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REVIEW ARTICLE

Commercial and potential applications of bacterial cellulose in Brazil: ten years review

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Abstract

In the last decade, bacterial cellulose (BC) has received considerable attention around the world, including in Brazil. The unique properties of BC, such as mechanical stability, tensile strength, thermostability, crystallinity, purity and biocompatibility make it a promising candidate for commercial applications in different areas. This article provides a comprehensive synthesis of commercial applications and studies related to BC around the world and shows the importance and development of Brazilian research during the last decade. In this review we present an overview of BC structure, biosynthesis and possible applications of BC mainly in the food, electronics, bioengineering, cosmetics and biomedical areas. The most significant contributions of Brazilian researchers using BC have been carried out in the biomedical area. Despite the increase in BC reserch, Brazil also needs to develop strategies to expand the use and commercialization of BC products, for which government financial support is extremely necessary.

Keywords: *bacterial cellulose, bacterial cellulose applications, biomedical, Brazil, electronics.*

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1. Introduction

During the last century, massive exploitation of fossil resources and pollution problems increased concerns related to the economy and the environment. In this context, polymers from renewable sources, like polysaccharides, among others, have received considerable and growing attention[1]. Cellulose $(C_6H_{10}O_5)_n$ is the most abundant renewable biopolymer produced in the biosphere, being basically composed of glucose monomers connected by β (1-4) glycosidic bonds. It can be synthesized by plants, animals and microorganisms[2,3].

Cellulose derived from plants is commonly incorporated into other biopolymers as hemicellulose and lignin; therefore, aggressive chemical treatments are necessary to remove these impurities[4]. On the other hand, BC produced by microbial fermentation is characterized by higher purity, and its purification is relatively simple, not requiring energy or chemically intensive processes. Furthermore, due its unique physical and chemical properties, BC has been successfully applied in the food, biomedicine, textile, and papermaking fields, as well as in biosorbent material and acoustic diaphragms[5,6].

However, the interest in cellulose is not only limited to industrial fields; it has also become increasingly relevant and interesting in academic areas[7]. Figure 1 shows an explosive increase in the number of publications related to BC since 2011. The years 2017, 2018, 2019 and 2020 have been the most productive in terms of publications. In Brazil, many groups have contributed and emerged in several areas of cellulose. In biomedicine, for example, Brazil was a pioneer, and gained prominence in employment of BC as an artificial skin to replace burned skin. Besides the commercial application in the biomedical area, potential applications of BC in Brazil include studies in the electronic/electrochemical/magnetic field, food and food packaging, cosmetic area, bioengineering, as well as in reinforcement material to make blends, composites and nanocomposites.

The aim of this review is to open with a sketch of the major cellulose-producing microorganisms and to provide a comprehensive discussion of cellulose synthesis. We then move to commercial applications of BC and the potential applications of biopolymers in Brazil and around the world by considering their uses ranging from food to medical industries.

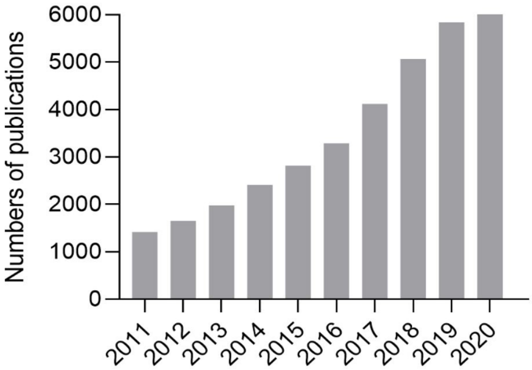

Figure 1. Evolution of the number of bacterial cellulose-related publications around the world, including Brazil, between 2011 and 2020. The searches were performed with Google scholar using the topics: bacterial cellulose, biocellulose, microbial cellulose and Brazil.

1.1 Synthesis, properties and production of BC

The BC consists of a transparent and gelatinous pellicle, produced in the vast majority by the Gram-negative bacterial cultures of *Acetobacter*, *Agrobacterium*, *Achromobacter*, *Aerobacter*, *Sarcina*, *Azotobacter*, *Rhizobium*, *Pseudomonas*, *Salmonella* and *Alcaligenes*[8]. Among them, the most efficient BC producer belongs to the group of acetic acid bacteria (AAB) previously known as *Acetobacter xylinum*[9], which was recently transferred to the newly proposed genus *Komagataeibacter* and named *Komagataeibacter xylinus*[10].

It is suggested that cellulose production is a bacterial defense mechanism that can provide protection from hazardous ultraviolet light radiation effect and can help bacteria move to the aerobic environment of surface. Furthermore, the pellicule retains moisture and confers mechanical, chemical, and physiological protection[11,12]. The biosynthesis of BC involves several biochemical processes containing a large number of enzymes, and the regulation of these key enzymes controls the cellulose production by means of three metabolic pathways: the pentose cycle, or branched hexose monophosphate pathway (HMP), for the oxidation of carbohydrates; the tricarboxylic acid cycle (TCA), for the oxidation of organic acids and other compounds; and the Embden-Meyerhof-Parnas pathway (EMP)[13,14].

Figure 2 briefly describes the biosynthetic pathway of *K. xylinus* cellulose production from glucose and fructose, although several other substrates can be used for this purpose[17].

Cellulose synthesis from glucose occurs initially through phosphorylation of hexose by GHK, thereby producing Glucose-6-Phosphate (G6P). G6P is metabolized by the Hexose monophosphate pathway, since Fructose-6-phosphate cannot be converted to Fructose-1,6-phosphate. Next, G6P is metabolized to UDP-glucose (UDPG), a direct precursor of the biopolymer, by PGM and UGP enzymatic action[15]. The fructose can be metabolized to cellulose following two paths: (I) phosphorylation of fructose to Fructose-6-phosphate (F6P) by FHK, and (II) phosphorylation of fructose to Fructose-1-phosphate (F1P) by PTS. In the first, PGI changes F6P to G6P, which can be used for cellulose production or can proceed to the Hexose monophosphate pathway. In (II) 1PFK transforms PF1 into Fructose-1,6-diphosphate (FDP) and it is later dephosphorylated to F6P. Through the EMP, FDP can also be metabolized, starting the whole process of cellulose synthesis[18].

Microscopic analysis has shown that from one bacterial cell a single cellulose ribbon is produced (Figure 3). The single ribbon is constituted of a structure of cellulose microfibrils that are excreted from a row of a complex of proteins called the terminal complexes (TCs), located in the outer cell membrane[19,20]. During the conversion of glucose to UDPG in cytoplasm, a row of 60 TCs conducts the recognition and synthesis of UDPG, as well as the crystallization and extrusion of cellulose fibers[19].

