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Recent research on economic production and water absorption improvement of bacterial cellulose: A review

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ABSTRACT:

Bacterial cellulose (BC) has been applied in various industries, like the hygiene and medical industry, due to its functional properties (water absorption properties, mechanical strength, biodegradability, and nanostructure). However, the use of BC instead of petrochemical products in the hygiene industry is associated with challenges such as high production costs and low water absorption performance. The type of microbial strain, the development of a cheap culture medium using food industry waste, and the optimization of the production type and conditions are among the most important strategies that have been used in recent years to increase production yield and reduce BC costs. Also, the presence of different hydroxyl functional groups in each repeating unit of the BC chain makes it possible to make various changes to increase its water absorption capacity by using in-situ and external modifications and adding different chemical or biological compounds to make it competitive with chemical polymers. In this article, an attempt has been made to encourage researchers and industries by reviewing the recent research related to reducing production costs and increasing the water absorption power of BC to make more efforts towards the possibility of replacing the chemical polymer with a biodegradable and environmentally friendly biopolymer.

KEYWORDS: *Bacterial Cellulose, culture medium, Production Yield, optimizing, Modification, water absorption*

INTRODUCTION

Due to the nanostructures, the simplicity of the production process, and better mechanical properties than plant cellulose, BC is rapidly expanding its use in some industries such as health and medicine, despite its higher price [1-3]. Because of functional groups, small diameter, large surface area, and high porosity, BC has held hydrophilic compound capacity. According to Kang et al., BC has 3.2 times higher water-holding capacity than commercial alpha-cellulose [4]. Also, BC is used in food as a fat or calorie reducer because it has high water absorption, cation exchange capacity, and cholesterol reduction effect [5]. BC gel has a hard structure like bone tissue. However, due to the increased water content, it becomes edible when combined with alginate, calcium chloride, or sugar alcohol. These features allow its use in foods [6]. Also, the hydroxyl functional groups in each repeating unit of the BC chain have enabled in-situ and ex-situ modifications. Tome et al. synthesized BC-esterified membranes with improved water barrier properties that can be used in the packaging industry [7]. Modification of BC improves its properties such as water absorption and can be used in health and medical industries as a natural superabsorbent and wound dressing [8,9].

Considering the BC properties and applications, BC industrial production has received more attention in the last decade. However, BC industrial production has challenges such as low production yield and high production price. Another BC limitation compared to the corresponding petrochemical materials is its water absorption properties. BC is swollen when cultivated from the culture medium and holds about 100 times water from its dry weight. After BC drying, its ability to absorb water decreases in comparison to the initial state [10]. Some strategies to overcome these limitations are using food industry waste as a BC culture medium, investigating BC bacterial strains producing capability and gene modification, optimizing BC production conditions, and combining some materials with BC during and after its cultivation (BC modification) [11]. The BC culture medium, especially the carbon source, determines the BC fermentation costs because the culture medium can take up to 65% of the total production costs. Many studies have been conducted to apply and optimize the rich and cost-effective BC culture medium [12]. Babeaipour et al. have used optimized food industry waste to produce BC, reduce production costs, and increase the BC production yield [13,14].

After maximizing BC production with an economical price, the improvement of BC water absorption by modifications was examined, in past studies. As mentioned, the functional group in BC chains makes in-situ and ex-situ modification possible. By improving the BC water absorption properties, and due to its excellent biocompatibility, porosity, water retention capacity, gas exchange ability, and thermal insulation, it can be used in hygiene and medical industries as wound dressing, artificial blood vessels, and vascular grafts [1-3,8,9]. Modified BC dressing prevents fluid loss and infection, while pure BC has more hydrogen bond networks between and within monomers and prevents gas permeability. Lin et al. examined the therapeutic effects of BC-chitosan composite and TegadermTM hydrocolloid dressing on wounds. This study showed that the BC-chitosan composite forms a better environment for wound healing than other samples due to its water absorption capacity and maintains a suitable moist environment [8]. Das et al. studied BC wet dressing containing antibiotics. They synthesized a composite by adding polycaprolactone to BC and functionalized with streptomycin and gentamicin antibiotics, which healed infected wounds as a wound dressing [9].

The purpose of this study is to present a database of different strategies for cost-effective increasing the BC production yield and then improving its water absorption properties for medical and hygiene applications such as wound dressing, sanitary pads, and baby diapers. In the following, the culture medium components, including additives, carbon and nitrogen sources, and modifiers, are reviewed.

CULTURE MEDIUM ADDITIVES TO INCREASE THE BC PRODUCTION YIELD

Some substances have been used in the BC culture medium to increase production yield. Table 1 summarizes these substances' effects on BC production yield [9,15-17]. The nutrient consumption metabolism of different strains has been the subject of various research to clarify the importance of the culture medium composition in the BC metabolic pathways production. For example, Zahan et al. reported that A. xylinum 0416 used part of glucose as a cellulose precursor and energy source, and another part converted into gluconic acid by dehydrogenase enzymes attached to the bacterial membrane [18]. This process reduces the overall BC production yield by lowering the pH to sub-optimal levels for cell viability. The use of acetic acid in the culture medium leads to a pH increase compared to the time not used, followed by a rise in BC production [19,20]. Some additives cause changes in the metabolism pathway of culture medium consumption and increase the BC production yield. These compounds include ethanol, citric acid, vegetable oils, vitamins, surfactants, pectin, etc., whose impacts on BC production yield are reviewed (Table 1) [9,15,17].

Table 1. The culture medium's additives effect on the BC production

Additive	*Effect of substance on BC production*	*Bc production yield*	*Reference*
Silicone polyether surfactant	***Reduction of surface tension and increase BC production yield.***	***Over 30% for BC wet mass and 15% for BC dry mass increased***	***[21]***
Acetate buffer	***Maintain proper PH to produce BC***	***Increased BC production from 0.66 g/L to 3.56 g/L***	***[22]***
Ethanol and sodium citrate	***Weakening the tricarboxylic acid (TCA) cycle and improving the efficiency of BC synthesis.***	***increased BC production 1.49 times***	***[23]***
Vegetable oil	***The oil reduces the friction force between the BC and the vessel wall and facilitates the process of membrane sinking. Hence the availability of nutrients and the formation of new cellulose layers on top of the primary layer increases.***	***Exceeding 500% increase in BC production***	***[24]***
Ethanol	***Reduction of byproducts that affect BC production negatively. (For example, by reducing the glycerol production.) Improve the production of ATP. Suppress the spontaneous mutation of the strain into non-cellulose-producing species. Lysing the cell wall, which leads to easier release of cellulose.***	***279% increase in BC production***	***[25]***
Acetic acid	***Acetobacter can oxidize acetic acid to CO_2 and water, so produce additional ATP and create a favorable pH range.***	***Reached to up to 28 g/L BC production***	***[19]***
Lignosulfonate	***Increasing crystallinity index and Ia cellulose in the case of BC produced by adding lignosulfonate***	***57% increase in BC production***	***[26]***

	Polyphenol compounds in lignosulfonate prevent the formation of gluconic acid		
Mesoporous halloysite nanotubes	***Increasing oxygen supply to bacteria and facilitating BC production by stabilizing bacteria on nanotubes***	***increased BC production from 2.2 to 5.9 g/L***	***[27]***

Another carbon source besides glucose in the BC culture medium increases the BC production yield due to the alternative carbon source consumption (for example, ethanol) and a change in the mechanism pathway consumption of the culture medium. This change in the metabolite pathway leads to the by-product production and BC reduction yield, so the carbon sources amount should be under control and optimized. For example, ethanol consumption by bacteria leads to acetic acid production, whose low level in the culture medium increases the BC production to protect the bacteria from acidic damage. While more than a certain level of acetic acid, despite the bacteria's resistance, it has a toxic effect on bacterial cells. Also, acetic acid acts as a catalyst in the glycogen synthesis pathway and arranges more glucose in this pathway [28]. In the study of Tian et al., the use of mesoporous halloysite nanotubes as an additive that increases BC production was investigated. They found that BC bacteria immobilization on mesoporous halloysite nanotubes improved fermentation and increased oxygen availability [27]. Polymers, including polyvinyl alcohol, alginate, polyethylene glycol, and carboxymethyl cellulose (CMC), were used as additives and modifiers in BC production [29]. These additives avoid clumping and coagulation of BC and increase the BC production yield [30]. On the other hand, this additive in culture medium increases the cost of production, so the use of a cost-effective culture medium rich in materials needed by bacteria for cellulose production was considered.

RICH AND COST-EFFECTIVE CULTURE MEDIUM FOR BC PRODUCTION

Cost-effective culture mediums like agricultural and food industry wastes or by-products are among these items. CSL is a byproduct of the wet corn milling process, containing 21%-45% protein, 20%-26% lactic acid, approximately 3% carbohydrates, and a small amount of fat (0.9%-1.2%). Lactate in CSL acts as an energy source and enhancer for BC synthesis. Lactate is converted into pyruvate by the oxidation process, and the energy from this process can increase BC production. This result was confirmed by Matsuoka et al. They concluded lactate helps bacterial growth by providing the required energy, improving the TCA cycle and the respiratory chain. In a study, CSL along grape pomace helped to increase BC production, because it not only acts as a nitrogen source (in the proteins, amino acids, amines, and ammonia form) but also organic acids, carbohydrates, vitamins and there are also minerals. It has also been reported that it has a buffering effect on the culture medium [31]. According to another report, adding 8% CSL to the culture medium increased BC production by 47%. Kim et al. reported that 4% glucose (carbon source) by 10% CSL (nitrogen source) increased BC production yield approximately three times more than HS.

Vinasse is a concentrated solution obtained from the anaerobic fermentation of sugarcane and the main waste of the fermentation ethanol industry. This black liquid is produced 10 to 15 times more than ethanol. Vinasse is a mixture of water, organic compounds, minerals, and elements that remain after various operations in ethanol production. According to a report, vinasse contains 93% water and 7% solids, of which 75% is organic matter [32]. Food waste due to glucose and fructose content are nutrients as BC culture medium. Various additives such

as fruit juices, minerals, and organic materials are used to modify the HS culture medium [33,34]. Babaipour et al. reduced production costs and increased BC production yield using food industry byproducts as a culture medium [35,36]. Beilichi et al. have used hydrolyzed beans and carob as a BC culture medium [37]. According to their study, Phaseolus vulgaris (bean) contains proteins, minerals (calcium and magnesium), and carbohydrates (75% sucrose, and the rest is fructose, maltose, and glucose). Carob also contains protein and minerals [37]. Gendi et al. investigated the hydrolyzed prickly pear peels culture medium for BC production, which supported about 2.94 g/L BC production and increased to 6.01 g/L by optimization of the BC production conditions [38]. Several studies have been examined in Table 2.

