(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
30 April 2015 (30.04.2015)

WIPO | PCT

(10) International Publication Number
WO 2015/061612 A1

(51) International Patent Classification:
A61L 26/00 (2006.01)

(21) International Application Number:
PCT/US2014/062041

(22) International Filing Date:
23 October 2014 (23.10.2014)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
14/061,993 24 October 2013 (24.10.2013) US
14/319,901 30 June 2014 (30.06.2014) US

(71) Applicant: **MEDTRONIC XOMED, INC.** [US/US]; 6743 Southpoint Drive North, Jacksonville, FL 32216-0980 (US).

(72) Inventors: **MEDINA, Jennifer, Gates**; 6743 Southpoint Drive North, Jacksonville, FL 32216-0980 (US). **SHERRMAN, Ethan, Glenn**; 6743 Southpoint Drive North, Jacksonville, FL 32216-0980 (US).

(74) Agent: **CLEVELAND, David, R.**; IPLM GROUP, P.A., Post Office Box 18455, Minneapolis, MN 55418 (US).

(81) Designated States *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IR, IS, JP, KE, KG, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) Designated States *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, ST, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, KM, ML, MR, NE, SN, TD, TG).

Declarations under Rule 4.17:

— *as to applicant's entitlement to apply for and be granted a patent (Rule 4.17(ii))*

— *as to the applicant's entitlement to claim the priority of the earlier application (Rule 4.17(iii))*

— *of inventorship (Rule 4.17(iv))*

Published:

— *with international search report (Art. 21(3))*

(54) Title: CHITOSAN PASTE WOUND DRESSING

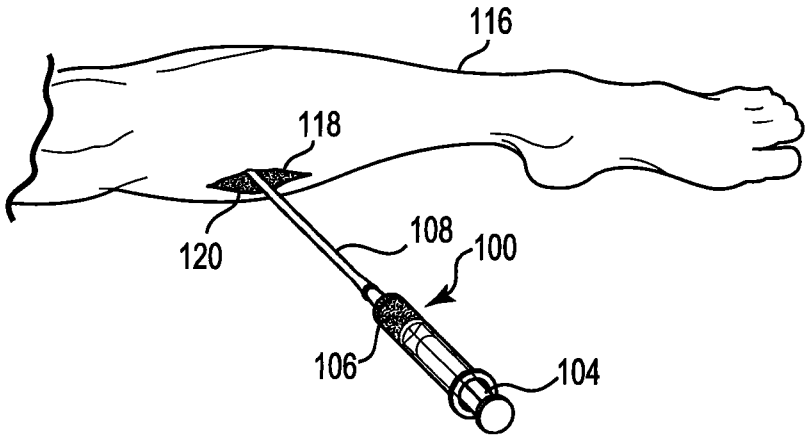

Fig. 1

(57) Abstract: Wounds are treated with a ready-to-use composition having a high concentration of a water-soluble chitosan in a phosphate-containing solution. The composition is a paste at room temperature, has a pH of at least 4, adheres to body tissue or a surgical site and has a residence time of at least 1 day.

WO 2015/061612 A1

CHITOSAN PASTE WOUND DRESSING

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority from U.S. Patent Application Serial No. 14/061,993, filed October 24, 2013 and entitled CHITOSAN STENTING PASTE and U.S. Patent Application Serial No. 14/319,901, filed June 30, 2014 and entitled CHITOSAN PASTE WOUND DRESSING, and is related to International Application No. (Attorney Docket No. C04881WO01), filed even date herewith and entitled CHITOSAN STENTING PASTE, the disclosures of each of which are incorporated herein by reference.

FIELD OF THE INVENTION

[0002] This invention relates to polysaccharide-based wound dressings.

BACKGROUND

[0003] A wound is an injury to the skin and may for example be a simple abrasion, burn, cut or a purposeful incision such as a surgical wound. Local wound treatments such as wound dressings may be applied to the wound to provide a barrier to micro-organisms and protect the wound from the external environment. Some wound dressings also support or promote wound healing mechanisms.

SUMMARY OF THE INVENTION

[0004] Wound dressings that include biodegradable materials are desirable because they may reduce the trauma associated with removal of the wound dressing from a wound surface. Desirably, the wound dressing also has positive therapeutic effects on wound healing.

[0005] The invention provides, in one aspect, a method for treating a wound comprising: applying to a wound a paste composition comprising a water-soluble chitosan or derivative thereof dissolved in a phosphate-containing solution, wherein the composition is a paste at room temperature and has a pH of at least 4, a viscosity of about 1 to about 15 Pa.s., and a residence time of at least 1 day.

[0006] The invention provides, in another aspect, use of a paste composition to treat a wound, the composition comprising a water-soluble chitosan or derivative thereof dissolved in a phosphate-containing solution, wherein the composition is a paste at room temperature and has a pH of at least 4, a viscosity of about 1 to about 15 Pa.s., and a residence time of at least 1 day.

[0007] The invention provides, in another aspect, a kit for treating a wound, the kit comprising sterile packaging containing a paste composition comprising a water-soluble chitosan or derivative thereof dissolved in a phosphate-containing solution, wherein the composition is a paste at room temperature and has a pH of at least 4, a viscosity of about 1 to about 15 Pa.s., and a residence time of at least 1 day; and printed instructions describing the use of the paste and kit for treating wounds.

BRIEF DESCRIPTION OF THE DRAWING

[0008] **Fig. 1** is a perspective view showing application of the disclosed paste to a wound.

[0009] **Fig. 2** and **Fig. 3** are perspective views respectively showing a syringe and bendable tip for use in dispensing the disclosed paste.

[0010] Like reference symbols in the various figures of the drawing indicate like elements. The elements in the drawing are not to scale.

DETAILED DESCRIPTION

[0011] The following detailed description describes certain embodiments and is not to be taken in a limiting sense. All weights, amounts and ratios herein are by weight, unless otherwise specifically noted. The terms shown below have the following meanings:

[0012] The term "adhesion" refers to the sticking together of a body structure or prosthetic material to tissue, to the sticking together of tissue to tissue with which it is in intimate contact for an extended period, or to the formation of tissue that connects body structures, prosthetic materials or tissues to one another across a normally open space.

[0013] The term "antimicrobial" when used in reference to a substance means that the substance can kill, significantly inhibit or control the growth of microbes, for example

bacteria such as *Staphylococcus aureus*, *Streptococcus epidermis*, *Pseudomonas aeruginosa* or *Escherichia coli*.

[0014] The term "biocompatible" when used in reference to a substance means that the substance presents no significant deleterious or untoward effects upon the body.

[0015] The term "biodegradable" when used in reference to a substance means that the substance will degrade or erode *in vivo* to form smaller chemical or physical species. Such degradation process may be enzymatic, chemical or physical.

[0016] The term "chitosan" refers to a polysaccharide polymer containing randomly distributed β-(1-4)-linked D-glucosamine (deacetylated) and optional N-acetyl-D-glucosamine (acetylated) monomer units, and includes chitosan derivatives in which one or more hydroxyl or amine groups of the polymer have been modified to alter the solubility or tissue adhesive characteristics of the derivative.

[0017] The term "conformal" when used in reference to a paste applied to tissue or other body structure means that the paste can form a substantially continuous layer over an area to which the paste has been applied.

[0018] The term "hemostat" means a device or material which stops blood flow.

