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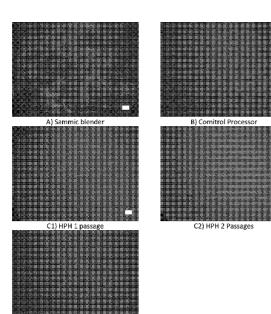
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(54) Title: BACTERIAL CELLULOSE FORMULATIONS, METHODS AND USES THEREOF



(57) Abstract: The present disclosure relates to powdered, rehydratable, bacterial cellulose formulations comprising methods of production and uses thereof. In particular the use of the formulation as a colloid stabilizer, foam stabilizer, or as a thickener, as a reinforcer material (as a filler), a dietary fibre, a foodstuff, a cosmetic or pharmaceutical composition, a composite, among others. An aspect of the present subject matter discloses a powdered formulation, comprising bacterial cellulose and an additional component (or third component) selected from the following list: sodium carboxymethyl cellulose, carboxymethyl cellulose, xanthan, methylcellulose, methyl cellulose, hydroxyethylcellulose, hydroxyethyl-cellulose, hydroxypropyl methyl cellulose, hydroxypropyl methylcellulose, tylose, glycerol, saccharose, or mixture thereof; wherein the powdered formulation is dispersible in an aqueous media, at 20 °C, with low shear mixing.

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BACTERIAL CELLULOSE FORMULATIONS, METHODS AND USES THEREOF

Technical field

[0001] The present disclosure relates to wet and powdered, rehydratable, bacterial cellulose formulations comprising methods of production and uses thereof.

[0002] The wet and powdered bacterial cellulose formulations of the present disclosure are useful in medicine, cosmetic, food, detergents, polymers and composites industries, among others.

Background

[0003] Bacterial cellulose (BC) is an exopolysaccharide synthesized by certain Gram-negative, obligate aerobic, acetic acid bacteria, the *Komagataeibacter* genus being the most important due to the high cellulose yield obtained. Morphologically BC is comprised of randomly assembled ribbon-shaped fibrils less than 100 nm wide and composed of aggregated bundles of elementary nanofibrils; these fibrils have a lateral size of 7-8 nm and several micrometres in length. As *Komagataeibacter* microorganisms are mandatory aerobes, under static conditions, BC is synthesized at the air/liquid interface of the culture medium [1-18].

[0004] For bulk commercial applications, dried BC formulations offer several advantages over aqueous suspensions, such as a decrease in the size and mass, thus lower storage space and transportation costs, improved storage stability, lower risk of contamination. It is known that, as with plant cellulose, the properties of BC are mostly lost upon drying. As water evaporates, the fine fibres from BC aggregates, establishing hydrogen bonds that are not easily broken upon resuspension in water [19]. Several attempts have been made to prepare dried BC formulations, some of these, addressing the restauration of the cellulose properties after aqueous dispersion, as will be reviewed below.

[0005] Document US 4960763 A [20] is related to the use of BC as a dietary fibre, to absorb/bind cholesterol (*in vitro*). BC, alone or in combination with sucrose, was either freeze-dried or frozen and cryomilled. While reasonably effective in preventing hydrogen bonding between cellulose fibres, these are rather costly solutions for industrial implementation. In this document, a comparison of the properties of BC, before and after drying, was made only in terms of the effect on cholesterol adsorption. Also, the authors recognize that, regardless of the drying method, drying BC alone strongly

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reduced the cholesterol adsorption properties of BC, as compared to the never-dried BC suspension. The addition of sucrose only partially mitigated such limitation.

[0006] Document US 5962676 A [21] describes a drying method for BC. High pressure homogenization and thin screw extrusion, in both cases with temperature treatment, were studied for the homogenization of BC. The BC was then dried by drum drying, spray drying, extrusion drying, oven drying and frying in oil. BC was also dried in the form of sheets and ground-milled. Regardless of the different drying method, maximum BC swelling took from 30 min to up to 18 hr. This is unpractical for industrial applications; also, this document did not establish whether the properties of the swelled BC are equivalent to those of the never-dried one.

[0007] Document JP 2873927 B2[22] proposes the addition of a third component to an aqueous BC suspension before drying, through several methods, such as oven drying, drum drying, infra-red drying. In this document, disintegration of BC was done with a kneader extruder at maximum speed, for up to 15 min, before drying. Examples of the tested third component included calcium carbonate, at 50 times the mass of BC, to obtain a dispersible product (under 30 sec, stirring with a spatula). A mixture of BC with calcium carbonate at 5 times the amount of BC and heat treated (120 °C, 20 min) after mixing in water with a spatula, results in a dispersible suspension. When glycerine was used as a third component, at a mass ratio BC:glycerine of 1:2, a dispersion could be achieved also by first heating the aqueous mixture to 100 °C, for 1 hr. Without heating, glycerine, at 50 times the amount of BC was required to allow obtaining a dispersible product. However, only when glycerine was added at 500 times the amount of BC (or 200 times if previous heating was used), nearly 80% of the initial product viscosity could be recovered. In all these cases, the stability of the dispersed products (as assessed by the precipitation degree following centrifugation) was low. BC was also mixed with carboxymethyl cellulose (CMC) and dried. A mixture composed of a mass ratio of BC:CMC of 1:5 was dispersible, with 71% of the initial product viscosity recovery and 100% stability recovery. Other polysaccharides (such as dextrin, xanthan, soluble starch) and combinations of polysaccharides and glycerin or polysaccharides and CMC, were tested as the "third" component, to be added to BC aqueous suspensions before drying. To the exception of BC:dextrin at 1:1 mass ratio, all mixtures required an excess of the third component, to allow for an almost complete viscosity and stability recovery, following aqueous dispersion. One example of that document, related to the preparation of a BC/dextrin mixture, addresses the grinding of the dried product, to a final particle size of 400 μm or less. Aqueous dispersion, at 0.2%, of the powder, with magnetic stirring, took 30 seconds, with complete recovery of the viscosity, as compared to the non-dried mixture. The dispersion time was defined as the time until large macroscopic lumps were no longer observable to the naked eye. However, no details were provided on the mass ratio of BC and dextrin, in this specific mixture. In all

these cases, after stirring in water, the dried material was submitted to heat treatment at 120 °C, for 10 min. Clearly, heat treatment (a time consuming and costly solution for industrial implementation) and excess of a third component (resulting actually in a small amount of BC in the final product) are required to obtain a dispersible product.

[0008] Document US 6153413 A [24] proposes several drying methods of BC alone (i.e. without the need of adding a third component to the aqueous BC suspension), allowing the re-stauration of its properties. According to this document, during drying and dehydrating of BC fibres, tensile stress is exerted on both the crystalline and amorphous parts of the fibres. Since elasticity is higher in the crystalline parts than in the amorphous ones, a degree of strain for the tensile stress is higher in the amorphous parts. As bonds in the amorphous parts of the cellulose fibres consist of hydrogen bonds between and within cellulose molecules, the distance between the hydrogen bonds will be enlarged due to the high degree of strain, allowing water molecules to penetrate. As a result, the hydrogen bonds between the BC fibrils with the high degree of strain will be easily broken upon the addition of water and the hydrogen bonds between BC and water molecules, which existed before dehydration and drying, will be easily re-formed. In such document, the "dry" state means a state in which water is present in an amount of about 25% or less on the basis of dry BC mass. To demonstrate the dispersibility of dried BC, a BC homogenate (blended at 18.000 Rpm, for 2 min, with a blender) was dried by pouring the BC homogenate into trays and using either air drying, oven drying, infra-red drying; a BC homogenate was also freeze dried and dried under vacuum or in a drum dryer. Then, a BC aqueous suspension was prepared by further homogenization of the dried BC, using an ultraturrax (Physcotron), at maximum speed (30.000 Rpm) for 1 min. At such rotational speeds, ultraturrax exerts quite a substantial shear stress, thus most likely inducing extensive BC fibre breakdown and/or defibrillation. This is further reinforced by a noticeable decrease in the mechanical properties of dried BC sheets that were previously submitted to ultraturrax treatment, as compared to BC that was only ground with a blender (control). Further, the authors claim a good recovery of the dispersibility and viscosity of dried BC (to the exception of spray-drying), but the never-dried BC samples (controls) were only submitted to homogenization at 18.000 Rpm for 2 min, not ultraturrax. The obtained results are therefore not comparable. From the above, regardless of the drying method, a high shear force is most likely responsible for the dispersion of the BC fibres. For industrial applications, this implies added capital investment (the need for a high shear mixing equipment) and operating costs.

[0009] Documents, US 20100016575 A1 and US 8053216 B2 [25, 26], describe the addition of polysaccharide hydrocolloids as co-agents, to BC, before drying. Dispersion of the dried material could be done by high shear mixing (e.g. high pressure homogenization at 2500 psi [25] or 8000 rpm for 10 min with a Silverson mixer, 1500 psi for 2 passes with an extensional homogenizer and 2500 rpm for

10 min [26]), with or without the addition of additional co-agents. These documents propose the use of alcoholic solutions, for the co-precipitation of the BC-hydrocolloid mixture; this is followed by concentration, drying and milling. However, for large-scale implementation, very high alcohol volumes are required for the co-precipitation. As such, the use of alcohols, the inevitable product losses (from the non-precipitated material), the need for alcohol recovery equipment, all contribute significantly to the total cost of production. The use of such organic solvents also poses additional safety and environmental concerns. In these documents, the fermented broth containing BC is treated with hypochlorite, glycosidases and proteases (to lyse the cells). Hydrocolloids are mixed and then organic solvents are added. However, co-precipitation of culture media components and bacterial cell hydrolysates will also occur, thus limiting the degree of purity (hence the quality) of the final product. The use of charged hydrocolloids further aggravates the co-precipitation of unwanted substances, due to ionic interactions.

[0010] Another document, WO 2001005838 A1 [27], discloses a process for drying reticulated (crosslinked) BC without co-agents: this involves dispersing a BC suspension in an organic solvent using high shear mixing and milling, concentrating the dispersion to substantially remove the solvent, and drying. Optionally, the dried material may be ground. Exemplary organic solvents useful in this disclosure include hydrocarbons, alkyl alcohols, alkyl sulfoxides, mixtures thereof or aqueous mixtures thereof. Preferably, the cellulose material is dried using a multi-step process comprising alternating drying and grinding processes. As with the previous documents, here too, a strong limitation exists regarding the use of organic solvents. The proposed method also presents superfluous unpractical steps for use at industrial scale, namely the addition followed by removal of the organic solvent and the multi-stage drying and grinding. Also, in all examples, co-agents (one or more) such as xanthan, CMC, sucrose, were mixed with dried BC before aqueous dispersion. Further, no details on the particle size, required time and energy requirements for dispersion of the dried material (only one example of such document describes the milling of dried BC to 20 mesh size (< 850 µm)). Finally, a comparison of the properties of the BC before and after drying was not presented.

[0011] From the above, several approaches have been considered to obtain redispersible BC formulations, some of which claiming the restoration of its properties, as compared to those of the never-dried formulations. However, the redispersion methods involve the use of high energy mixing and/or high temperature and/or long dispersion times and/or the use of excess amounts of third components, still with limited results regarding the recovery of the properties of the original material. Further, these energy and time intensive processes represents high capital and operating costs, when considering industrial applications.

