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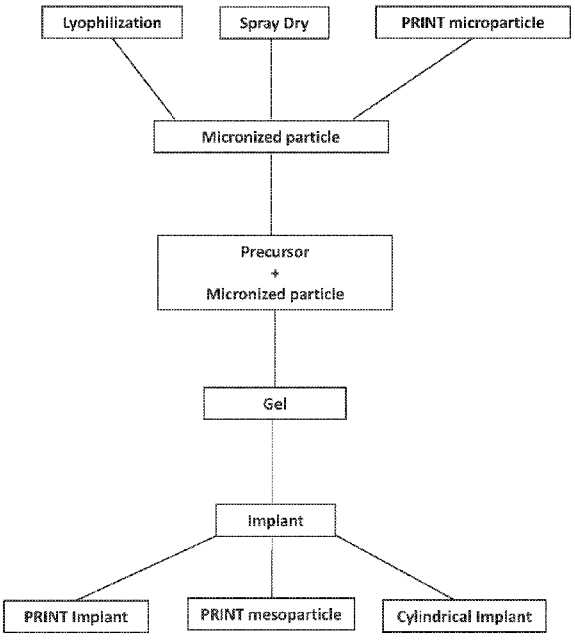
(54) Title: OCULAR PROTEIN DELIVERY

FIG. 1



(57) Abstract: The disclosure teaches methods of making ocular, sustained release pharmaceutical compositions. In embodiments, the disclosure teaches utilizing ocular, sustained release pharmaceutical compositions to treat ocular conditions. In aspects, the disclosure provides methods of treating Retinal Vein Occlusion (RVO), Age-Related Macular Degeneration (AMD), and/or Diabetic Macular Edema (DME).

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OCULAR PROTEIN DELIVERY

CROSS-REFERENCE TO RELATED APPLICATIONS

[001] The present Application claims priority to: U.S. Provisional Application No. 62/195,640, filed on July 22, 2015; U.S. Provisional Application No. 62/239,022, filed on October 8, 2015; U.S. Provisional Application No. 62/253,979, filed on November 11, 2015; U.S. Provisional Application No. 62/277,233, filed on January 11, 2016; U.S. Provisional Application No. 62/290,807, filed on February 3, 2016; U.S. Provisional Application No. 62/329,723, filed on April 29, 2016; and U.S. Provisional Application No. 62/352,259, filed on June 20, 2016, the entire contents of each of which are hereby incorporated by reference in their entirety.

TECHNICAL FIELD

[002] The present disclosure relates to the field of pharmaceutical compositions, implants formed from pharmaceutical compositions, methods of forming implants, and methods of treating ocular conditions. In some aspects, the disclosure relates to ocular delivery vehicles that are suitable for multi-month administration of biologics and small molecules.

BACKGROUND

[003] Neovascular (Wet) Age-related Macular Degeneration (AMD) affects approximately 1.7 million mainly adult patients over 40 years old in the United States. It is projected that in 2020 it could potential affect as many as 3.8 million patients. AMD is the third leading cause of blindness after cataracts and glaucoma. The disease progression leads to decreased quality of life and difficulty in daily functioning.

[004] AMD is a degenerative disease of the central portion of the retina. The degeneration is caused by abnormal vessel growth into the subretinal space usually from the choroidal circulation (choroidal neovascularization or CNV). This abnormal vessel growth is stimulated by vascular endothelial growth factor (VEGF) among other factors.

The abnormal vessels leak fluid leading to collection of subretinal fluid and/or blood beneath the retina leading to retinal thickening. Retinal thickening causes rapid distortion and loss of central vision over a period of weeks to months. Retinal thickening is reversible with therapy.

[005] Other diseases caused by abnormal vessel growth include Macular Degeneration (MD), Retinal Vein Occlusion (RVO) and Diabetic Macular Edema (DME) and Diabetic Retinopathy (DR).

[006] Anti-VEGF pharmacotherapy is the primary and most efficient treatment strategy. For treatment, the anti-VEGF agent is injected via an intravitreal injection (IVT) every four to twelve weeks. On average, a patient will receive seven injections annually. Additionally, diagnostic visits occur approximately monthly. This course of treatment is burdensome for patients and medical care providers alike.

[007] Therefore, there is a great need in the medical field for an alternative treatment using a sustained-release delivery system with an improved safety and efficacy profile.

SUMMARY OF DISCLOSURE

[008] The present disclosure addresses a crucial need in the art, by providing a sustained release pharmaceutical formulation that may be directly administered to the vitreous of the eye and that does not suffer from the drawbacks of the current monthly to semi-monthly treatment paradigm.

[009] In certain embodiments, the disclosure relates to precisely engineered biodegradable drug delivery systems and methods of making and utilizing such systems. The precisely engineered biodegradable drug delivery systems of the present invention include incorporating solid state protein microparticles (PuPs) into larger biodegradable implants for delivery and controlled release profile of the overall pharmaceutical composition.

[0010] The biodegradable drug delivery systems taught herein are, in some embodiments, engineered using PRINT® Technology (Particle Replication in Non-wetting Template) (Envisia Therapeutics Inc., North Carolina). The PRINT® Technology utilized in some

embodiments allows for uniform size, shape, and dose concentration in the disclosed drug delivery systems.

[0011] Further, the disclosure provides methods of utilizing the taught precisely engineered biodegradable drug delivery systems to treat, *inter alia,* conditions of the eye.

[0012] Some conditions treatable according to the present disclosure include AMD, Retinal Vein Occlusion (RVO), and Diabetic Macular Edema (DME).

[0013] In certain embodiments, the present disclosure relates to pharmaceutical compositions for treating an ocular condition, comprising: a biodegradable polymer matrix and at least one therapeutic agent.

[0014] In certain embodiments, the present disclosure provides for pharmaceutical compositions for treating an ocular condition, comprising: an ocular implant. In aspects, the ocular implant comprises a biodegradable polymer matrix that contains a therapeutic agent dispersed therein. In some embodiments, the ocular implant is a "non-extruded" ocular implant.

[0015] In embodiments, the biodegradable polymer matrix is formulated as a gel.

[0016] In some embodiments, the at least one therapeutic agent is selected from the group consisting of biologics and monoclonal antibodies (mAb).

[0017] In preferred embodiments, the at least one therapeutic agent is an anti-VEGF agent.

[0018] In some embodiments, the at least one therapeutic agent is selected from the group consisting of bevacizumab, ranibizumab, aflibercept, pegaptanib, ziv-aflibercept, adalimumab, eculizumab, efalizumab, lampalizumab, RG7716 (Roche), rinucumab, nesvacumab, pegpleranib, zimura, DS-7080a (Daiichi Sankyo), BCD-021 (Biocad), ocriplasmin, ABP-215 (Allergan/Amgen), FKB238, ONS-1045, BI695502, PF-06439535, BX2314, FYB201, FYB203, razumab, PF582, TK-001, sonepcizumab, and LFG316.

[0019] In particular embodiments, the at least one therapeutic agent is selected from the group consisting of bevacizumab and aflibercept.

[0020] In preferred embodiments, the at least one therapeutic agent is bevacizumab.

[0021] In certain embodiments, the disclosure provides for the manufacture and utilization of DARPins (an acronym for designed ankyrin repeat proteins), which are genetically engineered antibody mimetic proteins typically exhibiting highly specific and high-affinity target protein binding. They are derived from natural ankyrin proteins and consist of at least three, usually four or five repeat motifs of these proteins. Their molecular mass is about 14 or 18 kDa (kilodaltons) for four- or five-repeat DARPins, respectively. The disclosure envisions manufacture of these DARPins and utilization in the delivery systems taught herein.

[0022] In a particular embodiment, the disclosure provides a pharmaceutical composition: A) a biocompatible polymer matrix; and B) at least one therapeutic agent dispersed within the polymer matrix. In embodiments, the biocompatible polymer matrix is composed of two or more polymers. In embodiments, the two or more polymer are gel precursors, which may interact to for a gel. Thus, in embodiments, the pharmaceutical composition is gel comprising a micronized therapeutic agent. In embodiments, the gel is a hydrogel. In embodiments, the pharmaceutical composition can be formulated for the sustained delivery of therapeutic agents.

[0023] In embodiments, the pharmaceutical compositions described herein can be formulated for oral, parental, intramuscular, transdermal, intravenous, inter-arterial, nasal, vaginal, sublingual, and subungual. Further, the route also includes, but is not limited to auricular, buccal, conjunctival, cutaneous, dental, electro-osmosis, endocervical, endosinusial, endotracheal, enteral, epidural, extra-amniotic, extracorporeal, hemodialysis, infiltration, interstitial, intra-abdominal, intra-amniotic, intra-arterial, intra-articular, intrabiliary, intrachronchial, intrabursal, intracardiac, intracartilagenous, intracaudal, intracavernous, intracavitary, intracerebral, intracisternal, intracorneal, intracoronary, intracorporus cavernosum, intradermal, intradiscal, intraducatal, intraduodenal, intradural, intraepidermal, intraesophageal, intragastric, intragingival, intraileal, intralesional, intralumical, intralymphatic, intramedullary, intrameningeal, intraocular, intraovarian, intrapericardial, intraperitoneal, intrapleural, intrapulmonary, intrasinal, intrasynovial, intratendinous, intratesticular, intrathecal,

intrathoracic, intratubular, intratumor, intratympanic, intrauterine, intravascular, intravenous bolus, intravenous drip, intraventricular, intravesical, intravitreal, iontophoresis, irrigation, laryngeal, nasal, nasogastric, occlusive dressing technique, ophthalmic, oropharyngeal, percutaneous, periarticular, peridural, periodontal, rectal, respiratory, retrobulbar, soft tissue, subarachnoid, subconjunctival, subcutaneous, submucosal, topical, transmucosal, transplacental, transtracheal, transtympanic, ureteral, or urethal. In particular embodiments, the pharmaceutical compositions are formulated for intraocular administration, e.g., intravitreal.

[0024] In certain embodiments, the disclosure provides for a sustained delivery system for treating abnormal vascular growth in an eye of a patient in need thereof, comprising: A) a biocompatible polymer matrix based gel delivery system; and B) at least one therapeutic encapsulated in the polymer matrix, wherein said delivery system is formulated to deliver a therapeutic agent to the eye for at least about 2 months (i.e., about 3 months, about 4 months, about 5 months, about 6 months, etc.). In some aspects, the therapeutic agent is a water soluble biologic. In some aspects, the therapeutic agent is an anti-VEGF agent. In some aspects, the therapeutic agent is a protein, such as bevacizumab, ranibizumab, aflibercept, and pegaptanib. In some aspects the biocompatible polymer matrix comprises poly(ethylene glycol) (*i.e.* "PEG") polymers.

[0025] In embodiments, the pharmaceutical composition comprises as biocompatible polymer matrix content about 1 μg to about 100 mg, or about 1 μg to about 10 mg, or about 1 μg to about 1,000 μg, or about 1 μg to about 900 μg, or about 1 μg to about 800 μg, 1 μg to about 700 μg, or about 1 μg to about 600 μg, or about 1 μg to about 500 μg, 1 μg to about 400 μg, 1 μg to about 300 μg, or about 1 μg to about 200 μg, or about 1 μg to about 100 μg, or about 1 μg to about 90 μg, or about 1 μg to about 80 μg, or about 1 μg to about 70 μg, or about 1 μg to about 60 μg, or about 1 μg to about 50 μg, or about 1 μg to about 40 μg. or about 1 μg to about 40 μg, or about 1 μg to about 30 μg, or about 1 μg to about 20 μg, or about 1 μg to about 10 μg. In embodiments, the biodegradable polymer matrix forms a gel. In such embodiments, the biodegradable polymer matrix comprises gel precursors. In embodiments, the gel precursors are PEG polymers.

[0026] In embodiments, the biodegradable polymer matrix comprises as a weight percent of the pharmaceutical composition: about 1 wt % to about 99 wt %, or about 1 wt % to about 95 wt %, or about 1 wt % to about 90 wt %, or about 1 wt % to about 85 wt %, or about 1 wt % to about 80 wt %, or about 1 wt % to about 75 wt %, or about 1 wt % to about 70 wt %, or about 1 wt % to about 65 wt %, or about 1 wt % to about 60 wt %, or about 1 wt % to about 55 wt %, or about 1 wt % to about 50 wt %, or about 1 wt % to about 45 wt %, or about 1 wt % to about 40 wt %, or about 1 wt % to about 35 wt %, or about 1 wt % to about 30 wt %, or about 1 wt % to about 25 wt %, or about 1 wt % to about 20 wt %, or about 1 wt % to about 15 wt %, or about 1 wt % to about 10 wt %, or about 10 wt % to about 99 wt %, or about 10 wt % to about 95 wt %, or about 10 wt % to about 90 wt %, or about 10 wt % to about 85 wt %, or about 10 wt % to about 80 wt %, or about 10 wt % to about 75 wt %, or about 10 wt % to about 70 wt %, or about 10 wt % to about 65 wt %, or about 10 wt % to about 60 wt %, or about 10 wt % to about 55 wt %, or about 10 wt % to about 50 wt %, or about 10 wt % to about 45 wt %, or about 10 wt % to about 40 wt %, or about 10 wt % to about 35 wt %, or about 10 wt % to about 30 wt %, or about 10 wt % to about 25 wt %, or about 10 wt % to about 20 wt %, or about 10 wt % to about 15 wt %, or about 12% to about 55%. In embodiments, the biodegradable polymer matrix forms a gel. In such embodiments, the biodegradable polymer matrix comprises gel precursors. In embodiments, the gel precursors are PEG polymers.

[0027] In embodiments, the biodegradable polymer matrix comprises as a % w/w of the pharmaceutical composition: about 1 % to about 99 % w/w, or about 1 % to about 95 % w/w, or about 1 % to about 90 % w/w, or about 1 % to about 85 % w/w, or about 1 % to about 80 % w/w, or about 1 % to about 75 % w/w, or about 1 % to about 70 % w/w, or about 1 % to about 65 % w/w, or about 1 % to about 60 % w/w, or about 1 % to about 55 % w/w, or about 1 % to about 50 % w/w, or about 1 % to about 45 % w/w, or about 1 % to about 40 % w/w, or about 1 % to about 35 % w/w, or about 1 % to about 30 % w/w, or about 1 % to about 25 % w/w, or about 1 % to about 20 % w/w, or about 1 % to about 15 % w/w, or about 1 % to about 10 % w/w, or about 10 % to about 99 % w/w, or about 10 % to about 95 % w/w, or about 10 % to about 90 % w/w, or about 10 % to about 85 % w/w, or about 10 % to about 80 % w/w, or about 10 % to about 75 % w/w, or about 10 %

to about 70 % w/w, or about 10 % to about 65 % w/w, or about 10 % to about 60 % w/w, or about 10 % to about 55 % w/w, or about 10 % to about 50 % w/w, or about 10 % to about 45 % w/w, or about 10 % to about 40 % w/w, or about 10 % to about 35 % w/w, or about 10 % to about 30 % w/w, or about 10 % to about 25 % w/w, or about 10 % to about 20 % w/w, or about 10 % to about 15 % w/w. In embodiments, the biodegradable polymer matrix forms a gel. In such embodiments, the biodegradable polymer matrix comprises gel precursors. In embodiments, the gel precursors are PEG polymers.

[0028] In certain embodiments, the biodegradable polymer matrix includes a first polymer. In aspects, the first polymer comprises as a % w/w of the biodegradable polymer matrix: about 1% to about 100%, or about 1% to about 90% w/w, or about 1% to about 80%, or about 1% to about 70%, or about 1% to about 60%, or about 1% to about 50%, or about 1% to about 40%, or about 1% to about 30%, or about 1% to about 20%, or about 1% to about 15%, or about 1% to about 10%, or about 1% to about 5%, or about 10% to about 90%, or about 10% to about 80%, or about 10% to about 70%, or about 10% to about 60%, or about 10% to about 50%, or about 10% to about 45%, or about 10% to about 40%, or about 10% to about 35%, or about 20% to about 90%, or 40% to about 60%, or about 45% to about 55%, or about 50%, including all values and subranges in between. In embodiments, the first polymer is a PEG polymer. In embodiments, the PEG polymer has a molecular weight in the range of about 10,000 Da to about 15,000 Da (e.g., about 10,000 Da or about 15,000 Da). In embodiments, the PEG polymer includes at least one arm having at least one functional group located on said arm. In embodiments, the functional group is an NHS-ester. In embodiments, the PEG-NHS is an 8-arm PEG-NHS or a 4-arm PEG-NHS.

[0029] In certain embodiments, the biodegradable polymer matrix includes a first polymer. In aspects, the first polymer comprises as weight of the biodegradable polymer matrix: about 1 μg to about 1,000 μg, about 1 μg to about 500 μg, or about 1 μg to about 400 μg, or about 1 μg to about 300 μg, or about 1 μg to about 200 μg, or about 1 μg to about 100 μg, or about 1 μg to about 90 μg, or about 1 μg to about 80 μg, or about 1 μg to about 70 μg, or about 1 μg to about 60 μg, or about 1 μg to about 50 μg, or about 1 μg to about 40 μg, or about 1 μg to about 30 μg, or about 1 μg to about 20 μg, or about 1 μg to about 10 μg, including all values and subranges in between. In embodiments, the first

polymer comprises as weight of the biodegradable polymer matrix: about 5 μg to about 100 μg, or about 5 μg to about 90 μg, or about 5 μg to about 80 μg, or about 5 to about 70 μg, or about 5 μg to about 60 μg, or about 5 μg to about 50 μg, or about 5 μg to about 40 μg, or about 5 μg to about 30 μg. In embodiments, the first polymer is a PEG polymer. In embodiments, the PEG polymer has a molecular weight in the range of about 10,000 Da to about 15,000 Da (e.g., about 10,000 Da or about 15,000 Da). In embodiments, the PEG polymer includes at least one arm having a functional group located on said arm. In embodiments, the functional group is an NHS-ester. In embodiments, the PEG-NHS is an 8-arm PEG-NHS or a 4-arm PEG-NHS.

[0030] In certain embodiments, the biodegradable polymer matrix includes a second polymer. In aspects, the second polymer comprises as a % w/w of the biodegradable polymer matrix: about 1% to about 100%, or about 1% to about 90% w/w, or about 1% to about 80%, or about 1% to about 70%, or about 1% to about 60%, or about 1% to about 50%, or about 1% to about 40%, or about 1% to about 30%, or about 1% to about 20%, or about 1% to about 15%, or about 1% to about 10%, or about 1% to about 5%, or about 10% to about 90%, or about 10% to about 80%, or about 10% to about 70%, or about 10% to about 60%, or about 10% to about 50%, or about 10% to about 45%, or about 10% to about 40%, or about 10% to about 35%, or about 20% to about 90%, or 40% to about 60%, or about 45% to about 55%, or about 50%, including all values and subranges therein. In embodiments, the second polymer is a PEG polymer. In embodiments, the PEG polymer has a molecular weight in the range of about 10,000 Da to about 15,000 Da (e.g., about 10,000 Da or about 15,000 Da). In embodiments, the PEG polymer includes at least one arm having a functional group located on said arm. In embodiments, the functional group is an amine. In embodiments, the PEG-amine is an 8-arm PEG-amine or a 4-arm PEG-amine.

[0031] In certain embodiments, the biodegradable polymer matrix includes a second polymer. In aspects, the second polymer comprises as weight of the biodegradable polymer matrix: about 1 μg to about 1,000 μg, about 1 μg to about 500 μg, or about 1 μg to about 400 μg, or about 1 μg to about 300 μg, or about 1 μg to about 200 μg, or about 1 μg to about 100 μg, or about 1 μg to about 50 μg, or about 1 μg to about 40 μg, or about 1 μg to about 30 μg, or about 1 μg to about 20 μg, or about 1 μg to about 10 μg, or about

1 to about 5 μg. In embodiments, the first polymer comprises as weight of the biodegradable polymer matrix: about 5 μg to about 100 μg, or about 5 μg to about 90 μg, or about 5 μg to about 80 μg, or about 5 to about 70 μg, or about 5 μg to about 60 μg, or about 5 μg to about 50 μg, or about 5 μg to about 40 μg, or about 5 μg to about 30 μg. In embodiments, the second polymer is a PEG polymer. In embodiments, the PEG polymer has a molecular weight in the range of about 10,000 Da to about 15,000 Da (e.g., about 10,000 Da or about 15,000 Da). In embodiments, the PEG polymer includes at least one arm having a functional group located on said arm. In embodiments, the functional group is an amine. In embodiments, the PEG-amine is an 8-arm PEG-amine or a 4-arm PEG-amine.

[0032] In certain embodiments, the biodegradable polymer matrix includes a first polymer and a second polymer. In aspects, the first polymer and the second polymer comprise as a % w/w ratio of the biodegradable polymer matrix: about 1%/99% to about 99%/1%, or about 5%/95% to about 95%/5%, or about 10%/90% to about 90%/10%, or about 15%/85% to about 85%/15%, or about 20%/80% to about 80%/20%, or about 25%/75% to about 75%/25%, or about 30%/70% to about 70%/30%, or about 35%/65% to about 65%/35%, or about 40%/60% to about 60%/40%, or about 45%/55% to about 55%/45%, or about 50%/50%. In embodiments, the first polymer and the second polymer comprises as a % w/w ratio of the biodegradable polymer matrix: about 50%/50%. In embodiments, the first polymer is a PEG polymer. In embodiments, the PEG polymer has a molecular weight in the range of about 10,000 Da to about 15,000 Da (e.g., about 10,000 Da or about 15,000 Da). In embodiments, the PEG polymer includes at least one arm having at least one functional group located on said arm. In embodiments, the functional group is an NHS-ester. In embodiments, the PEG-NHS is an 8-arm PEG-NHS or a 4-arm PEG-NHS. In embodiments, the second polymer is a PEG polymer. In embodiments, the PEG polymer has a molecular weight in the range of about 10,000 Da to about 15,000 Da (e.g., about 10,000 Da or about 15,000 Da). In embodiments, the PEG polymer includes at least one arm having a functional group located on said arm. In embodiments, the functional group is an amine. In embodiments, the PEG-amine is an 8-arm PEG-amine or a 4-arm PEG-amine.

[0033] In certain embodiments, the biodegradable polymer matrix is comprised of a first polymer and a second polymer. In aspects, the first polymer and the second polymer respectively comprises as a weight of the biodegradable polymer matrix: about 1 μg to about 1000 μg and about 1 μg to about 1000 μg; or about 1 μg to about 900 μg and about 1 μg to about 900 μg; or about 1 μg to about 800 μg and about 1 μg to about 800 μg; or about 1 μg to about 700 μg and about 1 μg to about 700; or about 1 μg to about 600 μg and about 1 μg to about 600; or about 1 μg to about 500 μg and about 1 μg to about 500; or about 1 μg to about 400 μg and about 1 μg to about 400; or about 1 μg to about 300 μg and about 1 μg to about 300; or about 1 μg to about 200 μg and about 1 μg to about 200; or about 1 μg to about 100 μg and about 1 μg to about 100; or about 1 μg to about 90 μg and about 1 μg to about 90; or about 1 μg to about 80 μg and about 1 μg to about 80; or about 1 μg to about 70 μg and about 1 μg to about 70; or about 1 μg to about 60 μg and about 1 μg to about 60; or about 1 μg to about 50 μg and about 1 μg to about 50; or about 1 μg to about 40 μg and about 1 μg to about 40; or about 1 μg to about 30 μg and about 1 μg to about 30; or about 1 μg to about 20 μg and about 1 μg to about 20; or about 1 μg to about 10 μg and about 1 μg to about 10, including all values and subranges in between. In embodiments, the first polymer is a PEG polymer. In embodiments, the PEG polymer has a molecular weight in the range of about 10,000 Da to about 15,000 Da (e.g., about 10,000 Da or about 15,000 Da). In embodiments, the PEG polymer includes at least one arm having at least one functional group located on said arm. In embodiments, the functional group is an NHS-ester. In embodiments, the PEG-NHS is an 8-arm PEG-NHS or a 4-arm PEG-NHS. In embodiments, the second polymer is a PEG polymer. In embodiments, the PEG polymer has a molecular weight in the range of about 10,000 Da to about 15,000 Da (e.g., about 10,000 Da or about 15,000 Da). In embodiments, the PEG polymer includes at least one arm having a functional group located on said arm. In embodiments, the functional group is an amine. In embodiments, the PEG-amine is an 8-arm PEG-amine or a 4-arm PEG-amine.

[0034] In certain embodiments, the biodegradable polymer matrix includes a third polymer. In aspects, the third polymer comprises as a % w/w of the biodegradable polymer matrix: about 1% to about 99%, or about 1% to about 90% w/w, or about 1% to about 80%, or about 1% to about 70%, or about 1% to about 60%, or about 1% to about

50%, or about 1% to about 40%, or about 1% to about 30%, or about 1% to about 20%, or about 1% to about 10%; or 10% to about 100%, or about 10% to about 90% w/w, or about 10% to about 80%, or about 10% to about 70%, or about 10% to about 60%, or about 10% to about 50%, or about 10% to about 40%, or about 10% to about 30%, or about 10% to about 20%; or 20% to about 100%, or about 20% to about 90% w/w, or about 20% to about 80%, or about 20% to about 70%, or about 20% to about 60%, or about 20% to about 50%, or about 20% to about 40%, or about 20% to about 30%; or 30% to about 100%, or about 30% to about 90% w/w, or about 30% to about 80%, or about 30% to about 70%, or about 30% to about 60%, or about 30% to about 50%, or about 30% to about 40%; or 40% to about 100%, or about 40% to about 90% w/w, or about 40% to about 80%, or about 40% to about 70%, or about 40% to about 60%, or about 40% to about 50%; or 50% to about 100%, or about 50% to about 90% w/w, or about 50% to about 80%, or about 50% to about 70%, or about 50% to about 60%; or 60% to about 100%, or about 60% to about 90% w/w, or about 60% to about 80%, or about 60% to about 70%; or 70% to about 100%, or about 70% to about 90% w/w, or about 70% to about 80%; or 80% to about 100%, or about 80% to about 90% w/w; or 90% to about 100%; or about 2% to about 9%, or about 3% to about 8%, or about 4% to about 8%, or about 5% to about 7%; or about 5%; or about 7 %, or about 15%; or about 40%; or about 50%; or about 60%; or about 70%; or about 85%; or about 90%; or about 95%, including all values and subranges in between. In embodiments, the third polymer is a PEG polymer. In embodiments, the PEG polymer has a molecular weight in the range of about 10,000 Da to about 15,000 Da (e.g., about 10,000 Da or about 15,000 Da). In embodiments, the PEG polymer includes at least one arm having a functional group located on said arm. In embodiments, the functional group is an amine. In embodiments, the PEG-amine is an 8-arm PEG-amine or a 4-arm PEG-amine.

[0035] In certain embodiments, the biodegradable polymer matrix includes a third polymer. In aspects, the third polymer comprises as a % w/w of the pharmaceutical composition: about 1% to about 99%, or about 1% to about 90% w/w, or about 1% to about 80%, or about 1% to about 70%, or about 1% to about 60%, or about 1% to about 50%, or about 1% to about 40%, or about 1% to about 30%, or about 1% to about 20%, or about 1% to about 10%; or 10% to about 100%, or about 10% to about 90% w/w, or

about 10% to about 80%, or about 10% to about 70%, or about 10% to about 60%, or about 10% to about 50%, or about 10% to about 40%, or about 10% to about 30%, or about 10% to about 20%; or about 15% to about 100%, or about 15% to about 95%, or about 15% to about 90%, or about 15% to about 85%, or about 15% to about 80%, or about 15% to about 70%, or about 15% to about 60%, or about 15% to about 50%, or about 15% to about 40%, or about 15% to about 30%, or about 15% to about 20%, or 20% to about 100%, or about 20% to about 90% w/w, or about 20% to about 80%, or about 20% to about 70%, or about 20% to about 60%, or about 20% to about 50%, or about 20% to about 40%, or about 20% to about 30%; or 30% to about 100%, or about 30% to about 90% w/w, or about 30% to about 80%, or about 30% to about 70%, or about 30% to about 60%, or about 30% to about 50%, or about 30% to about 40%; or 40% to about 100%, or about 40% to about 90% w/w, or about 40% to about 80%, or about 40% to about 70%, or about 40% to about 60%, or about 40% to about 50%; or 50% to about 100%, or about 50% to about 90% w/w, or about 50% to about 80%, or about 50% to about 70%, or about 50% to about 60%; or 60% to about 100%, or about 60% to about 90% w/w, or about 60% to about 80%, or about 60% to about 70%; or about 2% to about 9%, or about 3% to about 8%, or about 4% to about 8%, or about 5% to about 7%; including all values and subranges in between. In embodiments, the third polymer is a PEG polymer. In embodiments, the PEG polymer has a molecular weight in the range of about 10,000 Da to about 15,000 Da (e.g., about 10,000 Da or about 15,000 Da). In embodiments, the PEG polymer includes at least one arm having a functional group located on said arm. In embodiments, the functional group is an amine. In embodiments, the PEG-amine is an 8-arm PEG-amine or a 4-arm PEG-amine.

[0036] In certain embodiments, the biodegradable polymer matrix includes a first polymer, a second polymer, and a third polymer. In aspects, the first polymer, the second polymer, and the third polymer comprise as a % w/w ratio of the pharmaceutical composition: about 1%/99% to about 99%/1%, or about 5%/95% to about 95%/5%, or about 10%/90% to about 90%/10%, or about 15%/85% to about 85%/15%, or about 20%/80% to about 80%/20%, or about 25%/75% to about 75%/25%, or about 30%/70% to about 70%/30%, or about 35%/65% to about 65%/35%, or about 40%/60% to about 60%/40%, or about 45%/55% to about 55%/45%, or about 50%/50%.

[0037] In certain embodiments, the biodegradable polymer matrix includes a third polymer. In aspects, the third polymer comprises as a weight of the biodegradable polymer matrix: about 1 μg to about 1000 μg and about 1 μg to about 1000 μg; or about 1 μg to about 900 μg and about 1 μg to about 900 μg; or about 1 μg to about 800 μg and about 1 μg to about 800 μg; or about 1 μg to about 700 μg and about 1 μg to about 700; or about 1 μg to about 600 μg and about 1 μg to about 600; or about 1 μg to about 500 μg and about 1 μg to about 500; or about 1 μg to about 400 μg and about 1 μg to about 400; or about 1 μg to about 300 μg and about 1 μg to about 300; or about 1 μg to about 200 μg and about 1 μg to about 200; or about 1 μg to about 100 μg and about 1 μg to about 100; or about 1 μg to about 90 μg and about 1 μg to about 90; or about 1 μg to about 80 μg and about 1 μg to about 80; or about 1 μg to about 70 μg and about 1 μg to about 70; or about 1 μg to about 60 μg and about 1 μg to about 60; or about 1 μg to about 50 μg and about 1 μg to about 50; or about 1 μg to about 40 μg and about 1 μg to about 40; or about 1 μg to about 30 μg and about 1 μg to about 30; or about 1 μg to about 20 μg and about 1 μg to about 20; or about 1 μg to about 10 μg and about 1 μg to about 10, including all values and subranges in between. In embodiments, the third polymer is a PEG polymer. In embodiments, the PEG polymer has a molecular weight in the range of about 10,000 Da to about 15,000 Da (e.g., about 10,000 Da or about 15,000 Da). In embodiments, the PEG polymer includes at least one arm having a functional group located on said arm. In embodiments, the functional group is an amine. In embodiments, the PEG-amine is an 8-arm PEG-amine or a 4-arm PEG-amine.

[0038] In embodiments in which the biodegradable polymer matrix includes a first polymer, a second polymer, and a third polymer, said polymers can be present in the biodegradable polymer matrix at the following ratios: from 1:1:1 to 100:1:1 to 1:100:1 to 1:1:100; or from 10:1:1 to 1:10:1 to 1:1:10; or from 5:1:1: to 1:5:1 to 1:1:5; or from 2:1:1 to 1:2:1 to 1:1:2, including all values and subranges in between.

[0039] In embodiments, the biodegradable polymer matrix is composed of gel precursors. In embodiments, the gel precursors comprise at least one PEG-NHS polymer having a molecular weight of about 10,000 Da to about 15,000 Da, and at least one PEG-amine polymer having a molecular weight of about 10,000 Da to about 15,000 Da. In embodiments, the gel precursors comprise at least one PEG-NHS polymer having a

molecular weight of about 10,000 Da and at least one PEG-amine polymer having a molecular weight of about 10,000 Da. In embodiments, the gel precursors comprise at least one PEG-NHS polymer having a molecular weight of about 15,000 Da and at least one PEG-amine polymer having a molecular weight of about 10,000 Da. In embodiments, the gel precursors comprise at least one PEG-NHS polymer having a molecular weight of about 10,000 Da and at least one PEG-amine polymer having a molecular weight of about 15,000 Da. In embodiments, the PEG-NHS polymers can have from about 1 to about 16 arms, e.g., about 4 arms or about 8 arms. In embodiments, the PEG-amine polymers can have from about 1 to about 16 arms, e.g., about 4 arms or about 8 arms.

[0040] In embodiments, the gel precursors comprise: (a) at least one 8-arm PEG-NHS having a molecular weight of from about 10,000-15,000 Da, at least one 4-arm PEG-NHS having a molecular weight of about 10,000-15,000 Da, or a combination thereof; and (b) at least one 8 arm PEG-amine having a molecular weight of about 10,000-15,000 Da, at least one 4 arm PEG-amine having a molecular weight of about 10,000-15,000 Da, or a combination thereof. For example, in embodiments, the gel precursors comprise at least one 8-arm PEG-NHS having a molecular weight of 10,000 Da and at least one 8 arm PEG-amine having a molecular weight of 10,000 Da; or at least one 8-arm PEG-NHS having a molecular weight of 10,000 Da and at least one 4 arm PEG-amine having a molecular weight of 10,000 Da; or at least one 4-arm PEG-NHS having a molecular weight of 10,000 Da and at least one 8 arm PEG-amine having a molecular weight of 10,000 Da; or at least one 8-arm PEG-NHS having a molecular weight of 10,000 Da, at least one 8 arm PEG-amine having a molecular weight of 10,000 Da, and at least one 4 arm PEG-amine having a molecular weight of 10,000 Da; or combinations thereof.

[0041] In embodiments, the pharmaceutical composition comprises as a therapeutic content: about 1 μg to about 100 mg, or about 1 μg to about 10 mg, or about 1 μg to about 1,000 μg, or about 1 μg to about 900 μg, or about 1 μg to about 800 μg, 1 μg to about 700 μg, or about 1 μg to about 600 μg, or about 1 μg to about 500 μg, 1 μg to about 400 μg, 1 μg to about 300 μg, or about 1 μg to about 200 μg, or about 1 μg to about 100 μg, or about 1 μg to about 90 μg, or about 1 μg to about 80 μg, or about 1 μg to about 70 μg, or about 1 μg to about 60 μg, or about 1 μg to about 50 μg, or about 1 μg

to about 40 μg. or about 1 μg to about 40 μg, or about 1 μg to about 30 μg, or about 1 μg to about 20 μg, or about 1 μg to about 10 μg. In embodiments, the therapeutic agent is present in the pharmaceutical composition in an amount: of about 0.001 ng to about 100 μg, or about 1 ng to about 10 μg, or about 1 ng to about 1,000 ng, or about 1 ng to about 900 ng, or about 1 ng to about 800 ng, 1 ng to about 700 ng, or about 1 ng to about 600 ng, or about 1 ng to about 500 ng, 1 ng to about 400 ng, 1 ng to about 300 ng, or about 1 ng to about 200 ng, or about 1 ng to about 100 ng, or about 1 ng to about 90 ng, or about 1 ng to about 80 ng, or about 1 ng to about 70 ng, or about 1 ng to about 60 ng, or about 1 ng to about 50 ng, or about 1 ng to about 40 ng, or about 1 ng to about 40 ng, or about 1 ng to about 30 ng, or about 1 ng to about 20 ng, or about 1 ng to about 10 ng, 0.001 ng to about 1 ng, or about 0.01 ng to about 1 ng, or about 0.1 ng to about 1 ng.

[0042] In embodiments, the therapeutic agent comprises as a weight percent of the pharmaceutical composition: about 1 wt % to about 99 wt %, or about 1 wt % to about 95 wt %, or about 1 wt % to about 90 wt %, or about 1 wt % to about 85 wt %, or about 1 wt % to about 80 wt %, or about 1 wt % to about 75 wt %, or about 1 wt % to about 70 wt %, or about 1 wt % to about 65 wt %, or about 1 wt % to about 60 wt %, or about 1 wt % to about 55 wt %, or about 1 wt % to about 50 wt %, or about 1 wt % to about 45 wt %, or about 1 wt % to about 40 wt %, or about 1 wt % to about 35 wt %, or about 1 wt % to about 30 wt %, or about 1 wt % to about 25 wt %, or about 1 wt % to about 20 wt %, or about 1 wt % to about 15 wt %, or about 1 wt % to about 10 wt %, or about 10 wt % to about 99 wt %, or about 10 wt % to about 95 wt %, or about 10 wt % to about 90 wt %, or about 10 wt % to about 85 wt %, or about 10 wt % to about 80 wt %, or about 10 wt % to about 75 wt %, or about 10 wt % to about 70 wt %, or about 10 wt % to about 65 wt %, or about 10 wt % to about 60 wt %, or about 10 wt % to about 55 wt %, or about 10 wt % to about 50 wt %, or about 10 wt % to about 45 wt %, or about 10 wt % to about 40 wt %, or about 10 wt % to about 35 wt %, or about 10 wt % to about 30 wt %, or about 10 wt % to about 25 wt %, or about 10 wt % to about 20 wt %, or about 10 wt % to about 15 wt %.

[0043] In embodiments, the therapeutic agent comprises as a % w/w of the pharmaceutical composition: about 1 % to about 99 % w/w, or about 1 % to about 95 % w/w, or about 1 % to about 90 % w/w, or about 1 % to about 85 % w/w, or about 1 % to

about 80 % w/w, or about 1 % to about 75 % w/w, or about 1 % to about 70 % w/w, or about 1 % to about 65 % w/w, or about 1 % to about 60 % w/w, or about 1 % to about 55 % w/w, or about 1 % to about 50 % w/w, or about 1 % to about 45 % w/w, or about 1 % to about 40 % w/w, or about 1 % to about 35 % w/w, or about 1 % to about 30 % w/w, or about 1 % to about 25 % w/w, or about 1 % to about 20 % w/w, or about 1 % to about 15 % w/w, or about 1 % to about 10 % w/w, or about 10 % to about 99 % w/w, or about 10 % to about 95 % w/w, or about 10 % to about 90 % w/w, or about 10 % to about 85 % w/w, or about 10 % to about 80 % w/w, or about 10 % to about 75 % w/w, or about 10 % to about 70 % w/w, or about 10 % to about 65 % w/w, or about 10 % to about 60 % w/w, or about 10 % to about 55 % w/w, or about 10 % to about 50 % w/w, or about 10 % to about 45 % w/w, or about 10 % to about 40 % w/w, or about 10 % to about 35 % w/w, or about 10 % to about 30 % w/w, or about 10 % to about 25 % w/w, or about 10 % to about 20 % w/w, or about 10 % to about 15 % w/w.

[0044] In embodiments, the therapeutic agent is formulated as a particle. In some embodiments, the particle comprises as a therapeutic content: about 0.001 ng to about 100 µg, or about 1 ng to about 10 µg, or about 1 ng to about 1,000 ng, or about 1 ng to about 900 ng, or about 1 ng to about 800 ng, 1 ng to about 700 ng, or about 1 ng to about 600 ng, or about 1 ng to about 500 ng, 1 ng to about 400 ng, 1 ng to about 300 ng, or about 1 ng to about 200 ng, or about 1 ng to about 100 ng, or about 1 ng to about 90 ng, or about 1 ng to about 80 ng, or about 1 ng to about 70 ng, or about 1 ng to about 60 ng, or about 1 ng to about 50 ng, or about 1 ng to about 40 ng, or about 1 ng to about 40 ng, or about 1 ng to about 30 ng, or about 1 ng to about 20 ng, or about 1 ng to about 10 ng, 0.001 ng to about 1 ng, or about 0.01 ng to about 1 ng, or about 0.1 ng to about 1 ng.

[0045] In embodiments, the particle comprises as a weight percent of the pharmaceutical composition: about 1 wt % to about 99 wt %, or about 1 wt % to about 95 wt %, or about 1 wt % to about 90 wt %, or about 1 wt % to about 85 wt %, or about 1 wt % to about 80 wt %, or about 1 wt % to about 75 wt %, or about 1 wt % to about 70 wt %, or about 1 wt % to about 65 wt %, or about 1 wt % to about 60 wt %, or about 1 wt % to about 55 wt %, or about 1 wt % to about 50 wt %, or about 1 wt % to about 45 wt %, or about 1 wt % to about 40 wt %, or about 1 wt % to about 35 wt %, or about 1 wt % to about 30 wt %, or about 1 wt % to about 25 wt %, or about 1 wt % to about 20 wt %, or about 1 wt % to

about 15 wt %, or about 1 wt % to about 10 wt %, or about 10 wt % to about 99 wt %, or about 10 wt % to about 95 wt %, or about 10 wt % to about 90 wt %, or about 10 wt % to about 85 wt %, or about 10 wt % to about 80 wt %, or about 10 wt % to about 75 wt %, or about 10 wt % to about 70 wt %, or about 10 wt % to about 65 wt %, or about 10 wt % to about 60 wt %, or about 10 wt % to about 55 wt %, or about 10 wt % to about 50 wt %, or about 10 wt % to about 45 wt %, or about 10 wt % to about 40 wt %, or about 10 wt % to about 35 wt %, or about 10 wt % to about 30 wt %, or about 10 wt % to about 25 wt %, or about 10 wt % to about 20 wt %, or about 10 wt % to about 15 wt %, or about 20 wt % to about 99 wt %, or about 20 wt % to about 95 wt %, or about 20 wt % to about 90 wt %, or about 20 wt % to about 85 wt %, or about 20 wt % to about 80 wt %, or about 20 wt % to about 75 wt %, or about 20 wt % to about 70 wt %, or about 20 wt % to about 65 wt %, or about 20 wt % to about 60 wt %, or about 20 wt % to about 55 wt %, or about 20 wt % to about 50 wt %, or about 20 wt % to about 45 wt %, or about 20 wt % to about 40 wt %, or about 20 wt % to about 35 wt %, or about 20 wt % to about 30 wt %, or about 20 wt % to about 25 wt %.

[0046] In embodiments, the particle comprises as a % w/w of the pharmaceutical composition: about 1 % to about 99 % w/w, or about 1 % to about 95 % w/w, or about 1 % to about 90 % w/w, or about 1 % to about 85 % w/w, or about 1 % to about 80 % w/w, or about 1 % to about 75 % w/w, or about 1 % to about 70 % w/w, or about 1 % to about 65 % w/w, or about 1 % to about 60 % w/w, or about 1 % to about 55 % w/w, or about 1 % to about 50 % w/w, or about 1 % to about 45 % w/w, or about 1 % to about 40 % w/w, or about 1 % to about 35 % w/w, or about 1 % to about 30 % w/w, or about 1 % to about 25 % w/w, or about 1 % to about 20 % w/w, or about 1 % to about 15 % w/w, or about 1 % to about 10 % w/w, or about 10 % to about 99 % w/w, or about 10 % to about 95 % w/w, or about 10 % to about 90 % w/w, or about 10 % to about 85 % w/w, or about 10 % to about 80 % w/w, or about 10 % to about 75 % w/w, or about 10 % to about 70 % w/w, or about 10 % to about 65 % w/w, or about 10 % to about 60 % w/w, or about 10 % to about 55 % w/w, or about 10 % to about 50 % w/w, or about 10 % to about 45 % w/w, or about 10 % to about 40 % w/w, or about 10 % to about 35 % w/w, or about 10 % to about 30 % w/w, or about 10 % to about 25 % w/w, or about 10 % to about 20 % w/w, or about 10 % to about 15 % w/w.

[0047] In embodiments, the therapeutic agent is formulated as a solution comprising one or more excipients. In some embodiments, the solution comprises as a therapeutic agent content: of from about 1 to about 100 g/L, or about 1 to 90 g/L, or about 1 to about 80 g/L, or about 1 to about 70 g/L, or about 1 to about 60 g/L, or about 1 to about 50 g/L, or about 1 to about 40 g/L, or about 1 to about 30 g/L, or about 1 to about 20 g/L, or about 1 to about 10 g/L, 10 to about 100 g/L, or about 10 to 90 g/L, or about 10 to about 80 g/L, or about 10 to about 70 g/L, or about 10 to about 60 g/L, or about 10 to about 50 g/L, or about 10 to about 40 g/L, or about 10 to about 30 g/L, or about 10 to about 20 g/L, or about 20 to about 100 g/L, or about 20 to 90 g/L, or about 20 to about 80 g/L, or about 20 to about 70 g/L, or about 20 to about 60 g/L, or about 20 to about 50 g/L, or about 20 to about 40 g/L, or about 20 to about 30 g/L.

[0048] In some embodiments, the solution comprises as a therapeutic agent content: of from about 1 % w/v to about 90 % w/v, or about 1 % w/v to about 80 % w/v, about 1 % w/v to about 70 % w/v, about 1 % w/v to about 60 % w/v, about 1 % w/v to about 50 % w/v, about 1 % w/v to about 40 % w/v, about 1 % w/v to about 30 % w/v, about 1 % w/v to about 20 % w/v, about 1 % w/v to about 10 % w/v, or about 10 % w/v to about 90 % w/v, or about 10 % w/v to about 80 % w/v, about 10 % w/v to about 70 % w/v, about 10 % w/v to about 60 % w/v, about 10 % w/v to about 50 % w/v, about 10 % w/v to about 40 % w/v, about 10 % w/v to about 30 % w/v, about 10 % w/v to about 20 % w/v, or about 20 % w/v to about 90 % w/v, or about 20 % w/v to about 80 % w/v, about 20 % w/v to about 70 % w/v, about 20 % w/v to about 60 % w/v, about 20 % w/v to about 50 % w/v, about 20 % w/v to about 40 % w/v, about 20 % w/v to about 30 % w/v.

[0049] In embodiments, the therapeutic agent comprises as a % weight of the particle: from about 1 to about 99 % wt, from about 1 to 90 % wt, or about 1 to about 80 % wt, or about 1 to about 70 % wt, or about 1 to about 60 % wt, or about 1 to about 50 % wt, or about 1 to about 40 % wt, or about 1 to about 30 % wt, or about 1 to about 20 % wt, or about 1 to about 10 % wt, or about 1 to about 99 % wt, or about 10 to 90 % wt, or about 10 to about 80 % wt, or about 10 to about 70 % wt, or about 10 to about 60 % wt, or about 10 to about 50 % wt, or about 10 to about 40 % wt, or about 10 to about 30 % wt, or about 10 to about 20 % wt, or about 20 to about 99 % wt, or about 20 to 90 % wt, or about 20 to about 80 % wt, or about 20 to about 70 % wt, or about 20 to about 60 % wt,

or about 20 to about 50 % wt, or about 20 to about 40 % wt, or about 20 to about 30 % wt, or about 30 to about 99 % wt, or about 30 to 90 % wt, or about 30 to about 80 % wt, or about 30 to about 70 % wt, or about 30 to about 60 % wt, or about 30 to about 50 % wt, or about 30 to about 40 % wt, or about 40 to about 99 % wt, or about 40 to 90 % wt, or about 40 to about 80 % wt, or about 40 to about 70 % wt, or about 40 to about 60 % wt, or about 40 to about 50 % wt, or about 50 to about 99 % wt, or about 50 to 90 % wt, or about 50 to about 80 % wt, or about 50 to about 70 % wt, or about 50 to about 60 % wt, or about 60 to about 99 % wt, or about 60 to 90 % wt, or about 60 to about 80 % wt, or about 60 to about 70 % wt, or about 70 to about 99 % wt, or about 70 to 90 % wt, or about 70 to about 80 % wt, or about 80 to about 99 % wt, or about 80 to 90 % wt, or about 90 to about 99 % wt.

[0050] In embodiments, the therapeutic agent is present in the particle in an amount: from about 0.001 μg to about 100 mg, or about 0. 01 μg to about 100 mg, about 0. 1 μg to about 100 mg, about 1 μg to about 100 mg, or about 1 μg to about 10 mg, or about 1 μg to about 1,000 μg, or about 1 μg to about 900 μg, or about 1 μg to about 800 μg, 1 μg to about 700 μg, or about 1 μg to about 600 μg, or about 1 μg to about 500 μg, 1 μg to about 400 μg, 1 μg to about 300 μg, or about 1 μg to about 200 μg, or about 1 μg to about 100 μg, or about 1 μg to about 90 μg, or about 1 μg to about 80 μg, or about 1 μg to about 70 μg, or about 1 μg to about 60 μg, or about 1 μg to about 50 μg, or about 1 μg to about 40 μg. or about 1 μg to about 40 μg, or about 1 μg to about 30 μg, or about 1 μg to about 20 μg, or about 1 μg to about 10 μg.

[0051] In embodiments, the therapeutic agent comprises as a % weight of the PRINT® microparticle (PuP): from about 1 to about 99 % wt, from about 1 to 90 % wt, or about 1 to about 80 % wt, or about 1 to about 70 % wt, or about 1 to about 60 % wt, or about 1 to about 50 % wt, or about 1 to about 40 % wt, or about 1 to about 30 % wt, or about 1 to about 20 % wt, or about 1 to about 10 % wt, or about 1 to about 99 % wt, or about 10 to 90 % wt, or about 10 to about 80 % wt, or about 10 to about 70 % wt, or about 10 to about 60 % wt, or about 10 to about 50 % wt, or about 10 to about 40 % wt, or about 10 to about 30 % wt, or about 10 to about 20 % wt, or about 20 to about 99 % wt, or about 20 to 90 % wt, or about 20 to about 80 % wt, or about 20 to about 70 % wt, or about 20 to about 60 % wt, or about 20 to about 50 % wt, or about 20 to about 40 % wt, or about 20

to about 30 % wt, or about 30 to about 99 % wt, or about 30 to 90 % wt, or about 30 to about 80 % wt, or about 30 to about 70 % wt, or about 30 to about 60 % wt, or about 30 to about 50 % wt, or about 30 to about 40 % wt, or about 40 to about 99 % wt, or about 40 to 90 % wt, or about 40 to about 80 % wt, or about 40 to about 70 % wt, or about 40 to about 60 % wt, or about 40 to about 50 % wt, or about 50 to about 99 % wt, or about 50 to 90 % wt, or about 50 to about 80 % wt, or about 50 to about 70 % wt, or about 50 to about 60 % wt, or about 60 to about 99 % wt, or about 60 to 90 % wt, or about 60 to about 80 % wt, or about 60 to about 70 % wt, or about 70 to about 99 % wt, or about 70 to 90 % wt, or about 70 to about 80 % wt, or about 80 to about 99 % wt, or about 80 to 90 % wt, or about 90 to about 99 % wt.

[0052] In embodiments, the therapeutic agent is present in the PuP in an amount: from about 0.001 μg to about 100 mg, or about 0. 01 μg to about 100 mg, about 0. 1 μg to about 100 mg, about 1 μg to about 100 mg, or about 1 μg to about 10 mg, or about 1 μg to about 1,000 μg, or about 1 μg to about 900 μg, or about 1 μg to about 800 μg, 1 μg to about 700 μg, or about 1 μg to about 600 μg, or about 1 μg to about 500 μg, 1 μg to about 400 μg, 1 μg to about 300 μg, or about 1 μg to about 200 μg, or about 1 μg to about 100 μg, or about 1 μg to about 90 μg, or about 1 μg to about 80 μg, or about 1 μg to about 70 μg, or about 1 μg to about 60 μg, or about 1 μg to about 50 μg, or about 1 μg to about 40 μg. or about 1 μg to about 40 μg, or about 1 μg to about 30 μg, or about 1 μg to about 20 μg, or about 1 μg to about 10 μg.

[0053] In embodiments, the therapeutic agent comprises as a % weight of the pharmaceutical composition: from about 1 to about 99 % wt, from about 1 to 90 % wt, or about 1 to about 80 % wt, or about 1 to about 70 % wt, or about 1 to about 60 % wt, or about 1 to about 50 % wt, or about 1 to about 40 % wt, or about 1 to about 30 % wt, or about 1 to about 20 % wt, or about 1 to about 10 % wt, or about 1 to about 99 % wt, or about 10 to 90 % wt, or about 10 to about 80 % wt, or about 10 to about 70 % wt, or about 10 to about 60 % wt, or about 10 to about 50 % wt, or about 10 to about 40 % wt, or about 10 to about 30 % wt, or about 10 to about 20 % wt, or about 20 to about 99 % wt, or about 20 to 90 % wt, or about 20 to about 80 % wt, or about 20 to about 70 % wt, or about 20 to about 60 % wt, or about 20 to about 50 % wt, or about 20 to about 40 % wt, or about 20 to about 30 % wt, or about 30 to about 99 % wt, or about 30 to 90 % wt,

or about 30 to about 80 % wt, or about 30 to about 70 % wt, or about 30 to about 60 % wt, or about 30 to about 50 % wt, or about 30 to about 40 % wt, or about 40 to about 99 % wt, or about 40 to 90 % wt, or about 40 to about 80 % wt, or about 40 to about 70 % wt, or about 40 to about 60 % wt, or about 40 to about 50 % wt, or about 50 to about 99 % wt, or about 50 to 90 % wt, or about 50 to about 80 % wt, or about 50 to about 70 % wt, or about 50 to about 60 % wt, or about 60 to about 99 % wt, or about 60 to 90 % wt, or about 60 to about 80 % wt, or about 60 to about 70 % wt, or about 70 to about 99 % wt, or about 70 to 90 % wt, or about 70 to about 80 % wt, or about 80 to about 99 % wt, or about 80 to 90 % wt, or about 90 to about 99 % wt.

[0054] In embodiments, the therapeutic agent is present in the implant in an amount: from about 0.001 μg to about 100 mg, or about 0. 01 μg to about 100 mg, about 0. 1 μg to about 100 mg, about 1 μg to about 100 mg, or about 1 μg to about 10 mg, or about 1 μg to about 1,000 μg, or about 1 μg to about 900 μg, or about 1 μg to about 800 μg, 1 μg to about 700 μg, or about 1 μg to about 600 μg, or about 1 μg to about 500 μg, 1 μg to about 400 μg, 1 μg to about 300 μg, or about 1 μg to about 200 μg, or about 1 μg to about 100 μg, or about 1 μg to about 90 μg, or about 1 μg to about 80 μg, or about 1 μg to about 70 μg, or about 1 μg to about 60 μg, or about 1 μg to about 50 μg, or about 1 μg to about 40 μg. or about 1 μg to about 40 μg, or about 1 μg to about 30 μg, or about 1 μg to about 20 μg, or about 1 μg to about 10 μg.

[0055] In embodiments, the therapeutic agent comprises as a % weight of the implant: from about 1 to about 99 % wt, from about 1 to 90 % wt, or about 1 to about 80 % wt, or about 1 to about 70 % wt, or about 1 to about 60 % wt, or about 1 to about 50 % wt, or about 1 to about 40 % wt, or about 1 to about 30 % wt, or about 1 to about 20 % wt, or about 1 to about 10 % wt, or about 1 to about 99 % wt, or about 10 to 90 % wt, or about 10 to about 80 % wt, or about 10 to about 70 % wt, or about 10 to about 60 % wt, or about 10 to about 50 % wt, or about 10 to about 40 % wt, or about 10 to about 30 % wt, or about 10 to about 20 % wt, or about 20 to about 99 % wt, or about 20 to 90 % wt, or about 20 to about 80 % wt, or about 20 to about 70 % wt, or about 20 to about 60 % wt, or about 20 to about 50 % wt, or about 20 to about 40 % wt, or about 20 to about 30 % wt, or about 30 to about 99 % wt, or about 30 to 90 % wt, or about 30 to about 80 % wt, or about 30 to about 70 % wt, or about 30 to about 60 % wt, or about 30 to about 50 %

wt, or about 30 to about 40 % wt, or about 40 to about 99 % wt, or about 40 to 90 % wt, or about 40 to about 80 % wt, or about 40 to about 70 % wt, or about 40 to about 60 % wt, or about 40 to about 50 % wt, or about 50 to about 99 % wt, or about 50 to 90 % wt, or about 50 to about 80 % wt, or about 50 to about 70 % wt, or about 50 to about 60 % wt, or about 60 to about 99 % wt, or about 60 to 90 % wt, or about 60 to about 80 % wt, or about 60 to about 70 % wt, or about 70 to about 99 % wt, or about 70 to 90 % wt, or about 70 to about 80 % wt, or about 80 to about 99 % wt, or about 80 to 90 % wt, or about 90 to about 99 % wt.

