Bacterial Cellulose: A Sustainable Source to Develop Value-Added Products – A Review

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In recent decades, worldwide economic and environmental issues have prompted research scientists to re-direct their interests to bio-based resources, which are sustainable in nature. In this context, microbial polysaccharides, such as bacterial cellulose (BC), also known as microbial cellulose (MC), are some of the upcoming and emergent resources and have potential application in various bio- and non-bio-based sectors of the modern world. Many researchers have already established novel BC/MC production methods, and many new studies have been published on lab-scale and large-scale production aspects of BC/MC to date. To further expand the novel use of this sustainable source, significant progress toward the development of BC/MC has appeared in recent years. Specifically, there have been many publications and/or research reports on the valorization of BC/MC in the food, paper, materials, biomedical, pharmaceutical, and cosmeceutical industries, among others. This review will address the novel application aspects of BC/MC today, with the aim of demonstrating the importance of this sustainable and novel source in the development of value-added products.

Keywords: Bacterial cellulose; Microbial cellulose; Food industry; Cosmetic industry; Green technology

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INTRODUCTION

Cellulose is a most abundant, renewable, and widely used natural material. During the last few years, the development of cellulose-based products, with multi-functional characteristics, has gained considerable attention. There has also been interest in utilizing cellulose as a reinforcement material, to impart new or improve the existing mechanical characteristics of a product. Cellulose is a major constituent of plants. It is a homogenous, linear polymer of D-glucopyranose sugar units (Kumar *et al*. 2009; Sánchez 2009; Bertero *et al*. 2012; Iqbal *et al*. 2013) that are connected through β linkages. The average cellulose chain has a degree of polymerization of about 9,000 to 10,000 units. Cellulose possesses excellent mechanical properties, such as tensile and elastic modulus strength of approximately 16.9 GPa and 2 GPa, respectively. Approximately 65% or more of cellulose is highly oriented and crystalline and therefore is not accessible to water or other solvents. Cellulose is protected from degradation because of its close association to a sheath of matrix polymers, which include lignin and hemicellulose (Iqbal 2015).

The biotransformation from a petrochemical-based economy to a bio-based green economy necessitates a novel exploitation of natural materials that are transformable into high-value-added products for bio- and non-bio sectors of the modern world. In this context, green biotechnology could provide a noteworthy solution to this problematic issue for both bio-based green economy development and a range of value-added products of interests. The present review article focuses on the potential of utilizing microbial-based BC/MC materials, as a sustainable source that aim at the generation of green, recyclable, and sustainable products. The sustainability concept is shown in Fig. 1 (Iqbal 2015). Following a brief introduction, a part of this review mainly focuses on the potential source and various characteristics of BC/MC. In the second part of this review, various industrial and biotechnological applications of BC/MC and/or BC/MC-based materials are discussed.

