

FULLY-HUMAN POST-TRANSLATIONALLY MODIFIED ANTIBODY THERAPEUTICS

0. SEQUENCE LISTING

[0000] The instant application contains a Sequence Listing which has been submitted electronically in ASCII format and is hereby incorporated by reference in its entirety. Said ASCII copy, created on April 24, 2020, is named 38013_0001P1_SL.txt and is 690,185 bytes in size.

1. INTRODUCTION

[0001] Compositions and methods are described for the delivery of a fully human post-translationally modified (HuPTM) therapeutic monoclonal antibody (“mAb”) or the HuPTM antigen-binding fragment of a therapeutic mAb—*e.g.*, a fully human-glycosylated (HuGly) Fab of the therapeutic mAb—to a human subject diagnosed with a disease or condition indicated for treatment with the therapeutic mAb.

2. BACKGROUND OF THE INVENTION

[0002] Therapeutic mAbs have been shown to be effective in treating a number of diseases and conditions. However, because these agents are effective for only a short period of time, repeated injections for long durations are often required, thereby creating considerable treatment burden for patients.

3. SUMMARY

[0003] Compositions and methods are described for the delivery of a HuPTM mAb or a HuPTM antigen-binding fragment of a therapeutic mAb (for example, a fully human-glycosylated Fab (HuGlyFab) of a therapeutic mAb) to a patient (human subject) diagnosed with a disease or condition indicated for treatment with the therapeutic mAb. Such antigen-binding fragments of therapeutic mAbs include a Fab, F(ab')₂, or scFv (single-chain variable fragment) (collectively referred to herein as “antigen-binding fragment”). “HuPTM Fab” as used herein may include other antigen binding fragments of a mAb. In an alternative example, full-length mAbs can be used. Delivery may be advantageously accomplished via gene therapy—*e.g.*, by administering a viral vector or other DNA expression construct encoding a therapeutic mAb or its antigen-binding fragment (or a hyperglycosylated derivative of either) to a patient (human subject) diagnosed with a condition indicated for treatment with the therapeutic mAb—to create a permanent depot in a tissue or organ of the patient that continuously supplies the HuPTM mAb or antigen-binding fragment of the therapeutic

[0006] Gene therapy constructs are designed such that both the heavy and light chains are expressed. The coding sequences for the heavy and light chains can be engineered in a single construct in which the heavy and light chains are separated by a cleavable linker or IRES so that separate heavy and light chain polypeptides are expressed. In certain examples, the coding sequences encode for a Fab or F(ab')₂ or an scFv. In certain examples the full length heavy and light chains of the antibody are expressed. In other examples, the constructs express an scFv in which the heavy and light chain variable domains are connected via a flexible, non-cleavable linker. In certain examples, the construct expresses, from the N-terminus, NH₂-V_L-linker-V_H-COOH or NH₂-V_H-linker-V_L-COOH.

[0007] Therapeutic antibodies delivered by gene therapy have several advantages over injected or infused therapeutic antibodies that dissipate over time resulting in peak and trough levels. Sustained expression of the transgene product antibody, as opposed to injecting an antibody repeatedly, allows for a more consistent level of antibody to be present at the site of action, and is less risky and more convenient for patients, since fewer injections need to be made. Furthermore, antibodies expressed from transgenes are post-translationally modified in a different manner than those that are directly injected because of the different microenvironment present during and after translation. Without being bound by any particular theory, this results in antibodies that have different diffusion, bioactivity, distribution, affinity, pharmacokinetic, and immunogenicity characteristics, such that the antibodies delivered to the site of action are “biobetters” in comparison with directly injected antibodies.

[0008] In addition, antibodies expressed from transgenes in vivo are not likely to contain degradation products associated with antibodies produced by recombinant technologies, such as protein aggregation and protein oxidation. Aggregation is an issue associated with protein production and storage due to high protein concentration, surface interaction with manufacturing equipment and containers, and purification with certain buffer systems. These conditions, which promote aggregation, do not exist in transgene expression in gene therapy. Oxidation, such as methionine, tryptophan, and histidine oxidation, is also associated with protein production and storage, and is caused by stressed cell culture conditions, metal and air contact, and impurities in buffers and excipients. The proteins expressed from transgenes in vivo may also oxidize in a stressed condition. However, humans, and many other organisms, are equipped with an antioxidation defense system, which not only reduces the

oxidation stress, but sometimes also repairs and/or reverses the oxidation. Thus, proteins produced in vivo are not likely to be in an oxidized form. Both aggregation and oxidation could affect the potency, pharmacokinetics (clearance), and immunogenicity.

[0009] Pharmaceutical compositions suitable for administration to human subjects comprise a suspension of the recombinant vector in a formulation buffer comprising a physiologically compatible aqueous buffer, a surfactant and optional excipients.

[0010] The disclosure is based, in part, on the following principles:

- (i) The mAb therapeutics currently on the market are of the immunoglobulin G (IgG) isotypes, such as IgG1, IgG2, and IgG4, which in general have pharmacokinetic (PK) characteristics, such as slow clearance, long half-life, and limited tissue distribution. After intravenous administration, typical mAb serum PK profiles are biphasic with a rapid distribution phase and a slower elimination phase; thus, repeat administration is required to maintain doses required to treat chronic conditions. Moreover, the distribution of mAbs is generally limited to the vascular and interstitial spaces due to their large size and hydrophilicity. The extent of mAb partitioning from circulation into most tissues generally ranges from about 5–15%, except for brain where it is much lower. (*See, e.g.*, Kamath, 2016, Drug Discovery Today: Technologies 21-22: 75-83, which is incorporated by reference herein in its entirety). Continuous production of HuPTMmAbs or HuPTM Fabs *in situ* avoids repeat administrations and allows the use of Fabs, which would otherwise have too short a systemic half-life to achieve efficacy; and the methods of administration described allow direct access to target tissues, such as the brain, where the delivery of higher doses to such tissues can be achieved.
- (ii) The Fab region of a number of therapeutic mAbs possesses glycosylation sites. For example, see FIGS. 2A-2C, 3, 4A-4C, 5, 6A-6C, 7A-7B, 8A-8C, 9A-9C, 10A-10D, 11, 12A-12C, 13, 14A-14B, 15, 16A-16I, 17, 18, 19, and 29A-29F which identify consensus and non-consensus asparaginal (“N”) glycosylation sites as well as glutamine (“Q”) residues that are glycosylation sites in the Fab region of certain therapeutic mAbs. (*See, e.g.*, Valliere-Douglass et al., 2009, J. Biol. Chem. 284: 32493-32506, and Valliere-

the addition of glycans that can improve stability, half-life and reduce unwanted aggregation and/or immunogenicity of the transgene product. (See, *e.g.*, Bovenkamp et al., 2016, J. Immunol. 196: 1435-1441 for a review of the emerging importance of Fab glycosylation; and FIG. 22 which identifies glycans that can be attached to HuGlyFab (adapted from Bondt et al., 2014, Mol & Cell Proteomics 13.1: 3029-2029)). The Fab and Fc portions of antibodies have been shown to have distinct glycosylation patterns, with Fab glycans being high in galactosylation, sialylation, and bisection (*e.g.*, with bisecting GlcNAc) but low in fucosylation with respect to Fc glycans. (*E.g.*, see Bondt et al., 2014, Mol. & Cell. Proteomics 13.11:3029-3039, incorporated by reference herein in its entirety for its disclosure of Fab-associated N-glycans).