Komagataeibacter species are able to produce two forms of cellulose: (I) cellulose I, the ribbon-like polymer; and (II) cellulose II, which is the most thermodynamically stable amorphous form of the polymer. While cellulose I is constituted of parallel β (1-4) glucan chains organized uniaxially with van der Waals forces, β (1-4) glucan chains of cellulose II are arranged in random way. The latter is mainly antiparallel, with a large number of hydrogen bonds that provide more stability to the structure[21]. According to Koizumi et al.[19], during the BC synthesis, the amorphous regions, interspersed among crystalline regions, occupy 90% of material total volume.

Due to its microfibrillar structure, several mechanical properties are attributed to BC, such as: tensile strength, which may vary between 200-300 MPa with Young's modulus, up to 15-35 GPa; and thermal stability, reaching temperatures above 100 °C without changes in their biophysical properties[17,22,23]. Furthermore, BC presents high purity, high degree of polymerization, water holding capacity up to a hundred times its weight, excellent biodegradability and biological affinity and non-allergenicity[11,24-27]. The structure, physical, and mechanical properties of BC are directly related to the cultivation technique employed, of which, the main methods of production are static, agitating culture, and the airlift reactor [16,28].

Despite its unique properties, the production of BC is still limited due to difficulties in the large-scale production, which are related to long-time cultivation methods, low production yields, bacterial strain mutation and high cost of the process. The static method is the simplest and widely used to produce BC, mainly in lab-scale[29]. This technique presents high cost, long cultivation time (~7-20 days), low productivity, and uneven distribution of nutrients, oxygen and cells, resulting in a material with non-uniform thickness[30]. On the other hand, the agitation method favors the diffusion of oxygen and the availability of nutrients, promoting greater production and shorter cultivation time (~5 days). However, it is associated with mutation and formation of non-cellulose-forming cells[31,32].

Studies have also been conducted to find strains with high capacity for BC synthesis, besides employing genetic engineering techniques to improve the production[33-35]. Alternatively, bioreactors can be employed for industrial-scale production of BC, since they show higher yield compared to static and agitated methods. Some bioreactors developed use oxygen, rotating disc and can produce BC through biofilm

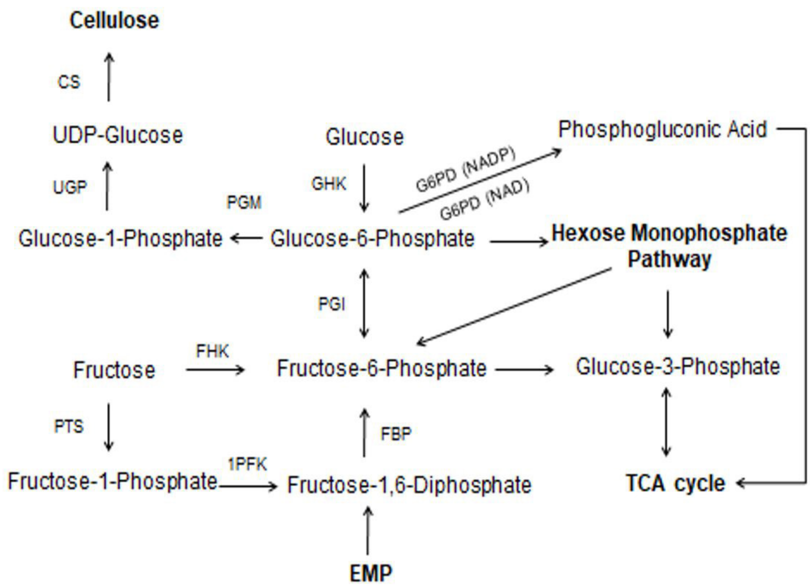

Figure 2. Pathway of cellulose synthesis from glucose and fructose in *K. xylinus*, also containing the direct path from Glucose-6-Phosphate to Glucose-1-Phosphate. GHK glucose hexokinase, FHK fructose hexokinase, 1PFK fructose-1-phosphate kinase, FBP fructose bisphosphatase, PGI phosphoglucose isomerase, PGM phosphoglucomutase, UGP UDP-glucose pyrophosphorylase, G6PD glucose-6-phosphate dehydrogenase, PTS phosphotransferase system; CS cellulose synthase; EMP Embden-Myerhoff pathway; HMP Hexose Monophosphate pathway[13,15,16].

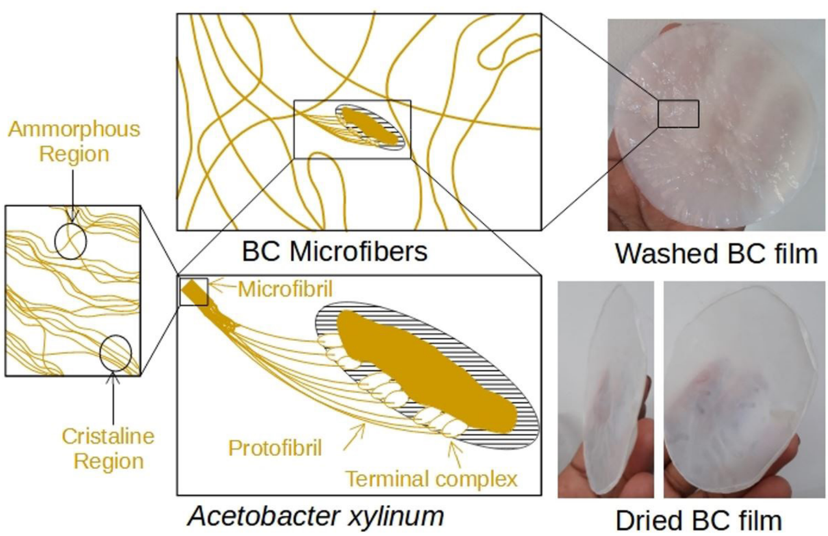

Figure 3. Schematic diagram of microbial cellulose organization. Adapted from Picheth et al.[8] and Koizumi et al.[19].

support. This method still requires studies that allow the use of controlled parameters and low-cost substrates[29,36].

