

INSULIN-LIKE GROWTH FACTOR-1 RECEPTOR (IGF1R) ANTIBODIES AND RELATED METHODS OF TREATMENT

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority to U.S. Provisional Application No. 63/671,695, filed July 15, 2024, and to U.S. Provisional Application No. 63/585,198, filed September 25, 2023, and the respective contents of each of which is hereby incorporated in its entirety.

REFERENCE TO SEQUENCE LISTING

[0002] The contents of the concurrently filed electronic sequence listing (“037525-00589_SequenceListing.xml”; size: 72,312 bytes; created: September 25, 2024) is hereby incorporated by reference in its entirety.

BACKGROUND

[0003] Thyroid eye disease (“TED”), also known as thyroid-associated ophthalmopathy (“TAO”), Graves' ophthalmopathy or orbitopathy (“GO”), thyrotoxic exophthalmos, dysthyroid ophthalmopathy, and several other terms, is an orbitopathy associated with thyroid dysfunction. TAO is divided into two types. Active, or acute, TED typically lasts 1-3 years and is characterized by an ongoing autoimmune/inflammatory response in the soft tissues of the orbit. Active TED is responsible for the expansion and remodeling of the ocular soft tissues. The autoimmune/inflammatory response of active TED spontaneously resolves eventually and the condition transitions into inactive, or chronic, TED. Inactive TED is the term used to describe the long-term/permanent sequelae of active TED. In TED, as a result of inflammation and expansion of orbital soft tissues, primarily eye muscles and adipose tissue, the eyes are forced forward (bulge) out of their sockets—a phenomenon termed proptosis or exophthalmos. Although most cases of TED do not result in loss of vision, this condition can cause vision-threatening exposure keratopathy, troublesome diplopia (double vision), and compressive dysthyroid optic neuropathy.

[0004] The cause of TED is unknown. However, antibodies that activate the insulin-like growth factor 1 receptor (“IGF1R”) have been detected and implicated in active TED. IGF1R is a widely expressed heterotetrameric protein involved in the regulation of proliferation and

metabolic function of many cell types. It is a tyrosine kinase receptor comprising two subunits. IGF1R- α contains a ligand-binding domain while IGF1R- β is involved in signaling and contains tyrosine phosphorylation sites. Monoclonal antibodies directed against IGF1R have been developed and assessed as a therapeutic strategy for several types of solid tumors and lymphomas.

[0005] Without being bound to any theory, it is believed that the thyroid stimulating hormone (“TSH”) receptor (“TSHR”) and IGF1R form a physical and functional complex in orbital fibroblasts, and that blocking IGF1R appears to attenuate both IGF1-dependent signaling and TSH-dependent signaling. It has been suggested that blocking IGF1R using an antibody antagonist might reduce IGF-1-dependent signaling and/or TSH-dependent signaling, thereby interrupting the pathological activities of autoantibodies acting as agonists on either receptor.

[0006] The current standard of care for Thyroid Eye Disease is teprotumumab (Tepezza[®]), an IGF1R inhibitor indicated for the treatment of TED, which is administered via intravenous (“IV”) infusion at 10 mg/kg for the first infusion and 20 mg/kg for subsequent infusions every 3 weeks for a total of 8 infusions over a 24-week period. These infusions are typically administered over a 60 to 90-minute period and result in systemic administration of Tepezza[®], leading to IGF1R inhibition in various parts of the body that can lead to adverse or undesired effects (e.g., infusion reactions, exacerbation of irritable bowel disease, hyperglycemia, or hearing loss).

SUMMARY OF THE DISCLOSURE

[0007] Provided herein are methods of treating or reducing the severity of TED, and achieving specific treatment endpoints in the treatment of TED, such as reducing proptosis, diplopia, TED clinical activity score and subsets and individual measures thereof, and of improving the quality of life of TED patients, using novel antibodies (or antigen-binding fragments thereof) that specifically bind to IGF1R. In some embodiments, these anti-IGF1R antibodies, or antigen-binding fragments thereof, are expressed by a viral or other vector administered to a subject in need of treatment. Such constructs may be designed, e.g., to express a single chain fragment variable (“scFv”), a recombinant antibody fragment comprising a light chain variable domain and a heavy chain variable domain of an immunoglobulin or a portion thereof, in various configurations. In some embodiments, the viral or other vector expresses one or more anti-IGF1R antibodies, or antigen-binding fragments, locally, e.g., in a specific tissue or tissues or specific cell type or cell types. The novel gene therapy methods described

herein may thus allow for localized expression of anti-IGF1R antibodies, or antigen-binding fragments. In some embodiments, the anti-IGF1R antibodies, or antigen-binding fragments described herein, may be directly administered as a pharmaceutical composition to at least one eye of a subject in need of treatment. Systemic administration, or at least clinically relevant systemic administration, may be avoided in both cases of vector delivery designed for local expression or direct administration (e.g., to the lacrimal gland), reducing or eliminating the burden and undesirable side effects associated with systemic administration of Tepezza[®], the current standard of care for TED.

