and UV-C ranges, respectively. The addition of PVOH reinforced the BC matrix, while glycerol acted as a plasticizer.

Several companies and research initiatives are developing BC for food-related applications, such as Cellugy in Denmark [40] and publicly owned research institutes such as VTT Technical Research Centre in Finland [41] and Fraunhofer Society in Germany [42], all of which are actively investigating BC’s potential in food applications.

2.3. Biomedical Application

BC has also garnered significant attention in fields such as wound care, tissue and organ regeneration tailored with 3D bioprinting, bone regeneration, drug delivery for anti-cancer therapies and antibiotic delivery, and medical implants [19,32,43].

Maurer et al. [44] assessed the effectiveness of BC dressings for treating pediatric thermal injuries, analyzing clinical outcomes such as dressing changes, hospital stays, and complications. The average length of hospital stay was 6.7 days, and children underwent an average of 2.4 dressing changes under anesthesia. The wound infection rate was low, with none of the children developing an infection. Approximately 82% of children were treated exclusively with BC-based dressings, and most patients indicated good epithelialization after 10 days. Most patients exhibited favorable outcomes in terms of scar formation, with 98% showing normal pigmentation, and high rates of adequate vascularization (91%), height (92%), and pliability (92%). Recent findings by Cañas-Gutiérrez et al. [45] investigated the biomineralization of BC scaffolds designed for bone tissue engineering. The study revealed significant findings regarding the mineralization process, including the deposition of calcium phosphates (CPs) onto the BC scaffolds over five immersion cycles. These cycles involved alternating between calcium and phosphate salts. The results showed that the crystals formed on the BC surface were rod-shaped and like the calcium phosphate composition of immature bone. As the number of cycles increased, the crystals' calcium phosphate ratio grew from 1.13 to 1.6, and their size expanded from 25.12 nm to 35.9 nm. Furthermore, *in vitro* results demonstrated that the scaffolds supported high cell viability (up to 95%) and promoted the osteogenic differentiation of human bone marrow mesenchymal stem cells, indicated by the expression of bone-related markers like alkaline phosphatase, osteocalcin, and osteopontin.

Notable companies and research institutes are developing biomedical applications of BC, such as BioModics (Denmark) [46], which is working on BC wound dressings suitable for advanced wound care. BC is also being explored for drug delivery systems, as demonstrated by the research on controlled antibiotic release for wound infections and implant-associated infections. In the public sector, the VTT Technical Research Centre (Finland) [41], a leading research institution, is exploring BC for 3D bioprinting, tissue engineering, and drug delivery applications in the biomedical field.

2.4. Cosmetic and Personal Care

The unique properties of bacterial cellulose (BC) have positioned it as an innovative ingredient in creams, lotions, and face masks aimed at improving skin texture and maintaining a supple, moisturized complexion [19,32,41]. Amorim et al. [47] developed a polymer-based mask designed for acne-prone skin. This BioMask incorporates BC as the primary material due to its biocompatibility, hypoallergenic properties, and high-water retention, making it particularly effective for skin hydration and inflammation reduction. The BioMask also contains a 2% propolis extract, chosen for its polyphenols and flavonoids, which have proven anti-inflammatory and healing properties. A characterization of the BC film was conducted using X-ray diffraction (XRD) and thermogravimetric analysis (TGA), confirming the stability and potential efficacy of the propolis-infused BC film as a cosmetic product. This mask is expected to improve skin hydration, promote faster healing, and enhance skin texture, offering a non-invasive solution for managing acne symptoms.

In another study, Fonseca et al. [48] discovered the development of microneedle patches made from BC combined with hyaluronic acid (HA). BC provides a strong, biocompatible scaffold with high mechanical strength, which makes it suitable for fabricating microneedles that can penetrate the skin. Meanwhile, hyaluronic acid, known for its hydration and biocompatibility, enhances the skin's moisture content and is beneficial for

therapeutic skin applications. In vitro evaluations of these microneedles on skin models demonstrated good skin penetration, a controlled release of bioactive compounds, and excellent biocompatibility. Then, based on in vivo evaluations, animal model studies further confirmed that the patches were safe and effective for delivering therapeutic agents. The research highlights the potential of BC-HA microneedle patches as a promising solution for minimally invasive skin treatments with enhanced efficacy and minimal side effects.

In a recent study, Kucuk et al. [49] examined the impact of a facial serum combining BC with herbal extracts and bioactive compounds on hyperpigmentation. Key ingredients, such as licorice root, white mulberry, and ascorbic acid, inhibited tyrosinase activity, thereby reducing melanin production and hyperpigmentation. The serum also promoted collagenase activity and showed no cytotoxic effects, making it suitable for cosmetic use and demonstrating effectiveness in skin-brightening applications.

As major cosmetic companies and research institutions increasingly invest in BC, its role in the industry is poised to expand. Companies like L'Oréal and Procter & Gamble are exploring BC for both product formulations and packaging, and companies like Cellugy [40] are leveraging BC for its hydrating and film-forming properties. Furthermore, research institutions, such as the University of Tokyo [50] and the Fraunhofer Society [42], are also advancing the applications of BC, paving the way for more sophisticated and sustainable personal care solutions. As the industry continues to evolve, BC stands out as a promising ingredient that combines performance with environmental consciousness, driving innovation in cosmetic and personal care products [19,32,43].

2.5. Paper and Packaging Industry

In the paper and packaging industry, BC also shows potential as a sustainable material. Chemically like plant cellulose but immensely finer in structure, BC provides strong inter-fiber hydrogen bonding, making it ideal for enhancing paper strength and durability. This improvement can increase the paper's resistance to wear and tear [19,32,51].