Table 2. Investigating BC production yield in various culture mediums

Waste	*Nutrients*	*Type of process*	*Production yield (g/L) (Based on Dry weight)*	*Time (day)*	*Reference*
Vinasse (a byproduct of the ethanol industry)	*-*	*Static*	*0.28*	*11*	*[32]*
Beverage industrial waste of citrus peel and pomace	*-*	*Static*	*5.7*	*8*	*[33]*
Fruit juice (apple, orange, pear ...)	*Peptone, Yeast extract, citric acid*	*Static*	*5.9*	*14*	*[34]*
Pecan nutshell		*Static*	*2.816*	*28*	*[39]*
Sugarcane and pineapple pulp	*-*	*Agitated*	*4*	*13*	*[40]*
Tobacco waste extract	*-*	*Static*	*5.2*	*16*	*[41]*
Durian shell hydrolysate	*-*		*2.67*	*10*	*[42]*
Carob and haricot bean	*-*	*Static*	*3.2*	*10*	*[37]*
Potato peel wastes	*-*		*4.7*	*6*	*[43]*
Rotten fruit	*-*	*Static*	*60 (wet weight)*	*4*	*[14]*
Coffee cherry husk	*CSL and urea*		*8.2*	*7*	*[44]*
Agrowastes	*Other HS components (except glucose)*		*7.7*	*15*	*[45]*
Tomato juice	*-*		*7.8*	*7*	*[46]*
Acetone- butanol- ethanol (ABE) fermentation wastewater		*Static*	*1.3*	*7*	*[47]*
Maple syrup		*Agitated*	*1.5*	*-*	*[48]*
Date syrup	*Other HS components (except glucose)*		*5.8*	*10*	*[49]*
Different fruit juice	*Other HS components (except glucose)*		*3.9*	*15*	*[50]*
Orange pulp	*-*		*2.8*	*7*	*[46]*
Konjac powder	*Peptone, and Yeast extract*	*Static*	*2.12*	*23*	*[51]*

Wastewater from rice wine distillery		***Static***	***10.4***	***-***	***[52]***
Distillery effluent	***HS components***		***8.5***	***8***	***[53]***
Waste from the beer culture broth	***-***		***8.46***	***14***	***[54]***
Wheat straw		***Static***	***8.3***	***-***	***[55]***
Wine industry residues	***-***		***6.7***	***30***	***[31]***
Glycerol recycled, and wine production residue		***Static***	***10*** ***8***	***-***	***[56]***
Acidic food industry byproducts	***-***		***6.19***	***3***	***[57]***
Spruce hydrolysate		***Static***	***8.2***	***7***	***[58]***
Byproducts of the cider production	***-***		***2.5***	***14***	***[59]***
Saccharified food wastes		***Static 30 l*** ***Agitated 10 l***	***18*** ***16.8***	***-***	***[60]***

Many of these culture mediums mentioned in the above table have additives such as ethanol, vegetable oils, citric acid, and lactic acid, which were mentioned in the previous section as BC production enhancers. For this reason, some of these culture mediums have a significant BC production yield [61]. Of course, other conditions besides the type and amount of rich culture medium, like strain type, environmental conditions, and process type (static or agitated), affected BC production. In the following, these factors were discussed.

TYPES OF BC PRODUCER SPICES

Different bacteria strains like *Gluconacetobacter, Achromobacter, Rhizobium, Alcaligenes, and Agrobacterium* can produce BC. Also, the free enzyme complex (without the cell) can synthesize BC [62,63]. The microorganism is effective in the BC production yield and its properties like mechanical strength, structure, fiber size, degree of polymerization, and Crystallinity index. According to Table 3, strain of *Gluconacetobacter. xylinus* PTCC1734 shows the highest production rate among different species.

Bacterial cellulose synthase (BCS) produces BC by catalyzing the uridine diphosphate glucose polymerization reaction. This enzyme operon is encoded and includes *bcsA, bcsB, bcsC,* and *bcsD* BC synthesis genes, which *bcsA* and *bcsB* belong to polypeptide chains, and *bcsC* and *bcsD* belong to crystallizing and transporting BC fibrils to the extracellular matrix, respectively. Several studies have been done to increase the BC yield through genetic manipulation. These studies are mostly regarded as adapting strains to the cost-effective culture medium, removing or modifying by-product-producing genes, and generally increasing BC production yield to the commercial level [76].

Table 3. Different species producing BC

Spices	*Culture medium*	*BC production yield (g/L)*	*Time (day)*	Reference
Acetobater aceti (AJ12368)	*HS*	*4/8g*	*7*	*[64]*
G. xylinus (PTCC1734)	*Date syrup*	*40/35*	*14*	*[65]*
A. xylinum ATCC 23769	*Rice bark*	*2/5*	*10*	*[66,67]*
G. xylinus (ATCC 53582)	*HS*	*3/5*	*7*	*[68]*
G. xylinum (ATCC 700178)	*CSL-fru*	*13*	*5*	*[69]*
G. xylinus (ATCC 10245)	*Molasses*	*17/5*	*7*	*[70]*
A.xylinum BPR2001	*+AgarHS*	*12/8*	*7*	*[71]*
sucrofermentans BPR3001	*Fru-ethanol*	*12/5*	*7*	*[72]*
A. xylinum NUST4.1	*HS + sodium alginate*	*6*	*5*	*[71]*
E. coli (ATCC 25922)	*Tryptone Soya Broth*	*30*	*8*	*[73]*
Staphylococcus aureus ATCC 6538	*TSP+agar*	*0/7*	*1*	*[74]*
A. xylinum GIM1.327	*HS*	*12*	*5*	*[75]*

THE ENVIRONMENTAL CONDITIONS EFFECT ON BC PRODUCTION

Microorganisms respond to environmental conditions by protein synthesis induction or inhibition, cell morphology changes, and metabolite pathway changes [77,78]. Therefore, ambient temperature, the culture medium pH, oxygen concentration, and incubation time affect BC production yield. Changing these factors from the optimal value leads to a decrease in BC production. Temperature is a primary parameter affecting bacterial growth and BC production. The optimum temperature for the *A. xylinum* and *Komagataeibacter* growth and BC production is 25°C – 30°C [79,80]. Different results have been reported for other strains. One study revealed that *A. xylinum* TISTR 975 cannot reproduce at 37°C even in an optimal culture medium [81]. In another study, the highest amount of BC production by *A. xylinum* was reported at 37 °C (Figs. 1a and b) [30]. At high temperatures, the cellular components of microbes, such as nucleic acid and cellulose-producer proteins, are denatured. High temperatures lead to culture medium denaturation, while low temperatures reduce cellular metabolism by supplying low energy for cell development [80]. Son et al. investigated the effect of temperature on BC production by Acetobacter sp., in HS culture medium. They reported the optimal temperature at 30°C [82]. Zahan et al. studied the temperature effect on BC production yield by Acetobacter xylinum 0416, and 28°C was selected as the best BC production temperature [83]. In these researches, the increase and decrease of the optimal temperature led to a reduction or lack of BC production.

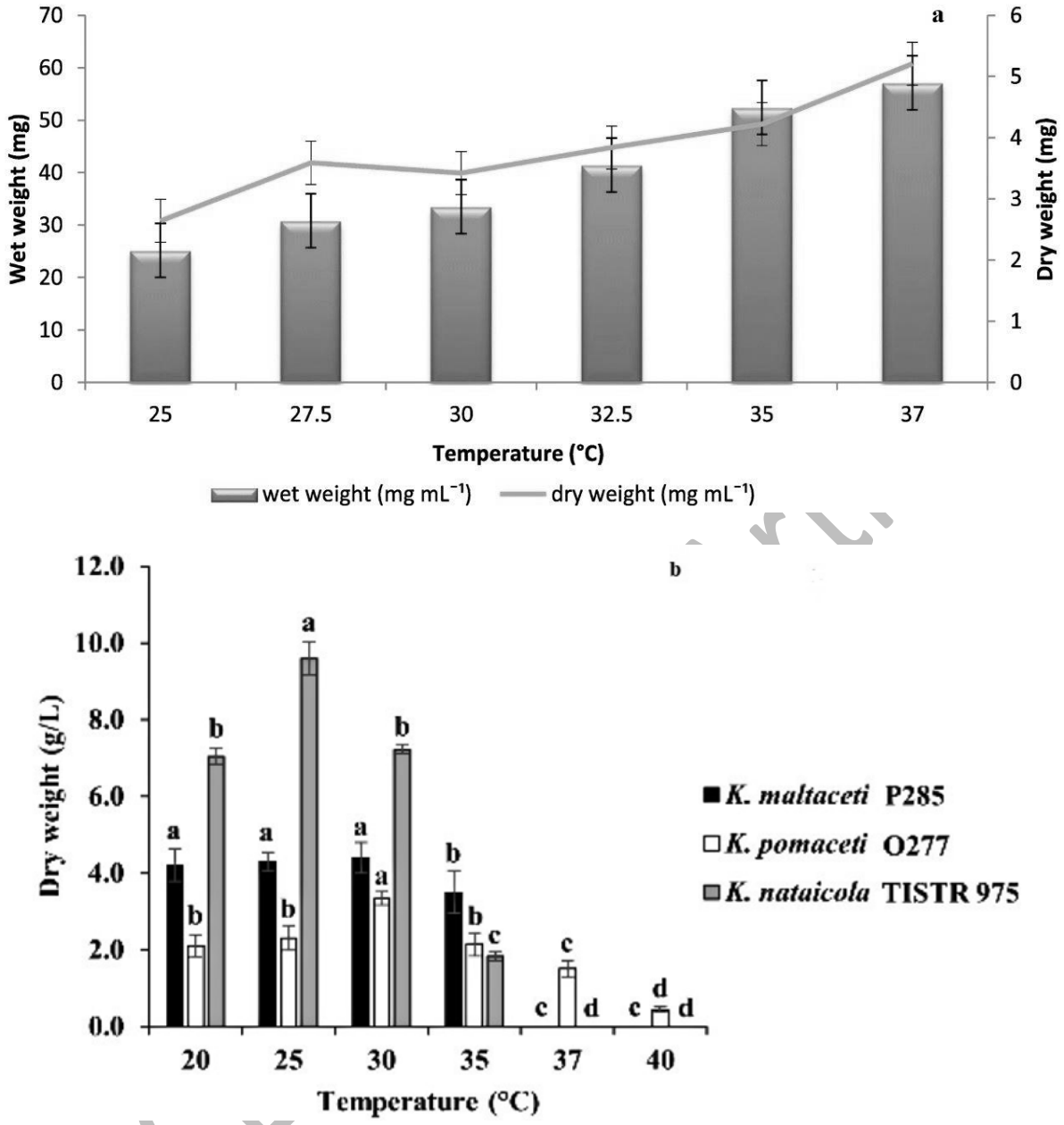

Fig. 1: Maximum BC production by different strains a) at 37 ℃ [30] ***and b) at 25*** ℃ [81]