To find new cost-effective fermentation media to replace the expensive Hestrin-Schramm medium (standard medium), researchers have checked the use of media containing either sugary materials or even agricultural and industrial waste products[37]. In such media containing high sugar content, are included the fruit juices[38] and related by-products such as fruit peels[12,39] and rotten fruit[40]. Molasses derived from the industries of sugarcane[41], beet[42] and soybean[43] have also been tested for production. Other several agricultural and industrial residues already tested include waste beer yeasts, fruit bagasse, cheese whey, glycerol and textile wastes[44]. These industrial by-products, rich in sugars and other nutrients could be suitable fermentative substrates for the production of BC, since they have low-cost and good sources of carbon[45]. By using these strategies, it is possible not only to make the BC production more economically feasible, but also make it more ecologically sustainable, which could help to reduce the release of waste by industries. Other strategies include screening of high-yield strains, inhibition of mutation, controlled side product formation, selection of suitable cultivation methods, optimization of medium composition, and searching for low-cost raw materials[31].

1.2 Overview of commercial applications of BC

Considering all the properties of BC, it is expected that this polymer will have several industrial applications. In fact, the number of patent applications registered around the world has been quite impressive, but unfortunately, few commercial applications have been pursued[15].

Among the most known industrial applications of BC is the "nata de coco", a traditional food consumed in the Philippines and other countries in Southeast Asia. The manufacturing process of this jelly-like product consists of using coconut water as a fermentation medium for cellulose production. The pellicule is then cleaned, washed, chopped and immersed in sugar syrup, to be served as a dessert. Originally from Southeast Asia, nata de coco became a very popular food worldwide[15,46,47]. Japan and the USA are the largest markets for nata de coco; between 2009 and 2011, the export volume from the Philippines reached an average of 6000 MT (Metric Tons)[48].

Potential application of BC has also been reported in the acoustic transducers area; BC presents remarkable shape retention ability, measured as the Young's Modulus, and also bears two essential properties for speaker diaphragms: high sonic velocity and low dynamics loss. Thus, novel diaphragms have been marketed by Sony Corp. as headphones[16,46].

Regarding applicability of BC in biomedicine, Brazil participated in one of the main applications associated with wound dressing, in which Fontana et al.[49] became pioneers in employing BC as an artificial skin to replace burned skin. BioFill® and Bioprocess® (BioFill Produtos Biotecnológicos – Curitiba, PR, Brazil; used as temporary skin in the treatment of burns and ulcers) and Gengiflex (BioFill Produtos Biotecnológicos; applied in periodontal diseases) are examples of Brazilian commercial products of BC-based wound healing systems that have been launched. Although these launches are no longer on the market, other products as NEXFILL, DERMAFILL and CUTICELL EPIGRAFT (Seven Indústria de Produtos Biotecnológicos Ltda – Londrina, PR, Brazil), Biocel (DMC Importação e Exportação de Equipamentos LTDA - São Carlos, SP, Brazil; Florida USA), Bionext® (Bionext® Produtos Biotecnológicos Ltda – São Paulo, Brazil), and Membracel® (Vuelo Pharma – Curitiba, PR, Brazil), emerged, thereby demonstrating the relevance of BC application in this area. Among other products available around the world are CelMat® Wound (BOWIL Biotech sp. - Poland), Bio-skinG (Coreleader Biotech Co. Ltd. - New Taipei City Taiwan) and XCell (Xylos Corporation – US; used to maintain an ideal moisture balance). The prerequisites for BC to be applied as a wound dressing include: high mechanical strength in a wet state, water vapor permeability, good adherence to the wound, low cost, good biocompatibility, durability, transparence and easy handling[15,16,50].

The increase in cardiovascular diseases has also led the researchers to reflect about the need for replacement blood vessels[51]. Klemm et al.[52] have developed a clinical product from BC and patented it as BASYC®-tubes (BActerial SYnthesized Cellulose). Besides their suitability for different inner diameter vascular conduits, studies show BASYC tubes have high mechanical strength in a wet state, great water retention properties and can be successfully used to replace carotid arteries in rats, pigs and sheep[50,53].

1.3 Reports applications of BC around the world and in Brazil

The studies related to the potential applications of BC in Brazil, as on the world stage, involve the biomedical area, electronic/electrochemical/magnetic field, food and food packaging, bioengineering and the cosmetics area (Figure 4 and 5). The studies also involve the use of reinforcement material to make blends, composites and nanocomposites, with possible application in many fields. For such applications, the properties of BC have usually been tailored by using various *in situ* techniques (such as addition of various substances and alteration of culture conditions) and *ex situ* strategies (physical and chemical modification)[54,55]. These applications are discussed in detail in the following sections.

1.3.1 Food and food packaging

1.3.1.1 World

Considered a dietary fiber and classified as "generally recognized as safe" (GRAS) by the USA Food and Drug Administration in 1992, BC can offer several health benefits, reducing the risk of chronic diseases such as cardiovascular disease, diabetes, obesity and diverticulitis[17,47]. Most of the approaches use BC as a raw material for obtaining new products and explore its use as food additive.

In this area, BC has been used as a fat substitute; as potential gelling, thickening, suspending and emulsion stabilizer, as solid support to immobilize cells and as food packaging. As a fat substitute, reports are found in the literature of BC application in meatballs[56], surimi products[57], cheese[58], ice cream[59] and mayonnaise[60]. BC also has been applied as a potential gelling, thickening, suspending and emulsion stabilizer to produce meat products[61], whey protein isolate[62], olive oil[63] and edible foam[64].

Recently, BC has gained prominence in studies related to the immobilization processes of cells, enzymes and probiotics for application in food[65]. Some authors immobilize yeast in BC for wine production. Immobilized yeast reduced expenses for inoculum preparation, since the yeast was recovered and separated at the end of the fermentation process[66]. Similarly, Fijałkowski et al.[65] obtained promising results in immobilizing probiotic strains of *Lactobacillus spp* in BC.

In the literature, the most explored food area worldwide is the development of films and packaging. BC has been incorporated into a wide variety of substances in order to increase the shelf life of food products[67]. Among the substances to which BC has been incorporated for the production of packaging can be mentioned: cotton fibers[68], postbiotics of lactic acid bacterium[69] and potato peel[70]. Other BC applications in the food field can be found in Table 1.