[0008] In a first general aspect, the disclosure provides an antibody or an antigen-binding fragment that specifically binds to a human IGF1R protein, comprising: a) a heavy chain variable domain, comprising a CDR1 region having a polypeptide sequence of SEQ ID NO: 3, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 3, a CDR2 region having a polypeptide sequence of SEQ ID NO: 4, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 4, and/or a CDR3 region having a polypeptide sequence of SEQ ID NO: 5, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 5; and/or b) a light chain variable domain, comprising a CDR1 region having a polypeptide sequence of SEQ ID NO: 7, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 7, a CDR2 region having a polypeptide sequence of SEQ ID NO: 8, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 8, and/or a CDR3 region having a polypeptide sequence of SEQ ID NO: 9, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 9.

[0009] In some embodiments, the heavy chain variable domain comprises a CDR1 region having a polypeptide sequence of SEQ ID NO: 3, the heavy chain variable domain comprises a CDR2 region having a polypeptide sequence of SEQ ID NO: 4, and/or the heavy chain variable domain comprises a CDR3 region having a polypeptide sequence of SEQ ID NO: 5; and/or the light chain variable domain comprises a CDR1 region having a polypeptide sequence of SEQ ID NO: 7, the light chain variable domain comprises a CDR2 region having a polypeptide sequence of SEQ ID NO: 8, and/or the light chain variable domain comprises a CDR3 region having a polypeptide sequence of SEQ ID NO: 9.

[0010] In some embodiments, the heavy chain variable domain comprises a CDR1 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 3, the heavy chain variable domain comprises a CDR2 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared

to SEQ ID NO: 4, and/or the heavy chain variable domain comprises a CDR3 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 5; and/or the light chain variable domain comprises a CDR1 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 7, the light chain variable domain comprises a CDR2 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 8, and/or the light chain variable domain comprises a CDR3 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 9.

[0011] In some embodiments, the heavy chain variable domain comprises a polypeptide sequence of SEQ ID NO: 2; and/or the light chain variable domain comprises a polypeptide sequence of SEQ ID NO: 6. In some embodiments, the heavy chain variable domain comprises a polypeptide sequence having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 2; and/or the light chain variable domain comprises a polypeptide sequence having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 6. In some embodiments, the heavy chain variable domain comprises a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 2; and/or the light chain variable domain comprises a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 6.

[0012] In some embodiments, the antibody or antigen-binding fragment specifically binds to: a) an IGF1R protein having a polypeptide sequence of SEQ ID NO: 1, or a fragment comprising at least 20 contiguous amino acids thereof; b) an alpha chain of an IGF1R protein having a polypeptide sequence comprising the sequence spanning positions 31-736 of SEQ ID NO: 1, or a fragment comprising at least 20 contiguous amino acids thereof; and/or c) a beta chain of an IGF1R protein having a polypeptide sequence comprising the sequence spanning positions 741-1367 of SEQ ID NO: 1, or a fragment comprising at least 20 contiguous amino acids thereof.

[0013] In some embodiments, the antibody is a rabbit antibody or a mouse antibody.

[0014] In some embodiments, the antibody is a monoclonal antibody.

[0015] In some embodiments, the antibody comprises a monoclonal antibody, a grafted antibody, a chimeric antibody, a human antibody, or a humanized antibody.

[0016] In some embodiments, the antigen-binding fragment comprises a Fab, a Fab', a F(ab')₂, a variable fragment (Fv), a triabody, a tetrabody, a minibody, a bispecific F(ab')₂, a

trisppecific F(ab')₂, a diabody, a bispecific diabody, a single chain variable fragment (scFv), a scFv-Fc, a Fab-Fc, a VHH, or a bispecific scFv.

[0017] In some embodiments, the antibody or antigen-binding fragment is operably linked to a signal peptide. In some embodiments, the signal peptide is configured to result in secretion of the antibody or antigen-binding fragment from a human cell.