In addition to temperature, pH is one of the most significant parameters in BC production yield. The optimal culture medium pH for BC production is in the 4-6 range, and the BC production yield decreases at a pH below four or more than six (Fig. 2) [84, 85]. When PH is out of proper range, like temperature, the proteins bent out of shape and denatured [86]. During the BC fermentation process, the pH decreases by gluconic acid and acetic acid production in the culture medium [87]. BC production was reported in both acidic and alkaline culture medium pH (more in acidic pH). For example, *Komagataeibacter intermedius* produces BC in the 4–9 pH range, with the highest production at pH 8 [88]. It was mentioned earlier that carbon consumption directly affects BC production. The consumption of some substances in the culture medium (glucose) by bacteria leads to a decrease in the culture medium pH below the optimal level, and due to the by-product production, BC production yield decreases. As carbon sources are generally available in culture mediums, the lack of air (dissolved oxygen) is a limiting factor

for cell metabolism and BC production, especially in static-type culture mediums [89]. It has been reported that BC acts as a layer to protect bacteria from environmental stress or to keep bacteria at the culture medium surface, where oxygen is available [90]. However, high oxygen concentration also helps to produce gluconic acid (a byproduct) [91]. Dissolved oxygen is affected by the S/V (surface-to-volume ratio or container shape). In static culture conditions, BC production is carried out at a higher oxygen level (higher S/V). Therefore, Erlen containers have more BC production than flat-shaped [92]. Higher or lower than the optimal S/V level for BC production reduces BC thickness and performance [92]. The S/V optimum ratio for BC production depends on the strain type and should be optimized to improve BC production yield under static conditions [13]. Table 4 shows the effect of changing S/V on BC production yield by *Acetobacter xylinium*.

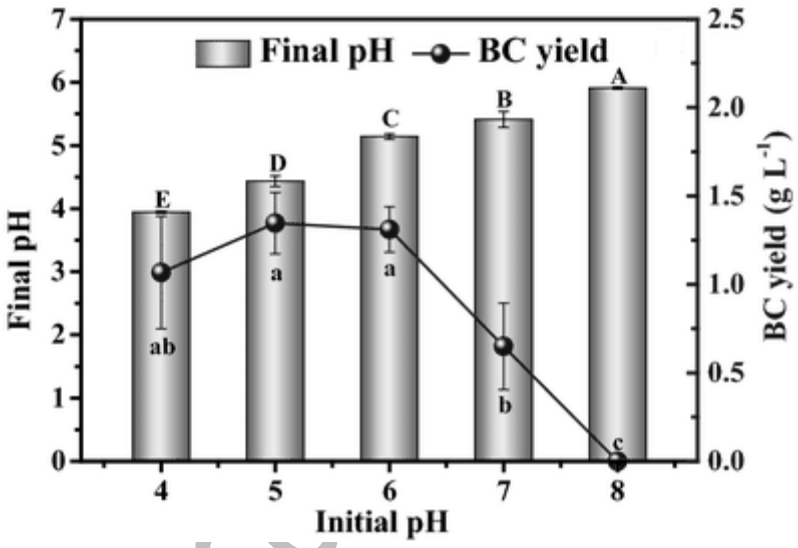

Fig. 2: Effects of initial and final pH on BC production [85]

Table 4. The effect of surface-to-volume ratio on BC production [92]

Thickness of medium layer (cm)	*Medium volume (ml)*	*Surface (cm^2)*	*$\frac{S}{V}(cm^{-1})$*	*Membrane thickness (cm)*	*Wet membrane mass (g)*	*Dry membrane mass (g)*	*BC yield (g/L)*
0.47	*200*	*425*	*2.13*	*-*	*-*	*0.54*	*2.7*
0.94	*400*	*425*	*1.06*	*0.8*	*203*	*0.96*	*3.1*
1.41	*600*	*425*	*0.71*	*1.1*	*-*	*1.2*	*3.5*
1.88	*800*	*425*	*0.53*	*1.2*	*440*	*2.4*	*3*
2.82	*1200*	*425*	*0.35*	*1.25*	*522*	*2.86*	*2.4*
3.74	*1600*	*425*	*0.27*	*1.2*	*621*	*3.13*	*1.93*

TYPES OF BC PRODUCTION PROCESS

The BC synthesis process is done using static and agitated methods. Several factors, including the production yield, application, and economic feasibility, are significant in determining the BC production method. The generative cell density in the agitated culture medium was reported more than the static type. Also, creating suitable turbulence leads to better oxygen availability. Therefore, BC production increases in agitated culture medium. On the other hand, BC granules produced through agitated fermentation have a lower degree of polymerization, mechanical strength, and crystallinity than the BC films produced in a static culture medium (Fig. 3) [93]. Therefore, BC granules are preferred for making in-situ and ex-situ modification composites from BC [68,94]. Also, in the agitated culture medium, gluconic acid (byproduct) production is dominated, and after the limited glucose source, the gluconic acid is turned into BC [95]. However, BC in agitated culture medium compared to the static one, has been inhibited due to the production of secondary metabolites. BC-producing strains are sensitive to stress (by stirring). It can cause mutations and reduce their production yield. On the other hand, adequate oxygen availability is another sensitivity of these strains, so the tension level due to stirring culture medium and oxygen supply should be balanced [95].

Static culture medium also has weaknesses such as long cultivation time, limitation of mass production, and in-situ composite manufacturing [94]. According to previous studies, the challenges of static culture medium can be reduced by air circulation, which leads to the development of BC culture in liquid culture vibration (due to airflow). According to these studies, in static cultures medium, cells have better contact with circulating air, which leads to better growth rates and increased BC production. The production of BC in these culture mediums is often associated with a significant increase in the viscosity of the fermentation culture medium, which prevents air penetration into it. So accumulation of glucuronic acid, acetic acid, and lactic acid significantly lowers the culture medium pH, below the optimum for bacterial growth and BC production [96]. In summary, both static and agitated culture methods have advantages and disadvantages, and depending on the application of productive BC and considering the economic feasibility, it is better to use the appropriate approach.

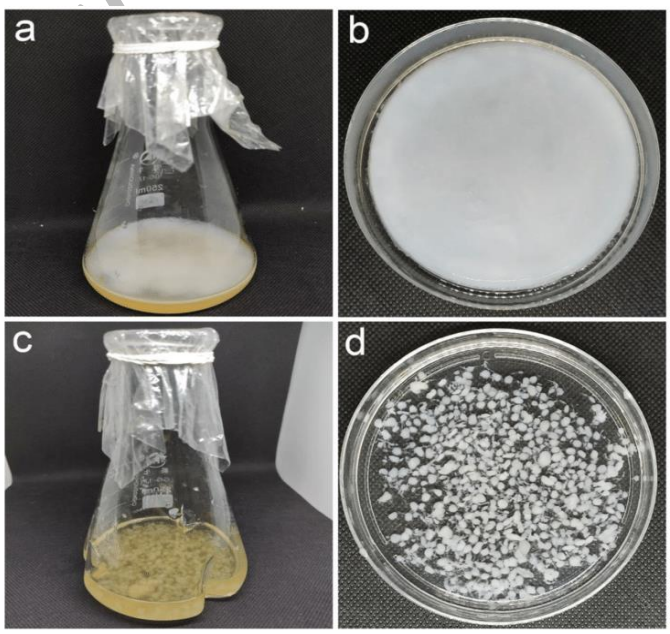

Fig. 3: Morphological difference of BC formed in a, b) static culture medium and c, d) agitated culture medium [93]

APPLICATION OF STATISTICAL DESIGN OF EXPERIMENTS IN BC PRODUCTION ENHANCEMENT

Another effective tool for reducing production costs, increasing production yield, and even improving product properties is process optimization. For example, two approaches have been proposed to optimize the culture medium, including one variable change at a time (OVAT) and optimization of numerical modeling through statistical designs. Statistical optimization is preferred as a more reliable and accurate tool for many optimization processes [38,97]. In general, to determine the culture medium conditions, such as the amounts of the components, and process type (static or agitated), and reduce the production costs or improve the characteristics of BC, it is necessary to use the statistical methods of screening and optimization. Environmental factors and culture medium conditions, such as temperature, pH, dissolved oxygen, stirring speed, time, carbon sources, nitrogen sources, and additives needed for BC production, should be in optimum amounts.

Statistical screening is used to determine the significant variables that affect BC production. Screening is applied to examine the effect of each variable on the response of the process, and the most important variables on the desired response selected. Insignificant factors are eliminated in screening to obtain a smaller set of reliable factors that can lead to optimal responses. Then, variables are optimized using statistical methods to maximize one or more responses. Researchers have used the Design Of Experiments to optimize BC production or characteristics [98]. Several experimental design methods, such as Plackett-Burman, Doehlert Design, Box-Behnken, and Response surface methodology (RSM), have been investigated to improve BC production and properties. Table 5 reviews some of these studies. The experimental design optimizes BC production by proposing and designing experiments with statistical significance and evaluating the importance of parameters. Finally, it evaluates the compatibility of the proposed model equation with experimental data. Also, these methods mention the results figures and help in more accurate visualization and analysis, interpretation of results by three-dimensional figures, and the ability to optimize and determine the range of all available factors to maximize the response [99].