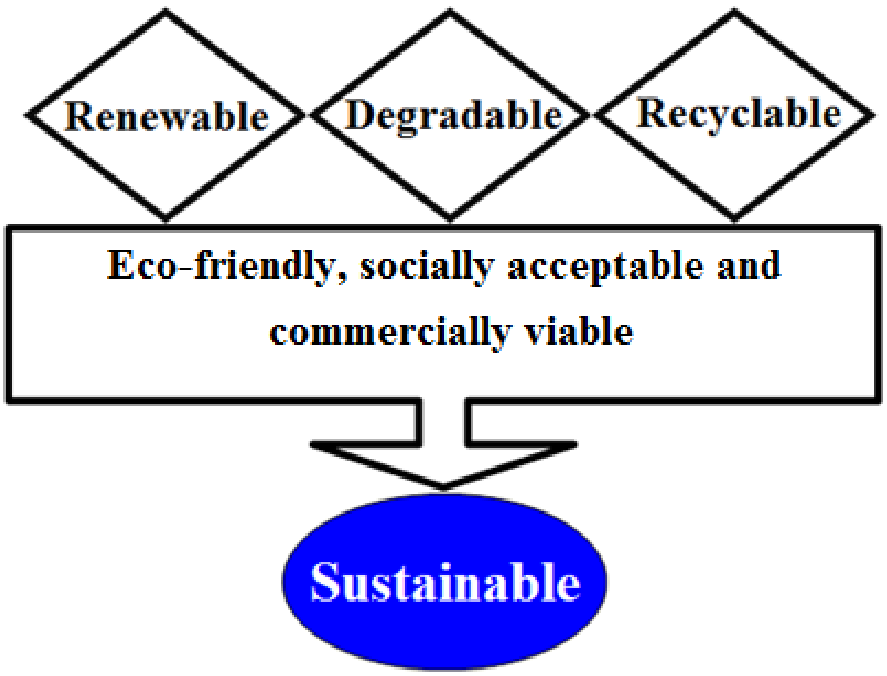

Fig. 1. Concept of "sustainability" (Iqbal 2015)

Bacterial Cellulose – Source and Characteristics

Apart from plants, cellulose is biosynthesized by certain bacteria, *e.g.*, *Rhizobium* spp., *Agrobacterium* spp., *Acetobacter* spp., and *Alcaligenes* spp. (Vandamme *et al*. 1998; Iqbal 2015; Iqbal *et al*. 2015a). The resulting cellulose is termed either bacterial cellulose (BC) or microbial cellulose (MC). Figure 2 illustrates an overview of the BC network produced by bacteria. The *Acetobacter xylinum* strain is able to produce cellulose within a temperature range of 25 to 30 °C and a pH range of 4.5 to 7.5 (Son *et al*. 2001). Many substrates have been analyzed for their potential to work as a carbon source in the production of bacterial cellulose. These include the monosaccharide, D-glucose; the disaccharide, lactose; the polysaccharide, starch; the organic acid, gluconic acid; and the alcohol, ethylene glycol (Jonas and Farah 1998; Iqbal 2015). Bacterial cellulose is a straight chain polysaccharide, with the same chemical structure as cellulose that is derived from plants. However, bacterial cellulose has the advantage of being devoid of lignin, pectin, hemicellulose, and other biogenic products that are normally associated with plant cell wall structures (Jonas and Farah 1998; Iqbal *et al*. 2014). Because of its high purity and special physicochemical characteristics, bacterial cellulose has applications in a wide range of sectors, including food, bio-medical (*e.g.*, wound care), and tissue engineering (*e.g.*, nanocomposites) (Svensson *et al*. 2005; Czaja *et al*. 2006; Shah *et al.* 2013; Silva *et al*. 2014; Iqbal *et al*. 2014; Iqbal 2015; Iqbal *et al*. 2015b, c). Therefore, in light of the aforementioned characteristics, BC/MC may be a promising candidate for the development of value-added products.

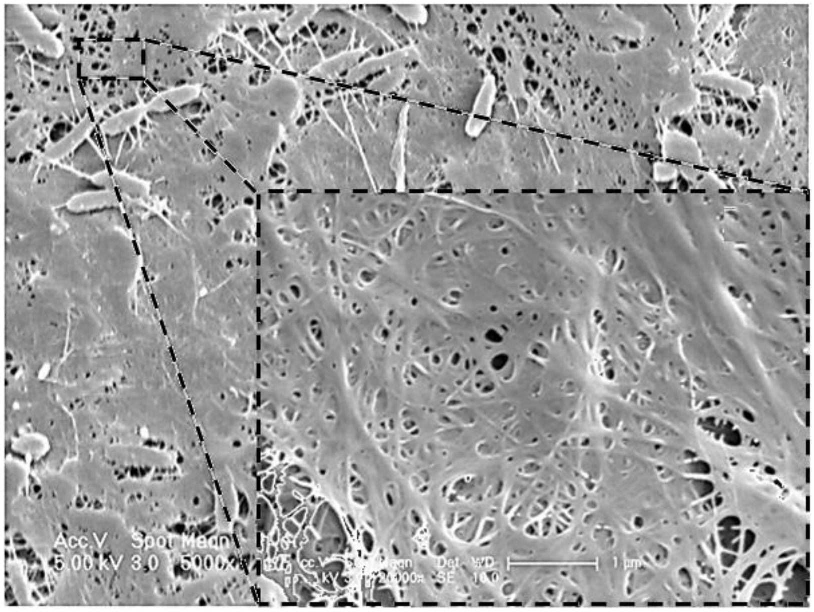

Fig. 2. Scanning electron microscope image of a bacteria-generated bacterial cellulose network (Iqbal 2015)