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[0012] Most of the mentioned documents do not fully demonstrate the recovery of the BC technological properties upon redispersion, being mainly focused on demonstrating the BC's redispersibility, which does not necessarily imply the recovery of the technological properties of the never-dried material. This is also the case with document US006069136A [28], that, instead of a dried product, provides a method of producing concentrated BC having improved properties such as dispersibility, suspensibility and viscosity, encompassing the homogenization (wet grinding) and concentration of BC. As observed for the other documents, the claimed restauration of the BC properties is limited to demonstrating the redispersibility as obtained by different concentration methods, different targeted BC final concentrations and different redispersing methods. No information was provided on the particle size range of the ground material, nor on the functional properties of the redispersed one.

[0013] As will be observed in the present disclosure, the apparent (macroscopic) BC redispersion or the recovery of a viscosity similar to never dried samples (a complex and potentially misleading property when dealing with heterogeneous systems) does not fully demonstrate the restauration of its properties. The technical properties, i.e., the functional properties of BC (e.g. the stabilization of multiphasic systems), provide more detailed information.

[0014] These facts are disclosed in order to illustrate the technical problem addressed by the present disclosure.

General description

[0015] The present disclosure relates to dried powdered and rehydratable bacterial cellulose (BC) formulations, comprising methods of production and uses thereof. In particular the use of the formulation as a colloid stabilizer, foam stabilizer, or as a thickener, as a reinforcer material (as a filler), a dietary fibre, a foodstuff, a cosmetic or pharmaceutical composition, a composite, among others.

[0016] The present disclosure concerns with the conditions by which dried powdered BC formulations, capable of being dispersed in aqueous media under 5 minutes, at room temperature, using low shear mixing, can be obtained. Such formulations preserve the technical properties of the non-dried material, in particular but not only, as a colloid stabilizer.

[0017] The present disclosure also relates to wet BC formulations, methods of production and uses thereof. The wet BC of the present disclosure can be used as a colloid stabilizer, foam stabilizer, as a

thickener, as a reinforcer material (as a filler), a dietary fibre, a foodstuff, a cosmetic or pharmaceutical composition, a composite, among others.

[0018] The present disclosure concerns with the conditions by which dried powdered BC formulations, capable of being dispersed in aqueous media under 5 minutes, at room temperature (20-25 °C), using low shear mixing (at most 1500 rpm, 1000 rpm), can be obtained. Such formulation preserves the technical properties of the non-dried material, in particular, but not only, its performance as a colloid stabilizer. More specifically, the level of grinding involved in the processing of the BC formulations must be controlled in such a way that full redispersion is possible, while not compromising the functional properties. The smaller the particle size, the better the redispersion. However, the comminution conditions required for optimal restoration of the BC technical properties may be achieved at non-optimal conditions concerning redispersion.

[0019] In an embodiment, the full restoration of the BC technical properties requires the addition of a third component but may be achieved regardless of the drying technology used or the amount of residual water in the dried formulation.

[0020] In an embodiment, the presence of a third component (or an additional component) may not always be desired, due to changes in the functional properties of native BC. However, dried BC (without the added third component) cannot be redispersed in aqueous media. Alternatively, a concentrated BC suspension can be prepared, also providing the full redispersion and restauration of the functional properties; however, the wet comminution of BC performed before the concentration step must also be controlled in such a way that the technical properties of BC are not compromised.

[0021] This disclosure encompasses the conditions by which dried BC formulations (containing preferably from 0% (w/w) to 30%(w/w) of water) can be obtained. Further, it relates to the conditions by which a concentrated wet BC formulation can be obtained. In both cases, the BC formulations disperse quickly in aqueous media, under low shear stress, at room temperature. Once dispersed, the properties of the dried material are equivalent to those of the never dried one.

[0022] For the purpose of the present invention, BC may be produced from a bacterium. The genus of the bacterium may be Acetobacter, Agrobacterium, Gluconacetobacter, Rhizobium, Achromobacter, Alcaligenes, Aerobacter, Azotobacter, Rhizobium, Salmonella, Escherichia and Sarcina. Preferably, BC is obtained using pure strains belonging to the genus Acetobacter or Komagataeibacter (formerly Gluconacetobacter), as these are the most productive, but any other cellulose-producing microbial strain, natural or mutated, may be used. The aerobic fermentation of BC can be done by any know static or agitated/aerated conditions. Any known culture fermentation method such as batch, fed

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batch, repeated batch or continuous fermentation may be used. Several feed stocks can be used for bacterial growth and cellulose production, provided they are free of contaminants (exogenous microorganisms or toxicants). For instance, carbon sources may be individual sugars or mixtures thereof, such as glucose and fructose, disaccharides such as sucrose, and mixtures of monosaccharide and disaccharides. In addition, the carbon source can be supplied as a complex mixture of sugars such as molasses, or plant biomass hydrolysates such as wood hydrolysate, straw hydrolysate, corn stalk hydrolysate, sorghum hydrolysate or other biomass wastes. Alternative carbon sources such as ethanol, glycerol or other sugar alcohols, alone or in combination with the previously mentioned, can also be used. Also, several nitrogen sources, organic or inorganic ones, can be used, alone or combined, such as - but not limited to – ammonium sulphate, ammonium chloride, ammonium phosphate, urea, sodium nitrate, yeast extract, corn steep liquor, whey hydrolysate, peptone, casein hydrolysate. Trace amounts of organic nutrients may be further added, including amino acids, vitamins, fatty acids, nucleic acids, 2,7,9-tricarboxy-1H pyrrolo [2,3,5]-quinoline-4,5-dione, ethanol, sulphite pulp waste liquor, lignin sulfonic acid and the like. Preferentially, the effective pH range of the culture media is in the range of 4-6. Optionally, during fermentation, the pH may be controlled using buffer solutions, such as, but not limited to, citrate or 3,3-dimethylglutaric acid, or the addition of base or acid to the medium in sufficient amounts, to maintain the pH within the desired range. Preferentially, the temperature for inoculum development and fermentation is in the range of 27-32 °C. Following fermentation, bacteria and remnants from the fermentation media can be removed by a variety of protocols. These include washing with water, dilute acid and/or alkali, bleaching with sodium hypochlorite or hydrogen peroxide, lysing the bacteria with lytic enzymes such as lysozyme, treatment with surfactants such as sodium lauryl sulphate or sodium deoxycholate. Washing procedures can be done at a temperature range between room temperature and 200 °C, with any combination of the above treatments. Those skilled in the art may optionally select other fermentation and purification conditions.

[0023] In an embodiment, BC obtained by any of the means described above, at a concentration in the range between 0.5% (m/v) and 10% solids (BC) preferably between 0.5% and 5% solids, is submitted to the following sequential steps, to yield a wet concentrated formulation or a dried powdered formulation:

• Wet comminution of BC (henceforward designated as "ground"), obtained by methods such as, but not limited to, mechanical shearing stress, wet grinding mills, crushing, cutting, trituration, aqueous counter-collision, ultrasound, high-pressure homogenization, hydrolysis with an acid or enzyme, or bleach, or combination thereof. Those skilled in the art may optionally select other methods and conditions. It was surprisingly observed that the degree of comminution plays an important role in the final properties of the concentrated or dried

material obtained through further processing: a coarsely ground BC yields long fibril aggregates (bundles); too extensive comminution will yield extensive individual BC fibres and/or extensive fibre break up. Thus, an optimal particle size or optimal particle size range must be achieved as to fully preserve the BC properties.

- The ground BC is concentrated to a final solids content within the range of 5%(w/v) to 70%(w/v), preferably in the range of 10%(w/v) to 30%(w/v) (wet BC formulation). For the purposes of the present invention, concentration of BC can be achieved by any solid-liquid separation method such as, but not limited to, centrifugation, press filter, sedimentation and settling, screw press, roller press, juice reamers, alone or combined in a multistep process and with reverse osmosis. Those skilled in the art may optionally select other concentration methods and conditions.
- The non-concentrated ground BC may alternatively be mixed by any of the existing mechanical shearing processes and equipment, with a third component encompassing any hydrophilic liquid, any water-soluble substance or any water-insoluble substance or a mixture of more than one of any of these substances. Examples of such substances include, but are not limited to, glycerin, ethylene glycol, dimethyl sulfoxide, dimethylformamide, lactic acid, gluconic acid and delta gluconolactone, a low molecular mass saccharide (e.g. glucose, fructose, galactose, xylose, mannose, arabinose, sucrose, lactose, cellobiose, palatinose, maltose, gentiobiose, trehalose, rhamnose, isomalto-oligosaccharide, soybean oligosaccharide, oligosaccharide, galactooligosaccharide, lactosucrose, coupling sugar, liquid sugar, cyclodextrin, sugar alcohols, sorbitol, erythritol, lactitol, maltitol, xylitol, mannitol, dulcit), salts (sodium sulfate, ammonium sulfate, sodium chloride, calcium chloride, sodium bicarbonate, sodium carbonate, Rossel salt), aminoacids, amino acid salts, acidic acids, organic acids, nucleic acids, nucleic acid salts, high molecular mass polysaccharides (e.g. xanthan gum, xyloglucan, dextrin, dextran, carrageenan, locust bean gum, tamarind gum, alginic acid, alginate, pullulan, starch, starch, CMC, methylcellulose, hydroxypropylcellulose, ethylcellulose, corn starch, gum arabic, guar gum, gellan gum, polydextrose, pectin), chitin, chitosan, casein, alpimine, soybean protein lysate, peptone, polyvinyl alcohol, polyacrylamide, polyacrylic acid sodium polyvinylpyrrolidone, a polyamide or polyamideimine or polyamine resin, polyethylene oxide, hydrophilic crosslinked polymer, polyacrylic acid copolymer, colloidal silica.
- The mixing of the ground BC and a third component can be done at a temperature range within room temperature to 80 °C, depending on the nature of the added substance. The third component can be added to ground BC in its dried form; alternatively, a stock solution can be prepared, before mixing with the ground BC. The amount of the third component to be added

may be optionally determined by those skilled in the art, depending on the kind of the material and the like, being usually in the range of from 2% by weight to 1000% by weight of BC, preferably between the range of 50% to 200% by weight (or mass) of BC. Preferably, the third component is a water-soluble polysaccharide or mixtures of these substances.

- The drying of BC mixtures with a third component, by means of any of the available drying processes and equipment, which include spray drying, drum drying, oven drying, vacuum drying, tunnel drying, infra-red drying, freeze drying. Those skilled in the art may select any other proper drying process and/or combination of methods. Contrary to what was observed in previous documents, the "dry" state may be a completely dried BC formulation, in which there is no residual water. The properties of the fully dried BC formulations could be restored to those of their original wet state.
- Grinding of the dried BC formulations, which can be achieved using equipment well known by those skilled in the art, such as roll crusher, roll mill, pan mill, strainer, hammer mill, hammer crusher, pin disk, colloidal mil, disintegrator, rotary grinder, impeller attritor, jet mil. As observed with the wet comminution of BC, the particle size of the dried powdered material (henceforward defined as the ground dry BC formulation) is determinant to the restauration of its original technical properties (those of the never dried, coarsely comminuted material), with an optimal particle size or range, being necessary to achieve such restauration.