In another interesting experiment, Soares da Silva et al. [52] explored the combination of BC with polyhydroxybutyrate-valerate (PHBV) to enhance its barrier properties and maintain its eco-friendliness. The composite was plasticized using glycerol or polyethylene glycol to improve its flexibility, then coated with PHBV to reduce water vapor permeability and increase hydrophobicity. This combination improved the mechanical strength of BC with poly(3-hydroxybutyrate-co-3-hydroxyvalerate) (PHBV), aiming to produce a biodegradable food packaging that requires moisture protection, such as for meat products. The resulting material exhibited a significant reduction in water vapor transmission, from 0.990 to 0.032 g/m²·day·Pa, offering a biodegradable alternative to conventional plastics used in food packaging. Companies like Ecovative Design [53] and Cellugy [40] are at the forefront of integrating BC into packaging solutions, driving eco-friendly alternatives that contribute to a circular economy.

In other research related to the enhancement of the quality of paper, Tabarsa et al. [54] investigated whether BC could improve the tensile strength and flexibility of paper made from softwood pulp. The study also examined the optimal BC content, including reinforcement (60%). BC improved the tensile strength and flexibility of the paper with an increment of water retention (20%), without significantly adding to its weight. Furthermore, Lourenço and colleagues [55] investigated the role of BC in improving the properties of fine paper, both as an internal additive and a coating agent. BC, due to its strength, high surface area, and film-forming capabilities, was found to enhance paper quality, particularly its printability. The application of a 50:50 BC coating to uncoated office paper significantly improved color reproduction, as indicated by a more than 25% increase in the color gamut area, compared to untreated paper. These findings suggest that BC could be a valuable material

for producing high-quality, eco-friendly fine papers for printing and other applications requiring a high-resolution output.

Table 1 highlights the advantages of BC over plant-based cellulose, particularly in terms of its finer structure, durability, and adaptability to innovative, eco-friendly applications.

Table 1. Comparison features in between BC and plant-based cellulose.

| Features | Plant Cellulose | Bacterial Cellulose (BC) | References |
|--------------------|---|---|------------|
| Quality | Chemically similar to BC but coarser in structure. | Highly refined, with a finer structure enabling superior inter-fiber bonding and enhanced material properties. | [32,51] |
| Durability | Moderate strength and durability. | Strong inter-fiber hydrogen bonding increases strength, tensile properties, and wear resistance in applications like paper. | [32,51] |
| Flexibility | Limited natural flexibility. | Can be modified with plasticizers or high filler content to improve flexibility. | [51] |
| Barrier properties | Limited barrier properties against water vapor and moisture. | Enhanced barrier properties, especially when combined with PHBV, reducing water vapor transmission and increasing hydrophobicity. | [32,51] |
| Eco-friendliness | Renewable and biodegradable. | Highly eco-friendly; supports biodegradable alternatives and a circular economy. | [32,51,55] |
| Cost | Generally lower production costs due to established large-scale agricultural systems. | Higher production costs, but ongoing research aims to make BC more cost-effective for industrial use. | [51] |
| Innovation | Limited adaptability to new material innovations. | Allows for tailored properties through combinations with other materials, such as PHBV, for specialized applications. | [52] |
| Weight addition | Adds noticeable weight to paper products when used for reinforcement. | Improves tensile strength and flexibility with minimal weight increase. | [54] |
| Printability | Moderate printability with limited surface properties. | Excellent printability due to high surface area, strength, and film-forming capabilities, enhancing color reproduction. | [55] |

2.6. Environmental and Energy Sector

BC-based materials have shown potential for use in energy storage devices such as advanced batteries and supercapacitors. Lokhande et al. [56] and Ma et al. [57] performed extensive research related to BC for separators, binders, and electrode materials (in its carbonized form) in supercapacitors and lithium-ion batteries. Through tuning BC into various fiber, layered, or porous forms, subsequent studies could enhance ion transport and storage capacity by up to 20–30%. The formation of micropores and mesopore structures in electrodes also increases electrode–electrolyte contact, boosting overall conductivity and energy density. BC-based materials are thus able to increase the energy storage capacity and improve device stability. Other environmentally friendly applications have been developed by Ecovative Design (Green Island, USA) [53], which focuses on using BC for creating eco-friendly alternatives to conventional materials, including energy-efficient insulation and filtration systems.

2.7. Automotive and Aerospace Sector

Aisyah et al. [58] and Nurazzi et al. [59] reviewed the use of nanocellulose composites to reinforce polymeric matrices, improving their mechanical and thermal properties. BC composites, when combined with synthetic nanofillers like carbon nanotubes or graphene,

exhibit enhanced stiffness, strength, and thermal stability, all of which are crucial for reducing vehicle weight and improving fuel efficiency in the automotive industry. These composites also offer excellent thermal stability, flexibility, and low thermal expansion, making them suitable for use in aerospace applications where high-temperature resistance is essential. BC composites are also being explored for their corrosion resistance, and, in automotive applications, BC is being used in components such as bumpers and foam pads, contributing to energy absorption and crash resistance.

Several companies, including Toyota Motor Corporation (Toyota City, Japan) [60], Boeing (Everett, WA, USA), and Airbus (Toulouse, France) [61], are integrating nanocellulose into automotive and aerospace technologies. These companies, alongside research institutions like the Georgia Institute of Technology [62], are driving forward the adoption of BC in automotive and aerospace applications, highlighting its transformative potential [61,62].

However, challenges related to the hydrophilic nature of nanocellulose and its dispersion in polymer matrices remain, but advances in processing technologies are expected to overcome these issues and further expand the use of nanocellulose composites.