An important component to consider for both forms of cellulose, either plant-based or microbial-based, is their biocompatibility and biodegradability under natural conditions. Various forms of cellulose, including pure cellulose or chemically-modified cellulose, have been widely used in the modern era of research and development. For this reason, the development of bio-based products has been a subject of interest in material science from both ecological and environmental perspectives (Bajpai *et al*. 2013). Among the possible alternatives, the development of composites, utilizing cellulose as a reinforcement material, are under investigation in almost every industry. There are various methods of manufacturing bio-based products, depending on the processing techniques; *e.g.*, surface casting, ultrasonic-assisted casting, pultrusion, extrusion, injection molding, press molding, hand lay-up, filament winding, sheet molding compounding, and enzymatic grafting (Fowler *et al*. 2006; Iqbal *et al*. 2014, 2015a). Synthetic fibres, such as glass and carbon fibers, are brittle, and they are often broken into smaller fragments (Iqbal *et al.* 2013); meanwhile, cellulose is flexible and will not fracture during processing over sharp curvatures. This permits high-volume fraction filling during processing of cellulose with other polymers, which results in improved mechanical properties, compared to the abrasive synthetic polymers. All of the aforementioned features enable cellulose to maintain its desired characteristics for good performance. Moreover, cellulose offers the ability for surface modification, eco-friendly processing, non-toxic nature, easy handling, and no health risks, while most synthetic polymers pose significant health risks, such as skin irritation and respiratory disease (Yang *et al*. 2004; Iqbal *et al*. 2014). Cellulose and cellulose-based materials can be used for different applications, including food, paper and packaging, tissue engineering, pharmaceutical, cosmeceutical, electronics, dentistry, and medicine (Wang and Chen 2011; Mathew *et al*. 2012; Ul-Islam *et al*. 2012).

Potential Applications of BC/MC

From an application standpoint, a wide spectrum of microbial strains is available for the production of BC/MC, and following careful characterization, it is possible to convert BC/MC into value-added products (Iqbal *et al*. 2014; Iqbal 2015). In this context, many research scientists have already focused on designing and engineering ideal BC-based products for targeted applications. The biomedical applications of BC and/or BC-based materials have already been reviewed and documented in the recent literature (Fu *et al*. 2013; Rajwade *et al*. 2015). Figure 3 illustrates various biomedical applications of BC-based materials. However, many other potential applications of BC/MC have not been comprehensively reviewed. Therefore, the current paper reviews the applications of BC/MC in the food, paper, composite, and cosmetic industries, specifically summarizing the present research on BCs for their traditional applications in current biotechnology.