Table 5. Application of statistical design of experiments in increasing BC production

Spices	Variables	Statical method	Conditions	Production yield (BC dry weight g/L)	Reference
Enterobacter hormaechei subsp. Steigerwaltii strain ZKE7	*Glycerol, and Glucose concentration*	*CCD*	*T=35 °C* *Time= 8 days* *Process= static* *Culture medium= HS*	*18.5*	*[100]*
Komagataeibacter sucrofermentans (DSM No. 15973)	*Sugar concentration, substrate pH level, Process Temperature*	*CCD*	*Time= 7 days* *T=30 °C*	*19.22*	*[101]*
Komactobacter intermedius (BCRC 910677)	*Fructose, and Peptone concentrations, pH*	*Box–Behnken design*	*T=28 °C* *Time= 6 days*	*3.906*	*[102]*
A. senegalensis MA1,	*Carbon, and nitrogen level, pH, Temperature, Polymer additives,*	*CCD*	*Time=30 days*	*Wet weight: 469.83*	*[30]*
G. xylinus (ATCC 700178)	*Protein amount, Incubation time, Inoculum ratio*	*CCD*	*Static culture 9 days, 30 ℃*	*3.2*	*[37]*
Komagatacibacter xylinus PTCC 1734.	*Vinasse concentration, Incubation Time*	*CCD*	*30 °C under static conditions for 11 days.*	*0.28*	*[32]*
A. xylinum BPR 2001	*Maple syrup concentration, Incubation period, Size of inoculum, Rotate speed*	*Box–Behnken design*	*25 ℃*	*1.51*	*[48]*
Gluconacetobacter xylinus TJU-D2	*Glucose, and Ethanol level, Initial pH*	*Box–Behnken design*	*8 day*	*4.82*	*[103]*
Gluconacetobacter xylinus (Komagataeibacter xylinus) (ATCC® 700178™)	*Inoculum Volumes, Fermentation period*	*Box–Behnken design*	*26℃*	*0.39*	*[104]*
Lactiplantibacillus plantarum	*Yeast extract, $MgSO_4$ level, pH*	*Box–Behnken design*	*30°C* *7 days*	*4.51*	*[105]*
Acetobacter xylinum NCIM 2526	*Incubation temperature, shaking rpm, pH of nitrogen source*	*Box–Behnken design*	*-*	*11.76*	*[106]*
Gluconacetobacter xylinus G29	*Yellow water, Citric acid $Na_2HPO_4 \cdot 12H_2O$*	*Box–Behnken design*	*28°C, Static culture 7 days*	*7.42*	*[107]*

Optimizing all the influential parameters to increase BC production and reduce the cost of BC production is applied to commercialize BC production. In addition to the mentioned cases, BC production on an industrial scale

requires modification or improvement of BC for practical purposes. For example, various studies examined the BC modification for medical applications, especially wound dressings. For this purpose, improving its water absorption properties to absorb infection and wound secretions or delivering medicine such as antibiotics is necessary. Improving BC water absorption properties can be used in various other applications such as diapers as a natural and non-toxic superabsorbent, agriculture to maintain soil moisture, and moisturizing creams and masks. The modification of BC properties, especially the water absorption, has been investigated further.

BC MODIFICATIONS

Due to the application of BC in food, pharmaceutical, electronic, textile, etc. industries, modification of its characteristics, in line with increasing production yield, has been proven by in situ and ex-situ modification. For example, the BC antimicrobial activity was created using inorganic materials, polymer compounds, and nanoparticles that show antimicrobial activity [108]. The unique three-dimensional structure, empty spaces in the BC network, hydrophilicity, crystallinity, and mechanical strength of BC make flexible and durable composites for tissue engineering [8,9]. In the hygiene industry, superabsorbent material using BC attracted attention. Dressings that can retain and absorb aqueous solutions are used to heal chronic wounds. Also, the water absorption and retention capacities allow drugs to load on the dressing structure. To prevent drying and sticking a dress to the wound, which causes severe pain and even damage, using a dressing that can maintain proper moisture or water on the wound is necessary. Therefore, the BC modification to improve water absorption and retention is mainly related to its application in health industries. Considering the importance of increasing BC production yield and its water absorption, studies were investigated in this review.

In-situ modification refers to the BC structure change during the cultivation, which is reachable by changing the cultivation conditions by adding additives to the culture medium or changing the carbon source. The simplicity of the modification process and its cost-effectiveness are two BC in-situ modification characteristics. This process is simultaneous with the BC production, which leads to the modification and improvement of various BC properties, including water absorption. In this regard, various substances were added to the culture medium like carboxymethyl cellulose, hydroxypropylmethylcellulose, xylan, chitosan, pectin, xyloglucan, dextrin, and lignosulfonate was applied. Increasing the concentration of the modification agent during BC production leads to problems such as the accumulation of additives in the BC network. Table 6 summarizes the BC in-situ modification materials and results in several studies. The results showed a 91.7% increase in water absorption of modified BC by adding only 1% carboxymethyl cellulose to the fermentation medium [109].

Table 6. Review of BC in-situ modification in past studies

culture medium additive for in-situ modification	*Result*	*Reference*
CMC	*Reduction of mechanical strength than BC.* *Denser cellulose network than BC.* *CMC decreased the crystallinity of BC.* *Higher rehydration of BC composite than BC.* *Lowest crystallinity at 1.0% CMC level.*	*[110]*

Fluorescent pigment Calcofluor White ST	*Decrease crystallinity from over 80% to about 50% by adding calcofluor-modified cellulose.* *Decreases crystallite size.* *Reduction of the microfibrils dimensions from 65 nm to about 30 nm by adding calcofluor.* *Reduction porosity by adding calcofluor.*	*[111]*
High methoxylated pectin	*Reduction of the water loss from 93% to 75% after 90 min by adding HMP.*	*[112]*
Polyacrylamide-co-acrylic acid	*Increase in BC production.* *The difference in appearance of fibers in the presence and absence of PA.*	*[113]*
Xyloglucan	*Reduction of cellulose Ia.* *XG interferes with the fibrillary units in regular ribbon assemblies*	*[114]*
Xyloglucan, *Pectin*	*Reduction in the Young's modulus.* *Decrease in tensile strength.* *Thinned the microfibrils' diameter.* *Increasing the pectin concentration in the culture medium decreased the average thickness of microfibrils.* *Increasing the concentration of xyloglucan caused the formation of thicker microfibrils.*	*[115]*
Tween 80, urea, fluorescent brightener, HPMC and CMC	*Decrease in mechanical strength of all BC composites except for urea addition.* *Increase in width of the cellulose bundles in the case of fluorescent, and HPMC addition.* *Decreases in the degree of crystallinity in the case of HPMC, and CMC addition.* *Increase in rehydration ability in the case of HPMC and CMC addition.*	*[116]*
Montmorillonite	*Increased storage modulus, swelling value, and thermal stability by adding montmorillonite to BC.* *More time is needed to dehydrate the montmorillonite-BC composite than BC.*	*[117]*
Aloe Vera	*Improve mechanical strength, crystallinity, water absorption capacity, and water vapor permeability.* *Reduction of average pore size of the modified film with a narrow pore size distribution.*	*[118]*
CMC	*Highest BC production by adding CMC.* *Decrease in the crystallinity and crystal size with increasing CMC. Increase in water retention by adding CMC.* *Higher Tmax of BC modified with CMC compared to the control sample.*	*[119]*
Xanthan gum	*Increase in fiber diameters combined with significantly improved hardness, flexibility, and tensile strength.*	*[120]*
Pectin *Gelatin*	*Increase in compressive modulus in the case of pectin addition.* *Increase in tensile modulus in case of gelatin addition.* *Decrease in crystal size and crystallinity in the presence of gelatin, while pectin only decreased crystallite size.* *Increase in microfibril accumulation in modified BCs.*	*[121]*

CMC	***looser structure and less crystalline in the case of BNC-CMC composite***	***[122]***
HEC	***compared to the control sample***	
	presence of HEC made cellulosic fibers thinner compared to control fibers	

Adding xyloglucan or sodium carboxymethyl cellulose to the culture medium led to a decrease in cellulose Ia from 64% to less than 30% [114]. Also, the carboxymethyl cellulose induced a smaller size of cellulose microfibers than the pure BC. Adding the fluorescent pigment Calcofluor White ST to the culture medium led to a non-crystalline cellulose network [123,124]. In a study, the addition of hydroxypropyl methylcellulose and carboxymethylcellulose reduced the crystallinity in modified BC from 70.54% to 52.23% and 45.38%, respectively, and carboxymethylcellulose can increase BC water absorption compared to the unmodified sample [125]. The paraffin microparticles addition to the BC culture medium for producing high-porosity cellulose has been investigated. After the BC formation, paraffin wax microparticles were removed by washing the product in water containing the active agent on the surface of Berol EZ-1 [126].

The disadvantage of this method is filling the BC pores with additives and reducing the water absorption [127]. In the study of Żywicka et al., only 1% rapeseed oil in culture medium increased BC weight by 604% and led to a high initial swelling ratio compared to the control sample (without rapeseed oil) [24]. Ma et al. reported that in situ modified BC with CMC resulted in a decrease in crystallinity of more than 39.8% and an increase in rehydration of up to 43.3% compared to control BC [109]. The hydrophilic functional groups of CMC facilitate the water absorption and the water diffusion molecules into the BC network during rehydration. However, a certain concentration of CMC in the composite leads to a dense network with less porosity than the control sample, which can significantly reduce the water absorption in the BC composite structure [109]. Other limitations of the in-situ modification method include the anti-microbial activity of some additives against BC strains and insolubility or low stability of suspension of some additives in the culture medium [126]. To overcome these limitations, ex-situ modification can be used. The ex-situ modification is done after BC harvesting from the culture medium. Dissolution and immersion are two usual methods for ex-situ modifications. In the dissolution method, BC and additive are dissolved in a solvent, and then BC composite is regenerated. The regenerated BC composite showed different structural features than BC. BC is immersed in a solution containing fine particles like nanocomposites, enzymes, and proteins in the immersion method, and this solution penetrates the BC structure [126,128,129]. Wahid et al. produced BC-zinc oxide composite by immersion modification method. For this process, the BC film was immersed in the nitrate solution for a certain period. The resulting composite showed the ability to degrade methyl orange, remarkable UV-blocking properties, and antibacterial activity [130].