1.3.1.2 Brazil

In Brazil, the research groups have focused more on the development of edible films and food packaging. Viana et al. [108] produced films with different ratios of nanofibrillated

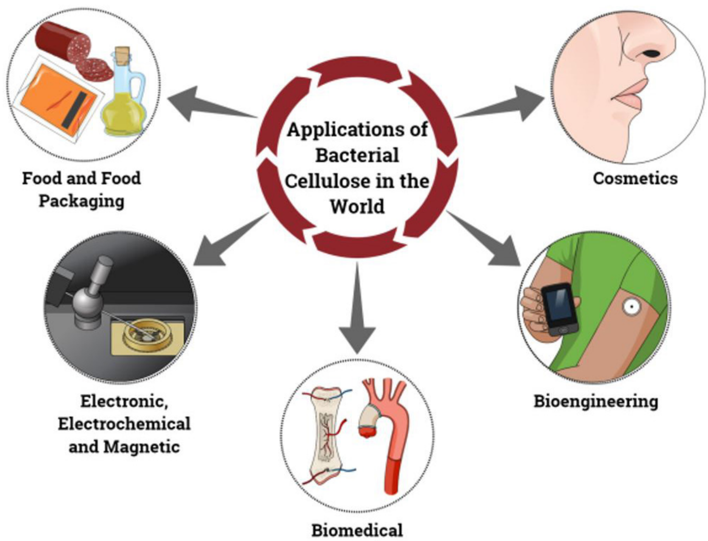

Figure 4. Main applications of bacterial cellulose in the world.

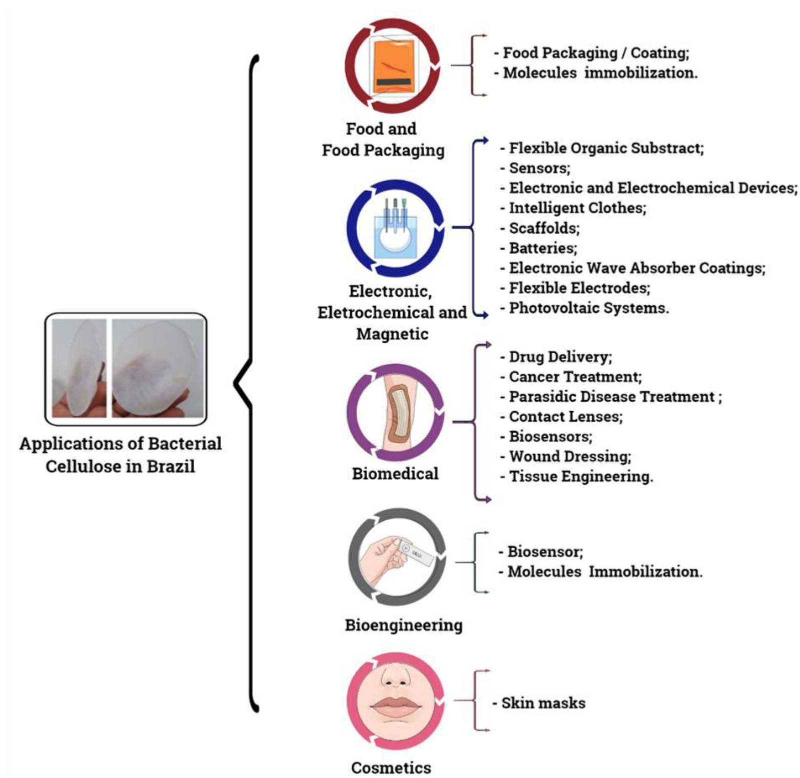

Figure 5. Main applications of bacterial cellulose in Brazil.

BC (NFBC) to pectin, with or without the addition of fruit purees. In their study, films (with or without purees) with higher NFBC contents showed improvement in physical properties and were proposed for use in food wrapping or coating.

Studies conducted by Malheiros et al.[109] show that the immobilization of antimicrobial peptides from *Lactobacillus sakei subsp. sakei* 2a in BC is a promising strategy for the control of *Listeria monocytogenes* in foods. Similarly, a new material composed of BC/poly(3-hydroxybutyrate) with the addition of clove essential oil demonstrated a reduction of 65% in microbial growth and attractive properties for active food packaging[110].

In order to develop active composite films from cashew by-products, Sá et al.[111] produced BC from cashew juice and then they added nanocrystals of lignin and cellulose (both from cashew pruning fiber) to produce the film. Although lignin gave brown color and opacity, the films showed good

Table 1. Exemples of BC applications in the world.

Type	Description	References
Food	BC can be used as a fat substitute; as solid support to immobilization of enzymes and cells; as food packaging and as potential gelling, thickening, suspending and emulsion stabilizer.	Akoğlu et al.[71], Bandyopadhyay et al.[72], Jayani et al.[73], Razavi et al.[74]
Electronics, electrochemical and magnetic fields	Due to its porous nanofibrous network structure, BC can be used as a flexible matrix for developing biomaterials.	Fei et al.[75], Guan et al.[76], Kim et al.[77], Xie et al.[78]
Antimicrobial activity	After being combined with substances with antimicrobial effects, BC can be used to protect against infections and contaminations.	Żywicka et al.[79], Adepu and Khandelwal[80], Horue et al.[81], Sajjad et al.[82]
Drug delivery	BC can be used for drug delivery due to its water holding capacity and controlled release of substances.	Badshah et al.[83], Beekmann et al.[84], Inoue et al.[85], Luo et al.[86], Weyell et al.[87]
Wound dressing	BC can be used to reduce pain, maintain moisture and serve as a bacteriological barrier in patients with burns and chronic wounds.	Gupta et al.[88], Faisul Aris et al.[89], Ye et al.[90], Moradi et al.[91]
Tissue engineering	Since BC's morphological structure has a high degree of purity, excellent biocompatibility and high tensile strength, BC can be used to support newly formed tissues, interact with cells and release substances necessary for cell growth.	Halib et al.[92], Frone et al.[93], Zhang et al.[94,95]
Bioengineering	BC can be used as biosensors to detect and quantify certain analytes in several areas. The immobilization of enzymes and cells into BC has been promising for effluent treatment, biomedical area and food manufacturing.	Hu et al.[96], Moradi et al.[97], Cai et al.[98]
Cosmetics	Due to its controlled release of substances, water holding capacity and ability to stabilize emulsions, BC can be used to produce cosmetic products.	Wang et al.[99], Muhsinin et al.[100], Amnuaikit et al.[101], Aramwit and Bang[102]
Filtration/ adsorption	BC combined with materials specific materials can be used to remove contaminants, heavy metal and impurities from water.	Urbina et al.[103], Zhuang and Wang[104], Núñez et al.[105], Shoukat et al.[106], Luo et al.[107]

mechanical properties and interesting antioxidant capacity. Other works involve the preparation of biodegradable films made from blends of potato starch/BC/ glycerol[112] and BC/ polycaprolactone acetone solution[113].