[0018] In a second general aspect, the disclosure provides a nucleic acid molecule encoding the antibody or antigen-binding fragment according to any of the embodiments described herein. In some embodiments, the nucleic acid molecule comprises a nucleotide sequence sharing at least 70, 75, 80, 85, 90 or 95% identity with a nucleotide sequence comprising SEQ ID NOs: 21 or 22. In some embodiments, the nucleic acid molecule comprises SEQ ID NO: 21 or SEQ ID NO: 22. In some embodiments, the nucleic acid molecule comprises a nucleotide sequence sharing at least 70, 75, 80, 85, 90 or 95% identity with SEQ ID NO: 21 and a nucleotide sequence sharing at least 70, 75, 80, 85, 90 or 95% identity with SEQ ID NO: 22. In some embodiments, the nucleic acid molecule comprises SEQ ID NO: 21 and SEQ ID NO: 22. In some embodiments, the nucleic acid molecule comprises a codon optimized version of SEQ ID NO: 21 and/or SEQ ID NO: 22, or a fragment thereof.

[0019] In some embodiments, the nucleic acid molecule further comprises a promoter or enhancer sequence operably linked to the sequence in the molecule encoding the antibody or antigen-binding fragment.

[0020] In some embodiments, the nucleic acid molecule further encodes a signal peptide sequence operably linked to the encoded antibody or antigen-binding fragment.

[0021] In some embodiments, the nucleic acid molecule may be contained within a liposome.

[0022] In a third general aspect, the disclosure provides a vector comprising a nucleic acid molecule according to any of the embodiments described herein.

[0023] In some embodiments, the vector comprises an adenoviral vector, an HIV vector, a lentiviral vector, an adeno-associated virus ("AAV") vector, a plasmid vector, or a C³DNA vector.

[0024] In some embodiments, the AAV vector comprises AAV2, AAV4, AAV8, or AAV9.

[0025] In some embodiments, the vector may be contained within a liposome.

[0026] In a fourth general aspect, the disclosure provides a recombinant vector comprising an expression cassette, the expression cassette comprising a polynucleotide encoding an antibody or antigen-binding fragment according to any of the embodiments described herein, the polynucleotide being operatively linked to a promoter. In some embodiments, the antibody

or antigen-binding fragment is an scFv. In some embodiments, the scFv comprises: a heavy chain variable domain comprising a polypeptide sequence of SEQ ID NO: 2; and/or a light chain variable domain comprising a polypeptide sequence of SEQ ID NO: 6. In some embodiments, the scFv further comprises a linker sequence (e.g., a GS linker such as SEQ ID NO: 42) in between the heavy and light variable chain domains. In other aspects, any other linker sequence may be used. In some embodiments, the heavy chain variable domain comprises a polypeptide sequence having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 2; and/or the light chain variable domain comprises a polypeptide sequence having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 6. In some embodiments, the heavy chain variable domain comprises a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 2; and/or the light chain variable domain comprises a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 6. In some embodiments, the recombinant vector is a recombinant adeno-associated virus (“rAAV”) vector comprising an AAV capsid.

[0027] In some embodiments, the scFv comprises: a) a heavy chain variable domain, comprising a CDR1 region having a polypeptide sequence of SEQ ID NO: 3, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 3, a CDR2 region having a polypeptide sequence of SEQ ID NO: 4, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 4, and/or a CDR3 region having a polypeptide sequence of SEQ ID NO: 5, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 5; and/or b) a light chain variable domain, comprising a CDR1 region having a polypeptide sequence of SEQ ID NO: 7, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 7, a CDR2 region having a polypeptide sequence of SEQ ID NO: 8, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 8, and/or a CDR3 region having a polypeptide sequence of SEQ ID NO: 9 or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 9.

[0028] In some embodiments (e.g., where the recombinant vector is an rAAV vector), the expression cassette may be flanked by two inverted terminal repeats (“ITRs”). In some embodiments, the ITRs are AAV2 ITRs. In some embodiments, the AAV capsid comprises a viral protein (“VP”) that shares at least 95%, 98%, or 100% identity with AAV2 capsid protein (SEQ ID NO: 13), AAV5 capsid protein (SEQ ID NO: 15), AAV8 capsid protein (SEQ ID NO: 17), or AAV9 capsid protein (SEQ ID NO: 19). In some embodiments, the AAV capsid comprises a VP that shares at least 95%, 98%, or 100% identity with AAV9 capsid protein

(SEQ ID NO: 19). In some embodiments, the AAV capsid comprises a VP used in the capsid of any other AAV serotype.