[0056] In embodiments, the pharmaceutical compositions described herein are fabricated as implants having dimensions in the range of about 100 μm x 100 μm x 200 μm to about 700 μm x 700 μm x 7,000 μm. In embodiments, the implants have dimensions of about 225 μm (± 50 μm) x 225 μm (± 50 μm) x 2,925 μm (± 100 μm). In embodiments, the implants have dimensions of about 225 μm (± 20%) x 225 μm (± 20%) x 2,925 μm(± 20%). In embodiments, the implants have dimensions of about 225 μm (± 10%) x 225 μm (± 10%) x 2,925 μm(± 10%). In embodiments, the implants have dimensions of about 225 μm (± 5%) x 225 μm (± 5%) x 2,925 μm(± 5%).

[0057] In embodiments, the implants have dimensions of about 311 μm (± 50 μm) x 395 μm (± 50 μm) x 6,045 μm (± 100 μm). In embodiments, the implants have dimensions of about 311 μm (± 20%) x 395 μm (± 20%) x 6,045 μm (±20%). In embodiments, the implants have dimensions of about 311 μm (± 10%) x 395 μm (± 10%) x 6,045 μm (±10%). In embodiments, the implants have dimensions of about 311 μm (± 5%) x 395 μm (± 5%) x 6,045 μm (± 5%).

[0058] In embodiments, the implants have dimensions of about 600 μm (± 50 μm) x 600 μm (± 50 μm) x 1,000 μm (± 100 μm). In embodiments, the implants have dimensions of about 600 μm (± 20%) x 600 μm (± 20%) x 1,000 μm (± 20%). In embodiments, the implants have dimensions of about 600 μm (± 10%) x 600 μm (± 10%) x 1,000 μm (± 10%). In embodiments, the implants have dimensions of about 600 μm (± 5%) x 600 μm (± 5%) x 1,000 μm (± 5%).

[0059] In embodiments, the implants are cylinder shaped having a diameter of about 0.01 mm to about 10 mm and a length of about 0.01 mm to about 10 mm. In embodiments,

the implants have a diameter of about 1 mm (± 20%) and a length of about 3 mm (± 20%). In embodiments, the implants have a diameter of about 1 mm (± 10%) and a length of about 3 mm (± 10%). In embodiments, the implants have a diameter of about 1 mm (± 5%) and a length of about 3 mm (± 5%). In embodiments, the implants have a diameter of about 0.425 mm (± 20%) and a length of about 3 mm (± 20%). In embodiments, the implants have a diameter of about 0.425 mm (± 10%) and a length of about 3 mm (± 10%). In embodiments, the implants have a diameter of about 0.425 mm (± 5%) and a length of about 3 mm (± 5%).

[0060] In embodiments, the therapeutic agent comprises as a % w/w of the mesoparticles: from about 1 to about 99 % w/w, from about 1 to 90 % w/w, or about 1 to about 80 % w/w, or about 1 to about 70 % w/w, or about 1 to about 60 % w/w, or about 1 to about 50 % w/w, or about 1 to about 40 % w/w, or about 1 to about 30 % w/w, or about 1 to about 20 % w/w, or about 1 to about 10 % w/w, or about 1 to about 99 % w/w, or about 10 to 90 % w/w, or about 10 to about 80 % w/w, or about 10 to about 70 % w/w, or about 10 to about 60 % w/w, or about 10 to about 50 % w/w, or about 10 to about 40 % w/w, or about 10 to about 30 % w/w, or about 10 to about 20 % w/w, or about 20 to about 99 % w/w, or about 20 to 90 % w/w, or about 20 to about 80 % w/w, or about 20 to about 70 % w/w, or about 20 to about 60 % w/w, or about 20 to about 50 % w/w, or about 20 to about 40 % w/w, or about 20 to about 30 % w/w, or about 30 to about 99 % w/w, or about 30 to 90 % w/w, or about 30 to about 80 % w/w, or about 30 to about 70 % w/w, or about 30 to about 60 % w/w, or about 30 to about 50 % w/w, or about 30 to about 40 % w/w, or about 40 to about 99 % w/w, or about 40 to 90 % w/w, or about 40 to about 80 % w/w, or about 40 to about 70 % w/w, or about 40 to about 60 % w/w, or about 40 to about 50 % w/w, or about 50 to about 99 % w/w, or about 50 to 90 % w/w, or about 50 to about 80 % w/w, or about 50 to about 70 % w/w, or about 50 to about 60 % w/w, or about 60 to about 99 % w/w, or about 60 to 90 % w/w, or about 60 to about 80 % w/w, or about 60 to about 70 % w/w, or about 70 to about 99 % w/w, or about 70 to 90 % w/w, or about 70 to about 80 % w/w, or about 80 to about 99 % w/w, or about 80 to 90 % w/w, or about 90 to about 99 % w/w.

[0061] In embodiments, the mesoparticle comprises as a therapeutic agent content: about 0.001 mg to about 100 mg, or about 0.01 mg to about 100 mg, or about 0.01 mg to about

90 mg, or about 0.01 mg to about 80 mg, or about 0.01 mg to about 70 mg, or about 0.01 mg to about 60 mg, or about 0.01 mg to about 50 mg, or about 0.01 mg to about 40 mg, or about 0.01 mg to about 30 mg, or about 0.01 mg, to about 20 mg, or about 0.01 mg to about 10 mg, or about 0.01 mg to about 5 mg, or about 0. 1 mg to about 100 mg, or about 0.1 mg to about 90 mg, or about 0.1 mg to about 80 mg, or about 0.1 mg to about 70 mg, or about 0.1 mg to about 60 mg, or about 0.1 mg to about 50 mg, or about 0.1 mg to about 40 mg, or about 0.1 mg to about 30 mg, or about 0.1 mg, to about 20 mg, or about 0.1 mg to about 10 mg, or about 0.1 mg to about 5 mg, or about 0.2 mg to about 5 mg, or about 0.3 mg to about 5 mg, or about 0.4 mg to about 5 mg, or about 0.5 mg to about 5 mg, or about 1 mg to about 4 mg.

[0062] In embodiments, the mesoparticle is fabricated as a delivery vehicle by suspending the mesoparticles in a pharmaceutically acceptable liquid. In embodiments, the liquid delivery vehicle has a volume of from about 1 μL to about 1 mL, or from about 10 μL to about 500 μL, about 10 μL to about 400 μL, about 10 μL to about 300 μL, about 10 μL to about 200 μL, or about 10 μL to about 100 μL.

[0063] In embodiments, the mesoparticles comprise as a % wt of the liquid delivery vehicle: from about 1 to about 99 % wt, from about 1 to 90 % wt, or about 1 to about 80 % wt, or about 1 to about 70 % wt, or about 1 to about 60 % wt, or about 1 to about 50 % wt, or about 1 to about 40 % wt, or about 1 to about 30 % wt, or about 1 to about 20 % wt, or about 1 to about 10 % wt, or about 1 to about 99 % wt, or about 10 to 90 % wt, or about 10 to about 80 % wt, or about 10 to about 70 % wt, or about 10 to about 60 % wt, or about 10 to about 50 % wt, or about 10 to about 40 % wt, or about 10 to about 30 % wt, or about 10 to about 20 % wt, or about 20 to about 99 % wt, or about 20 to 90 % wt, or about 20 to about 80 % wt, or about 20 to about 70 % wt, or about 20 to about 60 % wt, or about 20 to about 50 % wt, or about 20 to about 40 % wt, or about 20 to about 30 % wt, or about 30 to about 99 % wt, or about 30 to 90 % wt, or about 30 to about 80 % wt, or about 30 to about 70 % wt, or about 30 to about 60 % wt, or about 30 to about 50 % wt, or about 30 to about 40 % wt, or about 40 to about 99 % wt, or about 40 to 90 % wt, or about 40 to about 80 % wt, or about 40 to about 70 % wt, or about 40 to about 60 % wt, or about 40 to about 50 % wt, or about 50 to about 99 % wt, or about 50 to 90 % wt, or about 50 to about 80 % wt, or about 50 to about 70 % wt, or about 50 to about 60

% wt, or about 60 to about 99 % wt, or about 60 to 90 % wt, or about 60 to about 80 % wt, or about 60 to about 70 % wt, or about 70 to about 99 % wt, or about 70 to 90 % wt, or about 70 to about 80 % wt, or about 80 to about 99 % wt, or about 80 to 90 % wt, or about 90 to about 99 % wt.

[0064] In embodiments, the liquid delivery vehicle comprises as a mesoparticle content: from about 0.001 μg to about 100 mg, or about 0. 01 μg to about 100 mg, about 0. 1 μg to about 100 mg, about 1 μg to about 100 mg, or about 1 μg to about 10 mg, or about 1 μg to about 1,000 μg, or about 1 μg to about 900 μg, or about 1 μg to about 800 μg, 1 μg to about 700 μg, or about 1 μg to about 600 μg, or about 1 μg to about 500 μg, 1 μg to about 400 μg, 1 μg to about 300 μg, or about 1 μg to about 200 μg, or about 1 μg to about 100 μg, or about 1 μg to about 90 μg, or about 1 μg to about 80 μg, or about 1 μg to about 70 μg, or about 1 μg to about 60 μg, or about 1 μg to about 50 μg, or about 1 μg to about 40 μg. or about 1 μg to about 40 μg, or about 1 μg to about 30 μg, or about 1 μg to about 20 μg, or about 1 μg to about 10 μg.

[0065] In embodiments, the pharmaceutical compositions described herein are fabricated as implants having dimensions in the range of about 10 μm x 10 μm x 10 μm to about 100 μm x 100 μm x 100 μm. In embodiments, the mesoparticles have dimensions of about 12.5 μm (±10 μm) x 12.5 μm (±10 μm) x 25 μm (±10 μm). In embodiments, the mesoparticles have dimensions of about 12.5 μm (± 20%) x 12.5 μm (± 20%) x 25 μm (± 20%). In embodiments, the mesoparticles have dimensions of about 12.5 μm (± 10%) x 12.5 μm (± 10%) x 25 μm (± 10%). In embodiments, the mesoparticles have dimensions of about 12.5 μm (± 5%) x 12.5 μm (± 5%) x 25 μm (± 5%).

[0066] In embodiments, the mesoparticles have dimensions of about 25 μm (±10 μm) x 25 μm (±10 μm) x 25 μm (±10 μm). In embodiments, the mesoparticles have dimensions of about 25 μm (± 20 %) x 25 μm (± 20 %) x 25 μm (± 20%). In embodiments, the mesoparticles have dimensions of about 25 μm (± 10 %) x 25 μm (± 10 %) x 25 μm (± 10%). In embodiments, the mesoparticles have dimensions of about 25 μm (± 5 %) x 25 μm (± 5 %) x 25 μm (± 5%).

[0067] In embodiments, the mesoparticles have dimensions of about 50 μm (±10 μm) x 50 μm (±10 μm) x 50 μm (±10 μm). In embodiments, the mesoparticles have dimensions

of about 50 μm (±20 %) x 50 μm (±20 %) x 50 μm (±20 %). In embodiments, the mesoparticles have dimensions of about 50 μm (±10 %) x 50 μm (±10 %) x 50 μm (±10 %). In embodiments, the mesoparticles have dimensions of about 50 μm (±5 %) x 50 μm (±5 %) x 50 μm (±5 %).

[0068] In embodiments, the pharmaceutical composition comprises a gel comprising an encapsulated therapeutic agent. In embodiments, the gel comprises 20-70 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising: (i) 10-35 ± 5 wt % of at least one PEG-NHS having a molecular weight of about 10,000-15,000 Da; and (ii) 10-35 ± 5 wt % of at least PEG-amine having a molecular weight of about 10,000-15-000 Da. In embodiments, the therapeutic agent comprises about 10-60 ± 5 wt % of the pharmaceutical composition.

[0069] In embodiments, the gel comprises 20 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising: (i) 10 ± 3 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; and (ii) 10 ± 3 wt % of an 8 arm PEG-amine having a molecular weight of 10,000 Da. In embodiments, the therapeutic agent comprises about 35 ± 5 wt % of the pharmaceutical composition. In embodiments, the therapeutic agent is fabricated as a micronized particle. In embodiments, the micronized particle is a PuP having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimensions. The pharmaceutical composition of claim 137, wherein pharmaceutical composition is a rod-shaped implant having dimensions of 225 μm x 225 μm x 2925 μm (L x W x H) ± 10% of any dimension. In embodiments, the pharmaceutical composition is a rod-shaped implant having dimensions of 311 μm x 395 μm x 6045 μm (L x W x H) ± 10% of any dimension, or 600 μm x 600 μm x 1000 μm (L x W x H) ± 10% of any dimension. In embodiments, the pharmaceutical composition is a cylindrical implant having a diameter of 1 mm ± 10% and length of 3 mm ± 10%. In embodiments, the pharmaceutical composition is a mesoparticle having dimensions of about 12.5 μm x 12.5 μm x 25 μm ± 10% in any dimension, or about 25 μm x 25 μm x 25 μm ± 10% in any dimension, or about 50 μm x 50 μm x 50 μm ± 10% in any dimension. In embodiments, the mesoparticle is suspended in about 1-500 μL of a pharmaceutically acceptable liquid. In embodiments, the therapeutic agent is aflibercept

or bevacizumab. In embodiments, the pharmaceutical composition is administered in a 27 gauge needle or smaller.

[0070] In embodiments, the gel comprises 20 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising: (i) 10 ± 3 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; (ii) 5 ± 3 wt % of an 8-arm PEG-amine having a molecular weight of 10,000 Da; and (iii) 5 ± 3 wt % of a 4-arm PEG-amine having a molecular weight of 10,000 Da; and wherein the therapeutic agent comprises about 36 ± 5 wt % of the pharmaceutical composition. In embodiments, the therapeutic agent is fabricated as a micronized particle. In embodiments, the micronized particle is a PuP having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimensions. In embodiments, the pharmaceutical composition is a rod-shaped implant having dimensions of 225 μm x 225 μm x 2925 μm (L x W x H) ± 10% of any dimension. In embodiments, the pharmaceutical composition is a rod-shaped implant having dimensions of 311 μm x 395 μm x 6045 μm (L x W x H) ± 10% of any dimension, or 600 μm x 600 μm x 1000 μm (L x W x H) ± 10% of any dimension. In embodiments, the pharmaceutical composition is a cylindrical implant having a diameter of 1 mm ± 10% and length of 3 mm ± 10%. In embodiments, the pharmaceutical composition is a mesoparticle having dimensions of about 12.5 μm x 12.5 μm x 25 μm ± 10% in any dimension, or about 25 μm x 25 μm x 25 μm ± 10% in any dimension, or about 50 μm x 50 μm x 50 μm ± 10% in any dimension. In embodiments, the mesoparticle is suspended in about 1-500 μL of a pharmaceutically acceptable liquid. In embodiments, the therapeutic agent is aflibercept or bevacizumab. In embodiments, the pharmaceutical composition is administered in a 27 gauge needle or smaller.

[0071] In embodiments, the gel comprises 21 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising: (i) 10 ± 5 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; and (ii) 10 ± 5 wt % of an 8 arm PEG-amine having a molecular weight of 10,000 Da; and wherein the therapeutic agent comprises about 46 ± 5 wt % of the pharmaceutical composition. In embodiments, the therapeutic agent is fabricated as a micronized particle. In embodiments, the micronized particle is a PuP having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimensions. The pharmaceutical composition of claim 137, wherein

pharmaceutical composition is a rod-shaped implant having dimensions of 225 μm x 225 μm x 2925 μm (L x W x H) ± 10% of any dimension. In embodiments, the pharmaceutical composition is a rod-shaped implant having dimensions of 311 μm x 395 μm x 6045 μm (L x W x H) ± 10% of any dimension, or 600 μm x 600 μm x 1000 μm (L x W x H) ± 10% of any dimension. In embodiments, the pharmaceutical composition is a cylindrical implant having a diameter of 1 mm ± 10% and length of 3 mm ± 10%. In embodiments, the pharmaceutical composition is a mesoparticle having dimensions of about 12.5 μm x 12.5 μm x 25 μm ± 10% in any dimension, or about 25 μm x 25 μm x 25 μm ± 10% in any dimension, or about 50 μm x 50 μm x 50 μm ± 10% in any dimension. In embodiments, the mesoparticle is suspended in about 1-500 μL of a pharmaceutically acceptable liquid. In embodiments, the therapeutic agent is aflibercept or bevacizumab. In embodiments, the pharmaceutical composition is administered in a 27 gauge needle or smaller.

[0072] In embodiments, the gel comprises 50 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising: (i) 25 ± 5 wt % of an 8-arm PEG-NHS having a molecular weight of 15,000 Da; (ii) 25 ± 5 wt % of an 8-arm PEG-amine having a molecular weight of 10,000 Da; and wherein the therapeutic agent comprises about 22 ± 5 wt % of the pharmaceutical composition. In embodiments, the therapeutic agent is fabricated as a micronized particle. In embodiments, the micronized particle is a PuP having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimensions. In embodiments, the pharmaceutical composition is a rod-shaped implant having dimensions of 225 μm x 225 μm x 2925 μm (L x W x H) ± 10% of any dimension. In embodiments, the pharmaceutical composition is a rod-shaped implant having dimensions of 311 μm x 395 μm x 6045 μm (L x W x H) ± 10% of any dimension, or 600 μm x 600 μm x 1000 μm (L x W x H) ± 10% of any dimension. In embodiments, the pharmaceutical composition is a cylindrical implant having a diameter of 1 mm ± 10% and length of 3 mm ± 10%. In embodiments, the pharmaceutical composition is a mesoparticle having dimensions of about 12.5 μm x 12.5 μm x 25 μm ± 10% in any dimension, or about 25 μm x 25 μm x 25 μm ± 10% in any dimension, or about 50 μm x 50 μm x 50 μm ± 10% in any dimension. In embodiments, the mesoparticle is suspended in about 1-500 μL of a pharmaceutically acceptable liquid. In

embodiments, the therapeutic agent is bevacizumab. In embodiments, the pharmaceutical compositions is administered in a 27 gauge needle or smaller.

[0073] In embodiments, the gel comprises 50 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising: (i) 25 ± 5 wt % of an 4-arm PEG-NHS having a molecular weight of 10,000 Da; (ii) 25 ± 5 wt % of an 4-arm PEG-amine having a molecular weight of 10,000 Da; and wherein the therapeutic agent comprises about 18 ± 5 wt % of the pharmaceutical composition. In embodiments, the therapeutic agent is fabricated as a micronized particle. In embodiments, the micronized particle is a PuP having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimensions. In embodiments, the pharmaceutical composition is a rod-shaped implant having dimensions of 225 μm x 225 μm x 2925 μm (L x W x H) ± 10% of any dimension. In embodiments, the pharmaceutical composition is a rod-shaped implant having dimensions of 311 μm x 395 μm x 6045 μm (L x W x H) ± 10% of any dimension, or 600 μm x 600 μm x 1000 μm (L x W x H) ± 10% of any dimension. In embodiments, the pharmaceutical composition is a cylindrical implant having a diameter of 1 mm ± 10% and length of 3 mm ± 10%. In embodiments, the pharmaceutical composition is a mesoparticle having dimensions of about 12.5 μm x 12.5 μm x 25 μm ± 10% in any dimension, or about 25 μm x 25 μm x 25 μm ± 10% in any dimension, or about 50 μm x 50 μm x 50 μm ± 10% in any dimension. In embodiments, the mesoparticle is suspended in about 1-500 μL of a pharmaceutically acceptable liquid. In embodiments, the therapeutic agent is bevacizumab. In embodiments, the pharmaceutical compositions is administered in a 27 gauge needle or smaller.

[0074] In embodiments, the pharmaceutical composition is a mixture of a first pharmaceutical composition and a second pharmaceutical composition, wherein: (i) the first pharmaceutical composition comprises a first gel comprising 21 ± 5 % wt of the first pharmaceutical composition and contains a mixture of polymers comprising: (a) 10 ± 5 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; and (b) 10 ± 5 wt % of an 4 arm PEG-amine having a molecular weight of 10,000 Da; and wherein a first therapeutic agent comprises about 36 ± 5 wt % of the first pharmaceutical composition; and (ii) the second composition comprises a second gel comprising 21 ± 5 % wt of the first pharmaceutical composition and contains a mixture of polymers comprising: (a) 10 ±

5 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; and (b) 10 ± 5 wt % of an 8 arm PEG-amine having a molecular weight of 10,000 Da; and wherein a second therapeutic agent comprises about 36 ± 5 wt % of the second pharmaceutical composition. In embodiments, the first and second therapeutic agents are fabricated as a micronized particle. In embodiments, the micronized particle is a PuP having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimensions. The pharmaceutical composition of claim 137, wherein pharmaceutical composition is a rod-shaped implant having dimensions of 225 μm x 225 μm x 2925 μm (L x W x H) ± 10% of any dimension. In embodiments, the pharmaceutical composition is a rod-shaped implant having dimensions of 311 μm x 395 μm x 6045 μm (L x W x H) ± 10% of any dimension, or 600 μm x 600 μm x 1000 μm (L x W x H) ± 10% of any dimension. In embodiments, the pharmaceutical composition is a cylindrical implant having a diameter of 1 mm ± 10% and length of 3 mm ± 10%. In embodiments, the pharmaceutical composition is a mesoparticle having dimensions of about 12.5 μm x 12.5 μm x 25 μm ± 10% in any dimension, or about 25 μm x 25 μm x 25 μm ± 10% in any dimension, or about 50 μm x 50 μm x 50 μm ± 10% in any dimension. In embodiments, the mesoparticle is suspended in about 1-500 μL of a pharmaceutically acceptable liquid. In embodiments, the therapeutic agent is aflibercept or bevacizumab. In embodiments, the pharmaceutical compositions is administered in a 27 gauge needle or smaller.

[0075] In embodiments, the disclosure provides for a method for making an ocular sustained-release pharmaceutical composition for the delivery of a therapeutic agent to an eye of a subject in need thereof, comprising: formulating a solution comprising a therapeutic agent and one or more excipients; forming, from the solution, particles comprising the therapeutic agent and one or more excipients; combining the particles with gel precursors in a solvent to form a suspension comprising the particles and the gel precursors, wherein the solvent is an organic and hydrophilic solvent; and allowing the gel precursors to gel around the particles, thereby forming a gel comprising encapsulated particles; and thereby forming the pharmaceutical composition for sustained delivery of a therapeutic agent. In embodiments, the therapeutic agent does not substantially aggregate in the presence of the hydrophilic solvent.

[0076] In embodiments, the the solution comprising the therapeutic agent is added to a mold to thereby form the particles comprising the therapeutic agent and one or more excipients. In embodiments, the mold has a largest dimension of less than or equal to about 10 μm. In embodiments, the mold has dimensions of about 1 μm in diameter x about 1 μm in height . In embodiments, the particles formed utilizing the mold have dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension.

[0077] In embodiments, the particles may be formed via lyophilizing, spray drying, or jet milling the solution to thereby form the particles. In such embodiments, the particles have a largest dimension in the range of about 1 μm to about 25 μm.

[0078] In embodiments, the gel is a biodegradable gel. In embodiments, the gel is formed by covalently crosslinking the gel precursors. In embodiments, the gel comprises crosslinked polyethylene glycol polymers. In embodiments, the gel precursors comprise a first precursor having a first functional group and a second precursor having a second functional group, wherein the first functional group and the second functional group are reactive in the solvent. In embodiments, the gel precursors comprise a first precursor having a first functional group and a second precursor having a second functional group, wherein the first functional group interacts with the second functional group thereby forming a covalent bond.

[0079] In embodiments, the gel precursors comprise PEG polymers. In embodiments, the gel precursors comprise PEG polymers having a molecular weight in the range of about 10,000 Da to about 15,000 Da. In embodiments, the gel precursors comprise at least one PEG polymer having an N-hydroxy succinimidyl ester functional group (PEG-NHS) and at least one PEG polymer having an amine functional group (PEG-amine). In embodiments, the gel precursors comprise at least one PEG-NHS polymer having a molecular weight range of about 10,000 Da to about 15,000 Da, and at least one PEG-amine polymer having a molecular weight range of about 10,000 Da to about 15,000 Da. In embodiments, the gel precursors comprise at least one PEG-NHS polymer having a molecular weight of about 10,000 Da and at least one PEG-amine polymer having a molecular weight of about 10,000 Da.

[0080] The method of claim 1, wherein the gel is formed in a mold. In embodiments, the gel is formed in a PRINT® mold and wherein the solvent is ethyl lactate. In embodiments, the gel is formed in a cylindrical mold. In embodiments, the gel is formed in a cylindrical mold having a dimeter of about 1 mm, and wherein the solvent is acetonitrile.

[0081] In embodiments, the methods further comprise removing the solvent from the gel thereby forming a xerogel. In embodiments, the removing comprises lyophilizing the gel, thereby forming the xerogel.

[0082] In embodiments, the methods further comprise hydrating the gel in an aqueous solution thereby forming a hydrogel. In embodiments, the hydrating occurs in-vivo after administration to the eye of a subject in need thereof.

[0083] In embodiments, the therapeutic agent is a water soluble biologic. In embodiments, wherein the therapeutic agent is a protein, peptide, antibody. In embodiments, the therapeutic agent is a protein. In embodiments, the therapeutic agent is an anti-VEGF agent. In embodiments, the therapeutic agent is bevacizumab, ranibizumab, aflibercept, or pegaptanib. In embodiments, the therapeutic agent is bevacizumab. In embodiments, the therapeutic agent is aflibercept. In embodiments, the therapeutic agent is an antibody. In embodiments, the therapeutic agent is an anti-VEFG antibody.

[0084] In embodiments, therapeutic agent is bevacizumab, the bevacizumab is formed as a micronized particle having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, the solvent is ethyl lactate, and the gel comprises cross-linked PEG polymers.

[0085] In embodiments, therapeutic agent is bevacizumab, the bevacizumab is formed as a micronized particle having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, the solvent is ethyl lactate, the gel comprises cross-linked PEG polymers, and the pharmaceutical composition is a mesoparticle.

[0086] In embodiments, wherein therapeutic agent is bevacizumab, the bevacizumab is formed as a micronized particle having dimensions of 1 μm in diameter x 1 μm in height

± 10% in any dimension, the solvent is ethyl lactate, the gel comprises cross-linked PEG polymers, and the pharmaceutical composition is an implant.

[0087] In embodiments, therapeutic agent is aflibercept, the aflibercept is formed as a micronized particle having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, the solvent is ethyl lactate, and the gel comprises cross-linked PEG polymers.

[0088] In embodiments, the therapeutic agent is aflibercept, the aflibercept is formed as a micronized particle having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, the solvent is ethyl lactate, the gel comprises cross-linked PEG polymers, and the pharmaceutical composition is a mesoparticle.

[0089] In embodiments, therapeutic agent is aflibercept, the aflibercept is formed as a micronized particle having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, the solvent is ethyl lactate, the gel comprises cross-linked PEG polymers, and the pharmaceutical composition is an implant.

[0090] In certain embodiments, the disclosure provides for methods for making an ocular sustained-release pharmaceutical composition for the delivery of a therapeutic agent to an eye of a subject in need thereof, comprising: formulating a solution comprising a therapeutic agent and one or more excipients; fabricating solid state microparticles (PuPs) comprising the therapeutic agent; combining the particles with gel precursors in a solution to form a suspension comprising the particles and the gel precursors, wherein the solvent is an organic and hydrophilic solvent; and allowing the gel precursors to gel around the particles, thereby forming a gel comprising encapsulated particles; thereby forming the pharmaceutical composition for sustained delivery of a therapeutic agent.

[0091] In embodiments, the disclosure provides for sustained release delivery systems prepared by a process comprising the steps of: formulating a solution comprising a therapeutic agent and one or more excipients; forming, from the solution, particles comprising the therapeutic agent and one or more excipients; combining the particles with gel precursors in a solvent to form a suspension comprising the particles and the gel precursors, wherein the solvent is an organic and hydrophilic solvent; and allowing the gel precursors to gel around the particles, thereby forming a gel comprising encapsulated

particles; and thereby forming the pharmaceutical composition for sustained delivery of a therapeutic agent, wherein the pharmaceutical composition comprises: (A) a gel comprising a biocompatible polymer matrix; and (B) and at least one protein microparticle (PuP) comprising a therapeutic agent and at least one pharmaceutically acceptable excipient; wherein the PuP has a largest dimension of less than 10 μm; wherein the pharmaceutical composition is formulated to deliver the therapeutic agent for at least about 4 months. In embodiments, the PuP has dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension. In embodiments, the pharmaceutical composition can be an implant (e.g., rod-shaped implant or cylindrical implant) or a mesoparticle. In embodiments, the therapeutic agent bevacizumab, ranibizumab, aflibercept, or pegaptanib.

[0092] In embodiments, the pharmaceutical composition described herein are engineered such that the therapeutic agent does not substantially aggregate. Accordingly, in embodiments, the protein comprises % monomer at least about 90%, e.g., 91%, 92%, 93%, 94%, 96%, 97%, 98%, or 99%. In embodiments, the pharmaceutical compositions are formulated with excipients which prevent substantial aggregation of proteins. In some embodiments, the excipients may be an amino acid. In certain embodiments, the amino acid is histidine, leucine and trileucine.

[0093] In embodiments, the disclosure provides for method of treating an ocular condition associated with abnormal vessel growth in the eye of a patient in need thereof comprising: administering to the vitreous of an eye of a patient in need thereof an ocular sustained-release pharmaceutical composition comprising: (A) a gel comprising a biocompatible polymer matrix; and (B) at least one therapeutic agent fabricated as a microparticle having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension; and wherein the gel encapsulates the therapeutic agent, and wherein the ocular sustained-release pharmaceutical composition is formulated to release the therapeutic agent in the vitreous for at least about 4 months. In embodiments, the pharmaceutical composition can be an implant (e.g., rod-shaped implant or cylindrical implant) or a mesoparticle. In embodiments, the therapeutic agent bevacizumab, ranibizumab, aflibercept, or pegaptanib. In certain embodiments, the therapeutic agent is aflibercept and aflibercept is administered in an amount of less than or equal to about 2

mg, e.g., less than or equal to about 850 μg. In embodiments, the therapeutic agent maintained for about 4 months to about 6 months. In embodiments, the abnormal vessel growth is Retinal Vein Occlusion (RVO), Age-Related Macular Degeneration (AMD), or Diabetic Macular Edema (DME).

[0094] In embodiments, the disclosure provides for methods of maintaining an ocular vitreous concentration of an anti-Vascular Endothelial Growth Factor (anti-VEGF) agent of greater than or equal to about 1 ng/mL, comprising: administering an ocular sustained-release pharmaceutical composition to the vitreous of an eye of a human in need thereof, wherein said ocular sustained-release pharmaceutical composition comprises: (A) a gel comprising a biocompatible polymer matrix; and (B) at least one a therapeutic agent fabricated as a microparticle having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension; and wherein the vitreous concentration of the anti-VEGF agent in said human's eye is maintained at a concentration greater than or equal to about 1 ng/mL for at least about 3 months. In embodiments, the anti-VEGF agent is bevacizumab, ranibizumab, aflibercept, or pegaptanib. In embodiments, the anti-VEGF agent is aflibercept and wherein the ocular sustained-release pharmaceutical composition comprises less than or equal to about 2 mg of aflibercept, e.g., less than or equal to about 850 μg. In embodiments, the vitreous concentration of the anti-VEGF agent in said human's eye is maintained at a concentration greater than or equal to about 1 ng/mL for at least about 4 months or for about 4 months to about 6 months. In embodiments, the vitreous concentration of the anti-VEGF agent in said human's eye is from about 1 ng/mL to about 10 ng/mL. In embodiments, the vitreous concentration of the anti-VEGF agent in said human's eye is greater than or equal to about 10 ng/mL.

[0095] In embodiments, disclosure provides for an ocular, sustained-release pharmaceutical composition comprising: (A) a gel comprising a biocompatible polymer matrix; and (B) at least one protein microparticle (PuP) comprising bevacizumab and at least one pharmaceutically acceptable excipient; wherein the gel encapsulates the PuP; wherein the PuP has dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension; and wherein the pharmaceutical composition is formulated to achieve a score less than the negative control as determined with standardized fluorescein angiogram scoring using fluorescein angiography when administered to an eye of a rabbit. In

embodiments, the ocular, sustained-release pharmaceutical composition is fabricated as a cylindrical implant having a length of about 0.425 mm ± 10% and a dimeter of about 3 mm ± 10%.

BRIEF DESCRIPTION OF FIGURES

[0096] FIG. 1 illustrates a method of making a sustained release implant comprising a therapeutic agent.

[0097] FIG. 2 schematically illustrates a method of making a sustained release pharmaceutical composition comprising a micronized therapeutic agent.

[0098] FIG. 3 schematically illustrates a method of making gel comprising PEG precursors.

[0099] FIG. 4A-F shows the in-vitro release and release rate of aflibercept from implants in Example 12&13; **FIG. 4A** shows the in-vitro release of aflibercept from ENV-1P-0205-46-A measured with an ELISA; **FIG. 4B** shows the release rates of aflibercept from ENV-1P-0205-46-A measured with an ELISA; **FIG. 4C** shows the in-vitro release of aflibercept from ENV-1P-0205-46-A measured with a Bradford Assay; **FIG. 4D** shows the release rates of aflibercept from ENV-1P-0205-46-A measured with a Bradford Assay; **FIG. 4E** shows the in-vitro release of aflibercept from RES-PRO-0021-22-A, RES-PRO-0021-22-B, and RES-PRO-0021-22-C measured with an ELISA; **FIG. 4F** shows the release rate of aflibercept from RES-PRO-0021-22-A, RES-PRO-0021-22-B, and RES-PRO-0021-22-C measured with an ELISA; **FIG. 4G** shows the in-vitro release of aflibercept from RES-PRO-0021-68-A, RES-PRO-0021-68-B, RES-PRO-0021-68-C, and RES-PRO-0021-68-D measured with an ELISA; **FIG. 4H** shows the release rate of aflibercept from RES-PRO-0021-68-A, RES-PRO-0021-68-B, RES-PRO-0021-68-C, and RES-PRO-0021-68-D measured with an ELISA; **FIG. 4I** shows the in-vitro release of aflibercept from RES-PRO-0021-68-A, RES-PRO-0021-68-B, RES-PRO-0021-68-C, and RES-PRO-0021-68-D measured with a Bradford Assay; **FIG. 4J** shows the release rate of aflibercept from RES-PRO-0021-68-A, RES-PRO-0021-68-B, RES-PRO-0021-68-C, and RES-PRO-0021-68-D measured with a Bradford Assay; **FIG. 4K** shows in-vitro release of aflibercept from RES-PRO-0021-68-G, RES-PRO-0021-68-

H, and RES-PRO-0021-68-I measured with an ELISA; and **FIG. 4L** shows the release rate of aflibercept from RES-PRO-0021-68-G, RES-PRO-0021-68-H, and RES-PRO-0021-68-I measured with an ELISA.

[00100] **FIG. 5A-B** shows the in-vitro release of bevacizumab from implants in Example 17&18 measured with an ELISA. **FIG. 5A** shows the in-vitro release of bevacizumab from ENV-1A-0050-09-E, ENV-1A-0050-09-G, ENV-1A-0050-09-H, ENV-1A-0050-09-K, ENV-1A-0050-09-L, ENV-1A-0050-09-O, ENV-1A-0050-29-B, ENV-1A-0089-14-B, and ENV-1A-0089-14-C. **FIG. 5B** shows the in-vitro release of bevacizumab from ENV-1A-0089-14-C, ENV-1A-0103-03-D, and ENV-1A-0084-32-I.

[00101] **FIG. 6A-B** shows scanning electron micrographs (SEM) of a PRINT® cylindrical implants. **FIG. 6A** shows a low resolution SEM of a PRINT cylindrical implant containing 1 μm x 1 μm PRINT protein microparticles. **FIG. 6B** shows a high resolution SEM of the surface of the PRINT cylindrical implant showing the 1 μm x 1 μm PRINT bevacizumab microparticles.

[00102] **FIG. 7A-B** shows scanning electron micrographs (SEM) of a PRINT® rod shaped implant. **FIG. 7A** shows an SEM of a PRINT rod shaped implant containing 1 μm x 1 μm PRINT protein microparticles. **FIG. 7B** shows an SEM of the same implant loaded in a 27 Gauge needle.

[00103] **FIG. 8A-B** shows scanning electron micrographs (SEM) of PRINT® protein mesoparticles. **FIG. 8A** shows a low resolution SEM of PRINT® mesoparticles containing 1 μm x 1 μm PRINT® bevacizumab microparticles. **FIG. 8B** shows a high resolution SEM of the surface of the PRINT® mesoparticles showing the 1 μm x 1 μm PRINT bevacizumab microparticles.

[00104] **FIG. 9** shows a scanning electron micrographs (SEM) of the surface of the 1 μm x 1 μm PRINT aflibercept microparticles.

[00105] **FIG. 10** shows a low resolution scanning electron micrograph (SEM) of a PRINT cylindrical implant containing lyophilized aflibercept micronized particles.

[00106] **FIG. 11** shows a cylindrical PRINT® implant with aflibercept PRINT® microparticles. **FIG. 11A** shows low resolution SEM of a PRINT® cylindrical implant

containing 1 μm x 1 μm PRINT® aflibercept microparticles. **FIG. 11B** shows a high resolution SEM of a cross section of the PRINT® cylindrical implant showing the 1 μm x 1 μm PRINT® aflibercept microparticles.

[00107] **FIG. 12** depicts the results from a Size Exclusion Chromatography experiment with unprocessed aflibercept and PRINT® aflibercept.

[00108] **FIG. 13** depicts the results from a Size Exclusion Chromatography experiment with unprocessed bevacizumab and PRINT® bevacizumab.

[00109] **FIG. 14A** shows the in-vitro release of bevacizumab from a cylindrical PRINT implant containing 1 μm x 1 μm PRINT® bevacizumab microparticles.

[00110] **FIG. 14B** shows the in-vivo (IVT, rabbit) release bevacizumab from a cylindrical PRINT implant containing 1 μm x 1 μm PRINT® bevacizumab microparticles.

[00111] **FIG. 15** depicts the total ocular examination score over the course of the 12 days of study of certain embodiments and samples.

[00112] **FIG. 16** depicts the bevacizumab content in the aqueous humor at the termination of the study on day 12.

[00113] **FIG. 17** depicts the total ocular examination score over the course of the study of certain embodiments and samples.

[00114] **FIG. 18** depicts the bevacizumab content in the aqueous humor at the termination of the study on day 28.

[00115] **FIG. 19A** depicts the leakage score tested according to Example 1 post challenge on day 14.

[00116] **FIG. 19B** depicts the leakage score tested according to Example 1 post challenge on day 28.

[00117] **FIG. 20** depicts the total ocular examination score over the course of the study of certain embodiments and samples.

[00118] **FIG. 21** depicts the bevacizumab content in the aqueous humor at the termination of the study on day 28.

[00119] **FIG. 22A** depicts the leakage score tested according to Example 1 post challenge on day 28.

[00120] **FIG. 22B** depicts the leakage score tested according to Example 1 post challenge on day 56.

[00121] **FIG. 23** illustrates plasma concentration data from ENV1305-PRE-001: A Pharmacokinetics Study of ENV1305 in the African Green Monkey.

[00122] **FIG. 24** illustrates aqueous Humor concentration data from ENV1305-PRE-001: A Pharmacokinetics Study of ENV1305 in the African Green Monkey.

[00123] **FIG. 25** illustrates vitreous humor concentration data from ENV1305-PRE-001: A Pharmacokinetics Study of ENV1305 in the African Green Monkey.

[00124] **FIG. 26A-B** shows the in-vitro release of aflibercept from implants in Example 40. **FIG. 26A** shows the in-vitro release of aflibercept from RES-PRO-0041-44-A, RES-PRO-0041-44-B, RES-PRO-0041-44-C, RES-PRO-0041-44-D, RES-PRO-0041-44-E. **FIG. 26B** shows the in-vitro release rate of aflibercept from RES-PRO-0041-44-A, RES-PRO-0041-44-B, RES-PRO-0041-44-C, RES-PRO-0041-44-D, RES-PRO-0041-44-E.

[00125] **FIG. 27** illustrates the amount of protein monomer release from ENV-1P-0205-91-D, RES-PRO-0041-44-A, RES-PRO-0041-44-B, RES-PRO-0041-44-C, RES-PRO-0041-44-D, RES-PRO-0041-44-E.

DETAILED DESCRIPTION

[00126] Described herein are pharmaceutical compositions for delivering therapeutic agents to an eye of a subject. Said pharmaceutical composition may be used for treating or preventing an ocular condition. In embodiments, the pharmaceutical compositions can be formulated for the sustained delivery of a therapeutic agent. In embodiments, the pharmaceutical composition comprises: a biocompatible polymer matrix and a therapeutic agent, which is included in the polymer matrix. The therapeutic agent may be encapsulated in the polymer matrix.

[00127] As described herein, multiple pharmaceutical compositions have been fabricated and/or contemplated in the form of an implant or mesoparticle, resulting in highly effective pharmaceutically active products, including ocular therapeutic treatments, including sustained release ocular implants.

[00128] In various embodiments, these pharmaceutical compositions include a therapeutic agent dispersed throughout a polymer matrix formed into an ocular implant. In embodiments, the polymer matrix forms a gel which encapsulates the therapeutic agent. In some aspects, the ocular implant degrades slowly in a human eye and gradually, or in a sustained way, releases the active therapeutic agent.

[00129] In embodiments, the pharmaceutical composition of the present disclosure comprises: i) a biocompatible polymer or polymers, and ii) a therapeutic agent, for example, a protein effective for use in the prevention and/or treatment of an ocular condition, such as post-operative inflammation and/or pain. In particle embodiments, the biocompatible polymers form a gel which entraps the therapeutic agent, thereby forming delivery system. In particular embodiments, the gel is formulated for the sustained release of the therapeutic agent to the eye of a subject in need thereof.

[00130] In embodiments, the therapeutic agent is a protein or peptide which can be used to treat an ocular condition. In other embodiments, the therapeutic agent is an antibody which can be used to treat an ocular condition. In further embodiments, the therapeutic agent is a small molecule which can be used to treat an ocular condition.

[00131] Also described herein are methods for making pharmaceutical compositions comprising therapeutic agent. In embodiments, the methods include forming a micronized particle comprising the therapeutic agent. The micronized particle is then entrapped in a gel, and the gel comprising the entrapped micronized particle is fabricated as an implant.

[00132] As shown in **FIG. 1** shown, embodiments of these methods include forming solid state micronized particles comprising the therapeutic agent. In embodiments, the solid state micronized particle is fabricated in a mold of desired dimensions utilizing PRINT® technology. In other embodiments, the micronized particle is prepared via lyophilizing or spray drying a solution comprising the therapeutic agent.

The particles are mixed with gel precursors in a solution and allowed to react under appropriate conditions such that a gel is formed which encapsulates the therapeutic agent particles. In embodiments, the gel can be formed in a mold having desired dimensions to produce an implant. In particular embodiments, the mold is a PRINT mold (e.g., to fabricate an implant or a mesoparticle as disclosed herein) or a cylindrical mold. In embodiments, the implant is dehydrated and stored or administered. In embodiments, the gels or implants described herein can be hydrated in an aqueous solution to form hydrogels.

[00133] DEFINITIONS

[00134] "About" means plus or minus a percent (e.g., ±5%) of the number, parameter, or characteristic so qualified, which would be understood as appropriate by a skilled artisan to the scientific context in which the term is utilized. Furthermore, since all numbers, values, and expressions referring to quantities used herein, are subject to the various uncertainties of measurement encountered in the art, then unless otherwise indicated, all presented values may be understood as modified by the term "about."

[00135] As used herein, the articles "a," "an," and "the" may include plural referents unless otherwise expressly limited to one-referent, or if it would be obvious to a skilled artisan from the context of the sentence that the article referred to a singular referent.

[00136] Where a numerical range is disclosed herein, then such a range is continuous, inclusive of both the minimum and maximum values of the range, as well as every value between such minimum and maximum values. Still further, where a range refers to integers, every integer between the minimum and maximum values of such range is included. In addition, where multiple ranges are provided to describe a feature or characteristic, such ranges can be combined. That is to say that, unless otherwise indicated, all ranges disclosed herein are to be understood to encompass any and all subranges subsumed therein. For example, a stated range of from "1 to 10" should be considered to include any and all subranges between the minimum value of 1 and the maximum value of 10. Exemplary subranges of the range "1 to 10" include, but are not limited to, 1 to 6.1, 3.5 to 7.8, and 5.5 to 10.

[00137] As used herein, the term "polymer" is meant to encompass both homopolymers (polymers having only one type of repeating unit) and copolymers (a polymer having more than one type of repeating unit).

[00138] "Biodegradable polymer" means a polymer or polymers, which degrade in vivo, under physiological conditions. The release of the therapeutic agent occurs concurrent with, or subsequent to, the degradation of a biodegradable polymer over time.

[00139] The terms "biodegradable" and "bioerodible" are used interchangeably herein. A biodegradable polymer may be a homopolymer, a copolymer, or a polymer comprising more than two different polymeric units.

[00140] As used herein, the term "biocompatible" means a material, such as a polymer, that is compatible with living tissue. Biocompatible polymers may be biodegradable, bioerodible, or dissolvable.

[00141] As used herein, the term "polymer matrix" refers to a mixture of polymers. The polymer matrix includes polymers which interact to form a gel. In embodiments, the polymers may include functional groups. In embodiments, the functional groups may be attached to the polymer by a linker. The mixture of polymers may be of the same type, e.g. two different PEG polymers, or of different types, e.g. PEG polymers combined with PGA polymers.

[00142] "Ocular condition" means a disease, ailment, or condition, which affects or involves the ocular region.

[00143] The term "hot-melt extrusion" or "hot-melt extruded" is used herein to describe a process, whereby a blended composition is heated and/or compressed to a molten (or softened) state and subsequently forced through an orifice, where the extruded product (extrudate) is formed into its final shape, in which it solidifies upon cooling.

[00144] The term "non-extruded implant" or "non-hot melt extruded implant" refers to an implant that was not manufactured in a process that utilizes an extrusion step.

[00145] As used herein, "drug release" refers to the release of the at least one therapeutic agent, or drug, from an implant. The drug release may be sustained release, controlled release, or rapid release.

[00146] "Sustained release" or "controlled release" refers to the release of at least one therapeutic agent, or drug, from an implant at a predetermined rate. Sustained release implies that the therapeutic agent is not released from the implant sporadically, in an unpredictable fashion. The term "sustained release" may include a "burst phenomenon" associated with deployment. In some example embodiments, an initial burst of at least one therapeutic agent may be desirable, followed by a more gradual release thereafter. The release rate may be steady state (commonly referred to as "timed release" or zero order kinetics), that is the at least one therapeutic agent is released in even amounts over a predetermined time (with or without an initial burst phase), or may be a gradient release.

[00147] "Therapeutic agent" means an agent which possess activity to treat or prevent a disease or symptom of a disease. A therapeutic agent includes a small molecule, protein, peptide, amino acid, nucleic acid, antibodies, antibody fragments, short chain variable fragments (scFv), growth factors, angiogenic factors, insulin, nucleic acids, antisense nucleic acids, DNA, RNA, siRNA, aptamers, CRISPR complexes, and the like. Embodiments described herein which refer to protein are also operable with any of therapeutic agents described above.

[00148] "Solid state" means a particle comprising a therapeutic agent which is formed in a solid phase. A solid state particle is a stable form a therapeutic agent, having a fixed volume and shape.

[00149] "Micronized particles" have a largest dimension of less than about 20 μm.

[00150] "Therapeutically effective amount" means a level or amount of a therapeutic agent needed to treat an ocular condition; or the level or amount of a therapeutic agent that produces a therapeutic response or desired effect in the subject to which the therapeutic agent was administered. Thus, a therapeutically effective amount of a therapeutic agent, such as a aflibercept, is an amount that is effective in reducing at least one symptom of an ocular condition. Achieving a therapeutically effective amount may be achieved through sustained release of a therapeutic agent from an implant or mesoparticle.

[00151] "Pharmaceutical composition" means a composition comprising a therapeutic agent. In embodiments, the pharmaceutical composition can refer to a gel, a xerogel, and/or a hydrogel.

[00152] "Treatment," "treating," and the like mean the following actions: (i) preventing a particular disease or disorder from occurring in a subject who may be predisposed to the disease or disorder but has not yet been diagnosed as having it; (ii) curing, treating, or inhibiting the disease, i.e., arresting its development; or (iii) ameliorating the disease by reducing or eliminating symptoms, conditions, and/or by causing regression of the disease.

[00153] As used herein, "PuP" is used represent a protein microparticle.

[00154] As used herein, PEG is used to represent poly(ethylene glycol) of all molecular weights including polyethylene oxides.

[00155] **THERAPEUTIC AGENT**

[00156] Embodiments described herein provide for the sustained delivery of a therapeutic agent to the eye of a subject in need thereof.

[00157] **Anti-VEGF Agents**

[00158] Many disease of the eye, such as AMD, DR, DMA, and RVO, damage the retina and cause blindness when blood vessels around the retina grow abnormally and leak fluid, causing the layers of the retina to separate. This abnormal growth is caused by VEGF.

[00159] Anti-VEGF therapy is a treatment for AMD, DME, and RVO. Treatment is generally an intravitreal injection (IVT) of between 50 μL and 100 μL. The duration of action is between 4 and 8 weeks. However, the vitreous half-life of the anti-VEGF agents is much shorter, on the order of days. IVT can only be performed by medical doctors. Extending the duration of action to 4 to 6 months would have a tremendous value to patients, healthcare providers, and payers.

[00160] Monoclonal antibodies (mAb) are both thermally and pH sensitive. These materials can also dimerize and degrade and become very viscous when concentrated. Doses required are typically in the milligram range.

[00161] Examples of anti-VEGF agents include pegaptanib, bevacizumab, ranibizumab, aflibercept, lampalizumab.

[00162] Pegaptanib is a PEGylated aptamer of approximately 50 kDa. This material is provided as a single dose injection containing 1.6 mg of pegaptanib sodium. Ranibizumab is approved for the treatment of AMD. This molecule has a molecular weight of 48 kDa. The typical dose is 0.3 mg and has a duration of action of four weeks.

[00163] **Bevacizumab**

[00164] Bevacizumab (AVASTIN®), is an angiogenesis inhibitor, a drug that slows the growth of new blood vessels.

[00165] Bevacizumab is a recombinant, divalent mAb which has a molecular weight of approximately 149 kDa. A typical dose is 1.25 mg.

[00166] Bevacizumab blocks angiogenesis by inhibiting vascular endothelial growth factor A (VEGF-A). VEGF-A is a chemical signal that stimulates angiogenesis in a variety of diseases.

[00167] **Aflibercept**

[00168] Aflibercept (EYLEA) is a recombinant fusion protein consisting of vascular endothelial growth factor (VEGF)-binding portions from the extracellular domains of human VEGF receptors 1 and 2, that are fused to the Fc portion of the human IgG1 immunoglobulin. This mAb has a molecular weight of approximately 97 kDa. Aflibercept binds to circulating VEGFs and acts like a "VEGF trap". It thereby inhibits the activity of the vascular endothelial growth factor subtypes VEGF-A and VEGF-B inhibiting the growth of new blood vessels.

[00169] Aflibercept is administered for the treatment of AMD, MD, RVO, and DME. Typical dosing is about 2 mg. The dosing schedule for this material requires more frequent injections, one every four weeks for the first twelve weeks, followed by every eight weeks thereafter.

[00170] **DARPins**

[00171] In certain embodiments, the disclosure provides for the manufacture and utilization of DARPins (an acronym for designed ankyrin repeat proteins), which are genetically engineered antibody mimetic proteins typically exhibiting highly specific and high-affinity target protein binding. They are derived from natural ankyrin proteins and consist of at least three, usually four or five repeat motifs of these proteins. Their molecular mass is about 14 or 18 kDa (kilodaltons) for four- or five-repeat DARPins, respectively. The disclosure envisions manufacture of these DARPins and utilization in the delivery systems taught herein.

[00172] **MICRONIZED PARTICLE**

[00173] In embodiments, the therapeutic agent is prepared as a powder prior to incorporation into a pharmaceutical composition. "Powder" refers to a collection of dry particles. "Particle" includes any shape, such as cubes, spheres, cylinders, teardrop-shapes, small rods and other irregular shapes. In general, the powder is processed to provide a controlled particle composition with a known size, shape, or distribution (variance from a mean or average) thereof. Protein powders typically contain stabilizing sugars such as sucrose or trehalose. Powders may be prepared by, for example, grinding, ball milling, cryomilling, microfluidizing or mortar-and-pestle followed by sieving. Particle size reduction to the desired range may be achieved by, for example, grinding, ball milling, jet milling of a solid protein suspension in a compatible organic solvent.

[00174] In particular embodiments, the powder is prepared as solid state micronized particle prior to incorporation into a pharmaceutical composition. Micronization techniques include fabrication of PRINT® microparticles, lyophilization, spray drying, and jet milling, to name a few.

[00175] **PRINT® Microparticle**

[00176] In some embodiments, the therapeutic agent is prepared as a PRINT® microparticles (PuPs). PuPs are micronized solid state particles comprising a therapeutic agent (e.g., protein) made using the PRINT® technique. In embodiments, PuPs are

fabricated by casting a solution of protein as a thin film on a sheet. Molds are then placed on the thin film to form the PuPs.

[00177] The molds used to prepare the PuPs may be of any dimension and/or shape. In some embodiments, the PuP molds have a largest dimension of about 1 μm to about 10 μm, and a smallest dimension of about 1 μm to about 10 μm. In embodiments, the PuP molds have dimensions of about 1 μm in diameter x 1 μm in height ± 10% of any dimension.

[00178] The PuPs may then be added to a polymer matrix to form a hydrogel as discussed herein.

[00179] **Lyophilized Particle**

[00180] In embodiments, the therapeutic agent (e.g., protein) is prepared as a micronized powder utilizing lyophilization. Lyophilization (also known as freeze-drying or cryodesiccation) is a dehydration process. Lyophilization works by freezing a sample and then reducing the surrounding pressure to allow the frozen solvent in the sample to sublimate directly from the solid phase to the gas phase.

[00181] In embodiments, a stock solution comprising the protein is lyophilized to form protein particles. In embodiments, the lyophilized protein powder can be further processed to reduce the average particle size, such as by (1) sieving particles through mesh or series of mesh sizes (e.g., from about 250 μm to about 20 μm), and/or (2) breaking particles using a mortar and pestle.

[00182] In embodiments, the lyophilized protein powder is added to a polymer matrix to form a gel as described herein.

[00183] **Spray Dried Particle**

[00184] In embodiments, the therapeutic agent (e.g., protein) is prepared as a micronized powder utilizing a spray drying technique. Spray drying is a method of producing a dry powder from a liquid or slurry by rapidly drying with a hot gas. Spray dryers use an atomizer or spray nozzle to disperse the liquid or slurry into a controlled drop size spray. Spray drying produces a powder comprising polydisperse particles.