[0019] The term "opaque" when used in reference to a material means that ordinary overhead illumination is not transmitted through about a 4 mm thick layer of the material

[0020] The term "osmolality" means the number of osmoles of solute per kilogram of solvent, as measured using a freezing point depression osmometer.

[0021] The term "paste" when used in reference to a substance means the substance is a visibly homogenous, nonporous, opaque material having a soft, malleable, spreadable consistency, for example similar to toothpaste. An opaque gel may be a paste. A collection of free flowing dry solid particles, a non-malleable solid, a porous sponge, a translucent gel, a liquid or a sprayable composition would not be a paste.

[0022] The term "protective" when used in reference to a paste applied to tissue or other body structure means that the paste may assist in returning an injured, inflamed or surgically repaired tissue surface to a normal state, *e.g.*, through one or more healing mechanisms such as modulation of an inflammatory response, phagocytosis, mucosal remodeling, reciliation or other full or partial restoration of normal function.

[0023] The term "residence time" when used in reference to a paste applied to a wound means the time period during which the paste or portion thereof remains in place *in vivo* under gross observation.

[0024] The term "room temperature" means a temperature of 20-25° C.

[0025] The term "thin" when used in reference to a protective layer atop tissue or other body structure means having an average thickness less than about two millimeters.

[0026] The term "tissue adhesive" when used in reference to a substance means that the substance will adhere to tissue.

[0027] The term "tonicity" when used in reference to a cell's response to an external substance refers to the sum of the concentration of solutes having the capacity to exert an osmotic force across a given membrane. Solutes that cannot cross the cell membrane exert an osmotic force. Depending on the solute concentration of the substance in reference to the cell membrane, tonicity may be referred to as "hypertonic", "hypotonic" or "isotonic". "Hypertonic" refers to a substance with a higher solute concentration outside a cell membrane. As such, when the substance contacts the cell membrane, water in the cell will have a tendency to move out of the cell to balance the solute concentration outside the cell membrane. "Hypotonic" refers to substance with a lower solute concentration outside the cell membrane. As such, water from outside the cell will enter into the cell, causing swelling in an attempt to balance the solute concentration inside the cell. "Isotonic" refers to a substance's solute concentration that is the same as the cell to which it comes in contact. As such, it is considered physiological with the cell and hence there is no net flow of water.

[0028] The term "viscosity" when used in reference to a substance is the extent to which the substance resists a tendency to flow when subjected to stress. Viscosity may be measured with a cone and plate viscometer that imposes a specific stress on the substance and the resultant stress deformation or resistance is measured according to ASTM F2103-11 (Part 5). The units of viscosity are reported as Pascal-seconds (Pa.s). For the disclosed pastes, viscosity values are determined and reported after the paste has been sterilized.

[0029] The term "wound" means an opening in the skin through which dermal, subdermal or deeper tissue (*e.g.,* subcutaneous fat, muscle, bone or other tissue) is exposed, *viz.,* an external wound. The wound may be initiated in a variety of ways (*e.g.,*

pressure sores from extended bed rest, wounds induced by trauma, cuts, ulcers, burns, surgical incisions and the like).

[0030] The disclosed wound dressing or method includes a chitosan paste that includes a high concentration of a water-soluble chitosan (*e.g.,* chitosan salt) dissolved in a phosphate-containing solution, the paste having a pH of at least about 4. The disclosed paste desirably has an off white to yellowish coloration, which makes it easy to visualize when applied. The disclosed paste is also desirably provided in a ready-to-use, storage-stable, injectable or extrudable form, requiring no or minimal preparation. Because the paste desirably does not include crosslinkers, it can be stored for extended time periods and desirably does not require further hydration, mixing or other similar preparation steps before application.

[0031] The disclosed paste desirably is biocompatible, biodegradable and has bactericidal and hemostatic properties. The disclosed paste desirably may be used as a topical wound dressing to absorb a substantial amount of wound exudate without undue desiccation of the wound site. The paste also desirably provides antimicrobial activity or wound healing capability or both at the wound site.

[0032] The disclosed paste may be used as a wound dressing in a variety of surgical procedures, including neurosurgery, abdominal surgery, cardiovascular surgery, thoracic surgery, head and neck surgery, pelvic surgery, skin, subcutaneous tissue procedures and the like. The paste is also useful in treating ulcers, burns, cuts and the like. **Fig. 1** shows a dispensing syringe **100** equipped with a plunger **104**, barrel **106** and tip **108**. Patient leg 116 exhibits an external wound **118** that is being filled with a layer or mass of the disclosed paste **120**. Paste **120** is allowed to remain in place while healing takes place.

[0033] **Fig. 2** shows a dispensing syringe **200** for the disclosed paste held in the right hand **202** of a surgeon. When plunger **204** is depressed into syringe barrel **206**, the disclosed paste is dispensed through straight tip **208** and bendable tip **210**, whereupon it exits tip **210** as an opaque mass **220**. **Fig. 3** shows various bent positions (in phantom) that may be formed in bendable tip **210** to facilitate access to difficult to reach portions of a patient's anatomy.

[0034] The disclosed paste may be applied as a thin film or other conformal coating in which case the layer may be relatively thin and exposed to air or other nearby gases, and

with a substantially uniform thickness throughout the layer. The disclosed paste desirably is applied to at least an extent sufficient to cover healthy or healable tissue in the wound. In some instances it will be desirable to apply the paste within and not merely atop exposed tissue within the wound. The wound dressing desirably acts as a protective paste desirably adhering to tissues (*e.g.,* cartilage or bone) at the treatment site and resists detachment or other disruption until natural degradation or degradation initiated by irrigation or hydrolysis takes place. The wound dressing may be reapplied as often as needed. The residence time or treatment time may be for example from at least 1 day, at least 3 days, at least 5 days, at least 7 days, at least 15 days, up to about 3 weeks, up to about 4 weeks, up to about 45 days or up to about 60 days. The disclosed paste may be used alone or in conjunction with wound coverings that may include bandages, cotton gauze, absorptive pads or the like.

[0035] Applying the wound dressing may significantly reduce or prevent bacterial recolonization or reinfection, and may improve healing. The protective paste may provide various therapeutic advantages including but not limited to bacterial adhesion inhibition, anti-infective properties, local immune modulation, tissue protection, reduction or elimination of pain or bleeding, reduction in inflammation, reduction in adhesions to critical anatomy, and the like. These advantages may arise due to a variety of mechanisms including a) killing bacteria, b) inhibiting bacterial colonization, c) inhibiting the adherence of bacteria to tissue, d) reducing tissue morbidity or abscess formation, e) reducing or preventing disease recurrence (for example, specifically reducing the chronic inflammation related to bacterial toxin and extracellular polysaccharide matrix (*viz.*, biofilm) toxin), f) coating and protecting tissue during healing, such as by maintenance of a moist wound which promotes platelet aggregation, or by closure of a dry wound without excessive scabrous formation, g) hemostasis and h) delivering therapeutic agent(s) to the treatment site.

[0036] The disclosed paste may be prepared by mixing or dissolving the initially solid ingredients (*e.g.,* water-soluble chitosan) in a phosphate-containing solution (*e.g.,* phosphate-buffered saline (PBS)). A paste is formed at room temperature (*e.g.,* about 20° C to about 25° C) when the solid ingredients become solubilized.