[0029] In a fifth general aspect, the disclosure provides a composition comprising any recombinant vector as described herein. In some embodiments of the composition, the recombinant vector is an rAAV vector, wherein the rAAV vector comprises: (a) an AAV2, AAV5, AAV8, and/or AAV9 capsid, and (b) an expression cassette, wherein the expression cassette comprises a polynucleotide comprising a nucleotide sequence sharing at least 80, 85, 90, 95, or 100% identity with a nucleotide sequence comprising SEQ ID NOs: 21 or 22, and wherein the polynucleotide is linked to a promoter.

[0030] In some embodiments, e.g., where the composition comprises an rAAV vector, the rAAV vector may comprises: (a) an AAV2, AAV5, AAV8, and/or AAV9 capsid, and (b) an expression cassette, wherein the expression cassette comprises a polynucleotide sequence sharing at least 80, 85, 90, 95, or 100% identity with SEQ ID NOs: 21 or 22. In some embodiments, the AAV capsid is AAV2, AAV5, AAV8, or AAV9.

[0031] In some embodiments, a composition according to the disclosure comprises an rAAV vector, wherein the rAAV vector comprises: (a) an AAV capsid, and (b) an expression cassette, wherein the expression cassette comprises a polynucleotide encoding a polypeptide sequence sharing at least 80, 85, 90 or 95% sequence identity with SEQ ID NOs: 2 and/or 6, and wherein the polynucleotide is optionally linked to a promoter. In some embodiments, the polypeptide sequence comprises SEQ ID NO: 2 N-terminal to SEQ ID NO: 6, whereas in others SEQ ID NO: 6 is N-terminal to SEQ ID NO: 2.

[0032] In a sixth general aspect, the disclosure provides pharmaceutical compositions comprising: any antibody or antigen-binding fragment described herein, any nucleic acid molecule described herein, any vector described herein, or any composition described herein, and a pharmaceutically acceptable carrier. In some embodiments, the composition comprises about 1×10^7 to about 1×10^{14} genome copies per milliliter of the rAAV vector. In some embodiments, the composition comprises about 1×10^{12} to about 6.5×10^{12} genome copies per milliliter of the rAAV vector.

[0033] In a seventh general aspect, the disclosure provides a kit comprising the antibody or antigen-binding fragment of any embodiment described herein, the vector of any embodiment described herein, the composition of any embodiment described herein, or the pharmaceutical composition of any embodiment described herein, and a pharmaceutically acceptable carrier, and instructions for use in treating an ocular disease, disorder, or condition in a subject, the instructions comprising administering the antibody, antigen-binding fragment

of the antibody, vector, composition, or pharmaceutical composition to the eye of the subject (e.g., topically or via injection). In some embodiments, the ocular disease, disorder, or condition is Graves' Orbitopathy or TED.

[0034] In an eighth general aspect, the disclosure provides methods of treating an ocular disease, disorder, or condition in a subject in need thereof, such methods comprising administering an effective amount of: a) the antibody or antigen-binding fragment of any of the embodiments described herein, b) the vector of any embodiment described herein, c) the composition of any embodiment described herein, or d) the pharmaceutical composition of any embodiment described herein, to an eye of the subject. In some embodiments, the administration to an eye of a subject provides local administration of the IGF1R antibody or antigen-binding fragment to the eye.

[0035] In some embodiments, the pharmaceutical composition is delivered to an accessory lacrimal gland. In some embodiments, the accessory lacrimal gland is the meibomian glands. In some embodiments, the pharmaceutical composition is delivered to the trabecular meshwork. In some embodiments, the administration provides local administration of the IGF1R antibody or antigen-binding fragment to the accessory lacrimal gland or the trabecular meshwork.

[0036] In some embodiments, the administration is by an injection to the eye, accessory gland, or trabecular meshwork. In some embodiments the administration is topical to the eye.

[0037] In some embodiments, the ocular disease, disorder, or condition is TED. In some embodiments, the ocular disease, disorder, or condition comprises a) hypothyroid associated visual disturbance, b) a reduction in visual acuity due to lid retraction, c) lid wiper epitheliopathy, d) dry eye, e) aqueous deficiency, f) double vision, g) compressive optic neuropathy, h) ocular pain, i) proptosis, j) and/or diplopia.

[0038] In some embodiments, the ocular disease, disorder, or condition is treated by administering 10^8 to 10^{14} genome copies of any of the vectors described herein to one or more tissues of the subject (e.g., to at least one eye, or to an accessory lacrimal gland or the trabecular meshwork of the subject).

[0039] In some embodiments, the method results in: a) an improvement of one or more symptoms of the ocular condition; and/or b) a delay of the progression of the ocular condition.