Combining BC with different materials leads to the production of composites with diverse properties, such as antibacterial properties, higher physical strength, and better rehydration and water retention [131]. Materials such as silver, gold, ZnO, TiO_2, and montmorillonite have been combined by different methods with BC [131-135]. Montmorillonite-BC composites were synthesized in Menegasso et al.'s study by the displacement method and using a 0.1% montmorillonite suspension (immersing dry BC in a stirred solution of montmorillonite at 100 rpm for 24 hours at 28°C). This composite was then combined using simple processes with hydroxyethylcellulose, propylene glycol, and methyl 4-hydroxybenzoate, and a new composite was obtained for dressing application [131]. Gendi et al. modified BC with several fruit by-product extracts, and the product (BC-fruit by-product extract

composite) demonstrated better food preservatives compared to BC control as a plastic packaging. [38]. Due to the hydrophilic and porous structure of BC, plasticizers can be distributed through this matrix. Plasticizers reduce the fiber's intramuscular friction and enhance the polymer's flexibility. A low molecular weight plasticizer can penetrate the polymer structure and act as an internal lubricant. Plasticizers improve mechanical properties [136,137].

Almeida used glycerin to plasticize and modify BC. The resulting composite had improved water retention properties compared to pure BC [138]. In both modifications (in-situ and ex-situ), usually, BC crystallinity is reduced, leading to increased rehydration and BC swelling ratio [21]. Each of these modifications has advantages and disadvantages. For example, BC ex-situ modifications usually only affect the BC surface, while in-situ modification allows more intimate interactions between the growing cellulose fibers and the additive molecule to reach stable molecular films [139]. In general, BC modification processes are aimed at its application in industries. Improving the BC water absorption properties, making it a suitable option for use in the medical, health, hygiene, and cosmetic industries. The prerequisites for BC industrial production are improving production yield, reducing production cost, and improving water absorption properties, which were discussed in this review and shown in Fig. 4.

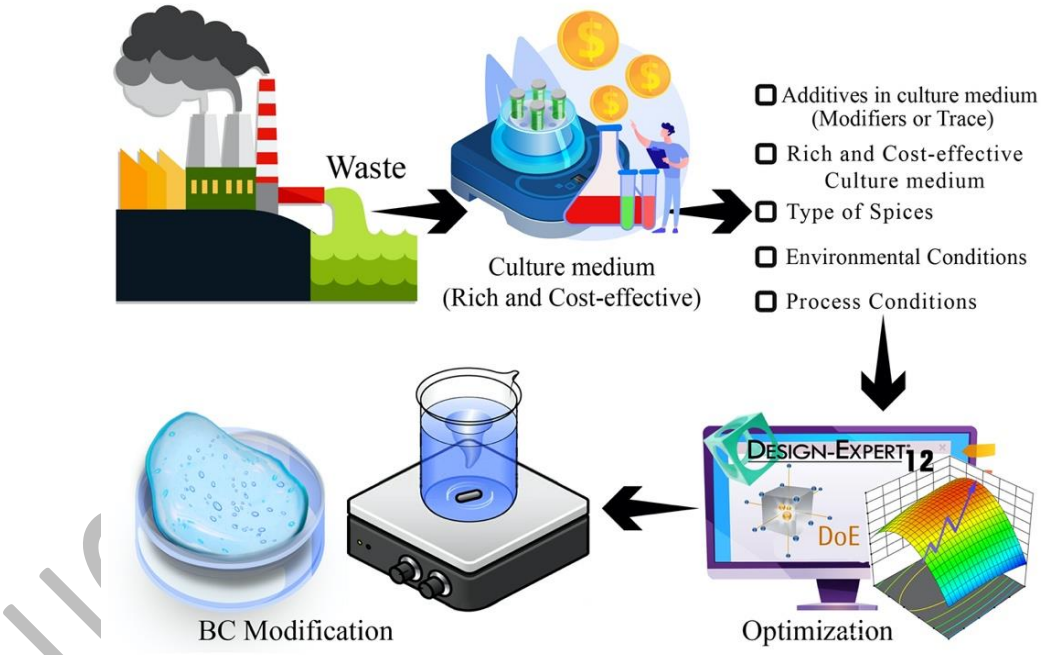

Fig. 4: Summarizing the BC industrialization process means increasing production yield, reducing production cost, and improving water absorption properties. (Graphical abstract)

CONCLUSION

Increasing BC production and reducing its production price are still challenges that lead to continuous research to discover rich and cost-effective culture mediums and optimization of effective parameters in the fermentation process. Common solutions to these challenges are to use industrial wastewater as a culture medium and enrich it with additives. Some industrial wastes or by-products of different industries

are vinasse, CSL, molasses, and rotten fruits, which are rich in minerals and organic. Adding substances such as ethanol and acetic acid to these culture mediums and investigating the effect of their metabolites on BC production has brought positive results in increasing production. Apart from the culture medium, other environmental and process conditions (temperature, pH, dissolved oxygen, and static or agitated process) are also very effective in the BC production yield, product price, and even product characteristics. Therefore, these parameters should be screened using statistical techniques and optimized using different designs of experiment methods. Improving the BC properties to make it suitable for various applications is as important as optimizing its production process. Properties related to BC water absorption are closely related to its applications in tissue engineering, wound dressing, and drug delivery systems. Therefore, BC in situ and ex-situ modification, especially those related to water absorption, was discussed in several studies. This paper is a database study of the past decade on the increasing BC production and modification using simple and low-cost methods that can be used to produce BC for medical, health, and high-scale industrial applications.

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CONFLICT OF INTEREST

All authors declare that they have no conflicts of interest.

REFERENCES

1. Jabbari F., Babaeipour V., Bacterial cellulose as a potential biopolymer for wound care: A review, *Int. J. Polym. Mater.*, **11**(3):11-17 (2023)
2. Mohammadi S., Jabbari F., Babaeipour V., Bacterial cellulose-based composites as vehicles for dermal and transdermal drug delivery: A review, *Int. J. Biol. Macromol.*, ***243*** *(3)* (2023)
3. Jabbari F., Babaeipour V. Bacterial Nanocellulose-A Remarkable Polymer for Biomedical Applications: Production, Engineering, and Recent Advances and Developments, *Iran. J. Chem. Eng.*, **42** (127): 1712-1730 (2023).
4. Kang J.H., Park S.C., Production and Characteristics of Cellulose from Saprolegnia ferax, *J. Korea Tech. Assoc. Pulp Pap. Ind.,* **39**: 77–83 (2007).
5. Ullah H., Santos H. A., Khan T., Applications of bacterial cellulose in food, cosmetics and drug delivery, *Cellulose.,* **23**: 2291–2314 (2016).
6. Okiyama A., Motoki M., Yamanaka S., Bacterial cellulose II. Processing of the gelatinous cellulose for food materials, *Food Hydrocoll.,* **6**: 479–487 (1992).
7. Tomé L. C., Brandao L., Mendes A. M., Silvestre A. J., Neto C. P., Gandini, A., Marrucho I. M.,

Preparation and characterization of bacterial cellulose membranes with tailored surface and barrier properties, *Cellulose.,* **17**: 1203–1211 (2010).

8. Andriani D., Apriyana A. Y., Karina M., The optimization of bacterial cellulose production and its applications: a review, *Cellulose.,* **27**: 6747–6766 (2020).
9. Chawla P. R., Bajaj I. B., Survase S. A., Singhal R. S., Microbial cellulose: fermentative production and applications, *Food Technol. Biotechnol.,* **47**: 107–124 (2009).
10. Choi S. M., Rao K. M., Zo S. M., Shin E. J., Han S. S., Bacterial Cellulose and Its Applications, *Polymers (Basel).,* **14**: 1080 (2022).
11. Lin W.-C., Lien C.-C., Yeh H.-J., Yu C.-M., Hsu S., Bacterial cellulose and bacterial cellulose–chitosan membranes for wound dressing applications, *Carbohydr. Polym.,* **94**: 603–611 (2013).
12. Das M., Zandraa O., Mudenur C., Saha N., Sáha P., Mandal B., & Katiyar V., Composite scaffolds based on bacterial cellulose for wound dressing application, *ACS Appl. Bio Mater.,* **5**: 3722–3733 (2022).
13. El-Gendi H., Taha T. H., Ray J. B., Saleh A. K., Recent advances in bacterial cellulose: a low-cost effective production media, optimization strategies and applications, *Cellulose.,* **29**: 7495–7533 (2022).
14. Jozala A. F., Pértile R. A. N., dos Santos C. A., de Carvalho Santos-Ebinuma V., Seckler M. M., Gama F. M., Pessoa, A., Bacterial cellulose production by Gluconacetobacter xylinus by employing alternative culture media, *Appl. Microbiol. Biotechnol.,* **99**: 1181–1190 (2015).
15. Islam M. U., Ullah M. W., Khan S., Shah N., Park J. K., Strategies for cost-effective and enhanced production of bacterial cellulose, *Int. J. Biol. Macromol.,* **102**: 1166–1173 (2017).
16. Cappa C., Lucisano M., Mariotti M., Influence of Psyllium, sugar beet fibre and water on gluten-free dough properties and bread quality, *Carbohydr. Polym.,* **98**: 1657–1666 (2013).
17. Cacicedo M.L., Castro M.C., Servetas I., Bosnea L., Boura K., Tsafrakidou P., Dima A., Terpou A., Koutinas A., Castro G.R., Progress in bacterial cellulose matrices for biotechnological applications, *Bioresour. Technol.,* **213**: 172–180 (2016).
18. Zahan K. A., Nordin K., Mustapha M., Mohd Zairi M. N., Effect of incubation temperature on growth of Acetobacter xylinum 0416 and bacterial cellulose production, *Appl. Mech. Mater.,* **815**: 3–8 (2015).
19. Vandamme E. J., De Baets S., Vanbaelen A., Joris K., De Wulf, P., Improved production of bacterial cellulose and its application potential, *Polym. Degrad. Stab.,* **59**: 93–99 (1998).
20. Zhong C., Zhang G.C., Liu M., Zheng X.T., Han P.P., Jia S.R., Metabolic flux analysis of Gluconacetobacter xylinus for bacterial cellulose production, *Appl. Microbiol. Biotechnol.,* **97**: 6189–6199 (2013).