3. Biofabrication of BC on Vegan Leather Industry

The biofabrication of BC in vegan leather synthesis represents an innovative and sustainable approach. It utilizes biodegradable waste materials as carbon sources for BC cultivation, allowing for natural decomposition by microorganisms after disposal. This process collaborates with eco-friendly tanning methods, reducing the impact of chemical waste pollution [10,12,63,64].

In a previous study, Yang et al. [63] employed waste coconut water and natural dyes from sources such as indigo, carrot peel, grape skin, and coffee grounds to create vegan leather. The production cost for these vegan leather shoes was approximately \$97.92/m² (excluding labor and operational costs), while the raw material cost was around \$7.52/m², while the commercial cowhide cost ranges from \$39.70 to \$220.80/m². Additionally, BC vegan leather crosslinked with oxidized edible oil demonstrated a remarkable tensile strength of 82.14 MPa, surpassing that of traditional cow leather for shoe uppers by 2.08 times. When plasticized with glycerol, the tensile strength reached 56.19 MPa, with an elongation at break of 36%. These properties make BC vegan leather an attractive eco-friendly alternative to conventional leather manufacturing. The workflow for the biofabrication of BC, from cultivation to the production of vegan leather, is schematically depicted in Figure 3.

The biofabrication of BC involves cultivating cellulose-producing bacteria in nutrient-rich media, typically containing glucose or other sugars as carbon sources [65]. Bacteria from the *Komagataeibacter* genus could synthesize cellulose from a wide range of carbon sources, including monosaccharides, disaccharides, oligosaccharides, and sugar alcohols [10,65,66]. Under both static and agitated conditions, these bacteria produce cellulose fibers that form a hydrogel-like matrix at the air–liquid interface. This nanocellulose can then be harvested, purified, and processed into various forms, such as films, sheets, or fibers, for multiple applications [67]. Extensive optimization efforts have focused on areas such as inoculum density, temperature, pH, agitation, and types of carbon and nitrogen sources to cultivate the bacteria effectively [65,66,68,69]. Additional factors, such as light, humidity, incubation period, and BC treatment method (static or agitated), significantly impact the cellulose growth and yield [68]. Studies have examined the effects of various carbon and nitrogen sources, as well as the initial pH, on the BC yield and its properties [10,68,69]. Glucose remains the most used carbon source for cultivating cellulose-producing bacteria [66,70]. Maintaining an optimal pH range is essential to prevent gluconic acid accumulation, which can inhibit cellulose production [66]. Optimizing the fermentation process is critical to improving the BC yield and reducing production

costs [70]. Generally, fermentation processes have been optimized by varying one independent factor at a time, while holding others constant [65,68]. However, at an industrial scale, the complexity of interacting variables requires more sophisticated optimization approaches, as conventional methods often fail to capture intricate relationships affecting BC production [11]. Consequently, this limits the effectiveness of scaling production rates and achieving high cellulose yields.