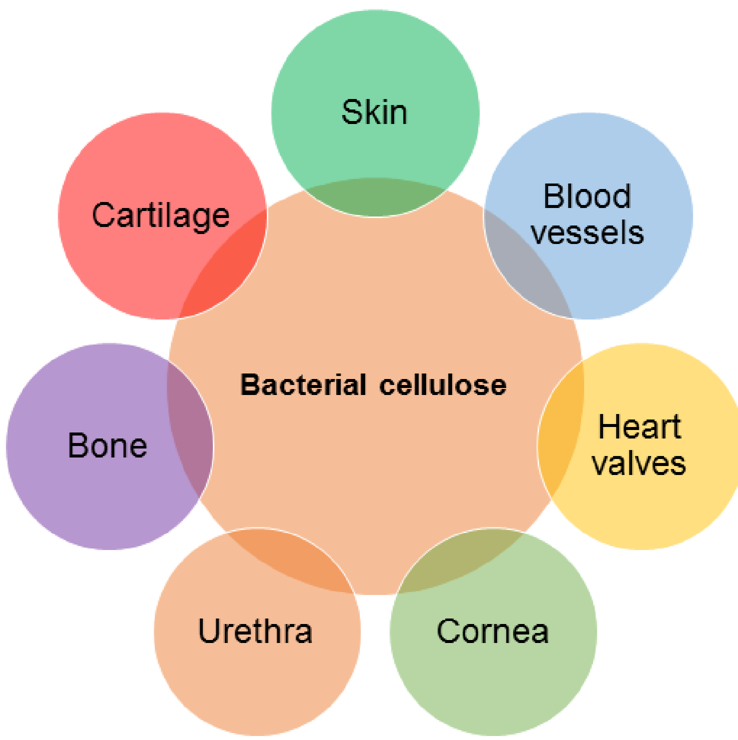

Fig. 3. Prospects for the various biomedical applications of BC and BC-based materials

Food Industry

The high level of purity, change in color, change in flavor, and enormous potential to develop various shapes and textures, makes BC/MC a potential candidate for the food industry. When compared with traditional dietary fibers, BC/MCs offer a wider range of health benefits, which is why BC has been classified as "generally recognized as safe" (GRAS) (Badel *et al*. 2011). This classification was officially accepted/approved by the United States Food and Drug Administration in 1992 (Shi *et al*. 2014).

Nata de coco and Nata de pina

The most popular use of BC in food is the production of Nata, originating from the Philippines; Nata is a traditional sweet dessert in Southeast Asia. Nata is a fermentation product of the bacteria, *Acetobacter xylinum*. Referred to as Nata de coco and Nata de pina, their flavors are controlled by the coconut water-based and pineapple water-based culture mediums, respectively (Shi *et al*. 2014; Jozala *et al*. 2016).