- (v) Significantly, glycans that are added to HuPTM mAb and HuGlyFab of the disclosure are highly processed complex-type N-glycans that contain 2,6-sialic acid. Such glycans are not present in (a) therapeutic mAbs produced in *E. coli* (which are not glycosylated at all); (b) in therapeutic antibodies produced in CHO cells that do not have the 2,6-sialyltransferase required to add 2,6-sialic acid during glycosylation; or (c) in therapeutic antibodies produced in either CHO or murine cell lines that add N-Glycolylneuraminic acid (“Neu5Gc” or “NeuGc”) which is not natural to humans (and potentially immunogenic), instead of N-Acetylneuraminic acid (“Neu5Ac”) the predominant human sialic acid. See, *e.g.*, Dumont et al., 2015, Crit. Rev. Biotechnol. 36(6):1110-1122; Huang et al., 2006, Anal. Biochem. 349:197-207 (NeuGc is the predominant sialic acid in murine cell lines such as SP2/0 and NS0); and Song et al., 2014, Anal. Chem. 86:5661-5666, each of which is incorporated by reference herein in its entirety.
- (vi) The human glycosylation pattern of the HuPTM mAb and HuGlyFab of the disclosure should reduce immunogenicity of the transgene product and improve efficacy. Importantly, when the full-length antibodies and antigen-binding fragments, used in accordance with the methods described herein are expressed in human target cells, the need for *in vitro* production in prokaryotic host cells (*e.g.*, *E. coli*) or eukaryotic host cells (*e.g.*, CHO cells or murine NS0 or SP2/0 cells) is circumvented. Instead, as a result of the methods described herein (*e.g.*, use of human target cells to express the antigen-binding

construct for the HuPTMmAb or HuPTM Fab should include a signal peptide that ensures proper co- and post-translational processing (glycosylation and protein sulfation) by the transduced human cells.

[0012] As an alternative, or an additional treatment to gene therapy, the full-length HuTPM mAb or HuPTM Fab can be produced in human cell lines by recombinant DNA technology, and the glycoprotein can be administered to patients.

[0013] Combination therapies involving delivery of the full-length HuPTM mAb or HuPTM Fab to the patient accompanied by administration of other available treatments are encompassed by the methods provided herein. The additional treatments may be administered before, concurrently or subsequent to the gene therapy treatment. Such additional treatments can include but are not limited to co-therapy with the therapeutic mAb.

[0014] Also provided are methods of manufacturing the viral vectors, particularly the AAV based viral vectors. In specific examples, provided are methods of producing recombinant AAVs comprising culturing a host cell containing an artificial genome comprising a cis expression cassette flanked by AAV ITRs, wherein the cis expression cassette comprises a transgene encoding a therapeutic antibody operably linked to expression control elements that will control expression of the transgene in human cells; a trans expression cassette lacking AAV ITRs, wherein the trans expression cassette encodes an AAV rep and capsid protein operably linked to expression control elements that drive expression of the AAV rep and capsid proteins in the host cell in culture and supply the rep and cap proteins in trans; sufficient adenovirus helper functions to permit replication and packaging of the artificial genome by the AAV capsid proteins; and recovering recombinant AAV encapsidating the artificial genome from the cell culture.

[0015] The inventors have also found that full length antibodies can be expressed from AAV based vectors (see Examples 36 and 37). The nucleotide sequence encoding the heavy and light chains of the full-length antibodies may be codon optimized for expression in human cells and may have reduced numbers of CpG dimers in the sequence. Accordingly, provided are compositions comprising AAV vectors that express a transgene encoding a full-length heavy chain (including an Fc domain) and light chain of a therapeutic antibody. Methods of administration and manufacture are also provided.

3.1 ILLUSTRATIVE EXAMPLES

Compositions of Matter

1. A pharmaceutical composition for treating Alzheimer's disease (AD), frontotemporal dementia (FD), tauopathies, progressive supranuclear palsy, chronic traumatic encephalopathy, Pick's Complex, and primary age-related tauopathy, Huntington's disease, juvenile Huntington's disease, Parkinson's disease, synucleinopathies, ALS, migraines, or cluster headaches in a human subject in need thereof, comprising an adeno-associated virus (AAV) vector having:

- (a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143), AAV9 capsid (SEQ ID NO: 144), AAVrh10 capsid (SEQ ID NO: 145), an AAVrh20 capsid, an AAVrh39 capsid or an AAVcy5 capsid; and
- (b) an artificial genome comprising an expression cassette flanked by AAV inverted terminal repeats (ITRs), wherein the expression cassette comprises a transgene encoding a substantially full-length or full-length mAb of an anti-amyloid beta (anti-A β), anti-sortilin, anti-Tau protein (anti-Tau), anti-semaphorin 4D (anti-SEMA4D), anti-alpha synuclein (anti-SNCA), anti-superoxide dismutase-1 (anti-SOD1) or anti-calcitonin gene-related peptide receptor (anti-CGRPR) monoclonal antibody (mAb), or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human CNS cells, human liver cells and/or human muscle cells;

wherein said AAV vector is formulated for administration to said subject, optionally wherein administration is intrathecal, intravenous, subcutaneous, intranasal, or intramuscular.

2. The pharmaceutical composition of paragraph 1, wherein the anti-A β mAb is solanezumab, lecanemab, or GSK933776; the anti-sortilin mAb is AL-001; the anti-Tau mAb is ABBV-8E12, UCB-0107, or NI-105 (BIIB076); the anti-SEMA4D mAb is VX15/2503; the anti-SNCA mAb is prasinezumab, NI-202 (BIIB054), or MED-1341; the anti-SOD1 mAb is NI-2041.10D12 or NI-204.12G7; and the anti-CGRPR mAb is eptinezumab, fremanezumab, or galcanezumab.