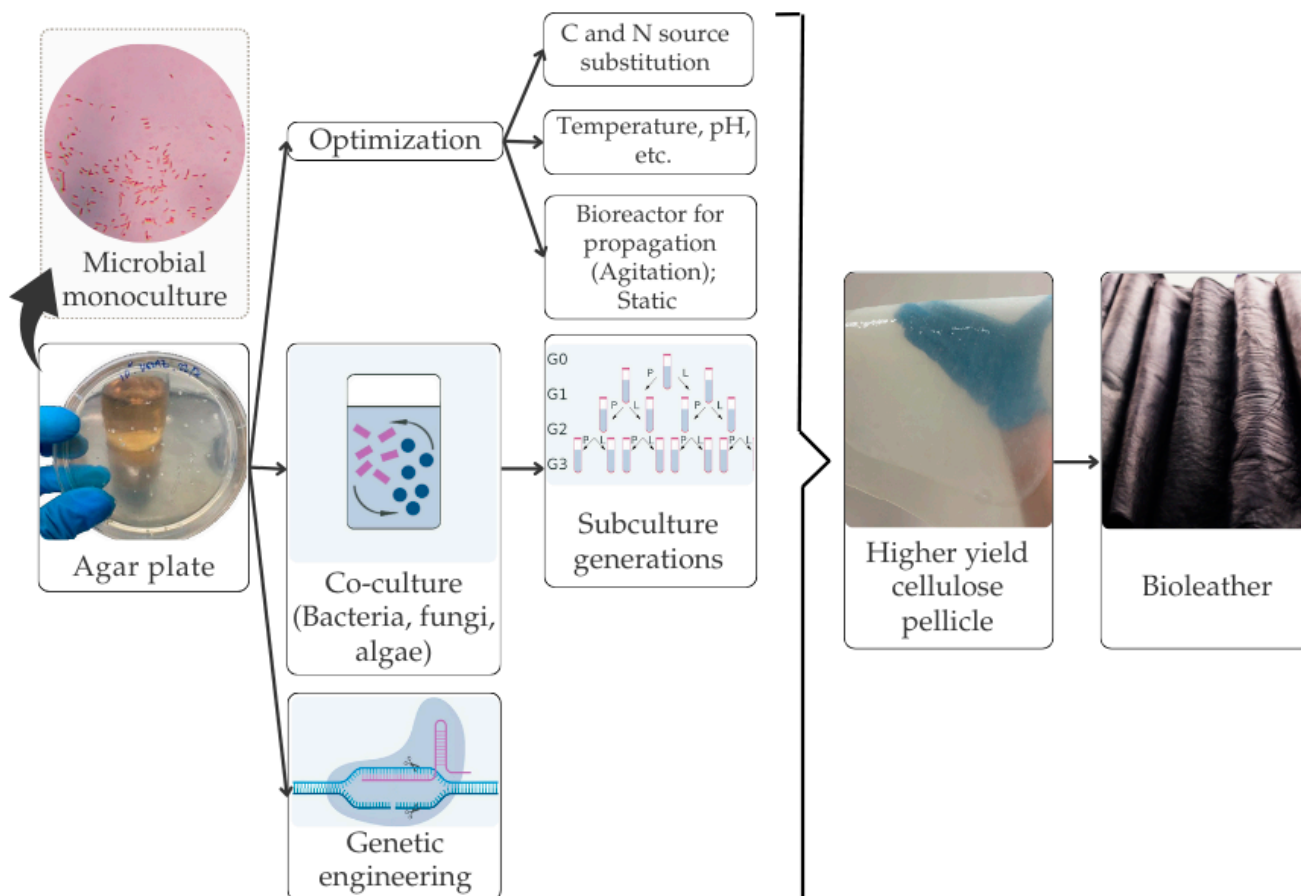


Figure 3. Biofabrication of BC for vegan leather products.

Coculture-based biofabrication of BC is another innovative approach, enhancing cellulose production by combining bacterial strains or microbial communities. Coculturing promotes synergistic interactions, improving the yield, structure, and functional properties of nanocellulose [15,22,71,72]. For example, coculturing *Komagataeibacter* with other cellulose- or non-cellulose-producing microbes can increase the cellulose output or produce bio-leather that mimics traditional leather in appearance, texture, and durability [73]. A previous study demonstrated that coculturing *K. hansenii*, a well-known BC producer, with *A. pullulans*, a producer of the polysaccharide pullulan, significantly improved the mechanical properties of BC. Optimized conditions led to a 22.4% increase in BC production and a 4.5- to 6-fold improvement in elastic modulus compared to baseline conditions in both shake-flask and bioreactor systems [22,74]. Similarly, another study reported an increase in BC production from 2.5 to 13.8 g/L by coculturing *K. nataicola* with *L. fermentum* [15]. The coculture approaches summarized in Table 2 highlight significant advancements in bacterial cellulose (BC) production through various strain consortia, substrate utilization, and additive applications. Among these pairings, the most effective coculture was *K. nataicola* Q2 and *L. fermentum* SR, yielding both the highest BC production and improved crystallinity. Lignocellulosic substrates and pullulan additives demonstrated their value

in enhancing sustainability and yield, respectively. These findings underscore the importance of strain compatibility, substrate selection, and additive usage in maximizing BC production. Coculture fermentation holds potential for (a) increased product yields, (b) the synthesis of novel compounds, and (c) metabolic symbiosis [74,75]. However, maintaining microbial community stability poses challenges, as cocultivated microorganisms often require distinct growth conditions and media.

Table 2. Studies on coculture approaches to improve bacterial cellulose production in the last 5 years.

| Cocultured Strains | | Result | Reference |
|--|-----------------------------------|--|-----------|
| <i>K. nataicola</i> Q2 | <i>L. fermentum</i> SR | Enhanced BC yield from 2.5 to 11.79 g/L (increasement 4.5 times higher) and 6.7% increased crystallinity of BC membrane | [15] |
| <i>K. intermedius</i> | <i>D. bruxellensis</i> | Promoting the BC synthesis reaching 1.09–1.27 g/L by coculture with <i>D. bruxellensis</i> on concentration 10 ⁶ CFU/mL | [17] |
| <i>K. hansenii</i> | <i>A. pullulans</i> | Increased (22.4%) BC yield | [22] |
| <i>K. hansenii</i> (ATCC 23769) | <i>A. pullulans</i> (ATCC 201253) | Increment of BC production after adding pullulan additive (1.5% and 2%), from 0.447 g/L to 0.814 g/L and 1.997 g/L, respectively | [72] |
| <i>K. xylinus</i> CGMCC 2955 Δ <i>gdh</i> | <i>B. cereus</i> | Increased BC yield in lignocellulosic enzymatic hydrolysate from 1.2 to 4.4 g/L | [73] |

Research into sub-cultured generations examines the consistency of BC production across multiple sub-cultured generations, focusing on whether repeated culturing and transfer to fresh media impact cellulose quality, fiber structure, or yield [74]. This research emphasized the importance of preserving bacterial strains capable of producing nanofibers with desirable properties, in terms of thickness, length, and strength, which are critical for evaluating the scalability of BC production in the fashion industry.

The bioindustry of BC is often constrained by low production yields, microbial strain instability, and high culture media costs, all of which hinder its scalability for large-scale manufacturing [12,32]. Previous studies have shown that engineered *K. xylinus* strains can produce thicker BC films than wild-type strains, with engineered strains achieving film thicknesses between 6.2 and 10.2 μ m compared to 5.1 μ m in wild-type strains [76]. Recently, Jang et al. [77] reported that the heterologous overexpression of the *pgi* gene in the *K. xylinus* 2325 strain led to an 115% increase in the BC yield, reaching approximately 3.15 g/L, compared to the 1.46 g/L yield of the wild-type strain. However, further research is critical to achieving the technological advancements required for large-scale production and commercialization. As shown in Table 3, improvements in BC production—such as enhanced thickness and crystallinity—have been achieved through gene overexpression, the inhibition of gluconic acid (a byproduct that can limit cellulose yield), and optimized oxygen availability to maintain essential oxygen levels for large-scale BC production. Recent developments in synthetic biology and CRISPR interference technologies have enabled the precise modulation of BC biosynthesis, resulting in BC-based materials with improved characteristics [78,79]. Additionally, genetic toolkits and CRISPR-based approaches, which have shown success in other microorganisms, hold potential for overcoming current challenges in BC production, including unrevealed gene functions.