1.3.2 Electronic, electrochemical and magnetic field

1.3.2.1 World

In electronic field, BC has attracted great attention mainly due to depletion of non-renewable resources[114]. Moreover, due to its porous nanofibrous network structure, BC is used as a flexible matrix for developing biomaterials with desired properties. BC-based materials have been developed using conducting polymers, graphene, graphene oxide, carbon nanotube and carbon nanofiber (Table 1). Their application occurs especially in flexible supercapacitors, ion battery, fuel cell, and other electrochemical devices[115].

Actually, several works have combined BC nanofibers with conducting polymers as polyaniline and polypyrrole. When Graphene/Carbon Nanotube/Bacterial Cellulose (RGO/CNT/BC) architecture was designed as substrate for loading polypyrrole (PPy), Bai et al.[116] showed that flexible supercapacitor exhibits stable electrochemical performance under bending and flat conditions. Similarly, Rebelo et al.[117] synthesized an electrically conductive BC/Polyvinylaniline/ Polyaniline (BC/PVAN/PANI) nanobiosensor for potential use in nerve regenerative medicine, which demands both electroactivity and biocompatibility. BC has also become a valuable for production of lithium-ion batteries (LIB).

Yuan et al.[118] designed free-standing films of tin sulfide (SnS) nanosheets distributed uniformly on carbonized BC (CBC) nanofibers. LIB using SnS/CBC as anode could be a promising electrode material, since exhibited high capacity, excellent cycling stability and high-rate capability. In order to develop proton exchange membranes (PEMs) for application in polymer electrolyte fuel cells (PEFC), Vilela et al.[119] combined BC (support), fucoidan (polyelectrolyte) and tannic acid (cross-linker). The membranes presented thermal-oxidative stability (180-200 °C), good dynamic mechanical performance and the protonic conductivity increased with the increase in relative humidity. The authors concluded that BC/fucoidan membranes have potential as eco-friendly alternatives to other PEMs for application in PEFCs.

1.3.2.2 Brazil

In Brazil, Müller et al.[120] carried out an *in situ* oxidative polymerization of EDOT on nanocellulose fibers to obtain PEDOT-nanocellulose flexible composites. The nanocomposites showed an increase in electrical conductivity and in elongation at break, with a lower thermal stability when compared to pure BC. Therefore, the authors concluded that the flexible membranes have potential use for flexible organic electronics.

In a recent study, Legnani et al.[121] produced multifunctional membranes based on BC and an organic-inorganic sol, composed of boehmite (Boe) nanoparticles and epoxy modified siloxane (GTPS), to be used as substrates in organic light-emitting diodes (OLEDs). Then, they covered the BC/Boe-GTPS with silicon dioxide (SiO2) and indium tin oxide (ITO), thereby obtaining eco-friendly, biocompatible substrates comparable to fabricated commercial glass substrate.

Conductive composite membranes of BC and polyaniline, using different oxidizing agents like FeCl3.6H2O[122] and ammonium peroxydisulfate[123], may allow for important technological applications, such as sensors, electronic devices, intelligent clothes, flexible electrodes and tissue engineering scaffolds. Other interesting approachs exploit the intrinsic properties of BC, combined with the physical and chemical characteristics of compounds such as nanoparticulated Boe and GTPS[124], polypyrrole[125] and laponite[126], to produce new materials that may be suitable for biomedical and electronic applications. Other studies are shown in Table 2.

1.3.3 Biomedical area

1.3.3.1 World

Most BC studies are currently focused in the biomedical field, where the multifunctionality of this polymer has been shown mainly through the development of biomaterials as wound dressing, scaffold for tissue engineering and drug delivery[135] (Table 1).

BC membranes have several advantages that allow it to be considered as an ideal wound dressing material. It forms a bacteriological barrier that allows for gas exchange and reduces pain, it maintains moisture of the environment, its transparency favors direct visualization of the wound, and it also reduces the treatment time and the costs of hospitalization of patients with burns and chronic wounds[8,136]. The direct use of BC itself has no antimicrobial activity to prevent wound infection[137]; therefore, recently, several studies have incorporated active compounds into its structure to improve the properties and functionalities.

Cao et al.[138] evaluated the potential use of a structured surface BC biomaterial incorporated with human urine-derived stem cells for use as wound dressing. The *in vivo* results were promising for this combination, since the healing rate was significantly higher compared to the control. BC was also incorporated with sodium alginate, chitosan and copper sulfate, in order to provide an antimicrobial for use as wound dressing. The material obtained showed antimicrobial activity against methicillin-resistant *Staphylococcus aureus* and *Escherichia coli*[139]. In another study, BC was combined with poly(methylmethacrylate) (PMMA) in order to obtain biocompatible and biodegradable bandages to support wound healing. Preliminary analyses of swelling characteristics and mechanical properties indicated that this biomaterial is a promising candidate for biomedical applications[140].

Tissue cells, scaffold and growth factors are extremely important factors in the success of tissue engineering. Scaffolding is used to support the newly formed tissues, interact with cells and release substances necessary for cell growth. Since BC's morphological structure has a high degree of purity, excellent biocompatibility and high tensile strength, BC has been extensively studied as a candidate for tissue engineering[141]. Ahn et al.[142] developed a structure of BC coated with hydroxyapatite (HA) for bone tissue regeneration. The bone regeneration capacity was assessed using a rat calvary defect model. The results showed that both scaffold (BC coated with BC and HA) were effective in bone formation derived from existing bone, in addition, a new bone was found inside the scaffold.

Torgbo and Sukyai[143] synthesized a nanocomposite by combining BC, hydroxyapatite (HA) and magnetite nanoparticles (Fe_3O_4) through ultrasonic irradiation. The scaffold was not toxic to mouse fibroblast cells and was also biocompatible to osteoblasts attachment and proliferation.

Table 2. Summary of studies of BC applications in electronics/electrochemical/magnetic field.