[00185] In embodiments, the spray dried protein powder is added to a polymer matrix to form a gel as described herein.

[00186] **Jet Milled Particle**

[00187] In some embodiments, the therapeutic agent (e.g., protein) is prepared as a micronized powder utilizing a jet mill. A jet mill grinds materials by using a high speed jet of compressed air or inert gas to impact particles into each other. Jet mills can be designed to output particles below a certain size, while continue milling particles above that size, resulting in a narrow size distribution of the resulting product.

[00188] In embodiments, the jet milled protein powder is added to a polymer matrix described herein to form a gel as discussed herein.

[00189] **POLYMERS**

[00190] The polymers used to form implants and/or mesoparticles of the disclosure have independent properties associated with them that when combined produce the properties needed to provide a sustained release of a therapeutically effective amount of a therapeutic agent.

[00191] A few of the primary polymer characteristics that control therapeutic agent release rates are the molecular weight, molecular weight distribution, polymer endgroup (i.e., acid or ester), and the ratio of polymers and/or copolymers in the polymer matrix. The present disclosure provides examples of polymer matrices that possess desirable therapeutic agent release characteristics by manipulating one or more of the aforementioned properties to develop a suitable ocular implant.

[00192] The degree of stability can be varied widely, depending upon the choice of monomer, whether a homopolymer or copolymer is employed, employing mixtures of polymers, and whether the polymer includes terminal acid groups.

[00193] In embodiments, the polymer materials used to form the implants described herein are biodegradable. In embodiments, the polymer materials may be any combination of polyethylene glycol, polylactic acid, glycolic acid, and co-polymers thereof that provides sustained-release of the therapeutic agent into the eye over time.

[00194] In particular embodiments, examples of useful polymeric materials include, without limitation, such materials derived from and/or including organic esters and organic ethers, which when degraded result in physiologically acceptable degradation products. Also, polymeric materials derived from and/or including, anhydrides, amides, orthoesters and the like, by themselves or in combination with other monomers, may also find use in the present disclosure. The polymeric materials may be addition or condensation polymers. The polymeric materials may be cross-linked or non-cross-linked. For some embodiments, besides carbon and hydrogen, the polymers may include at least one of oxygen and nitrogen. The oxygen may be present as oxy, e.g. hydroxy or ether, carbonyl, e.g. non-oxo-carbonyl, such as carboxylic acid ester, and the like. The nitrogen may be present as amide, cyano and amino.

[00195] In one embodiment, polymers of ethylene glycol are used in the implants.

[00196] Equally important to controlling the biodegradation of the polymer and hence the extended release profile of the implant is the relative average molecular weight of the polymeric composition employed in the implants. Different molecular weights of the same or different polymeric compositions may be included to modulate the release profile of the at least one therapeutic agent.

[00197] **PLA**

[00198] PLA polymers are known to degrade via backbone hydrolysis (bulk erosion) and the final degradation products are lactic and glycolic acids, which are non-toxic and considered natural metabolic compounds. Lactic and glycolic acids are eliminated safely via the Krebs cycle by conversion to carbon dioxide and water.

[00199] PLA is cleaved predominantly by non-enzymatic hydrolysis of its ester linkages throughout the polymer matrix, in the presence of water in the surrounding tissues. PLA polymers are biocompatible, because they undergo hydrolysis in the body to produce the original monomers, lactic acid and/or glycolic acid. Lactic and glycolic acids are nontoxic and eliminated safely via the Krebs cycle by conversion to carbon dioxide and water. The biocompatibility of PLA polymers has been further examined in animals and humans. The findings indicate that the polymers are well tolerated.

[00200] Examples of PLA polymers, which may be utilized in an embodiment of the disclosure, include the RESOMER® product line available from Evonik Industries identified as, but are not limited to, R 207 S, R 202 S, R 202 H, R 203 S, R 203 H, R 205 S, R 208, R 206, and R 104. Examples of suitable PLA polymers include both acid terminated (H) and ester terminated (S) polymers with inherent viscosities ranging from approximately 0.15 to approximately 2.2 dL/g when measured at 0.1% w/v in $CHCl_3$ at 25°C with an Ubbelhode size 0c glass capillary viscometer.

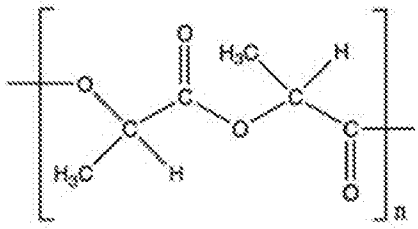
[00201] In one embodiment, ester terminated (S) PLA polymers with an inherent viscosity ranging from approximately 0.25 to approximately 2.2 dL/g when measured at 0.1% w/v in $CHCl_3$ at 25°C with an Ubbelhode size 0c glass capillary viscometer can be used in the present invention.

[00202] The synthesis of various molecular weights of PLA is possible. In one embodiment, PLA, such as RESOMER® R208, with an inherent viscosity of approximately 1.8 to approximately 2.2 dl/g (0.1% in chloroform, 25 °C), can be used. In another embodiment, PLA, such as RESOMER® R203S, with an inherent viscosity of approximately 0.25 to approximately 0.35 dl/g (0.1% in chloroform, 25 °C) can be used. In this embodiment, the R208 and R203S polymers can be ester end capped.

[00203] In one embodiment, the biodegradable matrix is comprised of a mixture of RESOMER® R208 and R203S polymers. In one such embodiment, R208 constitutes 67 +/- 5% of the biodegradable polymer matrix and R203S constitutes 33 +/- 5% of the biodegradable polymer matrix.

[00204] In some aspects, R203S comprises 21% ±10% and R208 comprises 44% ±10% and the API (e.g. travoprost) comprises 34% ±10% of the total intracameral implant.

[00205] Resomer's R203S and R208 are poly(D,L-lactide) or PLA ester-terminated polymers with the general structure (1):



(1)

[00206] **PLGA**

[00207] PLGA polymers are known to degrade via backbone hydrolysis (bulk erosion) and the final degradation products are lactic and glycolic acids, which are non-toxic and considered natural metabolic compounds. Lactic and glycolic acids are eliminated safely via the Krebs cycle by conversion to carbon dioxide and water.

[00208] PLGA is synthesized through random ring-opening co-polymerization of the cyclic dimers of glycolic acid and lactic acid. Successive monomeric units of glycolic or lactic acid are linked together by ester linkages. The ratio of lactide to glycolide can be varied, altering the biodegradation characteristics of the product. By altering the ratio it is possible to tailor the polymer degradation time. Importantly, drug release characteristics are affected by the rate of biodegradation, molecular weight, and degree of crystallinity in drug delivery systems. By altering and customizing the biodegradable polymer matrix, the drug delivery profile can be changed.

[00209] PLGA is cleaved predominantly by non-enzymatic hydrolysis of its ester linkages throughout the polymer matrix, in the presence of water in the surrounding tissues. PLGA polymers are biocompatible, because they undergo hydrolysis in the body to produce the original monomers, lactic acid and/or glycolic acid. Lactic and glycolic acids are nontoxic and eliminated safely via the Krebs cycle by conversion to carbon dioxide and water. The biocompatibility of PLGA polymers has been further examined in animals and humans. The findings indicate that the polymers are well tolerated.

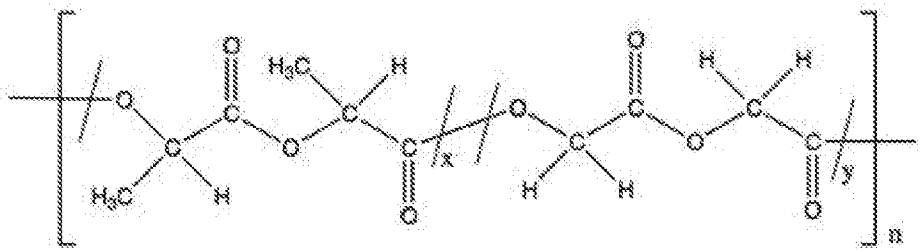
[00210] Examples of PLGA polymers, which may be utilized in an embodiment of the disclosure, include the RESOMER® Product line from Evonik Industries identified as, but are not limited to, RG 502, RG 502 H, RG 503, RG 503 H, RG 504, RG 504 H, RG 505, RG 506, RG 653 H, RG 752 H, RG 752 S, RG 753 H, RG 753 S, RG 755, RG 755 S, RG 756, RG 756 S, RG 757 S, RG 750 S, RG 858, and RG 858 S. Such PLGA

polymers include both acid and ester terminated polymers with inherent viscosities ranging from approximately 0.14 to approximately 1.7 dl/g when measured at 0.1% w/v in CHCl3 at 25°C with an Ubbelhode size 0c glass capillary viscometer. Example polymers used in various embodiments of the disclosure may include variation in the mole ratio of D,L-lactide to glycolide from approximately 50:50 to approximately 85:15, including, but not limited to, 50:50, 65:35, 75:25, and 85:15.

[00211] The synthesis of various molecular weights of PLGA with various D,L-lactide-glycolide ratios is possible. In one embodiment, PLGA, such as RESOMER® RG 752 S, with an inherent viscosity of approximately 0.16 to approximately 0.24 dl/g can be used

[00212] The synthesis of various molecular weights of PLGA with various D,L-lactide-glycolide ratios is possible. In one embodiment, PLGA, such as RESOMER® RG752S, with an inherent viscosity of approximately 0.16 to approximately 0.24 dl/g can be used

[00213] Resomer RG752S is a poly(D,L-lactide-co-glycolide) or ester-terminated PLGA copolymer (lactide:glycolide ratio of 75:25) with the general structure (2):



(2)

[00214] The polymers used to form the implants of the disclosure have independent properties associated with them that when combined provide the properties needed to provide sustained release of a therapeutically effective amount of a therapeutic agent.

[00215] A few of the primary polymer characteristics that control therapeutic agent release rates are the molecular weight distribution, polymer endgroup (*i.e.*, acid or ester), and the ratio of polymers and/or copolymers in the polymer matrix. The present disclosure provides examples of polymer matrices that possess desirable therapeutic

agent release characteristics by manipulating one or more of the aforementioned properties to develop a suitable ocular implant.

[00216] The biodegradable polymeric materials which are included to form the implant's polymeric matrix are often subject to enzymatic or hydrolytic instability. Water soluble polymers may be cross-linked with hydrolytic or biodegradable unstable cross-links to provide useful water insoluble polymers. The degree of stability can be varied widely, depending upon the choice of monomer, whether a homopolymer or copolymer is employed, employing mixtures of polymers, and whether the polymer includes terminal acid groups.

[00217] **PEG**

[00218] In some embodiments, the gels described here are crosslinked polyethylene glycol (PEG). In embodiments, the PEGs are long-chain polymers of ethylene oxide with a formula of $OH\text{-}(CH_2\text{-}CH_2\text{-}O)_n\text{-}H$, where n is the average number of oxyethylene groups present in the molecule. These polymers have been used extensively in approved products across multiple routes of administration including oral, topical, ophthalmic, rectal, vaginal, intrasynovial, intra-articular, intramuscular, and intravenous (Inactive Ingredients Database). PEGs are generally considered to have low toxicity by all routes of administration, and have been shown to be very safe in a large battery of tests using a large range of routes and molecular weights (Herold 1982, Smyth 1947, Smyth 1950, Smyth 1970, Webster 2009).

[00219] PEGs are prepared by polymerization of ethylene oxide and are available in a wide range of molecular weights. PEG has found medical use as an excipient in pharmaceutical products, in lubricating eye drops, film coatings, and ointment bases. PEG may be synthesized at a variety of molecular weights. Used herein are PEGs with molecular weights that include 100-100,000,000 Da, 400 Da, 500 Da, 100 Da, 500 Da, 1,000 Da, 2,000 Da, 3,000 Da, 5,000 Da, 7,000 Da, 10,000 Da, 15,000 Da, 20,000 Da, 30,00 Da, 40,000 Da, 50,00 Da, 100,000 Da, and 1,000,000 Da. In some aspects, PEGs with molecular weights from 500 to 1,000,000 Daltons are used. PEG is represented by the general structure (1).

(1)

[00220] PEG macromers (high molecular weight monomers) can be purchased directly from the supplier (JenKem USA, Sigma Aldrich, etc.). Such hydrogels can be degradable and non-degradable.

[00221] In one embodiment utilizing PEGs, micronized protein, the active ingredient, is added to the PEG, the inactive ingredients. In embodiments, the PEGs may have an average molecular weight of 10,000 daltons (Da) (PEG 10K) or 15,000 daltons (Da) (PEG 15K). These PEG function as both a diluent and a binder for the micronized protein to form an injectable pharmaceutical composition described herein (e.g., an implant or a mesoparticle).

[00222] **Functional Groups**

[00223] In embodiments, polymers described herein may interact to form a gel, e.g., through crosslinking interactions. In some such embodiments, such polymers are referred to herein "gel precursors" or "precursors." In embodiments, a first precursor has a first functional group and a second precursor has a second functional group. In embodiments, the first functional group and the second functional group are reactive in a solvent or melt phase to form the gel. In embodiments, the first functional group interacts with the second functional group, thereby forming a covalent bond.

[00224] In embodiments, the functional groups are configured to participate in other polymerization reactions or react with other functional groups in electrophile-nucleophile reactions. Various aspects of polymerization reactions are discussed below.

[00225] In some embodiments, precursors have electrophilic functional groups, such as carbodiimidazole, sulfonyl chloride, chlorocarbonates, n-hydroxysuccinimidyl ester, succinimidyl ester or sulfasuccinimidyl esters, and the like. In embodiments, the nucleophilic functional groups may be, for example, amine, hydroxyl, carboxyl, and thiol, and the like.

[00226] Certain functional groups, such as alcohols and carboxylic acids, do not normally react with other functional groups, such as amines, under physiological conditions (e.g., pH 7.2-11.0, 37° C.). However, such functional groups can be made more reactive by using an activating group such as N-hydroxysuccinimide (NHS). Non-limiting examples of activating groups include carbonyldiimidazole, sulfonyl chloride, aryl halides, sulfosuccinimidyl esters, N-hydroxysuccinimidyl ester, succinimidyl ester, epoxide, aldehyde, maleimides, imidoesters and the like. The N-hydroxysuccinimide esters or N-hydroxysulfosuccinimide (NHS) groups are useful groups for crosslinking of proteins or amine-containing polymers, e.g., amino terminated polyethylene glycol. An advantage of an NHS-amine reaction is that the reaction kinetics are favorable, but the gelation rate may be adjusted through pH or concentration. Sulfonated or ethoxylated forms of N-hydroxysuccinimide have a relatively increased solubility in water and hence their rapid clearance from the body. An NHS-amine crosslinking reaction may be carried out in aqueous solutions and in the presence of buffers, e.g., phosphate buffer (pH 5.0-7.5), triethanolamine buffer (pH 7.5-9.0), borate buffer (pH 9.0-12), or sodium bicarbonate buffer (pH 9.0-10.0).

[00227] In some embodiments, each precursor comprises only nucleophilic or only electrophilic functional groups and a crosslinker is used which comprises a compatible functional group. Thus, for example, if a crosslinker has nucleophilic functional groups such as amines, the precursor may have electrophilic functional groups such as N-hydroxysuccinimides. On the other hand, if a crosslinker has electrophilic functional groups such as N-hydroxysuccinimides, then the precursor may have nucleophilic functional groups such as amines or thiols. Thus, precursors such as proteins, poly(allyl amine), or amine-terminated di- or multifunctional poly(ethylene glycol) can be used.

[00228] In embodiments, a precursor can have from about 1 to about 1,000 functional groups, about 1 to about 500 functional groups, about 1 to about 400 functional groups, about 1 to about 300 functional groups, about 1 to about 200 functional groups, about 1 t about 100 functional groups, about 1 to about 90 functional groups, about 1 to about 80 functional groups, about 1 to about 70 functional groups, about 1 to about 60 functional groups, about 1 to about 50 functional groups, about 1 to about 40 functional groups, about 1 to about 30 functional groups, about 1 to about 25

functional groups, about 1 to about 20 functional groups, about 1 to about 15 functional groups, about 1 to about 10 functional groups, or about 1 to about 5 functional groups, including a values and subranges in between. Such functional groups can be the same or different.

[00229] The functional groups may be electrophiles reactable with nucleophiles, e.g., amines which form amide bonds with carboxyls or activated-acid functional groups. The functional groups may be, e.g., a strong electrophilic functional group, meaning an electrophilic functional group that effectively forms a covalent bond with a primary amine in aqueous solution at pH 9.0 at room temperature and pressure and/or an electrophilic group that reacts by a of Michael-type reaction.

[00230] Examples of strong electrophiles that do not participate in a Michaels-type reaction are: succinimides, succinimidyl esters, or NHS-esters. Examples of Michael-type electrophiles are acrylates, methacrylates, methylmethacrylates, and other unsaturated polymerizable groups.

[00231] **HYDROGEL**

[00232] A hydrogel is defined as a crosslinked hydrophilic polymer network that swells in water to at least 50%, 60%, 70%, 80%, 90%, 100%, 200%, 300 %, 400% or more of its mass or volume. Hydrogels may be made from precursors. The precursors are not hydrogels but are cross-linked with each other to form a hydrogel. Crosslinks can be formed by covalent or non-covalent bonds (e.g., ionic interactions), by hydrophobic association of precursor molecule segments, or by crystallization of precursor molecule segments. The precursors can be activated to react to form a crosslinked hydrogel. The precursors can be polymerizable and include crosslinkers that are often, but not always, polymerizable. Polymerizable precursors are thus precursors that have functional groups which react with each other to form polymers made of repeating units.

[00233] **Precursors**

[00234] Precursors may be polymers or monomers. Precursors can be have a molecular weight from 100 Da to about 100,000 Da. The precursors can have biologically inert, hydrophilic portions, e.g, a core. In the hydrophilic portion may be a

poly ether, such as polyalkylene oxides, e.g., polyethylene glycol (PEG), polyethylene oxide (PEO), polyethylene oxide-co-polypropylene oxide (PPO), co-polyethylene oxide block or random copolymers, polyvinyl alcohol (PVA), poly (vinyl pyrrolidinone) (PVP), poly (amino acids), dextran, or a protein. Thus, in embodiments, precursors may be a polymer described above comprising at least one functional group described herein.

[00235] Precursors may be made with a hydrophobic portion provided that the resultant hydrogel retains the requisite amount of water, e.g., at least about 20%. In some cases, the precursor is nonetheless soluble in water because it also has a hydrophilic portion. In other cases, the precursor makes a dispersion in the water (a suspension) but is nonetheless reactable to from a crosslinked material. Some hydrophobic portions may include a plurality of alkyls, polypropylenes, alkyl chains, or other groups. Some precursors with hydrophobic portions are sold under the trade names PLURONIC F68, JEFFAMINE, or TECTRONIC.

[00236] In embodiments, precursors may have multiple arms, e.g., 2-1,000 arms, with each arm having a terminus. An arm on a hydrogel precursor refers to a chain of chemical groups that connect a crosslinkable functional group to the core of the precursor. In some embodiments, precursors have between 1 and 500 arms, e.g., 4 to 100, 4 to 16, 4 to 8, or at least 4 arms, including all values and subranges in between.

[00237] Thus, hydrogels can be made, e.g., from a multi-armed precursor with a first set of functional groups and second precursor having a second set of functional groups. For example, four-armed precursor may have hydrophilic arms, such as a polyethylene glycol terminated with primary amines. Such precursors may be mixed with other precursors having a compatible functional group, e.g., carboxylic acid derivative, such an N-hydroxysuccinimidyl ester. The precursors described herein can have 1 or more than one functional group attached to the core, and such functional groups can be the same or different.

[00238] **Crosslinking of Precursors**

[00239] Polymerization reactions which can be used crosslink precursors are well known in the art. In some embodiments, the precursors can react by chain-growth polymerization. Chain-growth polymerization can occur through free-radical

polymerization, cationic polymerization, anionic polymerization, or coordination polymerization. Free-radical polymerization involves the processes of initiation, propagation, and termination. Initiation is the creation of free radicals necessary for propagation, as created from radical initiators (e.g., organic peroxides). Termination occurs when a radical reacts in a way that prevents further propagation. The most common method of termination is by coupling, where two radical species react with each other to form a single molecule. In cationic polymerization, a cationic initiator transfers charge to a monomer which then becomes reactive. This reactive monomer goes on to react similarly with other monomers to form a polymer. The types of monomers necessary for cationic polymerization are olefins with electron-donating substituents and heterocycles. Anionic polymerization involves the polymerization of vinyl monomers with strong electronegative groups. This polymerization is carried out through a carbanion active species. Coordination polymerization occurs when a monomer adds to a growing macromolecule through an organometallic active center. In other embodiments, precursors can react occur through step-growth polymerization. Step-growth polymerization occurs through the step wise formation of dimers, trimers, oligomers, and then polymers starting from a bifunctional or multifunctional precursor. In other embodiments, the precursors react through condensation polymerization. These reactions are typically achieved through reacting molecules which incorporate an alcohol or an amine functional groups on one species and carboxylic acid derivative (e.g., N-hydroxy succinimidyl ester) functional groups on a second species. When an amine reacts with a carboxylic acid, an amide or peptide bond is formed.

[00240] To form covalently crosslinked hydrogels, two or more precursors are crosslinked. In general, precursors will be joined to other precursors at two or more points, which each point being a linkage to the same or different precursor. Precursors with at least two reactive groups can serve as crosslinkers since each reactive group can participate in the formation of a different growing polymer chain. In embodiments in which the precursors are monomers, the monomers may be polymerized as described herein to form polymeric precursors, and such polymeric precursors may then form a crosslinked gel by reacting with other precursors.

[00241] In embodiments, the therapeutic agent is present during the cross-linking interaction. In such embodiments, a fine powder of a therapeutic agent (e.g., a water soluble biologic, such as a protein) is prepared and suspended in solvent. When the therapeutic agent is present during the cross-linking interaction, said agent becomes encapsulated in the resulting cross-linked matrix. In embodiments, the solvent is an organic solvent that does not solvate the protein. In particular embodiments, embodiments, the solvent is a hydrophilic solvent which does not solvate the protein. Gel precursors are prepared that have the capacity to form a crosslinked gel by reacting with each other in the solvent. The precursors are chosen to be substantially soluble in the solvent. The precursors and therapeutic agent are mixed in the solvent so that the therapeutic agent particles are dispersed through the matrix that forms upon crosslinking of precursors. The matrix formed in an organic is referred to as an organogel.

[00242] Typically, hydrogel systems are formed by a serial two-solvent process using an organic solvent (which can be hydrophobic or hydrophilic) to form the crosslinked system referred to as an organogel. The organic solvent is then removed to form a xerogel. The xerogel is hydrated to form a hydrogel. Prior to the inventors' discovery, it was well known that an organic, hydrophobic solvent was required to fabricate gels comprising water soluble therapeutic agents (e.g., proteins) a hydrophilic, non-aqueous solvent exposes the protein to interfaces which results in denaturation, aggregation, and therefore should result in reduced activity. Thus, prior art methods utilize a hydrophobic solvent to eliminate the possibility of interface exposure and maintain protein stability. *See, e.g.*, U.S. Patent No. 9,205,150.

[00243] Surprisingly, however, it has been discovered that proteins can be exposed to certain hydrophilic solvents during gelation without extensive denaturation, aggregation, or reduced activity. In the case of polyethylene glycol (PEG) precursors, solvents such as acetonitrile and ethyl lactate have been employed.

[00244] **Degradable hydrogels**

[00245] In some embodiments, the hydrogels described herein are degradable hydrogels. Degradable hydrogels are composed of multiarm PEGs which may be endcapped with a functional group. The degradable hydrogels can have from 1 to 1,000

arms (e.g., from about 1 to about 500 arms, or about 4 to about 100, about 4 to about 16, about 4 to about 8, or at least 4 arms).

[00246] In embodiments of this system, the PEGs have degradable spacers between the PEG and the functional group, such as a degradable spacer between a PEG and an NHS. In embodiments, the length of the spacer dictates the rate of degradation. PEGs with shorter spacers degrade faster.

[00247] In embodiments, the linker between the functional group and the core of the precursor can influence the rate of degradation. Electrophilic groups such as SG (N-hydroxysuccinimidyl glutarate), SS (N-hydroxysuccinimidyl succinate), SC (N-hydroxysuccinimidyl carbonate), SAP (N-hydroxysuccinimidyl adipate) or SAZ (N-hydroxysuccinimidyl azelate) have esteric linkages that are hydrolytically labile. As used herein "PEG-NHS" refers to a PEG polymer comprising an N-hydroxysuccinimidyl functional group, which may contain any of the above electrophilic groups. That is "PEG-NHS" includes PEG-SG-NHS, PEG-SS-NHS, PEG-SC-NHS, PEG-SAP-NHS, PEG-SAZ-NHS, and the like. More linear hydrophobic linkages such as pimelate, suberate, azelate or sebacate linkages may also be used, with these linkages being less degradable than succinate, glutarate or adipate linkages. Branched, cyclic or other hydrophobic linkages may also be used. Polyethylene glycols and other precursors may be prepared with these groups. The crosslinked hydrogel degradation may proceed by the water-driven hydrolysis of the biodegradable segment when water-degradable materials are used.

[00248] Thus it is possible to construct a hydrogel with a desired degradation profile, from a few days to many months, using a degradable segment.

[00249] In embodiments, the precursors include ester linkages to provide a desired degradation rate, and spacers may be added or subtracted near the esters to increase or decrease the rate of degradation. In embodiments, the spacers are derived from a family of dicarboxylic acids including but not limited to: Succinic (SS), glutaric (SG), adipic (SAP), and azelaic (SAZ) acids which have 2, 3, 4, and 7 methylene units between acid endgroups, respectively. The diacid forms an ester on the side proximal to the PEG

chain, and forms an activated NHS ester capable of reacting with amines on the distal end.

[00250] Non-degradable hydrogels

[00251] In some embodiments, hydrogels described herein are non-degradable hydrogels. 'Non-degradable' implants are composed of linear, or multi-arm, PEGs with acrylate functional end groups. In embodiments, the acrylate units are polymerized photochemically. The acrylate backbone crosslinks PEGs resulting in a crosslinked elastomeric network. The resulting crosslink density can be modified by changing the molecular weight of the PEGs. The ester of the acrylate degrades much more slowly than the esters described above for the degradable hydrogels. Therefore these materials are referred to as 'non-degradable.'

[00252] Wax-Fatty Acid Hydrogels

[00253] In some embodiments, the hydrogels described herein a wax-fatty acid based hydrogels. Components of wax-fatty acid mixtures may include, but are not limited to Beeswax, the triglyceride Tristearin, and fatty acids with carbon chains between 4 and 28, or between 6 and 18. These include, for example, Caproic acid, Caprylic acid, Lauric acid, Capric acid, Myristic acid, Palmitic acid, and Stearic acid. Different ratios and components may be used to control both initial release and overall sustained release. In one embodiment, the wax-fatty acid composition of the implant is formulated with Beeswax (about 30%wt), Tristearin (about 10%wt), Palmitic acid (about 30%wt), Caproic acid (about 25%wt) and Caprylic acid (about 5%wt).

[00254] PHARMACEUTICAL COMPOSITIONS

[00255] As shown in **FIG. 2** and discussed above, the disclosure provides for pharmaceutical compositions comprising a therapeutic agent. In embodiments, the pharmaceutical compositions are formulated as a hydrogel. In embodiments, the hydrogels can be fabricated as an ocular implant or mesoparticle. In some embodiments, said ocular implant or mesoparticle is manufactured utilizing PRINT® technology (Envisia Therapeutics, Inc.). Such implants and mesoparticles are formed by casting the hydrogels in a mold, thereby forming an implant or mesoparticle.

[00256] In embodiments, the pharmaceutical composition is comprised of the biocompatible polymer matrix and at least one therapeutic agent. The biocompatible polymer matrix is comprised of polymers meeting desired characteristics. For example, desired characteristics may include a specific therapeutic agent release rate or a specific duration of action. The biocompatible polymer matrix may be comprised of one polymer, two polymers, or many polymers, such as three, four, five polymers, or more polymers.

[00257] In some embodiments, the compositions may comprise polymers utilizing the same monomer, such as compositions comprising various PEG homopolymers. However, even if the polymers of the composition utilize the same monomer, the polymers may differ in other characteristics, such as, for example, inherent viscosity or molecular weight.

[00258] In other embodiments, the compositions may comprise a mixture of polymers, wherein the polymers may be the same or different, such as compositions comprising a amine-terminated PEG and NHS ester-terminated PEG. However, even if the polymers of the compositions are different, the polymers may be similar in other characteristics, such as for example, inherent viscosity or molecular weight.

[00259] In one embodiment, the pharmaceutical composition comprises a biocompatible polymer matrix and at least one therapeutic agent homogeneously dispersed throughout the polymer matrix. Further, the presently discussed pharmaceutical compositions comprising a biocompatible polymer matrix and at least one therapeutic agent, may, in certain embodiments, also exclude other polymers. That is, in some embodiments, the aforementioned polymer matrix only includes one polymer and active therapeutic agent.

[00260] In embodiments, the biocompatible polymer matrix comprises about 10 wt % to about 90 wt % of the pharmaceutical composition. In embodiments, the biocompatible polymer matrix comprises about 20 wt % to about 70 wt % of the pharmaceutical composition. In embodiments, the biocompatible polymer matrix forms a gel which encapsulates the therapeutic agent.

[00261] In embodiments, the therapeutic agent is formulated as a solid state micronized particle. In embodiments, the therapeutic agent is blended with the

biocompatible polymer matrix to form the pharmaceutical composition. In embodiments, the polymer matrix forms a hydrogel under appropriate conditions. The amount of therapeutic agent used in the pharmaceutical composition depends on several factors such as: biocompatible polymer matrix selection, therapeutic agent selection, rate of release, duration of release desired, configuration of pharmaceutical composition, and ocular pharmacokinetics, to name a few.

[00262] For example, the therapeutic agent may comprise approximately 0.1 to approximately 90.0 weight percent of the pharmaceutical composition. In some embodiments, the therapeutic agent comprises approximately 10.0 to approximately 60.0 weight percent of the pharmaceutical composition. In other embodiments, the therapeutic agent comprises approximately 13.0 to approximately 55.0 weight percent of the pharmaceutical composition.

[00263] Implants having various compositions, sizes, and shapes were fabricated and tested as set forth in the disclosure; however, it will be appreciated that these are non-limiting examples of implant designs contemplated by the present disclosure.

[00264] **Mesoparticles**

[00265] 'Mesoparticles' are implants of the present disclosure, of a particular size, in which a protein particle manufactured by a process disclosed herein (i.e., PRINT®, lyophilization, spray drying, etc) is embedded.

[00266] Thus, the term "mesoparticle" is intended to describe a particular size of *implant*, which is between a "nano and small micron" sized *particle* and a larger "100's of micron-sized" *implant*.

[00267] In particular aspects, the disclosure provides for hydrogel based mesoparticles that comprise a hydrogel matrix implant that has protein microparticles embedded therein.

[00268] In embodiments, a mesoparticle may have dimensions within the range of about 10 μm x 10 μm x 10 μm (or 10 μm diameter if cylindrical) to about 100 μm x 100 μm x 100 μm (or 100 μm diameter if cylindrical), including all values and subranges in between. In embodiments, the mesoparticles have dimensions of about 10 μm x 10 μm x

10 μm (or 10 μm diameter if cylindrical) to about 90 μm x 90 μm x 90 μm (or 90 μm diameter if cylindrical), or about 10 μm x 10 μm x 10 μm (or 10 μm diameter if cylindrical) to about 80 μm x 80 μm x 80 μm (or 80 μm diameter if cylindrical), or about 10 μm x 10 μm x 10 μm (or 10 μm diameter if cylindrical) to about 70 μm x 70 μm x 70 μm (or 70 μm diameter if cylindrical), or about 15 μm x 15 μm x 15 μm (or 15 μm diameter if cylindrical) to about 60 μm x 60 μm x 60 μm (or 60 μm diameter if cylindrical), or about 20 μm x 20 μm x 20 μm (or 20 μm diameter if cylindrical) to about 50 μm x 50 μm x 50 μm (or 50 μm diameter if cylindrical). In particular embodiments, the mesoparticles have dimensions of about 12.5 μm x 12.5 μm x 12.5 μm (or 12.5 μm diameter if cylindrical) to about 50 μm x 50 μm x 50 μm (or 50 μm diameter if cylindrical). In certain embodiments, the mesoparticles have dimensions of about 12.5 μm x 12.5 μm x 12.5 μm (or 12.5 μm diameter if cylindrical) ± 10% in any dimension, or 25 μm x 25 μm x 25 μm (or 25 μm diameter if cylindrical) ± 10% in any dimension, or about 50 μm x 50 μm x 50 μm (or 50 μm diameter if cylindrical) ± 10% in any dimension.

[00269] Protein particles embedded in the mesoparticle, *e.g.* hydrogel based mesoparticle, maybe of any size, so long as the protein particles are sufficiently sized so as to be entrapped within said mesoparticle sized hydrogel implant. In particular embodiments, the protein particles embodied in a mesoparticle may have dimensions within the range of about 1 μm x 1 μm x 1 μm (or 1μm diameter if cylindrical) to about 10 μm x 10 μm x 10 μm (or 10μm diameter if cylindrical). In other embodiments, the protein particles are less than or equal to about 20 μm in an dimension.

[00270] Due to the size of the mesoparticles, said mesoparticles are suspended in a pharmaceutically acceptable liquid vehicle for ocular delivery. In some embodiments, the volume of the liquid delivery vehicle may be about 1000 μL or less, about 900 μL or less, about 800 μL or less, about 700 μL or less, about 600 μL or less, about 500 μL or less, about 400 μL or less, about 300 μL or less, about 200 μL or less, about 100 μL or less, about 90 μL or less, about 80 μL or less, about 70 μL or less, about 60 μL or less, about 50 μL or less, about 40 μL or less, about 30 μL or less, about 20 μL or less, or about 15 μL or less.

[00271] In some embodiments, the volume of the liquid delivery vehicle may be in the range of from about 10 μL to about 500 μL, about 10 μL to about 400 μL, about 10 μL to about 300 μL, about 10 μL to about 200 μL, or about 10 μL to about 100 μL, including all values and subranges in between. In a particular embodiment, the volume of the liquid delivery vehicle containing the mesoparticle is about 100 μL or less. In some embodiments, the pharmaceutically acceptable liquid delivery vehicle comprises about 1 to about 1,000,000 mesoparticles, or about 1 to about 100,000 mesoparticles, or about 1 to about 10,000 mesoparticles, or about 1 to about 1,000 mesoparticles, or about 1 to about 900 mesoparticles, or about 1 to about 800 mesoparticles, or about 1 to about 700 mesoparticles, or about 1 to about 600 mesoparticles, or about 1 to about 500 mesoparticles, or about 1 to about 400 mesoparticles, or about 1 to about 300 mesoparticles, or about 1 to about 200 mesoparticles, or about 1 to about 100 mesoparticles, or about 1 to about 50 mesoparticles, including all values and subranges in between.

[00272] **Implants**

[00273] Implants of the present disclosure are pharmaceutical compositions having particular size, in which a protein particle manufactured by a process disclosed herein (i.e., PRINT®, lyophilization, spray drying, etc) is embedded.

[00274] Thus, the term "implant" is intended to describe a particular size of *implant*, which is larger than about 100 μm in all dimensions.

[00275] In particular aspects, the disclosure provides for hydrogel based implants that comprise a hydrogel matrix which has protein microparticles encapsulated therein.

[00276] In particular embodiments, the implant can be a cylindrical-shaped implant (produced by an extrusion) or a rod-shaped implant (produced by a PRINT® process). Cylindrical implants may have a diameter of about 0.001 mm to about 10 mm μm, and length of about 0.001 mm to about 10 mm. In particular embodiments, the cylindrical implants have a dimeter of about 1 mm ± 10% and a length of about 3 mm ± 10%. Rod-shaped implants may have dimensions within the range of about 100 μm x 100 μm x 100 μm to about 1,000 μm x 1,000 μm x 5,000 μm, including all values and subranges in between. In particular embodiments, the hydrogels may have dimensions

225 μm x 225 μm x 2925 μm (L x W x H) ± 10% of any dimension, or 311 μm x 395 μm x 6045 μm (L x W x H) ± 10% of any dimension, or 600 μm x 600 μm x 1000 μm (L x W x H) ± 10% of any dimension.

[00277] Protein particles embodied in the implant, *e.g.* hydrogel based implant, maybe of any size, so long as the protein particles are sufficiently sized so as to be entrapped within said implant. In particular embodiments, the protein particles embodied in an implant may have dimensions within the range of about 1 μm x 1 μm x 1 μm (or 1μm diameter if cylindrical) to about 10 μm x 10 μm x 10 μm (or 10μm diameter if cylindrical). In other embodiments, the protein particles are less than or equal to about 20 μm in an dimension.

[00278] **Compositions**

[00279] In embodiments, the pharmaceutical composition comprises: (A) gel comprising biocompatible polymer matrix; and (B) a particle comprising a therapeutic agent, wherein the gel encapsulates the therapeutic agent. In certain embodiments, the particle is a protein microparticle (PuP) comprising the therapeutic agent and at least one pharmaceutically acceptable excipient. In embodiments, the PuP has dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension.

[00280] In embodiments, the gel is a biodegradable gel. In embodiments, the gel is formed by covalently crosslinking the gel precursors. In embodiments, the gel comprises crosslinked polyethylene glycol polymers.

[00281] In embodiments, the gel precursors comprise at least one PEG polymer having an N-hydroxy succinimidyl ester functional group (PEG-NHS) and at least one PEG polymer having an amine functional group (PEG-amine). In embodiments, the PEG polymers can have molecular weight of about 10,000 Da to about 15,000 Da. In embodiments, the PEG-NHS polymers can have from about 1 to about 16 arms, e.g., about 4 arms or about 8 arms. In embodiments, the PEG-amine polymers can have from about 1 to about 16 arms, e.g., about 4 arms or about 8 arms.

[00282] In embodiments, the gel comprises 20-70 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising: (i) 10-35 ± 5 wt % of at

least one PEG-NHS having a molecular weight of 10,000 Da; and (ii) 10-35 ± 5 wt % of at least PEG-amine having a molecular weight of 10,000 Da. In embodiments, the therapeutic agent comprises about 10-60 ± 5 wt % of the pharmaceutical composition. In embodiments, the PEG-NHS has 4 arms or 8 arms. In embodiments, the least PEG-amine 10,000 Da has 4 arms or 8 arms.

[00283] In embodiments, the gel comprises 20-70 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising: (i) 10-35 ± 5 wt % of at least one PEG-NHS having a molecular weight of about 10,000-15,000 Da; and (ii) 10-35 ± 5 wt % of at least one PEG-amine having a molecular weight of about 10,000-15,000 Da; and wherein the therapeutic agent comprises about 10-60 ± 5 wt % of the pharmaceutical composition. In embodiments, the therapeutic agent is fabricated as a PuP having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension.

[00284] In particular embodiments, the pharmaceutical compositions may be rod-shaped having have dimensions 225 μm x 225 μm x 2925 μm (L x W x H) ± 10% of any dimension, or 311 μm x 395 μm x 6045 μm (L x W x H) ± 10% of any dimension, or 600 μm x 600 μm x 1000 μm (L x W x H) ± 10% of any dimension. In other embodiments, the pharmaceutical composition may be cylindrical having a diameter of about 1 mm ± 10% and a length of about 3 mm ± 10%. In still other embodiments, the pharmaceutical composition is cube-shaped having dimensions of about 12.5 μm x 12.5 μm x 25 μm ± 10% in any dimension, or 25 μm x 25 μm x 25 μm ± 10% in any dimension, or about 50 μm x 50 μm x 50 μm ± 10% in any dimension.

[00285] In embodiments, the gel comprises 20 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising: (i) 10 ± 3 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; and (ii) 10 ± 3 wt % of an 8 arm PEG-amine having a molecular weight of 10,000 Da; and wherein the therapeutic agent comprises about 35 ± 5 wt % of the pharmaceutical composition. In embodiments, the therapeutic agent is fabricated as a PuP having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension.

[00286] In embodiments, the gel comprises 20 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising: (i) 10 ± 3 wt % of an 8-

arm PEG-NHS having a molecular weight of 10,000 Da; (ii) 5 ± 3 wt % of an 8-arm PEG-amine having a molecular weight of 10,000 Da; and (iii) 5 ± 3 wt % of a 4-arm PEG-amine having a molecular weight of 10,000 Da, and wherein the therapeutic agent comprises about 36 ± 5 wt % of the pharmaceutical composition. In embodiments, the therapeutic agent is fabricated as a PuP having dimensions of 1 μm x 1 μm x 1 μm (L x W x H) ± 10% in any dimension.

[00287] In embodiments, the gel comprises 21 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising: (i) 10 ± 5 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; and (ii) 10 ± 5 wt % of an 8 arm PEG-amine having a molecular weight of 10,000 Da; and wherein the therapeutic agent comprises about 46 ± 5 wt % of the pharmaceutical composition. In embodiments, the therapeutic agent is fabricated as a PuP having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension.

[00288] In embodiments, the gel comprises a first pharmaceutical composition and a second pharmaceutical composition. In embodiments, the first pharmaceutical composition comprises a first gel comprising 21 ± 5 % wt of the first pharmaceutical composition and contains a mixture of polymers comprising: (a) 10 ± 5 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; and (b) 10 ± 5 wt % of an 4 arm PEG-amine having a molecular weight of 10,000 Da. In embodiments, the first therapeutic agent comprises about 36 ± 5 wt % of the first pharmaceutical composition. In embodiments, the first therapeutic agent is fabricated as a PuP having In embodiments, the second composition comprises a second gel comprising 21 ± 5 % wt of the first pharmaceutical composition and contains a mixture of polymers comprising: (a) 10 ± 5 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; and (b) 10 ± 5 wt % of an 8 arm PEG-amine having a molecular weight of 10,000 Da. In embodiments, the second therapeutic agent comprises about 36 ± 5 wt % of the second pharmaceutical composition. In embodiments, the second therapeutic agent is fabricated as a PuP having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension.

[00289] **Drug Release Profile Manipulation**

[00290] The rate of drug release from the mesoparticles and implants described herein depends on several factors. For example, the surface area of the implant, the therapeutic agent content, the water solubility of the therapeutic agent, and the speed of polymer degradation or dissolution.

[00291] The versatility of drug delivery polymer systems which can be used to fabricate an implant or mesoparticle (e.g., PCL, PEG, PGA, PLA, and PLGA) allows for construction of delivery systems to tailor the drug release for treating a variety of front and back of the eye diseases.

[00292] When the versatility of drug delivery polymer systems (e.g., PCL, PEG, PGA, PLA, and PLGA polymers) are combined with the manufacturing techniques of the present disclosure, for example PRINT® (Envisia Therapeutics Inc.) fabrication technology, then a host of custom tailored and highly consistent and predictable drug release profiles can be created, which were not possible based upon the technology of the prior art.

[00293] That is, with the present mold based particle fabrication technology, implants can be manufactured that exhibit a drug release profile that has highly reproducible characteristics implant to implant. The drug release profiles exhibited by various implants or mesoparticles of the present disclosure are consistent implant to implant and mesoparticle to mesoparticle and demonstrate variation that is not statistically significant. Consequently, the drug release profiles demonstrated by embodiments of the implants and mesoparticles exhibit coefficients of variation that are within a confidence interval and not biologically relevant. The ability to produce implants and mesoparticles that demonstrate such a high degree of consistent drug release is advancement over the state of the art.

[00294] In some embodiments, the release rate is dependent on the relationship between the therapeutic agent and mesh size of the hydrogel network. For bevacizumab (Dh=6.5nm), for example, slow release occurs when the mesh size below 4-5 nm. Once the mesh size reaches 4-5 nm, the release rate increases significantly. Thus, the composition of implants and mesoparticles described herein can be tuned to such that the therapeutic agent is released at a desired rate and duration.

[00295] The amount of therapeutic agent loading can have an effect on the release profile of the hydrogel. For example, in some embodiments, increasing the therapeutic agent loading (for example, above about 50 %) can cause a burst release.

[00296] In some embodiments, the ocular implants can have instant release, such as burst release (e.g., more than about 20% of the therapeutic agent released in about 1 day), a delayed release (e.g., a significant amount of the therapeutic agent is not released for at least about 1 day after administration), or a sustained release.

[00297] **Drug Release Kinetics**

[00298] Drug release is influenced by many factors including: polymer composition, therapeutic agent content, implant morphology, porosity, tortuosity, surface area, method of manufacture, and deviation from sink conditions, just to name a few. The present mold based manufacturing techniques, utilized in embodiments of the disclosure, are able to manipulate implant morphology, porosity, tortuosity, and surface area in ways that the prior art methods were incapable of doing. For instance, the highly consistent drug release profiles, highly consistent implant morphologies, and highly consistent homogeneous drug dispersions achievable with PRINT molds were not available to prior art practitioners.

[00299] In general, therapeutic agent release occurs in 3 phases: (a) an initial burst release of therapeutic agent from the surface, (b) followed by a period of diffusional release, which is governed by the inherent dissolution of therapeutic agent (diffusion through internal pores into the surrounding media) and lastly, (c) therapeutic agent release associated with biodegradation of the polymer matrix. The rapid achievement of high therapeutic agent concentrations, followed by a longer period of continuous lower-dose release, makes such delivery systems ideally suited for acute-onset diseases that require a loading dose of therapeutic agent followed by tapering doses.

[00300] More recent advancements in drug delivery systems have allowed for biphasic release characteristics with an initial high (burst) rate of therapeutic agent release followed by sustained zero-order (linear) kinetic release (*i.e.*, therapeutic agent release rate from the polymer matrix is steady and independent of the therapeutic agent concentration in the surrounding milieu) over longer periods. In addition, when desired

for treating chronic diseases such as AMD, RVO, and DME, these therapeutic agent delivery systems can be designed to have steady state release following zero order kinetics from the onset.

[00301] **FABRICATION OF PHARMACEUTICAL COMPOSITION**

[00302] **Protein Solution**

[00303] As shown in **FIG. 2**, as solution is prepared comprising the therapeutic agent (e.g., protein). The therapeutic agent can be prepared in any solution in which the protein is stable. Protein buffers can be used to formulate stable protein solutions. Non-limiting examples of protein buffers include bicarbonate/carbonic acid buffers, phosphate buffers, citric acid buffers, histidine buffers, tris buffers, succinate buffers, glutamate buffers, and acetate buffers. Excipients used to stabilize a protein in solution are well known in the art. Non-limiting examples of such excipients include sugars, such as sucrose, trehalose, lactose, maltose, mannitol, and sorbitol. In some embodiments, the protein solution is formulated with excipients trehalose, phosphate buffer (pH 6.2), and polysorbate 20 (tween 20). The excipients can be present in the protein solution at any concentration in which the protein is stable and may be different for different proteins or therapeutic agents.

[00304] In embodiments, the protein concentration in the solution is about 1-100 g/L, or about 10-100 g/L, or about 10-60g/L, or about 20-60 g/L, or about 30-60g/L.

[00305] **Micronization**

[00306] As discussed above, in embodiments, the solution comprising the therapeutic agent (e.g., a protein) may be processed to form particles comprising the therapeutic agent. Such particles may be incorporated into a pharmaceutical composition. In embodiments, the particles are solid state particles. In embodiments, the particles are micronized particles. In embodiments, such particles have a largest dimension of less than or equal to about 20 μm, e.g., from about 1 μm to about 20 μm. Such particles may be referred to herein as micronized particles, micronized therapeutic agents, micronized protein, and the like.

[00307] In embodiments, micronization of the therapeutic agent can be performed utilizing PRINT® mold, lyophilization, spray drying, jet milling, and the like. In embodiments which do not utilize a PRINT® mold to fabricate micronized particles, particle sizing can be performed using custom-made or standardized sieve mesh sizes. In addition to standard U.S. and Tyler mesh sizes, sieves are also commonly used in the Market Grade, Mill Grade, and Tensile Bolting Cloth.

[00308] Micronized particles may be formulated with sugars such as trehalose to stabilize the therapeutic agent. In some embodiment, these sugars may be allowed to persist in the gel. Thus, in embodiments, the micronized particle may comprise as a therapeutic agent content of from about 20% w/w to about 99% w/w.

[00309] **Solvent System**

[00310] As discussed above, the gel precursors and therapeutic agent particles are combined in a solvent to form a suspension comprising the gel precursors and particles comprising the therapeutic agent. The gel precursors and therapeutic agent particles can be combined in any order, and the components can be dissolved in the same or different solvent prior to combining such components to form the suspension. Any solvent can be used provided that such solvent does not substantially interfere with gel formation. In embodiments, the solvent does not substantially degrade the mold utilized in implant fabrication. In embodiments, the solvent does not substantially alter the activity of the protein.

[00311] Solvent compatibility can be determined using techniques known to those skilled in the art. For example, protein-solvent compatibility can be established experimentally by exposure followed by characterization testing to determine if the protein has been denatured, formed aggregates, and/or undergone substitution or alteration of one or more chemical groups. Solvent compatibility can be tested by immersing the subject protein in the subject solvent for an appropriate period of time, removing the protein, such as by filtration and vacuum drying, and then testing for recovery of the protein by HPLC or other appropriate analytical method. In embodiments, solvent compatibility was assessed by fabricating implants in different solvents and measuring monomer content of the protein released from said implant. A

compatible solvent shows substantially no degradation of protein during manufacturing and substantially no aggregation.

[00312] In embodiments, the solvent is an organic solvent, a hydrophobic solvent a hydrophilic solvent, a polar aprotic solvent, a polar protic solvent, or combinations thereof. Particular embodiments utilize an organic, hydrophobic solvent, such methylene dichloride (also referred to as dichloromethane). Other embodiment utilize an organic, hydrophilic solvent, such as acetonitrile and ethyl lactate. Non-limiting examples of hydrophilic solvents which may be used with the methods and compositions described herein include: tetrahydrofuran, ethyl acetate, acetone, dimethylformamide, acetonitrile, dimethyl sulfoxide, nitromethane, propylene carbonate, formic acid, n-butanol, isopropanol, n-propanol, ethanol, methanol, acetic acid, and water, and the like.

[00313] In embodiments, the solvent is an organic solvent, and the gel is an organogel. In embodiments in which the gel is formed in a cylindrical mold, and the solvent is: organic and polar aprotic; or organic, hydrophilic and polar aprotic; or organic and hydrophobic; or organic, hydrophobic, and polar aprotic. In particular embodiments, the gel is formed in a cylindrical mold and the solvent is acetonitrile.

[00314] In embodiments in which the gel is formed in a PRINT mold, the solvent is organic and polar protic; organic and hydrophilic; organic, hydrophilic, and polar protic. In particular embodiments, the gel is formed in a PRINT mold and the solvent is a ethyl lactate.

[00315] In embodiments which utilize a hydrophilic solvent, the solvent may include water, provided that when water is present it does not substantially interfere with the gelation reaction. For example, the hydrophilic may include less than about 20 wt % water (e.g., 15% or less, 10% or less, 9% or less, 8% or less, 7% or less, 6% or less, 5% or less, 4% or less, 3% or less, 2% or less, or 1% or less).

[00316] **Gel Implants**

[00317] Thus, processes for making gels that incorporate a therapeutic agent include, for example, making a gel in a solvent with the therapeutic agent present at the time of formation of the gel or added to the gel after its formation. In embodiments, the

therapeutic agent is a micronized protein, such as a PuP, lyophilized powder or spray dried powder.

[00318] One embodiment for making a gel is to make a preformed delivery device in a solvent in the presence of a therapeutic agent using a mold, e.g., a cylindrical or PRINT® mold. A first gel precursor having a first type functional group is dissolved with a second gel precursor having a second type of functional group in a solvent in the presence of a therapeutic agent. Depending on the solvent system, the therapeutic agent is either miscible in the solvent (e.g., a hydrophilic solvent) or immiscible in the solvent (e.g., an organic solvent). The solution is introduced into a mold (e.g., cylindrical or PRINT® molds) and left until the precursors crosslink with each other by covalent bond formation between the first functional groups and the second functional groups. The gel is fully or partially dried to form a dehydrated gel (i.e., a xerogel). In some embodiments, the organogel is frozen and lyophilized to remove the solvent (i.e., to dehydrate the gel). The gel is then removed from the PRINT® mold or cut or otherwise trimmed to size after extrusion in the cylindrical mold. An organic solvent may be used, e.g., for loading the hydrogel with non-water soluble agents or encapsulating agents tolerant to the organic phase. A hydrophilic solvent may be used, e.g., for loading the hydrogel with water soluble agents or encapsulating agents tolerant to the aqueous phase. As discussed above, the inventors surprisingly discovered that certain therapeutic agents, which were previously thought to be susceptible to degradation in the presence of an aqueous phase, can be fabricated in a gel in a hydrophilic solvent without substantial degradation or loss of function.

[00319] In one embodiment for making a pharmaceutical composition, a second precursor having nucleophilic functional groups is dissolved in an organic solvent comprising a first precursor having electrophilic functional groups and a therapeutic agent. In embodiments, the therapeutic agent is a micronized protein, such as a PuP, lyophilized powder or spray dried powder. The precursors are formed into a gel, dried (as discussed above), and shaped as desired. Optionally, the gel can be prepared in a mold (e.g., PRINT® mold or cylindrical mold) whereby the gel is fabricated with desired dimensions.

[00320] In another embodiment for making a pharmaceutical composition in an organic solvent, a hydrogel is formed first, then the hydrogel is loaded with a therapeutic agent. A first hydrogel precursor having a first functional group is dissolved with a second hydrogel precursor having a second functional group in an organic solvent. The solution is introduced into a mold and left until the precursors crosslink with each other by covalent bond formation between the first functional group and the second functional group. The hydrogel is fully or partially dried to form a dehydrated hydrogel. A solvent (the same or different from the one used during crosslinking; i.e., that solvent can be the same or different organic solvent or the solvent can be a non-organic solvent, such as an aqueous solvent) that swells the cross-linked hydrogels is added. This solvent contains the dissolved therapeutic agent at high concentrations. In embodiments, the therapeutic agent is a micronized protein, such as a PuP, lyophilized powder or spray dried powder. The hydrogels are allowed to swell with the drug solution, causing some drug to permeate into the hydrogel matrix. Gels are removed, and either dried (as above), or placed into a non-solvent, e.g., hexane. The non-solvent causes the organic solvent to leave the gel and the agent to precipitate-out in the gel matrix, leaving an agent-loaded gel. This embodiment may be used, for example, for loading of agents incompatible with the particular crosslinking functional groups, e.g., agents with primary amines when precursor amine functional groups are intended to be reacted during crosslinking. This separation of drug loading and crosslinking steps removes problems with chemical incompatibility between the therapeutic agent and the crosslinking reaction.

[00321] Thus, in another embodiment for making a pharmaceutical composition, a branched PEG having electrophilic functional groups at each arm terminus is mixed with a PEG having nucleophilic functional groups at each arm terminus. The precursors are mixed or otherwise activated to form the crosslinked gel in a mold and then dried of solvent. The gel is added to a solvent that swells the cross-linked gel. The solvent contains dissolved therapeutic agents at high concentrations. The gels are allowed to swell with the solvent-agent solution, causing some drug to permeate into the gel matrix. Gels are removed, and either dried again as above, or placed into a precipitating agent such as hexane. If the precipitating agent is compatible with the solvent, but incompatible with the gel network and the therapeutic agent, it causes the solvent to

migrate from the gel leaving the therapeutic agent to precipitate out in the gel matrix, forming a drug loaded delivery device (e.g., implant).