[0024] In an embodiment, both wet and dry comminution must be controlled in such a way that full redispersion is possible while not compromising the functional properties. Thus, the restoration of the BC technical properties may be achieved at non-optimal conditions concerning redispersion. Indeed, the smaller the particle size, the better the redispersion, but the same does not apply to the technical properties.

[0025] An aspect of the present disclosure relates with powdered formulations comprising BC dispersible in an aqueous solution, at 20 °C using low shear mixing, and an additional (third) component selected from the following list: sodium carboxymethyl cellulose, carboxymethyl cellulose, xanthan, methylcellulose, methyl cellulose, hydroxyethyl-cellulose, hydroxyethyl-cellulose, hydroxypropyl methyl cellulose, hydroxypropyl methylcellulose, glycerol, saccharose, or mixture thereof.

[0026] An aspect of the present subject matter discloses a powdered formulation comprising a powdered bacterial cellulose and an additional component (or third component) selected from the following list: sodium carboxymethyl cellulose, carboxymethyl cellulose, methyl

cellulose, hydroxyethyl-cellulose, hydroxyethyl-cellulose, hydroxypropyl methyl cellulose, hydroxypropyl methylcellulose, tylose, glycerol, saccharose, or mixture thereof; wherein the powdered formulation is dispersible in an aqueous media, at 20 °C using low shear mixing. In particular, the low shear mixing is at most 1500 rpm, preferably at most 1000 rpm

[0027] The measurement of the dispersibility of the BC powdered formulation may be carried out in various ways. In the present disclosure, samples are prepared at 0.5% (m/v) in water at room temperature (i.e. within the range of 20 °C). For this, a low mechanical shear dispersion using magnetic stirrer plate (Stuart SD162), at 900 Rpm, for up to 10 min. The dispersed materials are spread over a petri dish and observed at naked eye. Dispersibility of the formulations is classified as follows: 1-sample is homogeneous and no visible particles or aggregates are observable; 2 - sample contains some very small particles or aggregates; 3 – the sample contains some larger particles or aggregates; 4 – the water remains transparent and the well separated particles or aggregates are observed.

[0028] In an embodiment, for better results, the powdered BC formulation may be dispersed in an aqueous media in at most 10 min; preferably in at most 6 min; more preferably in at most 4 min.

[0029] In an embodiment, for better results, the powdered BC formulation is dispersed in an aqueous media in at most 2 min; preferably in at most 1 min. In particular, it may use particles of at least 100 μ m.

[0030] In an embodiment, for better results, the particles may comprise a size between 5-500 μ m, preferably between 80 – 400 μ m, more preferably 100-300 μ m. These dimensions are particularly suitable for foodstuff or for use as, more in particular beverages, dairy based beverages, in particular chocolate milk.

[0031] In an embodiment, for better results, the D50 of the particle is between 50-300 μ m, preferably the D50 of the particle is between 80-300 μ m, more preferably the D50 of the particle is between 100-200 μ m. These dimensions are particularly suitable for foodstuff or for use as, more in particular beverages, dairy based beverages, in particular chocolate milk.

[0032] In an embodiment for better results, the powdered BC formulation is dispersed in an aqueous media in at most 2 min and wherein the particles comprise a size between 5-100 μ m; more preferably 10-60 μ m. These dimensions are particularly suitable for composite reinforcement.

[0033] In an embodiment, for better results, the D50 of the particle is between 10-90 μ m, preferably the D50 of the particle is between 20-80 μ m, more preferably the D50 of the particle is between 30-60 μ m. These dimensions are particularly suitable for composite reinforcement.

[0034] The measurement of the particle size may be carried out in various ways, in this disclosure the measurement was carried out on the basis of the standard granulometry analysis by mechanical sieving. In particular concerning the sizes of the particles obtained by the sieves specified: Mat.Mesh:AISI 316 n^{o} 1/2871/1 N^{o} : 1/2871/1 with an opening of 0.300 mm, Endecotts, Ltd, aperture 212 μ m and Endecotts, Ltd, aperture 106 μ m.

[0035] In an embodiment, for better results, wherein the mass ratio between BC and the additional component varies between 1:5 to 1:0.2, preferably 1:2 to 1:0.5.

[0036] In an embodiment, for better results, the additional component (third component) may be selected from a list consisting in: carboxymethyl cellulose, carboxymethyl cellulose and xanthan; carboxymethyl cellulose and hydroxyethyl-cellulose and carboxymethyl cellulose and hydroxypropyl methylcellulose, or mixtures thereof.

[0037] In an embodiment, the powdered BC formulation is obtained from a bacterium of the following list: Acetobacter, Agrobacterium, Gluconacetobacter, Achromobacter, Alcaligenes, Aerobacter, Azotobacter, Rhizobium, Salmonella, Escherichia, Sarcina, Komagataeibacter, or combinations thereof. Preferably Acetobacter, Komagataeibacter, Gluconacetobacter or combinations thereof.

[0038] Another aspect of the present disclosure relates to a wet BC formulation comprising

5-70% (wt./v) of bacterial cellulose,

wherein the BC is dispersed in bundles;

wherein the bacterial cellulose bundles size varies between 20 μm to 2 mm.

[0039] The measurement of the BC bundle size may be carried out in various ways. In this disclosure it was achieved by aqueous dilution of BC to a final concentration of 0.01% (w/v); the suspension was left under mild agitation (orbital shaker at 100 rpm) for 2 hours and then vortexed for 1 minute prior to being stained with Calcofluor. The size of the bundles as expressed in this disclosure concerns the longer dimension as observed by fluorescence microscopy.

[0040] In an embodiment, for better results, the BC bundles size may vary between 20 μ m to 2 mm. Preferably the bundles size of BC may vary between 200 μ m to 1 mm. These dimensions are particularly suitable for foodstuff or for use as, more in particular smoothies and foams.

[0041] In an embodiment, for better results, the BC bundles size may vary between 20 μ m to 400 μ m. Preferably the bundles size of BC may vary between 30 μ m to 200 μ m. These dimensions are particularly suitable for nanocomposites and filler reinforcement.

[0042] In an embodiment, the wet formulation may comprise of 0.3-70% (w/v) BC solids. Preferably, the water dispersion may comprise of 5-40% (wt./v) BC solids. More preferably, the water dispersion may comprise of 10-30% (wt./v) BC solids.

[0043] Another aspect of the present disclosure relates to the use of the powdered BC formulation described in the present disclosure as a thickener. In particular, the particles may comprise a size between 5-500 μ m, preferably between 80-400 μ m, more preferably 100-300 μ m. Preferably, the formulation may be used in foodstuff, more in particular in beverages, dairy base beverages, even more in particular chocolate milk.

[0044] Another aspect of the present disclosure relates to the use of the powdered BC or the wet BC described in the present disclosure as a reinforcer material, namely as filler. Preferably, by the use of a powdered BC formulation wherein the particles comprise a size between 5-100 μ m; more preferably 10-60 μ m.

[0045] Another aspect of the present disclosure relates to a pharmaceutical or a cosmetic composition comprising at least an active ingredient and the powdered BC formulation, or the wet BC described in the present disclosure.

[0046] Another aspect of the present disclosure relates to a dietary fibre or a foodstuff comprising an effective amount of powdered BC formulation or the wet BC described in the present disclosure.

[0047] Another aspect of the present disclosure relates to a composite comprising an effective amount of powdered BC formulation or the wet BC described in the present disclosure.

[0048] Method for producing a BC formulation of dried powder comprising the following steps of: providing a suspension of BC bundles in water, with a size between 20 µm to 2 mm, preferably 100 µm to 1.5 mm, more preferably 200 µm to 1 mm; adding a third component at a temperature between 20-80 °C; drying the BC formulation until obtaining at least 60% of solids; grinding the BC formulation.

[0049] In an embodiment, the step of providing a suspension of BC bundles (i) comprises wet comminution.

[0050] In an embodiment, the BC powdered formulation comprises a size between 5-500 μ m, preferably between 100-300 μ m.

[0051] In an embodiment, the concentration of BC in the ground BC suspension varies between 0.3 - 10% (wt./v).

[0052] In an embodiment, the concentration of BC in the ground BC suspension varies between 0.5–5 % (wt./v).

[0053] In an embodiment, the additional (third) component may be selected from a list consisting of: glucose, fructose, galactose, xylose, mannose, arabinose, sucrose, lactose, cellobiose, palatinose, maltose, gentiobiose, trehalose, rhamnose, isomalto-oligosaccharide, soybean oligosaccharide, fructo-oligosaccharide, galactooligosaccharide, lactosucrose, coupling sugar, liquid sugar, cyclodextrin, sugar alcohols, sorbitol, erythritol, lactitol, maltitol, xylitol, mannitol, dulcit, xanthan gum, xyloglucan, dextrin, dextran, carrageenan, locust bean gum, tamarind gum, alginic acid, alginate, pullulan, starch, starch, carboxymethyl cellulose, methylcellulose, hydroxypropylcellulose, ethylcellulose, corn starch, gum arabic, guar gum, gellan gum, polydextrose, pectin or mixture thereof.

[0054] Another aspect of the present invention, relates to a method for producing a BC powdered formulation described in the present disclosure comprising the following steps of:

providing an aqueous suspension of ground BC bundles, with a size between 20 μ m to 2 mm, preferably 100 μ m to 1.5 mm, more preferably 200 μ m to 1 mm; adding a third component at a temperature between 20-80 °C; drying the BC formulation until obtaining at least 60% of solids; grinding the BC formulation until obtaining a particle size between 5-500 μ m.

[0055] In an embodiment, the concentration of BC in the ground BC suspension may vary between 0.3-10% (wt./v). Preferably the concentration of BC in the ground bacterial cellulose suspension varies between 0.5-5% (wt./v).

[0056] Another aspect of the present invention, relates to a method for producing wet formulation BC comprising the following steps of:

providing an aqueous suspension of ground BC bundles, wherein said bundles comprises a size between 20 μm to 2 mm, preferably 100 μm to 1.5 mm, more preferably 200 μm to 1 mm;

concentration of the aqueous ground BC suspension up to a concentration of 10-70%, preferably between 15-30%.

[0057] In an embodiment, the step of homogenizing the BC suspension may comprise wet comminution.

[0058] Where ranges are given, endpoints are included. Furthermore, it is to be understood that unless otherwise indicated or otherwise evident from the context and/or the understanding of one of ordinary skill in the art, values that are expressed as ranges can assume any specific value within the stated ranges in different embodiments of the invention, to the tenth of the unit of the lower limit of

the range, unless the context clearly dictates otherwise. It is also to be understood that unless otherwise indicated or otherwise evident from the context and/or the understanding of one of ordinary skill in the art, values expressed as ranges can assume any subrange within the given range, wherein the endpoints of the subrange are expressed to the same degree of accuracy as the tenth of the unit of the lower limit of the range

Brief Description of the Drawings

[0059] The following figures provide preferred embodiments for illustrating the description and should not be seen as limiting the scope of invention.

[0060] Figure 1 - Petri dish photographs of bacterial cellulose aqueous suspension, following homogenization with A) Sammic blender, B) Comitrol Processor (2 passages), High Pressure Homogenizer, after C1) 1 passage, C2) 2 passages and C3) 6 passages. White bar scale: 1 mm.

[0061] Figure 2 – Fluorescence photomicrographs of bacterial cellulose aqueous suspension, following homogenization with A) Sammic blender, B) Comitrol Processor (2 passages), High Pressure Homogenizer, after C1) 1 passage C2) 2 passages and C3) 6 passages. BC was stained with Calcofluor white and observed in an Olympus BX51 microscope. Scale: 100 µm.