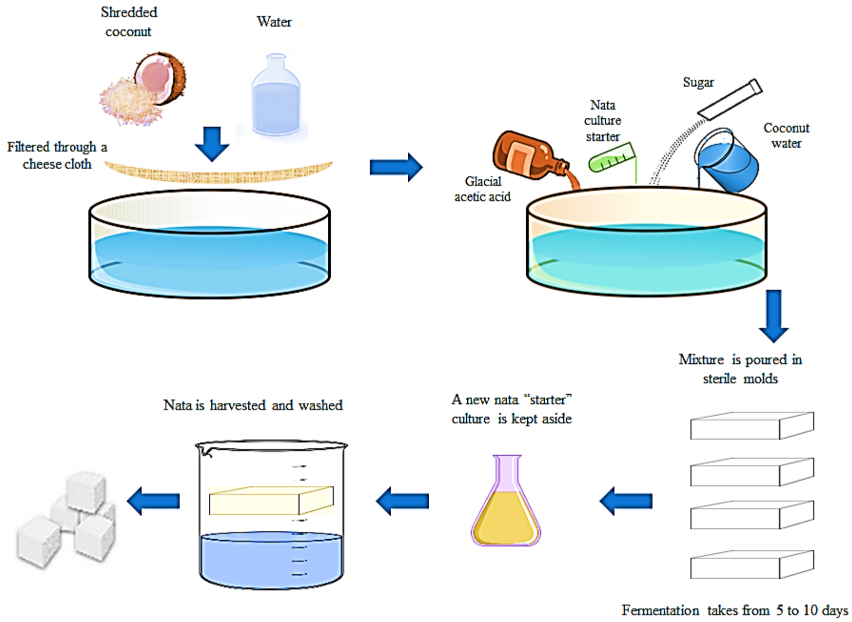

Fig. 4. Schematic representation of the Nata de coco production process

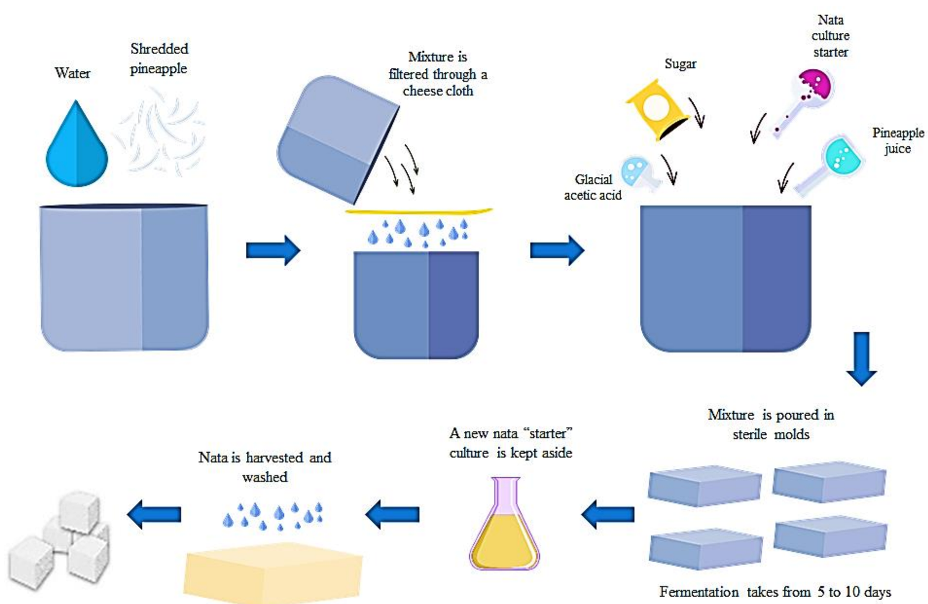

Fig. 5. Schematic representation of the Nata de pina production process

Figures 4 and 5 provide schematic representations of the production process for Nata de coco and Nata de pina, respectively. The usual way to produce Nata is a direct inoculation of *Acetobacter xylinum* into the fermentation medium. Darmawan *et al.* (2015) used a cell immobilization technique to trap *A. xylinum* in beads, and then used the beads for the fermentation of Nata de coco, repeatedly. The researchers found that the cell viability was maintained, and the production of Nata was favorable (Nugroho and Aji 2015). Lin *et al*. (2011) added Nata (with and without an alkali treatment) to mahi-mahi surimi to study its water absorption capacity and overall characteristics. They found that the addition of 5% AT-Nata (alkali-treated) improved the gel strength of the product.

Kombucha tea

Kombucha is a beverage made from fermented tea. It is prepared by soaking tea leaves, usually black tea, in boiling water, and then adding copious amounts of sugar (sucrose). Then, a "mother" biofilm is placed in the mixture. The biofilm is referred to as a symbiotic culture of bacteria and yeasts (SCOBY). The SCOBY is left in the mixture, and fermentation occurs. After several days of incubation, the tea becomes carbonated with a sweet and sour flavor and alcohol is produced. During fermentation, acetic acid is released by several bacteria, and a byproduct of this process is cellulose (Iguchi *et al*. 2000).

BC-based Nanocomposites, Silver Particles, and Self-Assembled Materials

The commonly accepted definition of a composite is a material that consists of two or more distinct materials/polymers in order to obtain tailor-made characteristics or to improve or impart ideal properties (specific strength, thermal properties, surface properties, bio-compatibility, and bio-degradability) that the individual, homogeneous material fails to demonstrate on its own (Iqbal *et al*. 2015b; 2016). When composite materials comprise one or more phases, derived from a biological origin, they are described as bio-composites (Fowler *et al*. 2006; Auras *et al*. 2011). A broad definition of a bio-composite is a composite material made up of natural or bio-derived polymers, *e.g.*, BC/MC, PHA, and PLA (John and Thomas 2008; Iqbal *et al*. 2016). So far, a range of methodologies have been successfully adopted for the production of BC and/or BC-based composites (Iqbal 2015). Furthermore, potential applications of BC and/or BC-based composites are also provided in Table 1.

Over the last few decades, there has been a continuous interest in the development of stronger, stiffer, lighter-weight, and multi-functional engineering materials for a variety of industrial and biotechnological applications. To address the demand for better performance, extensive research has been devoted to bio-based biomaterials, including BC and different polymer-based, green composites (Iqbal 2015). Research is underway on the commercial development of green technologies. Green technologies often promote new materials with high performance at affordable costs. The principle of “going green” has diverted this search towards eco-friendly materials, *i.e.*, BC/MC. Industrial ecology, eco-efficiency, and green engineering are guiding the next generation of processes and products (Markarian 2008). Bio-based polymers are moving into mainstream applications and changing the dynamics of 21st century materials. Biopolymers, bio-based, and biodegradable resources are terms that are becoming more important in the sector of industrial plastics. These materials have been a motivating factor for material scientists, because they provide potential opportunities for improving the standard of living (Nair and Laurencin 2007).