[00322] Another embodiment for making a pharmaceutical composition is to make a hydrogel in an aqueous solvent in the presence of a therapeutic agent. A first hydrogel precursor having a first functional group is dissolved with a second hydrogel precursor having a second functional group in an aqueous solvent in the presence of a therapeutic agent. In embodiments, the therapeutic agent is a micronized protein, such as a PuP, lyophilized powder or spray dried powder. The solution is introduced into a mold (e.g., cylindrical or PRINT® molds) and left until the precursors crosslink with each other by covalent bond formation between the first functional groups and the second functional groups. The hydrogel is fully or partially dried to form a dehydrated to form a xerogel. In embodiments during can occur by removing the solvent. In some embodiments, the organogel is lyophilized. The hydrogel is then removed from the mold or cut or otherwise trimmed to another shape or size. This embodiment may be used, for example, for loading the hydrogel with water soluble agents or encapsulated agents tolerant to the aqueous phase. The agent may be dispersed in the aqueous solvent, e.g., in solution or suspension. A suspension may be, for instance, a particle comprising the agent or a suspension of encapsulated agent. This embodiment is useful for, for example, loading of hydrogels with agents already encapsulated in other polymer systems. The aqueous based manufacture may also be used to avoid extraction of the encapsulated agent, which could occur with some organic solvents.

[00323] As shown in **FIG. 3**, an embodiment for making a hydrogel is thus dissolving a PEG having an electrophilic function group (e.g., an NHS ester) with a PEG having a nucleophilic functional group (e.g., a primary amine) in a solvent containing an therapeutic agent. In embodiments, the therapeutic agent is a micronized protein, such as a PuP, lyophilized powder or spray dried powder. The gel is formed in a mold (e.g., PRINT® or cylindrical mold) and the solvent is removed. The dried implants or mesoparticles are removed from the mold, and optionally further processed, e.g., for size or shape.

[00324] As discussed above, certain embodiments provide for carrying out the crosslinking chemistry in a hydrophilic solvent. Thus, one embodiment relates to combining in a hydrophilic solvent a first hydrogel precursor having a first type of functional group with a second hydrogel precursor having a second type of functional group in the presence of a therapeutic agent. The precursors are reacted to form a gel.

[00325] In embodiments, the first precursor is a PEG with an NHS-ester functional groups. In other embodiments, the second precursor is a PEG with an amine functional groups.

[00326] In embodiments, NHS-PEG and amine-PEG are gelled in an organic solvent that is a poor/non-solvent for the protein, e.g., dichloromethane. When the protein is not dissolved or substantially dissolved in the reaction solvent, the protein does not participate in the gelling reaction.

[00327] In embodiments, NHS-PEG and amine-PEG are gelled in an organic hydrophilic solvent, e.g., ethyl lactate or acetonitrile. In such embodiments, the protein is present in the reaction solvent, but the protein does not participate in the gelling reaction. Surprisingly, the protein does not aggregate and is not denatured when present in the reaction solvent.

[00328] In embodiments, the gels can be loaded with therapeutic agent ranging from 5-80% (w/w) of the gel, e.g., 10-50% (w/w). In some embodiments, gels are loaded with therapeutic agent in an amount less than or equal to about 50% (w/w) of the gel. In some embodiments, the gels can be loaded with therapeutic agent in the range of from 0.01 mg to about 100 mg, or about 0.1 mg to about 100 mg, or about 0.1 mg to about 10 mg, or about 0.1 mg to about 5 mg, or about 0.5 mg to about 1.0 mg.

[00329] **Fabrication of an Ocular Implant**

[00330] Various methods may be used to produce the implants. Methods include, but are not limited to, solvent casting, phase separation, interfacial methods, molding, compression molding, injection molding, extrusion, co-extrusion, heat extrusion, die cutting, heat compression, and combinations thereof. In certain embodiments, the implants are molded.

[00331] In some embodiments, the pharmaceutical compositions of the present disclosure are fabricated via extrusion in a cylindrical mold. In some embodiments, the implants and mesoparticles of the present disclosure are fabricated through the PRINT® particle fabrication technology (Envisia Therapeutics Inc., North Carolina). In particular, the implants, mesoparticles, or elements of the implants (for example, the PuP's) are made by molding components in polymeric mold cavities.

[00332] The molds can be polymer-based molds and the mold cavities can be formed into any desired shape and dimension. Uniquely, as the implants or elements of the implants are formed in the cavities of the mold, the implants or elements of the implants are highly uniform with respect to shape, size, and composition. Due to the consistency among the physical and compositional makeup of each implant of the present pharmaceutical compositions, the pharmaceutical compositions of the present disclosure provide highly uniform release rates and dosing ranges. The methods and materials for fabricating the implants or elements of the implants of the present disclosure are further described and disclosed in the following issued patents and co-pending patent applications, each of which are incorporated herein by reference in their entirety: U.S. Pat. Nos. 8,518,316; 8,444,907; 8,420,124; 8,268,446; 8,263,129; 8,158,728; 8,128,393; 7,976,759; U.S. Pat. Application Publications Nos. 2013-0249138, 2013-0241107, 2013-0228950, 2013-0202729, 2013-0011618, 2013-0256354, 2012-0189728, 2010-0003291, 2009-0165320, 2008-0131692; and pending U.S. Application Nos. 13/852,683 filed March 28, 2013 and 13/950,447 filed July 25, 2013.

[00333] The mold cavities can be formed into various shapes and sizes. For example, the cavities may be shaped with curved edges, sharp or square edges, circular or arched perimeters, flat sides, substantially parallel sides, pointed ends, cylindrical, prism, rectangular prism, triangular prism, pyramid, square pyramid, triangular pyramid, cone, cylinder, torus, or rod. The cavities may have the same shape or may have different shapes. In certain aspects of the disclosure, the shapes of the implants are a cylinder, rectangular prism, and rod. In a particular embodiment, the implant is a rod having substantially a square, rectangular, circular, or oval cross-section taken normal to the long axis of the implant.

[00334] The mold cavities can be dimensioned from nanometer to micrometer to millimeter dimensions and larger. For certain embodiments of the disclosure, mold cavities are dimensioned in the micrometer and millimeter range. For example, cavities may have a smallest dimension of between approximately 50 nanometers and approximately 750 μm. In some aspects, the smallest mold cavity dimension may be between approximately 1 μm and approximately 500 μm. In other aspects, the smallest mold cavity dimension may be between approximately 225 μm and approximately 400 μm. For example, mold cavities may have a largest dimension of between approximately 1,000 μm and approximately 10,000 μm. In other aspects, the largest mold cavity dimension may be between approximately 2,000 μm and approximately 6,000 μm. In other aspects, the largest mold cavity dimension may be between approximately 4,000 μm and approximately 6,000 μm.

[00335] In other embodiments, the implants can be fabricated through the application of additive manufacturing techniques. Additive manufacturing, such as disclosed in US published application US 2013/0295212 and the like can be utilized to either make the master template used in the PRINT® process, utilized to make the mold used into the PRINT® process otherwise disclosed herein or utilized to fabricate the implants directly.

[00336] The implants can have an aspect ratio of width-to-length from 1:1 to greater than 1:30. In some embodiments, the width-to-length aspect ratio of the implant is between 1:2 to 1:25. In some embodiments, the width-to-length aspect ratio of the implant is between 1:5 to 1:20. In some embodiments, the width-to-length aspect ratio of the implant is between 1:10 to 1:20. In some embodiments, the width-to-length aspect ratio of the implant is between 1:15 to 1:20.

[00337] In other embodiments, the micronized particles, e.g., PuPs, after being formed can be incorporated into larger implants as disclosed elsewhere herein. The larger implants can be in the size range of 10 micrometers in a broadest dimension or larger, depending on the size designed into the mold cavities (as further described herein and in the prior art incorporated herein by reference). Importantly, for intraorbital ophthalmic applications, the density of the implant is fabricated to be greater than the

density of the fluid environment in which the implant will be placed, such as for example the aqueous humor or the like, such that the implant settles and remains outside the field of view of the patient and the implant also remains in the eye. Furthermore, the larger surface area to volume ratio of the particles having smaller overall dimensions, for example, a 10 micron cube compared to a 100 micron cube, will degrade more rapidly. Likewise, a collection of, for example, 10 micron cube particles having total overall volume equal to a 100x100x2000 micron implant will conform to the shape of the space to which they are implanted much more closely than the 100x100x2000 micron implant.

[00338] In some embodiments the larger implants can have a cross-sectional dimension of 10 micrometers or larger and a density greater than that of the aqueous humor, vitreous humor, or the like such that the implant settles due to gravitational forces. In some embodiments the implants have a largest cross-sectional dimension of 20 micrometers and a density greater than that of the aqueous humor, vitreous humor, or the like such that the implant settles due to gravitational forces. In some embodiments the implants have a largest cross-sectional dimension of 50 micrometers and a density greater than that of the aqueous humor, vitreous humor, or the like such that the implant settles due to gravitational forces. In some embodiments the implants have a largest cross-sectional dimension of 100 micrometers and a density greater than that of the aqueous humor, vitreous humor, or the like such that the implant settles due to gravitational forces. In some embodiments the implants have a largest cross-sectional dimension of 200 micrometers and a density greater than that of the aqueous humor, vitreous humor, or the like such that the implant settles due to gravitational forces. In some embodiments the implants have a largest cross-sectional dimension of 500 micrometers and a density greater than that of the aqueous humor, vitreous humor, or the like such that the implant settles due to gravitational forces.

[00339] In one embodiment, a mold cavity with dimensions of 12.5 μm × 12.5 μm × 25 μm (W × H x L) is utilized to fabricate the mesoparticles of the present disclosure.

[00340] In one embodiment, a mold cavity with dimensions of 25 μm × 25 μm × 25 μm (W × H x L) is utilized to fabricate the mesoparticles of the present disclosure.

[00341] In one another embodiment, a mold cavity with dimensions of 25 μm × 25 μm × 50 μm (W × H x L) is utilized to fabricate the mesoparticles of the present disclosure.

[00342] In one another embodiment, a mold cavity with dimensions of 50 μm × 50 μm × 30 μm (W × H x L) is utilized to fabricate the mesoparticles of the present disclosure.

[00343] In one embodiment, a mold cavity with dimensions of 50 μm × 50 μm × 50 μm (W × H x L) is utilized to fabricate the mesoparticles of the present disclosure.

[00344] In one embodiment, a mold cavity having generally a rod shape with dimensions of 140 μm × 140 μm × 1325 μm (W × H x L) is utilized to fabricate the implants of the present disclosure.

[00345] In another embodiment, a mold cavity having generally a rod shape with dimensions of 395 μm × 311 μm × 6045 μm (W × H x L) is used to fabricate the implants of the present disclosure.

[00346] In one embodiment, a mold cavity having generally a rod shape with dimensions of 100 μm × 100 μm × 1500 μm (W × H x L) is utilized to fabricate the implants of the present disclosure.

[00347] In one embodiment, a mold cavity having a rod shape with dimensions of 150 μm × 150 μm × 3150 μm (W × H x L) is used to fabricate the implants of the present disclosure.

[00348] In one embodiment, a mold cavity having generally a rod shape with dimensions of 180 μm × 180 μm × 3000 μm (W × H x L) is used to fabricate the implants of the present disclosure.

[00349] In one embodiment, a mold cavity having generally a rod shape with dimensions of 200 μm × 200 μm × 2000 μm (W × H x L) is utilized to fabricate the implants of the present disclosure.

[00350] In one embodiment, a mold cavity having a rod shape with dimensions of 200 μm × 200 μm × 1000 μm (W × H x L) is used to fabricate the implants of the present disclosure.

[00351] In another embodiment, a mold cavity having generally a rod shape with dimensions of 225 μm × 225 μm × 2700 μm (W × H x L) is used to fabricate the implants of the present disclosure.

[00352] In another embodiment, a mold cavity having generally a rod shape with dimensions of 250 μm × 250 μm × 1500 μm (W × H x L) is used to fabricate the implants of the present disclosure.

[00353] In another embodiment, a mold cavity having generally a rod shape with dimensions of 200 μm × 200 μm × 4500 μm (W × H x L) is used to fabricate the implants of the present disclosure.

[00354] In another embodiment, a mold cavity having generally a rod shape with dimensions of 265 μm × 265 μm × 4500 μm (W × H x L) is used to fabricate the implants of the present disclosure.

[00355] In another embodiment, a mold cavity having generally a rod shape with dimensions of 255 μm × 255 μm × 4500 μm (W × H x L) is used to fabricate the implants of the present disclosure.

[00356] In another embodiment, a mold cavity having generally a rod shape with dimensions of 225 μm × 225 μm × 2925 μm (W × H x L) is used to fabricate the implants of the present disclosure.

[00357] In another embodiment, a mold cavity having generally a rod shape with dimensions of 311 μm × 395 μm × 6045 μm (W × H x L) is used to fabricate the implants of the present disclosure.

[00358] In another embodiment, a mold cavity having generally a rod shape with dimensions of 600 μm × 600 μm × 1000 μm (W × H x L) is used to fabricate the implants of the present disclosure.

[00359] Implants and mesoparticles may be stored in a dried state to improve stability during storage. Accordingly, in some embodiments, the solvent is removed from the gels to form xerogels. In embodiments, the gel is hydrated in an aqueous solution to form a hydrogel. Hydration can occur prior to administration and the implant is

administered as a hydrogel, or the implant can be administered as a xerogel and subsequently hydrated in-vivo to form a hydrogel.

[00360] The implants swell once immersed in water. The implants swell anisotropically. Generally they swell about 2x their initial size in all dimensions. As the esters degrade over time due to hydrolysis, the implant continues to swell. After this point, the implant breaks down into much smaller components. Eventually, once all of the esters have been hydrolyzed, the precursors revert to roughly their starting size and the polymer is gone. At this point the "implant" is a viscous aqueous solution of precursor.

[00361] The process of degradation can take anywhere from a few days to 6+ months. Full release of a therapeutic agent typically precedes the complete degradation of the implant. The lag time between complete release of the therapeutic agent and complete dissolution of the implant varies based on a variety of factors including but not limited to the degradation rate of the esters, % solids, the number of arms in the PEG, the molecular weight between crosslinks, % loading, etc. As the polymer degrades, the length between crosslinks or mesh size increases.

[00362] A needle may be used to deliver the implants disclosed herein to the desired site in the eye. In some embodiments, the needle used to deliver the implants disclosed herein is in the range of about a 34 gauge needle to about a 6 gauge needle, including all values and subranges in between.

[00363] In some embodiments, the needle used to deliver the implants disclosed herein can from be a 6 gauge to a 34 gauge needle, or from a 10 gauge to a 34 gauge, or a 15 gauge to a 34 gauge, or from a 17 gauge to a 34 gauge or from a 19 gauge to a 34 gauge, including all values and subranges therein. In some embodiments, the needle gauge is a 6 gauge or smaller, 7 gauge or smaller, 8 gauge or smaller, 9 gauge or smaller, 10 gauge or smaller, 11 gauge or smaller, 12 gauge or smaller, 13 gauge or smaller, 14 gauge or smaller, 15 gauge or smaller, 16 gauge or smaller, 17 gauge or smaller, 18 gauge or smaller, 19 gauge or smaller, 20 gauge or smaller, 21 gauge or smaller, 22 gauge or smaller, 23 gauge or smaller, 24 gauge or smaller, 25 gauge or smaller, 26 gauge or smaller, 27 gauge or smaller, 28 gauge or smaller, 29 gauge or smaller, 30

gauge or smaller, 31 gauge or smaller, 32 gauge or smaller, 33 gauge or smaller, or 34 gauge or smaller.

[00364] In a particular embodiment, the gauge of the needle used to deliver the implants disclosed herein is in the range of 19 or 34 gauge. Thus, in some embodiments, the needle gauge is 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, or 34.

[00365] In some embodiments, delivery of such implants disclosed herein include delivery through a 27 gauge needle or smaller. In preferred delivery methods the needed is 28 gauge, 29 gauge, 30 gauge, 31 gauge, 32 gauge, 33 gauge, or 34 gauge needle.

[00366] In some embodiments, the implants are sized and shaped to fit into a needle that is sufficiently small ($\leq$ 22G) to allow tissue to self-seal after injection.

[00367] **Delivery Devices**

[00368] In embodiments, a delivery device may be used to insert the implant into the eye or eyes for treatment of ocular diseases.

[00369] Suitable devices can include a needle or needle-like applicator. In some embodiments, the smallest dimension of an implant may range from approximately 50 μm to approximately 750 μm, and therefore a needle or needle-like applicator with a gauge ranging from approximately 22 to approximately 30 may be utilized. The delivery implant may be a syringe with an appropriately sized needle or may be a syringe-like implant with a needle-like applicator.

[00370] Delivery routes include punctual, intravitreal, subconjunctival, lens, intrascleral, fornix, anterior sub-Tenon's, suprachoroidal, posterior sub-Tenon's, subretinal, anterior chamber, and posterior chamber, to name a few.

[00371] In embodiments, an implant or implants are delivered intraviterally to a patient's eye to treat AMD.

[00372] In some aspects, the ocular implant is sized and structured to allow for administration with a needle for delivery. An important aspect of the present invention is the uniformity and control of overall size to the tolerances discussed herein to provide for use of the smallest needle gauge as possible. An implant will have between 10-50 micron clearance between overall maximum implant cross-sectional width and inside needle

diameter. In other embodiments, an implant-needle clearance shall be between 20-40 micron between overall maximum implant cross-sectional width and inside needle diameter. In other embodiments, an implant-needle clearance shall be not less than 40 micron between overall maximum implant cross-sectional width and inside needle diameter. In other embodiments, an implant-needle clearance shall be not less than 30 micron between overall maximum implant cross-sectional width and inside needle diameter. In other embodiments, an implant-needle clearance shall be not less than 20 micron between overall maximum implant cross-sectional width and inside needle diameter. In other embodiments, an implant-needle clearance shall be not less than 10 micron between overall maximum implant cross-sectional width and inside needle diameter. It will be appreciated by one of ordinary skill in the art that the three-dimensional shape of the implant can be designed to maximize the volume of the inner opening of the needle or to facilitate the desired loading, insertion, tissue deposition or other parameter of the implant or treatment. In some embodiments, the molds and implants of the present invention are designed as cylindrical implants. In some embodiments the cylindrical implants are fabricated with a cross-sectional diameter that is not less than 30 micrometers smaller than the inner diameter of the needle. In some embodiments the implant, mold, or master from which the mold is made is fabricated utilizing additive manufacturing techniques.

[00373] DELIVERY OF BIOLOGICS WITHOUT AGGREGATION OR DENATURATION

[00374] In embodiments, the pharmaceutical compositions and methods described may be used to formulate and deliver a protein (or other therapeutic agent) without causing substantial aggregation or substantial denaturation.

[00375] Proteins may aggregate, which reduces its biological activity. The pharmaceutical compositions described herein are used to formulate and deliver a protein without substantial aggregation. The term "without substantial aggregation" means that at least about 90% of the protein or protein microparticle (PuP) is delivered as monomer, e.g., 91%, 92%, 93%, 94%, 96%, 97%, 98%, or 99%.

[00376] In embodiments described herein, however, proteins may be fabricated in a pharmaceutical composition and delivered without substantial aggregation (i.e., as monomers). Excipients may be utilized in the methods and pharmaceutical compositions described herein to reduce aggregation. In embodiments, the excipients are amino acids or derivatives thereof. In certain embodiments, the excipients are leucine, trileucine, histidine, and the like. Proteins may be tested for aggregation by a variety of techniques, including size exclusion chromatography (SEC) and various mass spectrometry techniques.

[00377] Proteins are easily denatured. In embodiments described herein, however, proteins may be fabricated in a pharmaceutical composition and delivered without substantial denaturation. In embodiments, the term without substantial denaturation refers to a protein processed into a particle without modification of the protein's chemical structure (without addition of chemical groups or changes of the existing chemical groups), without changes to the protein's conformation, i.e., secondary and/or tertiary and/or quaternary structure.

[00378] Proteins may be tested for denaturation by a variety of techniques, including enzyme-linked immunosorbent assay (ELISA), isoelectric focusing (IEF), size exclusion chromatography (SEC), high-pressure liquid chromatography (HPLC), circular dichroism (CD), and Fourier Transform Infrared Spectroscopy (FTIR). These tests report parameters such as changes in molecular weight, change in end groups, changes in bonds, changes in hydrophobicity or volume exclusion, and revelation/hiding of antigenic sites. In general, a test by IEF and ELISA may be designed that is adequate to show native conformation after processing, although other tests and test combinations may alternatively be used.

[00379] **KITS**

[00380] In embodiments, the implant and delivery device may be combined and presented as a kit for use.

[00381] The implant may be packaged separately from the delivery device and loaded into the delivery device just prior to use.

[00382] Alternatively, the implant may be loaded into the delivery implant prior to packaging. In this case, once the kit is opened, the delivery implant is ready for use.

[00383] Components may be sterilized individually and combined into a kit, or may be sterilized after being combined into a kit.

[00384] Further, as aforementioned, a kit may include an array with implants bound thereon.

[00385] **METHODS OF TREATMENT**

[00386] **Use of Ocular Implant for Treatment**

[00387] In one aspect of the disclosure, there is presented a method of treating AMD, RVO, and DME. The method comprises placing a biodegradable implant in an eye, degrading the implant, releasing a therapeutic agent which is effective to treat neovascularization, and thereby treating AMD, RVO, or DME.

[00388] In aspects of the disclosure, the eye is that of an animal. For example, a dog, cat, horse, cow (or any agricultural livestock), or human.

[00389] **Course of Treatment**

[00390] Over the course of treatment, the biodegradable polymer matrix degrades releasing the therapeutic agent. Once the therapeutic agent has been completely released, the polymer matrix is expected to be gone. Complete polymer matrix degradation may take longer than the complete release of the therapeutic agent. Polymer matrix degradation may occur at the same rate as the release of the therapeutic agent.

[00391] Embodiments the present disclosure may be used treating abnormal vessel growth in the eye, such as AMD, RVO, and DME. Current treatments for AMD, RVO, and DME require the patient to receive intraviteral injections every 4 to 8 weeks. The pharmaceutical composition of the disclosure is designed for sustained release of an effective amount of therapeutic agent with sustained release greater than 8 weeks, thus eliminating the need for monthly injections. In embodiments, therapeutic agent is released into the vitreous and maintains therapeutically relevant concentrations of the therapeutic agent in the vitreous for at least about 3 months (e.g., about 4 months, about 5 months, about 6 months, etc.)

[00392] For example, the pharmaceutical composition may be designed to release an effective amount of therapeutic agent for more than two months, three months, four months, five months, six months, seven months, eight months, nine months, ten months, eleven months, or twelve months.

[00393] In embodiments, the inventors have discovered that the release profile of pharmaceutical compositions described herein may be engineered to release an amount of a therapeutic agent which has been determined to be efficacious based on half-life calculations and efficacy of immediate-release formulations. For example, 2 mg of aflibercept administered as a solution to the vitreous as a solution is known to be efficacious for about 2 months (e.g., about 57 days or about 64 days). Based on half-life calculations shown below for aflibercept in a human vitreous of 4.5mL, aflibercept is efficacious at concentrations of about 3.5 ng/mL and 1.7 ng/mL.. Half-life calculations are described in, e.g., Stewart, M. W. Eye Reports, 2011. 1:e5; 12-14, which is herein incorporated by reference in its entirety. Thus, in embodiments, the pharmaceutical compositions may be engineered for the sustained release of aflibercept at a concentration of at least about 1 ng/mL for at least about 3 months.

Aflilbercept

mass	day	total ng/mL
2	7.13	444.4444
1	14.26	222.2222
0.5	21.39	111.1111
0.25	28.52	55.55556
0.125	35.65	27.77778
0.0625	42.78	13.88889
0.03125	49.91	6.944444
0.015625	57.04	3.472222
0.007813	64.17	1.736111

[00394] Similarly, 0.5 mg of ranibizumab administered as a solution to the vitreous as a solution is known to be efficacious for about 1 month (e.g., about 28 days or about or 33 days). Based on half-life calculations shown below for ranibizumab in a human vitreous of 4.5mL, ranibizumab is efficacious at concentrations of about 3.5 ng/mL and 1.7 ng/mL.. Half-life calculations are described in, e.g., Stewart, M. W. Eye Reports, 2011. 1:e5; 12-14, which is herein incorporated by reference in its entirety. Thus, in embodiments, the pharmaceutical compositions may be engineered for the sustained release of ranibizumab at a concentration of at least about 1 ng/mL for at least about 3 months.

Ranibizumab

mass	day	total ng/mL
0.5	4.75	111.1111
0.25	9.5	55.55556
0.125	14.25	27.77778
0.0625	19	13.88889
0.03125	23.75	6.944444
0.015625	28.5	3.472222
0.007813	33.25	1.736111
0.003906	38	0.868056
0.001953	42.75	0.434028

[00395] Thus, the pharmaceutical composition may be designed to release an effective amount of the therapeutic agent per day during the sustained release of the therapeutic agent. In some embodiments, daily release rates may range from about 0.1 µg/day to about 50 µg/day, from about 0.1µg/day to about 40 µg/day, from about 0.1µg/day to about 30 µg/day, from about 0.1µg/day to about 25 µg/day, from about

0.1μg/day to about 20 μg/day, from about 0.1μg/day to about 15 μg/day, or from about 0.1μg/day to about 10 μg/day, including all values and subranges in between. In some embodiments, daily release rates may range from about 0.5 μg/day to about 40 μg/day, from about 0.5 μg/day to about 30 μg/day, from about 0.5 μg/day to about 25 μg/day, from about 0.5 μg/day to about 20 μg/day, from about 0.5 μg/day to about 15 μg/day, from about 0.5 μg/day to about 10 μg/day, from about 0.5 μg/day to about 5 μg/day, from about 0.5 μg/day to about 4 μg/day, from about 0.5 μg/day to about 3 μg/day, from about 0.5 μg/day to about 2 μg/day, or from about 0.5 μg/day to about 1 μg/day, including all values and subranges in between. In some embodiments, daily release rates may range from about 1 μg/day to about 30 μg/day, from about 1 μg/day to about 25 μg/day, from about 1 μg/day to about 20 μg/day, from about 1 μg/day to about 15 μg/day, from about 1 μg/day to about 10 μg/day, from about 1 μg/day to about 5 μg/day, from about 1 μg/day to about 4 μg/day, from about 1 μg/day to about 3 μg/day, or from about 1 μg/day to about 2 μg/day, including all values and subranges in between. In some aspects, the aforementioned daily release rates apply to utilization of Aflibercept as the therapeutic agent.

[00396] In some embodiments, daily release rates may range from about 2 μg/day to about 15 μg/day, including all values and subranges in between. Thus, in some embodiments, the daily release rate may be about 2 μg/day, about 3 μg/day, about 4 μg/day, about 5 μg/day, about 6 μg/day, about 7 μg/day, about 8 μg/day, about 9 μg/day, about 10 μg/day, about 11 μg/day, about 12 μg/day, about 13 μg/day, about 14 μg/day, or about 15 μg/day. In some aspects, the aforementioned daily release rates apply to utilization of Aflibercept as the therapeutic agent.

[00397] For example, in embodiments, the disclosure provides for Aflibercept daily release rates of 2 μg/day to about 15 μg/day, including all values and subranges in between. In some embodiments, the daily release rates of Aflibercept may be about 2 μg/day, about 3 μg/day, about 4 μg/day, about 5 μg/day, about 6 μg/day, about 7 μg/day, about 8 μg/day, about 9 μg/day, about 10 μg/day, about 11 μg/day, about 12 μg/day, about 13 μg/day, about 14 μg/day, or about 15 μg/day.

[00398] In some embodiments, the disclosure provides for methods of maintaining therapeutic relevant concentrations in the vitreous of an anti-VEGF agent for at least about 3 months (e.g., about 4 months, about 5 months, about 6 months or more). Accordingly, in some embodiments, the pharmaceutical compositions described herein may be formulated to release therapeutically relevant concentrations of an anti-VEGF agent for at least about 3 months (e.g., about 4 months, about 5 months, about 6 months or more). In embodiments, a therapeutically relevant concentration of an anti-VEGF agent in the vitreous of a human is in the range of about 0.1 ng/mL to about 50 ng/mL, or about 1 ng/mL to about 50 ng/mL, or about 1 ng/mL to about 40 ng/mL, or about 1 ng/mL to about 30 ng/mL, or about 1 ng/mL to about 20 ng/mL, or about 1 ng/mL to about 10 ng/mL, including all values and subranges in between. In certain embodiments, a therapeutically relevant concentration of an anti-VEGF agent in the vitreous of a human is at least about 1 ng/mL, or at least about 2 ng/mL, or at least about 3 ng/mL, or at least about 4 ng/mL, or at least about 5 ng/mL, or at least about 6 ng/mL, or at least about 7 ng/mL, or at least about 8 ng/mL, or at least about 9 ng/mL, or at least about 10 ng/mL, or at least about 11 ng/mL, or at least about 12 ng/mL, or at least about 13 ng/mL, or at least about 14 ng/mL, or at least about 15 ng/mL, or at least about 16 ng/mL, or at least about 17 ng/mL, or at least about 18 ng/mL, or at least about 19 ng/mL, or at least about 20 ng/mL.

[00399] Accordingly, in certain embodiments, a pharmaceutical composition described herein comprises aflibercept, and the pharmaceutical composition is formulated to maintain a concentration of aflibercept of at least about 1 ng/mL in the vitreous for at least about 3 months (e.g., about 4 months, about 5 months, about 6 months or more). In certain embodiments, the concentration of aflibercept in the vitreous is in the range of about 1 ng/mL to about 10 ng/mL, and such concentrations are maintained for at least about 3 months (e.g., about 4 months, about 5 months, about 6 months or more).

[00400] In a preferred embodiment, the pharmaceutical composition is dosed in a repetitive manner. The dosing regimen provides a second dose of the pharmaceutical composition is dosed following the substantial release of drug cargo by first dose . The dosing regimen also provides that a third dose of the pharmaceutical composition implants is not dosed until the polymer matrix of the implants of the second dosing are

sufficiently degraded. In a preferred embodiment the implant of the second dose fully degrade before the third dosing is administered.

[00401] Method of Treatment

[00402] Methods of the present invention for treating or preventing an ophthalmic condition include inserting more than 5 sustained release drug loaded gels (e.g., implants or mesoparticles) intraorbitally to treat or prevent the ophthalmic condition for more than 2 weeks. Methods of the present invention for treating or preventing an ophthalmic condition include inserting more than 10 sustained release drug loaded gels (e.g., implants or mesoparticles) intraorbitally to treat or prevent the ophthalmic condition for more than 2 weeks. Methods of the present invention for treating or preventing an ophthalmic condition include inserting more than 25 sustained release drug loaded gels (e.g., implants or mesoparticles) intraorbitally to treat or prevent the ophthalmic condition for more than 2 weeks. Methods of the present invention for treating or preventing an ophthalmic condition include inserting more than 50 sustained release drug loaded gels (e.g., implants or mesoparticles) intraorbitally to treat or prevent the ophthalmic condition for more than 2 weeks. Methods of the present invention for treating or preventing an ophthalmic condition include inserting more than 100 sustained release drug gels (e.g., implants or mesoparticles) intraorbitally to treat or prevent the ophthalmic condition for more than 2 weeks. Methods of the present invention for treating or preventing an ophthalmic condition include inserting more than 500 sustained release drug loaded gels (e.g., implants or mesoparticles) intraorbitally to treat or prevent the ophthalmic condition for more than 2 weeks. Methods of the present invention for treating or preventing an ophthalmic condition include inserting more than 1,000 sustained release drug loaded gels (e.g., implants or mesoparticles) intraorbitally to treat or prevent the ophthalmic condition for more than 2 weeks. Methods of the present invention for treating or preventing an ophthalmic condition include inserting more than 10,000 sustained release drug loaded gels (e.g., implants or mesoparticles) intraorbitally to treat or prevent the ophthalmic condition for more than 2 weeks. Methods of the present invention for treating or preventing an ophthalmic condition include inserting more than 100,000 sustained release drug loaded gels (e.g., implants or mesoparticles) intraorbitally to treat or prevent the ophthalmic condition for more than 2 weeks. Methods of the present

invention for treating or preventing an ophthalmic condition include inserting more than 1,000,000 sustained release drug loaded gels (e.g., implants or mesoparticles) intraorbitally to treat or prevent the ophthalmic condition for more than 2 weeks. The polymer composition and ratios of each implant/mesoparticle in these collections of small implants can be varied between implants within a single dose such that an aggregate degradation profile of the collection of implants is achieved for delivery of the active agent for greater than 2 weeks, greater than 1 month, greater than 2 months, greater than 3 months, greater than 4 months, greater than 6 months, greater than 9 months and greater than 12 months.

[00403] The following non-limiting examples illustrate certain aspects of the present disclosure.

[00404] **EXAMPLES**

[00405] **Example 1. Mold Fabrication for PRINT® Protein Microparticles (PuPs)**

[00406] A series of templated molds of various dimensions were fabricated by Envisia Therapeutics utilizing the PRINT® process.

[00407] Molds utilized had a cylindrical shape with a dimension of 1 μm in diameter x 1 μm in height.

[00408] **Example 2. Protein Micronization: PRINT® Protein Microparticles (PuPs)**

[00409] Protein was prepared as monodisperse microparticles.

[00410] **Reformulated Stock Solution**

[00411] Protein stock solutions were received from commercial manufacturers and reformulated with different ratios of excipients trehalose, phosphate buffer (pH 6.2), and polysorbate 20 (tween 20). Typically, the reformulated protein solution was prepared at 36 g/L trehalose, 50 mM sodium phosphate buffer (pH 6.2), and 1.6% w/w tween 20. The concentration of the protein in the reformulated protein solution is typically in the range of 30-60 g/L, e.g., 36 g/L.

[00412] A rinse buffer was prepared in 50mM sodium phosphate buffer (pH 6.0 - 6.2) with 36 g/L trehalose and 1.6% w/w tween 20. The rinse buffer was used to reformulate the protein stock solution using a tangential flow filtration (TFF) process or other similar buffer exchange process (dialysis, dead end filtration, etc.). Reformulation increases the protein to excipient ratio without significantly compromising the stability of the protein. This increases the downstream protein loading in the PuP. Reformulated protein solutions typically have protein to trehalose ratios of 1:1. The protein to trehalose ratio in the reformulated protein solution can be as high as 2:1 or higher. However, above a 2:1 ratio, stability of the protein may be lost. Generally, the protein concentration in the reformulated solution was 36 g/L. Concentrations ranging from 25-60 g/L have been used successfully. The relative amount of tween 20 was also increased to 1.6% (w/w). Increasing the concentration of tween 20 helps to stabilize the protein to interfaces and shear stresses that may occur during the PRINT® process. Tween 20 also helps with PuP release from the harvest array. The phosphate buffer was held mostly constant (relative to the stock solution) with a slight decrease in pH (6.0-6.2). Lowering the pH improved protein stability by increasing the electrostatic charge of the protein. In exemplary embodiments, the pH of phosphate buffer in the reformulated solution was 5.0-5.5. A number of solutions were made at this pH and showed improvements in protein stability. Phosphate and citrate buffers work well at 50 mM and pH 5.0. Once reformulated, the protein solution is stored at 4C until further use.

[00413] **Casting Protein**

[00414] The reformulated protein solution (80 g/L +/- 5 g/L total solids content) was cast on a continuous plastic sheet using a grooved mayer rod (#4). The film was at least partially dried and then at least partially re-humdified to aid in mold filling. Drying can be done under ambient forced air flow or under dry N_2 flow. Re-hydrating the film reduces the viscosity and processing temperature of the protein/excipient material. This allows the mold to be filled at 110F. The film was covered with the molds of Example 1. The mold can be filled at lower or higher temperatures (70-300F). However, temperatures above 140F (60C) can damage the protein due to thermal denaturation.

[00415] After the mold was filled, it was allowed to dry for between, for example, 2 minutes – 4 days. This rest period depended on the amount of water in the particles and parameters used to remove water. The rest period allowed water to diffuse out of the particles and into the polyethylene terephthalate (PET) and mold.

[00416] Next the mold and harvest array were split in a dry chamber (0-35%RH). Particles were removed from the harvest array. Particles were collected and stored at 4C. Water content in the particles was typically between 4-10 % (w/w).

[00417] **FIG. 9** depicts electron microscopy images of 1 μm x 1 μm particles containing either aflibercept or bevacizumab.

[00418] **Example 3. Protein Micronization: Lyophilization**

[00419] Protein was prepared as polydisperse microparticles using lyophilization.

[00420] **Reformulated Protein Solution**

[00421] Protein stock solutions were received from commercial manufacturers and reformulated with different ratios of excipients trehalose, phosphate buffer (pH 6.2), and polysorbate 20 (tween 20). Typically, the reformulated protein solution was prepared at 25 g/L trehalose, 50 mM phosphate buffered saline (pH 6.2), and 0.4% w/w tween 20. The concentration of the protein in the reformulated solution is typically in the range of 30-60 g/L, e.g., 56 g/L.

[00422] A rinse buffer was prepared in 50mM PBS (pH 6.2) with 25 g/L trehalose and 0.4% w/w tween 20. The rinse buffer was used to reformulate the protein stock solution using a tangential flow filtration (TFF) process or other similar buffer exchange process (dialysis, dead end filtration, etc.). Reformulation increases the protein to excipient ratio without significantly compromising the stability of the protein. This increases the downstream protein loading. Reformulated protein solutions typically have protein to trehalose ratios of 1:1 to 2:1. However, above a 2:1 ratio, stability of the protein may be lost. Generally, the protein concentration in the reformulated solution was 56 g/L. Concentrations ranging from 25-60 g/L have been used successfully. The relative amount of tween 20 and phosphate buffer were held mostly constant (relative to

the stock solution). Once reformulated, the protein solution is stored at 4C until further use.

[00423] The reformulated protein stock was aliquoted into small vials (1 – 20 mL) to aid in lyophilization. In some cases the protein solution was flash frozen in liquid nitrogen and then placed under vacuum overnight. In other instances the protein solution contained within vials was placed in the tray lyophilizer and frozen. Bulk water was removed using a primary vacuum. Lyophilization was run such that the liquid was substantially removed from the protein. Water content for lyophilized samples was typically lower than for PuPs (0.01-5.0% w/w).

[00424] Lyophilization of the protein was followed by reducing average particle size through:

A. Sieving particles through a series of decreasing mesh sizes, from 250 µm down to 20 µm.

B. Breaking apart particles using a mortar and pestle.

[00425] **Example 4. Protein Micronization: Spray dry**

[00426] Protein was prepared as polydisperse microparticles using spray drying. A reformulated solution of protein (as prepared in Example 3) was spray dried to produce particles 20 µm in length or smaller.

[00427] **Example 5. Mold Fabrication for Implants**

[00428] A series of templated molds of various dimensions were fabricated by Envisia Therapeutics utilizing the PRINT® process.

[00429] Molds utilized included: a) a rod shape with dimensions of 225 x 225 x 2925 µm; b) a rod shape with dimensions of 311 x 395 x 6045 µm; and c) a rod shape with dimensions of 600x600x1000 µm.

[00430] **Example 6. Mold Fabrication for Mesoparticles**

[00431] A series of templated molds of various dimensions were fabricated by Envisia Therapeutics utilizing the PRINT® process.

[00432] Molds utilized included: a) a cube shape with dimensions of 50 x 50 x 50 μm; b) a cube shape with dimensions of 25 x 25 x 25 μm; and c) a rectangular prism shape with dimensions of 12.5 x 12.5 x 25 μm.

[00433] **Example 7. Hydrogel-Based Implant Fabrication**

[00434] **Hydrogels**

[00435] Solutions comprising the first precursor, the second precursor, and the therapeutic agent were prepared. The solutions were added to a mold (e.g., cylindrical or PRINT®) of desired dimensions, and mixed to form hydrogels. Optionally, an initiator was added to catalyze the reaction. Different solvent systems were used.

[00436] **Degradable Hydrogels**

[00437] *Organic, Hydrophobic Solvent*

[00438] Approximately 50 mg of PEG-NHS (e.g., 4-10-SG, 4 arm-10,000g/mol PEG-NHS) were weighed into a 2 mL glass vial. The PEG-NHS was then dissolved in dry dichloromethane (DCM) to a concentration of 90 g/L (concentrations from 50-250 g/L have been used). An equimolar, or slight molar excess (1.01:1), of PEG amine (e.g. 4-10-NH2, 4 arm-10,000 g/mol PEG-NH2) was measured into a separate glass vial. PuPs were weighed into the same glass vial as the PEG amine. PuP to total PEG ratio was used to determine percent theoretical loading. For example 50 mg PuPs plus 50 mg total PEG would give a loading of 50% (w/w). DCM was then added to this vial to obtain the same concentration as the PEG-NHS. Triethylamine (TEA) was added to the second vial containing the PEG-NH2. The TEA activates the base and helps the reaction proceed. Typically an equimolar ratio of TEA to NH2 is used. However, lower and higher levels have been used (e.g., 0.1:1 and 4:1 TEA:NH2).

[00439] The suspension of PuP, PEG-NH2, and TEA in DCM was vortexed for 30+ seconds to substantially break down any aggregates of PuPs. Next the PEG-NHS/DCM solution was transferred into the PuP suspension. The single suspension was vortexted for 5-15 seconds. The suspension was then drawn up into a cylindrical mold of specific inner diameter. The diameter is typically 1 mm, however diameters smaller and larger have been used as well (0.1 – 3 mm diameter). The suspension can also be used to

fill a PRINT® mold. Initial onset of the gel suspension can take anywhere from seconds to hours. Typically onset takes about 40-90 seconds. The gelling onset time can be tuned by adding adjusting the amount TEA to the reaction. Higher TEA concentrations make the reaction proceed more quickly. The gelling reaction was then allowed to proceed overnight in the dark at room temperature. The gelling reaction can be further accelerated by increasing the heat.

[00440] *Organic, Hydrophilic Solvent*

[00441] Approximately 50 mg of PEG-NHS (e.g., 4-10-SG, 4 arm-10,000g/mol PEG-NHS) were weighed into a 2 mL glass vial. The PEG-NHS was then dissolved in either Acetonitrile (ACN) or Ethyl Lactate (EL) to a concentration of 90 g/L (concentrations from 50-250 g/L have been used). An equimolar, or slight molar excess (1.01:1), of PEG amine (e.g. 4-10-NH2, 4 arm-10,000 g/mol PEG-NH2) was measured into a separate glass vial. PuPs were weighed into the same glass vial as the PEG amine. PuP to total PEG ratio was used to determine percent theoretical loading. For example 50 mg PuPs plus 50 mg total PEG would give a loading of 50% (w/w). ACN or EL was then added to this vial to obtain the same concentration as the PEG-NHS. Triethylamine (TEA) was added to the second vial containing the PEG-NH2. The TEA activates the base and helps the reaction proceed. Typically an equimolar ratio of TEA to NH2 is used. However, lower and higher levels have been used (e.g., 0.1:1 and 4:1 TEA:NH2).

[00442] The suspension of PuP, PEG-NH2, and TEA in ACN or EL was vortexed for 30+ seconds to substantially break down any aggregates of PuPs. Next the PEG-NHS/ACN or EL solution was transferred into the PuP suspension. The single suspension was mixed by vortex for 5-15 seconds. The suspension was then drawn up into a tube of specific inner diameter. The diameter is typically 1 mm, however diameters smaller and larger have been used as well (0.1 – 3 mm diameter). The suspension can also be used to fill a PRINT® mold. Initial onset of the gel suspension can take anywhere from seconds to hours. Typically onset takes about 40-90 seconds. The gelling onset time can be tuned by adding adjusting the amount TEA to the reaction. Higher TEA concentrations make the reaction proceed more quickly. The gelling

reaction was then allowed to proceed overnight in the dark at room temperature. The gelling reaction can be further accelerated by increasing the heat.

[00443] Non-Degradable Hydrogels

[00444] To a glass vial, approximately 100 mg of PEG-acrylate (or blend of PEG-acrylates) and an appropriate amount of 100 mg/mL bovine gamma globulin (BGG) solution in 1X PBS were massed such that the wt.% of BGG to PEG was about 20 wt.% (e.g. 250 uL of a 100 mg/mL BGG solution to 100 mg of PEG-acrylate). Additionally, a porogen, such as PEG400 was added to the system. To the liquid suspension, liquid DPT was added such that the wt.% of DPT to (DPT + PEG-acrylate) is 1 wt.%. The suspension was vortexed to mix the cloudy liquid suspension. Using a clear cylindrical mold with an inner diameter of ~1 mm, a syringe was used to pull the suspension up through the tubing. The tubing was exposed to UV-light for ~10s to photochemically cure. The solid implants were removed and the tubbing was cut to appropriate lengths. Tweezers or a push-rod were used to remove the elastomer from the cylindrical mold.

[00445] In embodiments, low-molecular weight PEG macromers (e.g. PEG-diacrylate Mw=400) were photochemically crosslinked by adding a photoinitiator (such as Diphenyl(2,4,6-trimethylbenzoyl)phosphine oxide/2-hydroxy-2-methylpropiophenone, blend (DPT))to the liquid pre-polymer and exposing to appropriate wavelength of light (typically peak λ=365 nm). High-molecular weight PEG macromers (e.g. Mw>1000) were dissolved in water at an appropriate wt.% (e.g. 1-50 wt.%), photoiniator added, and exposed to the appropriate wavelength of light to form a crosslinked hydrogel. Additionally, unfunctional PEG can be added to the system (e.g. PEG400) that results in the formation of a softer gel.

[00446] Protein was incorporated into the polymer system by either adding an aqueous solution of protein (e.g. Avastin, BGG in 1X PBS) to the liquid PEG or aqueous PEG solution (containing photoinitiator), or, by adding solid state protein powder to the liquid PEG or aqueous PEG solution. The PEG causes the protein to precipitate, and a cloudy suspension is formed. It is important that the protein phase separates from the liquid PEG suspension, to protect the protein from being exposed to UV light while in solution.

[00447] The resulting suspension was then extruded through a clear cylindrical mold whereupon it was exposed to UV light to initiate the crosslinking reaction. The resulting tubular elastomers has an outer diameter that is equal to the inner diameter of the tubing it was extruded through. By altering the tubing size, it is possible to change the diameter of the resulting implant. Cylindrical implant length is controlled by cutting the tubing after formation to the appropriate length (e.g. 5 mm).

[00448] Therapeutic agent, or protein, release is dependent upon many factors, including but not limited to the overall crosslink density of the polymer, the phase domain size of the protein in the crosslinked hydrogel network, and the size of the surface area to volume ratio of the implant.

[00449] **Example 8. Wax Implant Fabrication**

[00450] Fabricating wax-fatty acid implants generally include three steps: (1) weighing and mixing wax and fatty acid components; (2) mixing the wax-fatty acid mixture with solid state PRINT® protein particles (described herein) under elevated temperature; and (3) forming the wax-fatty acid/solid state PRINT® protein microparticle (PuPs) mixture into a desired final form factor.

[00451] Once the wax and fatty acid materials are weighed and put into a vial, they are heated up past 70C so as to become a non-viscous liquid. The solution is kept in a heat bath at 70C for forming into final form factor when needed. Solid state PRINT® protein particle Pups are weighed out at 5-15% wt of the total formulation and mixed into the melted wax-fatty acid solution prior to forming the final form factor of the implant for delivery.

[00452] In one embodiment, once the protein particles were homogeneously distributed throughout the liquid solution the total solution was melt extruded into tubing of various diameters, for example, about 700-800 micrometers. The solution with suspended protein particles was allowed to solidify at lower temperature thereby forming the final implants which are removed and characterized or used in vitro or in vivo. Waxy-fatty acid implants can also be fabricated utilizing PRINT® molds.

[00453] **Example 9. Implant Fabrication: PRINT® Cylindrical Implants**

[00454] PRINT® cylindrical implants were prepared as disclosed in Example 7 using DCM, ACN, or EL. The PEG/protein solution was drawn into plastic tubing to form a cylindrical shaped implant. The cylindrical tube had an inner diameter of 1 mm. The gelling reaction was allowed to proceed, and the implants were cut to size, e.g., 3 mm in length. This method was used to prepare PRINT® cylindrical implants comprising bevacizumab and PRINT® cylindrical implants comprising aflibercept.

[00455] **Example 10 Implant Fabrication: PRINT® Implants**

[00456] PRINT® rod shaped implants were prepared as disclosed in Example 7 using EL. The PEG/protein in EL was formed as rod shaped implants using the PRINT® process and utilizing the molds in Example 5. This method was used to prepare PRINT® implants containing bevacizumab from Example 2 and 3.

[00457] **FIG. 7A-B** shows scanning electron micrographs (SEM) of a PRINT® rod shaped implant containing 1 μm x 1 μm PRINT bevacizumab microparticles.

[00458] **Example11. Mesoparticle Fabrication**

[00459] Mesoparticles were prepared as disclosed in Example 7 using EL. The PEG/protein in EL was formed as cube and rectangular prism shaped mesoparticles using the PRINT® process and utilizing the molds in Example 6. This method was used to prepare PRINT® mesoparticles containing bevacizumab from Example 2 or aflibercept from Example 4.

[00460] **FIG. 8A-B** shows scanning electron micrographs (SEM) of PRINT® mesoparticles containing 1 μm x 1 μm PRINT bevacizumab microparticles.

[00461] **Example 12. PRINT® Cylindrical Implant with Lyophilized Aflibercept**

[00462] A series of implants were prepared using the polydisperse aflibercept particles of Example 3 and the cylindrical implant process of Example 9. The implants are shown in **FIG. 10** and below in **Tables 1A** and **1B**.

[00463] Table 1A. Composition and Mold Size for AFB13

Sample ID	PEG Ester	PEG Amine	PEG Ester wt%	PEG Amine wt%	Lyophilized particle wt%	Target Protein wt%	Mold Diameter (mm)
ENV-1P-0205-46-A	8ARM-SAZ-10K	8ARM-NH2-10K/ 4ARM-NH2-10K (50/50)	10	10	80	52.8	1 mm

[00464] Table 1B. Composition and Mold Size for AFB19C

Sample ID	PEG Ester	PEG Amine	PEG Ester wt%	PEG Amine wt%	Lyophilized particle wt%	Target Protein wt%	Mold Diameter (mm)
RES-PRO-0021-22-C	8ARM-SAZ-10K	8ARM-NH2-10K/ 4ARM-NH2-10K (50/50)	10	10	80	53.4	1 mm

[00465] Example 13. PRINT® Cylindrical Implant + PRINT® Aflibercept Microparticles

[00466] A series of implants were prepared using the monodisperse aflibercept particles of Example 2 and the cylindrical implant process of Example 9. These implants are shown in **FIG. 11** and below in **Tables 2A-2C**.

[00467] **Table 2A. Composition and Mold Size for AFB19A/B**

Sample ID	PEG Ester	PEG Amine	PEG Ester wt%	PEG Amine wt%	PRINT particle wt%	Target Protein wt%	Mold Diameter (mm)
RES-PRO-0021-22-A	8ARM-SAZ-10K	8ARM-NH2-10K	10	10	80	36.3	1 mm
RES-PRO-0021-22-B	8ARM-SAZ-10K	8ARM-NH2-10K/ 4ARM-NH2-10K (50/50)	10	10	80	36.3	1 mm

[00468] **Table 2B. Composition and Mold Size for AFB 22**

Sample ID	PEG Ester	PEG Amine	PEG Ester wt%	PEG Amine wt%	PRINT particle wt%	Target Protein wt%	Mold Diameter (mm)
RES-PRO-0021-68-A	8ARM-SAZ-10K	8ARM-NH2-10K/ 4ARM-NH2-10K (50/50)	10	10	80	36.3	1 mm
RES-PRO-0021-68-B	8ARM-SAZ-10K	4ARM-NH2-10K	10	10	80	36.3	1 mm
RES-PRO-0021-68-C	8ARM-SAZ-10K	8ARM-NH2-10K/ 4ARM-NH2-10K (25/75)	10	10	80	36.3	1 mm
RES-PRO-0021-68-D	8ARM-SAZ-15K	4ARM-NH2-10K	10	10	80	36.3	1 mm

[00469] Table 2C. Composition and Mold Size for AFB 23

Sample ID	PEG Ester	PEG Amine	PEG Ester wt%	PEG Amine wt%	PRINT particle wt%	Target Protein wt%	Mold Diameter (mm)
RES-PRO-0021-68-G	4ARM-SG-10K	4ARM-NH2-10K	10	10	80	36.3	1 mm
RES-PRO-0021-68-H	4ARM-SAP-10K	8ARM-NH2-10K	10	10	80	36.3	1 mm
RES-PRO-0021-68-I	Implant 1: 4ARM-SG-10K Implant 2: 4ARM-SAP-10K	Implant 1: 4ARM-NH2-10K Implant 2: 8ARM-NH2-10K	10	10	80	36.3	1 mm

[00470] Example 14. Aflibercept In-vitro Release Analysis

[00471] In vitro release of aflibercept was determined for the implants of Example 11 - 12. Three implants cut to 3 mm length were placed into a 2 mL HPLC vial and were incubated at 37C in 1 mL of 1X PBS. At each time point of interest, the media was removed for analysis. The media was then replaced with 1 mL of fresh media. The media that was removed was analyzed for aflibercept released by $VEGF_{165}$ ELISA.. See **FIG. 4E-4L**. for in vitro release of select formulations.

[00472] Example 15. Analysis of Aflibercept Monomer and Aggregates

[00473] Protein particles of Example 2 were analyzed for monomer, dimer, and higher molecular weight aggregates. A weight of 15-20 mg of protein particles were weighed into a 2mL HPLC vial and dissolved in 1 mL of PBS. The dissolved powder was analyzed by size exclusion chromatography (SEC). The results are presented below in **Table 4** and in **FIG. 12**

[00474] Table 4. SEC Analysis of Aflibercept PRINT® Microparticles

Sample ID	% Monomer	% Dimer	% High Molecular Weight
RES-PRO-0025-4-1	96.8	3.2	0

[00475] The results of this experiment indicate that protein with high monomeric content (e.g. > 95% monomer) and minimal aggregation can be formed with the PRINT process.

[00476] Example 16. ENV1305 Aflibercept Intravitreal Implant Nonclinical Studies

[00477] The present example demonstrates an embodiment of the disclosure, termed ENV1305, which is an aflibercept intravitreal implant for the treatment of AMD, RVO, and DME. Embodiments of the aflibercept intravitreal implant are provided above in Example 13 and **Tables 2A-2C**.

[00478] ENV1305 aflibercept intravitreal implant is an injectable aflibercept implant formulation using a biocompatible hydrogel-based drug delivery system. The implant is designed for ophthalmic administration via intravitreal injection with a duration of action of 4-6 months. The drug delivery system is comprised of a blend of PEGs that function as binder and release modifier, and was designed for slow erosion of the implant with concurrent release of aflibercept.

[00479] The bioavailability and sustained therapeutic effect of ENV1305 over 4-6 months is governed by multiple factors, including route of administration, erosion of the implant, and the physicochemical properties of the drug substance aflibercept.

[00480] The present example illustrates the functionality of various formulation of ENV1305 intravitreal implants.

[00481] **Characterization and Overview of ENV1305**

[00482] ENV1305 is a biocompatible implant formulation containing aflibercept in a PEG-based drug delivery system. ENV1305 is formulated as a solid, rod-shaped implant of dimensions between 350 μm to 400 μm in diameter by 3,000 μm in length as a dry (non-hydrated) implant.

[00483] ENV1305 implants can be loaded into the needle of a single-use implant applicator and delivered directly into the vitreous. ENV1305 was designed to deliver therapeutic concentrations of aflibercept for approximately 4-6 months in certain aspects.

[00484] Following intravitreal insertion, ENV1305 implants are retained in the vitreous, remain largely immobile, and disintegrate over time.

[00485] **Potential Mechanism of Action for ENV1305 Embodiments**

[00486] Without wishing to be bound to a particular mechanistic theory of action, the following description provides one possible mechanism of action for the ENV1305 ocular implants disclosed herein.