[0037] Water-soluble chitosans, preferably chitosan salts may be used to form the paste. For example, high concentrations of a chitosan salt may be mixed in a phosphate-containing solution (*e.g.,* PBS, glycerol phosphate disodium salt hydrate or any combination thereof) to provide a ready-to use paste. The high chitosan salt concentration contributes both to the osmolality and opacity of the resulting paste. Without intending to be bound by theory, the phosphate and chitosan may react via an ionic reaction to help form the paste. The chitosan desirably is sufficiently water-soluble so that all of the desired chitosan amount can be dissolved in the disclosed paste. A portion of the chitosan may however be dispersed in the disclosed paste. The chosen chitosan preferably has a water solubility of at least about 3 wt. %, at least about 5 wt. %, at least about 8 wt. %, at least about 10 wt. %, at least about 15%, at least about 18% or at least about 20 wt. %. Exemplary chitosan concentrations may be from about 3 wt. % to about 20 wt. %, from about 5 wt. % to about 20 wt. % , from about 8 wt. % to about 18 wt. %, from about 10 wt. % to about 18 wt. %, or from about 15 wt. % to about 18 wt. % of the total paste weight. The high chitosan concentrations used in the paste result in desirable viscosities and acceptable syringe delivery force. Desired viscosities range from about 1 to about 15 Pa.s. when tested at 25° C and a shear rate of $221s^{-1}$. This shear rate correlates to the approximate average shear rate the substance may experience as it is dispensed through a standard 30 ml BD™ syringe with a LUER LOCK™ connector at a rate of 1 ml/s.

[0038] Exemplary unmodified, water-soluble chitosans and their salts (including chloride, citrate, nitrate, lactate, phosphate, and glutamate salts) may be obtained from a variety of commercial sources including sources described in U.S. Patent Application Publication No. US 2009/0291911 A1.

[0039] Chitosan may also be synthesized by deacetylation of chitin (poly-N-acetyl-D-glucosamine) to eliminate acetyl groups on the nitrogen atom by hydrolysis. The resulting polymer has a plurality of repeating units (*e.g.,* about 30 to about 3000 repeating units, about 60 to about 600 repeating units, or such other amount as may be desired for the chosen end use) some or all of which contain deacetylated amino groups (*e.g.,* about 30 to about 100% or about 60 to about 95% of the total repeating units), with the remaining repeating units (if any) containing acetylated amino groups. The polymer is cationic and may be regarded as being composed from glucosamine monomers.

[0040] The chitosan may have a variety of number average molecular weights, *e.g.*, about 5 to about 2000 kDa, about 10 to about 500 kDa, or about 10 to about 100 kDa. The chitosan may for example be an ultralow molecular weight material having a number average molecular weight less than or about 30 kDa, a low molecular weight material having a number average molecular weight of about 30 to about 400 kDa, a medium molecular weight material having a number average molecular weight of about 200 to about 500 kDa or a high molecular weight material having a number average molecular weight greater than about 500 kDa. A low molecular weight chitosan is preferred. The disclosed molecular weights are weights before sterilization of the paste. The chitosan desirably is in dry particulate form prior to mixing with the phosphate solution, for example, as free-flowing granules whose average particle diameter is less than about 1 mm, less than about 100 μm, about 1 to about 80 μm, or less than 1 μm.

[0041] Chitosan derivatives may also be employed, for example derivatives in which one or more hydroxyl or amino groups have been modified for the purpose of altering the solubility or tissue adhesive characteristics of the derivative. Exemplary derivatives include thiolated chitosans, and non-thiolated chitosan derivatives such as acetylated, alkylated or sulfonated chitosans (for example O-alkyl ethers, O-acyl esters, cationized trimethyl chitosans and chitosans modified with polyethylene glycol). Chitosan derivatives may be obtained from a variety of sources. For example, thiolated chitosans may be obtained from ThioMatrix Forschungs Beratungs GmbH and Mucobiomer Biotechnologische Forschungs-und Entwicklungs GmbH or prepared by reaction of chitosan with a suitable thiolated reactant, *e.g.*, as described in Published PCT Application No. WO 03/020771 A1 or in Roldo et al., *Mucoadhesive thiolated chitosans as platforms for oral controlled drug delivery: synthesis and in vitro evaluation*, European Journal of Pharmaceutics and Biopharmaceutics, 57, 115–121 (2004); Krauland et al., *Viscoelastic Properties of a New in situ Gelling Thiolated Chitosan Conjugate*, Drug Development And Industrial Pharmacy, 31, 885-893 (2005); Bernkop-Schnürch, *Thiomers: A new generation of mucoadhesive polymers*, Advanced Drug Delivery Reviews, 57, 1569-1582 (2005); and Bernkop-Schnürch et al., *Thiomers: Preparation and in vitro evaluation of a mucoadhesive nanoparticulate drug delivery system*, International journal of Pharmaceutics, 317, 76-81 (2006).

[0042] The paste desirably has a pH appropriate for contacting human tissue, *e.g.,* a pH of at least 4, a near-neutral pH, or a pH less than 10. An acid, base or buffering agent may for example be included to help maintain an appropriate pH. Buffering agents are preferred and phosphate-containing buffers are most preferred. Exemplary buffering agents include mixtures of barbitone sodium, glycinamide, glycine, potassium chloride, potassium phosphate, potassium hydrogen phthalate, sodium acetate, sodium citrate or sodium phosphate with their conjugate acids (for example a mixture of sodium citrate and citric acid).

[0043] Exemplary phosphate-containing buffers are derived from phosphoric acid and a base selected from potassium hydroxide, sodium hydroxide, the potassium or sodium salts of phosphoric acid, mixtures thereof and the like. Exemplary phosphate salts include sodium phosphate dibasic and monobasic, potassium phosphate dibasic and monobasic and mixtures thereof. The concentration of phosphoric acid and base or salt in the disclosed buffering agent may be varied to achieve the desired pH.

[0044] Exemplary phosphate-containing solutions include phosphate-containing buffers such as PBS. PBS solutions typically include a combination of one or more phosphate salts and one or more chloride salts. Exemplary phosphate salts include disodium phosphate, potassium dihydrogen phosphate or a combination thereof. Exemplary chloride salts include sodium chloride, potassium chloride or a combination thereof. The salts used to prepare the PBS solution are optionally hydrates. An exemplary combination of salts employs disodium phosphate heptahydrate ($Na_2HPO_4.7H_2O$) and potassium dihydrogen phosphate (KH_2PO_4), in a so-called 1X PBS solution with concentrations of about 0.01M phosphate, about 0.0027M KCl, about 0.137 M NaCl and a pH of 7.4 at 25° C. PBS buffer solutions may be prepared in other strengths such as 2X, 3X, 5X, 10X or any other suitable strength. For example, a 10X PBS buffer may be prepared by adding 10 times the ingredients described for a 1X PBS buffer to result in concentrations of about 0.1M phosphate, about 0.027M KCl, and about 1.37 M NaCl. Preferably, the phosphate-containing solution is a PBS solution and more preferably is greater than a 1X PBS solution. Preferably, the PBS solution has a pH between about 9 to about 12, and more preferably is a 3X solution having a pH of about 11.

[0045] Phosphates may also be provided as salts of glycerol-3-phosphate (GlyPhos) (*e.g.,* as sodium, potassium, calcium or magnesium salts). Stereoisomeric forms of GlyPhos, preferably the racemic, meso, α and β blends or other forms or blends, may also be used. In some embodiments, the phosphate may be provided by a buffering agent (*e.g.,* PBS), by a salt of GlyPhos or both.