[0062] Figure 3 – Effect of the addition of bacterial cellulose, plant nanocellulose and hydrocolloids on the overrun of whipped egg whites.

[0063] Figure 4 – Petri dish photographs of bacterial cellulose aqueous suspensions, following homogenization with Sammic blender, Comitrol Processor (6 passages) and High Pressure Homogenizer. Samples were concentrated to 20% solids and diluted and redispersed in water. White bar scale: 1 mm.

[0064] Figure 5 – Fluorescence photomicrographs of bacterial cellulose aqueous suspensions following homogenization, concentration to 20% solids and dispersion dilution in water. BC was stained with Calcofluor white and observed in an Olympus BX51 microscope. Scale: $100 \, \mu m$.

[0065] Figure 6 – Stability of whipped egg whites over time, as prepared by the addition of bacterial cellulose ground with Sammic blender (at 0.1, 0.2 and 0.3%), xanthan or Bioplus.

[0066] Figure 7 — Classification of the dispersibility of never-dried mixtures of BC and third components. 1 - sample is homogeneous and well dispersed; 2 - sample contains some very small BC aggregates; 3 - sample contains some larger BC aggregates

[0067] Figure 8 – Fluorescence photomicrographs of never-dried BC:CMC aqueous suspension, following homogenization in a A) Sammic blender, B) Comitrol Processor (2 passages) and High Pressure Homogenizer, after E) 6 passages. Following mixing, BC:CMC was dispersed with either I) magnetic stirrer or II) ultraturrax. Samples were stained with Calcofluor white and observed in an Olympus BX51 microscope. Scale: 50 µm.

[0068] Figure 9 – Rheological profile of 0.5% (m/v) BC:CMC samples diluted and dispersed using I) ultraturrax and II) magnetic stirrer.

[0069] Figure 10 – Rheological profile of 0.5% (m/v) BC:CMC samples diluted and dispersed using I) ultraturrax treatment and II) magnetic stirring.

[0070] Figure 11 -Rheological profile of 0.5% (m/v) BC:CMC samples ground at different particle sizes.

[0071] Figure 12 – Examples of % stability.

Detailed description of the invention

[0072] The present disclosure is also further described, in particular, using embodiments of the disclosure. Therefore, the disclosure is not limited to the descriptions and illustrations provided. These are used so that the disclosure is sufficiently detailed and comprehensive. Moreover, the intention of the drawings is for illustrative purposes and not for the purpose of limitation.

[0073] The present disclosure relates to powdered and wet BC formulations, methods of production and uses thereof.

[0074] The powdered BC formulation of the present disclosure is useful for use in medicine, food, cosmetic, among others.

[0075] The present disclosure concerns with the conditions by which a dried powdered BC formulation, capable of being dispersed in aqueous media under 5 minutes, at room temperature, using low shear mixing, can be obtained. Such formulation preserves the technical properties of the non-dried material, in particular but not only, the potential as a colloid stabilizer.

[0076] The present disclosure also relates to a never-dried BC, methods of production and uses thereof.

[0077] In an embodiment, the never-dried BC (or wet BC) of the present disclosure is useful for use in medicine, food, cosmetic, among others.

[0078] The present disclosure also concerns with the conditions by which a wet BC suspension can be obtained, also capable of quick and low energy dispersion, comprising the comminution of BC to a

specific size range, concentrating the aqueous suspension of BC to a final solids content of between 10% by dry mass or more and less than 70% by mass and dispersing it again into an aqueous solution.

[0079] For clarification purposes, the description of the present disclosure is divided into the following sequential steps:

- 1. Wet comminution of BC (production of "ground" BC);
- 2. Mixing of the ground BC with a third component;
- 3. Drying of BC formulations;
- 4. Grinding of BC formulations;
- 5. Alternatively to the steps 2-4, ground BC may be concentrated without being mixed with a third component.

Example 1 - Wet grinding of bacterial cellulose

[0080] In an embodiment, a BC obtained by any means as described in the previous section, in an amount of the range between 0.5% (m/v) and 10% solids (BC) preferably between 0.5% and 5% solids, preferably in the range of 0.7-1% (w/v) was wet ground by three different methods:

- A) Using a Sammic fixed speed blender, model TR250 at 9000 Rpm (Samic, S.L.) until a homogeneous pulp was obtained, as observed by visual inspection;
- B) By submitting BC to 2 passages through a Comitrol® Processor, Model 1700 (Urschel Laboratories Inc.). In each passage, grinding of BC was done at 900 Rpm, using a labyrinth verycut impeller and a cutting head 160 at 5° inclination;
- C) By High Pressure Homogenization (HPH) using a GEA Niro Soavi, model Panther NS3006L, at 600 Bar. Homogenized BC was collected after 1 passage (HPH 1 Pass), 2 passages (HPH 2 Pass) and 6 passages (HPH 6 Pass), through the High-Pressure Homogenizer. Before HPH processing, BC suspensions were first ground described in method A, to facilitate feeding of the suspension to the homogenizer.

[0081] BC suspensions were spread over petri dishes and observed with a SZ40 Zoom Stereo Microscope (Olympus). Photographs were taken with a camera SONY AVC-D5CE and adapter CMA-D5CE, at a magnification from 0.67x to 5x (Figure 1). BC suspensions were also stained with Calcofluor white and observed in an Olympus BX51 microscope.

[0082] Well-dispersed suspensions are a prerequisite in many industrial applications. BC suspensions are known to exhibit pronounced aggregation in aqueous media, due to strong interfibrillar hydrogen bonds and Van der Waals attraction, an effect that is concentration dependent. These attractive interactions, in combination with the long aspect ratio of the fibres, cause the formation of extended

networks when BC is dispersed in water. A ball of threads is obtained irrespective of whether BC is produced in a stirred tank or by static fermentation and then wet grinded. These highly heterogeneous dispersions consist of fibre bundles, flocs, and voids spanning tens to hundreds of micrometres depending on concentration and shear homogenization conditions (Figure 1 and Figure 2). In this example, three BC homogenization methods were used, with increasingly shear stress, from a blade blender (A), to high pressure homogenizer (C).

[0083] It was observed that with the increase in the shearing stress (from A to C), an increase in the BC defibrillation/fragmentation occurred. Homogenization of BC with Sammic blender showed larger fibre aggregates and BC fragments within 200 μ m to 1-1.5 mm size. BC homogenized with Comitrol Processor and HPH, lower amounts of millimetric fragments were observed, with most of them being in the range of 200-600 μ m (Comitrol), 100-200 μ m (HPH 1 pass) and 20-100 μ m (HPH 6 pass).

[0084] Increasing the number of passes by HPH resulted in the appearance of shorter fibres alongside with their increased separation. Due to their attractive forces and crosslinking, despite extensive deagglomeration, BC fibres always appear to be interconnected to neighbouring fibres, creating a 3D network of very fine fibres and other void areas containing only water. This effect occurred even at low BC concentrations (as low as 0.5%(w/v)) so that stable dispersions of well spread single fibres was never achieved.

Example 2 - Effect of bacterial cellulose on the overrun and foam stabilization of whipped egg whites [0085] Many food products are emulsions, foams or whipped emulsions (e.g. milk, cream, ice cream, butter, mayonnaise, finely minced meat like sausage meat, whipped egg white, whipped cream bread, etc). Foam and emulsion properties are strongly influenced by the interfacial properties of the emulsifier in the system. Polysaccharides (e.g. modified starches) and proteins, often play an important role in the formation and stabilization of these colloidal systems. These macromolecules provide the interface with physicochemical and rheological properties (steric hindrance, electrostatic repulsion, viscoelasticity), which determines the resistance to droplet coalescence. During the beating of egg whites, proteins adsorb on the newly-formed interface, denature and form a "gel" network that traps air bubbles and whose rheological properties govern the stability of the foam.

[0086] These non-dairy aerated products pose special concerns and constraints because the bubble wall is thin, relatively weak and unsupported (in contrast to oil droplets, which are supported by the mass of the oil). In these systems, two requirements exist. The first, a physical stabilization of the liquid within the interstitial regions of the foam. The second, a strengthening of the foam walls, accomplished using gums and other ingredients in the mix. Microcrystalline cellulose (MCC) and its derivatives,

hydroxypropyl cellulose (HPC), hydroxypropyl methylcellulose (HPMC), methyl etylcellulose (MEC) and other polysaccharide gums, such as xanthan have been used for the stabilization of foams.

[0087] In this embodiment, the effect of BC on the overrun and foam stabilization of whipped egg whites was evaluated. For this, BC from different sources (from *K. xylinus* strain ATCC 700178 and "nata de coco" from HTK Food CO. Ltd. (Vietnam), ground with a Sammic blender as described in Example 1, were used (code samples: 700178, S; Nata de coco, S. BC from ATCC 700178 was further homogenized with Comitrol and High Pressure Homogenization (Example 1) (code samples: BC Comitrol, BC HPH 1 Pass, BC HPH 6 Pass).

[0088] To compare the performance of BC against other hydrocolloids and plant nanocelluloses, several other samples were used in parallel: CelluForce NCC, Arbocel B 600, Arbocel BE 600/30 (from Celluforce Inc.) and Bioplus (from American Process Inc.) as sources of plant nanocellulose; xanthan, carboxymethyl cellulose, carrageenan (from Sigma) as examples of hydrocolloids.

[0089] As previously mentioned, BC suspensions are known to exhibit pronounced aggregation in aqueous media, due to strong interfibrillar hydrogen bonds and Van der Waals attraction, an effect that is more pronounced as the concentration of BC in aqueous media increases. To evaluate if concentrating BC affects the fibres dispersibility and functional properties, all BC samples were concentrated to 20%(wt/v) solids (code samples: 700178, S-P and Nata de coco, S-P), diluted and redispersped in water, under magnetic stirring at 900 Rpm for 2hr. According to the present embodiment, the BC concentrate may be prepared by dehydration with any known means such as a filter press, belt-press, centrifugation, vacuum filtration or any other method know to those of ordinary skill in the art.

[0090] Dried egg whites (12%, wt/v) were dispersed in water and whipped (using a Silvercrest kitchen blender). During whipping, each BC aqueous suspension or any other plant cellulose or hydrocolloid sample, was added to a final concentration of 0.1-0.3% (wt/v). A control was also done using only dried egg whites. The whipped cream's overrun was determined by measuring the volume increase of the whipped cream, relative to the control.

[0091] As observed in Figure 3, at 0.3%(wt/v) addition to the mixture, all BC samples ground with Sammic blender (700178, S and Nata de coco, S) showed a remarkable increase in the whipped cream's overrun. Contrarily, to the exception of Celluforce, with a marginal increase of 6% over the control, all plant nanocelluloses and hydrocolloids decreased the whipped cream's overrun. Increasing the extent of the homogenization of BC (HPH), which, as observed, decreases the size of the BC aggregates, decreases the capability of BC to stabilize the air/liquid interface, thus decreasing the cream's overrun. Finally, following compression of BC to 20%(wt/v) solids, low shear dispersion does not allow the full

lengthening of the BC fibre aggregates (Figure 5) when diluted and redispersed in water; however, provided the BC aggregates size is unaffected, compressing BC (Figure 4) does not affect the capacity to stabilize the foam.