Table 3. Genetic modifications of cellulose-producing bacteria.

| Strains | Genetic Modifications | Result | Reference |
|----------------------------------|--|---|-----------|
| <i>G. xylinus</i> | Overexpressing <i>bcsC</i> , <i>bcsD</i> , and <i>bcsCD</i> genes | To increase BC yield 5.4, 6.2, and 6.8 g/L, respectively. | [18] |
| <i>K. xylinus</i> DSM 2325 | Overexpressing of glucose 6-phosphate isomerase (PGI) gene from <i>E. coli</i> | Increased (115.8%) BC production from 1.46 g/L to 3.15 g/L. | [77] |
| <i>K. xylinus</i> CGMCC 2955 | Modulating <i>gallU</i> gene expression with CRISPRi (CRISPR interference) | Faster production rate with 2-to-4-fold higher yield of BC production. | [78] |
| <i>K. xylinus</i> DSM 2325 | Post-transcriptional repression of <i>bcsD</i> , <i>bcsZ</i> , and <i>ccpA</i> genes using genome-integrated mRFP1 | Modulation of the crystalline structure of the BC yield. | [79] |
| <i>K. hansenii</i> KO28 | Low doses (0.5, dual 0.5, 1, and dual 1 kGy) of gamma radiation treatment | Substantial enhancement in BC yield from 2.2 g/L to 4.3, 10.5, 5.5, and 7 g/L, respectively. | [80] |
| <i>K. hansenii</i> ATCC 23769 | Overexpression of <i>motA+</i> and <i>motB+</i> genes | The <i>motA+</i> and <i>motB+</i> mutants produced cellulose layers that were 6% and 20% thicker, respectively, compared to the wild-type (WT) and pTI99. | [81] |
| <i>K. hansenii</i> ATCC 53582 | Disruption of <i>motA</i> and <i>motAB</i> genes | Reduction in motility by approximately 65% in the <i>motA</i> mutant and 50% in the <i>motAB</i> mutant resulted in a corresponding decrease in the thickness of cellulose fibers. | [82] |
| <i>K. oboediens</i> MSKU 3 | Restoring the original function/phenotype of <i>bcsC</i> mutant gene | The R37-9 revertant produced ultrafine BC fibers with an average diameter of 34.6 nm, representing a 50.9% thinner than parental strain's fiber (70.5 nm). | [83] |
| <i>K. medellinensis</i> ID 13488 | Upregulating cellulose synthesis genes, the <i>bcs</i> clusters under acidic stress | This study contributes to a deeper understanding of the genetic mechanisms governing bacterial cellulose (BC) biosynthesis in <i>Komagataeibacter medellinensis</i> ID13488, with a specific emphasis on its functionality under acidic conditions. | [84] |
| <i>G. xylinus</i> | Insertion of <i>crdS</i> gene from <i>Agrobacterium</i> sp. | The production yields of cellulose and curdlan by the genetically modified (<i>crdS</i> -expressing) strain reached 46.38% of the wild type on day 5, improved to 71.76% on day 10, and reached 80.22% by day 15. Although the genetically modified strain starts with a lower yield, it nearly matches the wild type by day 15. | [85] |
| <i>G. hansenii</i> ATCC 53582 | Co-expression of <i>cmcax</i> , <i>ccpAx</i> , and <i>bcsABCD</i> genes from bacterial cellulose within <i>E. coli</i> , combined with IPTG induction | Increased BC production (0.3–0.92 mg/L). | [86] |
| <i>K. xylinus</i> CGMCC 2955 | Modifications to the phosphoenolpyruvate-dependent glucose phosphotransferase system (PTS _{Glc}) to improve bacterial cellulose (BC) production by optimizing glucose uptake | The engineered strain GX08PTS03 produced 7.74 g/L of BC after six days of static fermentation, which represents an 87.41% increase compared to the BC production level of the wild-type strain. | [87] |

Table 3. Cont.

| Strains | Genetic Modifications | Result | Reference |
|---|---|--|-----------|
| <i>A. xylinum</i> ATCC23769 | Overproduction carboxymethyl cellulose (CMCax) on <i>A. xylinum</i> | Enhancing cellulose production 1.2-fold. | [88] |
| <i>K. xylinus</i> DSM 2325 | IS element insertion on <i>bcsA</i> gene | Prevention of an irreversible genotype shift to a cellulose-negative (Cel-) strain which could increase cellulose yield of approximately 3.23 g/L compared to the unmodified strain. | [89] |
| <i>K. xylinus</i> K2G30 | Pyrrroloquinoline quinone-dependent glucose dehydrogenase (PQQ-GDH) suppression | The Δ <i>gdh</i> K2G30 strain achieved the highest BC yield (12.26 g/L) in HS medium by day 11, reflecting slower but enhanced production relative to the wild type, emphasizing the impact of carbon source preferences on BC synthesis. | [90] |
| <i>K. xylinus</i> DSM 2325 | Knocking-out pyrroloquinoline quinone (PQQ) dependent glucose dehydrogenase (GDH) gene | BC yield 2.3-fold higher. | [91] |
| <i>K. xylinus</i> (formerly was <i>A. xylinus</i>) | Inserting <i>Vitreoscilla</i> hemoglobin (<i>vhb</i>) gene | The Vhb-expressing strain produced a cellulose pellicle concentration of 11 g/L on static condition. | [92] |
| <i>K. xylinus</i> ATCC 23770 | Incorporating genes from <i>Escherichia coli</i> K-12 strain (mannose kinase and phosphomannose isomerase) to improve mannose utilization | Increased pellicle thickness (84%) and increased BC yield. | [93] |
| <i>K. rhaeticus</i> | Inserting and expressing tyrosinase gene within <i>Komagataeibacter</i> | Self-dyeing of BC with melanin pigmentation. | [94] |

As shown in Figure 4, over 50% of genetic modifications are concerned with enhancing the thickness of the cellulose pellicle, addressing a critical industry challenge in meeting consumer demands. Additionally, 13.6% of these genetic modifications aim to reduce gluconic acid formation, leading to increased BC production. Other modifications target improvements in the crystallinity, which influences the durability and robustness of the cellulose. Another biofabrication strategy involves the integration of the tyrosinase enzyme, which allows for in situ dyeing by catalyzing the oxidation of phenolic compounds to generate color [94]. Although this self-dyeing process presents technical challenges, such as regulating the color intensity and ensuring a uniform dye distribution, ongoing research is working to overcome these obstacles for broader applications.