[0046] When a high chitosan salt concentration is used, the paste may have a corresponding high osmolality and may for example exceed 2000 mOsm/kg. In some embodiments, the paste's osmolality may be up to about 3000 mOsm/kg. Such wound dressings may be desirable to provide a dehydrating wound environment. In other wound treatments, it may be desirable that the paste be physiologically compatible with the cells or tissues to which the paste may be applied. In such cases, the paste desirably has an osmolality such that the paste will be considered isotonic or hypertonic to the cells or tissues to which the paste is applied. In these applications, it is preferable that the paste have an osmolality of less than about 2000 mOsm/kg, for example from about 270 to about 1500 mOsm/kg, while still retaining a viscosity of about 1 to about 15 Pa.s.

[0047] To maintain or lower the osmolality without significantly altering or reducing the desired viscosity or the paste-like consistency, it was found that one or more osmolality reducing agents may be optionally used. The osmolality reducing agent desirably is sufficiently water-soluble so that all of the desired amount of the osmolality reducing agent can be dissolved in the disclosed paste. A portion of the osmolality reducing agent may however be dispersed in the disclosed paste. The chosen osmolality reducing agent preferably has a water solubility of at least about 1 wt. %, at least about 2 wt. %, at least about 8 wt. %, at least about 10 wt. %, or at least about 20 wt. %. The osmolality reducing agent desirably contains few or no salt groups as such groups may increase rather than decrease the osmolality of the disclosed paste. Recommended amounts of the osmolality reducing agent may for example be about 1 to about 20 wt. %, about 1 to about 10 wt. % or about 2 to about 8% wt. of the total paste weight. Examples of suitable osmolality reducing agents include polysaccharides other than chitosans that are biocompatible and which reduce osmolality in the disclosed paste but which do not crosslink the chitosan. Examples of such polysaccharides include agars, alginates, carrageenans, celluloses, dextrans, galactomannans, glycogens, hyaluronic acids, and

starches, and especially those that are water-soluble to the desired extent without containing salt groups.

[0048] Preferred polysaccharides include hydroxyl-functional or alkyl-modified celluloses. Exemplary cellulose materials include methylcellulose, ethylcellulose, hydroxybutyl methylcellulose, hydroxyethylcellulose, hydroxypropylcellulose, hydroxypropyl methylcellulose, carboxymethylcellulose, and mixtures thereof. Without intending to be bound by theory, it is believed that the osmolality reducing agent serves as a "salt scavenger" that may help reduce the paste's osmolality and maintain the desired paste-like consistency.

[0049] The paste may for example contain chitosan (and an osmolality reducing agent, if employed) in a combined amount representing about 1 to about 20 wt. %, about 10 to about 20 wt. % or about 10 to about 15 wt. % of the total paste composition. When an osmolality reducing agent is employed, the chitosan and osmolality reducing agent may for example be combined in a ratio of about 10:1 to about 1:20, about 5:1 to about 1:10, or about 3:1 to about 1:5. The chitosan (and the osmolality reducing agent, if employed) are at least partially and desirably fully dissolved in the phosphate-containing solution.

[0050] The disclosed paste may consist of or consist essentially of the above-mentioned water-soluble chitosan or derivative, phosphate-containing solution and, if used, the osmolality reducing agent, optionally together with any one or more of a variety of adjuvants. Exemplary adjuvants include lubricants, wetting agents, therapeutic agents, antimicrobial agents, dyes, pigments or other colorants, indicators, flavoring or sweetening agents, antioxidants, antifoam agents, thixotropes, release agent modifiers for sustained or delayed release of therapeutic agents, and other ingredients that will be known to persons having ordinary skill in the art.

[0051] Lubricants and wetting agents are especially desirable adjuvants that may help maintain paste consistency, and may aid in dispensing the paste into or onto a desired treatment site. Desirably, the paste should be able to be dispensed by an operator from a suitable delivery device (for example a syringe) using a single gloved hand. One preferred class of lubricants and wetting agents includes hydroxy compounds having two or more hydroxyl groups with the presence of 1,2-diol grouping being desirable. Hydroxy compounds having 2-4 carbon atoms have been found to be particularly useful lubricants.

Glycerol is especially preferred. Other compounds include ethane-1,2-diol; propane-1,2-diol; butane-1,3-diol and butane-1,4-diol. Mixtures of hydroxy compounds may be employed, especially mixtures of glycerol and one or more diols. Desired amounts of the lubricants and wetting agents may for example be about 1 to about 15 wt. % or about 2 to about 12 wt. % of the total paste weight.

[0052] Exemplary therapeutic agents include any material suitable for use at the intended treatment site including analgesics, anti-cholinergics, anti-fungal agents, antihistamines, steroidal or non-steroidal anti-inflammatory agents, anti-parasitic agents, antiviral agents, biostatic paste, chemotherapeutic agents, antineoplastic agents, cytokines, hemostatic agents (*e.g.,* thrombin), immunosuppressors, mucolytics, nucleic acids, peptides, proteins, steroids, vasoconstrictors, vitamins, mixtures thereof, and other therapeutic materials that will be known to those skilled in the art. A useful list of such therapeutic agents may also be found, for example, in U.S. Patent No. 7,959,943 B2 (Hissong et al).

[0053] The disclosed paste desirably is inherently antimicrobial without requiring addition of a separate antimicrobial agent. Antimicrobial activity may be influenced by the proportion of chitosan in the paste. A separate antimicrobial agent may be employed if desired. A useful list of such antimicrobial agents may be found, for example, in U.S. Patent No. 7,959,943 B2.

[0054] Exemplary dyes, pigments or other colorants include FD & C Red No. 3, FD & C Red No. 20, FD & C Yellow No. 6, FD & C Blue No.2, D & C Green No. 5, D & C Orange No. 4, D & C Red No. 8, caramel, titanium dioxide, fruit or vegetable colorants such as beet powder or beta-carotene, turmeric, paprika and other materials that will be known to those skilled in the art). Exemplary indicators; flavoring or sweetening agents include anise oil, cherry, cinnamon oil, citrus oil (*e.g.,* lemon, lime or orange oil), cocoa, eucalyptus, herbal aromatics (*e.g.,* clove oil, sage oil or cassia oil), lactose, maltose, menthol, peppermint oil, saccharine, sodium cyclamate, spearmint oil, sorbitol, sucrose, vanillin, wintergreen oil, xylitol and mixtures thereof.

[0055] The disclosed paste typically will be placed in suitable sealed packaging (for example, a syringe, a vial, or pouch made of suitable materials, or a kit containing such packaging and optional printed instructions) and subjected to sterilization before being

further packaged if need be with printed instructions describing the proper use of the paste or kit in the treatment of wounds. Sterilization methods that do not unduly discolor (*e.g.*, brown), affect the adhesive strength or viscosity or otherwise unduly affect the paste's consistency are desirable. Suitable sterilization methods include steam or ionizing radiation (*e.g.*, gamma radiation and electron beam (E-Beam)). E-Beam sterilization appears to prevent or limit paste discoloration. E-beam sterilization may be performed at reduced temperatures as described in U.S. Patent No. 8,653,319 B2 (Amery et al). E-beam or gamma sterilization may for example be used at doses in the range of about 12 to about 40 kGy. In some embodiments the disclosed paste may be translucent before sterilization and opaque after sterilization.