21. Szymańska M., Hoppe J., Dutkiewicz M., Sobolewski P., Palacz M., Janus E., Zielińska B., Drozd R., Silicone polyether surfactant enhances bacterial cellulose synthesis and water holding capacity, *Int. J. Biol. Macromol.,* **208**: 642–653 (2022).
22. Kuo C.-H., Chen J.-H., Liou B.-K., Lee C.-K., Utilization of acetate buffer to improve bacterial cellulose production by Gluconacetobacter xylinus, *Food Hydrocoll.,* **53** 98–103 (2016).
23. Li Y., Tian, C., Tian H., Zhang J., He X., Ping W., Lei H., Improvement of bacterial cellulose production by manipulating the metabolic pathways in which ethanol and sodium citrate involved, *Appl. Microbiol. Biotechnol.,* **96**: 1479–1487 (2012).
24. Żywicka A., Junka, A.F., Szymczyk P., Chodaczek G., Grzesiak, J., Sedghizadeh P.P., Fijałkowski K., Bacterial cellulose yield increased over 500% by supplementation of medium with vegetable oil, *Carbohydr. Polym.,* **199**: 294–303 (2018).
25. Molina-Ramírez C., Enciso C., Torres-Taborda M., Zuluaga R., Gañán P., Rojas O.J., Castro, C., Effects of alternative energy sources on bacterial cellulose characteristics produced by Komagataeibacter medellinensis, *Int. J. Biol. Macromol.,* **117**: 735–741 (2018).
26. Keshk S. M. A. S., Bacterial cellulose production and its industrial applications. J Bioprocess Biotech, **4** (2). (2014).
27. Tian D., Shen F., Hu J., Renneckar S., Saddler J. N., Enhancing bacterial cellulose production via adding mesoporous halloysite nanotubes in the culture medium, *Carbohydr. Polym.,* **198**: 191–196 (2018).
28. Gullo M., Sola A., Zanichelli G., Montorsi M., Messori M., Giudici P., Increased production of bacterial cellulose as starting point for scaled-up applications, *Appl. Microbiol. Biotechnol.,* **101**: 8115–8127 (2017).
29. Cazón P., Vázquez M., Improving bacterial cellulose films by ex-situ and in-situ modifications: a review, *Food Hydrocoll.,* **113**: 106514 (2021).
30. Aswini K., Gopal N. O., Uthandi S., Optimized culture conditions for bacterial cellulose production by Acetobacter senegalensis MA1, *BMC Biotechnol.,* **20**: 1–16 (2020).
31. Cerrutti P., Roldán P., García R.M., Galvagno M.A., Vázquez A., Foresti M.L., Production of bacterial nanocellulose from wine industry residues: I mportance of fermentation time on pellicle characteristics, *J. Appl. Polym. Sci.,* **133**: (2016).
32. Barshan S., Rezazadeh-Bari M., Almasi H., Amiri S., Optimization and characterization of bacterial cellulose produced by Komagatacibacter xylinus PTCC 1734 using vinasse as a cheap cultivation medium, *Int. J. Biol. Macromol.,* **136**: 1188–1195 (2019).
33. Fan X., Gao Y., He W., Hu H., Tian M., Wang K., Pan, S., Production of nano bacterial cellulose from beverage industrial waste of citrus peel and pomace using Komagataeibacter xylinus,

Carbohydr. Polym., **151**: 1068–1072 (2016).

34. Kurosumi A., Sasaki C., Yamashita Y., Nakamura Y., Utilization of various fruit juices as carbon source for production of bacterial cellulose by Acetobacter xylinum NBRC 13693, *Carbohydr. Polym.,* **76**: 333–335 (2009).
35. Hamid M., Babaeipour V., Imani M., Bahrami A., Dissolution and Regeneration of the Produced Nano Bacterial Cellulose of Food Industries Wastewaters by a Cost-Benefit Method, *Iran. J. Chem. Chem. Eng.,* **38**: 9–21 (2019).
36. Ebrahimi E., Babaeipour V., Meftahi A., Alibakhshi S., Effects of bio-production process parameters on bacterial cellulose mechanical properties, *J. Chem. Eng. Japan.,* **50**: 857–861 (2017).
37. Bilgi E., Bayir E., Sendemir-Urkmez A., Hames E. E., Optimization of bacterial cellulose production by Gluconacetobacter xylinus using carob and haricot bean, *Int. J. Biol. Macromol.,* **90**: 2–10 (2016).
38. El-Gendi H., Salama A., El-Fakharany E. M., Saleh A. K., Optimization of bacterial cellulose production from prickly pear peels and its ex situ impregnation with fruit byproducts for antimicrobial and strawberry packaging applications, *Carbohydr. Polym.,* **302**: 120383 (2023).
39. Dórame-Miranda R.F., Gámez-Meza N., Medina-Juárez L.Á., Ezquerra-Brauer J.M., Ovando-Martínez M., Lizardi-Mendoza J., Bacterial cellulose production by Gluconacetobacter entanii using pecan nutshell as carbon source and its chemical functionalization, *Carbohydr. Polym.,* **207**: 91–99 (2019).
40. Algar I., Fernandes S.C., Mondragon G., Castro C., Garcia‐Astrain C., Gabilondo N., Retegi, A., Eceiza A., Pineapple agroindustrial residues for the production of high value bacterial cellulose with different morphologies, *J. Appl. Polym. Sci.,* **132** (2015).
41. Ye J., Zheng S., Zhang Z., Yang F., Ma K., Feng Y., Zheng J., Mao, D., Yang X., Bacterial cellulose production by Acetobacter xylinum ATCC 23767 using tobacco waste extract as culture medium, *Bioresour. Technol.,* **274**: 518–524 (2019).
42. Luo M.T., Zhao C., Huang C., Chen X.F., Huang Q.L., Qi G.X., Tian L.L., Xiong L., Li H.L., Chen X.D., Efficient using durian shell hydrolysate as low-cost substrate for bacterial cellulose production by Gluconacetobacter xylinus, *Indian J. Microbiol.,* **57**: 393–399 (2017).
43. Abdelraof M., Hasanin M. S., El-Saied H., Ecofriendly green conversion of potato peel wastes to high productivity bacterial cellulose, *Carbohydr. Polym.,* **211**: 75–83 (2019).
44. Rani M. U., Appaiah K. A. A., Production of bacterial cellulose by Gluconacetobacter hansenii UAC09 using coffee cherry husk, *J. Food Sci. Technol.,* **50**: 755–762 (2013).
45. Adebayo-Tayo B. C., Akintunde M., Alao S., Comparative effect of agrowastes on bacterial

cellulose production by Acinetobacter sp. BAN1 and Acetobacter pasteurianus PW1, *Turk J Agric Nat Sci.,* **4**: 145–154 (2014).

46. Kumar V., Sharma D.K., Bansal V., Mehta D., Sangwan R.S., Yadav S.K., Efficient and economic process for the production of bacterial cellulose from isolated strain of Acetobacter pasteurianus of RSV-4 bacterium, *Bioresour. Technol.,* **275**: 430–433 (2019).
47. Huang C., Yang X.Y., Xiong L., Guo H.J., Luo J., Wang B., Zhang H.R., Lin X.Q., Chen X.D., Evaluating the possibility of using acetone-butanol-ethanol (ABE) fermentation wastewater for bacterial cellulose production by G luconacetobacter xylinus, *Lett. Appl. Microbiol.,* **60**: 491–496 (2015).
48. Zeng X., Small D. P., Wan W., Statistical optimization of culture conditions for bacterial cellulose production by Acetobacter xylinum BPR 2001 from maple syrup, *Carbohydr. Polym.,* **85**: 506–513 (2011).
49. Lotfiman S., Awang Biak D. R., Ti T. B., Kamarudin S., Nikbin S., Influence of date syrup as a carbon source on bacterial cellulose production by Acetobacter xylinum 0416, *Adv. Polym. Technol.,* **37**: 1085–1091 (2018).
50. Adebayo-Tayo B. C., Akintunde M. O., Sanusi J. F., Effect of different fruit juice media on bacterial cellulose production by Acinetobacter sp. BAN1 and Acetobacter pasteurianus PW1, *J. Adv. Biol. Biotechnol.* 1–9 (2017).
51. Hong F., Qiu K., An alternative carbon source from konjac powder for enhancing production of bacterial cellulose in static cultures by a model strain Acetobacter aceti subsp. xylinus ATCC 23770, *Carbohydr. Polym.,* **72**: 545–549 (2008).
52. Wu J.-M., Liu R.-H., Thin stillage supplementation greatly enhances bacterial cellulose production by Gluconacetobacter xylinus, *Carbohydr. Polym.,* **90**: 116–121 (2012).
53. Jahan F., Kumar V., Saxena R. K., Distillery effluent as a potential medium for bacterial cellulose production: a biopolymer of great commercial importance, *Bioresour. Technol.,* **250**: 922–926 (2018).
54. Ha J.H., Shehzad O., Khan S., Lee S.Y., Park J.W., Khan T., Park J.K., Production of bacterial cellulose by a static cultivation using the waste from beer culture broth, *Korean J. Chem. Eng.,* **25**: 812 (2008).
55. Chen L., Hong F., Yang X., Han S., Biotransformation of wheat straw to bacterial cellulose and its mechanism, *Bioresour. Technol.,* **135**: 464–468 (2013).
56. Vazquez A., Foresti M. L., Cerrutti P., Galvagno, M., Bacterial cellulose from simple and low cost production media by Gluconacetobacter xylinus, *J. Polym. Environ.,* **21**: 545–554 (2013).
57. Revin V., Liyaskina E., Nazarkina M., Bogatyreva A., Shchankin M., Cost-effective production of

bacterial cellulose using acidic food industry by-products, *brazilian J. Microbiol.,* **49**: 151–159 (2018).