Further advancements in BC biofabrication highlight its versatility for sustainable design. The integration of materials such as mesh or woolen yarn during growth enables the creation of complex textures and hybrid forms. Post-cultivation treatments, including dyeing and laser cutting, further enhance functionality. Optimized conditions, such as a six-day growth period with a 15 g/L tea concentration, allow for the efficient production of 2D sheets and seamless 3D forms, minimizing waste and exemplifying sustainable innovation in modern fashion [95].

Another innovative project, Ava, demonstrates the fusion of traditional craftsmanship and modern technology. By leveraging origami-inspired folds, it overcomes BC's rigidity and incorporates LEDs and conductive threads for interactive designs. This approach addresses limitations such as water resistance and rigidity, while also showcasing BC's potential for avant-garde fashion and sculptural accessories. The emphasis on adaptability,

minimal environmental impact, and artistic expression positions microbial 3D fiber art as a pioneering field in sustainable and futuristic design [96].

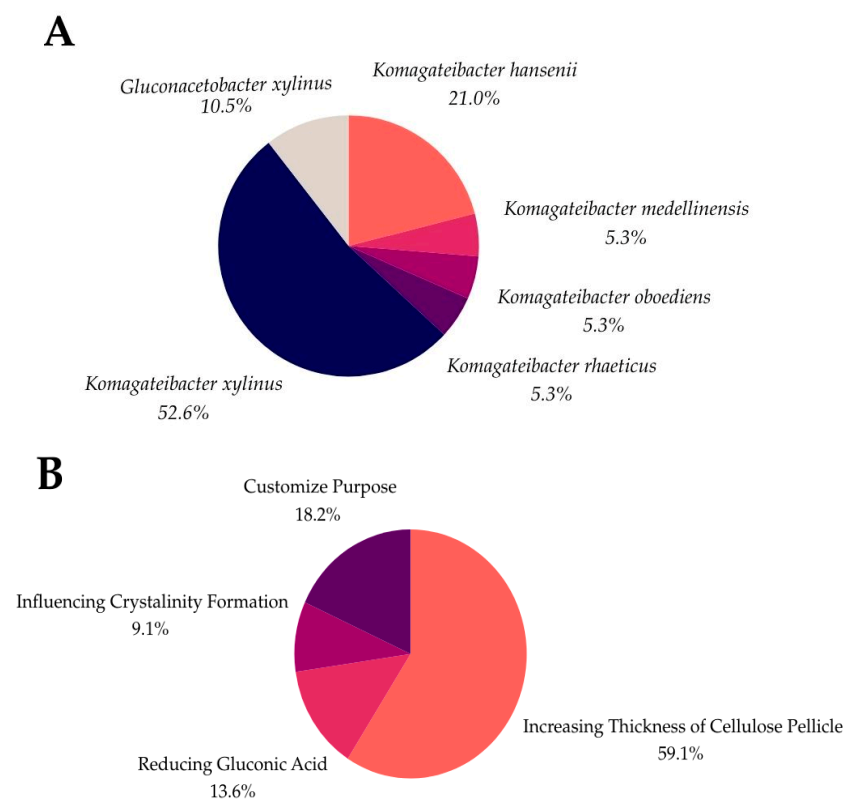


Figure 4. Strains of BC (A) and genetic modification purposes in BC (B).

BC, a biopolymer produced through microbial fermentation, offers high tensile strength, biocompatibility, and customizable properties, making it highly suitable for integration into 3D printing processes. In a study on 3D biofabrication, Polewka et al. [97] explored the use of 3D printing technologies with biomaterials as sustainable alternatives for textile and fashion production. Their research highlights how additive manufacturing (AM) techniques, such as fused filament fabrication (FFF), utilize bio-based materials like polylactic acid (PLA), cellulose, and lignin, which are renewable and biodegradable resources. These materials can be employed to produce textile-like structures, garments, and prototypes with minimal waste, contributing significantly to the eco-friendliness of fashion manufacturing.

The combination of BC and 3D printing represents a transformative approach in fashion, blending biofabrication with cutting-edge technology to create eco-friendly, customizable garments and textile products. Wang et al. [98] investigated cellulose nanofibrils (CNFs)-reinforced polylactic acid (PLA) biocomposite filaments for fused deposition modeling (FDM) 3D printing. CNFs, prepared through enzymatic hydrolysis and high-pressure homogenization, were integrated into PLA to improve the composite's mechanical, thermal, and water absorption properties. Similar to CNFs, BC could provide analogous benefits while potentially expanding functionality. Its hydrophilic nature supports post-processing, such as dyeing, and its ability to integrate into biodegradable matrices aligns with the emphasis on sustainability and environmental consciousness in consumer products. Both materials represent promising paths for eco-friendly solutions in 3D biofabrication, particularly for textiles and apparel, where customization, functionality, and sustainability are paramount.

Data that were collected from previous studies indicate that *K. xylinus* is the most used strain for genetic modification (52.6%), followed by *K. hansenii* (21.0%) and *G. xylinus* (10.5%). Less commonly used strains for genetic engineering are *K. rhaeticus*, *K. oboediens*, and *K. medellinensis* at 5.3%, respectively.