367. The pharmaceutical composition of any one of paragraphs 364 to 366, wherein the AAV viral capsid is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143), an AAV9 capsid (SEQ ID NO: 144), an AAVrh10 capsid (SEQ ID NO: 145).

368. The pharmaceutical composition of any of paragraphs 364 to 3673, wherein the antibody is sevacizumab, LKA-651, ravulizumab, adalimumab, infliximab, golimumab, elezanumab, NI-301, PRX-004, pamrevlumab, siltuximab, clazakizumab, sirukumab, olokizumab, gerilimzumab, satralizumab, sarilumab, tocilizumab, inebilizumab, etrolizumab, romosozumab, lanadelumab, benralizumab, reslizumab, tralokinumab, nemolizumab, omalizumab, or tezepelumab.

369. The pharmaceutical composition of any of paragraphs 364 to 366, wherein the transgene comprises nucleotide sequence SEQ ID NO; 443.

In one aspect, the present disclosure refers to a pharmaceutical composition for treating non-infectious uveitis in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV2.7m8 (SEQ ID NO: 142), an AAV8 capsid (SEQ ID NO: 143), or AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a substantially full-length or full-length anti-tumor necrosis factor-alpha (anti-TNF α) mAb or an antigen-binding fragment thereof, a substantially full-length or full-length anti-complement component 5 (C5) mAb or an antigen-binding fragment thereof, a substantially full-length or full-length anti-interleukin-6 (IL-6) mAb or an antigen-binding fragment thereof, or a substantially full-length or full-length anti-interleukin-6 receptor (IL-6R) mAb or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human retina cells;

wherein said AAV vector is formulated for subretinal, intravitreal, intranasal, or suprachoroidal administration to the subject.

In another aspect, the present disclosure refers to a polynucleotide having the nucleotide sequence of SEQ ID NO: 451 or 453 flanked by AAV ITR nucleotide sequences.

In another aspect, the present disclosure refers to a recombinant AAV comprising an artificial genome comprising the polynucleotide of claim 17 and an AAV8 capsid.

In another aspect, the present disclosure refers to a method of producing recombinant AAVs comprising:

(a) culturing a host cell containing:

(i) an artificial genome comprising a *cis* expression cassette flanked by AAV ITRs, wherein the *cis* expression cassette comprises a transgene encoding adalimumab or an antigen binding fragment thereof operably linked to expression control elements that will control expression of the transgene in human cells;

(ii) a *trans* expression cassette lacking AAV ITRs, wherein the *trans* expression cassette encodes an AAV rep and an AAV8 capsid protein operably linked to expression control elements that drive expression of the AAV rep and the AAV capsid protein in the host cell in culture and supply the AAV rep and the AAV capsid protein in *trans*; and

(iii) sufficient adenovirus helper functions to permit replication and packaging of the artificial genome by the AAV capsid protein; and

(b) recovering recombinant AAV encapsidating the artificial genome from the cell culture.

In another aspect, the present disclosure refers to a host cell comprising

(a) an artificial genome comprising a *cis* expression cassette flanked by AAV ITRs, wherein the *cis* expression cassette comprises a transgene encoding adalimumab or an antigen binding fragment thereof operably linked to expression control elements that will control expression of the transgene in human cells;

(b) a *trans* expression cassette lacking AAV ITRs, wherein the *trans* expression cassette encodes an AAV rep and an AAV8 capsid protein operably linked to expression control elements that drive expression of the AAV rep and the AAV capsid protein in the host cell in culture and supply the AAV rep and the AAV capsid protein in *trans*; and

(c) sufficient adenovirus helper functions to permit replication and packaging of the artificial genome by the AAV capsid protein.

In another aspect, the present disclosure refers to a pharmaceutical composition for treating angioedema including hereditary angioedema in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of AAV8 capsid (SEQ ID NO: 143); AAVrh10 capsid (SEQ ID NO: 145); or AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-kallikrein (anti-pKal) mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for administration to the subject, optionally wherein administration is intravenous, subcutaneous, intranasal, or intramuscular.

In another aspect, the present disclosure refers to use of a pharmaceutical composition for delivering lanadelumab to the bloodstream in the manufacture of a medicament for treating hereditary angioedema in a human subject in need thereof, said composition comprising a recombinant AAV comprising a transgene encoding lanadelumab operably linked to one or more regulatory sequences that control expression of the transgene in muscle and/or liver cells, wherein said recombinant AAV is to be administered to said human subject at a dose sufficient to result in expression from the transgene and secretion of lanadelumab into the bloodstream of the human subject to produce lanadelumab plasma levels of at least 5 µg/ml to at least 35 µg/ml lanadelumab in said subject.

In another aspect, the present disclosure refers to a pharmaceutical composition for delivery of an antibody or antigen binding fragment thereof to the bloodstream of a human subject in need thereof, comprising an AAV vector comprising:

(a) An AAV viral capsid that infects liver and/or muscle cells; and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a full-length antibody or an antigen-binding fragment thereof, operably linked to a chimeric promoter that directs expression in muscle and liver cells;

wherein said AAV vector is formulated for intramuscular administration.

In another aspect, the present disclosure refers to a pharmaceutical composition for treating Alzheimer's disease, frontotemporal dementia (FD), tauopathies, progressive supranuclear palsy, chronic traumatic encephalopathy, Pick's Complex, and primary age-related tauopathy, Huntington's disease, juvenile Huntington's disease, Parkinson's disease, synucleinopathies, ALS, migraines, or cluster headaches in a human subject in need thereof, comprising an adeno-associated virus (AAV) vector having:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143), AAV9 capsid (SEQ ID NO: 144), AAVrh10 capsid (SEQ ID NO: 145), an AAVrh20 capsid, an AAVrh39 capsid or an AAVcy5 capsid; and

(b) an artificial genome comprising an expression cassette flanked by AAV inverted terminal repeats (ITRs), wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-amyloid beta (anti-A β), anti-sortilin, anti-Tau protein (anti-Tau), anti-semaphorin 4D (anti-SEMA4D), anti-alpha synuclein (anti-SNCA), anti-superoxide dismutase-1 (anti-SOD1) or anti-calcitonin gene-related peptide receptor (anti-CGRPR) mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human CNS, muscle, or liver cells;

wherein said AAV vector is formulated for administration to the subject, optionally wherein administration is intrathecal, intravenous, subcutaneous, intranasal, or intramuscular.