[0056] The paste desirably is provided as a ready-to-use composition requiring little or no mixing, stirring, hydration or other preparation. The paste desirably is provided in a dispenser from which the paste may be injected or extruded, and may for example be packaged in unit doses of about 5 to about 100 g. The paste desirably has good shelf life as determined by adhesive strength, viscosity and pH, and preferably may be stored for more than 12 months. In some embodiments, the paste may be stored for more than 15 months, more than 18 months or up to 24 months while still maintaining a viscosity of about 1 to about 15 Pa.s. If the paste appears to have separated, re-mixing (*e.g.*, moving the composition back and forth between two syringes) typically will return the paste to a more homogenous consistency. However, the paste preferably does not separate during storage. The paste desirably is stable at temperatures ranging from about 2° C to about 60° C. In addition, the paste desirably remains a paste after exposure to extreme temperature ranges imposed during ISTA-2A testing (*e.g.*, about -29°C to about 60°C).

[0057] The disclosed paste also may have desirable tissue adhesion. In other words, the disclosed paste preferably will adhere or stick to the specific body tissue or passageway to which it is applied without having to fully pack the passageway to obtain adequate retention in the passageway. Desirably, the paste has an adhesive strength such that a separation force of about 5 grams to about 80 grams, about 20 to about 50 grams or about 15 to about 30 grams may be required. The separation force may be measured as the force required when using a tensile testing machine (*e.g.*, an MTS™ tensile testing machine) operated at a separation rate of 1 mm/s to separate two collagen-coated, rubber

hemispheres compressed against one another, with a sample of the paste between them, using about a 4.4 Newton (1 pound) compression force. The rubber hemispheres desirably are made from ultra soft Shore OO, 30 durometer black rubber balls with a diameter of about 5 cm (2 inches) that have been bisected to a height of about 2.5 cm (1 inch). The hemispheres desirably are mounted in the tensile tester in opposing convex relationship with a piece of sausage casing wrapped over each hemisphere. About 0.2 to about 0.5 ml of the disclosed paste desirably is applied to the center of the lower hemisphere, the hemispheres are compressed together using the specified compression force and the force required to separate the hemispheres at the specified separation rate is recorded. Adhesion strength values are reported for sterilized paste. Desirably, the paste has a residence time in the applied passage or structure of at least 1 day, at least 3 days, at least 5 days, or at least 7 days with or without irrigation. The paste may degrade naturally or by irrigation (*e.g.,* saline solution).

[0058] The disclosed paste desirably is non-cytotoxic with cytotoxicity scores of 0, 1 or 2 as measured by ISO Guideline 10993-5, Biological Evaluation of Medical Devices-Part 5: Tests for *in vitro* Cytotoxicity. Desirably, the paste may have a cytotoxicity score of 1 or less.

[0059] The disclosed paste desirably is substantially collagen-free. Desirably the paste is sufficiently free of collagen (*e.g.,* containing no collagen at all) so as to be saleable worldwide for use without restriction in humans. The disclosed paste desirably does not contain other ingredients which might potentially harm tissue in or near wounds.

[0060] The disclosed paste may be used as a part of a multi-step treatment regimen. For example, a series of steps that may be broadly classified as Cleansing/Disrupting, Killing, Protecting/Coating, and Healing may be carried out. The Cleansing/Disrupting step may be carried out by administering a solvating system like those described in U.S. Patent Nos. 7,976,873 B2 and 7,976,875 B2 and in U.S. Patent Application Publication No. 2011/0245757A1. The Killing step may be carried out by applying a suitable antimicrobial agent to the treatment site. This may for example be accomplished by including an antimicrobial agent in the solvating system, as a separately-applied agent, or in both the solvating system and the separately-applied agent. An antimicrobial agent may also be applied or administered post operatively. The Protecting/Coating step may be

carried out by coating at least part of the thus-treated tissue with the disclosed paste. The Healing step may be carried out by allowing the cleansed, protected and sealed tissue surface to undergo a return to a normal state, *e.g.,* through one or more healing mechanisms such as modulation of an inflammatory response, phagocytosis, or full or partial restoration of normal function. The multi-step treatment regimen may include or be followed by a Clearing step in which the disclosed paste is sufficiently biodegradable to disappear from the treatment site in a desired time period, *e.g.,* more than 5 days, or about 7 to 15 days or by irrigation.

[0061] The invention is further illustrated in the following non-limiting examples.

Example 1

Paste Formulations

[0062] A 3X PBS (pH 11-12) solution was prepared by dissolving PBS tablets in water and the pH was adjusted to pH 11-12 using 1N NaOH. Glycerol if used was then added to the PBS solution to form a PBS/Glycerol solution. To either the PBS solution or PBS/Glycerol solution were added varying amounts of dry ingredients, namely a 30-400 kDa molecular weight chitosan or glycerol phosphate disodium salt hydrate solid and mixed at room temperature to form a paste. The paste was then gamma sterilized. All formulations formed a paste and remained a paste at room temperature. Table 1 shows the percentage of the ingredients in the total volume of liquid. Table 1 also shows the osmolality, viscosity and adhesion values for each formulation after sterilization.

Table 1

Formulation	Chitosan HCl (%)	Glycerol (%l)	BGlyPh (%)	Osmolality (mOsm/kg)	Sterile Viscosity (Pa.s.) at Shear Rate 221 (1/s)	Average Sterile Adhesion Force (grams)
1	13	0.6		2087	1.9	26.6
2	17	0.6		1730	3.2	26.4 – 37.7
3	17	0.6	6	2882	4.9	38.3

Example 2

[0063] 0.6 ml of 10 % glycerol and 5.4 ml of 3X PBS solution (pH 11) were mixed in a 10 ml syringe. To this PBS/Glycerol solution was added varying amounts of glycerol phosphate, chitosan *HCl* and a hydroxypropyl cellulose (HPC) in solid form. After all the ingredients in the syringe were fully mixed at room temperature, the resultant paste was gamma or E-beam sterilized. The formulations are shown below in Table 2. All the formulations formed a paste at room temperature and remained a paste at room temperature. Table 2 shows the percentage of the various ingredients reported as a percentage of the total volume of liquid. The osmolality, viscosity and adhesion values for each formulation are shown below in Table 3 and are values after sterilization.

Table 2

Formulation	Chitosan HCl (%)	HPC (%)	Glycerol Phosphate (%)
4	8.5	3	1
5	13	4	2
6	10	3	1
7	10	3	1.5
8	13	2	2

Table 3

Formulation	Osmolality (mOsm/kg)	Sterile Viscosity (Pa.s.) at Shear Rate 221 (1/s) Gamma	Sterile Viscosity (Pa.s.) at Shear Rate 221 (1/s) E-Beam	Sterile Adhesion Force (grams) Gamma	Sterile Adhesion Force (grams) E-Beam
4	Less than 1492	3	3.2	13.5	40.6
5	Greater than or equal to 1492	1. 7	0.4	14.5	45.5
6	525	1.8	2.3	22.8	65.0
7	Less than 1492	1.6	1.4	17.4	67.3
8	Greater than or equal to 1492	2.2	3.2	19.8	50.5

Example 3

Antimicrobial Properties

[0064] Formulations 4, 5 and 6 from Table 2 were evaluated to determine their antimicrobial activity against four common bacterial strains (*S. aureus*, *S. epidermis*, *E. coli* and *P. aeruginosa* using a zone of inhibition screening technique.