These companies are leading the advancement of sustainable fashion by using *Komagataeibacter* strains, renowned for their high cellulose yield, in the production of vegan leather. This innovative approach emphasizes the potential of BC as a viable and eco-friendly alternative to traditional leather materials within the fashion industry (Figure 5).



Figure 5. Forefront of biotechnology companies utilizing biofabrication of bacterial nanocellulose for the vegan leather industry.

4. Patent Search

A patent search was conducted using the European Espacenet database, with further verification through Google Patents. The search employed keywords such as “*Komagataeibacter*”, “bacterial cellulose”, “leather”, “sustainable textile”, and “biofabrication”, resulting in 23 published patents from 2017 to 2023 (Table 4). Patents in this domain are geographically diverse, although the number of applications has declined since 2017 (Figure 6A). South Korea holds the largest share of patents (36.4%), followed by China and Japan (18.2% each), with additional contributions from the United States, Poland, and Turkey (Figure 6B). Modern Meadow Inc. leads in patent publications, holding 18.2% of the patents, followed by Samsung Electronics Co., and Sookmyung Women’s University Industry-Academic Cooperation Foundation (13.6% each). Other notable contributors include Gozen Bioworks Corp. and SK Bioland Co. (9.1% each), along with Dongguan Chengyuan Leather Technology Co., Ltd., Shandong Baochen Big Data Services Co., Ltd., Columbia University, DePuy Synthes Product Inc., RespectBio Co., Ltd., Kyungpook National University Industry-Academic Cooperation Foundation, and Song Ji-eun et al. (4.5% each) (Figure 6C). Figure 6 illustrates the fluctuation in the number of registrations and publications of applications utilizing bacterial cellulose (BC) for biofabrication purposes between 2017 and 2023. While the data initially show a consistent high trend from 2017 to 2019, with four applications each year, there was a noticeable decline starting in 2020, where the number dropped to three applications. This

downward trend continued in 2022 and 2023. Based on this observed pattern, it is projected that this decreasing trend may persist in subsequent years, potentially reflecting challenges in the development and commercialization of BC-based biofabrication technologies.

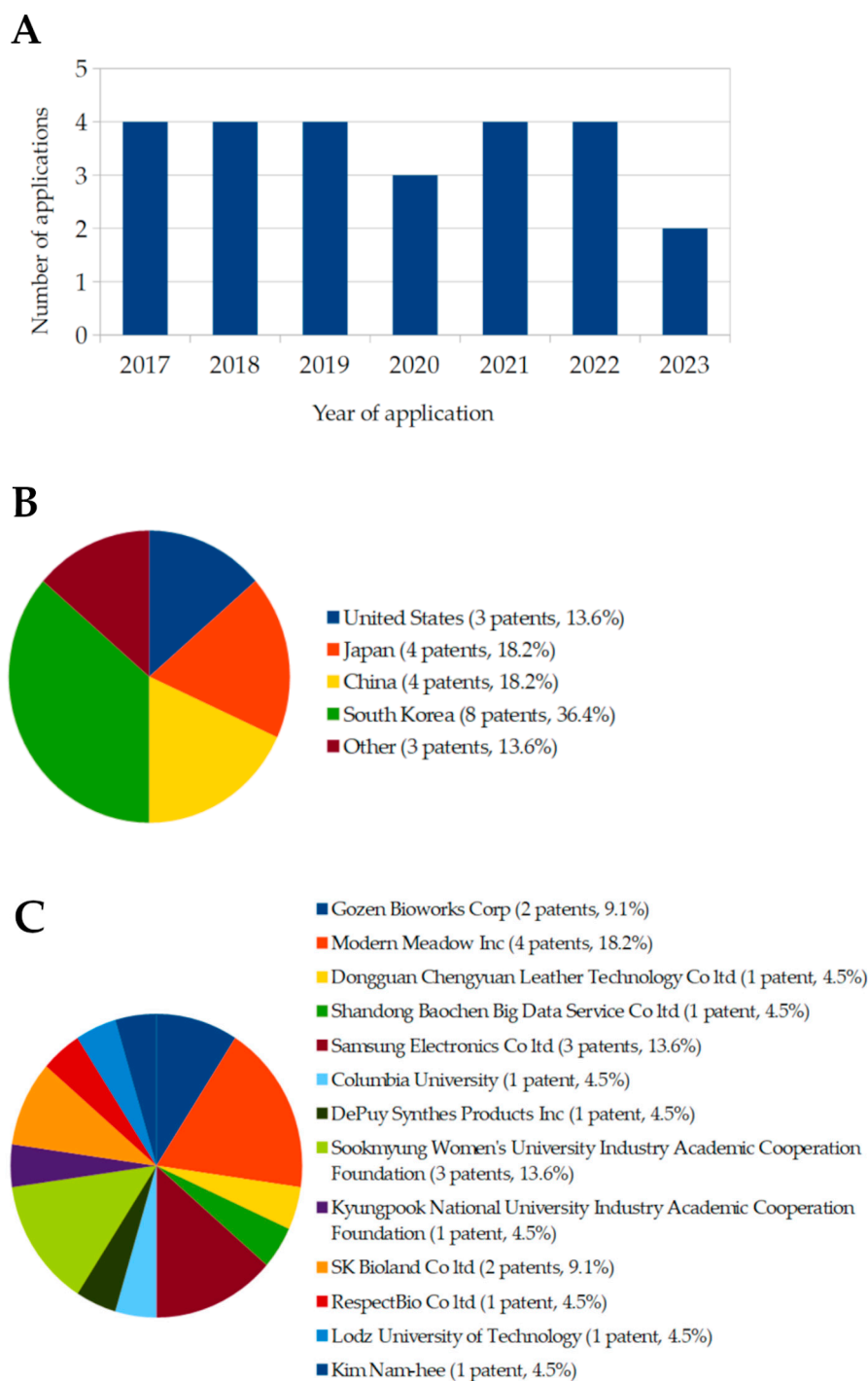


Figure 6. Number of patents (A), publication locations (B), and patent ownership (C) related to BC-based technologies and associated fields between 2017 and 2023. The data were obtained through searches conducted on the European database Espacenet and Google Patents, accessed on 15 September 2024.