[00487] Vascular endothelial growth factor-A (VEGF-A) and placental growth factor (PlGF) are members of the VEGF family of angiogenic factors that can act as mitogenic, chemotactic, and vascular permeability factors for endothelial cells. VEGF acts via two receptor tyrosine kinases, VEGFR-1 and VEGFR-2, present on the surface of

endothelial cells. PlGF binds only to VEGFR-1, which is also present on the surface of leucocytes. Activation of these receptors by VEGF-A can result in neovascularization and vascular permeability. Aflibercept acts as a soluble decoy receptor that binds VEGF-A and PlGF, and thereby can inhibit the binding and activation of these cognate VEGF receptors.

[00488] The ENV1305 embodiments can decrease neovascularization and vascular permeability associated with AMD, RVO, and DME for approximately 4-6 months following a single administration via intravitreal injection, by effectively delivering aflibercept to targeted areas of the eye in a sustained manner.

[00489] **Nonclinical Pharmacokinetics of ENV1305 in Non-Human Primate**

[00490] One nonclinical pharmacokinetics study of ENV1305, ENV1305-PRE-001, was conducted in the African Green Monkey to assess the duration of aflibercept exposure following a single administration of ENV1305. African Green Monkeys, 1-2 males/group/terminal time point, were administered a single bilateral intravitreal injection of either ENV1305 or EYLEA and were followed for up 1, 1.5, 2, 3, and/or 5 months.

[00491] **Bioanalytical Methods in Support of ENV1305-PRE-001**

[00492] Bioanalytical methods for aflibercept were developed and qualified in albino rabbit matrices by Intertek Pharmaceutical Services using enzyme-linked immunosorbent assay (ELISA). Methods in monkey aqueous humor, vitreous humor, and plasma were qualified for range of reliable response, selectivity, carryover assessment, and precision and accuracy. The formulation tested in the ENV1305-PRE-001 study are provided below in **Table 5**.

[00493] Table 5. Formulations Tested in Nonclinical Pharmacokinetics Study in NHP

Formulation ID	µg Aflibercept/ Implant	No. of Implants / Eye	Total Dose (µg / Eye)	Study Number	Matrices Analyzed
ENV1305 Placebo (ENV-1P-0185-060-002)	0	2	0	ENV1305-PRE-001	Aqueous humor, vitreous humor, plasma
ENV1305 (ENV-1P-0185-045-003)	424	2	848		

[00494] Ocular and Systemic Pharmacokinetics From Pharmacokinetics Study ENV1305-PRE-001

[00495] African Green Monkeys (1-2 males per group per terminal time point) were administered a single bilateral administration of either ENV1305 or EYLEA. Animals were followed for up to 3 months, with ocular matrices (aqueous humor, vitreous humor) and plasma collected to determine pharmacokinetics. A summary of the study design is presented below in **Table 6**.

[00496] Table 6. Study Design for ENV1305-PRE-003 Nonclinical Tolerability and Pharmacokinetics Study

Group	Number of Animals	Test Article	Dose (ug/eye)	No. of Implants /Eye	Terminal Time Points	Terminal Samples
1	2	ENV1305 Placebo	0	2	NA	Aqueous Humor, Vitreous Humor, Plasma
2	2	EYLEA	2000	0	Month 1, Week 6	
3	4	ENV1305	848	2	Months , 2, 3	

[00497] The concentration data is provided below in **Tables 7**, **8**, **9**, and **13**, and in **FIG. 23-25**.

[00498] Table 7. Aflibercept Concentration Data From Nonclinical Study ENV1305-PRE-001: Plasma

Days	Group 1			Group 2			Group 3		
	Mean	SD	N	Mean	SD	N	Mean	SD	N
0	BLQ	NA	2	BLQ	0.000	1	BLQ	NA	4
1	BLQ	NA	2	1000.500	713.1172	2	BLQ	NA	4
7	BLQ	NA	2	1443.500	226.2742	2	BLQ	NA	4
14	BLQ	NA	2	551.800	524.249	2	BLQ	NA	4
21	BLQ	NA	2	ND	ND	ND	BLQ	NA	4
28	BLQ	NA	2	BLQ	NA	2	BLQ	NA	4
42	BLQ	NA	2	BLQ	NA	1	BLQ	NA	3
56	ND	ND	ND	ND	ND	ND	BLQ	NA	3
70	ND	ND	ND	ND	ND	ND	BLQ	NA	2
84	ND	ND	ND	ND	ND	ND	BLQ	NA	2

Note: BLQ: below limit of quantification; NA: not applicable; ND: no data

[00499] Table 8. Aflibercept Concentration Data From Nonclinical Study ENV1305-PRE-001: Aqueous Humor

Days	Group 1			Group 2			Group 3		
	Mean	SD	N	Mean	SD	N	Mean	SD	N
14.000	BLQ	NA	4	3730.000	6210.000	4	229.600	169.1918	8
28.000	BLQ	NA	4	357.1875	422.5273	4	134.050	114.8178	8
31.000	ND	ND	ND	ND	ND	ND	BLQ	NA	2
42.000	BLQ	0.000	4	36.2125	42.19515	4	219.2417	107.5501	6
56.000	ND	ND	ND	ND	ND	ND	96.03333	50.80379	6
63.000	ND	ND	ND	ND	ND	ND	BLQ	NA	2
70.000	ND	ND	ND	ND	ND	ND	108.5875	89.81283	4
84.000	ND	ND	ND	ND	ND	ND	144.700	114.8544	4
85.000	ND	ND	ND	ND	ND	ND	BLQ	NA	4

Note: BLQ: below limit of quantification; NA: not applicable

[00500] Table 9. Aflibercept Concentration Data From Nonclinical Study ENV1305-PRE-002: Vitreous Humor

Days	Group 2			Group 3		
	Mean	SD	N	Mean	SD	N
28.000	10360.000	3712.311	2	ND	ND	ND
31.000	ND	ND	ND	8060.000	11398.560	2
42.000	626.875	73.36233	2	ND	ND	ND
63.000	ND	ND	ND	113025.000	43310.290	2
85.000	ND	ND	ND	54657.150	62694.340	4

Note: BLQ: below limit of quantification; NA: not applicable; ND: no data

[00501] Data collected to date demonstrate that aflibercept concentrations in vitreous humor were at or above therapeutically relevant concentrations in both EYLEA and ENV1305 groups at Month 1. By Week 6, aflibercept decreased significantly in the eyes administered EYLEA. In contrast, high concentrations remained in the vitreous humor samples in the ENV1305 group at Months 2 and 3, indicating that the implants had not yet released all test article at that last time point.

[00502] Exposure to aflibercept in the aqueous humor was observed at high concentrations on Days 14 and 28 in the EYLEA group and had decreased by Day 28, whereas the concentrations detected following ENV1305 administration wwere low and consistent over time. This indicates decreased exposure to aflibercept in the anterior segment at the early time points and a decreased Cmax following ENV1305 administration when compared with EYLEA.

[00503] Aflibercept was detected through Month 1 in the EYLEA group (mean Cmax = 1334 ng/mL), and was below the limit of quantitation in all other samples from all groups at all time points. These data demonstrate increased systemic exposure following EYLEA administration compared with ENV1305, as there were no quantifiable plasma samples after ENV1305 dose administration.

[00504] Data from this study indicate that ENV1305 retains and releases aflibercept for as many as about 3 months with the potential to continue releasing beyond the last time point in this study, while EYLEA shows minimal aflibercept remaining by Week 6. Exposure to aflibercept in aqueous humor for ENV1305 was decreased when compared to EYLEA, but sustained over time, while systemic exposure for ENV1305 was not quantifiable, in contrast to the higher concentrations seen with EYLEA. The trend suggests that ENV1305 implants will continue to release aflibercept for about 4-6 months, and may provide a superior safety profile compared with EYLEA.

[00505] **Example 17. PRINT® Cylindrical Implant with Lyophilized Bevacizumab**

[00506] A series of implants were prepared using the lyophilized polydisperse bevacizumab particles of Example 3 and the cylindrical implant process of Example 9. Said implant formulations are provided below in **Table 10**.

[00507] Table 10. PRINT® Cylindrical Implant with Lyophilized Bevacizumab

Sample ID	PEG Ester	PEG Amine	PEG Ester wt%	PEG Amine wt%	Lyophilized particle wt%	Target Protein wt%	Mold Diameter (mm)
ENV-1A-0084-32-I	8ARM-SAZ-10K	8ARM-NH2-10K	12	12	76	37.8	1 mm
ENV-1A-0103-03-D	8ARM-SAZ-10K	8ARM-NH2-10K	25	25	50	13.0	1 mm

[00508] Example 18. PRINT® Cylindrical Implant with PRINT® Bevacizumab Microparticles

[00509] A series of implants were prepared using the PRINT® monodisperse bevacizumab particles of Example 2 and the cylindrical implant process of Example 9. Said implants are shown in **FIG. 6A-B** and below in **Table 11**.

[00510] **Table 11. PRINT® Cylindrical Implants with PRINT® Bevacizumab Microparticles.**

Sample ID	PEG Ester	PEG Amine	PEG Ester wt%	PEG Amine wt%	PRINT particle wt%	Target Protein wt%	Mold Diameter (mm)
ENV-1A-0050-09-E	8ARM-SAZ-15K	4ARM-NH2-10K	35	35	30	13.1	1 mm
ENV-1A-0050-09-G	8ARM-SAZ-15K	8ARM-NH2-10K/ 4ARM-NH2-10K (50/50)	35	35	30	13.0	1 mm
ENV-1A-0050-09-H	8ARM-SAZ-15K	8ARM-NH2-10K/ 4ARM-NH2-10K (75/25)	35	35	30	12.9	1 mm
ENV-1A-0050-09-K	8ARM-SAZ-15K	8ARM-NH2-10K	35	35	30	12.8	1 mm
ENV-1A-0050-09-L	8ARM-SAZ-15K	8ARM-NH2-10K	35	35	30	12.8	1 mm
ENV-1A-0050-09-O	4ARM-SAP-10K	8ARM-NH2-10K	35	35	30	12.7	1 mm
ENV-1A-0050-29-B	8ARM-SAZ-15K	8ARM-NH2-10K	25	25	50	21.7	1 mm
ENV-1A-0089-14-B	8ARM-SAZ-10K	8ARM-NH2-10K	15.5	15.5	69	40.0	1 mm
ENV-1A-0089-14-C	8ARM-SAZ-10K	8ARM-NH2-10K	10.5	10.5	79	46.0	1 mm

[00511] **Example 20. In-vitro Release of Bevacizumab**

[00512] In vitro-release of bevacizumab was determined for the implants of Examples 17 and 18. Three implants cut to 3 mm length were placed into a 2 mL HPLC vial and were incubated at 37C in 1 mL of 1X PBS. At each time point of interest, the media was removed for analysis. The media was then replaced with 1 mL of fresh media. The media that was removed was analyzed for bevacizumab released by $VEGF_{165}$ ELISA. See **FIG. 5A-5B**. for in vitro release of select formulations.

[00513] **FIG. 5B** demonstrates the improved linearity of release as demonstrated with the micronized PRINT formulation vs the lyophilized formulation.

[00514] **Example 21. Analysis of Bevacizumab Monomer and Aggregates**

[00515] Bevacizumab particles of Example 2 were analyzed for monomer, dimer, and higher molecular weight aggregates. A weight of 10 mg of protein particles were weighed into a 2mL HPLC vial and dissolved in 1 mL of PBS. The dissolved powder was analyzed by size exclusion chromatography (SEC). The results are presented below in **Table 12** and **FIG. 13**.

[00516] **Table 12. Analysis of Bevacizumab Monomer and Aggregates**

Sample ID	% Monomer	% Aggregates (dimer and HMW)
ENV-1A-0084-83B	95.7	4.3

[00517] **Example 22. Bevacizumab In-vivo Release Studies: RV to complete**

[00518] **Example 23. Bevacizumab In-vitro/in-vivo Correlation**

[00519] **FIG. 14** depicts the in-vitro release as related to the in-vivo release of protein in rabbit eyes for sample 050-09-D.

[00520] **Example 24. Leakage Measured from Fluorescein Angiograms**

[00521] Leakage of blood from retinal vasculature and neovascularization of the retina was measured was measured using fluorescein angiography. Assessments were conducted and the images were graded according to **Table 13**.

[00522] **Table 13. Standardized Fluorescein Angiogram Scoring**

Score	Characteristics
0	Major vessels very straight with some tortuosity of smaller vessels
1	Increased tortuosity of major vessels and or vessel dialation
2	Leakage between major vessels
3	Leakage between major and minor vessels, minor vessels still visible
4	Leakage between major and minor vessels, minor vessels are not visible

[00523] **Example 25: In vitro Release of Active Agent**

[00524] Individual implants were incubated in 1 mL of 1X phosphate buffered saline (PBS) at pH 7.4 at 37°C. The PBS was removed at the time point of interest and replaced with fresh PBS. PBS was analyzed for total protein release using a bicinchoninic assay (BCA) assay and functional protein release using enzyme-linked immunosorbent assay (ELISA).

[00525] **Example 26: Total Protein Bradford Assay**

[00526] Total protein was detected using a Coomassie (Bradford) Protein Assay Kit (Thermo Scientific). A standard curve was generated using aflibercept solution diluted to 100 μg/mL and then serial diluted two fold.

[00527] A volume of in vitro release sample was combined with an equal volume of the Bradford reagent and incubated for 5 minutes at room temperature. Samples were analyzed using the Spectramax M5 plate reader at 595 nm. The resulting data was analyzed using analytical software.

[00528] The amount of aflibercept released as measured using a Bradford Assay is shown in **FIG. 4C**, **4D**, **4H**, and **4I**.

[00529] **Example 27: Enzyme-linked Immunosorbent Assay (ELISA)**

[00530] Functional protein was determined using a VEGF Sandwich ELISA.

[00531] Well plates were coated with VEGF A_{165} protein in Coating Buffer (0.05 M carbonate-bicarbonate, pH 9.6) overnight at 4°C at a specified concentration. Plates were washed with Wash Buffer (50 mM Tris, 0.14 M NaCl, 0..05% Tween 20, pH 8.0) and blocked for one hour at room temperature in Blocking Buffer (50 mM Tris, 0.14 M NaCl, 1% BSA, pH 8.0). Samples were diluted using Diluting Buffer (50 mM Tris, 0.14 M NaCl, 1% BSA, 0.05% Tween 20, pH 8.0) and added to the plates according to a plate map. A standard curve was generated using Avastin® (bevacizumab) Solution for Intravenous Infusion (Genentech, Inc.) diluted to 0.15 µg/mL and serially diluted. Plates were incubated for one hour at room temperature and washed.

[00532] The secondary antibody, HRP Conjugated Human IgG Detection Antibody, (Bethyl Labs) was added at a dilution of 1:100,000 and plates were incubated in the dark. After incubation, the plates were washed and then developed using TMB Substrate (Bethyl Labs). The substrate reaction was stopped after ten minutes using 0.18 M sulfuric acid and the plates were read using a Spectramax M5 at 450 nm. The resulting data was analyzed using analytical software.

[00533] **Example 28: Reverse Phase High Performance Liquid Chromatography (RP-HPLC) of Implant Content**

[00534] Individual implant samples were weighed and hydrolyzed at 120°C using 5.2 N trifluoroacetic acid (TFA) or 2.0 N hydrochloric acid (HCl). A standard curve was created using comparably hydrolyzed standards. Results were calculated on a percent by weight basis where applicable. Chromatography parameters are listed in **Table 14** and **Table 15**.

[00535] **Table 14. Chromatography Parameters for Hydrolyzed Implants**

Flow rate	1.5 mL/min
Injection volume	20 µL
Column Temperature	50.0°C
Detector	VWD or DAD, 280 nm
Run Time	10 min
HPLC Column	BetaBasic Phenyl: 5 µm, 150 Å, 4.6 X 150 mm
Mobile Phase A	0.1% TFA in water
Mobile Phase B	0.1% TFA in methanol

[00536] **Table 15. Chromatography Gradient**

Time (min)	% Mobile Phase A	% Mobile Phase B
0	95	5
3	95	5
4	5	95
7	5	95
7.5	95	5
9.5	95	5

[00537] **Example 29: Initial Content of Active Agent via Extraction**

[00538] Bevacizumab was extracted from implants using one of the following methods dependent upon implant type.

[00539] For hydrogel implants, individual implants were added to HPLC vials and 1 mL Tris buffer, pH 8.5 was added. The sample was placed in a rotating incubator at 51°C until the implant was dissolved, generally three to four days. Samples were then diluted and analyzed via RP-HPLC.

[00540] For wax implants, individual implants were added to HPLC vials and dissolved in 500 µL hexane. 1 mL of 10% NaCl in DI water was added and a liquid extraction was performed. Isopropanol was added to the matrix and the water layer was sampled for analysis by RP-HPLC.

[00541] For implants containing PLGA, individual implants were added to HPLC vials and dissolved in 100 µL acetonitrile. 500 µL of DI water was added and the solution was mixed by vortex. Samples were centrifuged to remove any solids and the supernatant was analyzed by RP-HPLC.

[00542] **Example 30: In vivo Release of Active Agent in Vitreous Humor (VH)**

[00543] At study termination, animal eyes were enucleated, frozen, and stored at -80°C until analysis. Prior to analysis, samples were removed from the freezer and thawed on ice for approximately one hour. Vitreous humor (VH) was removed and transferred into a petri-dish. Implants were isolated, retrieved, and transferred into a silonized glass vial until analysis. VH volume was measured using a positive displacement pipette and transferred to a 5 mL low protein binding Eppendorf cylindrical.

[00544] VH was homogenized mechanically. A volume of VH was transferred to 5 mL low protein binding Eppendorf cylindricals containing an equal volume of 1X PBS pH 7.4. Tungsten beads were added to aid in mixing. Cylindricals were shaken vigorously for 60 seconds to homogenize the VH.

[00545] The resulting sample was analyzed via ELISA.

[00546] **Example 31: In vivo Release of Active Agent: Retrieved Implant**

[00547] Retrieved implants were extracted as described previously in Example 29. Samples were analyzed via RP-HPLC and ELISA. PLGA samples were not analyzed via ELISA due to inactivation of the protein during extraction.

[00548] **Example 32: Model Protein Implants**

[00549] A series of implants were produced using bovine gamma globulin. **Table 16** details the implant composition. **Table 17** details the implant masses and active agent content.

[00550] **Table 16. Model Protein Implant Composition**

ID	Type	Composition
706-12-2B	Hydrogel	PEG-20k Tetraacrylate
706-12-3B	Hydrogel	PEG-20k Tetraacrylate/PEF-400 Diacrylate (50/50 by wt)
706-12-4B	Hydrogel	PEG-400 Diacrylate
706-14-3C	Hydrogel	PEG-10K Tetraacrylate w/20% by wt of PEG-400

[00551] **Table 17. Model Protein Implant Mass and Active Agent Content**

ID	Implant Mass mg			Active Agent %	Dose per Implant mg
	AVERAGE	STDEV	N		
706-12-2B	0.828	0.381	3	14.00%	0.116
706-12-3B	2.172	0.058	3	10.00%	0.217
706-12-4B	4.126	0.337	3	17.00%	0.701
706-14-3C	1.644	0.251	3	17.00%	0.279

[00552] **Example 33: Wax Implants**

[00553] A series of implants were produced using various waxes. **Table 18** details the composition of these implants.

[00554] **Table 18 – Wax Implant Composition**

ID	Type	Composition
708-87	Wax	Beeswax:Palmitic acid:Stearic acid:Caprylic acid 35:35:20:10, 10% PuPs
708-91	Wax	Beeswax:Tristearin:Palmitic acid:Capric acid:Caprylic acid 30:10:30:25:5, 10% PuPs
708-82-2-D	Wax	Beeswax:Palmitic acid:Stearic acid:Caprylic acid 35:35:20:10, 10% PuPs

[00555] **Example 34: In vivo Study: New Zealand White Rabbits**

[00556] A series of ocular implants were tested in a New Zealand White rabbit model. **Table 19** identifies the composition and size of the implants. **Table 20** shows the implant mass and active agent content. **Table 21** identifies the implant schedule for the study.

[00557] Implants were placed through a scleral incision into the vitreous of anesthetized eyes using forceps. One or two simple interrupted 9-0 vicryl sutures were used to close the conjunctival incision. For a positive control, 50 µL Avastin® (Genentech, Inc.) was injected. For a negative control, the procedure was conducted but no implant was inserted. For sample 708-69, one eye only was injected for the second animal. Also, for sample 708-69, one of the 10 implants for one eye fractured upon insertion. As a result, 9.5 implants were inserted.

[00558] **Table 19 – Implant Composition and Sizes**

ID	Type	Composition	Size Diameter X Length
1A-011-64	Hydrogel	8-10/4-10 SG, 5% solids, 15% PuPs	1.1 mm X 3.6 mm
708-69	Wax	Beeswax:Stearic acid 80:20, 10% PuPs	600-800 µm X 5 mm
1A-011-91-G	Hydrogel	8-10/4-10 SG, 15% solids, 15% PuPs	~1.3 mm X 3.6 mm
1A-003-15-A	PLGA	80:20 RG752S/Eicosanol, 15% lyophilized bevacizumab	380 µm X 6,000 µm
Avastin® (Genentech, Inc.)	Injection Solution	Vehicle	NA

Table 20 – Implant Mass and Active Agent Content

ID	Implant Mass mg	Active Agent %	Dose per Implant mg
1A-011-64	4.3	6.43%	0.276
708-69	2.5	6.29%	0.157
1A-011-91-G	12.7	4.60%	0.584
1A-003-15-A	1.1	7.62%	0.084
Avastin® (Genentech, Inc.)	50 µL	NA	1.250

[00559] **Table 21. New Zealand White Rabbit In Vivo Study**

ID	Number of Animals	Number of Eyes Total	Implants/Eye
1A-011-64	2	4	5
708-69	2	3	10

1A-011-91G	2	4	2
1A-003-15A	2	4	3
Avastin® (Genentech, Inc.)	2	4	50 µL/1.25 mg
Negative control	2	4	0

[00560] **FIG. 15** depicts the total ocular examination score over the course of the study. The total ocular examination score was determined using the McDonald-Shadduck Scoring System.

[00561] **FIG. 16** depicts the bevacizumab content in the aqueous humor at the termination of the study on day 12. It is shown in the figure that varying the composition, size, and/or shape of the implant varies the release of the bevacizumab.

[00562] **Example 35: In vivo Study: Dutch Belted Rabbits with $VEGF_{165}$ Challenge**

[00563] A series of ocular implants were tested in a Dutch Belted rabbit model. **Table 22** identifies the composition and size of the implants. **Table 23** shows the implant mass and active agent content. **Table 24** identifies the implant schedule for the study.

[00564] In this study, animals also experienced a challenge with $VEGF_{165}$ on day 12 of the study. Leakage was evaluated at both day 14 and day 28 of the study.

[00565] Implants were placed through a scleral incision into the vitreous of anesthetized eyes using forceps. One or two simple interrupted 8-0 vicryl sutures were used to close the conjunctival incision. For a positive control, 50 µL Avastin® (Genentech, Inc.) was injected. In a second set of animals, a second bevacizumab control sample was also injected. Positive controls were injected on day 10 of the study. For a negative control, the procedure was conducted but no implant was inserted.

[00566] **Table 22 – Implant Compositions and Sizes**

ID	Type	Composition	Size Diameter (mm)
706-130	PLGA	80:20 RG752S/Eicosanol, 15% lyophilized bevacizumab	0.54
706-130	PLGA	80:20 RG752S/Eicosanol, 15% lyophilized bevacizumab	0.54
1A-0029-86-A	Hydrogel	4-10/4-10 SG, 9% solids, 15% PuPs	1.65
1A-0029-86-A	Hydrogel	4-10/4-10 SG, 9% solids, 15% PuPs	1.65
708-78-3-A	Wax	Beeswax:Stearic acid 80:20, 10% PuPs	0.72
708-78-3-A	Wax	Beeswax:Stearic acid 80:20, 10% PuPs	0.72
1A-029-86-C-3	Hydrogel	8-10/4-10 SAP, 9% solids, 15% PuPs	1.5
1A-029-72-D	Hydrogel	4-10/4-10 SG, 10% solids, 30% PuPs	0.5
Avastin® (Genentech, Inc.)	Injection Solution	Vehicle	NA

[00567] **Table 23– Implant Mass and Active Agent Content**

ID	Implant Mass mg			Active Agent %	Dose per Implant mg
	AVERAGE	STDEV	N		
706-130	1.1664	0.1574	64	4.46%	0.052
706-130	1.1029	0.0801	8	4.46%	0.049
1A-0029-86-A	5.2489	0.3674	16	5.68%	0.298
1A-0029-86-A	4.9165	0.2314	8	5.68%	0.279
708-78-3-A	3.0882	0.3388	40	4.16%	0.128
708-78-3-A	2.9264	0.2543	8	4.16%	0.122
1A-029-86-C-3	5.1321	0.5016	24	6.37%	0.327
1A-029-72-D	0.5565	0.034	28	12.27%	0.068
Avastin® (Genentech, Inc.)	50 µL	NA	NA	NA	1.250

[00568] **Table 24 – Dutch Belted Rabbit In Vivo Study With Challenge**

ID	Number of Animals	Number of Eyes Total	Implants/Eye
706-130	4	8	8
706-130	4	8	1
1A-0029-86-A	4	8	2
1A-0029086-A	4	8	1
708-78-3-A	4	8	5
708-78-3-A	4	8	1
1A-0029-86-C-3	4	8	3
1A-0029-72-D	4	8	5
Avastin® (Genentech, Inc.)	2	4	50 µL/1.25 mg
Bezvcizumab control	2	4	1.25 mg
Negative control	2	4	0
$VEGF_{165}$ Challenge	All	All	25 µL/500 ng

[00569] **FIG. 17** depicts the total ocular examination score over the course of the study. The total ocular examination score was determined using the McDonald-Shadduck Scoring System.

[00570] **FIG. 18** depicts the bevacizumab content in the aqueous humor at the termination of the study on day 28. It is shown in the figure that varying the composition, size, and/or shape of the implant varies the release of the bevacizumab.

[00571] **FIG. 19A** depicts the leakage score tested according to Example 24 post challenge on day 14.

[00572] **FIG. 19B** depicts the leakage score tested according to Example 24 post challenge on day 28.

[00573] **Example 36: In vivo Study: Dutch Belted Rabbits with VEGF165 Challenge-56 Day Study**

[00574] A series of ocular implants were tested in a Dutch Belted rabbit model. **Table 25** identifies the composition and size of the implants. **Table 26** shows the implant mass and active agent content. **Table 27** identifies the implant schedule for the study.

[00575] In this study, animals also experienced a challenge with VEGF165 on day 26 or day 54 of the study. Leakage was evaluated at both day 28 and day 56 of the study.

[00576] Implants were placed through a scleral incision into the vitreous of anesthetized eyes using forceps. One or two simple interrupted 8-0 vicryl sutures were used to close the conjunctival incision. For a positive control, three doses of bevacizumab were injected into three separate animal groups. Doses included 1.25 mg, 25 μg, and 2.5 μg. Positive controls were injected on day 24 The positive control for the longer duration group was 1.25 mg and was injected on day 52 of the study. For a negative control, the procedure was conducted but no implant was inserted on day 24 or day 52 of the study.

[00577] **Table 25. Implant Composition and Sizes**

ID	Type	Composition	Size Diameter X Length
708-78-2	Wax	Beeswax:Palmitic acid:Stearic acid:Caprylic acid 35:35:20:10, 10 % PuPs	0.75 mm X 6 mm
050-09-D	Hydrogel	50-09-D, 4-10/4-10 SAP, 50% PuPs	0.425 mm X 3 mm
050-09-F	Hydrogel	50-09-F, 4-10(75)/8-10(25)/8-15 SAZ, 30% PuPs	0.425 mm X 3 mm
050-29-B	Hydrogel	050-29-B, 8-10/8-15 SAZ, 9% solids, 50% PuPs	0.425 mm X 3 mm
050-09-H	Hydrogel	50-09-H, 4-10(25)/8-10(75)/8-15 SAZ, 30% PuPs	0.430 mm X 3 mm

[00578] **Table 26. Implant Mass and Active Agent Content**

ID	Implant Mass mg			Active Agent %	Dose per Implant mg
	Average	STDEV	N		
708-78-2	3.063	0.0248	60	4.10%	0.126
050-09-D	0.545	0.045	120	18.10%	0.099
050-09-F	0.504	0.042	121	10.70%	0.054
050-29-B	0.562	0.058	120	20.70%	0.116
050-09-H	0.600	0.083	61	9.80%	0.059
Positive control 1	50 µL	NA	NA	NA	1.250
Positive control 2	50 µL	NA	NA	NA	0.025
Positive control 3	50 µL	NA	NA	NA	0.0025

[00579] Table 27. Dutch Belted Rabbit In Vivo Study With Challenge-56 Day Study

ID	Number of Animals	Number of Eyes Total	Implants/Eye	Study Group
708-78-2	4	8	5	28 day
050-09-D	4	8	5	28 and 56 day
050-09-F	4	8	5	28 and 56 day
050-29-B	4	8	5	28 and 56 day
050-09-H	4	8	5	56 day
Bezvcizumab control 1	2	4	1.25 mg	28 day
Bezvcizumab control 2	2	4	25 µg	28 day
Bezvcizumab control 3	2	4	2.5 µg	28 day
Negative control	2	4	0	28 and 56 day
$VEGF_{165}$ Challenge	All	All	25 µL/500 ng	28 and 56 day

[00580] **FIG. 20** depicts the total ocular examination score over the course of the study. The total ocular examination score was determined using the McDonald-Shadduck Scoring System.

[00581] **FIG. 21** depicts the bevacizumab content in the aqueous humor at the termination of the study on day 28. It is shown in the figure that varying the composition, size, and/or shape of the implant varies the release of the bevacizumab.

[00582] **FIG. 22A** depicts the leakage score tested according to Example 24post challenge on day 28.

[00583] **FIG. 22B** depicts the leakage score tested according to Example24 post challenge on day 56.

[00584] Implants recovered on day 28 were analyzed for residual bevacizumab using both RP-HPLC and ELISA. **Table 28** shows the results for analysis via RP-HPLC. **Table 29** shows the results for analysis via ELISA.

[00585] **Table 28. Active Agent Release at 28 Days-RP-HPLC Results**

ID	% Recovered				% Released	
	Total dosed AVE	Total dosed STDEV	Ave Implant AVE	Ave Implant STDEV	Of total dosed	Of average implant dosed
708-78-2	63	43	67	40	37	33
050-09-D	54	21	61	12	46	39
050-09-F	42	20	56	15	58	44
050-29-B	41	15	57	5	59	43

[00586] **Table 29. Active Agent Release at 28 Days-ELISA**

ID	% Recovered			
	Total dosed AVE	Total dosed STDEV	Ave Implant AVE	Ave Implant STDEV
708-78-2	63	43	67	40
050-09-D	41	18	46	13
050-09-F	15	15	20	18
050-29-B	30	14	40	6

[00587] **Example 37. Additional PRINT® Cylindrical Implants with Aflibercept Microparticles Formulations**.

[00588] Additionally, a series of implants were prepared using the polydisperse aflibercept particles of Example 3 and the cylindrical implant process of degradable hydrogel of Example 7 formed in the DCM solvent. The implants are shown below in **Tables 30**.

[00589] **Table 30. PRINT® Cylindrical Implants with Aflibercept Microparticles using DCM solvent**.

Sample ID	**PEG Ester**	**PEG Amine (w/w)**	**Polymer (PEG) wt%**	**Afliberceept particle wt%**	**Target Protein wt%**	**Mold Diameter (mm)**
AFB 1A	8ARM-SAZ-10K	8ARM-NH2-10K	50	50	17.5	1 mm
AFB 1B	8ARM-SAZ-10K	8ARM-NH2-10K/4ARM-NH2-10K (75/25)	37.5	62.5	21.9	1 mm
AFB 1C	8ARM-	8ARM-NH2-	25	75	26.3	1 mm

	SAZ-10K	10K/4ARM-NH2-10K (75/25)				
AFB 3A	8ARM-SAZ-10K	8ARM-NH2-10K	50	50	17.5	1 mm
AFB 3B	8ARM-SAZ-10K	8ARM-NH2-10K/4ARM-NH2-10K (75/25)	37.5	62.5	21.9	1 mm
AFB 5A	8ARM-SAZ-10K	8ARM-NH2-10K	25	75	47.5	1 mm
AFB 5B	8ARM-SAZ-10K	8ARM-NH2-10K/4ARM-NH2-10K (75/25)	25	75	47.5	1 mm
AFB 5C	8ARM-SAZ-10K	8ARM-NH2-10K/4ARM-NH2-10K (50/50)	25	75	47.5	1 mm
AFB 5D	8ARM-SAZ-10K	8ARM-NH2-10K	25	75	26.9	1 mm
AFB 5E	8ARM-SAZ-10K	8ARM-NH2-10K/4ARM-NH2-10K (75/25)	25	75	26.9	1 mm
AFB 5F	8ARM-SAZ-10K	8ARM-NH2-10K/4ARM-NH2-10K (50/50)	25	75	26.9	1 mm

[00590] Example 38. Additional PRINT® Cylindrical Implants with PRINT® Aflibercept Microparticles Formulations

[00591] Additionally, a series of implants were prepared using the polydisperse aflibercept particles of Example 3 and the cylindrical implant process of degradable hydrogel of Example 7 using EL as the solvent. The implants are shown below in **Tables 31.**

Table 31. PRINT® Cylindrical Implants with PRINT® Aflibercept Microparticles using EL solvent.

Sample ID	PEG Ester	PEG Amine (w/w)	PEG Ester wt%	Afliberce pt particle wt%	Target Protein wt%	Mold Diameter (mm)
AFB 8A	8ARM-SAZ-10K	8ARM-NH2-10K	50	50	28	1 mm
AFB 8B	8ARM-SAZ-10K	8ARM-NH2-10K/4ARM-NH2-10K (75/25)	35	65	36.4	1 mm
AFB 8C	8ARM-SAZ-10K	8ARM-NH2-10K/4ARM-NH2-10K (75/25)	20	80	44.8	1 mm
AFB 8D	8ARM-SAZ-10K	8ARM-NH2-10K/4ARM-NH2-10K (50/50)	20	80	44.8	1 mm

[00592] Example 39. Additional PRINT® Cylindrical Implants with PRINT® Bevacizumab Microparticles Formulations

[00593] A series of implants were prepared using the polydisperse bevacimab particles of Example 3 and the cylindrical implant process of degradable hydrogel of Example 9 using DCM as the solvent. The implants are shown below in **Tables 32**

[00594] Table 32. PRINT® Cylindrical Implants with PRINT® Bevacizumab Microparticles using DCM solvent.

Sample ID	PEG Ester	PEG Amine (w/w)	PEG wt%	Afliberce pt particle wt%	Target Protein wt%	Mold Diameter (mm)
AMD 80A	8ARM-SAZ-15K	8ARM-NH2-10K	50	50	13.5	1 mm

AMD 80B	8ARM-SAZ-15K	8ARM-NH2-10K	40	60	16.2	1 mm
AMD 80C	8ARM-SAZ-15K	8ARM-NH2-10K	30	70	18.9	1 mm
AMD 80D	8ARM-SAZ-15K	8ARM-NH2-10K	20	80	21.6	1 mm

[00595] Example 40. Improved Monomer Content of Released Aflibercept

[00596] A series of implants were prepared using the polydisperse aflibercept particles of Example 4 and the cylindrical implant process of Example 9. These implants are shown in **Table 33**.

[00597] Table 33. PRINT® Cylindrical Implants with Spray Dried Aflibercept

Sample ID	PEG Ester	PEG Amine	PEG Ester wt%	PEG Amine wt%	Protein particle wt%	Target Protein wt%	Mold Diameter (mm)
ENV-1P-0205-91-D	8ARM-SAZ-10K	8ARM-NH2-10K/ 4ARM-NH2-10K (50/50)	10	10	80	35.8	1 mm
RES-PRO-0041-44-A	8ARM-SAZ-10K	8ARM-NH2-10K/ 4ARM-NH2-10K (50/50)	10	10	80	35.8	1 mm
RES-PRO-0041-44-B	8ARM-SAZ-10K	8ARM-NH2-10K/ 4ARM-NH2-10K (50/50)	10	10	80	32.7	1 mm
RES-PRO-0041-44-C	8ARM-SAZ-10K	8ARM-NH2-10K/ 4ARM-NH2-10K (50/50)	10	10	80	24.7	1 mm
RES-PRO-0041-44-D	8ARM-SAZ-10K	8ARM-NH2-10K/ 4ARM-NH2-10K (50/50)	10	10	80	34.2	1 mm
RES-PRO-0041-44-E	8ARM-SAZ-10K	8ARM-NH2-10K/ 4ARM-NH2-10K (50/50)	10	10	80	26.3	1 mm

[00598] Implants were prepared with spray dried aflibercept made with additional excipients added to the protein solution prior to spray drying. These additional excipients are shown in **Table 34**.

[00599] **Table 34. Spray Dried Aflibercept Excipients**

Sample ID	Additional Excipients in Spray Dried Protein	Excipient wt.%
RES-PRO-0041-44-B	Trileucine	15
RES-PRO-0041-44-C	Trileucine	15
RES-PRO-0041-44-D	Leucine	25
RES-PRO-0041-44-E	Leucine	25

[00600] In vitro release rates were measured according to Example 14 and shown in **FIG.26 A and B**.

Monomer content of in vitro released aflibercept was measured by size exclusion chromatography (SEC). Results are shown in **FIG 27**. Protein formulated with additional excipients leucine and trileucine maintains high monomer content (>98%) through 23 days of in vitro release, whereas protein formulated without these additional excipients begins to show a decrease in monomer content. These additional excipients are thought to aid in protecting the protein from aggregation during protein micronization, implant fabrication, and in vitro release. Applicants do not wish to be bound to any particular theory, it is one theory that both leucine and trileucine protect proteins during spray dry processes by replacing proteins at the water/air interface and therefore reducing surface tensions that can induce protein aggregation during spray dry particle formation. It is possible that utilizing spray dried particles containing leucine or trileucine minimize the interfacial surface tension at the particle/water interface during the initial hydration of the extended release product and therefore reduce the formation of protein aggregates that occur upon initial contact with water.

INCORPORATION BY REFERENCE

All references, articles, publications, patents, patent publications, and patent applications cited herein are incorporated by reference in their entireties for all purposes. However, mention of any reference, article, publication, patent, patent publication, and patent application cited herein is not, and should not be taken as, an acknowledgment or any form of suggestion that they constitute valid prior art or form part of the common general knowledge in any country in the world.

CLAIMS

What is claimed is:

1. A method for making an ocular sustained-release pharmaceutical composition for the delivery of a therapeutic agent to an eye of a subject in need thereof, comprising:

formulating a solution comprising a therapeutic agent and one or more excipients;

forming, from the solution, particles comprising the therapeutic agent and one or more excipients;

combining the particles with gel precursors in a solvent to form a suspension comprising the particles and the gel precursors, wherein the solvent is an organic and hydrophilic solvent; and

allowing the gel precursors to gel around the particles, thereby forming a gel comprising encapsulated particles; and

thereby forming the pharmaceutical composition for sustained delivery of a therapeutic agent.

2. The method of claim 1, wherein the solution comprises a therapeutic agent content of from about 20 to about 60 g/L.

3. The method of claim 1, wherein the forming comprises adding the solution to a mold to thereby form the particles comprising the therapeutic agent and one or more excipients.

4. The method of claim 3, wherein the mold has a largest dimension of less than or equal to about 10 μm.

5. The method of claim 3, wherein the mold has dimensions of about 1 μm x about 1 μm x about 1 μm.

6. The method of claim 3, wherein particles have dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension.

7. The method of claim 1, wherein the forming comprises lyophilizing, spray drying, or jet milling the solution to thereby form the particles.

8. The method of claim 1, wherein the particles are formed as micronized particles.

9. The method of claim 1, wherein the particles have a largest dimension in the range of about 1 μm to about 25 μm.

10. The method of claim 1, wherein the particles have a largest dimension of about 1 μm.

11. The method of claim 1, wherein the particles comprise about 20 wt % to about 80 wt % of the pharmaceutical composition.

12. The method of claim 1, wherein the therapeutic agent comprises about 40 wt % to about 60 wt % of the particles.

13. The method of claim 1, wherein the therapeutic agent comprises about 10 wt % to about 60 wt % of the pharmaceutical composition.

14. The method of claim 1, wherein the gel comprises about 20 wt % to about 70 wt % of the pharmaceutical composition.

15. The method of claim 1, wherein the therapeutic agent comprises about 10 wt % to about 60 wt % of the pharmaceutical composition, and the gel comprises about 20 wt % to about 70 wt % of the pharmaceutical composition.

16. The method of claim 1, wherein the gel is a biodegradable gel.

17. The method of claim 1, wherein the gel is formed by covalently crosslinking the gel precursors.

18. The method of claim 1, wherein the gel comprises crosslinked polyethylene glycol polymers.

19. The method of claim 1, wherein the gel precursors comprise a first precursor having a first functional group and a second precursor having a second functional group, wherein the first functional group and the second functional group are reactive in the solvent.

20. The method of claim 1, wherein the gel precursors comprise a first precursor having a first functional group and a second precursor having a second functional group, wherein the first functional group interacts with the second functional group thereby forming a covalent bond.

21. The method of claim 1, wherein the precursors comprise a first precursor having a first functional group and a second precursor having a second functional group, wherein the first precursor and the second precursor are present in the gel in ratio in a range of from about 1 to about 10 to about 10 to about 1.

22. The method of claim 1, wherein the precursors comprise a first precursor having a first functional group and a second precursor having a second functional group, wherein the first precursor and the second precursor are present in the gel at a ratio of about 1:1.

23. The method of any of claims 19-22, wherein the second precursor includes a third precursor, the third precursor having a third functional group which is reactive with the first functional group.

24. The method of claim 23, wherein the third functional group and the second functional groups can be the same or different.

25. The method of claim 1, wherein the gel precursors comprise PEG polymers.

26. The method of claim 1, wherein the gel precursors comprise PEG polymers having a molecular weight in the range of about 10,000 Da to about 15,000 Da.

27. The method of claim 1, wherein the gel precursors comprise at least one PEG polymer having an N-hydroxy succinimidyl ester functional group (PEG-NHS) and at least one PEG polymer having an amine functional group (PEG-amine).

28. The method of claim 1, wherein the gel precursors comprise at least one PEG-NHS polymer having a molecular weight range of about 10,000 Da to about 15,000 Da, and at least one PEG-amine polymer having a molecular weight range of about 10,000 Da to about 15,000 Da.

29. The method of claim 1, wherein the gel precursors comprise at least one PEG-NHS polymer having a molecular weight of about 10,000 Da and at least one PEG-amine polymer having a molecular weight of about 10,000 Da.

30. The method of claim 1, wherein the gel is formed in a mold.

31. The method of claim 1, wherein the gel is formed in a mold and wherein the solvent is a ethyl lactate.

32. The method of claim 1, wherein the gel is formed in a mold having dimensions in the range of about 10 μm x 10 μm x 10 μm to about 100 μm x 100 μm x 100 μm.

33. The method of claim 1, wherein the gel is formed in a mold having dimensions of about 12.5 μm x 12.5 μm x 25 μm.

34. The method of claim 1, wherein the gel is formed in a mold having dimensions of about 25 μm x 25 μm x 25 μm.

35. The method of claim 1, wherein the gel is formed in a mold having dimensions of about 50 μm x 50 μm x 50 μm.

36. The method of claim 1, wherein the gel is formed in a mold having dimensions in the range of about 100 μm x 100 μm x 200 μm to about 700 μm x 700 μm x 7,000 μm, and wherein the solvent is a ethyl lactate.

37. The method of claim 1, wherein the gel is formed in a mold having dimensions of about 225 μm x 225 μm x 2,925 μm.

38. The method of claim 1, wherein the gel is formed in a mold having dimensions of about 311 μm x 395 μm x 6,045 μm.

39. The method of claim 1, wherein the gel is formed in a mold having dimensions of about 600 μm x 600 μm x 1,000 μm.

40. The method of claim 1, wherein the gel is formed in a cylindrical mold.

41. The method of claim 1, wherein the gel is formed in a cylindrical mold having a dimeter of about 1 mm.

42. The method of claim 1, wherein the gel is formed in a cylindrical mold having a dimeter of about 1 mm, and wherein the solvent is acetonitrile.

43. The method of claim 1, further comprising removing the solvent from the gel thereby forming a xerogel.

44. The method of claim 46, wherein the removing comprises lyophilizing the gel, thereby forming the xerogel.

45. The method of claim 1, further comprising hydrating the gel in an aqueous solution thereby forming a hydrogel.

46. The method of claim 48, wherein the hydrating occurs in-vivo after administration to the eye of a subject in need thereof.

47. The method of claim 1, wherein the gel is suspended in a pharmaceutically acceptable liquid to form a liquid vehicle for ocular delivery.

48. The method of claim 47, wherein the volume of the liquid is from about 10 μL to about 1,000 μL.

49. The method of claim 47, wherein the volume of the liquid is from about 10 μL to about 500 μL.

50. The method of claim 47, wherein the volume of the liquid is from about 10 μL to about 100 μL.

51. The method of claim 45, wherein the xerogel has dimensions of 12.5 μm × 12.5 μm × 25 μm ± 20 % in any dimension, of about 25 μm × 25 μm × 25 μm ± 20 % in any dimension, or 25 μm × 25 μm × 50 μm ± 20 % in any dimension, and the xerogel is suspended in a pharmaceutically acceptable liquid to form a liquid vehicle for ocular delivery.

52. The method of claim 51, wherein the volume of the liquid is from about 10 μL to about 1,000 μL.

53. The method of claim 51, wherein the volume of the liquid is from about 10 μL to about 500 μL.

54. The method of claim 51, wherein the volume of the liquid is from about 10 μL to about 100 μL.

55. The method of claim 1, wherein the therapeutic agent is a water soluble biologic.

56. The method of claim 1, wherein the therapeutic agent is a protein, peptide, antibody.

57. The method of claim 1, wherein the therapeutic agent is a protein.

58. The method of claim 1, wherein the therapeutic agent is an anti-VEGF agent.

59. The method of claim 1, wherein the therapeutic agent is bevacizumab, ranibizumab, aflibercept, or pegaptanib.

60. The method of claim 1, wherein the therapeutic agent is bevacizumab.

61. The method of claim 1, wherein the therapeutic agent is aflibercept.

62. The method of claim 1, wherein the therapeutic agent is an antibody.

63. The method of claim 1, wherein the therapeutic agent is an anti-VEFG antibody.

64. The method of claim 1, wherein therapeutic agent is bevacizumab, the bevacizumab is formed as a micronized particle having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, the solvent is ethyl lactate, and the gel comprises cross-linked PEG polymers.

65. The method of claim 1, wherein therapeutic agent is bevacizumab, the bevacizumab is formed as a micronized particle having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, the solvent is ethyl lactate, the gel comprises cross-linked PEG polymers, and the pharmaceutical composition is a mesoparticle.

66. The method of claim 1, wherein therapeutic agent is bevacizumab, the bevacizumab is formed as a micronized particle having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, the solvent is ethyl lactate, the gel comprises cross-linked PEG polymers, and the pharmaceutical composition is an implant.

67. The method of claim 1, wherein therapeutic agent is aflibercept, the aflibercept is formed as a micronized particle having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, the solvent is ethyl lactate, and the gel comprises cross-linked PEG polymers.

68. The method of claim 1, wherein therapeutic agent is aflibercept, the aflibercept is formed as a micronized particle having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, the solvent is ethyl lactate, the gel comprises cross-linked PEG polymers, and the pharmaceutical composition is a mesoparticle.

69. The method of claim 1, wherein therapeutic agent is aflibercept, the aflibercept is formed as a micronized particle having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, the solvent is ethyl lactate, the gel comprises cross-linked PEG polymers, and the pharmaceutical composition is an implant.

70. The method of claim 1, further comprising delivering the pharmaceutical composition to the eye of the subject to prevent or treat a condition of the eye.

71. The method of claim 1, wherein the pharmaceutical composition is formulated to deliver the therapeutic agent to the eye of the subject in need thereof for about 4 to about 6 months.

72. The method of claim 1, wherein the therapeutic agent does not substantially aggregate.

73. The method of claim 1, wherein the excipient is leucine, trileucine, valine, or combinations thereof.

74. A method for making an ocular sustained-release pharmaceutical composition for the delivery of a therapeutic agent to an eye of a subject in need thereof, comprising:

formulating a solution comprising a therapeutic agent and one or more excipients;

fabricating solid state microparticles (PuPs) comprising the therapeutic agent;

combining the particles with gel precursors in a solution to form a suspension comprising the particles and the gel precursors, wherein the solvent is an organic and hydrophilic solvent; and

allowing the gel precursors to gel around the particles, thereby forming a gel comprising encapsulated particles;

thereby forming the pharmaceutical composition for sustained delivery of a therapeutic agent.

75. The method of claim 74, wherein the PuPs are fabricated in a mold.

76. The method of claim 74, wherein the therapeutic agent comprises about 40 to 60 wt % of the PuP.

77. The method of claim 74, wherein the PuP comprises about 60 to 80 wt % of the pharmaceutical composition.

78. The method of claim 74, wherein the mold has a largest dimensions of less than or equal to about 10 μm.

79. The method of claim 74, wherein the mold has dimensions of about 1 μm x about 1 μm x 1 about μm.

80. The method of claim 74, wherein the PuPs have dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension.

81. The method of claim 74, wherein the PuPs are fabricated in a mold, said PuPs having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension.

82. The method of claim 74, wherein the gel is a biodegradable gel.

83. The method of claim 74, wherein the gel is formed by covalently crosslinking the gel precursors.

84. The method of claim 74, wherein the gel precursors comprise a first precursor having a first functional group and a second precursor having a second functional group, wherein the first functional group and the second functional group are reactive in the solvent.

85. The method of claim 74, wherein the gel precursors comprise a first precursor having a first functional group and a second precursor having a second functional group, wherein the first functional group interacts with the second functional group thereby forming a covalent bond.

86. The method of claim 74, wherein the precursors comprise a first precursor having a first functional group and a second precursor having a second functional group, wherein the first precursor and the second precursor are present in the gel in ratio in a range of from about 1 to about 10 to about 10 to about 1.

87. The method of claim 74, wherein the precursors comprise a first precursor having a first functional group and a second precursor having a second functional group, wherein the first precursor and the second precursor are present in the gel at a ratio of about 1:1.

88. The method of any of claims 83-87 wherein the second precursor includes a third precursor, the third precursor having a third functional group which is reactive with the first functional group.

89. The method of claim 88, wherein the third functional group and the second functional groups can be the same or different.

90. The method of claim 74, wherein the gel comprises crosslinked polyethylene glycol polymers.

91. The method of claim 74, wherein the gel precursors comprise PEG polymers having a molecular weight in the range of about 10,000 Da to about 15,000 Da.

92. The method of claim 74, wherein the gel precursors comprise at least one PEG polymer having an N-hydroxy succinimidyl ester functional group (PEG-NHS) and at least one PEG polymer having an amine functional group (PEG-amine).

93. The method of claim 74, wherein the gel precursors comprise at least one PEG-NHS polymer having a molecular weight range of about 10,000 Da to about 15,000 Da, and at least one PEG-amine polymer having a molecular weight range of about 10,000 Da to about 15,000 Da.

94. The method of claim 74, wherein the gel precursors comprise at least one PEG-NHS polymer having a molecular weight of about 10,000 Da and at least one PEG-amine polymer having a molecular weight of about 10,000 Da.

95. The method of claim 74, wherein the gel is formed in a mold.

96. The method of claim 74, wherein the gel is formed in a mold, and wherein the solvent is a ethyl lactate.

97. The method of claim 74, wherein the gel is formed in a mold having dimensions in the range of about 10 μm x 10 μm x 10 μm to about 100 μm x 100 μm x 100 μm.

98. The method of claim 74, wherein the gel is formed in a PRINT mold having dimensions in the range of about 100 μm x 100 μm x 200 μm to about 700 μm x 700 μm x 7,000 μm.

99. The method of claim 74, wherein the gel is formed in a mold having dimensions of about 225 μm x 225 μm x 2,925 μm.

100. The method of claim 74, wherein the gel is formed in a mold having dimensions of about 311 μm x 395 μm x 6,045 μm.

101. The method of claim 74, wherein the gel is formed in a mold having dimensions of about 600 μm x 600 μm x 1,000 μm.

102. The method of claim 74, wherein the gel is formed in a mold having dimensions of about 12.5 μm × 12.5 μm × 25 μm.

103. The method of claim 74, wherein the gel is formed in a mold having dimensions of about 25 μm × 25 μm × 25 μm.

104. The method of claim 74, wherein the gel is formed in a mold having dimensions of about 25 μm × 25 μm × 50 μm.

105. The method of claim 74, wherein the gel is formed in a cylindrical mold.

106. The method of claim 74, wherein the gel is formed in a cylindrical mold having a dimeter of about 1 mm.

107. The method of claim 74, wherein the gel is formed in a cylindrical mold having a dimeter of about 1 mm, and wherein the solvent is acetonitrile.

108. The method of claim 74, further comprising removing the solvent from the gel thereby forming a xerogel.

109. The method of claim 108, wherein the removing comprises lyophilizing the gel, thereby forming the xerogel.

110. The method of claim 74, further comprising hydrating the gel in an aqueous solution thereby forming a hydrogel.

111. The method of claim 110, wherein the hydrating occurs in-vivo after administration to the eye of a subject in need thereof.

112. The method of claim 74, wherein the gel is suspended in a pharmaceutically acceptable liquid to form a liquid vehicle for ocular delivery.

113. The method of claim 112, wherein the volume of the liquid is from about 10 μL to about 1,000 μL.

114. The method of claim 112, wherein the volume of the liquid is from about 10 μL to about 500 μL.

115. The method of claim 112, wherein the volume of the liquid is from about 10 μL to about 100 μL.

116. The method of claim 108, wherein the xerogel has dimensions of 12.5 μm × 12.5 μm × 25 μm ± 20 % in any dimension, of about 25 μm × 25 μm × 25 μm ± 20 % in any dimension, or 25 μm × 25 μm × 50 μm ± 20 % in any dimension, and the xerogel is suspended in a pharmaceutically acceptable liquid to form a liquid vehicle for ocular delivery.

117. The method of claim 116, wherein the volume of the liquid is from about 10 μL to about 1,000 μL.

118. The method of claim 116, wherein the volume of the liquid is from about 10 μL to about 500 μL.

119. The method of claim 116, wherein the volume of the liquid is from about 10 μL to about 100 μL.

120. The method of claim 74, wherein the therapeutic agent is a protein, peptide, antibody.

121. The method of claim 74, wherein the therapeutic agent is bevacizumab, ranibizumab, aflibercept, pegaptanib.

122. The method of claim 74, wherein the therapeutic agent is bevacizumab.

123. The method of claim 74, wherein the therapeutic agent is aflibercept.

124. The method of claim 74, wherein the therapeutic agent is an antibody.

125. The method of claim 74, wherein the therapeutic agent is an anti-VEFG antibody.

126. The method of claim 74, wherein therapeutic agent is bevacizumab, the bevacizumab is formed as a PuP having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, and the gel comprises cross-linked PEG polymers.

127. The method of claim 74, wherein therapeutic agent is bevacizumab, the bevacizumab is formed as a PuP having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, the gel comprises cross-linked PEG polymers, and the pharmaceutical composition is a mesoparticle.

128. The method of claim 74, wherein therapeutic agent is bevacizumab, the bevacizumab is formed as a PuP having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, the gel comprises cross-linked PEG polymers, and the pharmaceutical composition is an implant.

129. The method of claim 74, wherein therapeutic agent is aflibercept, the aflibercept is formed as a PuP having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, the solvent is ethyl lactate, and the gel comprises cross-linked PEG polymers.

130. The method of claim 74, wherein therapeutic agent is aflibercept, the aflibercept is formed as a PuP having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, the solvent is ethyl lactate, the gel comprises cross-linked PEG polymers, and the pharmaceutical composition is a mesoparticle.