[0065] The four bacteria were grown on Muller Hinton agar plates. Under sterile conditions, approximately 0.1 to 0.2 ml of each formulation was directly placed on the agar plates. The agar plates were incubated at 35 °C for 12 hours. After incubation, the plates were observed for bacterial growth. The use of the term "zone of inhibition" denotes an area around the formulations where bacterial growth was inhibited. The term "bacteriostatic" denotes that the bacteria grew to the edge of the formulation but no further growth was observed. In other words, the term "bacteriostatic" refers to an ability to prevent bacteria from growing and multiplying but possibly not killing them.

[0066] The results shown in Table 4 below are based on triplicates per formulation.

Table 4

Antimicrobial Properties

	Zone of Inhibition or Bacteriostatic		
Bacterial Strains	**Formulation 14**	**Formulation 15**	**Formulation 16**
S. aureus	zone of inhibition	zone of inhibition	zone of inhibition
S. epidermis	zone of inhibition	zone of inhibition	zone of inhibition
E. coli	zone of inhibition	zone of inhibition	zone of inhibition
P. aeruginosa	bacteriostatic	bacteriostatic	bacteriostatic

[0067] The results show that the formulations were antimicrobial and produced zones of inhibition.

[0068] Formulation 6 was evaluated according to United States Pharmacopeia (USP) Guidelines for Performing Antimicrobial Effectiveness Testing, Chapter <51> and Validation of Microbial Recovery, Chapter <1227> to determine antibacterial efficacy

against common organisms typically found in treatment application areas. The measured antimicrobial properties are shown below in Table 5:

Table 5

Antimicrobial Properties, USP

Bacterial strain	ATCC No.	Gram stain	Antibacterial -Log Reduction			
			1 hour	24 hours	3 days	7 days
Pseudomonas aeruginosa	9027	neg	3	4.8	5.3	>5.3
Staphylococcus aureus	25923	pos	0	2.7	4	>5.0
Staphylococcus epidermidis	12228	pos	1.2	5.1	5	>5.3
Echerichia coli	25922	neg	0	1.6	3.9	>5.1
Citrobacter freundii	8090	neg	N/A	4.7	5	>5.3
Enterobacter aerogenes	13048	neg	N/A	2	2.3	>5.1
Klebsiella pneumonia	4352	neg	N/A	4	4	>5.2
Proteus mirabilis	4630	neg	N/A	2	4	>5.3
Serratia marcescens	13880	neg	N/A	1.5	2.8	>5.0
Haemophilus influenzae	53782	neg	N/A	>4.9	>4.9	>4.9
Moraxella catarrhalis	8193	neg	N/A	2.9	4	>5.0
Staphylococcus aureus(MRSA)	33591	pos	N/A	0.6	3.7	>5.1
Staphylococcus saprophyticus	15305	pos	N/A	4.6	4.9	>5.1
Micrococcus luteus	49732	pos	N/A	2.8	4	>5.0
Streptococcus mutans	25175	pos	N/A	3.4	4.4	>5.5
Streptococcus pneumoniae	10015	pos	N/A	2.5	3.6	>5.0
Corynebacterium diphtheriae	296	pos	N/A	1.6	>4.5	1.5
Corynebacterium tuberculostearicum	35693	pos	N/A	1.6	>5.1	3.9

Example 4

Cytotoxicity

[0069] Formulations 4 and 6 were either E-beam or gamma sterilized and were evaluated for potential cytotoxic effects following ISO Guideline 10993-5, Biological Evaluation of Medical Devices - Part 5: Tests for *in vitro* Cytotoxicity. Formulations 4 and 6 were extracted in purified water (PW) at 37° C for 24 hours. The PW extract was mixed with double strength Minimum Essential Medium (2X MEM) to a 50% concentration. A negative control (high density polyethylene) and reagent control (*e.g.*, PW) were similarly prepared. A positive control (powder-free latex gloves which include natural rubber latex, zinc carbamate accelerators, zinc oxide and titanium dioxide) was extracted in single strength MEM (1X MEM) at 37° C for 24 hours. Triplicates of a mammalian cell culture monolayer having L-929 mouse fibroblast cells were dosed with each extract (formulations 14, 16, positive, negative and reagent controls) and incubated at 37° C in the presence of 5% CO_2 for 48 hours. Following incubation, the monolayers were examined microscopically (100X) for abnormal cell morphology and cellular degeneration.

[0070] To confirm the scores, 0.2 ml of trypan blue stain was added to wells containing the test samples. The trypan blue molecule is large and cannot readily be absorbed by live cells. Only dead cells or those with compromised cell membranes take up the blue colored stain. Table 6 describes the scoring and visual characteristics.

Table 6

Grade/ Score	Reactivity	Conditions of all Cultures
0	None	Discrete intracytoplasmic granules, no cell lysis, no reduction of cell growth.
1	Slight	Not more than 20% of the cells are round, loosely attached and without intracytoplasmic granules, or show changes in morphology; occasional lysed cells are present; only slight growth inhibition observable.
2	Mild	Not more than 50% of the cells are round, devoid of intracytoplasmic granules; no extensive cell lysis; not more than 50% growth inhibition observable.
3	Moderate	Not more than 70% of the cell layers contain rounded cells or are lysed; cell layers not completely destroyed, but more than 50% growth inhibition observed.
4	Severe	Nearly complete or complete destruction of the cell layers.

[0071] Set out below in Table 7 are the results for either E-beam (e) or gamma (g) sterilized versions of Formulations 4 and 6, with the subscripts e or g denoting the sterilization method:

Table 7

Cytotoxicity

Sample	Percent Rounding	Percent Cells Without Intracytoplasmic Granules	Percent Lysis	Grade	Reactivity
Formulation 4_e	0	0	0	0	None
Formulation 4_g	0	0	0	0	None
Formulation 6_e	10	10	10	1	Slight
Formulation 6_g	0	0	0	0	None
Negative Control	0	0	0	0	None
Reagent Control	0	0	0	0	None
Positive Control	Not Applicable	Not Applicable	100	4	Severe

[0072] Formulation 6_e showed slight cell lysis or toxicity but is generally considered to be non-cytotoxic with a cytoxicity score of 1. Formulations 6_g, 4_e and 4_g were shown to be nontoxic with each having a cytoxicity score of 0.

[0073] Some additional non-limiting embodiments are provided below to further exemplify the present invention.

1. A paste in sterile packaging comprising a paste composition comprising a water-soluble chitosan or derivative thereof and a lubricating or wetting agent dissolved in a phosphate-containing solution wherein the composition is a paste at room temperature and has a pH of at least 4, a viscosity of about 1 to about 15 Pa.s., and the paste adheres to a surgical site and has a residence time of at least 1 day.

2. A wound dressing comprising a paste composition comprising a water-soluble chitosan or derivative thereof and a lubricating or wetting agent dissolved in a phosphate-containing solution wherein the composition is a paste at room temperature and has a pH of at least 4, a viscosity of about 1 to about 15 Pa.s., and the paste adheres to a surgical site and has a residence time of at least 1 day.