In another aspect, the present disclosure refers to a pharmaceutical composition for treating retinal disorders including diabetic retinopathy, myopic choroidal neovascularization (mCNV), macular degeneration (e.g., neovascular (wet) or dry age-related macular degeneration (nAMD)), macular edema (e.g., macular edema following a retinal vein occlusion (RVO) or diabetic macular edema (DME)), retinal vein occlusion, diabetic retinopathy (DR), non-infectious uveitis, or glaucoma, or

abnormal vascularization of the retina in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV2.7m8 capsid (142), an AAV8 capsid (SEQ ID NO: 143), or an AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-vascular endothelial growth factor (anti-VEGF), anti-erythropoietin receptor (anti-EPOR), anti-A β , anti-activin receptor like kinase 1 (anti-ALK1), anti-complement component 5 (anti-C5), anti-endoglin (anti-ENG), anti-complement component 1Q (anti-CC1Q), anti-tumor necrosis factor-alpha (anti-TNF α), or anti-pKal mAb or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human retinal cells;

wherein said AAV vector is formulated for subretinal, intravitreal, suprachoroidal, or intranasal administration to said subject.

In another aspect, the present disclosure refers to a pharmaceutical composition for treating multiple sclerosis in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143), AAV9 capsid (SEQ ID NO: 144), AAVrh10 capsid (SEQ ID NO: 145), an AAVrh20 capsid, an AAVrh39 capsid or an AAVcy5 capsid; and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-repulsive guidance molecule-A (anti-RGMA) mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human CNS, liver, or muscle cells;

wherein said AAV vector is formulated for administration to the subject, optionally wherein administration is intrathecal, intravenous, subcutaneous, intranasal, or intramuscular.

In another aspect, the present disclosure refers to a pharmaceutical composition for treating amyloidosis (ATTR), familial amyloid cardiomyopathy (FAC), or familial amyloid polyneuropathy (FAP) in a human subject in need thereof, comprising AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143), an AAV9 capsid (SEQ ID NO: 144), an AAVrh10 capsid (SEQ ID NO: 145), an AAVrh20 capsid, an AAVrh39 capsid, or an AAVcy5 capsid; and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-TTR mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for administration to the subject, optionally wherein administration is intravenous, subcutaneous, intranasal, or intramuscular.

In another aspect, the present disclosure refers to a pharmaceutical composition for treating fibrotic disorders, pulmonary fibrosis, cystic fibrosis (CF), idiopathic pulmonary fibrosis (IPF), liver cirrhosis, atrial fibrosis, endomyocardial fibrosis, old myocardial infarction, arthrofibrosis, Crohn's disease, ulcerative colitis, mediastinal fibrosis, myelofibrosis (MF), nephrogenic systemic fibrosis (NSF), progressive massive fibrosis (PMF), and retroperitoneal fibrosis (RPF) in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143), an AAV9 capsid (SEQ ID NO: 144), or AAVrh10 (SEQ ID NO: 145); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-connective tissue growth factor (anti-CTGF) mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for administration to said subject, optionally wherein administration is intravenous, subcutaneous, intranasal, or intramuscular.

In another aspect, the present disclosure refers to a pharmaceutical composition for treating non-infectious uveitis, neuromyelitis optica (NMO), diabetic retinopathy (DR), or diabetic macular edema (DME) in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143), an AAV2.7m8 capsid (SEQ ID NO: 142) or an AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV inverted terminal repeats (ITRs) wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-interleukin-6 receptor (anti-IL6R), anti-interleukin 6 (IL6), or anti-cluster of differentiation 19 (anti-CD19) mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human retinal cells;

wherein said AAV vector is formulated for administration to said subject, optionally wherein administration is intranasal, subretinal, intravitreal, or suprachoroidal.

In another aspect, the present disclosure refers to a pharmaceutical composition for reducing, inhibiting or ameliorating a detrimental immune response in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143), an AAV9 capsid (SEQ ID NO: 144), an AAVrh10 capsid (SEQ ID NO: 145); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a substantially full-length or full-length mAb of an anti-interleukin-6 receptor (anti-IL6R) or anti-interleukin-6 (IL6), or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or muscle cells;

wherein said AAV vector is formulated for subcutaneous, intramuscular, intravenous or pulmonary administration to the subject.

In another aspect, the present disclosure refers to a pharmaceutical composition for treating inflammatory bowel disease (IBD) including UC and CD in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of AAV8 capsid (SEQ ID NO: 143); AAV9 capsid (SEQ ID NO: 144); or AAVrh10 capsid (SEQ ID NO: 145); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-integrin $\beta 7$ subunit (anti-ITGB7) mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for administration to the subject, optionally wherein administration is intravenous, subcutaneous, intranasal, or intramuscular.

In another aspect, the present disclosure refers to a pharmaceutical composition for treating osteoporosis or abnormal bone loss or weakness (e.g., treating giant cell tumor of bone, treating treatment-induced bone loss, slowing the loss of (or increasing) bone mass in breast and prostate cancer patients, preventing skeletal-related events due to bone metastasis, or for decreasing bone resorption and turnover in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143); AAVrh10 capsid (SEQ ID NO: 145); or an AAV9 capsid (SEQ ID NO: 144);

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-sclerostin (anti-SOST) mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for administration to the subject, optionally wherein administration is intravenous, subcutaneous, intranasal, or intramuscular.

In another aspect, the present disclosure refers to a pharmaceutical composition for treating atopic dermatitis in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143) or AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding an anti-IL13 mAb or anti-IL31RA, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for intravenous administration to the liver or muscle of said subject.

In another aspect, the present disclosure refers to a pharmaceutical composition for treating eosinophilic asthma in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143) or an AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding an anti-IL5R mAb or anti-IgE mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for intravenous administration to the liver or muscle of said subject.

In another aspect, the present disclosure refers to a pharmaceutical composition for treating asthma or chronic obstructive pulmonary disease (COPD) in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143) or an AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding an anti-IL5, anti-IL-5R, anti-IgE, or anti-TSLP mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for intravenous administration to the liver or muscle of said subject.

In another aspect, the present disclosure refers to a pharmaceutical composition for treating chronic idiopathic urticaria in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO:143) or an AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding an anti-IgE mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for intravenous administration to the liver or muscle of said subject.

In another aspect, the present disclosure refers to a pharmaceutical composition for treating myasthenia gravis in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143) or AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding an anti-C5 mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for intravenous administration to the liver or muscle of said subject.