Table 1. Potential/Proposed Applications of Some Bacterial Cellulose-based "Green" Composite Materials

BC-based Materials	Methodology	New/improved functionalities	Potential/Proposed Applications	References
BC/Chi/Alg	Molding	Physical, mechanical, Biocompatibility	Wound dressing	Chang and Chen 2016
BC-Vaccarin	Immersion	Physical, mechanical, and biocompatibility	Wound dressing	Qiu *et al*. 2016
BC–xGnP	Impregnation	Thermal properties and electrical conductivity	Biosensors, tissue engineering	Kiziltas *et al*. 2016
BC-Fe_2O_3	Immersion	Magnetic behavior	Magnetic paper, loudspeaker membranes	Barud *et al*. 2015
BC–HA	Immersion	Biocompatibility	Bone tissue regeneration	Duarte *et al*. 2015
P(3HB)-*g*-BC	Laccase-assisted grafting	Thermo-mechanical strength	Bio-plastics, Biomedical	Iqbal *et al*. 2014
AMPS-*g*-BC	Ultraviolet-induced polymerization	Conductivity, effective methanol barrier	Fuel cells	Lin *et al*. 2013
BC-MMTs	Immersion	Antibacterial properties	Wound dressing, regeneration materials	Ul-Islam *et al*. 2013
BC/GO	Vacuum-assisted self-assembly	Thermal, mechanical, conducting properties	Biochemical and electrochemical devices	Feng *et al*. 2012
BC-PAni	Immersion	Electrical conductivity	Flexible electrodes, flexible display devices, bio-sensors etc.	Shi *et al*. 2012
BC-MMT	Impregnation	Physical and mechanical properties	Biomedical	Ul-Islam *et al*. 2012b
PANI/BC	Oxidative polymerization	Thermal, mechanical, conductivity	Flexible electrodes, display, sensors	Hu *et al*. 2011
BC/Chi	Immersion	Physical, mechanical, Biocompatibility	Wound dressing	Kim *et al*. 2011
ε-PL/BC	Immersion	Physical, Antibacterial	Packaging	Zhu *et al*. 2010

By controlling the culture medium, several researchers have used oriented fermentation to produce BCs in a particular shape, thickness, and structure. Some examples include BC nanotubes and a honeycomb-like structure (Zhu *et al*. 2010). Polylactide (PLA), also referred to as poly-lactic acid, is primarily used in packaging, film, and fiber applications. Despite its usefulness, there are considerable limitations regarding its potential industrial applications, mainly because of its rate of degradation and mechanical and thermal properties. γ-Methacryloxypropyltrimethoxysilane (MPS) is well known for providing adhesion to inorganic materials when coupled with other substances. For example, a copolymer with MPS-g-PLA served as a compatibilizer for solid surfaces. When bacterial cellulose was treated with the MPS-g-PLA copolymer, the results indicated that this modification provided BC with an increasing hydrophobic nature (Li *et al*. 2010).

Since PLA is used primarily for food packaging, this modification expands the range of applications in this field for bacterial cellulose (Li *et al*. 2010). Moreover, BC may also provide the ideal matrix for metals incorporation (Barud *et al*. 2008). Silver is often incorporated because of its antimicrobial properties, and adding it to BC membranes opens the possibility for the development of antibacterial textiles, medical devices, food packaging, antimicrobial filters, among others.

Barud and coworkers (2008) have reported the preparation of a BC membrane containing silver nano-particles by cultivating BC membranes from *Acetobacter xylinum* cultures and treating them with a 0.01mol/L $AgNO_3$ solution. The resulting membranes exhibited well-dispersed, spherical silver particles on the surface (Barud *et al*. 2008). Zhu *et al*. (2010) evaluated a sausage casing made of BC embedded with Ɛ-polylysine, and they found that the casing exhibited bactericidal activities against several Gram-positive and Gram-negative bacteria, thus extending the shelf life of the product. Nguyen *et al.* (2008) produced a BC film containing nisin to investigate its potential as an antimicrobial packaging device. It was tested against *Listeria monocytogenes* on the surface of vacuum-packaged sausages. Films that were produced with a high concentration of nisin (2500 IU /mL) decreased in *L. monocytogenes* count on the packaging after 14 days of storage (Nguyen *et al*. 2008).