Table 4. List of patents related to BC-based application and patent ownership from 2017 to 2023.

| Patent Number | Title of Patent | Patent Ownership |
|-----------------|---|--|
| US20240117564A1 | A bioleather production method | Gozen Bioworks Corp. (San Francisco, CA, USA) |
| TR2021002376A2 | A bio-leather coated fabric and manufacturing method | Gozen Bioworks Corp. (USA) |
| JP2019530757A | Biofabricated leather article and method | Modern Meadow, Inc. (Nutley, NJ, USA) |
| JP6971039B2 | Methods for biofabrication of composites | Modern Meadow, Inc. (USA) |
| US20190092838A1 | Recombinant yeast strains for producing recombinant protein or collagen | Modern Meadow, Inc. (USA) |
| JP2019123982A | Bio-fabricated leather product having zone characteristic | Modern Meadow, Inc. (USA) |
| CN112771119A | Biological production of leather products and method thereof | Modern Meadow, Inc. (USA) |
| CN216139597U | Degradable biological leather | Dongguan Chengyuan Leather Technology Co., Ltd. (Dongguan, China) |
| CN117625395A | Growth type biological leather containing multiple-headed chorionic villus and preparation method and application thereof | Shandong Baochen Big Data Service Co., Ltd. (Jining, China) |
| US10465175B2 | Recombinant microorganism including genetic modification that increases pyruvate, phosphate dikinase activity and use thereof | Samsung Electronics Co., Ltd. (Seoul, South Korea) |
| CN109385390A | Recombinant microorganism of genus <i>Komagataeibacter</i> , method of producing cellulose by using same, and method of producing microorganism | Samsung Electronics Co., Ltd. (South Korea) |
| KR20190052888A | <i>Komagataeibacter</i> genus microorganism having enhanced cellulose productivity, method for producing cellulose using the same, and method for producing the microorganism | Samsung Electronics Co., Ltd. (South Korea) |
| US2023146427A1 | Biofabrication and microbial cellulose bio textile | Columbia University (New York, NY, USA) |
| JP2022528183A | Bacterial-derived nanocellulose fabric material | DePuy Synthes Products Inc. (Warsaw, IN, USA) |
| KR20230169689A | Bacteria cellulose-milk protein complex and method | Sookmyung Women's University Industry Academic Cooperation Foundation (Seoul, South Korea) |
| KR102192110B1 | Bio-leather material containing bacterial cellulose fiber and a preparation method | Sookmyung Women's University Industry Academic Cooperation Foundation (South Korea) |
| KR102501458B1 | Bacteria cellulose-mushroom derive particle complex and artificial leather manufactured by using the same | Sookmyung Women's University Industry Academic Cooperation Foundation (South Korea) |
| KR102524357B1 | Bacterial cellulose with improved mechanical properties and manufacturing method thereof | Kyungpook National University Industry Academic Cooperation Foundation (Sangju, South Korea) |
| WO2020060164A1 | <i>K. rhaeticus</i> strain and cellulose sheet production method using same | SK Bioland Co., Ltd. Bioland Haemun Branch (Cheongju, South Korea) |
| WO2020060165A1 | Composition for culturing strain for producing bacterial cellulose | SK Bioland Co., Ltd. Bioland Haemun Branch (South Korea) |
| KR102089538B1 | <i>K. intermedius</i> rb-b-24 and bio cellulose preparing method using the same | RespectBio Co., Ltd. (Seoul, South Korea) |
| PL443677A1 | Method of producing modified bacterial cellulose in the form of bio-leather | Lodz University of Technology (Lodz, Poland) |
| KR102192110B1 | Bio-leather material containing bacterial cellulose fiber and a preparation method thereof | Song Ji-eun, Kim Hyun-jin, Kim Hye-rim (South Korea) |

5. Conclusions and Future Perspectives

The pursuit of biomaterials as a reliable solution to mitigate the ecological impact of microplastic pollution and facilitate the recycling of agricultural waste has gained considerable attention. The biofabrication of BC offers transformative potential for the fashion industry, providing a sustainable, versatile, and high-performing alternative to

traditional textiles. This aligns with the growing consumer demand for environmentally conscious fashion products. Over the past two decades, biofabrication technologies utilizing microorganisms, such as fungi and bacteria grown on agricultural residues, have been developed to generate natural cellulose. This review underscores the potential of BC across various industries, focusing on alternative biofabrication processes, the strains used, and the promising outcomes related to scalability and innovation in sustainable textiles. BC presents key advantages over conventional synthetic materials, including cost-effectiveness, safety, biodegradability, and a reduced environmental footprint. Nevertheless, challenges remain in scaling production to industrial levels, while ensuring the stability and uniformity of the final product. Future research should prioritize the optimization of substrate parameters to better understand their interactions and enhance the quality of cellulose pellicles. Moreover, relevant patent trends of BC have also been summarized. Collaborative efforts combining optimization, metabolic and genetic engineering, and autonomous technologies will be essential in advancing the bio-industry from lab-scale to commercial production, and are particularly important for the development of bio-leather as a sustainable alternative in the fashion industry.

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