In another aspect, the present disclosure refers to a method of determining human anti-pKal antibody activity in a sample, said method comprising

- a. Incubating the sample with activated human pKal;
- b. Subsequently incubating the sample having been incubated with the activated human pKal with the synthetic substrate Pro-Phe-Arg-AMC
- c. Measure the release of AMC over three hours as compared to a control sample.

4. BRIEF DESCRIPTION OF THE DRAWINGS

[0016] **FIG. 1.** A schematic of an rAAV vector genome construct containing an expression cassette encoding the heavy and light chains of the Fab region of a therapeutic mAb controlled by expression elements, flanked by the AAV ITRs.

[0017] **FIGS. 2A-C.** The amino acid sequence of a transgene construct for the Fab region of therapeutic antibodies to amyloid β peptides: solanezumab Fab (**FIG. 2A**), GSK933776 (**FIG. 2B**) and lecanemab (**FIG. 2C**). Glycosylation sites are boldface. Glutamine glycosylation sites; asparaginal (N) glycosylation sites, non-consensus asparaginal (N) glycosylation sites; and tyrosine-O-sulfation sites (italics) are as indicated in the legend. Complementarity-determining regions (CDR) are underscored. The heavy chain hinge regions are highlighted in grey.

[0018] **FIG. 3.** The amino acid sequence of a transgene construct for the Fab region of therapeutic antibodies to sortilin: AL-001 (**FIG. 3**). Glycosylation sites are boldface. Glutamine glycosylation sites; asparaginal (N) glycosylation sites, non-consensus asparaginal (N) glycosylation sites; and tyrosine-O-sulfation sites (italics) are as indicated in the legend. Complementarity-determining regions (CDR) are underscored. The heavy chain hinge regions are highlighted in grey.

[0019] **FIGS. 4A-4C.** The amino acid sequence of a transgene construct for the Fab region of therapeutic antibodies to tau: ABBV-8E12 (**FIG. 4A**), UCB-0107 (**FIG. 4B**), and NI-105 (**FIG. 4C**).

What is claimed is:

1. A pharmaceutical composition for treating non-infectious uveitis in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV2.7m8 (SEQ ID NO: 142), an AAV8 capsid (SEQ ID NO: 143), or AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a substantially full-length or full-length anti-tumor necrosis factor-alpha (anti-TNF α) mAb or an antigen-binding fragment thereof, a substantially full-length or full-length anti-complement component 5 (C5) mAb or an antigen-binding fragment thereof, a substantially full-length or full-length anti-interleukin-6 (IL-6) mAb or an antigen-binding fragment thereof, or a substantially full-length or full-length anti-interleukin-6 receptor (IL-6R) mAb or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human retina cells;

wherein said AAV vector is formulated for subretinal, intravitreal, intranasal, or suprachoroidal administration to the subject.

2. The pharmaceutical composition of claim 1 wherein the anti-TNF α mAb is adalimumab, infliximab or golimumab; the anti-C5 mAb is tesidolumab or ravulizumab; the anti-IL-6 mAb is siltuximab, clazakimzumab, sirukumab, olokizumab or gerilimzumab; or the anti-IL-6R mAb is satralizumab, sarilumab or tocilizumab.

3. The pharmaceutical composition of claim 1 or 2, wherein the antigen-binding fragment is a Fab, a F(ab')₂, or an scFv.

4. The pharmaceutical composition of any of claims 1 to 3, wherein the full-length mAb or the antigen-binding fragment comprises a heavy chain with an amino acid sequence of SEQ ID NO: 45 and optionally an Fc polypeptide with an amino acid sequence of SEQ ID NO: 303 and a light chain with an amino acid sequence of SEQ ID NO: 46; or a heavy chain with an amino acid sequence of SEQ ID NO: 47 and optionally an Fc polypeptide with an amino acid sequence of SEQ ID NO: 304 and a light chain with an amino acid sequence of SEQ ID NO: 48; or a heavy chain with an amino

acid sequence of SEQ ID NO: 49 and optionally an Fc polypeptide with an amino acid sequence of SEQ ID NO: 305 and a light chain with an amino acid sequence of SEQ ID NO: 50; a heavy chain with an amino acid sequence of SEQ ID NO: 39 and optionally an Fc polypeptide with an amino acid sequence of SEQ ID NO: 301 and a light chain with an amino acid sequence of SEQ ID NO: 40; a heavy chain with an amino acid sequence of SEQ ID NO: 362 and optionally an Fc polypeptide with an amino acid sequence of SEQ ID NO: 393 and a light chain with an amino acid sequence of SEQ ID NO: 363; a heavy chain with an amino acid sequence of SEQ ID NO: 331 and optionally an Fc polypeptide with an amino acid sequence of SEQ ID NO: 355 and a light chain with an amino acid sequence of SEQ ID NO: 332; a heavy chain with an amino acid sequence of SEQ ID NO: 333 and optionally an Fc polypeptide with an amino acid sequence of SEQ ID NO: 356 and a light chain with an amino acid sequence of SEQ ID NO: 334; a heavy chain with an amino acid sequence of SEQ ID NO: 335 and optionally an Fc polypeptide with an amino acid sequence of SEQ ID NO: 357 and a light chain with an amino acid sequence of SEQ ID NO: 336; a heavy chain with an amino acid sequence of SEQ ID NO: 337 and optionally an Fc polypeptide with an amino acid sequence of SEQ ID NO: 358 and a light chain with an amino acid sequence of SEQ ID NO: 338; a heavy chain with an amino acid sequence of SEQ ID NO: 339 and a light chain with an amino acid sequence of SEQ ID NO: 340; a heavy chain with an amino acid sequence of SEQ ID NO: 59 and optionally an Fc polypeptide with an amino acid sequence of SEQ ID NO: 309 and a light chain with an amino acid sequence of SEQ ID NO: 60; a heavy chain with an amino acid sequence of SEQ ID NO: 61 and optionally an Fc polypeptide with an amino acid sequence of SEQ ID NO: 310 and a light chain with an amino acid sequence of SEQ ID NO: 62; and a heavy chain with an amino acid sequence of SEQ ID NO: 341 and optionally an Fc polypeptide with an amino acid sequence of SEQ ID NO: 359 and a light chain with an amino acid sequence of SEQ ID NO: 342.