58. Guo X., Cavka A., Jönsson L. J., Hong F., Comparison of methods for detoxification of spruce hydrolysate for bacterial cellulose production, *Microb. Cell Fact.,* **12**: 1–14 (2013).
59. Urbina L., Hernández-Arriaga A.M., Eceiza A., Gabilondo N., Corcuera M.A., Prieto M.A., Retegi, A., By-products of the cider production: an alternative source of nutrients to produce bacterial cellulose, *Cellulose.,* **24**: 2071–2082 (2017).
60. Moon S.-H., Park J.-M., Chun H.-Y., Kim S.-J., Comparisons of physical properties of bacterial celluloses produced in different culture conditions using saccharified food wastes, *Biotechnol. Bioprocess Eng.,* **11**: 26 (2006).
61. Rouhi M., Khanchezar S., Babaeipour V., Economical Optimization of Industrial Medium Culture for Bacterial Cellulose Production, *Appl. Biochem. Biotechnol.* 1–19 (2022).
62. Yamada Y., Yukphan P., Vu H.T.L., Muramatsu Y., Ochaikul D., Tanasupawat S., Nakagawa Y., Description of Komagataeibacter gen. nov., with proposals of new combinations (Acetobacteraceae), *J. Gen. Appl. Microbiol.,* **58**: 397–404 (2012).
63. Ullah M. W., Ul-Islam M., Khan S., Kim Y., Park J. K., Innovative production of bio-cellulose using a cell-free system derived from a single cell line, *Carbohydr. Polym.,* **132**: 286–294 (2015).
64. Thi T., Sugiyama J., Bulone V., Bacterial Cellulose-based Biomimetic Composites, *Biopolymers.,* **602**: 3–11 (2010).
65. Moosavi-nasab M., Yousefi A. R., Investigation of Physicochemical Properties of the Bacterial Cellulose Produced by Gluconacetobacter xylinus from date syrup, World Academy of Science, Engineering and Technology,, **4**: 1258–1263 (2010).
66. Goelzer F. D. E., Faria-Tischer P. C. S., Vitorino J. C., Sierakowski M. R., Tischer, C. A., Production and characterization of nanospheres of bacterial cellulose from Acetobacter xylinum from processed rice bark, *Mater. Sci. Eng. C.,* **29**: 546–551 (2009).
67. Lee C. M., Gu J., Kafle K., Catchmark J., Kim S. H., Cellulose produced by Gluconacetobacter xylinus strains ATCC 53524 and ATCC 23768: Pellicle formation, post-synthesis aggregation and fiber density, *Carbohydr. Polym.,* **133**: 270–276 (2015).
68. Czaja W., Romanovicz D., Malcolm Brown R., Structural investigations of microbial cellulose produced in stationary and agitated culture, *Cellulose.,* **11**: 403–411 (2004).
69. Shah N., Ul-Islam M., Khattak W. A., Park J. K., Overview of bacterial cellulose composites: A multipurpose advanced material, *Carbohydrate Polymers* vol. 98 1585–1598 (2013).
70. Moosavi-nasab M., Yousefi A., Biotechnological production of cellulose by Gluconacetobacter

xylinus from agricultural waste, *Iran. J. Biotechnol.,* **9**: 94–101 (2011).

71. Zhou L. L., Sun D. P., Hu L. Y., Li Y. W., Yang J. Z., Effect of addition of sodium alginate on bacterial cellulose production by Acetobacter xylinum, *J. Ind. Microbiol. Biotechnol.,* **34**: 483–489 (2007).
72. Santos S. M., Carbajo J. M., Villar J. C., The effect of carbon and nitrogen sources on bacterial cellulose production and properties from gluconacetobacter sucrofermentans CECT 7291 focused on its use in degraded paper restoration, *BioResources.,* **8**: 3630–3645 (2013).
73. Lin S., Chen L., Huang L., Cao S., Luo X., Liu K., Huang Z., PREPARATION AND CHARACTERIZATION OF CHITOSAN / CELLULOSE BLEND FILMS USING ZnCl2·3H2O AS A SOLVENT, *BioResources* **7**: 5488–5499 (2012).
74. Fernandes S.C., Sadocco P., Alonso-Varona A., Palomares T., Eceiza A., Silvestre A.J., Mondragon I., Freire C.S., Bioinspired antimicrobial and biocompatible bacterial cellulose membranes obtained by surface functionalization with aminoalkyl groups, *ACS Appl. Mater. Interfaces.,* **5**: 3290–3297 (2013).
75. Shao W., Wang S., Wu J., Huang M., Liu H., Min H., Synthesis and antimicrobial activity of copper nanoparticle loaded regenerated bacterial cellulose membranes, *RSC Adv.,* **6**: 65879–65884 (2016).
76. Lu T., Gao H., Liao B., Wu J., Zhang W., Huang J., Liu M., Huang J., Chang Z., Jin M., Yi Z., Characterization and optimization of production of bacterial cellulose from strain CGMCC 17276 based on whole-genome analysis, *Carbohydr. Polym.,* **232**: 115788 (2020).
77. Yang D. C., Blair K. M., Salama N. R., Staying in shape: the impact of cell shape on bacterial survival in diverse environments, *Microbiol. Mol. Biol. Rev.,* **80**: 187–203 (2016).
78. Karasz D. C., Weaver A. I., Buckley D. H., Wilhelm R. C., Conditional filamentation as an adaptive trait of bacteria and its ecological significance in soils, *Environ. Microbiol.* **24**: 1–17 (2022).
79. Pa'e N., Salehudin M. H., Hassan N. D., Marsin A. M., Muhamad I. I., Thermal behavior of bacterial cellulose-based hydrogels with other composites and related instrumental analysis, *Cellul. Superabsorbent Hydrogels.,* **1**: 763–787 (2018).
80. Lahiri D., Nag M., Dutta B., Dey A., Sarkar T., Pati S., Edinur H.A., Abdul Kari Z., Mohd Noor N.H., Ray R.R., Bacterial cellulose: Production, characterization, and application as antimicrobial agent, *Int. J. Mol. Sci.,* **22**: 12984 (2021).
81. Thongwai N., Futui W., Ladpala N., Sirichai B., Weechan A., Kanklai J., Rungsirivanich P., Characterization of bacterial cellulose produced by Komagataeibacter maltaceti P285 isolated from contaminated honey wine, *Microorganisms.,* **10**: 528 (2022).

82. Son H., Heo M., Kim Y., Lee S., Optimization of fermentation conditions for the production of bacterial cellulose by a newly isolated Acetobacter, *Biotechnol. Appl. Biochem.* **33**: 1–5 (2001).
83. Zahan K. A., Pa'e N., Muhamad I. I., Monitoring the effect of pH on bacterial cellulose production and Acetobacter xylinum 0416 growth in a rotary discs reactor, *Arab. J. Sci. Eng.*, **40**: 1881–1885 (2015).
84. Jahan F., Kumar V., Rawat G., Saxena R. K., Production of microbial cellulose by a bacterium isolated from fruit, *Appl. Biochem. Biotechnol.,* **167**: 1157–1171 (2012).
85. Han Y.H., Mao H.L., Wang S.S., Deng J.C., Chen D.L., Li M., Ecofriendly green biosynthesis of bacterial cellulose by Komagataeibacter xylinus B2-1 using the shell extract of Sapindus mukorossi Gaertn. as culture medium. *Cellulose.,* **27**: 1255–1272 (2020).
86. Hu Y., Catchmark J. M., Integration of cellulases into bacterial cellulose: toward bioabsorbable cellulose composites, *J. Biomed. Mater. Res. Part B Appl. Biomater.,* **97**: 114–123 (2011).
87. Yanti N. A., Ahmad S. W., Muhiddin N. H., Evaluation of inoculum size and fermentation period for bacterial cellulose production from sago liquid waste, in *Journal of Physics: Conference Series*., **1**: 11-16 (2018).
88. Lin S.P., Huang Y.H., Hsu K.D., Lai Y.J., Chen Y.K., Cheng K.C., Isolation and identification of cellulose-producing strain Komagataeibacter intermedius from fermented fruit juice, *Carbohydr. Polym.,* **151**: 827–833 (2016).
89. Son.H.J., Kim.H.G., Kim.K.K., Kim.H.S., K. Y., Increased production of bacterial cellulose by Acetobacter sp. V in synthetic media under shaking culture conditions, *Technol, Bioresour.*, **I86**: 200–215 (2003).
90. Oshima.T., Kondo.K., Ohto.K, Inoue.K., and B. Y., Preparation of phosphorylated bacterial cellulose as an adsorbent for metal ions, *React. Funct. Polym.,* **42**: 23–34 (2007).
91. Azeredo H., Barud H., Farinas C. S., Vasconcellos V. M., Claro A. M., Bacterial cellulose as a raw material for food and food packaging applications, *Front. Sustain. Food Syst.,* **3**: 7 (2019).
92. Krystynowicz A., Czaja W., Wiktorowska-Jezierska A., Gonçalves-Miśkiewicz M., Turkiewicz M., Bielecki S., Factors affecting the yield and properties of bacterial cellulose, *J. Ind. Microbiol. Biotechnol.,* **29**: 189–95 (2002).
93. Zhong C., Industrial-scale production and applications of bacterial cellulose, *Front. Bioeng. Biotechnol.,* **8**: 605374 (2020).
94. Son H.J., Kim H.G., Kim K.K., Kim H.S., Kim Y.G., Lee S.J., Increased production of bacterial cellulose by Acetobacter sp. V6 in synthetic media under shaking culture conditions, *Bioresour. Technol.,* **86**: 215–219 (2003).
95. Liu M., Zhong C., Wu X.Y., Wei Y.Q., Bo T., Han P.P., Jia S.R., Metabolomic profiling coupled with

metabolic network reveals differences in Gluconacetobacter xylinus from static and agitated cultures, *Biochem. Eng. J.,* **101**: 85–98 (2015).