Material	Main results of biomaterials based on BC	References
Castor oil based polyurethane (PU)	The BC/PU composites offer new opportunities as substrates for flexible electronic displays, since they exhibit great transparency and excellent mechanical properties.	Pinto et al.[127]
PEG (polyethylene glycol)-Fe2O3 magnetic nanoparticles	Atomic force microscopy height and phase images of BC/Fe_2O_3 nanocomposites confirmed that the addition of Fe_2O_3 nanoparticles did not change the ability of the BC to form nanofiber networks.	Barud et al.[128]
Boehmite	The impregnation of Boehmite through BC pores improves transparency of pure BC, suggesting applications in optics and electronics fields.	Salvi et al.[129]
Triethanola-mine (TEA)	BC–TEA membranes showed potential application as ion-conducting membranes, since the addition of TEA avoids complete dryness, ensuring humidity necessary for maintaining high ion-conductivity of the membranes.	Salvi et al.[130]
Switchable photoluminescence liquid crystal	4'-(hexyloxy)-4-biphenyl carbonitrile, 4'-(hexyl)-4-biphenyl-carbonitrile and multicomponent nematic mixture were used to impregnate BC film. This resulted in a transparent film with 3D rigid structure.	Tercjak et al.[131]
Polyaniline and Ammonium persulfate (APS) or iron(III) chloride (FeCl3)	BC membranes were polymerized with polyaniline in the presence of APS or $FeCl_3$. The membranes with $FeCl_3$ displayed higher conductivity and better mechanical performance than membranes pure or prepared with APS.	Marins et al.[132]
Polyurethane (PU)	BC/PU nanocomposites present higher thermal stability, a non-crystalline character and low water absorption when compared to control.	Pinto et al.[133]
Cadmium tellurite quantum dots (CdTe) capped with glutathione (CdTe-GSH)	CdTe-GSH/BC membranes were tested as photoelectrodes and exhibited maximum photocurrent around 530nm, indicating potential application as flexible electrodes, sensors and photovoltaic systems	Pinheiro et al.[134]

Aiming to promote the formation of blood vessels, Wang et al.[144] modified BC/gelatin (BC/G) scaffold with heparin and studied its skill of promoting angiogenesis in terms of vascular endothelial growth factor (VEGF) release. The scaffold provided prolonged VEGF release. Proliferation and migration (*in vitro* cellular assay) were observed in the presence of VEGF. Furthermore, heparinized scaffolds loaded with VEGF (V-BC/G/H) improved the angiogenesis compared to BC/G scaffold.

During the past few years, the number of studies related to the incorporation of drugs into BC membranes has increased. The most common drugs to be loaded in BC are non-steroidal anti-inflammatory drugs (NSAIDs) and antibiotics[30]. Shao et al.[145] loaded BC membrane with tetracycline hydrochloride (TCH). The biomaterial showed biocompatibility, high antibacterial activity and it was not toxic in HEK293 cells. Similarly, BC, PVA and chitosan mono and multilayer films were effective for controlled release of ibuprofen sodium salt[146].

1.3.3.2 Brazil

In Brazil, although considerable research has been devoted to wound dressings and tissue engineering, the incorporation and modification of BC with countless substances has further expanded the spectrum of applications in the biomedical area (Table 3). For example, BC and carboxymethylcelullose (CMC) biocomposites loaded with methotrexate were developed in order to evaluate their effectiveness as a drug delivery system, as an alternative for the topical treatment of psoriasis. The results showed that different amounts of DS-CMC can generate distinct biomaterials, to be applied through the cutaneous route at different stages of evolution of a pathology[160].

In the case of application for cancer therapeutics, photodynamic therapy (PDT) has emerged as an innovative therapeutic modality, focused mainly on skin cancer treatment. Peres et al.[161] utilized chloroaluminum phthalocyanine (ClAlPc) as a photosensitizer and impregnated BC membranes with it, with the goal of developing such applications as a drug delivery system for PDT skin cancer protocols. Photophysical studies of BC-ClAlPc showed that the properties of photosensitizer were not affected, and the result of the cell viability test using Chinese hamster ovary cells (CHO-K1) demonstrated their potential for safe biological use. Another study of great relevance is related to Leishmaniasis, a group of parasitic diseases caused by protozoa of the genus Leishmania. Brazil, together with nine other countries, is responsible for 70-75% of the global Cutaneous Leishmaniasis (CL) occurrence. In this context, Celes et al.[162] described a topical formulation by incorporating Diethyldithiocarbamate (DETC), a superoxide dismutase 1 inhibitor, into BC membranes for CL treatment. BC-DETC did not cause noticeable toxic effects and resulted in parasite killing. The topical formulation significantly reduced the lesion size, inflammatory response at the infection site and parasite load, highlighting its availability for treatment of CL.

Dengue is an endemic disease widespread throughout the tropics, in which the regions most affected are the Americas, South-East Asia and Western Pacific[163,164]. According to the World Health Organization[164], in Brazil, the average of number of reported dengue infections was over 100 thousand between 2010-2016. Since misdiagnosis is still a problem[165], Picheth et al.[163] proposed that diagnostic assays were more sensible, faster and less expensive for this disease. For this purpose, piezoelectric sensors were coated with thin BC nanocrystals (CN) in order to facilitate anchoring of monoclonal immunoglobulin G (IgGNS1) against NS1 dengue antigen. The authors observed that biosensors, compared to cellulosic surfaces, increased the total IgGNS1 immobilized mass by twofold and reduced the need for sample dilution by tenfold. Lastly, they concluded the sensors can be used qualitatively in clinical diagnosis after suitable validation.

In the ophthalmological area, Coelho et al.[166] developed and evaluated the cytoxicity, genotoxicity and mutagenicity of contact lenses based on BC, coated with either glycidoxypropyltrimethoxysilane (H) or chitosan (Q), incorporating ciclodextrin (CD) to release diclofenac sodium (DS) or ciprofloxacin (CP). Functionalized BC lenses safely allowed for the bioavailability of ophthalmic drugs, in which only BC-H-CD-DS presented cytotoxic and genotoxic effects and BC-Q-CD-DS showed cytotoxic effects. Therefore, the authors suggested other specific tests with corneal lineage to ensure safe ophthalmologic use. Additionally, unmodified BC also proved to be a useful material in medical displacement procedures of the vocal folds. Souza et al.[167] demonstrated that laryngeal medialization with BC in the larynx of rabbits did not cause rejection or absorption and it was stable over a long period.

In order to obtain antimicrobial properties, Brazilian researchers have incorporated different antimicrobial to BC. For example, silver nanoparticles composites[137,168] (cerium nitrate and silver nanoparticles[136], copper nitrate ($Cu(NO_3)_2$)[169] and ceftriaxone[170]) have been incorporated in BC membranes and exhibited strong antimicrobial activity against Gram-negative (*Pseudomonas aeruginosa*, *Salmonella* and *Escherichia coli*) and Gram-positive (*Staphylococcus aureus*) bacteria, which are commonly found in skin infections.

The antimicrobial activity and the wound healing properties of novel BC containing Brazilian propolis was also demonstrated by Wei et al.[171], Marquele-Oliveira et al.[172] and Picolotto et al.[173]. The first two authors proved the antimicrobial efficacy of *in vitro* BC/propolis membranes against Gram-negative and Gram-positive bacteria; their *in vitro* studies suggested that the biomaterial may promote fast re-epithelization and tissue organization, setting up a potential therapy for infected wounds. The accelerated wound healing process in a diabetic mouse model was also evidenced by Picolotto et al.[173].