131. The method of claim 74, wherein therapeutic agent is aflibercept, the aflibercept is formed as a PuP having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, the solvent is ethyl lactate, the gel comprises cross-linked PEG polymers, and the pharmaceutical composition is an implant.

132. The method of claim 74, further comprising delivering the pharmaceutical composition to a patient's eye in need thereof to prevent or treat a condition of the eye.

133. The method of claim 74, wherein the pharmaceutical composition prevents or treats the condition of the eye for about 4 months to about 6 months.

134. The method of claim 74, wherein the therapeutic agent does not substantially aggregate.

135. The method of claim 74, wherein the excipient is leucine, trileucine, valine, or combinations thereof.

136. A sustained release pharmaceutical composition comprising:

(A) a gel comprising a biocompatible polymer matrix; and

(B) and at least one protein microparticle (PuP) comprising a therapeutic agent and at least one pharmaceutically acceptable excipient;

wherein the gel encapsulates the PuP;

wherein the PuP has a largest dimension of less than about 10 μm;

wherein the pharmaceutical composition is formulated to deliver the therapeutic agent for at least about 4 months.

137. The pharmaceutical composition of claim 136, wherein PuP has dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension.

138. The pharmaceutical composition of claim 136, wherein the PuP comprise about 60 wt % to about 80 wt % of the pharmaceutical composition.

139. The pharmaceutical composition of claim 136, wherein the therapeutic agent comprises about 40 wt % to about 60 wt % of the PuP.

140. The pharmaceutical composition of claim 136, wherein the therapeutic agent comprises about 10 wt % to about 60 wt % of the pharmaceutical composition.

141. The pharmaceutical composition of claim 136, wherein the gel comprises about 20 wt % to about 70 wt % of the pharmaceutical composition.

142. The pharmaceutical composition of claim 136, wherein the therapeutic agent comprises about 10 wt % to about 60 wt % of the pharmaceutical composition, and the gel comprises about 20 wt % to about 70 wt % of the pharmaceutical composition

143. The pharmaceutical composition of claim 136, wherein the gel is a biodegradable gel.

144. The pharmaceutical composition of claim 136, wherein the gel is formed by covalently crosslinking the gel precursors.

145. The pharmaceutical composition of claim 136, wherein the gel comprises crosslinked polyethylene glycol polymers.

146. The pharmaceutical composition of claim 136, wherein the gel comprises at least one PEG polymer having an N-hydroxy succinimidyl ester functional group (PEG-NHS) and at least one PEG polymer having an amine functional group (PEG-amine).

147. The pharmaceutical composition of claim 136, wherein the gel comprised at least one PEG-NHS polymer having a molecular weight of about 10,000-15,000 Da and at least one PEG-amine polymer having a molecular weight of about 10,000-15,000 Da.

148. The pharmaceutical composition of claim 136, wherein the gel comprises at least one 8-arm PEG-NHS having a molecular weight of 10,000-15,000 Da, at least one 4-arm PEG-NHS having a molecular weight of about 10,000-15,000 Da, at least one 8 arm PEG-amine having a molecular weight of about 10,000-15,000 Da, or at least one 4 arm PEG-amine having a molecular weight of about 10,000-15,000 Da

149. The pharmaceutical composition of claim 136, wherein the gel comprises 20-70 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising:

(i) 10-35 ± 5 wt % of at least one PEG-NHS having a molecular weight of about 10,000-15,000 Da; and

(ii) 10-35 ± 5 wt % of at least PEG-amine having a molecular weight of about 10,000-15-000 Da.

150. The pharmaceutical composition of claim 136, wherein the gel comprises 20-70 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising:

(i) 10-35 ± 5 wt % of at least one PEG-NHS having a molecular weight of about 10,000-15,000 Da; and

(ii) 10-35 ± 5 wt % of at least one PEG-amine having a molecular weight of about 10,000-15,000 Da;

and

wherein the therapeutic agent comprises about 10-60 ± 5 wt % of the pharmaceutical composition.

151. The pharmaceutical composition of claim 136, wherein the gel comprises 20 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising:

(i) 10 ± 3 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; and

(ii) 10 ± 3 wt % of an 8 arm PEG-amine having a molecular weight of 10,000 Da;

and

wherein the therapeutic agent comprises about 35 ± 5 wt % of the pharmaceutical composition.

152. The method of claim 136, wherein the gel comprises 20 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising:

(i) 10 ± 3 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da;

(ii) 5 ± 3 wt % of an 8-arm PEG-amine having a molecular weight of 10,000 Da; and

(iii) 5 ± 3 wt % of a 4-arm PEG-amine having a molecular weight of 10,000 Da

and

wherein the therapeutic agent comprises about 36 ± 5 wt % of the pharmaceutical composition

153. The pharmaceutical composition of claim 136, wherein the gel comprises 21 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising:

(i) 10 ± 5 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; and

(ii) 10 ± 5 wt % of an 8 arm PEG-amine having a molecular weight of 10,000 Da;

and

wherein the therapeutic agent comprises about 46 ± 5 wt % of the pharmaceutical composition.

154. The pharmaceutical composition of claim 136, wherein the the gel comprises 50 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising:

(i) 25 ± 5 wt % of an 8-arm PEG-NHS having a molecular weight of 15,000 Da;

(ii) 25 ± 5 wt % of an 8-arm PEG-amine having a molecular weight of 10,000 Da; and

wherein the therapeutic agent comprises about 22 ± 5 wt % of the pharmaceutical composition.

155. The pharmaceutical composition of claim 136, wherein the gel comprises 50 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising:

(i) 25 ± 5 wt % of an 4-arm PEG-NHS having a molecular weight of 10,000 Da;

(ii) 25 ± 5 wt % of an 4-arm PEG-amine having a molecular weight of 10,000 Da; and

wherein the therapeutic agent comprises about 18 ± 5 wt % of the pharmaceutical composition.

156. The pharmaceutical composition of claim 136, wherein pharmaceutical composition is a mixture of a first pharmaceutical composition and a second pharmaceutical composition, wherein:

(i) the first pharmaceutical composition comprises a first gel comprising 21 ± 5 % wt of the first pharmaceutical composition and contains a mixture of polymers comprising:

(a) 10 ± 5 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; and

(b) 10 ± 5 wt % of an 4 arm PEG-amine having a molecular weight of 10,000 Da; and

wherein a first therapeutic agent comprises about 36 ± 5 wt % of the first pharmaceutical composition;

and

(ii) the second composition comprises a second gel comprising 21 ± 5 % wt of the first pharmaceutical composition and contains a mixture of polymers comprising:

(a) 10 ± 5 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; and

(b) 10 ± 5 wt % of an 8 arm PEG-amine having a molecular weight of 10,000 Da; and

wherein a second therapeutic agent comprises about 36 ± 5 wt % of the second pharmaceutical composition;

157. The pharmaceutical composition of claim 136, wherein the gel comprises 20-70 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising:

(i) 10-35 ± 5 wt % of at least one PEG-NHS having a molecular weight of about 10,000-15,000 Da; and

(ii) 10-35 ± 5 wt % of at least one PEG-amine having a molecular weight of about 10,000-15,000 Da; and

wherein the PuP has dimensions of 1 µm in diameter x 1 µm in height ± 10% in any dimension, and the therapeutic agent comprises about 10-60 ± 5 wt % of the pharmaceutical composition.

158. The pharmaceutical composition of claim 136, wherein the gel comprises 20 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising:

(i) 10 ± 3 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; and

(ii) 10 ± 3 wt % of an 8 arm PEG-amine having a molecular weight of 10,000 Da; and

wherein the PuP has dimensions of 1 µm in diameter x 1 µm in height ± 10% in any dimension, and the therapeutic agent comprises about 35 ± 5 wt % of the pharmaceutical composition.

159. The pharmaceutical composition of claim 136, wherein the gel comprises 20 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising:

(i) 10 ± 3 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da;

(ii) 5 ± 3 wt % of an 8-arm PEG-amine having a molecular weight of 10,000 Da; and

(iii) 5 ± 3 wt % of a 4-arm PEG-amine having a molecular weight of 10,000 Da; and wherein the PuP has dimensions of 1 µm in diameter x 1 µm in height ± 10% in any dimension, and the therapeutic agent comprises about 36 ± 5 wt % of the pharmaceutical composition.

160. The pharmaceutical composition of claim 136, wherein the gel comprises 21 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising:

(i) 10 ± 5 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; and

(ii) 10 ± 5 wt % of an 8 arm PEG-amine having a molecular weight of 10,000 Da; and

wherein the PuP has dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, and the therapeutic agent comprises about 46 ± 5 wt % of the pharmaceutical composition.

161. The pharmaceutical composition of claim 136, wherein the the gel comprises 50 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising:

(i) 25 ± 5 wt % of an 8-arm PEG-NHS having a molecular weight of 15,000 Da;

(ii) 25 ± 5 wt % of an 8-arm PEG-amine having a molecular weight of 10,000 Da; and

wherein the PuP has dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension,

wherein the therapeutic agent comprises about 22 ± 5 wt % of the pharmaceutical composition.

162. The pharmaceutical composition of claim 136, wherein the gel comprises 50 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising:

(i) 25 ± 5 wt % of an 4-arm PEG-NHS having a molecular weight of 10,000 Da;

(ii) 25 ± 5 wt % of an 4-arm PEG-amine having a molecular weight of 10,000 Da; and

wherein the PuP has dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, and wherein the therapeutic agent comprises about 18 ± 5 wt % of the pharmaceutical composition.

163. The pharmaceutical composition of claim 136, wherein pharmaceutical composition is a mixture of a first pharmaceutical composition and a second pharmaceutical composition, wherein:

(i) the first pharmaceutical composition comprises a first gel comprising 21 ± 5 % wt of the first pharmaceutical composition and contains a mixture of polymers comprising:

(a) 10 ± 5 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; and

(b) 10 ± 5 wt % of an 4 arm PEG-amine having a molecular weight of 10,000 Da; and

wherein a first PuP has dimensions of 1 μm in diameter x 1 μm in heigh ± 10% in any dimension, and a first therapeutic agent comprises about 36 ± 5 wt % of the first pharmaceutical composition;

and

(ii) the second composition comprises a second gel comprising 21 ± 5 % wt of the first pharmaceutical composition and contains a mixture of polymers comprising:

(a) 10 ± 5 wt % of an 8-arm PEG-NHS having a molecular weight of 10,000 Da; and

(b) 10 ± 5 wt % of an 8 arm PEG-amine having a molecular weight of 10,000 Da; and

wherein a second PuP has dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension, and a second therapeutic agent comprises about 36 ± 5 wt % of the second pharmaceutical composition.

164. The pharmaceutical composition of claim 136, wherein pharmaceutical composition is a rod-shaped implant having dimensions of 225 μm x 225 μm x 2925 μm ± 10% of any dimension.

165. The pharmaceutical composition of claim 136, wherein pharmaceutical composition is a rod-shaped implant having dimensions of 311 μm x 395 μm x 6045 μm ± 10% of any dimension.

166. The pharmaceutical composition of claim 136, wherein pharmaceutical composition is a rod-shaped implant having dimensions of or 600 μm x 600 μm x 1000 μm ± 10% of any dimension.

167. The pharmaceutical composition of claim 136, wherein pharmaceutical composition is a cylindrical implant having a diameter of 1 mm ± 10% and length of 3 mm ± 10%.

168. The pharmaceutical composition of claim 136, wherein pharmaceutical composition is a mesoparticle having dimensions of about 12.5 μm x 12.5 μm x 25 μm ± 10% in any dimension

169. The pharmaceutical composition of claim 168, wherein the pharmaceutical composition is suspended in about 1-500 μL of a pharmaceutically acceptable liquid.

170. The pharmaceutical composition of claim 136, wherein pharmaceutical composition is a mesoparticle having dimensions of about 25 μm x 25 μm x 25 μm ± 10% in any dimension.

171. The pharmaceutical composition of claim 170, wherein the pharmaceutical composition is suspended in about 1-500 μL of a pharmaceutically acceptable liquid.

172. The pharmaceutical composition of claim 136, wherein pharmaceutical composition is a mesoparticle having dimensions of about 50 μm x 50 μm x 50 μm ± 10% in any dimension.

173. The pharmaceutical composition of claim 172, wherein the pharmaceutical composition is suspended in about 1-500 μL of a pharmaceutically acceptable liquid.

174. The pharmaceutical composition of claim 136, wherein pharmaceutical composition is a cylindrical implant.

175. The pharmaceutical composition of claim 136, wherein pharmaceutical composition is a cylindrical implant having a diameter of about 1 mm ± 10 % and a length of about 3 mm ± 10%.

176. The pharmaceutical composition of claim 136, wherein the therapeutic agent is a protein, peptide, antibody.

177. The pharmaceutical composition of claim 136, wherein the therapeutic agent is bevacizumab, ranibizumab, aflibercept, pegaptanib.

178. The pharmaceutical composition of claim 136, wherein the therapeutic agent is bevacizumab.

179. The pharmaceutical composition of claim 136, wherein the therapeutic agent is aflibercept.

180. The pharmaceutical composition of claim 136, wherein the therapeutic agent is an antibody.

181. The pharmaceutical composition of claim 136, wherein the therapeutic agent is an anti-VEFG antibody.

182. The pharmaceutical composition of claim 136,, wherein the therapeutic agent does not substantially aggregate.

183. The pharmaceutical composition of claim 136, wherein the excipient is leucine, trileucine, valine, or combinations thereof.

184. A sustained release delivery system prepared by a process comprising the steps of:

formulating a solution comprising a therapeutic agent and one or more excipients;

forming, from the solution, particles comprising the therapeutic agent and one or more excipients;

combining the particles with gel precursors in a solvent to form a suspension comprising the particles and the gel precursors, wherein the solvent is an organic and hydrophilic solvent; and

allowing the gel precursors to gel around the particles, thereby forming a gel comprising encapsulated particles; and

thereby forming the pharmaceutical composition for sustained delivery of a therapeutic agent,

wherein the pharmaceutical composition comprises:

(A) a gel comprising a biocompatible polymer matrix; and

(B) and at least one protein microparticle (PuP) comprising a therapeutic agent and at least one pharmaceutically acceptable excipient;

wherein the PuP has a largest dimension of less than 10 μm;

wherein the pharmaceutical composition is formulated to deliver the therapeutic agent for at least about 4 months.

185. The sustained release delivery system of claim 184, wherein PuP has dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension.

186. The sustained release delivery system of claim 184, wherein the PuP comprise about 60 wt % to about 80 wt % of the pharmaceutical composition.

187. The sustained release delivery system of claim 184, wherein the therapeutic agent comprises about 40 wt % to about 60 wt % of the PuP.

188. The sustained release delivery system of claim 184, wherein the therapeutic agent comprises about 10 wt % to about 60 wt % of the pharmaceutical composition.

189. The sustained release delivery system of claim 184, wherein the gel comprises about 20 wt % to about 70 wt % of the pharmaceutical composition.

190. The sustained release delivery system of claim 184, wherein the therapeutic agent comprises about 10 wt % to about 60 wt % of the pharmaceutical composition, and the gel comprises about 20 wt % to about 70 wt % of the pharmaceutical composition

191. The sustained release delivery system of claim 184, wherein the gel is a biodegradable gel.

192. The sustained release delivery system of claim 184, wherein the gel is formed by covalently crosslinking the gel precursors.

193. The sustained release delivery system of claim 184, wherein the gel comprises crosslinked polyethylene glycol polymers.

194. The sustained release delivery system of claim 184, wherein the gel comprises at least one PEG polymer having an N-hydroxy succinimidyl ester functional group (PEG-NHS) and at least one PEG polymer having an amine functional group (PEG-amine).

195. The sustained release delivery system of claim 184, wherein the gel is formed in a mold.

196. The sustained release delivery system of claim 184, wherein the gel is formed in a mold and wherein the solvent is a ethyl lactate.

197. The sustained release delivery system of claim 184, wherein the gel is formed in a mold having dimensions in the range of about 10 μm x 10 μm x 10 μm to about 100 μm x 100 μm x 100 μm.

198. The sustained release delivery system of claim 184, wherein the gel is formed in a mold having dimensions of about 12.5 μm x 12.5 μm x 25 μm.

199. The sustained release delivery system of claim 184, wherein the gel is formed in a mold having dimensions of about 25 μm x 25 μm x 25 μm.

200. The sustained release delivery system of claim 184, wherein the gel is formed in a mold having dimensions of about 50 μm x 50 μm x 50 μm.

201. The sustained release delivery system of claim 184, wherein the gel is formed in a mold having dimensions in the range of about 100 μm x 100 μm x 200 μm to about 700 μm x 700 μm x 7,000 μm, and wherein the solvent is a ethyl lactate.

202. The sustained release delivery system of claim 184, wherein the gel is formed in a mold having dimensions of about 225 μm x 225 μm x 2,925 μm.

203. The sustained release delivery system of claim 184, wherein the gel is formed in a mold having dimensions of about 311 μm x 395 μm x 6,045 μm.

204. The sustained release delivery system of claim 184, wherein the gel is formed in a mold having dimensions of about 600 μm x 600 μm x 1,000 μm.

205. The sustained release delivery system of claim 184, wherein the gel is formed in a cylindrical mold.

206. The sustained release delivery system of claim 184, wherein the gel is formed in a cylindrical mold having a dimeter of about 1 mm.

207. The sustained release delivery system of claim 184, wherein the gel is formed in a cylindrical mold having a dimeter of about 1 mm, and wherein the solvent is acetonitrile.

208. The sustained release delivery system of claim 184, wherein the therapeutic agent is a protein, peptide, antibody.

209. The sustained release delivery system of claim 184, wherein the therapeutic agent is a bevacizumab, ranibizumab, aflibercept, pegaptanib.

210. The sustained release delivery system of claim 184, wherein the therapeutic agent is bevacizumab.

211. The sustained release delivery system of claim 184, wherein the therapeutic agent is aflibercept.

212. The sustained release delivery system of claim 184, wherein the therapeutic agent is an antibody.

213. The sustained release delivery system of claim 184, wherein the therapeutic agent is an anti-VEFG antibody.

214. A method of treating an ocular condition associated with abnormal vessel growth in the eye of a patient, comprising: administering to the vitreous of an eye of a patient in need thereof an ocular sustained-release pharmaceutical composition comprising:

(A) a gel comprising a biocompatible polymer matrix; and

(B) at least one therapeutic agent fabricated as a microparticle having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension; and

wherein the gel encapsulates the therapeutic agent, and

wherein the ocular sustained-release pharmaceutical composition is formulated to release the therapeutic agent in the vitreous of the eye for at least about 4 months.

215. The method of claim 214, wherein the therapeutic agent is bevacizumab, ranibizumab, aflibercept, or pegaptanib.

216. The method of claim 214, wherein the therapeutic agent is aflibercept.

217. The method of claim 214, wherein the therapeutic agent is aflibercept and aflibercept is administered in an amount of less than or equal to about 2 mg.

218. The method of claim 214, wherein the therapeutic agent is aflibercept and aflibercept is administered in an amount of less than or equal to about 850 μg.

219. The method of claim 214, wherein the therapeutic agent is released into the vitreous of the eye for about 4 months to about 6 months.

220. The method of claim 214, wherein the ocular condition is Retinal Vein Occlusion (RVO).

221. The method of claim 214, wherein the ocular condition is Age-Related Macular Degeneration (AMD).

222. The method of claim 214, wherein the a ocular condition is Diabetic Macular Edema (DME).

223. The method of claim 214, wherein the pharmaceutical composition is a mesoparticle.

224. The method of claim 214, wherein the pharmaceutical composition is an implant.

225. A method of maintaining an ocular vitreous concentration of an anti-Vascular Endothelial Growth Factor (anti-VEGF) agent of greater than or equal to about 1 ng/mL in an eye of a human, comprising: administering an ocular sustained-release pharmaceutical composition to the vitreous of an eye of a human in need thereof, wherein said ocular sustained-release pharmaceutical composition comprises:

(A) a gel comprising a biocompatible polymer matrix; and

(B) at least one a therapeutic agent fabricated as a microparticle having dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension; and wherein the vitreous concentration of the anti-VEGF agent in said human's eye is maintained at a concentration greater than or equal to about 1 ng/mL for at least about 3 months.

226. The method of claim 225, wherein the anti-VEGF agent is bevacizumab, ranibizumab, aflibercept, or pegaptanib.

227. The method of claim 225, wherein the anti-VEGF agent is aflibercept.

228. The method of claim 225, wherein the anti-VEGF agent is aflibercept and wherein the ocular sustained-release pharmaceutical composition comprises less than or equal to about 2 mg of aflibercept.

229. The method of claim 225, wherein the anti-VEGF agent is aflibercept and wherein the ocular sustained-release pharmaceutical composition comprises less than or equal to about 850 μg of aflibercept.

230. The method of claim 225, wherein the vitreous concentration of the anti-VEGF agent in said human's eye is maintained at a concentration greater than or equal to about 1 ng/mL for at least about 4 months.

231. The method of claim 225, the vitreous concentration of the anti-VEGF agent in said human's eye is maintained at a concentration greater than or equal to about 1 ng/mL for about 4 months to about 6 months.

232. The method of claim 225, wherein the anti-VEGF agent is aflibercept and the vitreous concentration in said human's eye of aflibercept is from about 1 ng/mL to about 10 ng/mL.

233. The method of claim 225, wherein the anti-VEGF agent is aflibercept and the vitreous concentration in said human's eye of aflibercept is greater than or equal to about 10 ng/mL.

234. An ocular, sustained-release pharmaceutical composition comprising:

(A) a gel comprising a biocompatible polymer matrix; and

(B) at least one protein microparticle (PuP) comprising bevacizumab and at least one pharmaceutically acceptable excipient;

wherein the gel encapsulates the PuP;

wherein the PuP has dimensions of 1 μm in diameter x 1 μm in height ± 10% in any dimension; and

wherein the pharmaceutical composition is formulated to achieve a score less than the negative control as determined with standardized fluorescein angiogram scoring using fluorescein angiography when administered to an eye of a rabbit.

235. The ocular, sustained-release pharmaceutical composition of claim 234, wherein the gel comprises 50 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising:

(i) 25 ± 5 wt % of an 8-arm PEG-NHS having a molecular weight of 15,000 Da;

(ii) 25 ± 5 wt % of an 8-arm PEG-amine having a molecular weight of 10,000 Da; and

wherein the therapeutic agent comprises about 22 ± 5 wt % of the pharmaceutical composition.

236. The ocular, sustained-release pharmaceutical composition of claim 234, wherein the gel comprises 50 ± 5 % wt of the pharmaceutical composition and contains a mixture of polymers comprising:

(i) 25 ± 5 wt % of an 4-arm PEG-NHS having a molecular weight of 10,000 Da;

(ii) 25 ± 5 wt % of an 4-arm PEG-amine having a molecular weight of 10,000 Da; and

wherein the therapeutic agent comprises about 18 ± 5 wt % of the pharmaceutical composition.

237. The ocular, sustained-release pharmaceutical composition of claim 234, wherein the ocular, sustained-release pharmaceutical composition is fabricated as a rod-shaped implant, a cylindrical implant, or a mesoparticle.

238. The ocular, sustained-release pharmaceutical composition of claim 234, wherein the ocular, sustained-release pharmaceutical composition is fabricated as a cylindrical implant having a length of about 0.425 mm ± 10% and a dimeter of about 3 mm ± 10%.

FIG. 1

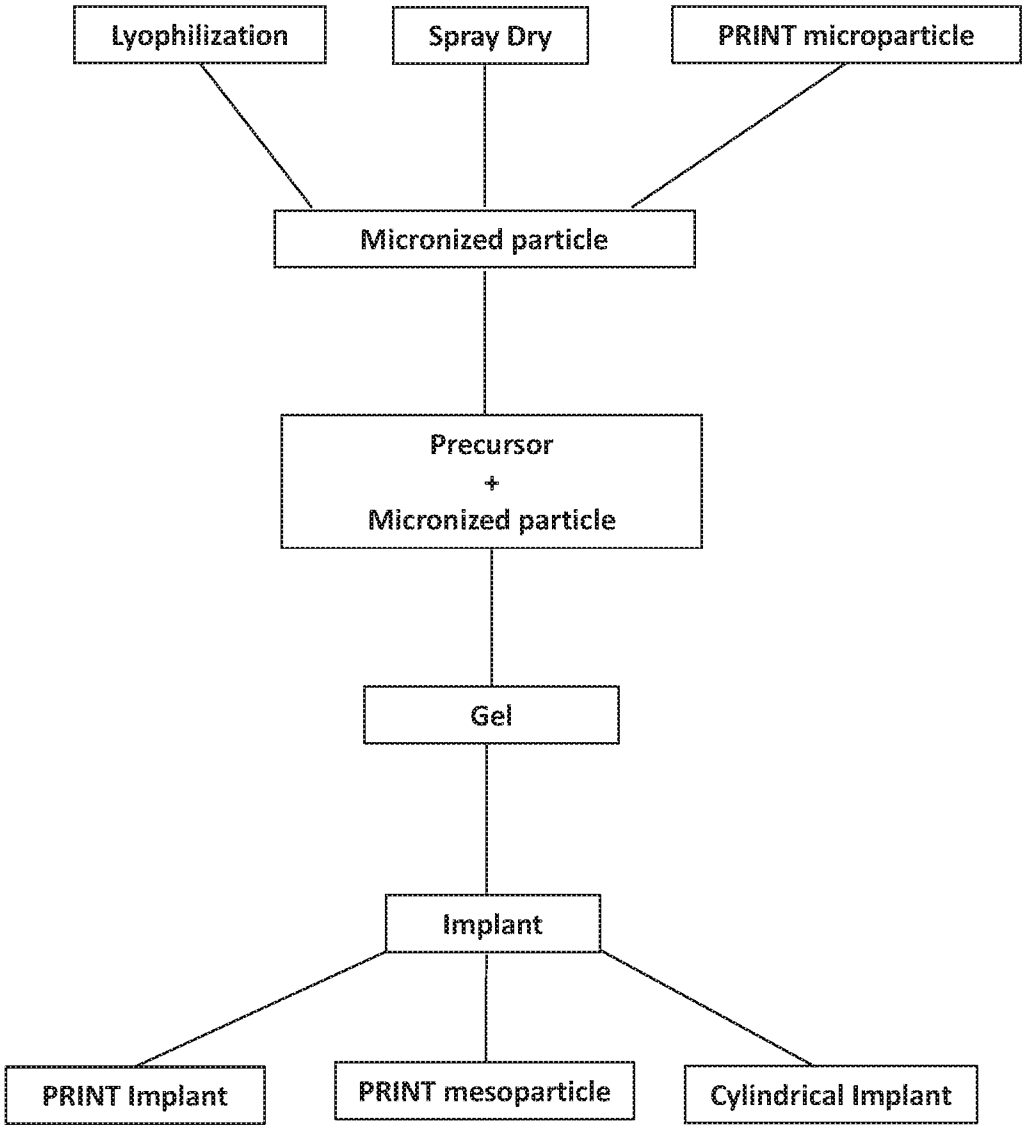

Lyophilization
Spray Dry
PRINT microparticle
Micronized particle
Precursor
+
Micronized particle
Gel
Implant
PRINT Implant
PRINT mesoparticle
Cylindrical Implant

FIG. 2

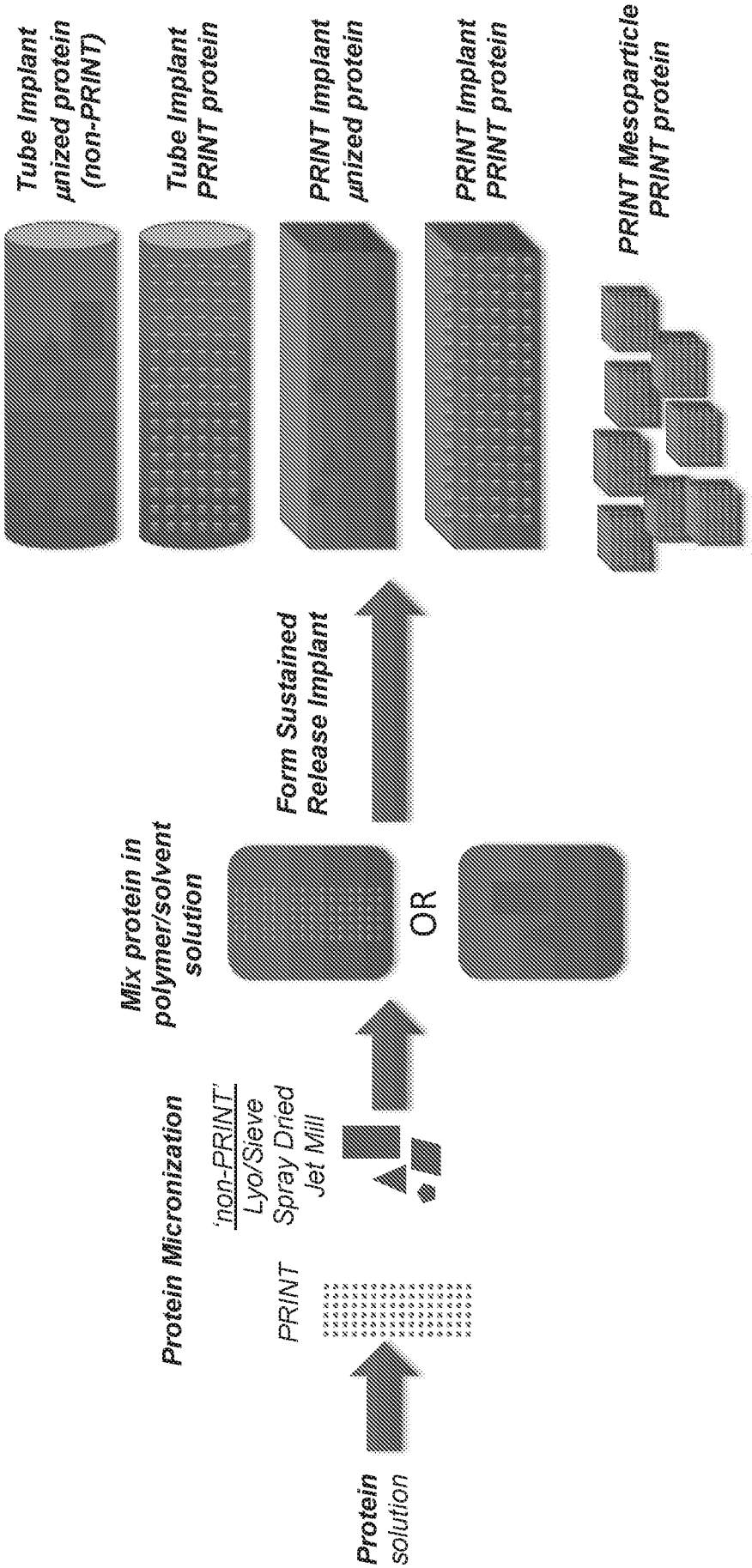

Protein solution
PRINT
'non-PRINT'
Lyo/Sieve
Spray Dried
Jet Mill
Protein Micronization
Mix protein in polymer/solvent solution
OR
Form Sustained Release Implant
Tube Implant µnized protein (non-PRINT)
Tube Implant PRINT protein
PRINT Implant µnized protein
PRINT Implant PRINT protein
PRINT Mesoparticle PRINT protein

FIG. 3

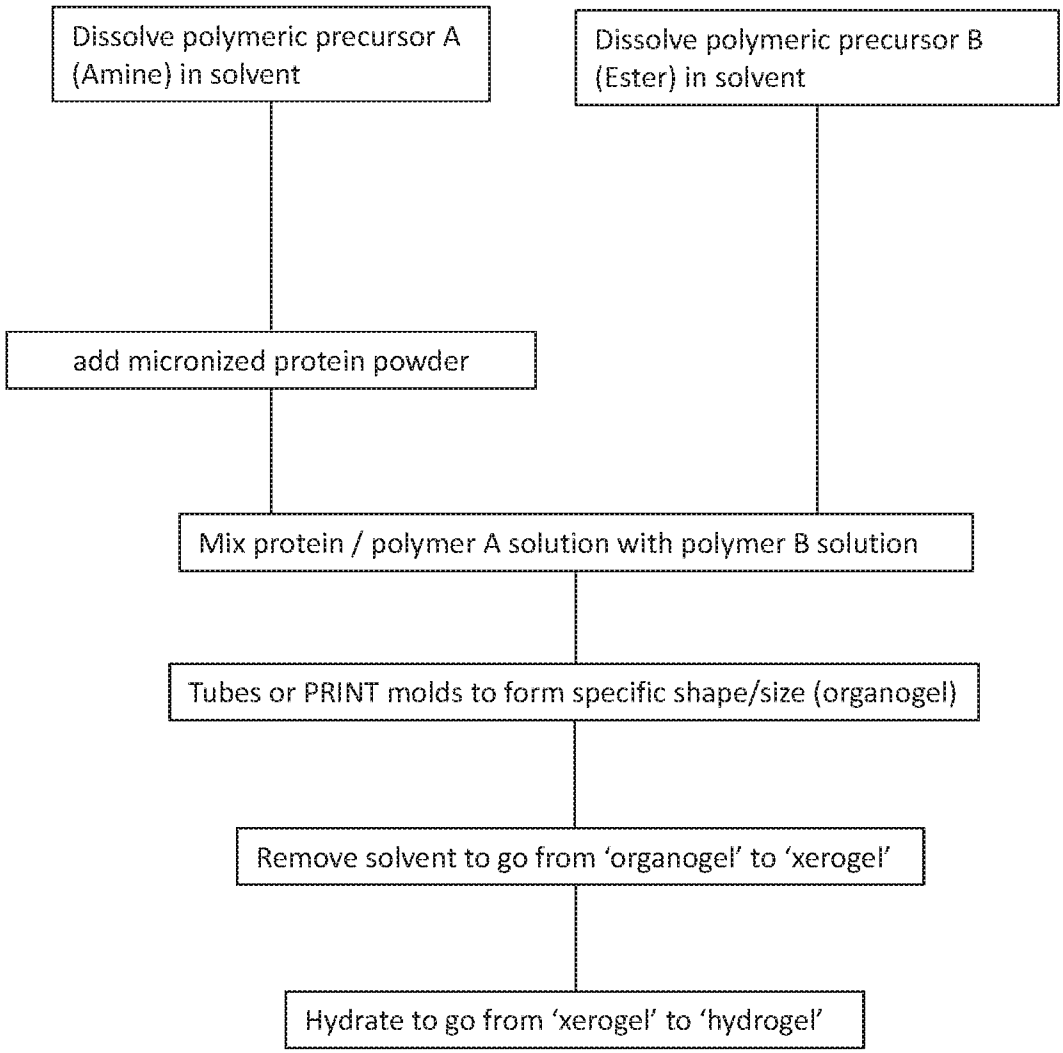

Dissolve polymeric precursor A (Amine) in solvent
Dissolve polymeric precursor B (Ester) in solvent
add micronized protein powder
Mix protein / polymer A solution with polymer B solution
Tubes or PRINT molds to form specific shape/size (organogel)
Remove solvent to go from 'organogel' to 'xerogel'
Hydrate to go from 'xerogel' to 'hydrogel'

FIG. 4A

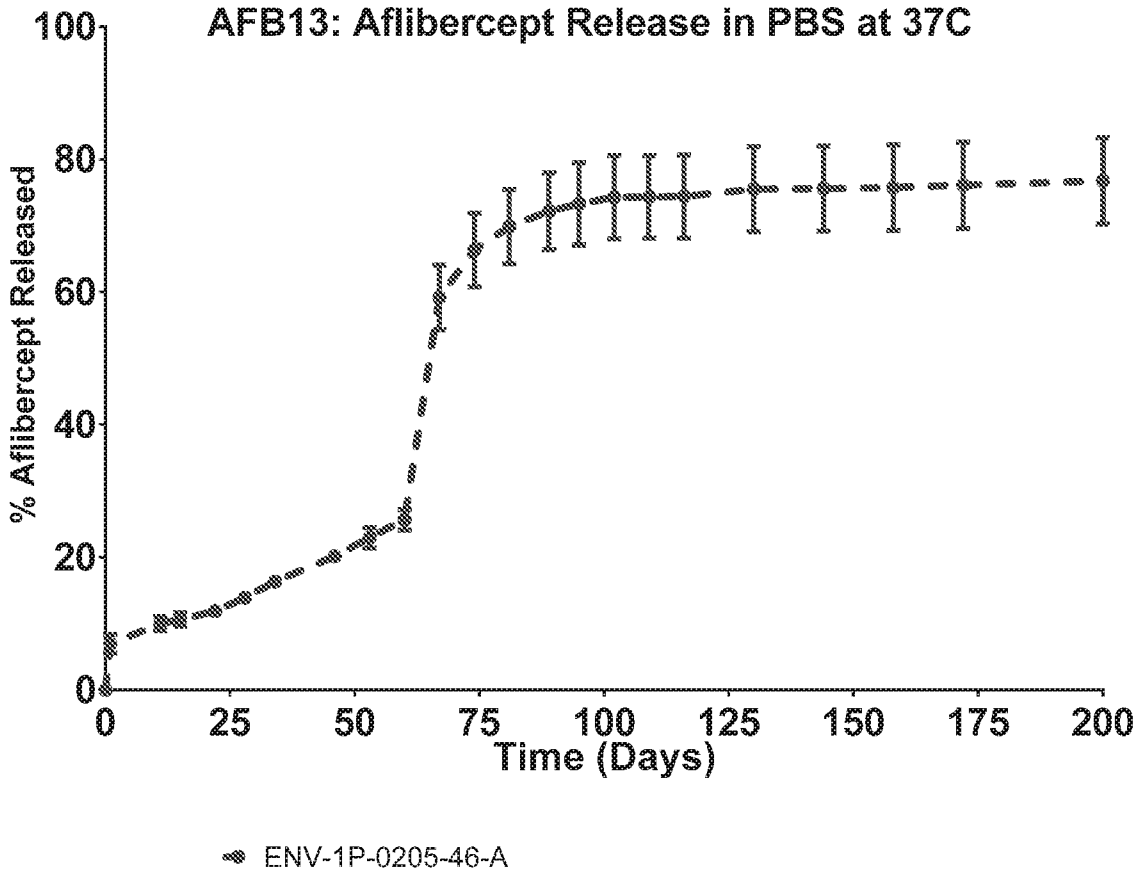

AFB13: Aflibercept Release in PBS at 37C
% Aflibercept Released
100
80
60
40
20
0
0
25
50
75
100
125
150
175
200
Time (Days)
ENV-1P-0205-46-A

FIG. 4B

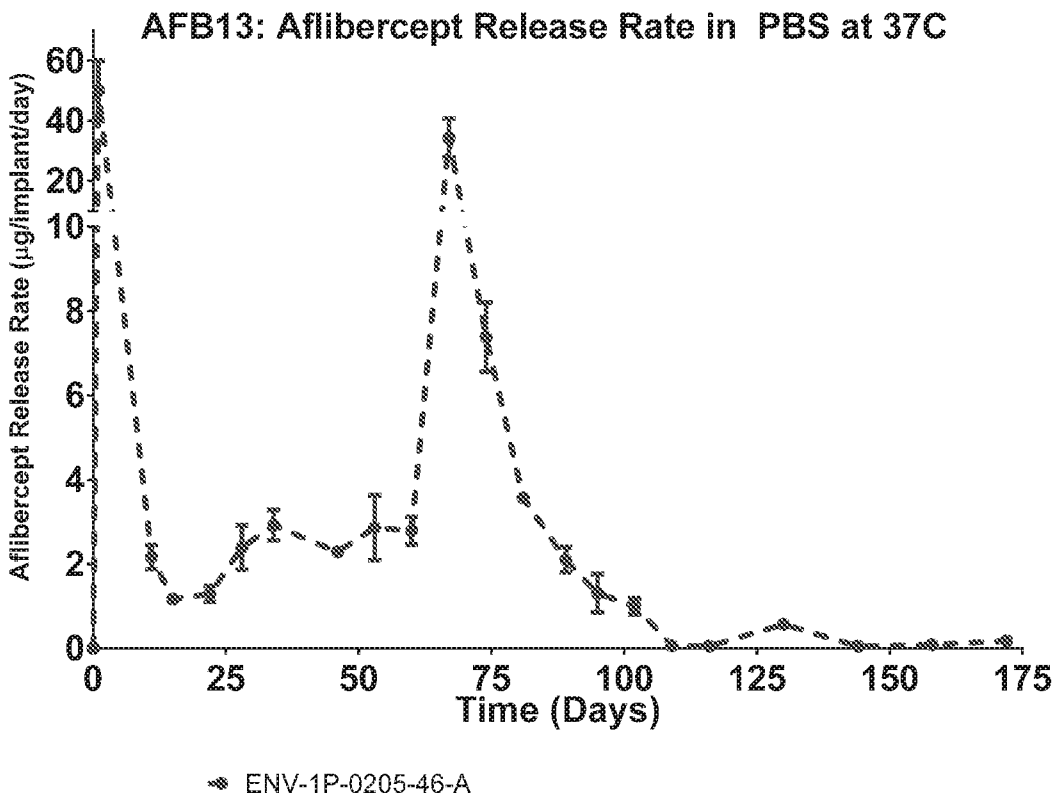

AFB13: Aflibercept Release Rate in PBS at 37C
Aflibercept Release Rate (µg/implant/day)
60
40
20
10
8
6
4
2
0
0
25
50
75
100
125
150
175
Time (Days)
ENV-1P-0205-46-A

FIG. 4C

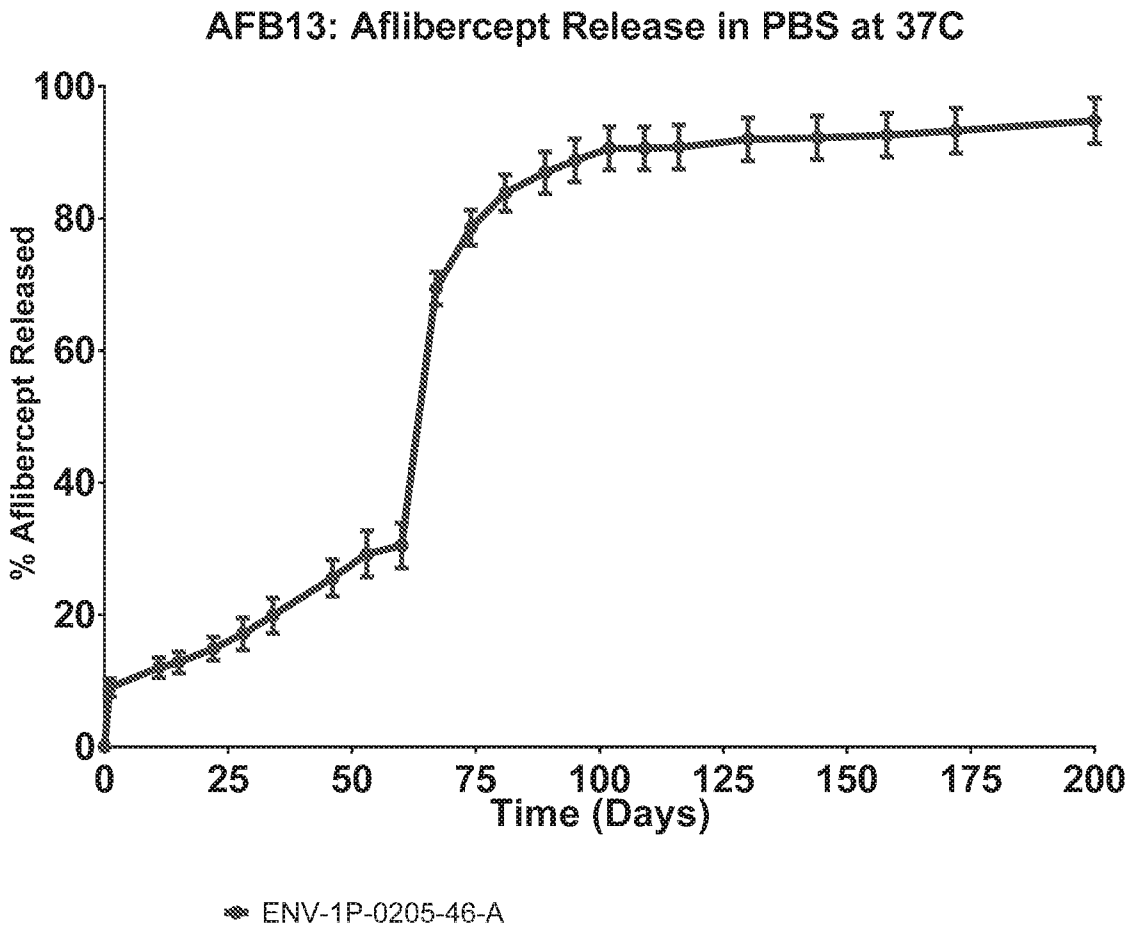

AFB13: Aflibercept Release in PBS at 37C
% Aflibercept Released
100
80
60
40
20
0
0
25
50
75
100
125
150
175
200
Time (Days)
ENV-1P-0205-46-A

FIG. 4D

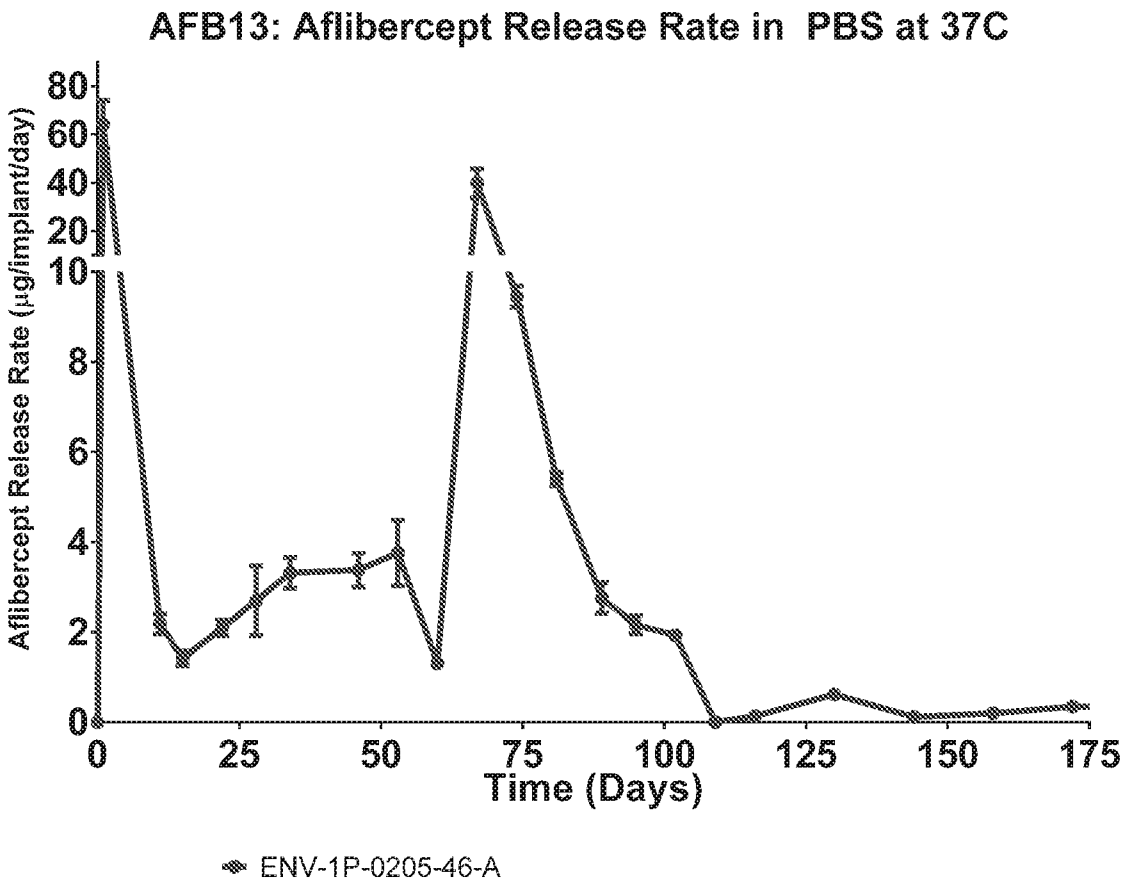

AFB13: Aflibercept Release Rate in PBS at 37C
Aflibercept Release Rate (µg/implant/day)
80
60
40
20
10
8
6
4
2
0
0
25
50
75
100
125
150
175
Time (Days)
ENV-1P-0205-46-A

FIG. 4E

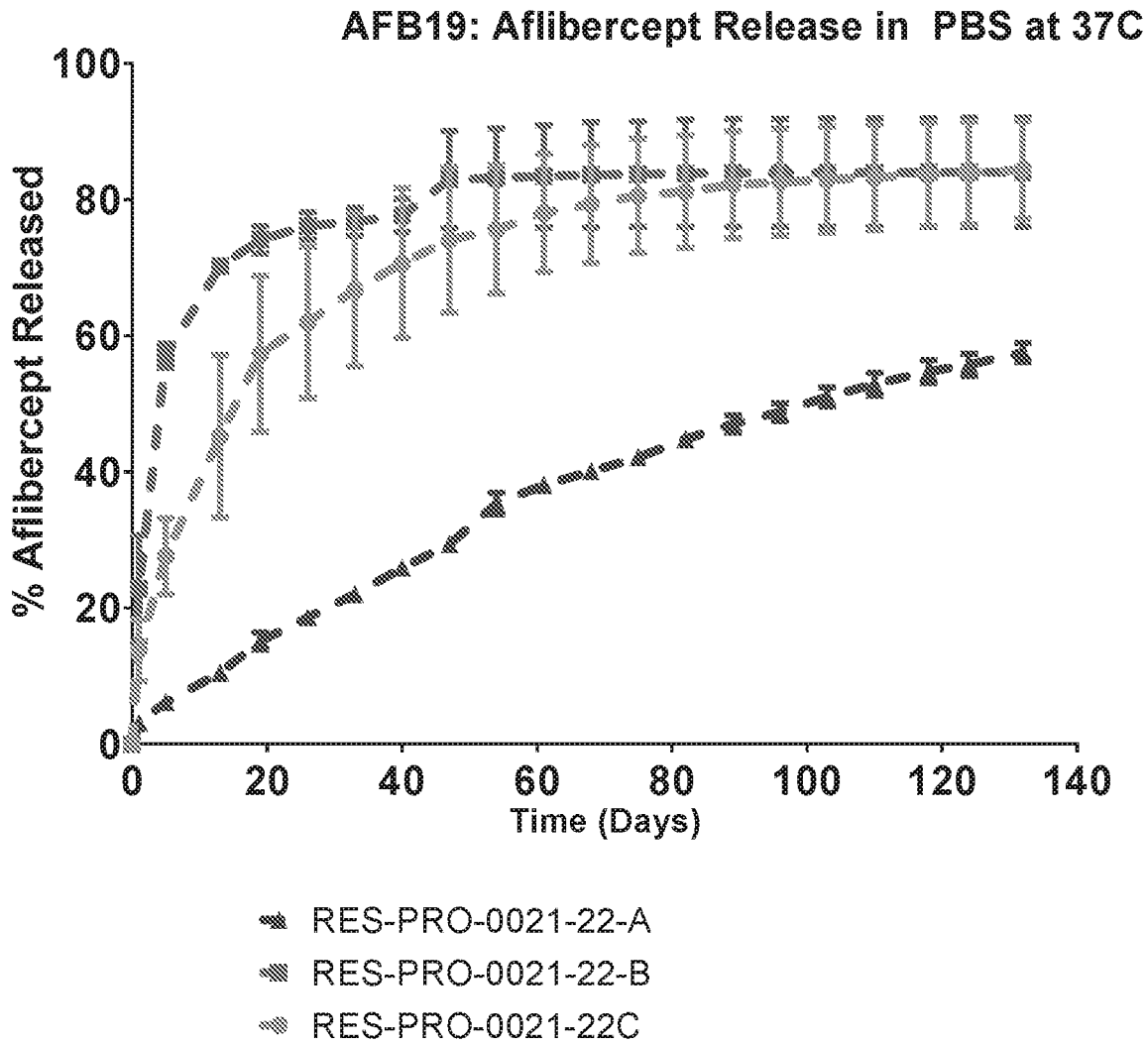

AFB19: Aflibercept Release in PBS at 37C
% Aflibercept Released
100
80
60
40
20
0
0
20
40
60
80
100
120
140
Time (Days)
RES-PRO-0021-22-A
RES-PRO-0021-22-B
RES-PRO-0021-22C

FIG. 4F

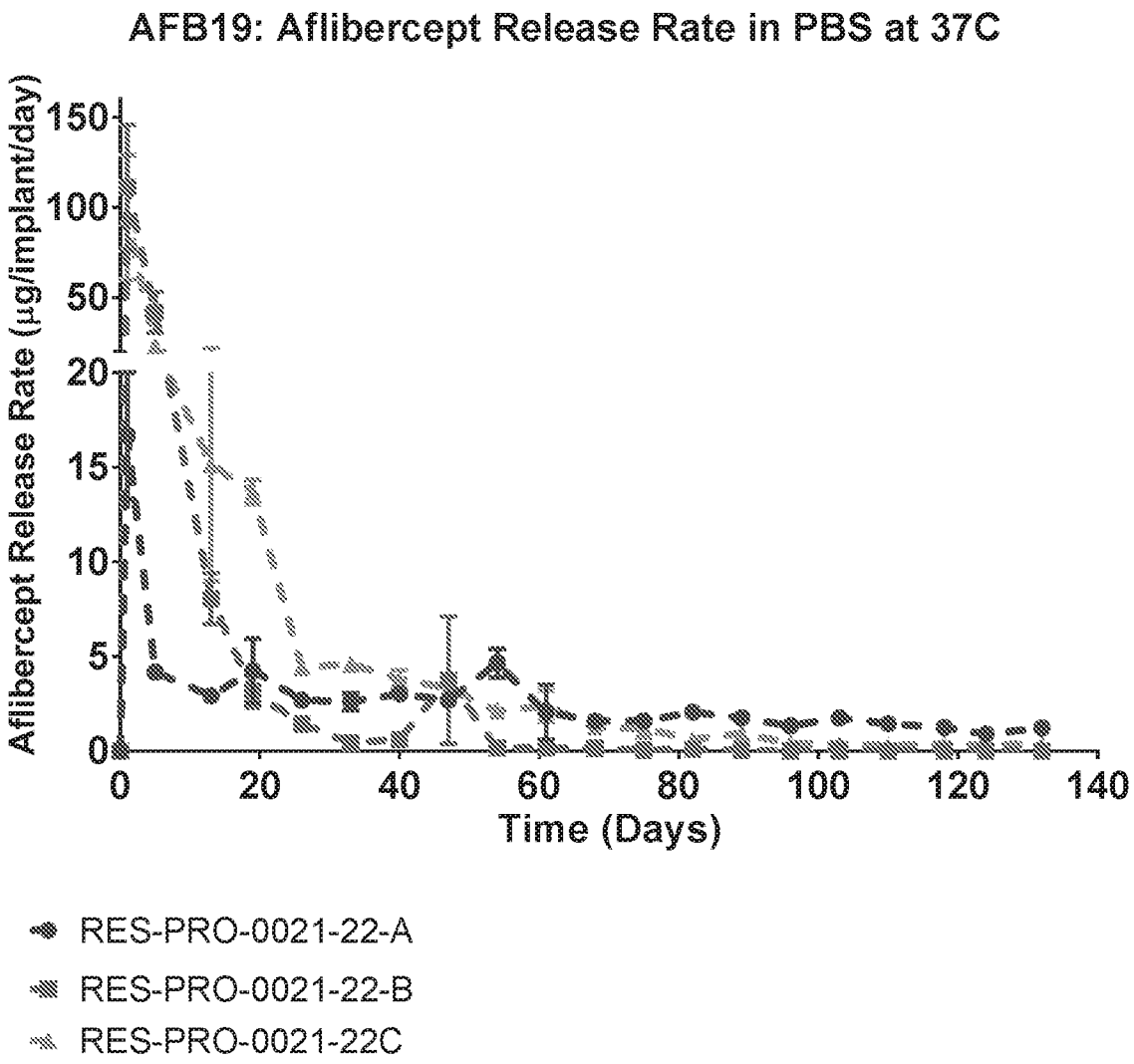

AFB19: Aflibercept Release Rate in PBS at 37C
Aflibercept Release Rate (µg/implant/day)
150
100
50
20
15
10
5
0
0
20
40
60
80
100
120
140
Time (Days)
RES-PRO-0021-22-A
RES-PRO-0021-22-B
RES-PRO-0021-22C