Another technique for modifying BC membranes consists of forming nano-composites by incorporating reinforced particles into the structure of the membrane, in order to grant new properties to the BC membrane. A composite is formed by a matrix (scaffold) and the reinforcement with another substance that imparts new physio-chemical and biological properties to the matrix. For example, an electrically conducting BC has been synthesized by incorporating carbon nanotubes into its porous structure by immersing the BC membrane into an aqueous solution; the nanotubes then infiltrate the membranes' pores and provide it with electrical conductivity (Vitta and Thiruvengadam 2012). Nogi and Yano (2008) developed a BC-acrylic resin composite and demonstrated its capacity to be used as a substrate for flat-panel displays by fabricating an organic LED using the composite.

Bacterial cellulose can be used as a template for hybrid nanocomposites, especially magnetic nanoparticles that provide the BC membrane with the ability to be sensed by an external magnetic field (Barud *et al.* 2015). This type of hybrid is known as a magnetic-BC nanohybrid, and it can be used in several applications, such as filtration or purification, toxic waste remediation, and loudspeaker membranes. Recently, Barud *et al.* (2015) incorporated PEG-Fe_2O_3 magnetic nanoparticles into BC membranes, and the resulting hybrid presented magnetic behavior as expected. The MFM (magnetic force microscopy) phase images of the BC-Fe_2O_3 membrane confirmed that the nanoparticles retained their magnetic properties among the BC nanofibers network (Barud *et al*. 2015). Carbon paper has both excellent conductivity and permeability. These characteristics favor its use in fuel cells, or more specifically, in the gas diffusion layer of the polymer electrolyte fuel cell. Miyajima *et al.* (2016) used a BC-based fiber to produce carbon paper. First, metal ions were impregnated in the BC gel, and then it was dried and carbonized to obtain the carbon paper. The final product was confirmed to be electrochemically active (Miyajima *et al*. 2016).

Paper Industry

Paper is a ubiquitous product that can be used for many applications in our daily lives (Manda *et al.* 2012). The pulp and paper industry processes large quantities of cellulosic materials every year. With an increasing demand for paper and improvements in the processing technology (Singh *et al*. 2012), paper can be produced from many different cellulosic materials, including BC/MC. One of the emergent applications of bulk BC/MC is as a strength additive. Bacterial cellulose can impart or improve paper gloss, and it can reduce the grammage of paper and paper-based cardboard products. Apart from routine paper production processes, there has been a revolution in the development of BC-based electronic papers with novel characteristics through different techniques. Recently, Miyajima *et al*. (2016) developed electrically conductive bacterial cellulose-based carbon paper. Furthermore, they have also proposed this newly developed bacterial cellulose-based carbon paper as a binder-less porous carbon electrode for electrochemical applications following its electrochemical characterization *via* cyclic voltammetry. In another study, Barud and co-workers (2015) have also used bacterial cellulose as a novel material to develop bio-cellulose-based flexible magnetic paper (Barud *et al*. 2015). Mautner *et al*. (2015) demonstrated that bacterial cellulose-based nanopaper was suitable for tight ultrafiltration operations, while Li *et al*. (2015) developed a low-cost and environmentally friendly paper-based device using bacterial cellulose along with other suitable materials. Earlier, Basta and El-Saied (2009) prepared another type of flame retardant bacterial cellulose-based functional paper. In 2005, Shah and Brown used various strains of *Acetobacter* for the production of BC. In the same study, it was found that BC exhibited several advantages over synthetic paper, *i.e.*, it was completely pure, had higher dimensional stability, and exhibited higher water retention. Their research sought to improve the conductivity of BC-based membranes so they could be used as substrates for electro-optic effects, to integrate electronic dyes into the structure for color change, and to fabricate a single-pixel device using BC film to represent a full-scale study (Shah and Brown 2005). Yet earlier, Serafica *et al*. (2002) produced BCs in a rotating disk bioreactor, and added several types of particles to the medium to form new composite materials. Among them, calcium carbonate and talc were both immobilized within the BC membrane and produced a stronger yet suppler material. In addition, scraps of copier paper and newspaper (without any ink removal) were added and it was determined that a composite film, containing up to 90% scrap paper on a daily basis, could be produced (Serafica *et al*. 2002).