5. The pharmaceutical composition of claim 4, wherein the transgene comprises a nucleotide sequence of SEQ ID NO: 115, 444 or 445 encoding the heavy chain and a nucleotide sequence of SEQ ID NO: 116, 448 or 449 encoding the light chain; or a nucleotide sequence of SEQ ID NO: 117 encoding the heavy chain and a nucleotide sequence of SEQ ID NO: 118 encoding the light chain; a nucleotide sequence of SEQ ID NO: 119 encoding the heavy chain and a nucleotide sequence of SEQ ID NO: 120 encoding the light chain; nucleotide sequence of SEQ ID NO: 109 encoding the heavy

chain and a nucleotide sequence of SEQ ID NO: 110 encoding the light chain; or a nucleotide sequence of SEQ ID NO: 378 encoding the heavy chain and a nucleotide sequence of SEQ ID NO: 379 encoding the light chain; a nucleotide sequence of SEQ ID NO: 343 encoding the heavy chain and a nucleotide sequence of SEQ ID NO: 344 encoding the light chain; nucleotide sequence of SEQ ID NO: 345 encoding the heavy chain and a nucleotide sequence of SEQ ID NO: 346 encoding the light chain; a nucleotide sequence of SEQ ID NO: 347 encoding the heavy chain and a nucleotide sequence of SEQ ID NO: 348 encoding the light chain; a nucleotide sequence of SEQ ID NO: 349 encoding the heavy chain and a nucleotide sequence of SEQ ID NO: 350 encoding the light chain; nucleotide sequence of SEQ ID NO: 351 encoding the heavy chain and a nucleotide sequence of SEQ ID NO: 352 encoding the light chain; a nucleotide sequence of SEQ ID NO: 129 encoding the heavy chain and a nucleotide sequence of SEQ ID NO: 130 encoding the light chain; a nucleotide sequence of SEQ ID NO: 131 encoding the heavy chain and a nucleotide sequence of SEQ ID NO: 132 encoding the light chain; or nucleotide sequence of SEQ ID NO: 341 encoding the heavy chain and a nucleotide sequence of SEQ ID NO: 342 encoding the light chain.

6. The pharmaceutical composition of any of claims 1 to 5, wherein the mAb or antigen-binding fragment thereof is a hyperglycosylated mutant or wherein the Fc polypeptide of the mAb is glycosylated or aglycosylated.

7. The pharmaceutical composition of any of claims 1 to 6, wherein the transgene encodes a signal sequence at the N-terminus of the heavy chain and the light chain of said antigen-binding fragment that directs secretion and post translational modification in said human retina cells.

8. The pharmaceutical composition of claim 7, wherein said signal sequence is MYRMQLLLLLIALSLALVTNS (SEQ ID NO: 146) or a signal sequence from Table 2, Table 3 or Table 4.

9. The pharmaceutical composition of any of claims 1 to 8 wherein the transgene comprises a Furin/T2A linker between the nucleotide sequences coding for the heavy and light chains of mAb.

10. The pharmaceutical composition of any of claims 1 to 9, wherein the AAV capsid is AAV8.

11. The pharmaceutical composition of any of claims 1 to 10 wherein the anti-TNF α mAb is adalimumab.

12. The pharmaceutical composition of claim 11 wherein the heavy chain is encoded by a nucleotide sequence comprising the nucleotide sequence of SEQ ID NO: 115, 444 or 445 and the light chain is encoded by a nucleotide sequence comprising the nucleotide sequence of SEQ ID NO: 116, 448 or 449.

13. The pharmaceutical composition of claim 12, wherein the heavy chain, light chain and Furin/T2A linker are encoded by the nucleotide sequence of SEQ ID NO: 452 or 454.

14. The pharmaceutical composition of any of claims 11 to 13 wherein the regulatory sequence includes a CAG, EF1alpha, or mU1 promoter.

15. The pharmaceutical composition of any of claims 11 to 14, wherein the expression cassette comprises the nucleotide sequence of SEQ ID NO: 451 or 453.

16. The pharmaceutical composition of any of claims 11 to 15 wherein the viral capsid is an AAV8 viral capsid.

17. A polynucleotide having the nucleotide sequence of SEQ ID NO: 451 or 453 flanked by AAV ITR nucleotide sequences.

18. A recombinant AAV comprising an artificial genome comprising the polynucleotide of claim 17 and an AAV8 capsid.

19. A method of producing recombinant AAVs comprising:

(a) culturing a host cell containing:

(i) an artificial genome comprising a cis expression cassette flanked by AAV ITRs, wherein the cis expression cassette comprises a transgene encoding adalimumab or an antigen binding fragment thereof operably linked to expression control elements that will control expression of the transgene in human cells;

(ii) a *trans* expression cassette lacking AAV ITRs, wherein the trans expression cassette encodes an AAV rep and an AAV8 capsid protein operably linked to expression control elements that drive expression of the AAV rep and the AAV capsid protein in the host cell in culture and supply the AAV rep and the AAV capsid protein in *trans*; and

(iii) sufficient adenovirus helper functions to permit replication and packaging of the artificial genome by the AAV capsid protein; and

(b) recovering recombinant AAV encapsidating the artificial genome from the cell culture.

20. The method of claim 19 wherein the expression cassette comprises the nucleotide sequence of SEQ ID NO: 451 or 453.

21. A host cell comprising

(a) an artificial genome comprising a *cis* expression cassette flanked by AAV ITRs, wherein the *cis* expression cassette comprises a transgene encoding adalimumab or an antigen binding fragment thereof operably linked to expression control elements that will control expression of the transgene in human cells;

(b) a *trans* expression cassette lacking AAV ITRs, wherein the *trans* expression cassette encodes an AAV rep and an AAV8 capsid protein operably linked to expression control elements that drive expression of the AAV rep and the AAV capsid protein in the host cell in culture and supply the AAV rep and the AAV capsid protein in *trans*; and

(c) sufficient adenovirus helper functions to permit replication and packaging of the artificial genome by the AAV capsid protein.

22. The host cell of claim 21 wherein the expression cassette comprises the nucleotide sequence of SEQ ID NO: 451 or 453.

23. A pharmaceutical composition for treating angioedema including hereditary angioedema in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of AAV8 capsid (SEQ ID NO: 143); AAVrh10 capsid (SEQ ID NO: 145); or AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-kallikrein (anti-pKal) mAb, or an antigen-binding fragment thereof, operably linked to one or more

regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for administration to the subject, optionally wherein administration is intravenous, subcutaneous, intranasal, or intramuscular.