Other approaches favoring wound healing have also been developed. Picheth et al.[51] assembled a novel wound dressing sensitive to lysozyme by depositing nanopolymeric chitosan and alginate films onto oxidized BC membranes incorporated with epidermal growth factor (EGF). The proposed system proved to be effective as wound dressing and presented a local delivery mechanism to recognize infections and to respond with a burst of EGF release. Wound dressing based on BC/collagen hydrogel in rat dorsum stimulated better wound healing than commercial collagenase and control group (untreated wound)[174].

As previously commented, BC has also become a promising biopolymer for tissue engineering and regenerative

Table 3. Summary of studies of potential applications of BC in the biomedical area.

Purpose	Therapeutic materials	Main results of biomaterials based on BC	References
General biomedical applications	Hydrocolloids (guar gum and hyaluronic acid) and collagen	Films developed from BC, hydrocolloids and collagen showed potential for applications as bioactive wound dressings, scaffolds for cellular growth and sustained drug release systems.	Woehl et al.[147]
	Bionanocomposites based on alternative sources (cashew juice and sisal liquid waste) and hydroxyapatite (HA)	BC from agro-waste juices were similar to the standard culture medium. *In vitro* studies showed that materials were biocompatible and proved to be suitable for the preparation of nanocomposites for applications in the biomedical field.	Duarte et al.[148]
	Silk fibroin (SF)	Presence of SF in BC nanostructure induced a significant increase in cell adhesion and could be an excellent option in bioengineering, since the material was non-genotoxic and safe for medical applications.	Barud et al.[149]
	Succinic anhydride	Succinylation over the surface of the BC showed the potential for conjugations with molecules of medical interest, such as hydrolyzed collagen.	Ribeiro-Viana et al.[150]
	Aloe vera portions	The incorporation of *Aloe vera* extract fractions during the BC synthesis resulted in biocomposites with mechanical properties and chemical composition distinct from pure BC. The material could be used as a scaffold for skin substitution and regeneration and cell culture substrates.	Godinho et al.[151]
	Polyethylene glycol (PEG)	The BC/PEG films showed more hydrophilic properties, supporting possible use as biomedical devices.	Silva et al.[152]
	Arabinogalactan (AG) / Xyloglucan (XG)	Incorporation of AG and/or XG into BC mechanically defibrillated membranes caused a change in the hydrophilic surface characteristics. AG inclusion also showed viability and cellular adhesion profiles similar to those of commercial BC.	Lucyszyn et al.[153]
	Resistant starch/pectin (RS/P)	CB-RS/P nanocomposite films showed an increase in thermal stability. The films also enhanced the drug dissolution rates, and therefore may be a potent material used as a carrier of poor solubility drugs, which may enhance oral bioavailability.	Meneguin et al.[154]
	BC membranes with IKVAV peptide	Mimics the structural architecture and biological functions of the extracellular matrix for vasculogenic mimicry of human melanoma cells.	Reis et al.[155]
Tissue engineering	Dimethylacetamide/lithium chloride	Modification of BC structure by acetylation and/or oxidation has developed films with more degradability *in vivo* and potential applications as scaffold material for tissue engineering.	Lima et al.[156]
	Otoliths/collagen (O/C)	O/C/CB new scaffolds should be applied as: inducers of vascularization; facilitators of deposition of otoliths in predefined regions; guides for the regeneration of tissue, allowing the development of different tissues; or inhibitors of calcification and cell adhesion.	Olyveira et al.[157]
	Glycidyltrimethylammonium chloride (GTMAC)	BC/GTMAC films showed a significant increase in cell attachment and spreading compared to unmodified membranes.	Courtenay et al.[158]
	BC and collagen associated with osteogenic growth peptide.	The membranes promoted cell proliferation and alkaline phosphatase activity in osteoblastic cell cultures, in addition to better bone regeneration *in vivo* than the control group.	Saska et al.[159]

medicine applications; many studies have been conducted to synthesize new biomaterials based on BC (Table 3). Saska et al.[175] developed and evaluated the biological properties of bacterial cellulose-hydroxyapatite (BC-HA) nanocomposite membranes in noncritical bone defects in rat tibiae at 1, 4, and 16 weeks. BC-HA composites have presented properties similar to that of physiological bone and have accelerated new bone formation of rat tibiae, without showing inflammatory reaction. Furthermore, the authors concluded that the membranes exhibited slow reabsorption, suggesting that the material takes longer to be completely reabsorbed. Afterwards, Saska et al.[176] demonstrated that the peptide (osteogenic growth peptide (OGP) and its C-terminal pentapeptide OGP (10-14)) incorporation did not change the BC properties. Furthermore, *in vitro* assays revealed BC membranes influenced osteogenic cell proliferation and do not present cytotoxic, genotoxic or mutagenic action.

Similarly, Coelho et al.[177] associated BC, HA and anti-bone morphogenetic protein antibody (anti-BMP-2) (BC-HA-anti-BMP-2), and did not observe toxicity of the membranes in MC3T3-E1 cells. BC-HA-anti-BMP-2 increased the expression of genes related to bone repair, the mineralization nodules and the levels of alkaline phosphatase activity when compared to the control group. Biocompatibility tests of BC, BC-HA and PTFE (polytetrafluoroethylene) using rats (Wistar) complemented prior studies[175,176] which displayed that BC and BC/HA materials have the same inflammatory pattern when compared to PTFE, thus proving to be biocompatible materials[178].

Composites based on collagen have demonstrated improvement of the biological and mechanical properties in bone tissue engineering. This protein is plentiful in the natural extracellular matrix (ECM) and in the human body, in addition to stimulating the regeneration process[179]. Based on this, Saska et al.[180] developed a composite based on BC and type I collagen (COL) and evaluated the *in vitro* bone regeneration. BC-COL presented a more flexible structure than BC membranes, and showed osteoblastic differentiation that was observed by way of higher levels of alkaline phosphatase activity. Aiming to functionalize BC-COL with other proteins and/or peptides and promote bone formation, the group later synthesized and evaluated *in vitro* the biomaterial based on BC, COL, apatite (Ap) and OGP or OGP(10-14). The nanocomposites (OGP/OGP(10-14)-BC-COL-Ap) produced did not display cytotoxic, genotoxic or mutagenic action, and *in vitro* tests showed a synergism between the elements that provided cell growth regarding the BC-Ap nanocomposite. The authors consider that (BC-COL)-Ap associated with OGP peptides might be potential candidates for bone tissue engineering applications .