3. The embodiment 1 or 2 wherein the water-soluble chitosan comprises a salt.

4. Any of embodiments 1 to 3 wherein the water-soluble chitosan comprises a hydrochloric acid salt.

5. Any of embodiments 1 to 4 wherein the water-soluble chitosan is about 3 wt. % to about 20 wt. % of total paste weight.

6. Any of embodiments 1 to 5 wherein the water-soluble chitosan is about 15 wt. % to about 18 wt. % of total paste weight.

7. Any of embodiments 1 to 6 wherein the phosphate-containing solution is a phosphate-buffered saline (PBS).

8. Any of embodiments 1 to 7 wherein the phosphate-containing solution has a pH of 9 to 12.

9. Any of embodiments 1 to 8 wherein the lubricating or wetting agent comprises glycerol.

10. Any of embodiments 1 to 9 having a pH of 4 to 7.

11. Any of embodiments 1 to 10 wherein the residence time is at least 3 days.

12. Any of embodiments 1 to 11 having an adhesive strength of about 5 grams and about 80 grams of force required when using a tensile testing machine operated at a separation rate of 1 mm/s to separate two collagen-coated rubber hemispheres that have been compressed against one another, with a sample of the paste between them, using about a 4.4 Newton compression force.

13. Any of embodiments 1 to 12 further comprising glycerol phosphate.

14. Any of embodiments 1 to 13 further comprising an osmolality reducing agent.

15. The embodiment 14 wherein the osmolality reducing agent comprises a hydroxyl-functional or alkyl-modified cellulose.

16. The embodiment 15 wherein the hydroxyl-functional or alkyl-modified cellulose is hydroxypropyl cellulose, methyl cellulose or hydroxyethyl cellulose.

17. Any of embodiments 14 to 16 wherein the osmolality is about 270 to about 2000 mOsm/kg.

18. Any of embodiments 1 to 17 that is non-cytotoxic.

19. The embodiment 18 having a cytotoxicity score of 1 or less.

20. Any of embodiments 1 to 19 wherein the sterile packaging is electron-beam or gamma sterilized.

21. The embodiment 20 wherein the sterilization energy is about 12 to about 40 kGy.

22. A wound dressing formed from any of embodiments 1 to 21.

[0074] All patents, patent applications and literature cited in the specification are hereby incorporated by reference in their entirety. In the case of any inconsistencies, the present disclosure, including any definitions therein will prevail. Although specific embodiments have been illustrated and described herein for purposes of description of the preferred embodiments, it will be appreciated by those of ordinary skill in the art that a wide variety of alternate or equivalent implementations calculated to achieve the same purposes may be substituted for the specific embodiments shown and described without departing from the present invention. This application is intended to cover any adaptations or variations of the preferred embodiments discussed herein. Therefore, it is manifestly intended that this invention be limited only by the claims and the equivalents thereof.

We claim:

1. A method for treating a wound comprising:

applying to a wound a paste composition comprising a water-soluble chitosan or derivative thereof dissolved in a phosphate-containing solution, wherein the composition is a paste at room temperature and has a pH of at least 4, a viscosity of about 1 to about 15 Pa.s., and a residence time of at least 1 day.

2. Use of a paste composition to treat a wound, the composition comprising a water-soluble chitosan or derivative thereof dissolved in a phosphate-containing solution, wherein the composition is a paste at room temperature and has a pH of at least 4, a viscosity of about 1 to about 15 Pa.s., and a residence time of at least 1 day.

3. A kit for treating a wound, the kit comprising sterile packaging containing a paste composition comprising a water-soluble chitosan or derivative thereof in a phosphate-containing solution, wherein the composition is a paste at room temperature and has a pH of at least 4, a viscosity of about 1 to about 15 Pa.s., and a residence time of at least 1 day; and printed instructions describing the use of the paste and kit for treating wounds.

4. A method according to claim 1, use according to claim 2 or kit according to claim 3 wherein the water-soluble chitosan comprises a hydrochloric acid salt.

5. A method, use or kit according to any preceding claim wherein the water-soluble chitosan is about 3 wt. % to about 20 wt. % of the total paste weight.

6. A method, use or kit according to any preceding claim wherein the water-soluble chitosan is about 15 wt. % to about 18 wt. % of the total paste weight.

7. A method, use or kit according to any preceding claim wherein the water-soluble chitosan has a number average molecular weight of about 5 to about 2000 kDa.

8. A method, use or kit according to any preceding claim wherein the water-soluble chitosan has a number average molecular weight of about 30 to about 400 kDa.

9. A method, use or kit according to any preceding claim wherein the phosphate-containing solution is a phosphate-buffered saline.

10. A method, use or kit according to any preceding claim wherein the phosphate-containing solution is a 3X phosphate-buffered saline.

11. A method, use or kit according to any preceding claim wherein the phosphate-containing solution has a pH of 9 to 12.

12. A method, use or kit according to any preceding claim wherein the phosphate-containing solution comprises a glycerol phosphate.

13. A method, use or kit according to any preceding claim further comprising a lubricant or wetting agent.

14. A method, use or kit according to claim 12 wherein the lubricant or wetting agent is glycerol.

15. A method, use or kit according to claim 12 wherein the lubricant or wetting agent is about 1 wt. % to about 10 wt. % of total paste weight.

16. A method, use or kit according to any preceding claim wherein the paste is sterilized.

17. A method, use or kit according to claim 15 wherein the sterilization is electron-beam radiation or gamma radiation.

18. A method, use or kit according to any preceding claim wherein the paste has an adhesive strength of about 5 grams to about 80 grams of force required when using a tensile testing machine operated at a separation rate of 1 mm/s to separate two collagen-coated rubber hemispheres that have been compressed against one another, with a sample of the paste between them, using about a 4.4 Newton compression force.

19. A method, use or kit according to any preceding claim wherein the paste is non-cytotoxic with a cytotoxicity score of 1 or less.