[0092] These results clearly demonstrate the influence of the BC's particle size, as obtained by wet grinding, on the stabilization of protein foam.

[0093] Also, the stability of the foam was registered over time, at room temperature by evaluating, qualitatively, the accumulation liquid at the bottom of the flasks (Figure 6). Regardless of pressing or not the BC, results also showed that BC ground with the Sammic blender, at 0.3%(wt/v) in the final mixture, results best in the stabilization of whipped egg whites (as no liquid accumulation was observed) as compared to the control, showing better performance than the other materials tested.

Example 3 - Mixing bacterial cellulose with a third component

[0094] In an embodiment, the BC obtained by any of the three different comminution methods described in Example 1 was mixed with either sodium carboxymethyl cellulose, CMC (90 KDa, or 250 KDa or 700 KDa, Sigma), xanthan (Sigma), methylcellulose, MC (Sigma), hydroxyethyl-cellulose, HEC (Sigma), hydroxypropyl methyl cellulose (Sigma), HPMC (Sigma), tylose (Sigma), glycerol (Sigma) and saccharose (Sigma); BC was also mixed with combinations of CMC:Xanthan, CMC:HEC and CMC:HPMC, as described in the following table:

[0095] Table 1 - Combination of bacterial cellulose and third components

Mixture	Mass ratio		
	1:1		
BC:CMC	1:0.75		
	1:0.25		
BC:Xanthan			
BC:MC			
BC:HEC	1:1		
BC:HPMC	1.1		
BC:Tylose			
BC:Saccharose			
P.C.Chucorol	1:1.6		
BC:Glycerol	1:1.3		
BC:CMC:Xanthan	1:0.25:0.75		
CB:CMCHEC	1:0.5:0.5		
CB:CMC:HPMC	1:0.75:0.25		

[0096] In all of these cases, the components to be added to BC were first dissolved in water. Those of ordinary skill in the art can appropriately select the addition amount of the third components, as a

redispersing aid, according to the type of substance and the like, usually within 2 to 1,000%(wt/v) by mass of BC.

[0097] To evaluate the dispersibility of the BC mixtures, samples were prepared at 0.5% (wt/v) in water to at room temperature. For this, two dispersion method were used:

- I) a high shear dispersion mixing, using an ultraturrax CAT Unidrive 1000D, at 15000 Rpm using a dispersing shaft CAT 20F, also for 5 min and
- II) a low mechanical shear dispersion using magnetic stirrer plate (Stuart SD162), at 900 Rpm, for 5 min

[0098] As a control, BC, as obtained by any of methods A, B and C mentioned above (Example 1), was used at the same final solids concentration. The dispersed materials were spread over a petri dish and observed at naked eye. Also, fluorescent microscopy was used to better visualize the BC fibres. As an example, the following table, Figure 7 and Figure 8 summarize the main observations pertaining to the BC and CMC mixtures.

[0099] Table 2 - Dispersibility of never-dried mixtures of BC and third components.

Component		Dispersion	Dispersibility Classification*	
BC:CMC 90 KD	oa (1:1):			
A) Sammic blender B) Comitrol Processor		I) UltraTurrax II) Magnetic	1 2	
		stirrer	Z	
НРН	C) 1, 2, 6 Pass	I) UltraTurrax II) Magnetic stirrer	1 2	
BC:CMC HPMC A) Sammic BC:CMC:HEC A) Sammic	1:0.5:0.5 1:0.75:0.25	I) UltraTurrax	2	
BC:Xanthan B) Comitrol	1:1			

^{*} According to embodiments of paragraphs [0027] and [0066]

[00100] While samples dispersed using an UT showed complete dispersibility (minimum amount of fibre bundles were observed), samples dispersed using a magnetic stirrer still showed fibre bundles and agglomerates (Figure 8 and Table 2). However, the presence of the third component improves the dispersion of the aggregates, more homogeneous samples being observed (Figure 8 vs Figure 2). Also, further homogenization with ultraturrax for up to 60 min, does not change the morphology nor the rheological profile of the BC:CMC mixtures (data not shown).

[00101] It was observed that when CMC or xanthan were added to BC, at preferably a mass ratio of 1:1, a dispersible and stable suspension was obtained, following high shear treatment with ultraturrax.

Regarding the control, BC alone, it could not be dispersed by any of the dispersion methods.

[00102] From these results, the water-soluble anionic polyelectrolyte CMC plays a determinant role in ensuring the dispersibility of the BC fibres and allowing their stabilization in aqueous media. The negative charge of CMC may contribute to the improved dispersion of the BC fibres due to steric hindrance. Also, CMC has a strong affinity for water molecules. Non-adsorbed CMC may also prevent the agglomeration of BC fibres due to the creation of a hydration shell in aqueous media, around CMC and BC thus also contributing to the improved dispersibility and stability in aqueous media.

[00103] The BC:CMC samples were further characterized by rheological assays (Figure 9). For this, stress-strain curves were obtained using a TA instruments rheometer, model 5332-1179 and a disk geometry. Shear rate versus viscosity graphs were drawn in semi-log scale to better visualise the different rheological profiles at low shear rates. From these assays, it may be concluded that the use of higher shear rate stress during wet comminution decreases the dynamic viscosity of the samples (from A to E, Figure 9). As previously mentioned, this effect is caused by some fragmentation of the fibres with increasing shear stress. Also, to the exception of samples ground with B) Comitrol, which show a coincidental rheological profile, all other samples showed higher rheological profile after dispersion with I) ultraturrax, which suggests that the high shear rates used by ultraturrax, exerted a better dispersion of the BC fibres, without compromising its viscosity – actually improving it.

Example 4 - Drying of bacterial cellulose mixtures

[00104] In an embodiment, the BC was wet ground by three different methods, as described in Example 1 (methods A, B and C). The obtained BC was mixed with a third component, as described in Example 3. The obtained BC mixtures can be dried by means of any of the available drying processes and equipment, which include spray drying, drum drying, oven drying, vacuum drying, tunnel drying, infra-red drying, freeze drying. Those skilled in the art may select any other proper drying process and/or combination of methods. In the present disclosure, BC mixtures were dried by four methods:

- i) Drying on a hot plate, a fast drying process (within minutes), whereby a thin layer of BC samples was spread over a hot plate (130 °C) of an Ariette Crepes Maker, model 183 and dried; the "dry" state is an absolute dried BC product (100% TS (total solids)), in which there is no residual water in the final product.
- ii) Drying in an oven, a slower drying process, in the range of 4-6hr. For this, samples were placed in aluminium crucibles and dried at 80 °C. To evaluate the effect of residual moisture on the dispersibility

of BC formulations wet ground with Comitrol Processor, drying was done to a final solid content of a) 80% (m/m) (80% TS) and b) 100% (m/m) (100% TS).

iii) Drum drying, also a fast drying process, was also used. For this, samples were fed to a lab-scale chrome plated cast iron double drum dryer, from Tummer Simon Dryers (drum size 0.3m x 0.3m giving a drying surface area of 0.28 m² per roll (0.56 m² total). Feed was hand fed ('jugged') into the nip at room temperature, at a rate was 11.9Kg/hr. The nip gap was set at 0.1 mm. Drying was done at 2-4 bar steam pressure at a roll speed of 2.5 Rpm. Thin product sheets were obtained at a rate of 0.86 Kg/hr at around 20% residual moisture.

iv) Samples were also dried by spray drying. For this, a sample at 1% (m/v) was fed to a MOBILE MINOR™ R&D Spray Dryer (GEA Group AG) at a flow rate of 240 Kg/h. The inlet and outlet temperatures were set to 220 and 115°C, respectively and a pressure gas nozzle of 3.5 bar was used. In this case, samples were dried to 100%TS.

[00105] All dried material was ground (with a High Power Herb Grain Grinder Cereal Mill Powder Grinding Machine Flour 600G) and sieved (Mat.Mesh:AISI 316 nº 1/2871/1 Nº: 1/2871/1 with an opening of 0.300 mm) to a final particle size preferably in the range of <300 μ m. As in the previous examples, the dried powders were dispersed at a final solids concentration of 0.5% in water, using:

- I) a high shear dispersion mixing, using an ultraturrax CAT Unidrive 1000D, at 15000 Rpm using a dispersing shaft CAT 20F, for 5 min and
- II) a low mechanical shear dispersion using magnetic stirrer plate (Stuart SD162), at 900 Rom, also for 5 min.

[00106] Rheological assays were done using a TA instruments rheometer, model 5332-1179 and a disk geometry. Shear rate versus viscosity graphs were drawn in semi-log scale to better visualise the different rheological profiles at low shear rates.

[00107] The following table summarizes the main observations regarding the aqueous dispersibility of the dried and ground BC:CMC 90 KDa mixture.

[00108] Table 3 - Dispersibility of BC:CMC mixtures after drying and grinding to a particle size of <300 μm .

Homogenization	Drying	Dispersion	Dispersibility Classification*
A) Sammic blender	i) Hot plate	- I) UltraTurrax	
	ii) Oven dried	,	1
B)Comitrol	i) Hot plate	II) Magnetic	2
Processor	ii) Oven 80% TS	- stirrer	2
	dried 100% TS		

HPH		_		
	1 2 and 6		I) UltraTurrax	1
	1, 2 and 6 Pass i) Hot plate	II) Magnetic	2	
	. 200		stirrer	

^{*} According to embodiments of paragraphs [0027] and [0066]

[00109] As was observed for Figure 8, water dispersion of dried and ground samples under low shear (magnetic stirring) showed a few fibre blundes, whereas with a high shear rate (ultraturrax), the BC was completely dispersed. The highest viscosity profile of BC:CMC was obtained from BC ground with a A) Sammic blender and dried on a hot plate to 100%TS (Figure 10). The rheological profile was actually higher (higher dynamic viscosity) than the one of non-dried sample. As for the remaining samples, all of the ones dispersed with low shear stress (II - magnetic stirrer) showed a similar rheological profile to those of the never-dried ones. These results suggest that the use of low shear stress dispersion mixing, at room temperature, under 5 minutes is enough to allow for a recovery or even slight increase in the rheological profile of BC:CMC dried formulation. However, the presence of small aggregates of BC in the suspension may contribute to the increase in the apparent viscosity profile. Contrarily to the never-dried samples, dispersion of the dried ones with ultraturrax reduced the rheological profile (as compared to the samples dispersed in the magnetic stirrer), possibly due the reduction of some fibre bundles and agglomerates. Further homogenization for up to 60 min with ultraturrax does not change the rheologic profile of the samples (data not show).

[00110] Contrarily to what has been proposed in other documents, when a dispersing aid is added to BC, such as CMC, drying can be done to the extent of the full removal of water molecules, as dried samples with 80 and 100%TS, showed an identical rheological profile (Figure 10B). A similar observation was recorded for samples dried using a drum dryer (80 and 100%TS) and spray dryer (100%TS).

[00111] Too extensive wet comminution of the BC suspension such as with HPH (Figure 10 C, D, E) strongly affected the rheological properties of the dried material, confirming the effect previously observed on the never dried samples.

[00112] A dry control without the addition of CMC was also prepared. Results showed that BC alone is not dispersible, regardless of the homogenization, drying and dispersion method used.