FIG. 4G

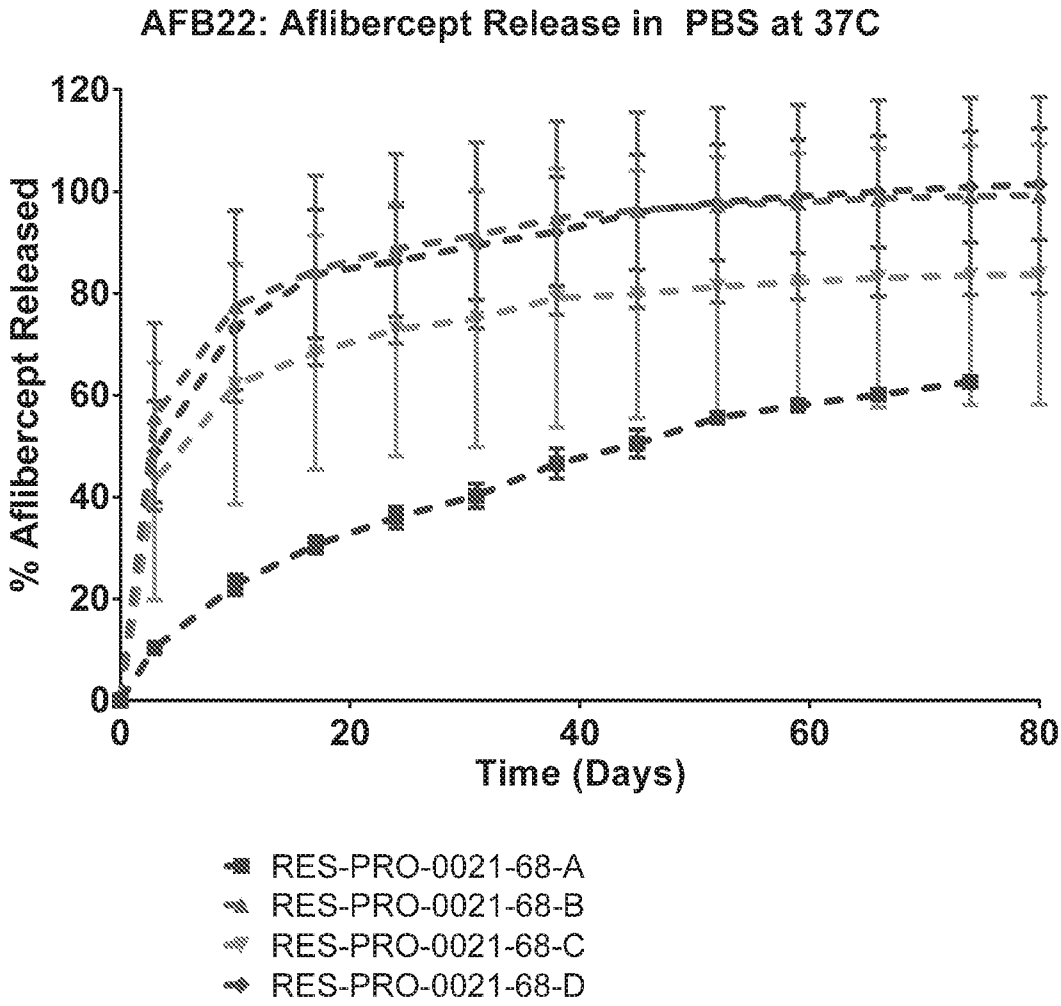

AFB22: Aflibercept Release in PBS at 37C
% Aflibercept Released
120
100
80
60
40
20
0
0
20
40
60
80
Time (Days)
RES-PRO-0021-68-A
RES-PRO-0021-68-B
RES-PRO-0021-68-C
RES-PRO-0021-68-D

FIG. 4 H

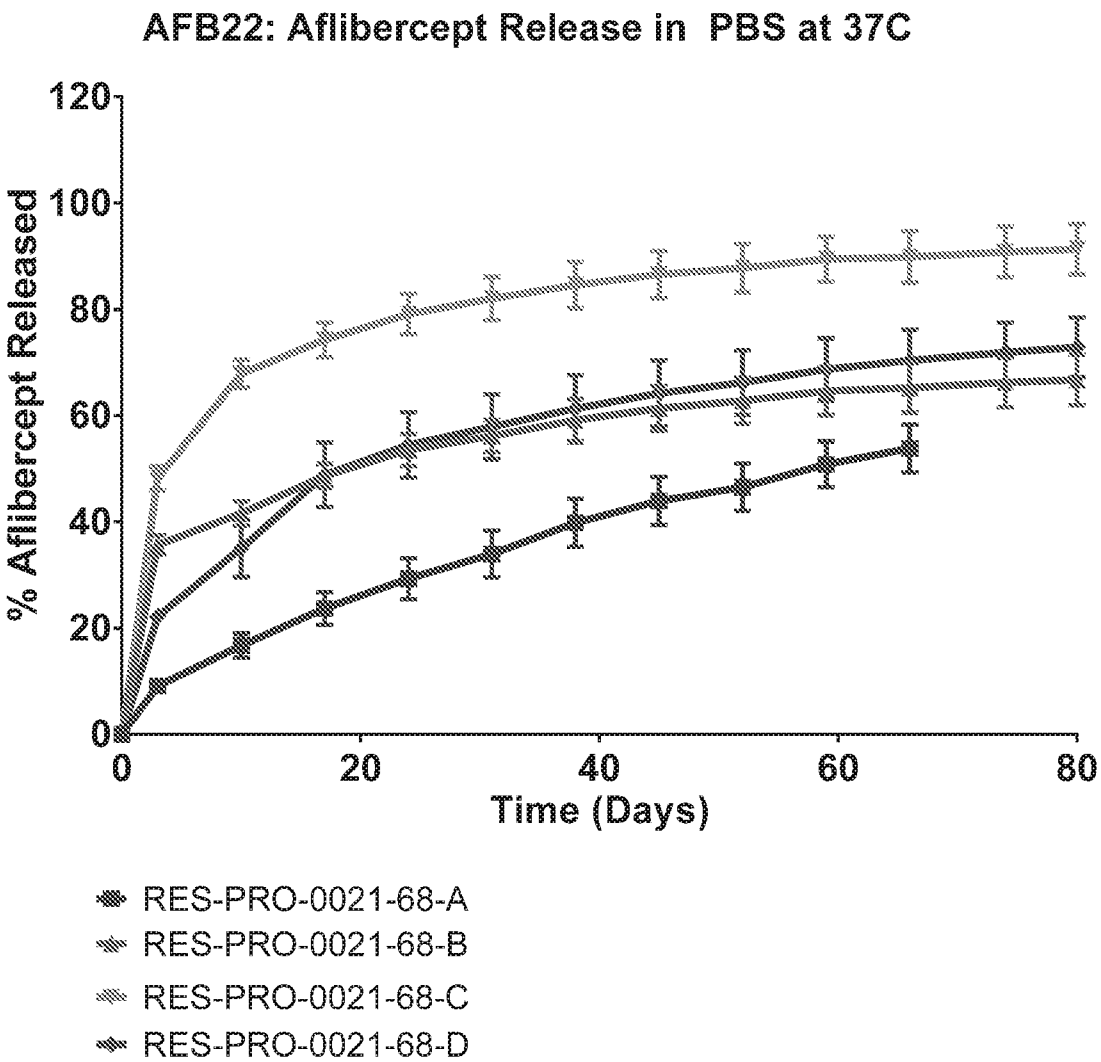

AFB22: Aflibercept Release in PBS at 37C
% Aflibercept Released
120
100
80
60
40
20
0
0
20
40
60
80
Time (Days)
RES-PRO-0021-68-A
RES-PRO-0021-68-B
RES-PRO-0021-68-C
RES-PRO-0021-68-D

FIG. 4I

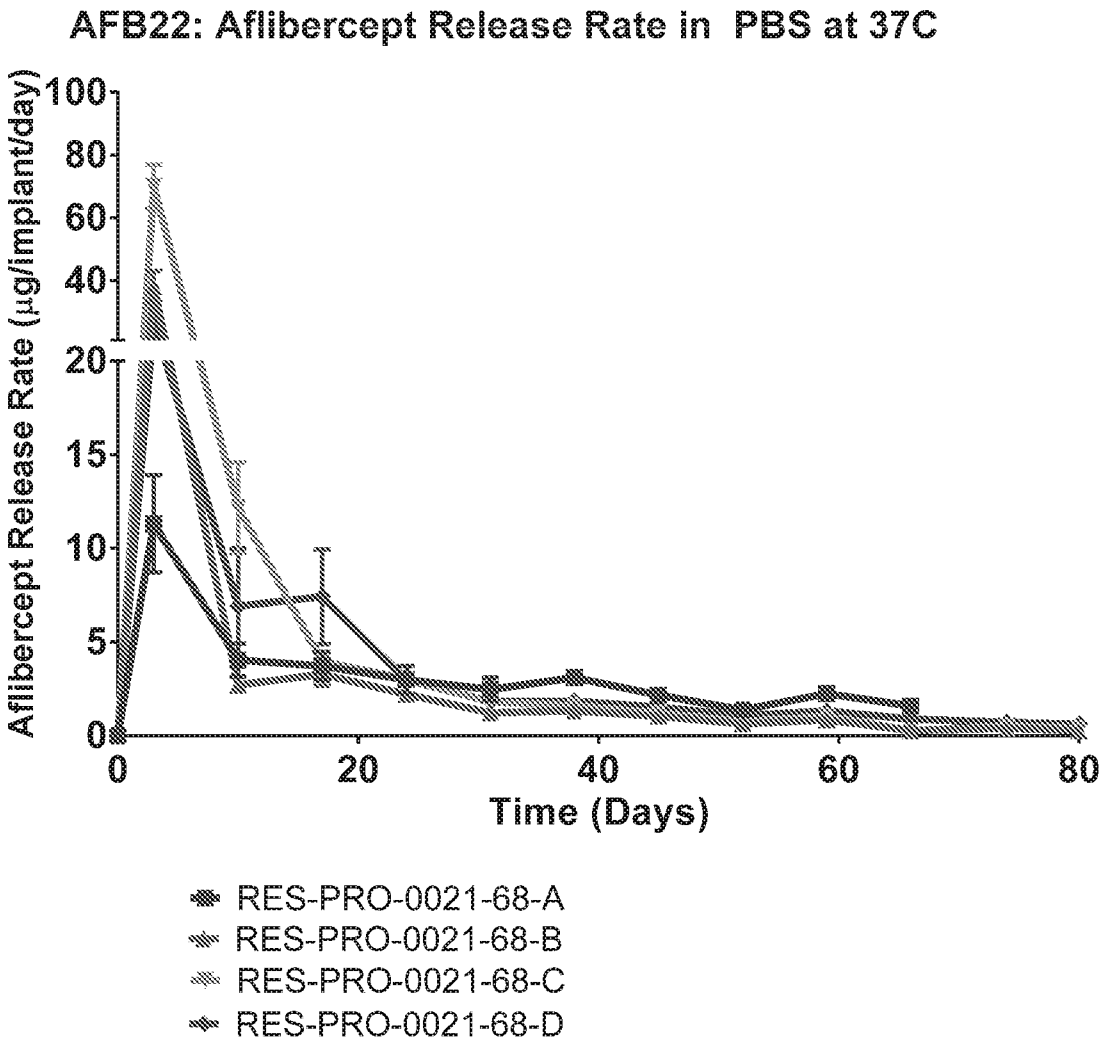

AFB22: Aflibercept Release Rate in PBS at 37C
Aflibercept Release Rate (µg/implant/day)
100
80
60
40
20
15
10
5
0
0
20
40
60
80
Time (Days)
RES-PRO-0021-68-A
RES-PRO-0021-68-B
RES-PRO-0021-68-C
RES-PRO-0021-68-D

FIG. 4J

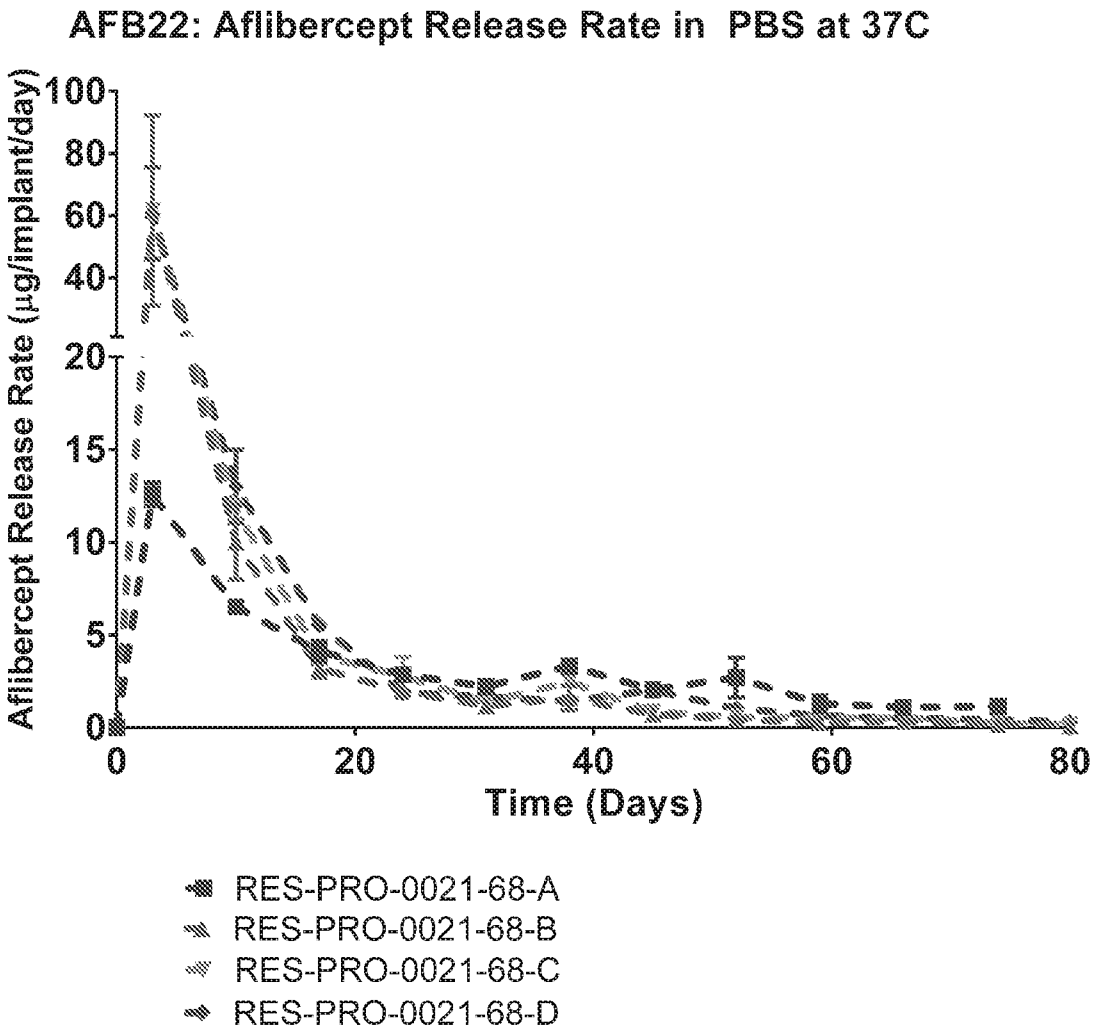

AFB22: Aflibercept Release Rate in PBS at 37C
Aflibercept Release Rate (µg/implant/day)
100
80
60
40
20
15
10
5
0
0
20
40
60
80
Time (Days)
RES-PRO-0021-68-A
RES-PRO-0021-68-B
RES-PRO-0021-68-C
RES-PRO-0021-68-D

FIG. 4K

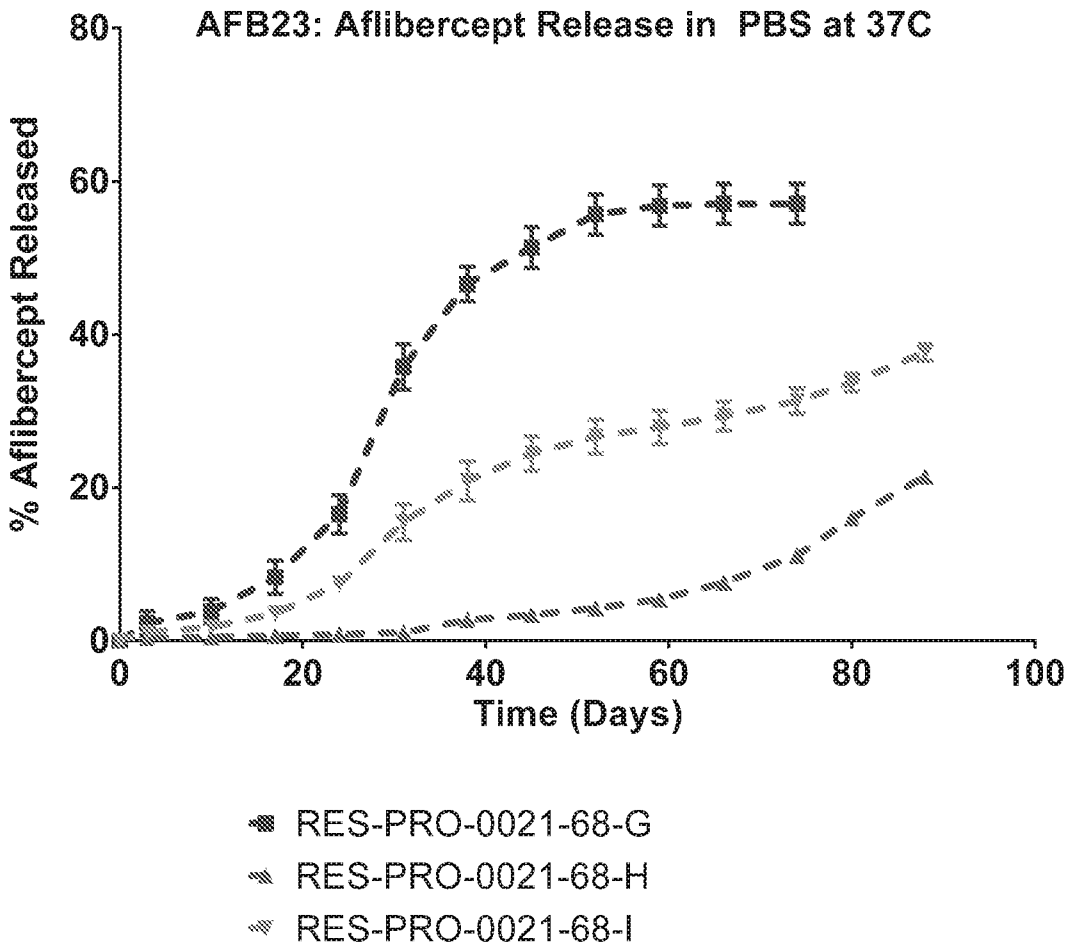

AFB23: Aflibercept Release in PBS at 37C
% Aflibercept Released
0
20
40
60
80
0
20
40
60
80
100
Time (Days)
RES-PRO-0021-68-G
RES-PRO-0021-68-H
RES-PRO-0021-68-I

FIG. 4L

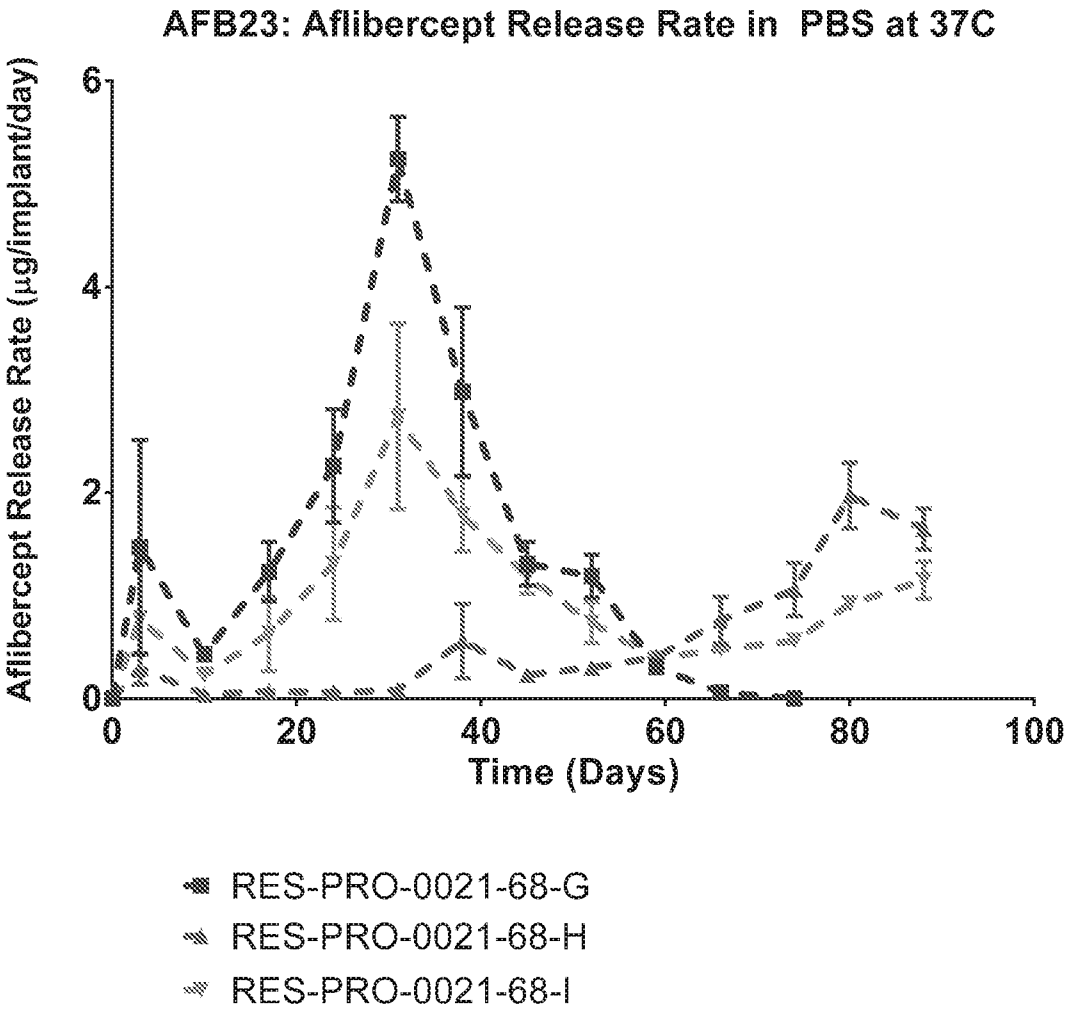

AFB23: Aflibercept Release Rate in PBS at 37C
Aflibercept Release Rate (µg/implant/day)
0
2
4
6
0
20
40
60
80
100
Time (Days)
RES-PRO-0021-68-G
RES-PRO-0021-68-H
RES-PRO-0021-68-I

FIG. 5A

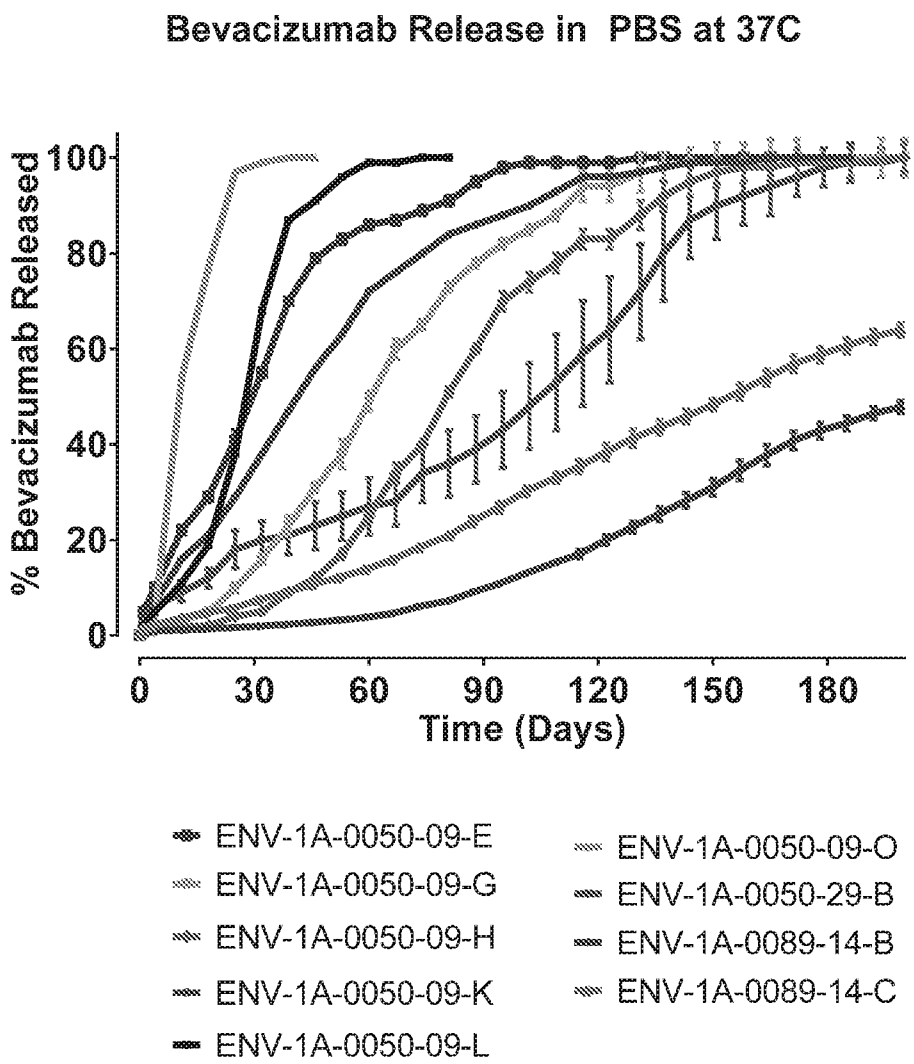

Bevacizumab Release in PBS at 37C
% Bevacizumab Released
100
80
60
40
20
0
0
30
60
90
120
150
180
Time (Days)
ENV-1A-0050-09-E
ENV-1A-0050-09-G
ENV-1A-0050-09-H
ENV-1A-0050-09-K
ENV-1A-0050-09-L
ENV-1A-0050-09-O
ENV-1A-0050-29-B
ENV-1A-0089-14-B
ENV-1A-0089-14-C

FIG. 5B

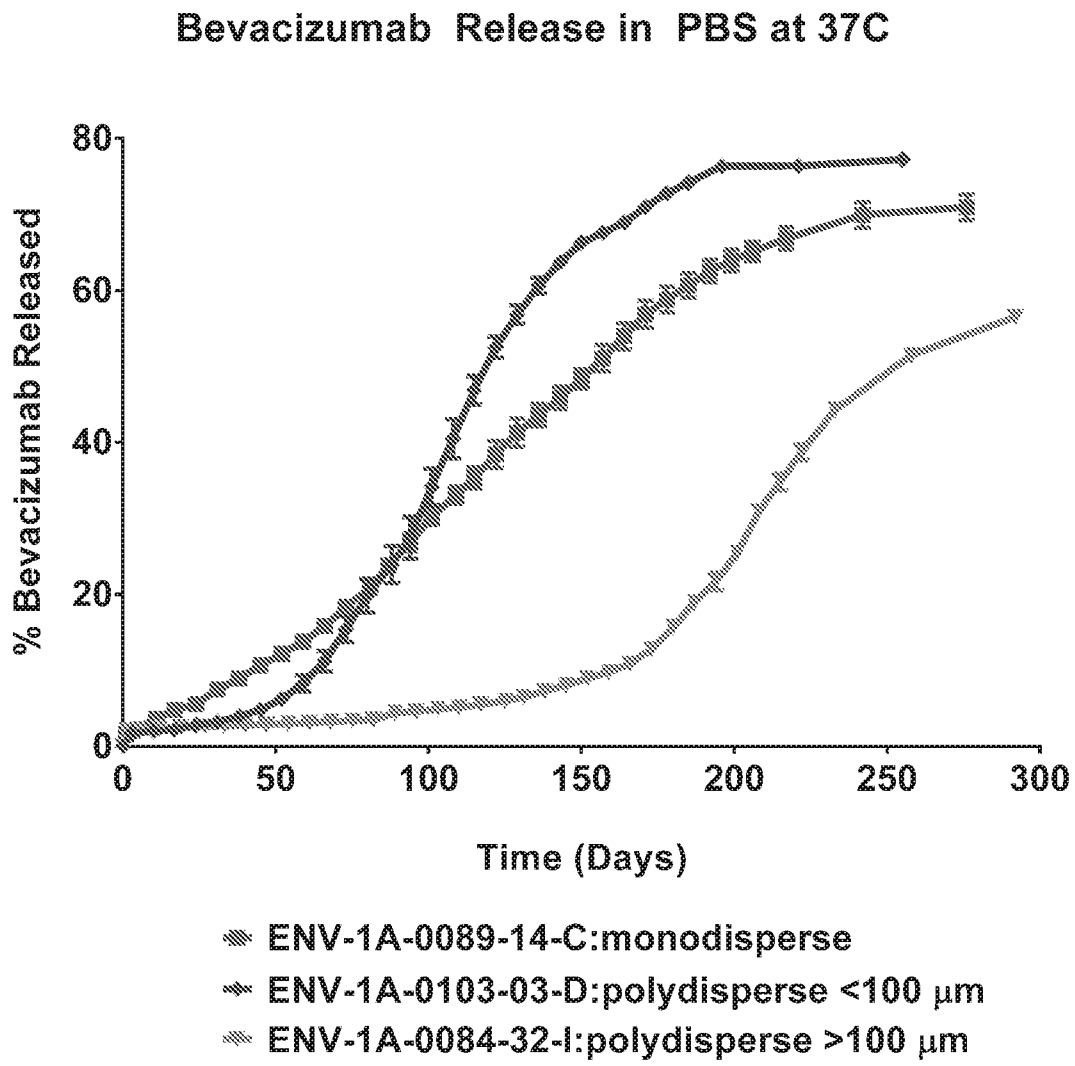

Bevacizumab Release in PBS at 37C
% Bevacizumab Released
0
20
40
60
80
0
50
100
150
200
250
300
Time (Days)
ENV-1A-0089-14-C:monodisperse
ENV-1A-0103-03-D:polydisperse <100 μm
ENV-1A-0084-32-I:polydisperse >100 μm

FIG. 6

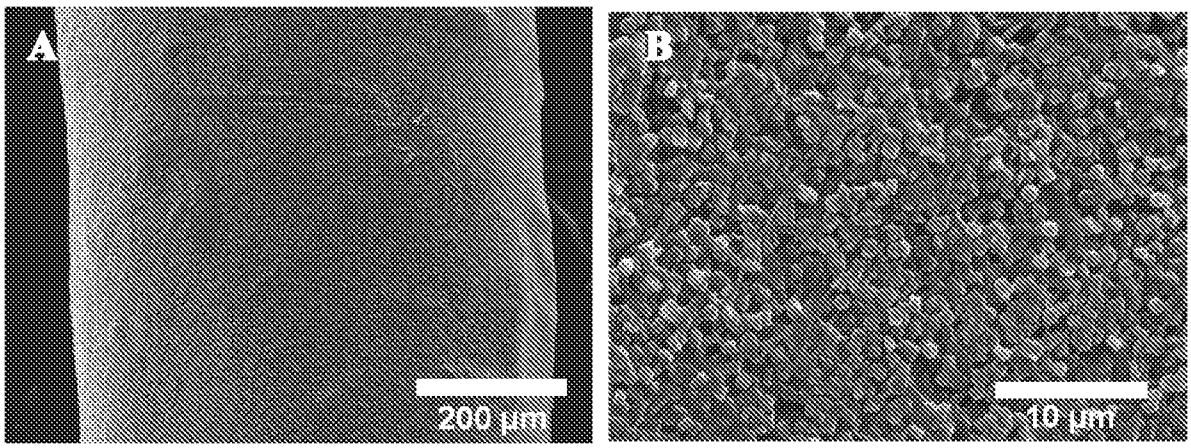


FIG. 7

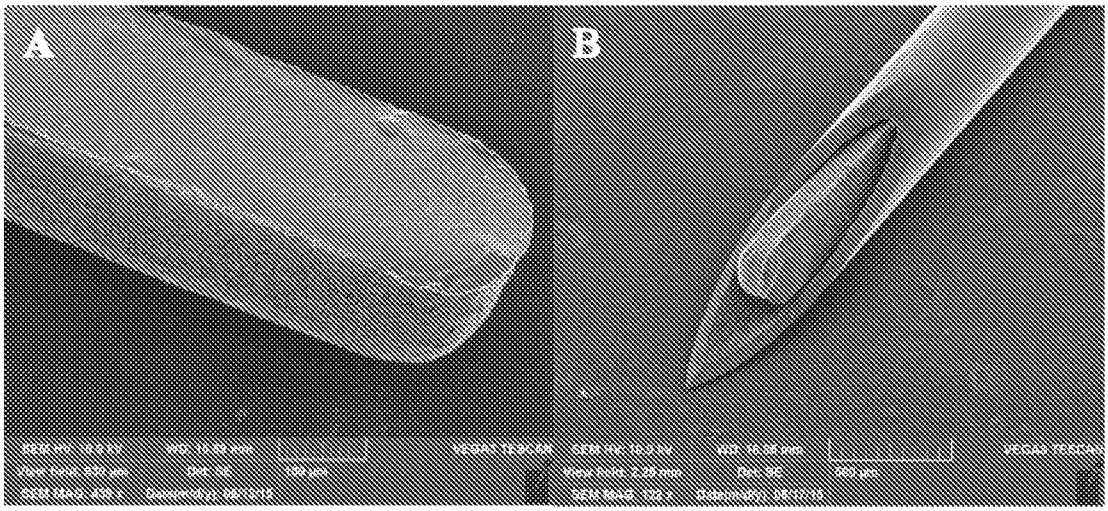


FIG. 8

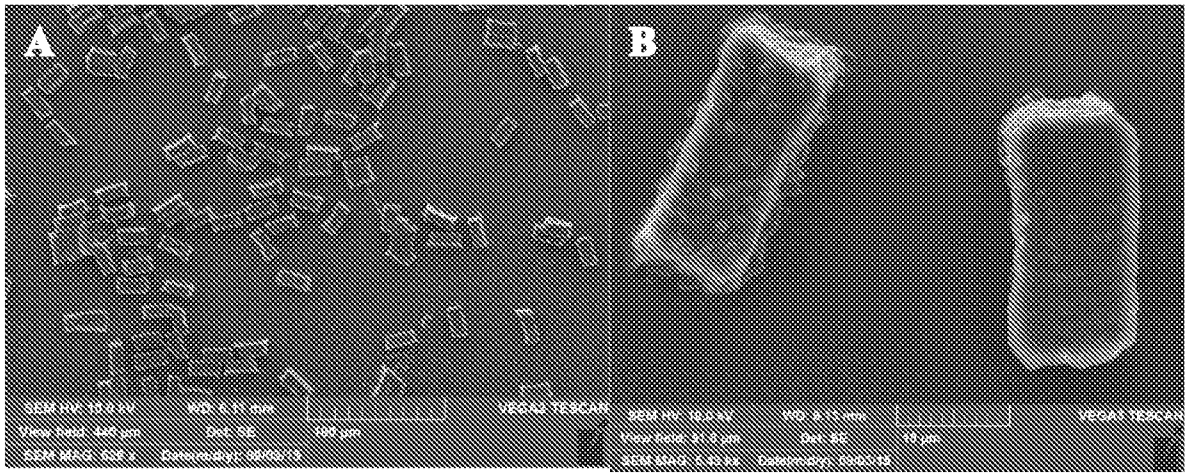


FIG. 9

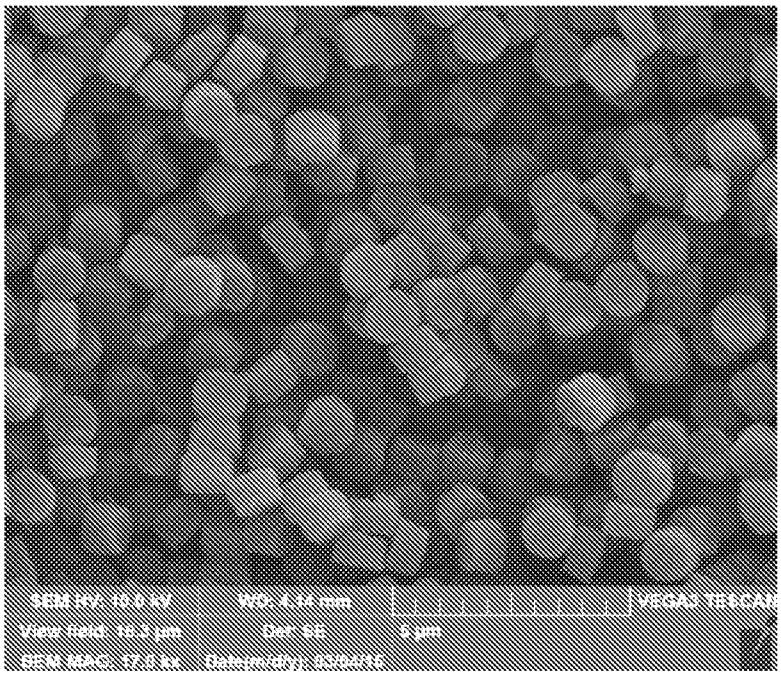

SEM HV: 10.0 kV
WD: 9.74 mm
View field: 19.3 μm
Det: SE
9 μm
SEM MAG: 17.8 kx
Chamber: 0.08 mTorr

FIG. 10

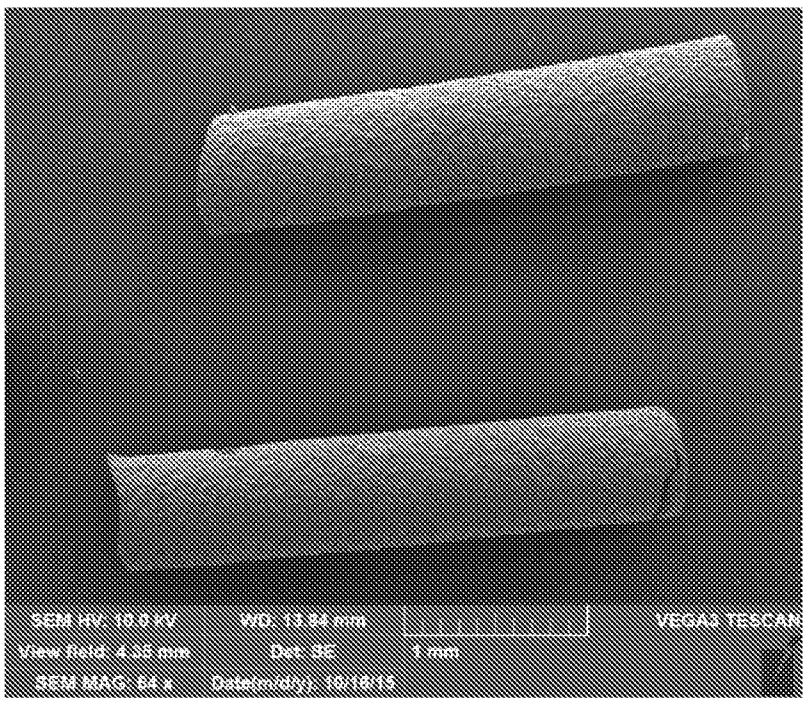

SEM HV: 10.0 kV
WD: 19.24 mm
VEGA3 TESCAN
View field: 4.35 mm
Det: SE
1 mm
SEM MAG: 64 x
Date(m/d/y): 10/10/13

FIG. 11

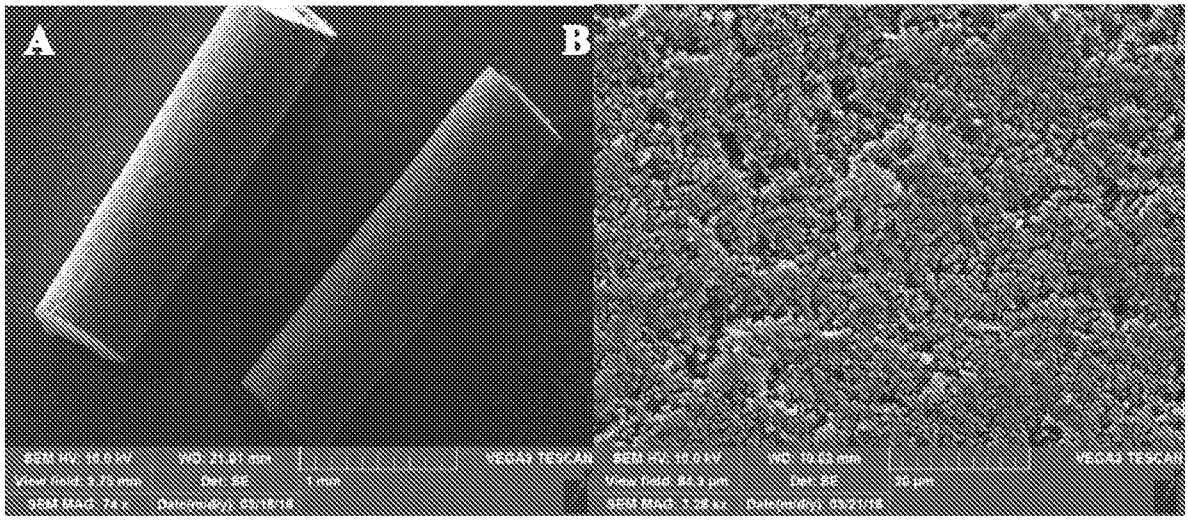


FIG. 12

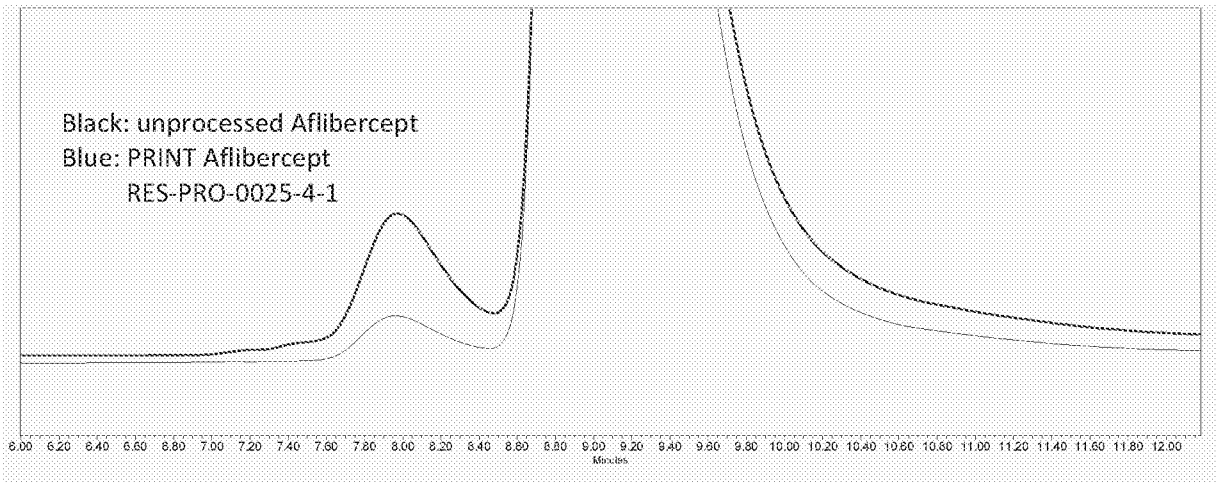

Black: unprocessed Aflibercept
Blue: PRINT Aflibercept
RES-PRO-0025-4-1
6.00 6.20 6.40 6.60 6.80 7.00 7.20 7.40 7.60 7.80 8.00 8.20 8.40 8.60 8.80 9.00 9.20 9.40 9.60 9.80 10.00 10.20 10.40 10.60 10.80 11.00 11.20 11.40 11.60 11.80 12.00
Minutes

FIG. 13

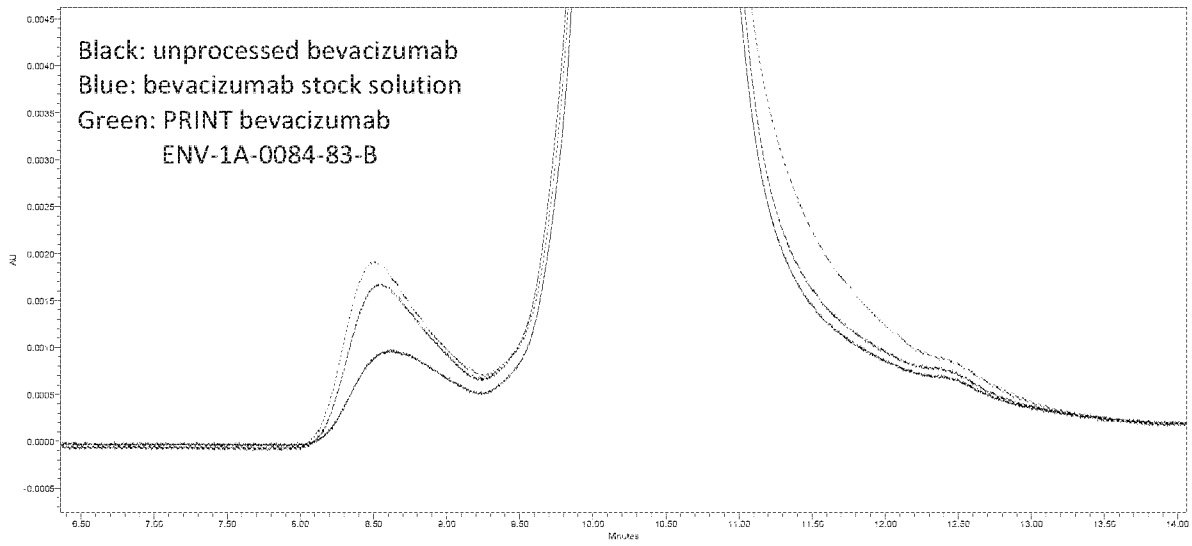
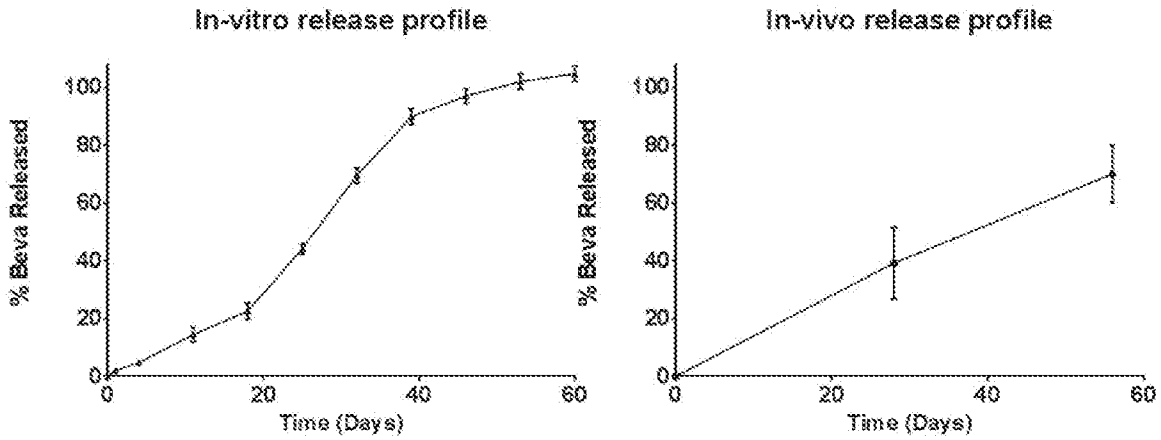

Black: unprocessed bevacizumab
Blue: bevacizumab stock solution
Green: PRINT bevacizumab
ENV-1A-0084-83-B
AU
Minutes

FIG. 14


In-vitro release profile
% Beva Released
100
80
60
40
20
0
0
20
40
60
Time (Days)
In-vivo release profile
% Beva Released
100
80
60
40
20
0
0
20
40
60
Time (Days)

A B

FIG. 15

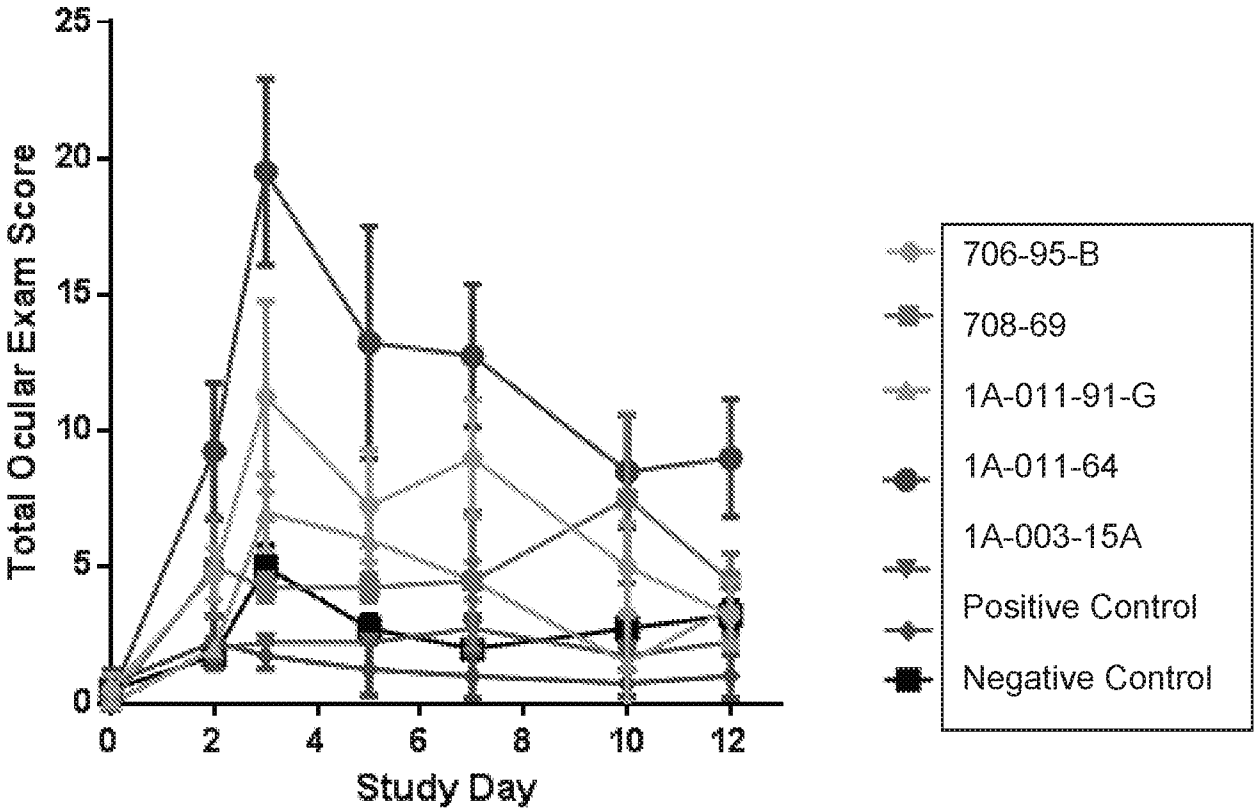

Total Ocular Exam Score
25
20
15
10
5
0
0
2
4
6
8
10
12
Study Day
706-95-B
708-69
1A-011-91-G
1A-011-64
1A-003-15A
Positive Control
Negative Control

FIG. 16

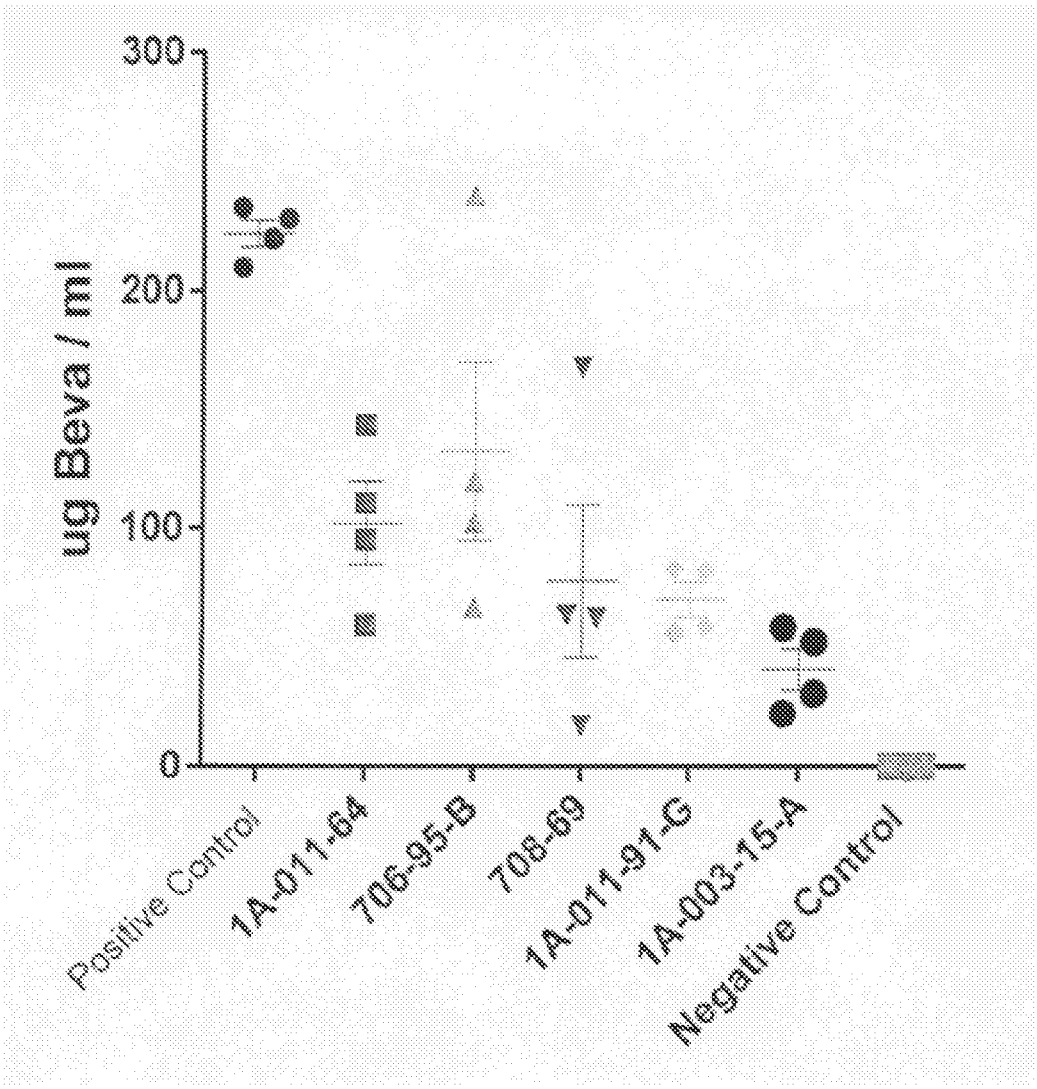

300
200
100
0
ug Beva / ml
Positive Control
1A-011-64
706-95-B
708-69
1A-011-91-G
1A-003-15-A
Negative Control