Cosmetics Industry

In recent years, interest in BC/MC has increased steadily because of its enormous potential for many applications in the biomedical, pharmaceutical, and/or cosmeceutical sectors (Hornung *et al*. 2009; Fu *et al*. 2013). A cosmetic is defined as a "product that is applied to the human body for cleansing, beautifying, promoting attractiveness, or altering the appearance without affecting the body structure or functions" (Hasan *et al*. 2012). However, skin allergies and contact dermatitis from exposure to cosmetics are a constant problem. According to a study published in the *Journal of the American Academy of Dermatology*, almost 70% *i.e.*, 661 of 945 patients had at least one positive reaction to skin allergens from personal skincare (Wetter *et al*. 2010). Thus, natural skincare products have been introduced to counteract these issues. It has been reported that bacterial cellulose can be used in cosmetic formulations to produce stable oil-in-water emulsions without irritating the skin. Because BCs provide a high degree of hydration and can penetrate the skin, they can be added to moisturizing creams. Other applications for BCs are as a component of fingernail polish and artificial nails. The rheological properties of BCs provide stability to suspensions. There are also studies reporting the use of cellulose in powders. Additional patents describe the use of bacterial cellulose for dissolving cosmetic compositions in face masks, using BC produced by *A. xylinum*, and mentions that the BC was able to dissolve powder faster than an acylic hydrogel (Gama *et al*. 2013). Furthermore, work by Procter & Gamble describes a cleansing composition containing a network of BCs that, along with a cationic polymer, produce a cleansing product that provides good lathering and can be easily rinsed off without producing an “undesirable slimy” feel (Gama *et al*. 2013).

There is little information available on the use of BCs in the cosmetic industry. However, the Hainan Guangyu Biotechnology Co. Ltd. (Hainan, China) is one of the few bacterial cellulose producers and is becoming one of the first traders of BC for cosmetic application. Hansan *et al*. (2012) have analyzed the rheological behavior of a facial scrub with added BC. The results indicate that the final product has a similar viscosity as a commercial facial scrub, with the added benefit of having ingredients that do not harm the skin (Hasan *et al*. 2012). Numata *et al.* (2015) studied the possibility of introducing PEO-b-PCL (poly (ethylene oxide)-b-poly caprolactone) nanoparticles in a BC-based gel to encapsulate hydrophobic particles. If successful, the BC gel could release nanoparticles and the encapsulated hydrophobic particles. A BC gel from *Gluconacetobacter xylinus* was obtained, and PEO-b-PCL nanoparticles were prepared using solvent evaporation and acetone. Finally, the retinol-loaded PEO-b-PCL particles were analyzed based on their stability and release of encapsulated retinol. Their results indicated that this product has the capacity to release retinol, thus allowing it to be used in the cosmetic field (Numata *et al*. 2015).

CONCLUDING REMARKS AND FUTURE PROSPECTS

1. The microbial polysaccharide, bacterial cellulose (BC), has the potential for many applications in industries, such as food, paper, and cosmetics, and it has the potential to meet various demands/trends of the modern world.
2. Although there are noteworthy applications for bacterial cellulose/microbial cellulose or bacterial cellulose-based materials, particularly in the food industry, many of these applications have not been fully explored, especially the use of bacterial cellulose in Nata.
3. Bacterial cellulose is a sustainable source material that offers significant opportunities in the area of industrial biotechnology and bio-medical sectors to produce value-added products.
4. Recent trends indicate that bacterial cellulose-based novel materials are improving products through the green chemistry route.
5. To overcome the current demands for green, value-added products, bacterial cellulose should remain the focus of future research.

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