Example 5 - Effect of the particle size on the dispersibility and rheological profile of bacterial cellulose mixtures

[00113] In the previous example, the dried BC formulations were ground to a particle size <300 μ m. It is important, however, to better understand the effect of the particle size in the restauration of the properties of BC mixtures. For this, the BC mixtures obtained in the previous example were further

ground and sieved to a final particle size <200 μ m (Endecotts, Ltd, aperture 212 μ m) and <100 μ m (Endecotts, Ltd, aperture 106 μ m). Also, samples were fractioned into the following size ranges: 100<x<300 μ m, 300<x<500 μ m. As in the previous examples, the dried powders were dispersed to a final solids' concentration of 0.5% in water, evaluated by spreading over a petri dish, fluorescent microscopy and through rheological profile.

[00114] As an example, table 4 and Figure 11 summarize the main observations pertaining to the dispersibility and rheological behaviour of BC:CMC 90 KDa. To simplify the demonstration of the results, only those obtained using hot plate drying are shown. The same profile was observed for samples dried in an oven.

[00115] Table 4 - Dispersibility of BC:CMC mixtures after drying (in a hot plate) and grinding.

Homogenizatio n	Particle size	Dispersion	Dispersibility (Classification)*
A) Sammic blender	< 100 μm		
B)Comitrol Processor	< 100 μm	-	
	< 200 μm	- I) UltraTurrax	1
НРН		 II) Magnetic stirrer 	
C) 1 Pass	-		
D) 2 Pass	< 100 μm		
E) 6 Pass			
A) Sammic	100 <x<300 td="" μm<=""><td>II) Magnetic</td><td>2</td></x<300>	II) Magnetic	2
blender	300 <x<500 td="" μm<=""><td>stirrer**</td><td>3</td></x<500>	stirrer**	3

^{*} According to embodiments of paragraphs [0027] and [0066]

[00116] Results show that, decreasing the particle size of the BC:CMC allows for a faster dispersion of BC mixtures (under 1 min in the case of particles ground to < 100 μ m) at low shear rates (magnetic stirring). Increasing the particle size (from e.g. <100 μ m, to 300<x<500 μ m), decreases the capability of fully dispersion under low shear mixing conditions, even after mixing for 30 min. However, with the decrease in particle size, a decrease in the viscosity profile was also observed (Figure 11). In addition, as examples below will show, this impacts the technological performance of BC mixtures.

^{**} Stirring for 30 min

Example 6 -Effect of particle size of dried BC formulations on the Suspension Stability of Cocoa Beverage

[00117] In an embodiment, the suspension stability of water-insoluble solid particles such as cocoa, powdered green tea, and calcium carbonate are important aspects for the development of certain commercial beverages. In the case of chocolate milk, cocoa particles tend to precipitate soon after the initial mixing.

[00118] In this example, the effect of particle size of dried formulations of BC:CMC 90 KDa, obtained as described in Example 5, on the stabilization of cocoa particles suspensions in a chocolate milk beverage was evaluated. For this, BC:CMC at different particle sizes and final concentrations in the range of 0.075-0.15% and pure cocoa (1.2%) were weighted. Medium-skimmed milk, 15 mL, was added to each BC:CMC concentration. The mixture was stirred in a vortex (2,800 rpm) for 3 min at room temperature and then pasteurized at 75 °C for 15 seconds. Samples were stored at room temperature and the sedimentation of cocoa was evaluated. Further, the effect of ultraturrax on the functional properties of the BC:CMC samples was assessed. For this, BC samples were homogenized with ultraturrax (CAT Unidrive 1000D, at 15000 Rpm using a dispersing shaft CAT 20F) for 4 and 30 min before being added to the milk. In parallel, the same test was done using xanthan, carboxymethyl cellulose, colloidal plant celluloses: Avicel (RT 1133, LM 310 and CM 2159, FMC Biopolymers), Novagel (RCN-10 and RCN-15, FMC Biopolymers), Bioplus Fibrils (a microfibrillated cellulose, not used in food applications, American Process Inc.) and plant nanocelluloses: CelluForce NCC, Arbocel B 600, Arbocel BE 600/30 (from Celluforce Inc.). All Avicel and Novagel celluloses were previously activated, for 30 min, at 23,800 Rpm, according to the specifications sheets. A control assay, where no stabilizers were added, was also made. Also controls with never-dried BC (ground using methods A, B, C and D described in Example 1) to which CMC was added to, were done. In this case, CMC was mixed with BC using I) a low shear dispersion mixing, as described in Example 3. The stabilization of the chocolate drinks was assessed by calculating the percentage of sedimentation, according to the following equation:

Stability (%) = (Milk with chocolate
$$mL \times 100$$
)/(total milk volume) (1)

[00119] The higher the stability (%), the more stabilized the suspension is.

[00120] The results from this study are displayed in the following table and Figure 12, using 0.15% BC:CMC (other concentrations were tested with similar observations).

[00121] Table 5 - Stabilization of cocoa particles in chocolate milk, with BC:CMC (redispersed under low shear rate unless otherwise specified), added at 0.15%, at room temperature.

Homogenization	Drying		Particle size	Stability/%		%
			μm	1h	24h	4days
A) Sammic blender	i) Hot plate		<100	100	100	83
			<300	100	100	100
	Never dried			100	100	100
	Never dried		4 min	100	100	100
	(UT)		30 min	100	100	100
B) Comitrol Processor	i) Hot plate		<100	100	100	100
			<300	100	100	100
	Never dried			100	100	100
HPH						
C) 1 Pass	i) Hot plate		<100	100	67	50
			<300	100	100	100
	Never dried			100	100	100
D) 2 Pass						
	i) Hot plate		<100	100	67	50
			<300	100	100	83
	Never dried			100	100	83
E) 6 Pass						
	i) Hot plate		<100	100	27	33
			<300	100	100	50
	Never dried			87	83	67
E) 6 Pass,	i) Hot plate					
<100 μm	(UT)	4 min		100	67	50
		30 min		30	30	27
Avicel CM 2159				95	0	0
Novagel RC-15				96	0	0
Novagel RC-10				96	0	0
Bioplus Fibrils	0	Γ30 min	-	70	40	35
CelluForce NCC	<u> </u>		-	100	100	100
Arbocel B600	_		-	30	30	27
Arbocel BE600/30				30	30	27

[00122] These results show that an extensive homogenization through High Pressure Homogenization and extensive grinding of dried material to a particle size $< 100 \mu m$, reduces the stabilizing effect of

BC:CMC. Treatment with UT of never dried BC:CMC processed with the Sammic blender, does not change its stabilizing effect on cocoa particles. However, under extreme processing conditions such as HPH 6 passages, drying and comminution to <100 μ m, UT treatment affects the cocoa stability achieved with the BC:CMC.

[00123] To the exception of Celluforce, all colloidal plant celluloses and nanocelluloses showed a very low stabilizing effect on cocoa particles.

[00124] Where singular forms of elements or features are used in the specification of the claims, the plural form is also included, and vice versa, if not specifically excluded. For example, the term "a polysaccharide" or "the polysaccharide" also includes the plural forms "polysaccharides" or "the polysaccharides," and vice versa. In the claims articles such as "a," "an," and "the" may mean one or more than one unless indicated to the contrary or otherwise evident from the context. Claims or descriptions that include "or" between one or more members of a group are considered satisfied if one, more than one, or all of the group members are present in, employed in, or otherwise relevant to a given product or process unless indicated to the contrary or otherwise evident from the context. The disclosure includes embodiments in which exactly one member of the group is present in, employed in, or otherwise relevant to a given product or process. The disclosure also includes embodiments in which more than one, or all of the group members are present in, employed in, or otherwise relevant to a given product or process.

[00125] Furthermore, it is to be understood that the disclosure encompasses all variations, combinations, and permutations in which one or more limitations, elements, clauses, descriptive terms, etc., from one or more of the claims or from relevant portions of the description is introduced into another claim. For example, any claim that is dependent on another claim can be modified to include one or more limitations found in any other claim that is dependent on the same base claim.

[00126] Furthermore, where the claims recite a composition, it is to be understood that methods of using the composition for any of the purposes disclosed herein are included, and methods of making the composition according to any of the methods of making disclosed herein or other methods known in the art are included, unless otherwise indicated or unless it would be evident to one of ordinary skill in the art that a contradiction or inconsistency would arise.

[00127] The disclosure should not be seen in any way restricted to the embodiments described and a person with ordinary skill in the art will foresee many possibilities to modifications thereof.

[00128] The above described embodiments are combinable.

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CLAIMS

1. A powdered formulation, comprising

bacterial cellulose;

and an additional component selected from the following list: sodium carboxymethyl cellulose, carboxymethyl cellulose, xanthan, methylcellulose, methyl cellulose, hydroxyethyl-cellulose, hydroxyethyl-cellulose, hydroxypropyl methyl cellulose, hydroxypropyl methylcellulose, tylose, glycerol, saccharose, or mixture thereof; wherein the powdered formulation is dispersible in an aqueous media, at 20 °C, with low shear mixing.

- 2. The powdered formulation according to the previous claim, wherein the low shear mixing is at most 1500 rpm, preferably at most 1000 rpm.
- 3. The powdered formulation according to any of the previous claims, wherein the powdered bacterial cellulose is dispersed in an aqueous media in at most 10 min; preferably in at most 6 min; more preferably in at most 4 min.
- 4. The powdered formulation according to any of the previous claims, wherein the powdered bacterial cellulose is dispersed in an aqueous media in at most 2 min; preferably in at most 1 min.
- 5. The powdered formulation according to any of the previous claims, wherein the particles comprises a size between 5-500 μ m.
- 6. The powdered formulation according to any of the previous claims, wherein the particles comprises a size between 80-400 μ m, more preferably 100-300 μ m.
- 7. The powdered formulation according to any of the previous claims 5-6, wherein the D50 of the particle is 50-300 μ m, preferably the D50 of the particle is 80-250 μ m, more preferably the D50 of the particle is 100-200 μ m.
- 8. The powdered formulation according to the previous claim 5, wherein the D50 of the particle is 10- $90 \, \mu m$, preferably the D50 of the particle is 20-80 μm , more preferably the D50 of the particle is 30- $60 \, \mu m$.

9. The powdered formulation according to the previous claim 5, wherein the bacterial cellulose is dispersed in an aqueous media in at most 2 min and wherein the particles comprise a size between 5-100 μ m; more preferably 10-60 μ m.

- 10. The powdered formulation according to the previous claims, wherein the mass ratio between bacterial cellulose and the additional component varies between 1:5 to 1:0.2, preferably 1:2 to 1:0.5.
- 11. The powdered formulation according to any of the previous claims, wherein the additional component is carboxymethyl cellulose, carboxymethyl cellulose and xanthan; carboxymethyl cellulose and hydroxyethyl-cellulose and carboxymethyl cellulose and hydroxypropyl methylcellulose, or mixture thereof.
- 12. The powdered formulation according to any of the previous claims, wherein the powdered bacterial cellulose is obtained from a bacterium of the following list: Acetobacter, Agrobacterium, Gluconacetobacter, Achromobacter, Alcaligenes, Aerobacter, Azotobacter, Rhizobium, Salmonella, Escherichia, Sarcina, Komagataeibacter, or combinations thereof, preferably the powdered bacterial cellulose is obtained from a bacterium of the following list: Acetobacter, Komagataeibacter, Gluconacetobacter or combinations thereof.
- 13. A wet bacterial cellulose formulation comprising

5-70 % (wt./v) of bacterial cellulose, wherein the bacterial cellulose is dispersed in bundles; wherein the bacterial cellulose bundles size varies between 20 μm to 2 mm.