24. The pharmaceutical composition of claim 23, wherein the anti-pKal mAb is lanadelumab.

25. Use of a pharmaceutical composition for delivering lanadelumab to the bloodstream in the manufacture of a medicament for treating hereditary angioedema in a human subject in need thereof, said composition comprising a recombinant AAV comprising a transgene encoding lanadelumab operably linked to one or more regulatory sequences that control expression of the transgene in muscle and/or liver cells, wherein said recombinant AAV is to be administered to said human subject at a dose sufficient to result in expression from the transgene and secretion of lanadelumab into the bloodstream of the human subject to produce lanadelumab plasma levels of at least 5 µg/ml to at least 35 µg/ml lanadelumab in said subject.

26. The use of claim 25, wherein the lanadelumab plasma levels are 20 µg/ml to 35 µg/ml and the lanadelumab plasma levels are maintained for at least three months.

27. The use of claim 25 or 26 wherein the lanadelumab antibody secreted into the plasma exhibits greater a greater than at least 40%, 45%, 50%, 55%, 60%, 65% or 70 reduction in pKal activity as measured by a kinetic enzymatic functional assay wherein the activity of the lanadelumab antibody is measured at 2 weeks, 3 weeks, 4 weeks, 5 weeks, 6 weeks, 7 weeks, 8 weeks, 9 weeks, 10 weeks, 11 weeks or 12 weeks after said administration.

28. A pharmaceutical composition for delivery of an antibody or antigen binding fragment thereof to the bloodstream of a human subject in need thereof, comprising an AAV vector comprising:

(a) An AAV viral capsid that infects liver and/or muscle cells; and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a full-length antibody or an antigen-binding

fragment thereof, operably linked to a chimeric promoter that directs expression in muscle and liver cells;

wherein said AAV vector is formulated for intramuscular administration.

29. The pharmaceutical composition of claim 28 wherein the chimeric promoter is LMTP6 (SEQ ID NO: 320), LMTP13 (SEQ ID NO: 321), LMTP14 (SEQ ID NO: 322), LMTP15 (SEQ ID NO: 323), LMTP18 (SEQ ID NO: 324), LMTP19 (SEQ ID NO: 325), or LMTP20 (SEQ ID NO: 326).

30. A pharmaceutical composition for treating Alzheimer's disease, frontotemporal dementia (FD), tauopathies, progressive supranuclear palsy, chronic traumatic encephalopathy, Pick's Complex, and primary age-related tauopathy, Huntington's disease, juvenile Huntington's disease, Parkinson's disease, synucleinopathies, ALS, migraines, or cluster headaches in a human subject in need thereof, comprising an adeno-associated virus (AAV) vector having:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143), AAV9 capsid (SEQ ID NO: 144), AAVrh10 capsid (SEQ ID NO: 145), an AAVrh20 capsid, an AAVrh39 capsid or an AAVcy5 capsid; and

(b) an artificial genome comprising an expression cassette flanked by AAV inverted terminal repeats (ITRs), wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-amyloid beta (anti-A β), anti-sortilin, anti-Tau protein (anti-Tau), anti-semaphorin 4D (anti-SEMA4D), anti-alpha synuclein (anti-SNCA), anti-superoxide dismutase-1 (anti-SOD1) or anti-calcitonin gene-related peptide receptor (anti-CGRPR) mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human CNS, muscle, or liver cells;

wherein said AAV vector is formulated for administration to the subject, optionally wherein administration is intrathecal, intravenous, subcutaneous, intranasal, or intramuscular.

31. The pharmaceutical composition of claim 30, wherein the anti-A β mAb is solanezumab, lecanemab, or GSK933776; the anti-sortilin mAb is AL-001; the anti-Tau mAb is ABBV-8E12, UCB-0107, or NI-105 (BIIB076); the anti-SEMA4D mAb is VX15/2503; the anti-SNCA mAb is

prasinezumab, NI-202 (BIIB054), or MED-1341; the anti-SOD1 mAb is NI-2041.10D12 or NI-204.12G7; and the anti-CGRPR mAb is eptinezumab, fremanezumab, or galcanezumab.

32. A pharmaceutical composition for treating retinal disorders including diabetic retinopathy, myopic choroidal neovascularization (mCNV), macular degeneration (e.g., neovascular (wet) or dry age-related macular degeneration (nAMD)), macular edema (e.g., macular edema following a retinal vein occlusion (RVO) or diabetic macular edema (DME)), retinal vein occlusion, diabetic retinopathy (DR), non-infectious uveitis, or glaucoma, or abnormal vascularization of the retina in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV2.7m8 capsid (142), an AAV8 capsid (SEQ ID NO: 143), or an AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-vascular endothelial growth factor (anti-VEGF), anti-erythropoietin receptor (anti-EPOR), anti-A β , anti-activin receptor like kinase 1 (anti-ALK1), anti-complement component 5 (anti-C5), anti-endoglin (anti-ENG), anti-complement component 1Q (anti-CC1Q), anti-tumor necrosis factor-alpha (anti-TNF α), or anti-pKal mAb or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human retinal cells;

wherein said AAV vector is formulated for subretinal, intravitreal, suprachoroidal, or intranasal administration to said subject.

33. The pharmaceutical composition of claim 32 wherein the anti-VEGF mAb is sevacizumab; anti-EPOR mAb is LKA-651 (NSV2) or LKA-651 (NSV3); anti-A β mAb is solanezumab, lecanemab, or GSK933776; anti-ALK1 mAb is ascrinvacumab; anti-C5 mAb is tesidolumab or ravulizumab; anti-ENG mAb is carotuximab; the anti-CC1Q mAb is ANX-007; wherein anti-TNF α mAb is adalimumab, infliximab or golimumab; and the anti-pKal mAb is lanadelumab.

34. A pharmaceutical composition for treating multiple sclerosis in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143), AAV9 capsid (SEQ ID NO: 144), AAVrh10 capsid (SEQ ID NO: 145), an AAVrh20 capsid, an AAVrh39 capsid or an AAVcy5 capsid; and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-repulsive guidance molecule-A (anti-RGMA) mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human CNS, liver, or muscle cells;

wherein said AAV vector is formulated for administration to the subject, optionally wherein administration is intrathecal, intravenous, subcutaneous, intranasal, or intramuscular.

35. The pharmaceutical composition of claim 34, wherein the anti-RGMA mAb is elezanumab.

36. A pharmaceutical composition for treating amyloidosis (ATTR), familial amyloid cardiomyopathy (FAC), or familial amyloid polyneuropathy (FAP) in a human subject in need thereof, comprising AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143), an AAV9 capsid (SEQ ID NO: 144), an AAVrh10 capsid (SEQ ID NO: 145), an AAVrh20 capsid, an AAVrh39 capsid, or an AAVcy5 capsid; and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-TTR mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for administration to the subject, optionally wherein administration is intravenous, subcutaneous, intranasal, or intramuscular.