96. Ha J. H., Shah N., Ul-Islam M., Khan T., Park J. K., Bacterial cellulose production from a single sugar α-linked glucuronic acid-based oligosaccharide, *Process Biochem.,* **46**: 1717–1723 (2011).
97. Do C.V.T., Nguyen N.T.T., Pham M.H.T., Pham T.Y.T., Le T.G., Tran T.D., Central composite design for simultaneously optimizing biomass and lutein production by a mixotrophic Chlorella sorokiniana TH01, *Biochem. Eng. J.,* **177**: 108231 (2022).
98. Mandenius C., Brundin A., Bioprocess optimization using design-of-experiments methodology. *Biotechnol, Prog.,* **24**: 1191–1203 (2008).
99. Fatemi S. M., Robatjazi S. M., Zarei A. R., Hosseini S. G., Optimization of a Bleaching Process for the Conversion of Sugar Beet Pulp Alpha-Cellulose with a High Whiteness Index and Purity, *Iran. J. Chem. Chem. Eng.,* **42**: 90–99 (2023).
100. Bagewadi Z.K., Dsouza V., Mulla S.I., Deshpande S.H., Muddapur U.M., Yaraguppi D.A., Reddy V.D., Bhavikatti J.S., More S.S., Structural and functional characterization of bacterial cellulose from Enterobacter hormaechei subsp. steigerwaltii strain ZKE7, *Cellulose.,* **27**: 9181–9199 (2020).
101. Bekatorou A., Plioni I., Sparou K., Maroutsiou R., Tsafrakidou P., Petsi, T., Kordouli E., Bacterial cellulose production using the corinthian currant finishing side-stream and cheese whey: process optimization and textural characterization, *Foods.,* **8**: 193 (2019).
102. SSantoso S.P., Chou C.C., Lin S.P., Soetaredjo F.E., Ismadji S., Hsieh C.W., Cheng K.C., Enhanced production of bacterial cellulose by Komactobacter intermedius using statistical modeling, *Cellulose.,* **27**: 2497–2509 (2020).
103. Du R., Wang Y. Zhao F., Qiao X., Song Q., Li S., Kim R.C., Pan L., Han Y., Xiao H., Zhou Z., Production, optimization and partial characterization of bacterial cellulose from Gluconacetobacter xylinus TJU-D2, *Waste and Biomass Valorization.,* **11**: 1681–1690 (2020).
104. Bodea I.M., Beteg F.I., Pop C.R., David A.P., Dudescu M.C., Vilău C., Stănilă A., Rotar A.M., Cătunescu G.M., Optimization of moist and oven-dried bacterial cellulose production for functional properties, **22**:110-129 (2021).
105. Saleh A. K., El-Gendi H., Soliman N. A., El-Zawawy W. K., Abdel-Fattah Y. R., Bioprocess development for bacterial cellulose biosynthesis by novel Lactiplantibacillus plantarum isolate along with characterization and antimicrobial assessment of fabricated membrane, *Sci. Rep.,* **12**: 1–17 (2022).
106. Singh M. K., Singh A., Pandey A., Optimization of Bacterial Cellulose Production Using Jaggery

Waste by Response Surface Method, **66**:1-12 (2022).

107. He F., Yang H., Zeng L., Hu H., Hu C., Production and characterization of bacterial cellulose obtained by Gluconacetobacter xylinus utilizing the by-products from Baijiu production, *Bioprocess Biosyst. Eng.,* **43**: 927–936 (2020).
108. Gromovykh T. I., Creation of composites of bacterial cellulose and silver nanoparticles: Evaluation of antimicrobial activity and cytotoxicity, *Int. J. Nanotechnol.,* **16**: 408–420 (2019).
109. Ma T., Zhao Q. Q., Ji K. H., Zeng B., Li G. Q., Homogeneous and porous modified bacterial cellulose achieved by in situ modification with low amounts of carboxymethyl cellulose, *Cellulose.,* **21**: 2637–2646 (2014).
110. Chen H.-H., Chen L.-C., Huang H.-C., Lin S.-B., In situ modification of bacterial cellulose nanostructure by adding CMC during the growth of Gluconacetobacter xylinus, *Cellulose.,* **18**: 1573–1583 (2011).
111. Khandelwal M., Windle A. H., Hessler N., In situ tunability of bacteria produced cellulose by additives in the culture media, *J. Mater. Sci.,* **51**: 4839–4844 (2016).
112. Cacicedo M. L., Hybrid bacterial cellulose–pectin films for delivery of bioactive molecules, *New J. Chem.,* **42**: 7457–7467 (2018).
113. Joseph G., Rowe G. E., Margaritis A., Wan W., Effects of polyacrylamide-co-acrylic acid on cellulose production by Acetobacter xylinum, *J. Chem. Technol. Biotechnol. Int. Res. Process. Environ. Clean Technol.,* **78**: 964–970 (2003).
114. Yamamoto H., Horn F., In Situ crystallization of bacterial cellulose I. Influences of polymeric additives, stirring and temperature on the formation celluloses I α and I β as revealed by cross polarization/magic angle spinning (CP/MAS) 13 C NMR spectroscopy, *Cellulose.,* **1**: 57–66 (1994).
115. Szymańska-Chargot M., Chylińska M., Cybulska J., Kozioł A., Pieczywek P.M., Zdunek A., Simultaneous influence of pectin and xyloglucan on structure and mechanical properties of bacterial cellulose composites, *Carbohydr. Polym.,* **174**: 970–979 (2017).
116. Huang H.-C., Chen L.-C., Lin S.-B., Hsu C.-P., Chen H.-H., In situ modification of bacterial cellulose network structure by adding interfering substances during fermentation, *Bioresour. Technol.,* **101**: 6084–6091 (2010).
117. Khodamoradi N., Babaeipour V., Sirousazar M., Bacterial cellulose/montmorillonite bionanocomposites prepared by immersion and in-situ methods: structural, mechanical, thermal, swelling and dehydration properties, *Cellulose.,* **26**: 7847–7861 (2019).
118. Saibuatong O.-A., Phisalaphong M., Novo aloe vera–bacterial cellulose composite film from biosynthesis, *Carbohydr. Polym.,* **79**: 455–460 (2010).

119. Cheng K.-C., Catchmark J. M., Demirci A., Effect of different additives on bacterial cellulose production by Acetobacter xylinum and analysis of material property, *Cellulose.,* **16**: 1033–1045 (2009).

120. Gao G., In situ production of bacterial cellulose/xanthan gum nanocomposites with enhanced productivity and properties using Enterobacter sp. FY-07, *Carbohydr. Polym.,* **248**: 116788 (2020).

121. Dayal M. S., Catchmark J. M., Mechanical and structural property analysis of bacterial cellulose composites, *Carbohydr. Polym.,* **144**: 447–453 (2016).

122. Cielecka I., Szustak M., Kalinowska H., Gendaszewska-Darmach E., Ryngajłło M., Maniukiewicz W., Bielecki S., Glycerol-plasticized bacterial nanocellulose-based composites with enhanced flexibility and liquid sorption capacity, *Cellulose.,* **26**: 5409–5426 (2019).

123. Hernández-Hernández H. M., Acetosolv treatment of fibers from waste agave leaves: Influence of process variables and microstructural study, *Ind. Crops Prod.,* **86**: 163–172 (2016).

124. Hernández-Varela J. D., Chanona-Pérez J. J., Benavides H. A. C., Sodi F. C., Vicente-Flores M., Effect of ball milling on cellulose nanoparticles structure obtained from garlic and agave waste, *Carbohydr. Polym., **1**:* 117-147 (2020).

125. Huang H.-C., Chen L.-C., Lin S.-B., Hsu C.-P., Chen H.-H., In Situ crystallization of bacterial cellulose I. Influences of polymeric additives, stirring and temperature on the formation celluloses Io~and I/3 as revealed by cross polarization/magic angle spinning (CP/MAS) 13CNMR spectroscopy, *Bioresour. Technol.,* **101**: 6084–6091 (2010).

126. Stumpf T. R., Yang X., Zhang J., Cao X., In situ and ex situ modifications of bacterial cellulose for applications in tissue engineering, *Mater. Sci. Eng. C.,* **82**: 372–383 (2018).

127. Ul-Islam M., Khan T., Park J. K. Water holding and release properties of bacterial cellulose obtained by in situ and ex situ modification, *Carbohydr. Polym.,* **88**: 596–603 (2012).

128. Fatima A., Ex situ development and characterization of green antibacterial bacterial cellulose-based composites for potential biomedical applications, *Adv. Compos. Hybrid Mater.,* **5**: 307–321 (2022).

129. Cazon P., Vazquez M., Improving bacterial cellulose films by ex-situ and in-situ modifications: a review, *Food Hydrocoll.,* **113**: 106514 (2021).

130. Wahid F., A facile construction of bacterial cellulose/ZnO nanocomposite films and their photocatalytic and antibacterial properties, *Int. J. Biol. Macromol.,* **132**: 692–700 (2019).

131. Menegasso J.F., Moraes N.A.C., Vasquez T.P., Felipetti F.A., Antonio R.V., Dutra R.C., Modified montmorillonite-bacterial cellulose composites as a novel dressing system for pressure

injury, *Int. J. Biol. Macromol.,* **194**: 402–411 (2022).

132. Horue M., Antimicrobial activities of bacterial cellulose–Silver montmorillonite nanocomposites for wound healing, *Mater. Sci. Eng. C.,* **116**: 111152 (2020).
133. Li Y., Composites of bacterial cellulose and small molecule-decorated gold nanoparticles for treating Gram-Negative bacteria-infected wounds, *Small.,* **13**: 1700130 (2017).
134. Janpetch N., Saito N., Rujiravanit R., Fabrication of bacterial cellulose-ZnO composite via solution plasma process for antibacterial applications, *Carbohydr. Polym.,* **148**: 335–344 (2016).
135. Khalid A., Bacterial cellulose–TiO 2 nanocomposites promote healing and tissue regeneration in burn mice model, *RSC Adv.,* **7**: 47662–47668 (2017).
136. Sun Y., The effects of two biocompatible plasticizers on the performance of dry bacterial cellulose membrane: a comparative study, *Cellulose.,* **25**: 5893–5908 (2018).
137. Cobos M., González B., Fernández M. J., Fernández M. D., Study on the effect of graphene and glycerol plasticizer on the properties of chitosan-graphene nanocomposites via in situ green chemical reduction of graphene oxide, *Int. J. Biol. Macromol.,* **114**: 599–613 (2018).
138. Almeida I. F., Bacterial cellulose membranes as drug delivery systems: An in vivo skin compatibility study, *Eur. J. Pharm. Biopharm.,* **86**: 332–336 (2014).
139. Ray D., Sain S., In situ processing of cellulose nanocomposites. *Compos. Part A Appl. Sci. Manuf.,* **83**: 19–37 (2016).