- 14. The wet bacterial cellulose of the previous claim wherein the bacterial cellulose bundle size varies between 20 μ m to 2 mm.
- 15. The wet bacterial cellulose of the previous claim wherein the bacterial cellulose bundle size varies between 200 μm to 1 mm.
- 16. The wet bacterial cellulose of claim 16 wherein the bacterial cellulose bundle size varies between 20 $400~\mu m$, preferably 30 - $200~\mu m$.

17. The wet bacterial cellulose in any of the previous claims 14-17 comprising 5-40 % (wt./v) of bacterial cellulose solids.

- 18. The wet bacterial cellulose of the previous claim comprising 10–30 % (wt./v) of bacterial cellulose solids.
- 19. The wet bacterial cellulose in any of the previous claims 14-19, wherein the bacterial cellulose is obtained from a bacterium of the following list: *Acetobacter*, *Agrobacterium*, *Gluconacetobacter*, *Achromobacter*, *Alcaligenes*, *Aerobacter*, *Azotobacter*, *Rhizobium*, *Salmonella*, *Escherichia*, *Sarcina*, *Komagataeibacter*, or combinations thereof, preferably the powdered bacterial cellulose is obtained from a bacterium of the following list: *Acetobacter*, *Komagataeibacter*, *Gluconacetobacter* or combinations thereof.
- 20. Use of the powdered bacterial cellulose formulation described in any of the previous claims as a colloid stabilizer, foam stabilizer or as a thickener.
- 21. Use of the powdered bacterial cellulose formulation or wet bacterial cellulose described in any of the previous claims as a reinforcer material.
- 22. A pharmaceutical or a cosmetic composition comprising at least an active ingredient and the powdered bacterial cellulose, or the wet bacterial cellulose described according to any of the previous claims as an excipient.
- 23. A dietary fiber or a foodstuff comprising an effective amount of powdered bacterial cellulose or the wet bacterial cellulose described according to any of the previous claims.
- 24. A composite comprising an effective amount of the bacterial cellulose powdered formulation or the wet bacterial cellulose as described according to any of the previous claims.
- 25. A method for producing a bacterial cellulose powdered formulation described in any of the previous claims 1-13 comprising the following steps of:

providing an aqueous suspension of ground bacterial cellulose bundles, with a size between 20 μm to 2 mm, preferably 100 μm to 1.5 mm, more preferably 200 μm to 1 mm;

adding a third component at a temperature between 20-80°C;

drying the bacterial cellulose until obtaining at least 60% of solids; grinding the bacterial cellulose until obtain a particle size between 5-500 μm .

- 26. The method according to any of the previous claim, wherein the concentration of bacterial cellulose in the ground bacterial cellulose suspension varies between 0.3-10 % (wt./v).
- 27. The method according to the previous claims 26-27, wherein the concentration of bacterial cellulose in the ground bacterial cellulose suspension varies between 0.5-5 % (wt./v).
- 28. A method for producing a wet bacterial cellulose formulation comprising the following steps of: providing a ground bacterial cellulose suspension, in water, with a size between 20 μ m to 2 mm, preferably 100 μ m to 1.5 mm, more preferably 200 μ m to 1 mm; concentration of the ground bacterial cellulose suspension in water up to a concentration of 10-70% (wt./v), preferably between 15-30% (wt./v).
- 29. The method according to the previous claim, wherein the step of homogenizing the bacterial cellulose suspension comprises wet comminution.

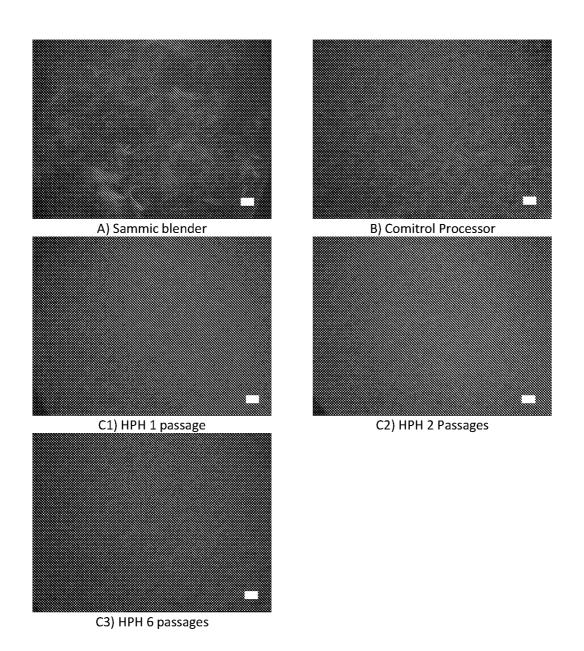
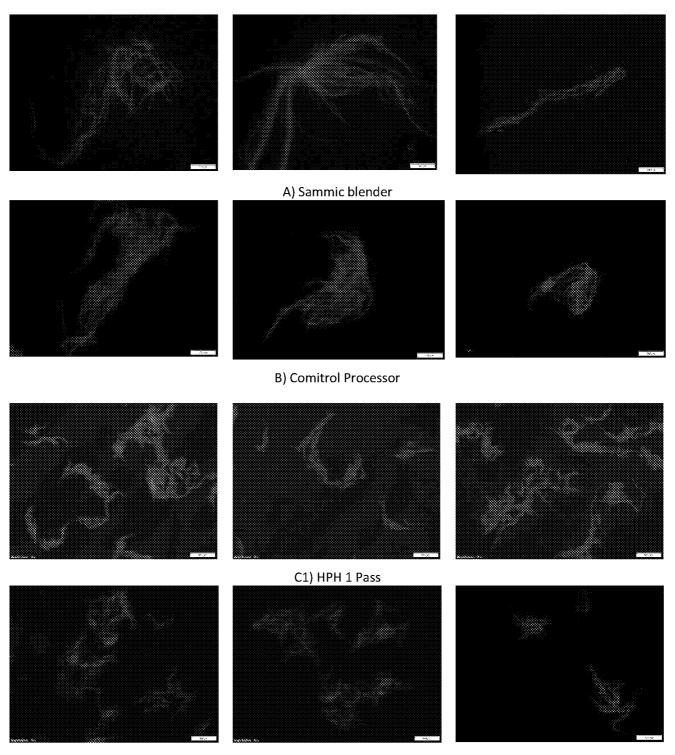
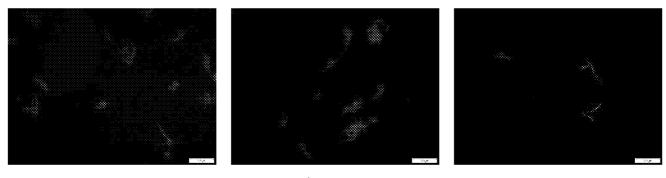


Fig. 1



C2) HPH 2 Pass Fig. 2 - A



C3) HPH 6 Pass

Fig. 2 - B

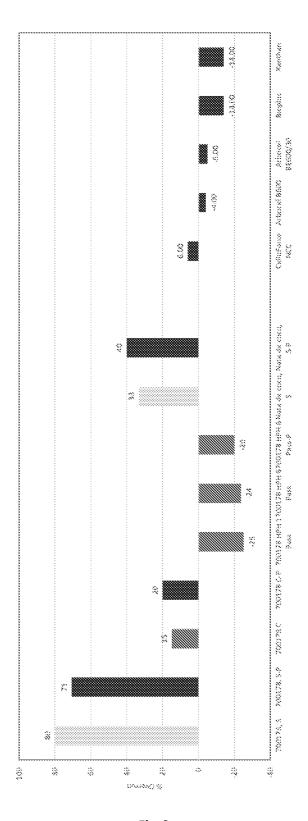


Fig. 3

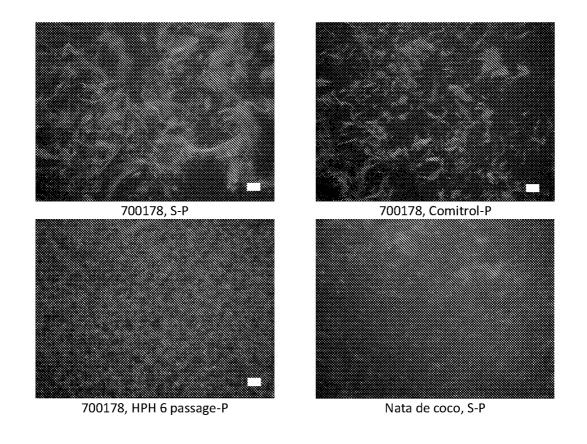
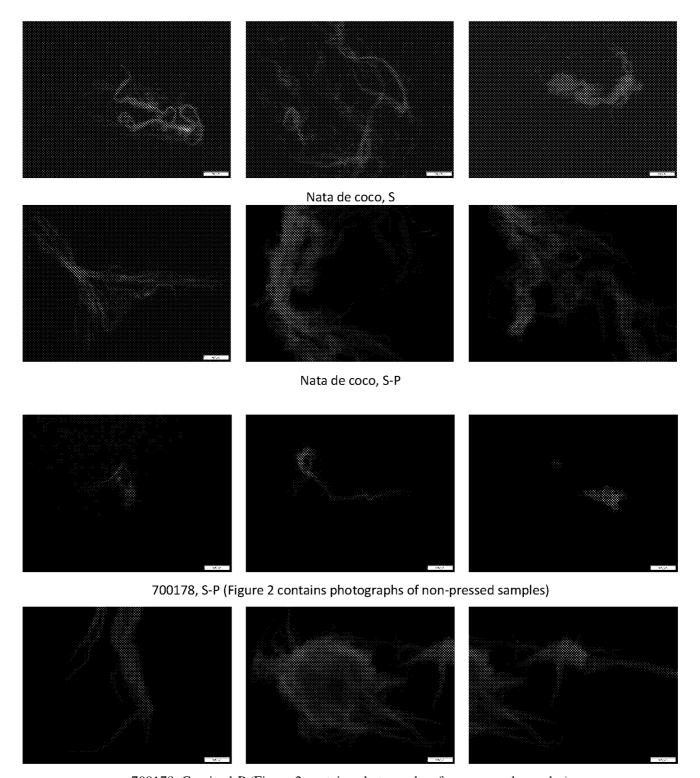
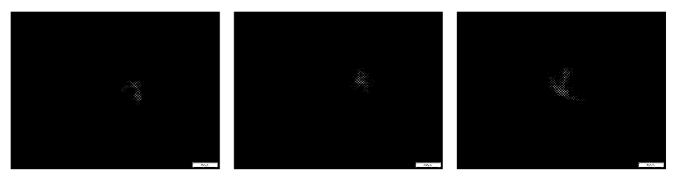


Fig. 4



700178, Comitrol-P (Figure 2 contains photographs of non-pressed samples)

Fig 5 - A



700178, HPH 6 Pass-P (Error! Reference source not found. 2 contains photographs of non-pressed samples)