FIG. 17

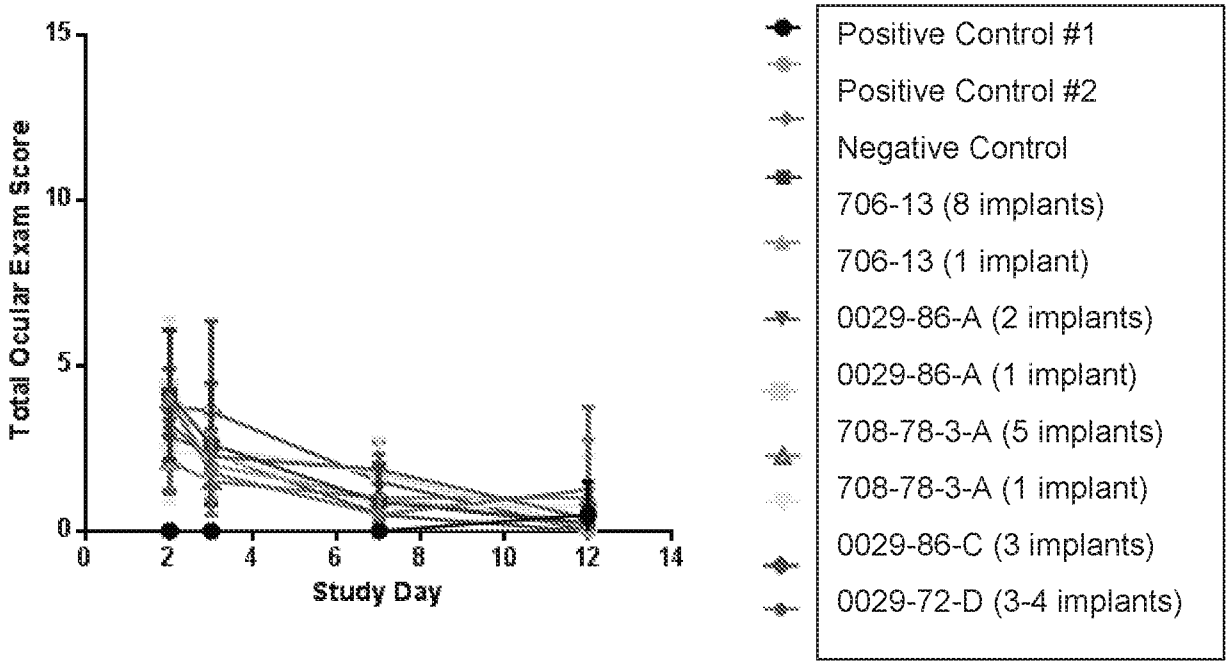

Total Ocular Exam Score
15
10
5
0
0
2
4
6
8
10
12
14
Study Day
Positive Control #1
Positive Control #2
Negative Control
706-13 (8 implants)
706-13 (1 implant)
0029-86-A (2 implants)
0029-86-A (1 implant)
708-78-3-A (5 implants)
708-78-3-A (1 implant)
0029-86-C (3 implants)
0029-72-D (3-4 implants)

FIG. 18

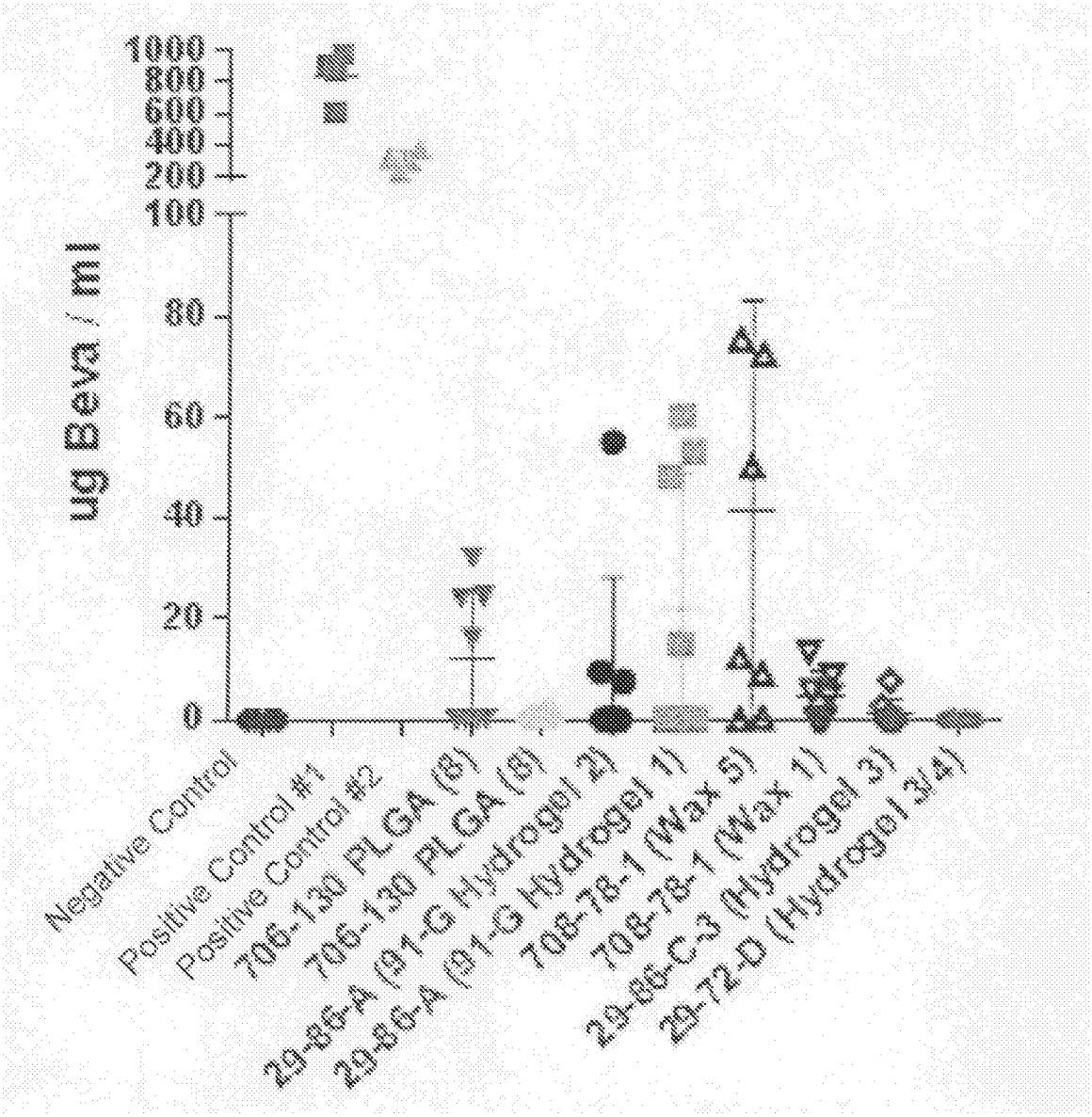

ug Bevi / ml
1000
800
600
400
200
100
80
60
40
20
0
Negative Control
Positive Control #1
Positive Control #2
706-130 PLGA (6)
706-130 PLGA (6)
29-86-A (91-G Hydrogel 2)
29-86-A (91-G Hydrogel 1)
708-78-1 (Wax 5)
708-78-1 (Wax 1)
29-86-C-3 (Hydrogel 3)
29-72-D (Hydrogel 3/4)

FIG. 19A

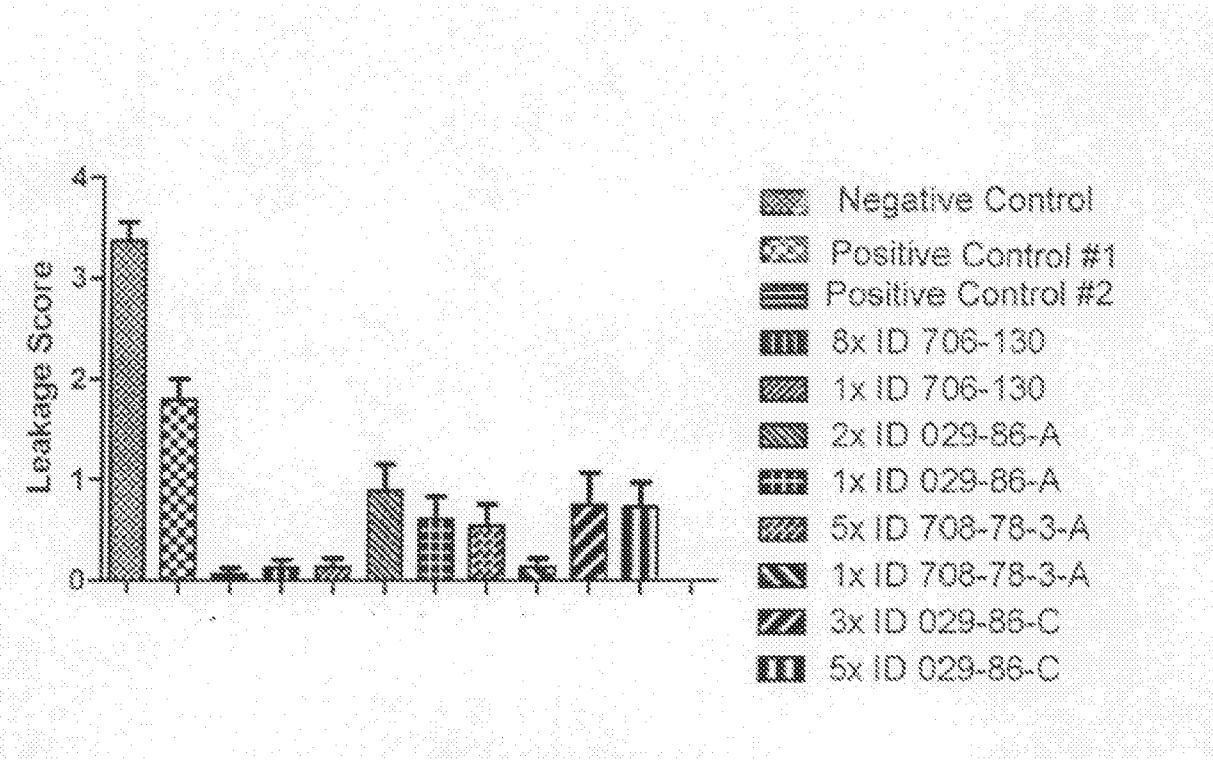

Leakage Score
0
1
2
3
4
Negative Control
Positive Control #1
Positive Control #2
8x ID 706-130
1x ID 706-130
2x ID 029-86-A
1x ID 029-86-A
5x ID 708-78-3-A
1x ID 708-78-3-A
3x ID 029-86-C
5x ID 029-86-C

FIG. 19B

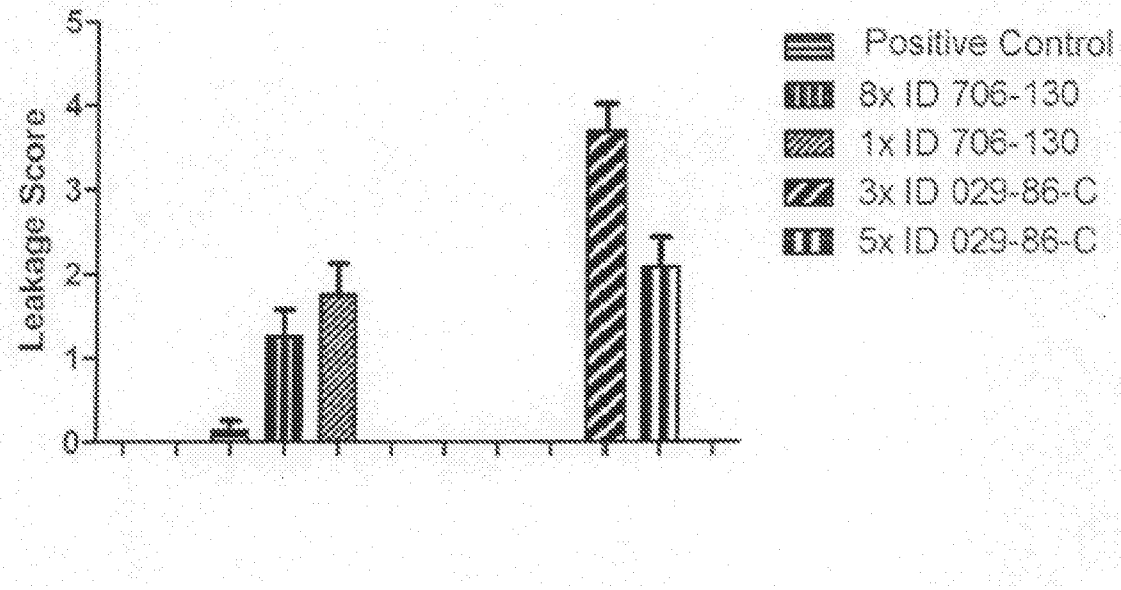

Leakage Score
0
1
2
3
4
5
Positive Control
8x ID 706-130
1x ID 706-130
3x ID 029-86-C
5x ID 029-86-C

FIG. 20

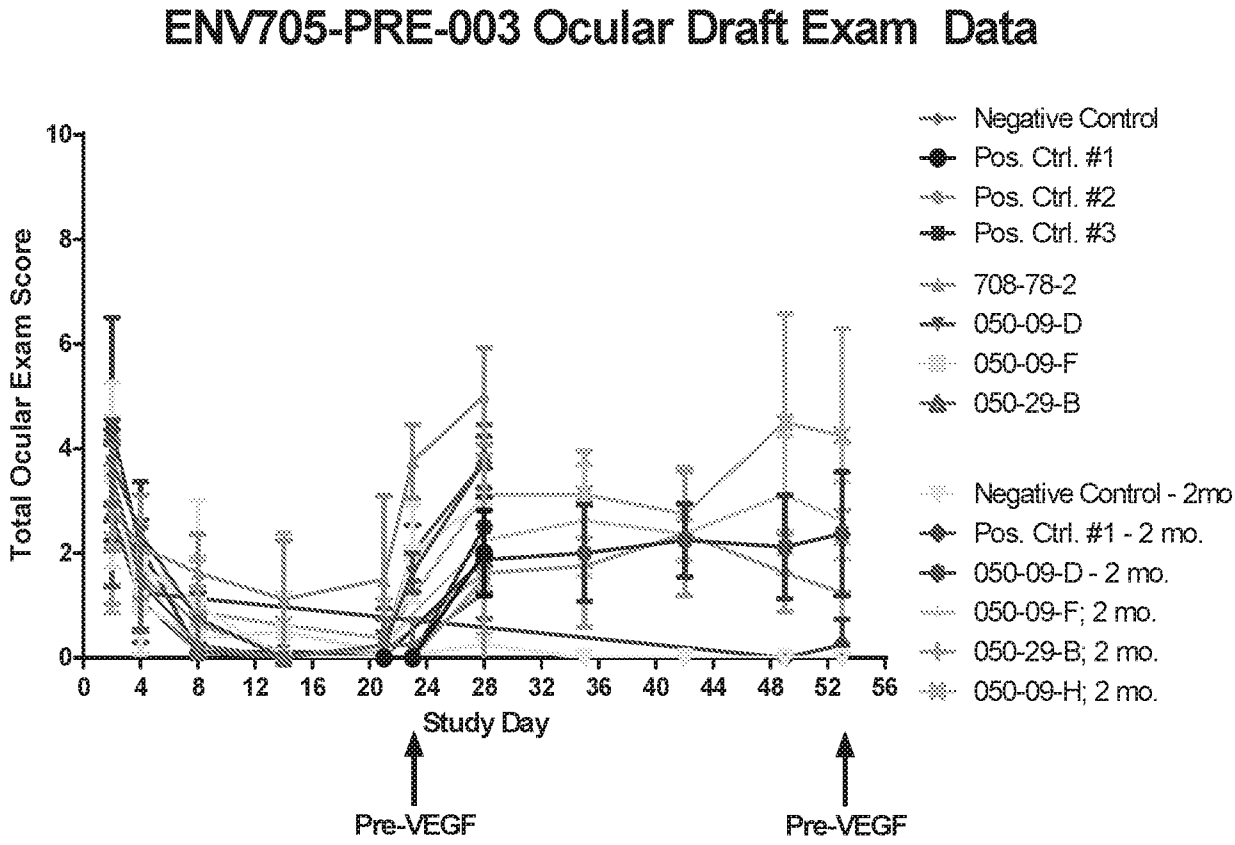

ENV705-PRE-003 Ocular Draft Exam Data
Total Ocular Exam Score
10
8
6
4
2
0
0 4 8 12 16 20 24 28 32 36 40 44 48 52 56
Study Day
Pre-VEGF
Pre-VEGF
Negative Control
Pos. Ctrl. #1
Pos. Ctrl. #2
Pos. Ctrl. #3
708-78-2
050-09-D
050-09-F
050-29-B
Negative Control - 2mo
Pos. Ctrl. #1 - 2 mo.
050-09-D - 2 mo.
050-09-F; 2 mo.
050-29-B; 2 mo.
050-09-H; 2 mo.

FIG. 21

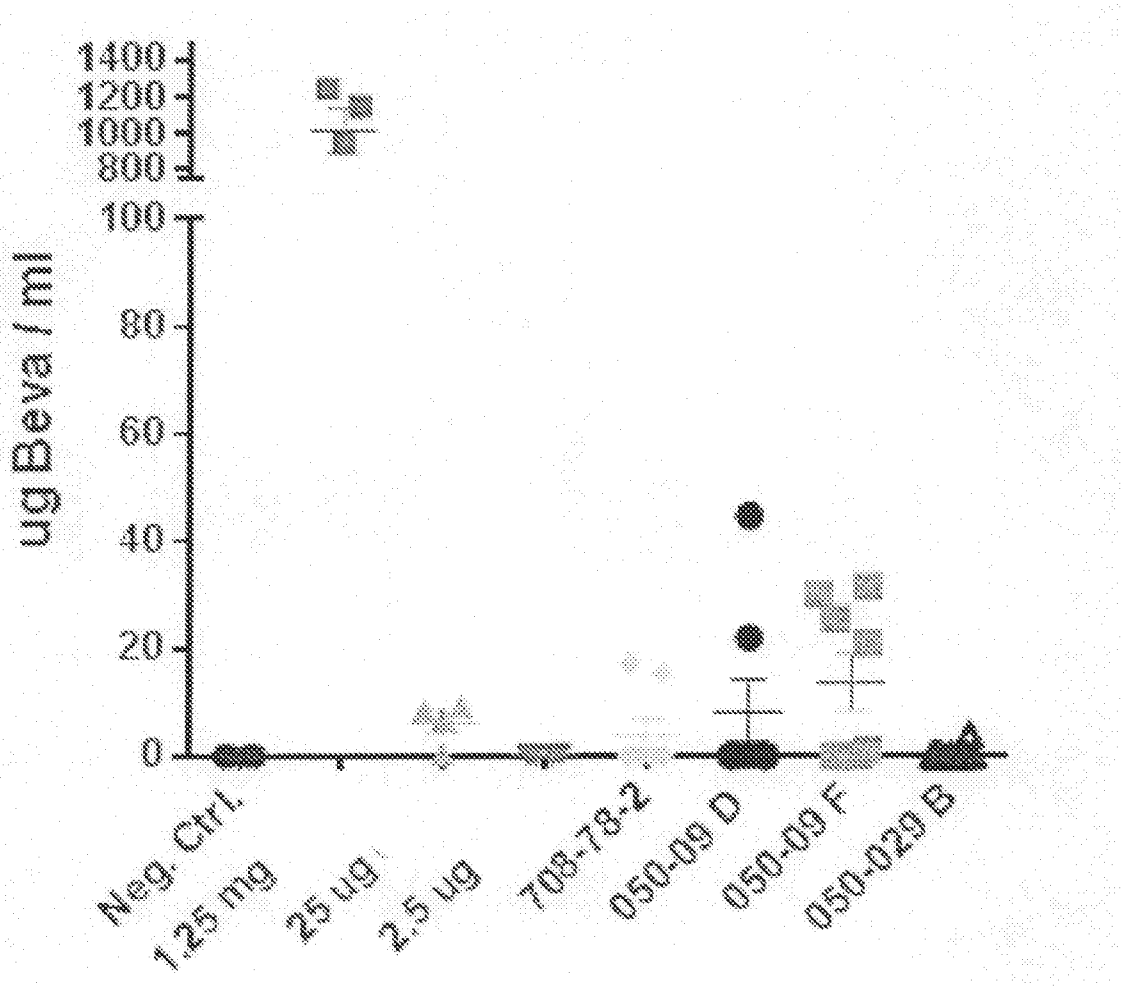

1400
1200
1000
800
100
80
60
40
20
0
ug Beva / ml
Neg. Ctrl.
1.25 ng
25 ug
2.5 ug
708-78-2
050-09 D
050-09 F
050-029 B

FIG. 22A

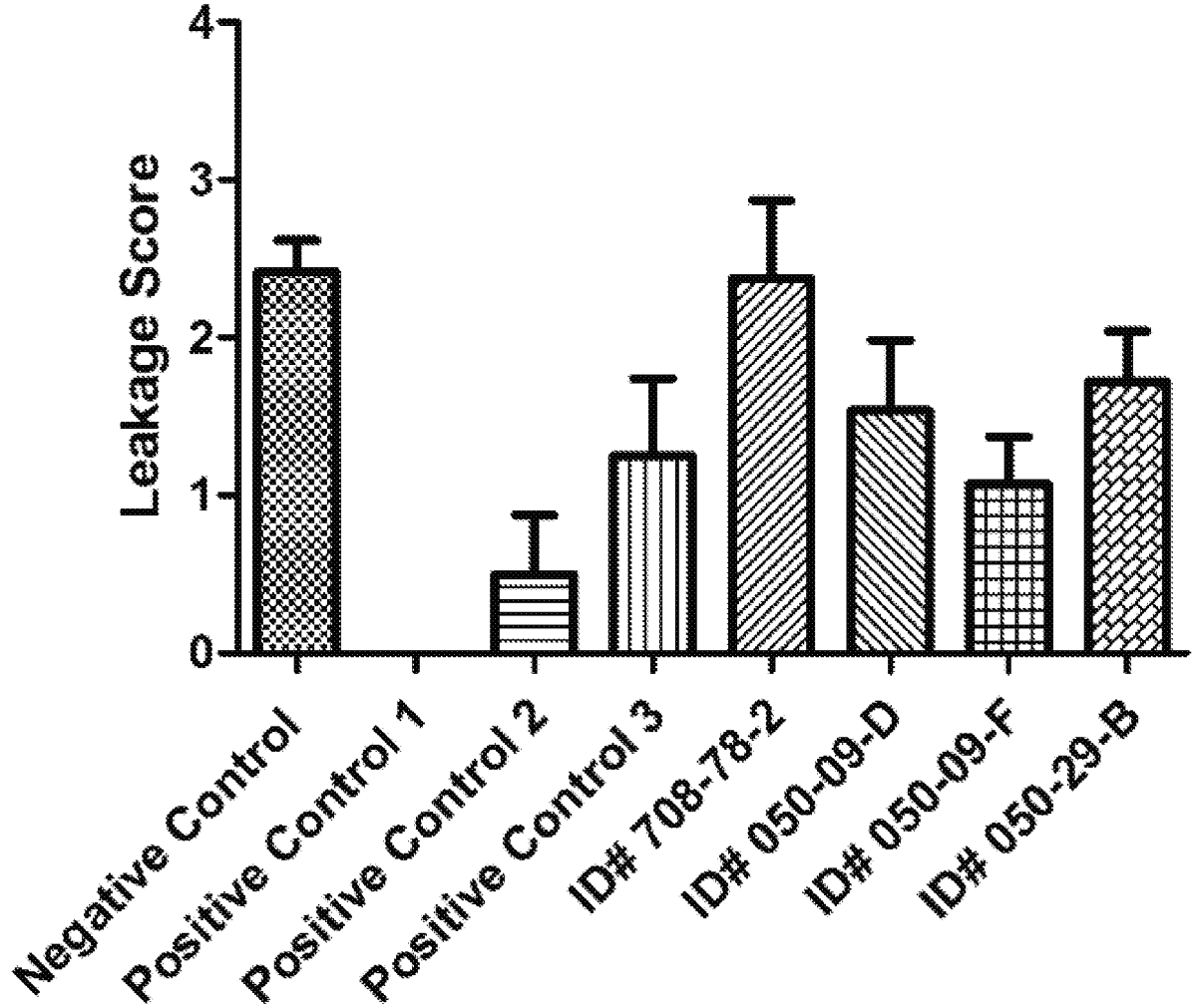

Leakage Score
0
1
2
3
4
Negative Control
Positive Control 1
Positive Control 2
Positive Control 3
ID# 708-78-2
ID# 050-09-D
ID# 050-09-F
ID# 050-29-B

FIG. 22B

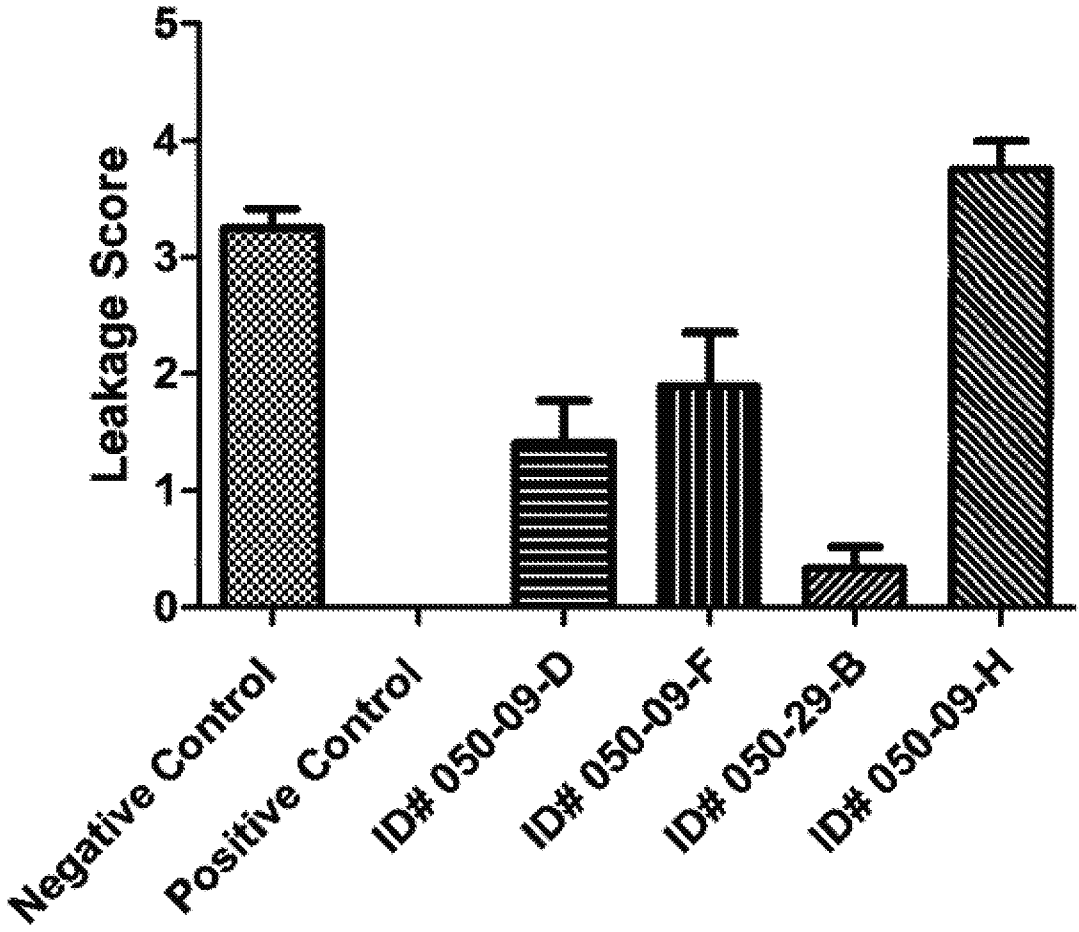

Leakage Score
5
4
3
2
1
0
Negative Control
Positive Control
ID# 050-09-D
ID# 050-09-F
ID# 050-29-B
ID# 050-09-H

FIG. 23

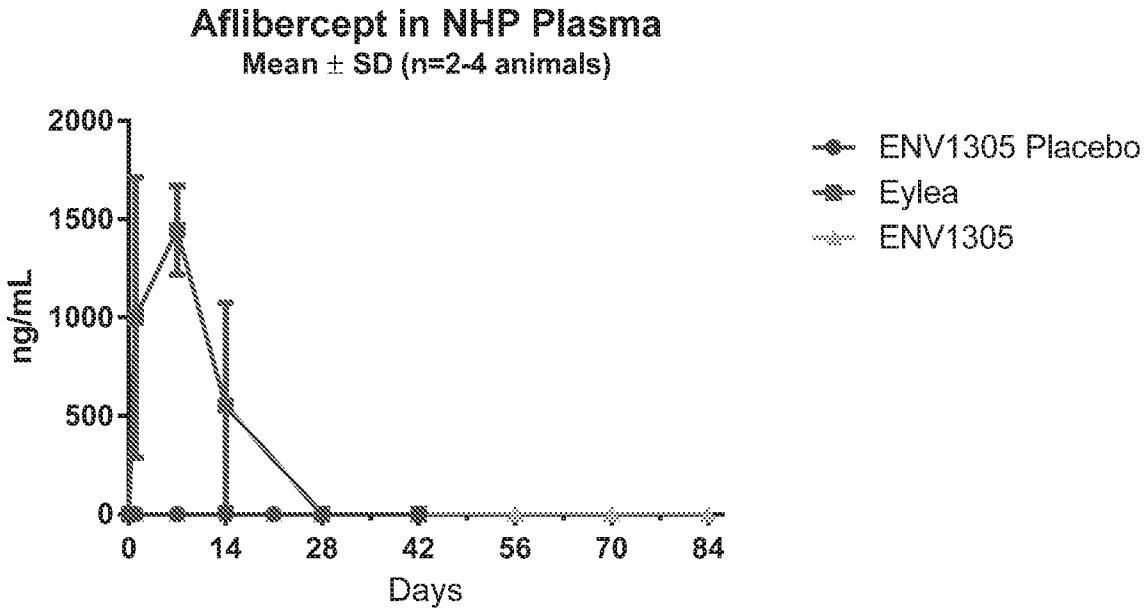

Aflibercept in NHP Plasma
Mean ± SD (n=2-4 animals)
2000
1500
1000
500
0
ng/mL
0
14
28
42
56
70
84
Days
ENV1305 Placebo
Eylea
ENV1305

FIG. 24

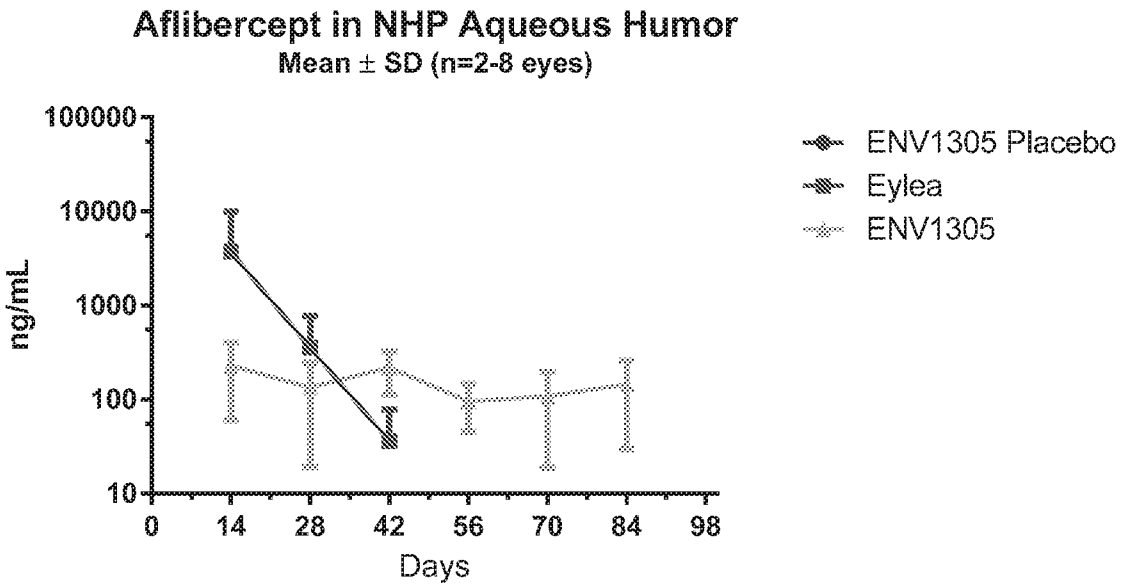

Aflibercept in NHP Aqueous Humor
Mean ± SD (n=2-8 eyes)
100000
10000
1000
100
10
ng/mL
0
14
28
42
56
70
84
98
Days
ENV1305 Placebo
Eylea
ENV1305

FIG. 25

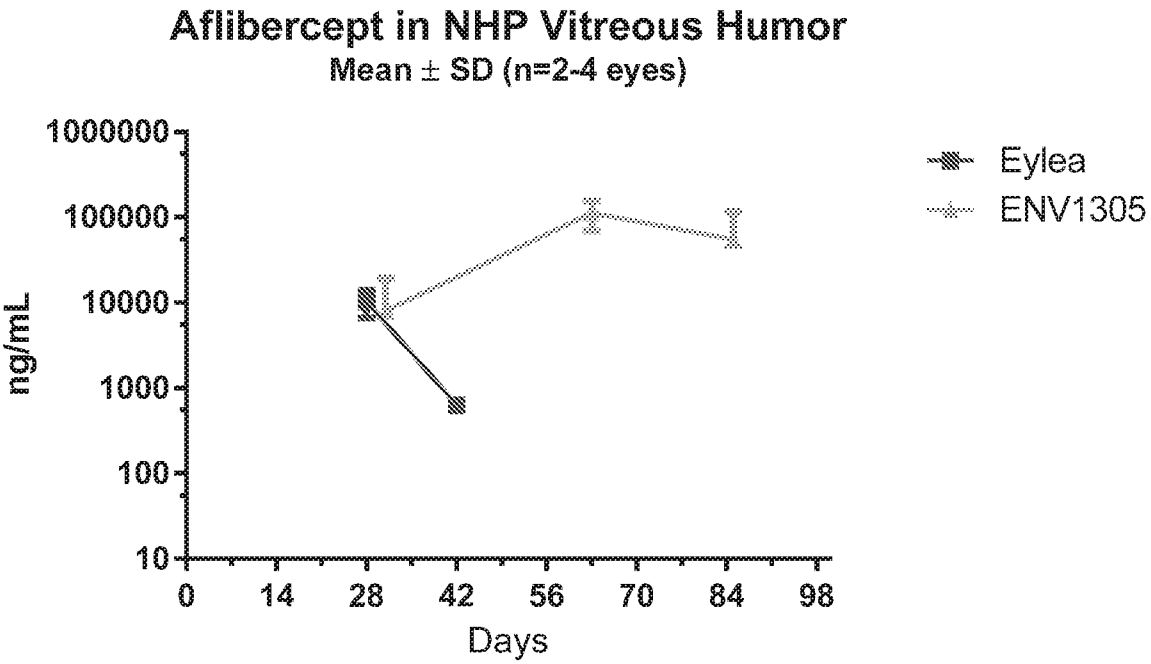

Aflibercept in NHP Vitreous Humor
Mean ± SD (n=2-4 eyes)
1000000
100000
10000
1000
100
10
ng/mL
0
14
28
42
56
70
84
98
Days
Eylea
ENV1305

FIG. 26A

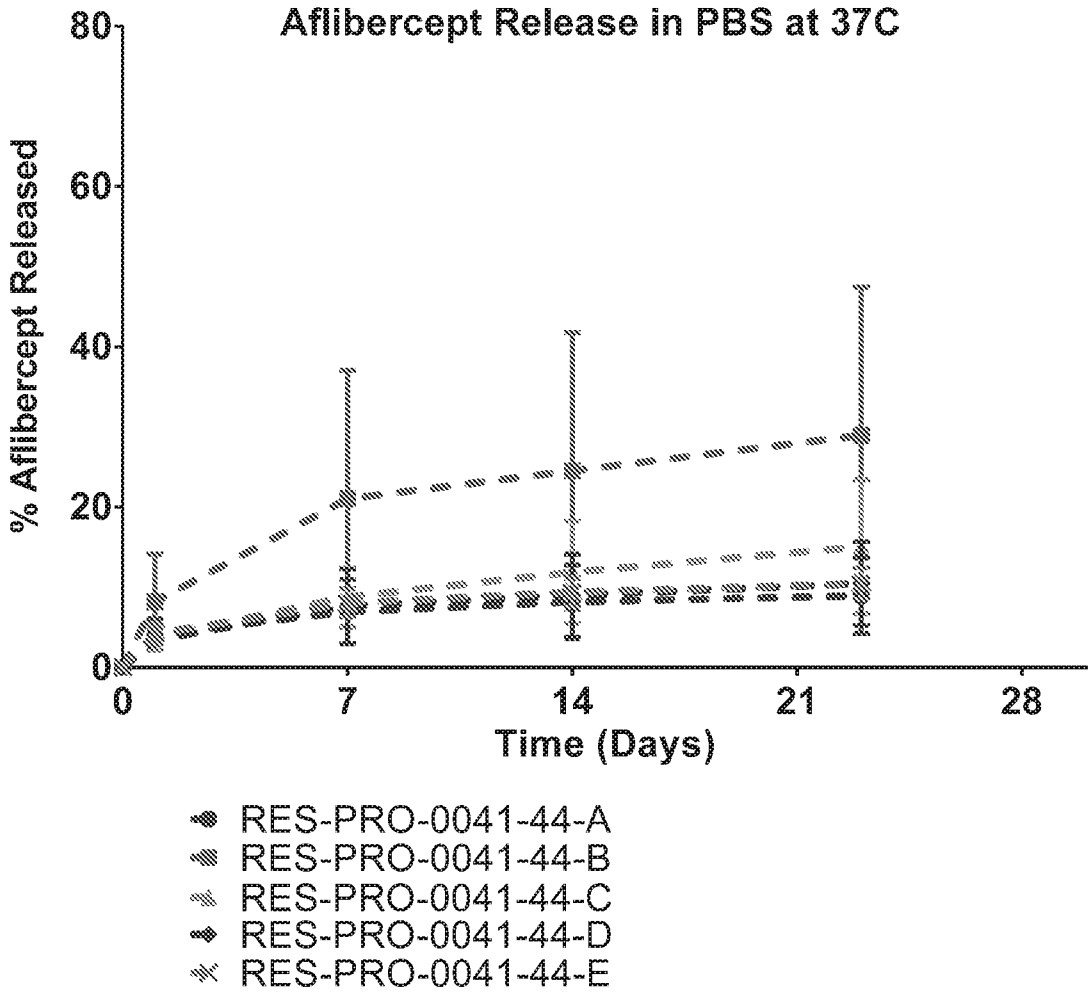

Aflibercept Release in PBS at 37C
% Aflibercept Released
80
60
40
20
0
0
7
14
21
28
Time (Days)
RES-PRO-0041-44-A
RES-PRO-0041-44-B
RES-PRO-0041-44-C
RES-PRO-0041-44-D
RES-PRO-0041-44-E

FIG. 26B

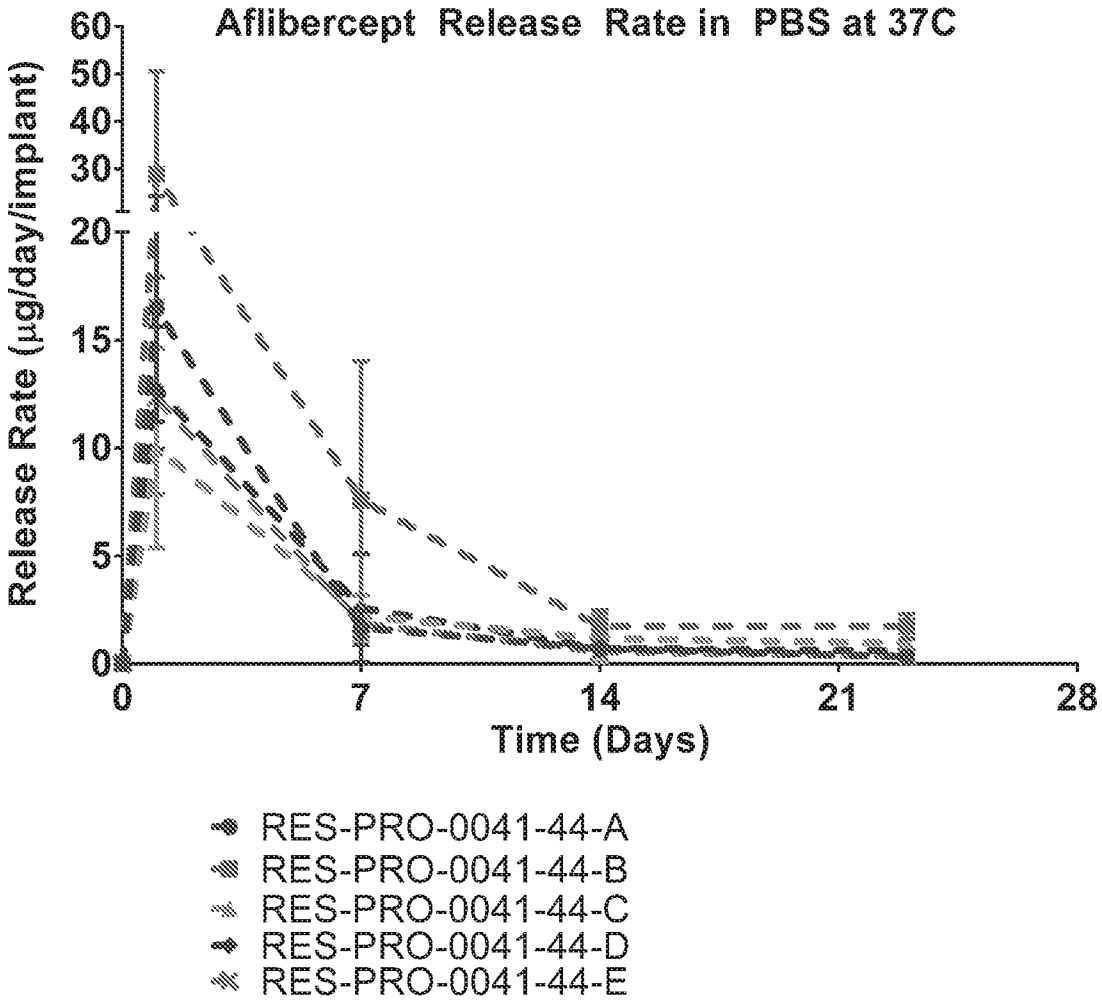
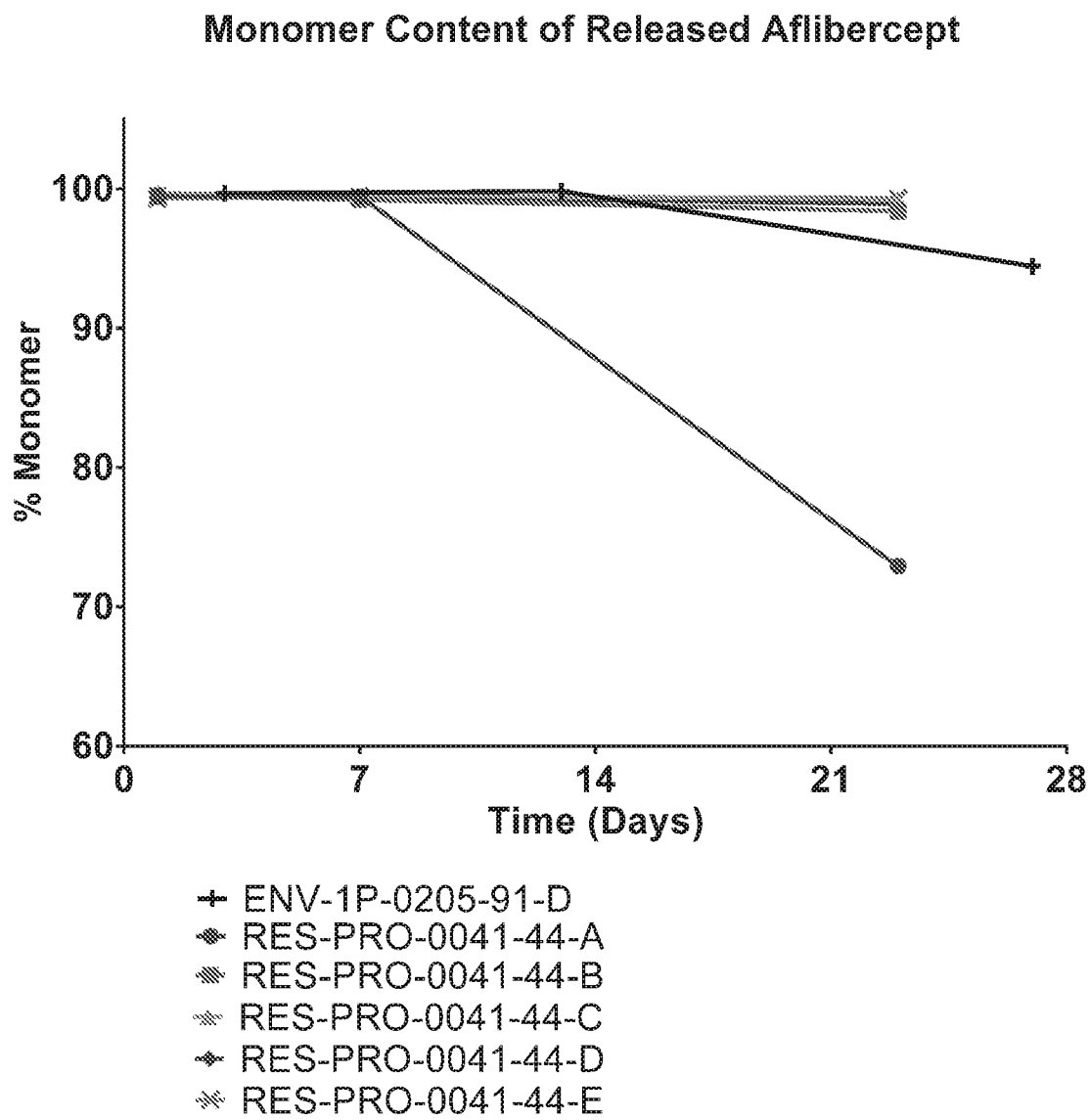

Aflibercept Release Rate in PBS at 37C
Release Rate (µg/day/implant)
60
50
40
30
20
15
10
5
0
0
7
14
21
28
Time (Days)
RES-PRO-0041-44-A
RES-PRO-0041-44-B
RES-PRO-0041-44-C
RES-PRO-0041-44-D
RES-PRO-0041-44-E

FIG. 27


Monomer Content of Released Aflibercept
% Monomer
100
90
80
70
60
0
7
14
21
28
Time (Days)
ENV-1P-0205-91-D
RES-PRO-0041-44-A
RES-PRO-0041-44-B
RES-PRO-0041-44-C
RES-PRO-0041-44-D
RES-PRO-0041-44-E

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 16/43697

A. CLASSIFICATION OF SUBJECT MATTER
IPC(8) - A61K 35/44 (2016.01)
CPC - A61K 35/44; A61B 19/00; A61B 2017/00969
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)
IPC(8) - A61K 35/44 (2016.01)
CPC - A61K 35/44; A61B 19/00; A61B 2017/00969

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched
UC - 424/571

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
Minesoft Patbase, Google Scholar, keywords: making ocular sustained-release pharmaceutical composition therapeutic agent eye excipients excipients gel precursors solvent suspension hydrophilic solvent gel precursors

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X ------ Y	US 2013/0071462 A1 (Jarrett et al.) 21 March 2013 (21.03.2013) para [0005], [0019], [0027], [0030]-[0032], [0037], [0050], [0051] - [0058], [0095], [0097], [0098], [0101], [0111], [0127], [0130], [0131], [0132], [0138]	1-3, 6-10, 16-26, 30, 45-50, 55-63, 70, 71, 74, 75, 80-91, 95, 110, 111, 120-128, 132, 133 ---------- 4, 5, 11-15, 27-29, 31-44, 51-54, 64-69, 72, 73, 76-79, 92-94, 96-109, 112-119, 129-131, 134, 135
Y	US 2011/0264030 A1 (DeSimone et al.) 27 October 2011 (27.10.2011) para [0033], [0054], [0256]	4, 5,12,13,15, 76, 78, 79, 98
Y	US 2006/0257488 A1 (Hubbard) 16 November 2006 (16.11.2006) para [0002], [0033]	11, 13-15, 77
Y	WO 2012/136016 A1 (GUANGZHOU ORACLE PHARMATECH CO., LTD) 11 October 2012 (11.10.2012) (abstract)	27-29, 92-94
Y	US 2007/0053870 A1 (Tae et al.) 08 March 2007 (08.03.2007) para [0020], [0023], [0028]	31, 36, 64- 69, 96, 116-119,129-131
Y	US 2009/0098380 A1 (Henn et al.) 16 April 2009 (16.04.2009) para [0085], [0091]	32-35, 97, 98, 102-104
Y	US 2012/0040397 A1 (Luo et al.) 16 February 2012 (16.02.2012) para [0407]	36-42, 99-101, 105-107,129-131

☒ Further documents are listed in the continuation of Box C. ☐

* 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
10 November 2016	29 NOV 2016
Name and mailing address of the ISA/US Mail Stop PCT, Attn: ISA/US, Commissioner for Patents P.O. Box 1450, Alexandria, Virginia 22313-1450 Facsimile No. 571-273-8300	Authorized officer: Lee W. Young PCT Helpdesk: 571-272-4300 PCT OSP: 571-272-7774

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 16/43697

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	US 2013/0156752 A1 (Jarrett et al.) 20 June 2013 (20.06.2013) para [0005], [0019], [0024], [0025], [0037], claim 37	43, 44, 51-54, 72, 108, 109, 112-119, 134
Y	US 2008/0280975 A1 (Badul) 13 November 2008 (13.11.2008) para [0060], [0076]	73, 135
Y	US 2014/0081012 A1 (DeSimone et al.) 20 March 2014 (20.03.2014) para [0294], [0407]	42, 107

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 16/43697

Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☐ Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:
This application contains the following inventions or groups of inventions which are not so linked as to form a single general inventive concept under PCT Rule 13.1. In order for all inventions to be examined, the appropriate additional examination fees must be paid.

Group I: Claims 1-135 directed a method for making an ocular sustained-release pharmaceutical composition
Group II: Claims 136-183 and 234-238, directed to a sustained release pharmaceutical composit1on
Group III: Claims 184-213, directed to a sustained release delivery system prepared by a process
Group IV: Claim 214-224, directed to a method of treating an ocular condition associated with abnormal vessel growth in the eye
Group V: Claim 225-233, directed to a method of maintaining an ocular vitreous concentration of an anti-Vascular Endothelial Growth Factor (anti-VEGF) agent

----------See supplemental form below ----------

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. ☐ As all searchable claims could be searched without effort justifying additional fees, this Authority did not invite payment of additional fees.

3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☒ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:
1-135

Remark on Protest

☐ The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.

☐ The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.

☐ No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 16/43697

Supplemental Box III (unity of invention)

The inventions listed as Groups I-V do not relate to a single general inventive concept under PCT Rule 13.1 because, under PCT Rule 13.2, they lack the same or corresponding special technical features for the following reasons:

Special Technical Features:

Group I requires a method for making an ocular sustained-release pharmaceutical composition, not required by group II-V.

Group III requires a sustained release delivery system, not required by group I-II and IV-V.

Group IV requires a method of treating an ocular condition associated with abnormal vessel growth in the eye, not required by group I-III and V

Group V requires a method of maintaining an ocular vitreous concentration of an anti-Vascular Endothelial Growth Factor (anti-VEGF) agent, not required by group I-IV

Common Technical Features:

Groups I and II share the technical feature of an ocular sustained-release pharmaceutical composition for the delivery of a therapeutic agent to an eye of a subject in need thereof, comprising: formulating a solution comprising a therapeutic agent and one or more excipients; forming, from the solution, particles comprising the therapeutic agent and one or more excipients; combining the particles with gel precursors in a solvent to form a suspension comprising the particles and the gel precursors, wherein the solvent is an organic and hydrophilic solvent: and allowing the gel precursors to gel around the particles, thereby forming a gel comprising encapsulated particles; and thereby forming the pharmaceutical composition for sustained delivery of a therapeutic agent. However, these shared technical features do not represent a contribution over prior art, because these technical feature is being anticipated by US 2013/0071462 A1 to Jarrett et al. (hereinafter 'Jarrett'). Jarrett discloses a method for making an ocular sustained-release pharmaceutical composition (para [0097], [0111]: intraocular drug delivery; controlled release in the intraocular space) for the delivery of a therapeutic agent to an eye of a subject in need thereof (para [0111]: delivery and retention in the eye), comprising:formulating a solution comprising a therapeutic agent and one or more excipients (para [0130]: Ovalbumin was ground to a fine powder using a mortar and pestle. 200 mg of the ground ovalbumin was added to 800 mg of methyl stearate and heated to 40 degrees centigrade in a 20 mL scintillation vial while stirring with a magnetic stir bar to uniformly suspend the ovalbumin particles. The vial was then cooled to solidify the methyl stearate phase. The solidified mixture was then ground to a powder using a mortar and pestle); forming, from the solution, particles comprising the therapeutic agent and one or more excipients; (para [0130], [0131]: particles and one excipient); combining the particles with gel precursors in a solvent to form a suspension comprising the particles and the gel precursors, wherein the solvent is a hydrophilic solvent:
(para [0132]: The powder from Examples 1, 2 or 3 were combined with a polyethylene glycol (PEG) precursor to form hydrogel samples for testing; para [0127]: The hydrogel particle solvent may be essentially water, meaning about 99 percent v/v of the solvent is water, with salts or buffers being present as desired. Other solvents may be used that are safe and biocompatible, e.g., dimethylsulfoxide. The hydrogel particles may further comprise therapeutic agent-loaded lipophilic particles). and allowing the gel precursors to gel around the particles, thereby forming a gel comprising encapsulated particles (para [0037]: matrices may be prepared and used to encapsulate the particles; hydrogels are an embodiment of such a matrix. Hydrogels are materials that do not dissolve in water and retain a significant fraction (more than 20 percent) of water within their structure; para [0132]: gel precursors around particlesles); and thereby forming the pharmaceutical composition for sustained delivery of a therapeutic agent {para [0138]: Example 8, controlled release).

Groups II-V share the technical feature of a gel comprising a biocompatible polymer matrix; and and at least one protein microparticle (PuP) comprising a therapeutic agent and at least one pharmaceutically acceptable excipient; wherein the gel encapsulates the PuP: wherein the PuP has a largest dimension of less than about 10 ?m; wherein the pharmaceutical composition is formulated to deliver the therapeutic agent for at least about 4 months. However, these shared technical features do not represent a contribution over prior art, because these technical feature is being obvious over US 5,529,914 A to Hubbell et al. (hereinafter ?Hubbell?). Hubbell teaches gel comprising a biocompatible polymer matrix (col 3, ln 11-21; col 10, ln 20-35)); and at least one protein microparticle comprising a therapeutic agent (col 3, ln 32-67); and at least one pharmaceutically acceptable excipient; wherein the gel encapsulates the protein microparticle wherein the pharmaceutical composition is formulated to deliver the therapeutic agent for at least about 4 months (col 3, ln 32-67, col 5, ln 44-50), but does not specifically teach wherein the PuP has a largest dimension of less than about 10 ?m. It would have been obvious to one of skill in the art to optimize protein microparticle size by routine experimentation.

As the shared technical features were known in the art at the time of the invention, they cannot be considered common technical features that would otherwise unify the groups. Therefore, Groups I-V lack unity under PCT Rule 13.