Recently, Birkheur et al.[181] prepared aminoaryl mannoside and conjugated it to a succinic group of BC without disrupting the microfibril network. The use of glycoconjugates to BC showed good fibroblast compatibility. Conversely, Souza et al.[182] developed films from mechanical defibrillation of BC followed by the dry-cast generation and incorporation of the xyloglucan (XGT), extracted from tamarind seeds, at various percentages. According to the authors, both mannosylated cellulose[181] and BC combined with hydrocolloids[182] demonstrated promise as biomaterials for this area.

1.3.4 Bioengineering area

1.3.4.1 World

In bioengineering area, BC has been used mainlny for bioanalysis, enzyme and cell immobilization and to produce biosensors. BC based biosensors have been explored for different applications. These biomaterials have been used in the food and biomedical areas and also to detect contaminants in the environment. Several studies have provided advantageous results to detect contaminants in aqueous matrices, such as bisphenol A in effluent[183] and heavy metal traces as Cd (II) and Pb (II) in drinking water[184]. Pollutant biosensors for detection of H_2O_2 in the environment[185] and formaldehyde vapors in houses and workplaces[186] were also developed showing low detection limits. In foods, BC has been used as an optical sensor for detection and determination of ethylene concentration in fruits[187] and potential biosensors for detecting and measuring the growth of pathogenic bacteria[188]. Other studies are related to the development of colorimetric pH indicators through the incorporation of anthocyanin from several sources into BC. These indicators demonstrate to be ideal candidates to monitor the freshness/spoilage of foods and beneficial for further development of smart indicator films for practical use[189].

Many researchers have also applied BC as a carrier of enzymes and cells since the immobilization method provides greater stability, reusability, more tolerance to changes in environmental conditions and less vulnerability to toxic substances present in the surrounding[190]. In biomedical area, BC based biosensor for dopamine detection in human urine was used successfully[191]. When enzymes such as lysozyme were immobilized onto BC, Bayazidi et al.[192] verified the system obtained good antimicrobial activity against several microorganisms, with potential applications in water treatment or food industry.

Aiming to develop an innovative treatment to remove the paracetamol from wastewater, Żur et al.[190] immobilized *Pseudomonas moorei* KB4 onto BC, since this strain is one of the few bacteria able to degrade the analgesic drug. Using the Real-Time PCR technique, they verified that paracetamol exposure influenced the expression of the selected genes encoding the degradation enzymes and KB4 strain was able to degrade 150 mg L-1 of paracetamol in the three cycles. Żywicka et al.[193] immobilized rod-shaped bacteria *Lactobacillus delbruecki*, spherical-shaped yeast *Saccharomyces cerevisiae* and hyphae forms of *Yarrowia lipolytica*) onto BC. As a result, the authors concluded that carrier must be individually combined to the cell type, considering mainly the carrier's porosity parameter.

1.3.4.2 Brazil

In Brazil, studies in this area still need to be explored. However, recently, Vasconcelos et al.[194] developed a new process of purification (via alkaline treatment with K2CO3) and chemical modification (NaIO4 oxidation) for covalent immobilization of papain into BC. Oxidized BC (OxBC) demonstrated no cytotoxicity and a greater amount of immobilized enzyme than BC alone, with recovered enzyme activity of 93.1%. The authors immobilized papain in BC by surface response methodology, exhibiting 53% of the generalized papain activity. They concluded that the biomaterial facilitate debridement of skin wounds[195].

In another study, Gomes et al.[196] developed a flexible biosensor for detection of lactate in artificial sweat by immobilizing lactate oxidase (Lox) into BC. The biosensor displayed excellent amperometric response to lactate in artificial sweat, high sensitivity, superior mechanical resistance and biocompatibility; offering new opportunities for the development of wearable devices.

1.3.5 Cosmetics

1.3.5.1 World

In addition to the bioengineering area, scientists have studied the cosmetic application of BC in face masks for delivery of active compounds and increased skin hydration, as well as an emulsion stabilizer[197]. *In vivo* studies conducted by Perugini et al.[198] demonstrated that BC masks loaded with different bioactive ingredients (peptides, natural extracts and biopolymers) can be used as an effective delivery method for revitalization of facial tissue.

Likewise, Stasiak-Różańska and Płoska[199] used BC as a biocarrier for 1,3-dihydroxy 2-propanone (DHA) also for cosmetic purposes. The biomaterial showed as an alternative in masking the effects of vitiligo, without leaving unpleasant odor, typical of commercial cosmetics containing DHA. The application of BC is attracting attention of the cosmetics industry and scientific community[197] (Table 1).

1.3.5.2 Brazil

In Brazil, as well as in the world, this area has been few explored[200]. BC was successfully loaded with different cosmetic actives for skin treatment and then evaluated through sensorial tests carried out by humans. The sensory tests revealed that masks based on BC were effective for skin adhesion and handling, and the actives improved the skin moisture of the volunteers[201]. Amorim et al.[114] aiming to develop a biomask that helps in the healing of inflammations caused by acne, loaded BC with natural propolis extract. The dermatological and cosmetic products improved the hydration and texture skin, accelerated the healing process and improved the self-esteem of acne patients.

2. Conclusions

BC stands out as a versatile biomaterial that allows promising applications in the areas of food, electronics, bioengineering, cosmetics and biomedics. In Brazil, the areas of food, bioengeering and cosmectics are still scarce for economic reasons. In the food field, Brazilian studies are essentially focused on the development of food packaging. Although a large amount of research have been developed worldwide in the electronic/ electrochemical/ magnetic fields, in Brazil, research in this area has only started to be more explored recently. Certainly, the most significant contributions of Brazilian researchers using BC have been made in the biomedical area. We highlight here the use of BC to treat psoriasis and cutaneous leishmaniasis, and the development of sensors in the clinical diagnosis of dengue. In addition, the incorporation of antimicrobials, polysaccharides, proteins / peptides and other compounds into BC has shown promising results in wound healing properties. In tissue engineering and regenerative medicine, the immobilization of biomolecules and their potential *in vitro* and *in vivo* is still being explored for greater activity and stability. In conclusion, Brazil is one of the countries that most develops research using BC. However, the expansion of the use and commercialization of BC products could be increased through improvements in its productivity, using, for example, residues generated in Brazilian agribusiness, which could contribute to an environmentally friendly society. Studies with new methods and technologies for the production of cellulose need to be explored, in addition to new biochemical and genetic investigations. Greater government financial support for Brazilian research is also sorely needed.

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