Fig. 5 - B

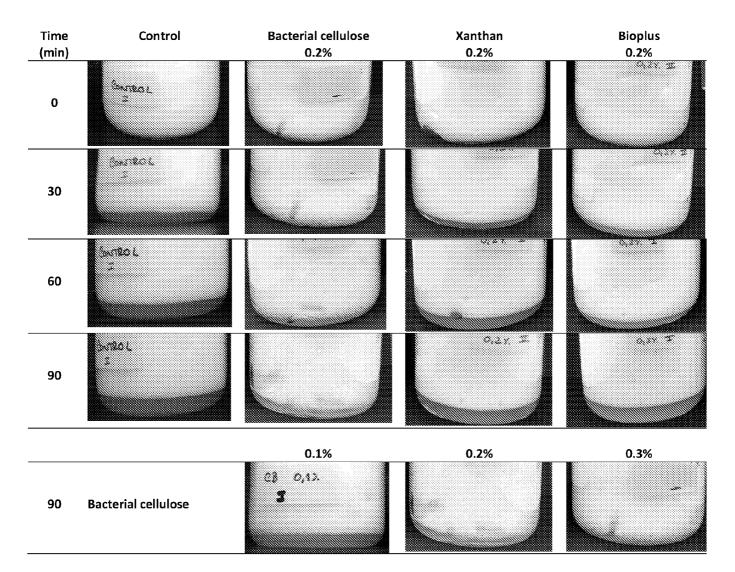
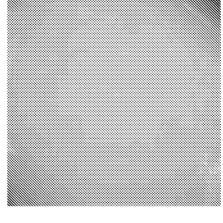
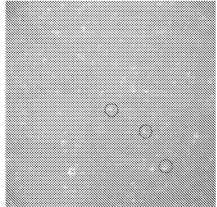


Fig. 6



1



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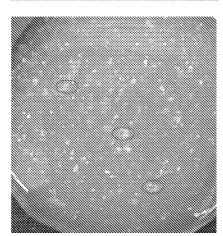


Fig. 7

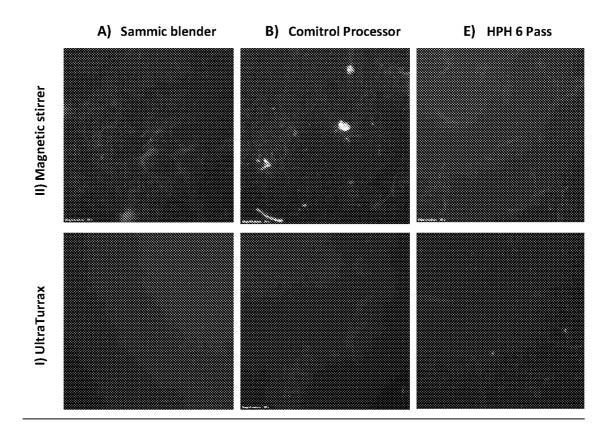
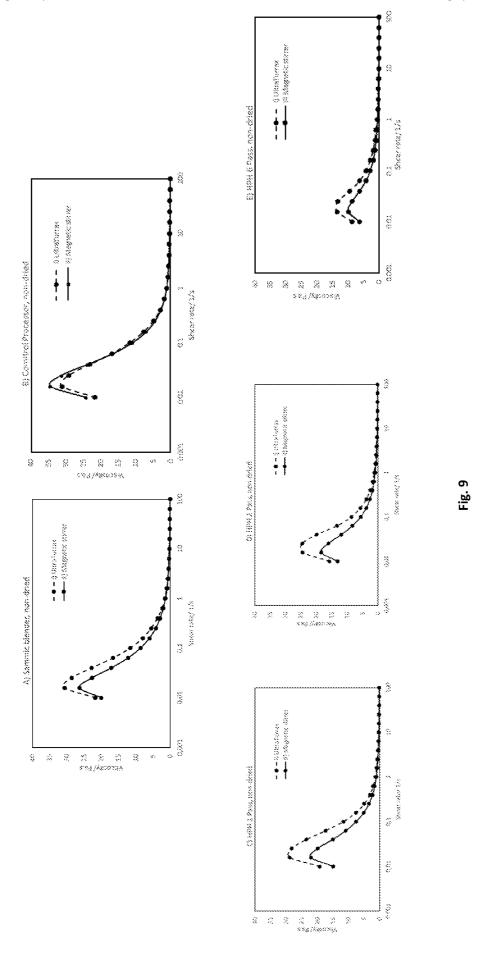
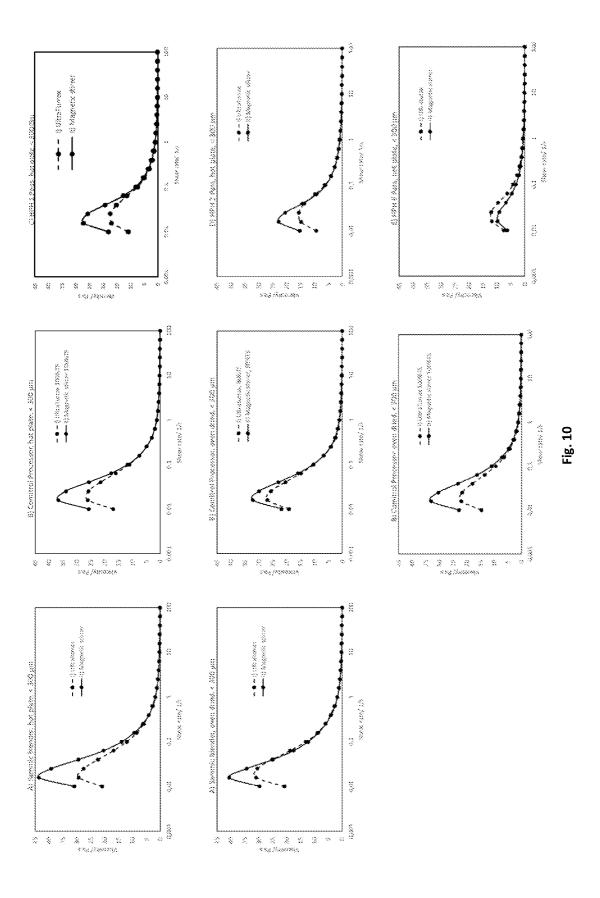


Fig. 8





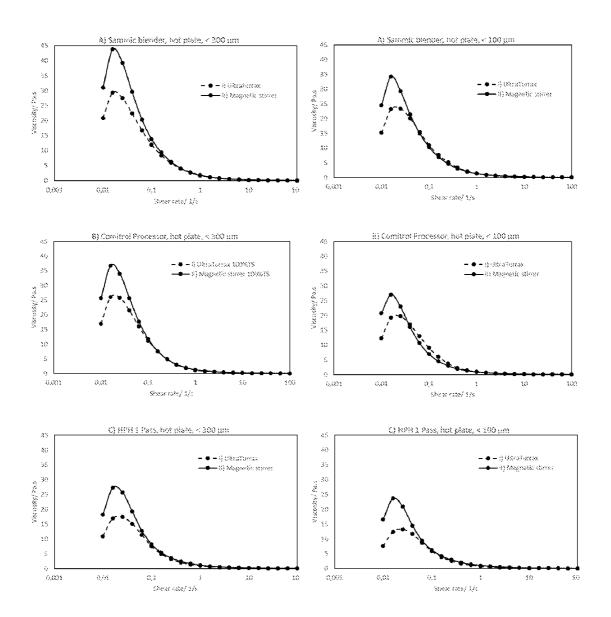
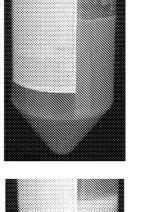
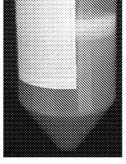


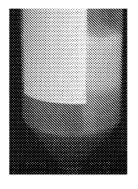
Fig. 11



100



83



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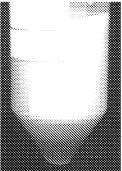


Fig. 12

INTERNATIONAL SEARCH REPORT

International application No PCT/IB2019/061452

A. CLASSIFICATION OF SUBJECT MATTER INV. C08L1/02

ADD.

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols) C08L

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

EPO-Internal, BIOSIS, EMBASE, WPI Data

C. DOCUM	NTS CONSIDERED TO BE RELEVANT	
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 2018/038055 A1 (KUSANO SAKKO INC [JP]) 1 March 2018 (2018-03-01)	1-4, 12-16, 19,22-24
Υ	paragraphs [0007], [0036]; claims 1-10	1-29
Y	US 8 053 216 B2 (CP KELCO US INC [US]) 8 November 2011 (2011-11-08) cited in the application column 6, paragraph 3/	1-29

Further documents are listed in the continuation of Box C.	X See patent family annex.
* Special categories of cited documents: "A" document defining the general state of the art which is not considered to be of particular relevance "E" earlier application or patent but published on or after the international filing date "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) "O" document referring to an oral disclosure, use, exhibition or other means "P" document published prior to the international filing date but later than the priority date claimed	"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art "&" document member of the same patent family
Date of the actual completion of the international search	Date of mailing of the international search report
5 May 2020	26/05/2020
Name and mailing address of the ISA/ European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Authorized officer Friedrich, Christof

INTERNATIONAL SEARCH REPORT

International application No
PCT/IB2019/061452

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Υ	MAZHARI MOUSAVI S M ET AL: "Cellulose nanofiber/carboxymethyl cellulose blends as an efficient coating to improve the structure and barrier properties of paperboard", CELLULOSE, SPRINGER NETHERLANDS, NETHERLANDS, vol. 24, no. 7, 27 April 2017 (2017-04-27), pages 3001-3014, XP036249461, ISSN: 0969-0239, DOI: 10.1007/S10570-017-1299-5 [retrieved on 2017-04-27] page 3003, column 1, paragraph 1	1-29
Y	NADERI ET AL: "Can redispersible low-charged nanofibrillated cellulose be produced by the addition of carboxymethyl cellulose?", NORDIC PULP AND PAPER RESEARCH JOURNAL, vol. 30, no. 04, 1 December 2015 (2015-12-01), pages 568-577, XP055456727, SE ISSN: 0283-2631, DOI: 10.3183/NPPRJ-2015-30-04-p568-577 the whole document	1-29
Y	NÚRIA BUTCHOSA ET AL: "Water redispersible cellulose nanofibrils adsorbed with carboxymethyl cellulose", CELLULOSE, vol. 21, no. 6, 25 September 2014 (2014-09-25), pages 4349-4358, XP055463678, Netherlands ISSN: 0969-0239, DOI: 10.1007/s10570-014-0452-7 the whole document	1-29
X,P	MARTINS D ET AL: "A dry and fully dispersible bacterial cellulose formulation as a stabilizer for oil-in-water emulsions", CARBOHYDRATE POLYMERS, APPLIED SCIENCE PUBLISHERS, LTD. BARKING, GB, vol. 230, 22 November 2019 (2019-11-22), XP085972479, ISSN: 0144-8617, DOI: 10.1016/J.CARBPOL.2019.115657 [retrieved on 2019-11-22] the whole document	1-29

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/IB2019/061452

Patent document cited in search report		Publication date	Patent family member(s)		Publication date
WO 2018038055	A1	01-03-2018	JP W0201803805 W0 201803805		20-06-2019 01-03-2018
US 8053216	В2	08-11-2011	CN 101203619 US 200719777 ZA 20071098	9 A1	18-06-2008 23-08-2007 26-11-2008