20. A method, use or kit according to any preceding claim packaged as a ready-to-use composition.

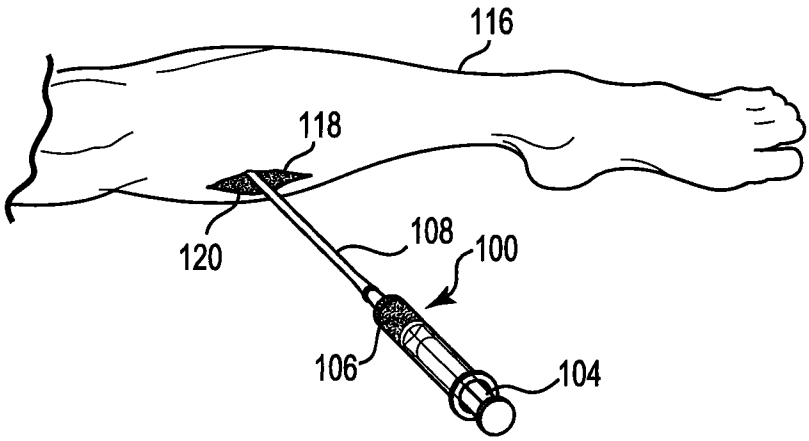

Fig. 1

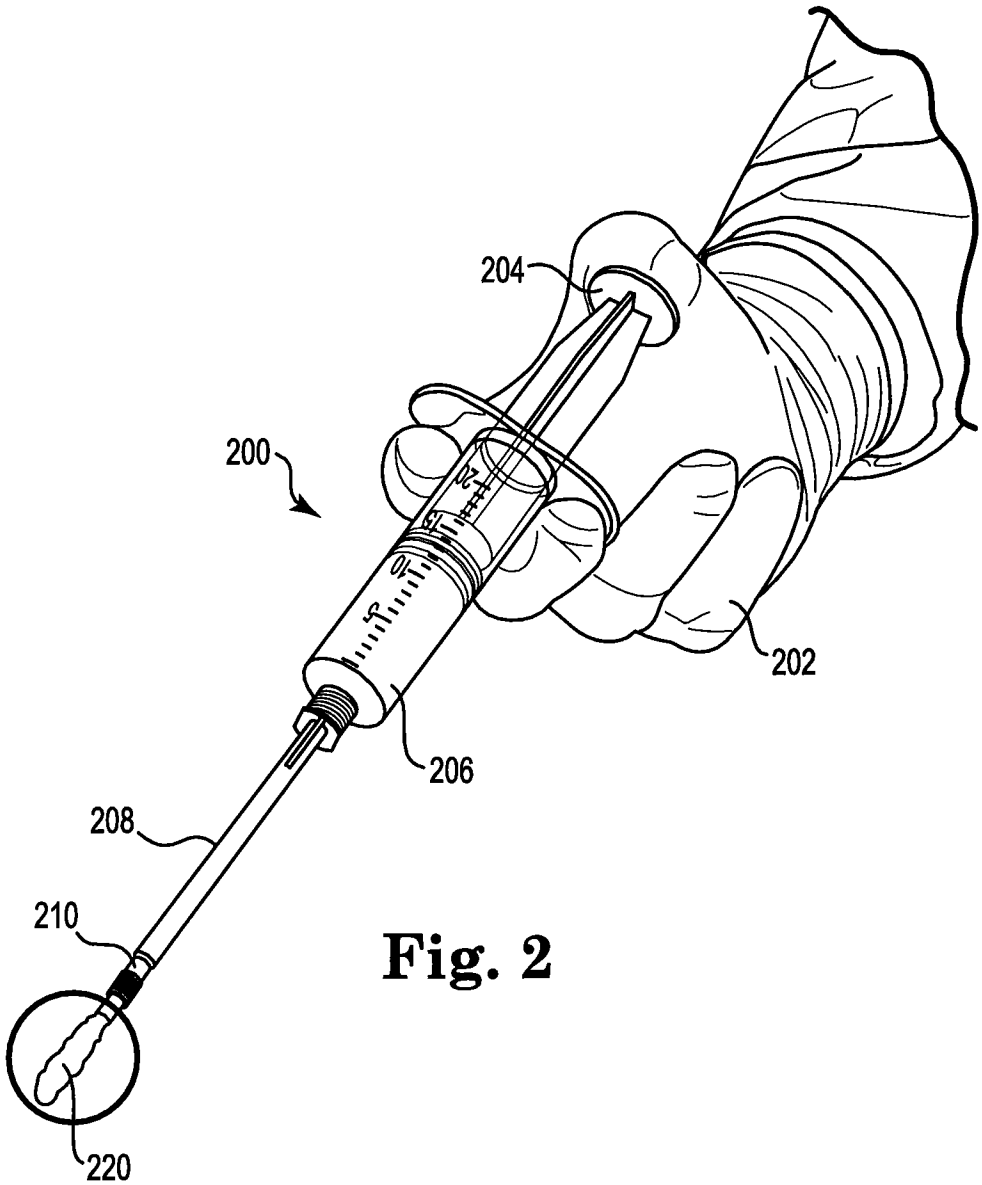

Fig. 2

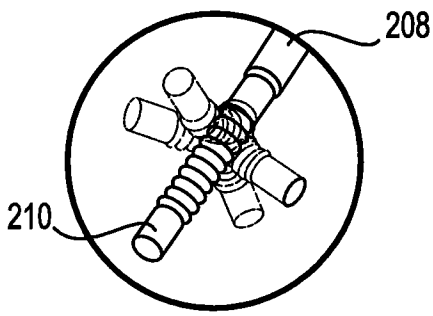

Fig. 3

INTERNATIONAL SEARCH REPORT

International application No
PCT/US2014/062041

A. CLASSIFICATION OF SUBJECT MATTER
INV. A61L26/00
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)
A61L A61K

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, WPI Data, BIOSIS, EMBASE, INSPEC

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 2011/060545 A1 (ECOLE POLYTECH [CA]; OUYANG WEI [CA]; BUSCHMANN MICHAEL [CA]; CHEVRIER) 26 May 2011 (2011-05-26) paragraphs [0001], [0010] claims 49, 56, 59-63 -----	1-5,7,8, 12,16, 18-20

-/--

[X] 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
18 December 2014	05/01/2015

Name and mailing address of the ISA/	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Cadamuro, Sergio

1

Form PCT/ISA/210 (second sheet) (April 2005)

INTERNATIONAL SEARCH REPORT

International application No
PCT/US2014/062041

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	RAHELEH AHMADI ET AL: "Biocompatibility and gelation of chitosan-glycerol phosphate hydrogels", JOURNAL OF BIOMEDICAL MATERIALS RESEARCH PART A, vol. 86A, no. 3, 1 September 2008 (2008-09-01), pages 824-832, XP055159287, ISSN: 1549-3296, DOI: 10.1002/jbm.a.31676 abstract page 824, column 2, paragraph 2 page 825, column 1, paragraph 3 page 827, column 1, paragraph 2 - column 2, paragraph 1 page 831, column 2, paragraph 3-4 -----	1-3,7, 12,16, 18-20
X	WO 99/07416 A1 (BIO SYNTECH LTD [CA]; CHENITE ABDELLATIF [CA]; CHAPUT CYRIL [CA]; COMB) 18 February 1999 (1999-02-18) page 14, lines 10-14 claims 1-2, 12, 17, 26 -----	1-3,5,7, 12-14, 16,18-20
X	WO 02/40072 A2 (BIOSYNTECH CANADA INC [CA]; DESROSIERS ERIC ANDRE [CA]; CHENITE ABDELL) 23 May 2002 (2002-05-23) page 1, paragraph 2 page 2, paragraph 2-4 page 3, paragraph 2 claims 1, 2, 4-7, 11, 12-14 -----	1-20

1

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/US2014/062041

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 2011060545 A1	26-05-2011	NONE	
WO 9907416 A1	18-02-1999	AT 246524 T	15-08-2003
		AU 724878 B2	05-10-2000
		AU 6915098 A	01-03-1999
		CA 2212300 A1	04-02-1999
		DE 69816996 D1	11-09-2003
		DE 69816996 T2	22-07-2004
		DK 1003567 T3	01-12-2003
		EP 1003567 A1	31-05-2000
		ES 2205471 T3	01-05-2004
		IL 134368 A	25-09-2005
		JP 3634748 B2	30-03-2005
		JP 2001513367 A	04-09-2001
		NO 20000593 A	29-03-2000
		NZ 502919 A	26-04-2002
		PT 1003567 E	31-12-2003
		US 6344488 B1	05-02-2002
		WO 9907416 A1	18-02-1999
WO 0240072 A2	23-05-2002	AU 2039602 A	27-05-2002
		CA 2429165 A1	23-05-2002
		EP 1333869 A2	13-08-2003
		WO 0240072 A2	23-05-2002