37. The pharmaceutical composition of claim 36, wherein the anti-TTR mAb is NI-301 or PRX-004.

38. A pharmaceutical composition for treating fibrotic disorders, pulmonary fibrosis, cystic fibrosis (CF), idiopathic pulmonary fibrosis (IPF), liver cirrhosis, atrial fibrosis, endomyocardial

fibrosis, old myocardial infarction, arthrofibrosis, Crohn's disease, ulcerative colitis, mediastinal fibrosis, myelofibrosis (MF), nephrogenic systemic fibrosis (NSF), progressive massive fibrosis (PMF), and retroperitoneal fibrosis (RPF) in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143), an AAV9 capsid (SEQ ID NO: 144), or AAVrh10 (SEQ ID NO: 145); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-connective tissue growth factor (anti-CTGF) mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for administration to said subject, optionally wherein administration is intravenous, subcutaneous, intranasal, or intramuscular.

39. The pharmaceutical composition of claim 38, wherein the anti-CTGF mAb is pamrevlumab.

40. A pharmaceutical composition for treating non-infectious uveitis, neuromyelitis optica (NMO), diabetic retinopathy (DR), or diabetic macular edema (DME) in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143), an AAV2.7m8 capsid (SEQ ID NO: 142) or an AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV inverted terminal repeats (ITRs) wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-interleukin-6 receptor (anti-IL6R), anti-interleukin 6 (IL6), or anti-cluster of differentiation 19 (anti-CD19) mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human retinal cells;

wherein said AAV vector is formulated for administration to said subject, optionally wherein administration is intranasal, subretinal, intravitreal, or suprachoroidal.

41. The pharmaceutical composition of claim 40, wherein the anti-IL6R mAb is satralizumab, sarilumab, or tocilizumab, or the anti-IL6 mAb is siltuximab, clazakizumab, sirukumab, olokizumab, or gerilimzumab, or the anti-CD19 mAb is inebilizumab.

42. A pharmaceutical composition for reducing, inhibiting or ameliorating a detrimental immune response in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143), an AAV9 capsid (SEQ ID NO: 144), an AAVrh10 capsid (SEQ ID NO: 145); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a substantially full-length or full-length mAb of an anti-interleukin-6 receptor (anti-IL6R) or anti-interleukin-6 (IL6), or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or muscle cells;

wherein said AAV vector is formulated for subcutaneous, intramuscular, intravenous or pulmonary administration to the subject.

43. The pharmaceutical composition of claim 42, wherein the anti-IL6R mAb is satralizumab, sarilumab, or tocilizumab, or the anti-IL6 mAb is siltuximab, clazakizumab, sirukumab, olokizumab, or gerilimzumab.

44. A pharmaceutical composition for treating inflammatory bowel disease (IBD) including UC and CD in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of AAV8 capsid (SEQ ID NO: 143); AAV9 capsid (SEQ ID NO: 144); or AAVrh10 capsid (SEQ ID NO: 145); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-integrin β 7 subunit (anti-ITGB7) mAb, or an antigen-binding fragment thereof, operably linked to one

or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for administration to the subject, optionally wherein administration is intravenous, subcutaneous, intranasal, or intramuscular.

45. The pharmaceutical composition of claim 44, wherein the anti-ITGB7 mAb is etrolizumab.

46. A pharmaceutical composition for treating osteoporosis or abnormal bone loss or weakness (e.g., treating giant cell tumor of bone, treating treatment-induced bone loss, slowing the loss of (or increasing) bone mass in breast and prostate cancer patients, preventing skeletal-related events due to bone metastasis, or for decreasing bone resorption and turnover in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143); AAVrh10 capsid (SEQ ID NO: 145); or an AAV9 capsid (SEQ ID NO: 144);

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding a full-length or substantially full-length anti-sclerostin (anti-SOST) mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for administration to the subject, optionally wherein administration is intravenous, subcutaneous, intranasal, or intramuscular.

47. The pharmaceutical composition of claim 46, wherein the anti-SOST mAb is romosozumab.

48. A pharmaceutical composition for treating atopic dermatitis in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143) or AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding an anti-IL13 mAb or anti-IL31RA, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for intravenous administration to the liver or muscle of said subject.

49. The pharmaceutical composition of claim 48 wherein the anti-IL13 or the IL31RA is tralokinumab or nemolizumab.

50. A pharmaceutical composition for treating eosinophilic asthma in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143) or an AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding an anti-IL5R mAb or anti-IgE mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for intravenous administration to the liver or muscle of said subject.

51. The pharmaceutical composition of claim 50 wherein the anti-IL5R or anti-IgE mAb is reslizumab or omalizumab.

52. A pharmaceutical composition for treating asthma or chronic obstructive pulmonary disease (COPD) in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143) or an AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding an anti-IL5, anti-IL-5R, anti-IgE, or anti-TSLP

mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for intravenous administration to the liver or muscle of said subject.

53. The pharmaceutical composition of claim 52 wherein the anti-IL-5, anti-IL5R, anti-IgE, or anti-TSLP mAb is benralizumab, reslizumab, omalizumab, or tezepelumab. .

54. A pharmaceutical composition for treating chronic idiopathic urticaria in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO:143) or an AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding an anti-IgE mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for intravenous administration to the liver or muscle of said subject.

55. The pharmaceutical composition of claim 54, wherein the anti-IgE mAb is omalizumab.

56. A pharmaceutical composition for treating myasthenia gravis in a human subject in need thereof, comprising an AAV vector comprising:

(a) a viral capsid that is at least 95% identical to the amino acid sequence of an AAV8 capsid (SEQ ID NO: 143) or AAV9 capsid (SEQ ID NO: 144); and

(b) an artificial genome comprising an expression cassette flanked by AAV ITRs wherein the expression cassette comprises a transgene encoding an anti-C5 mAb, or an antigen-binding fragment thereof, operably linked to one or more regulatory sequences that control expression of the transgene in human liver cells or human muscle cells;

wherein said AAV vector is formulated for intravenous administration to the liver or muscle of said subject.

57. The pharmaceutical composition of claim 56 wherein the anti-C5 is ravulizumab.

58. A method of determining human anti-pKal antibody activity in a sample, said method comprising

- a. Incubating the sample with activated human pKal;
- b. Subsequently incubating the sample having been incubated with the activated human pKal with the synthetic substrate Pro-Phe-Arg-AMC
- c. Measure the release of AMC over three hours as compared to a control sample.