[0040] In some embodiments, the subject is a human subject.

[0041] In a ninth general aspect, the disclosure provides a recombinant cell transfected or transduced with the vector of any of the embodiments described herein.

[0042] In a tenth general aspect, the disclosure provides a kit comprising the pharmaceutical composition of any embodiment described herein and instructions for use in treating a condition in a human subject, wherein the instructions comprise administering the pharmaceutical composition to an eye of the human subject. In some embodiments, the disclosure provides a pharmaceutical composition according to any of the embodiments described herein, for use in the manufacture of a medicament for treating a condition in a human subject in need thereof.

[0043] In an eleventh general aspect, the disclosure provides a method of treating a disease, disorder, or condition in a subject in need thereof with a therapeutic antibody or antigen-binding fragment (e.g., an scFv), wherein the method comprises administering an effective amount of a vector comprising a nucleic acid molecule encoding the antibody or antigen-binding fragment to the subject, and wherein the administration results in local expression of the antibody or antigen-binding fragment in a specific cell type or tissue of the subject. In some embodiments, the cell type or tissue is selected from: eye cells and/or tissue, accessory lacrimal gland cells and/or tissue, trabecular meshwork cells and/or tissue. In some embodiments, the cell type or tissue is selected from: periorbital tissue, periorbital adipose, scleral fibroblasts, conjunctival epithelial cells, and/or cornea keratocytes.

[0044] In some embodiments, administering an effective amount of a vector comprising a nucleic acid molecule encoding the antibody or antigen-binding fragment may be used to treat eye redness (e.g., caused by lid retraction) in a subject suffering from TED. In some aspects, administering an effective amount of a vector comprising a nucleic acid molecule encoding the antibody or antigen-binding fragment may be used to treat compressive optic neuropathy (e.g., compression of the optic nerve due to the extrinsic compressive forces from the adipose accumulation behind the globe) in a subject suffering from TED.

BRIEF DESCRIPTION OF THE FIGURES

[0045] **FIG. 1** shows a vector map of an rAAV plasmid vector (“pAAV-VH-VL-Stuffer-2”) configured to express and package an scFv comprising a heavy chain variable domain and a light chain variable domain of one of the anti-IGF1R antibodies described herein.

[0046] **FIGs. 2A-2B** show examples of delivery of a viral vector to the lacrimal gland of a human subject.

[0047] **FIG. 3** is an annotated photograph of an ELISA microplate, showing results described in Example 2.

[0048] FIGs. 4-12 contain graphs showing binding data for the VL-VH and VH-VL scFv constructs, and other analytes, tested in the experiments described in Example 4.

[0049] FIGs. 13-14 are photographs of SDS-PAGE gels showing the purified VL-VH and VH-VL scFv constructs generated in the experiments described in Example 5.

[0050] FIGs. 15-23 contain graphs showing binding data for the purified VL-VH and VH-VL scFv constructs, and other analytes, tested in the experiments described in Example 5.

DETAILED DESCRIPTION

[0051] The present disclosure provides novel antibodies, and fragments thereof, configured to specifically bind to IGF1R, as well as vectors, expression constructs, and formulations that may be used to treat GO and/or TED via gene therapy. Treatment may comprise transduction of human cells and/or tissue (e.g., exocrine glands, endocrine glands, adipose tissue, epithelial, endothelial, stromal cells, etc.) with an adenoviral-associated vector, adenoviral vector, lentiviral vector and/or plasmid DNA encoding a transgene expressing an scFv of an antibody to IGF1R and an appropriate promoter and/or enhancer, ITRs and polyA tail to drive transgene expression, resulting in protein translation. Additionally, a signal peptide (e.g., the IgK leader sequence) may be included in the transgene construct to induce secretion out of the cell following expression. In some embodiments, the expression and secretion of the gene product (e.g., IGF1R-scFv) will end up diffusing into the extraorbital fat pads, thereby blocking IGF-1 from binding to IGF1R and inhibiting IGF1R downstream signaling. Specifically, IGF1R activation by IGF-1 results in AKT activation in orbital adipocytes and orbital fibroblasts. Activated AKT inhibits FOXO1, which subsequently activates the mTOR signaling cascade to induce PPAR γ transcription factor activation leading to expression and secretion of hyaluronic acid (HA) and adipogenesis. Increased expression of HA and adipose leads to expansion of the fat pad behind the orbit resulting in increased force exerted on the posterior section of the globe that results in forward movement, or “bulging”, of the eyeball. This embodiment would provide constitutive inhibition of IGF1R signaling and the subsequent HA and adipose expansion behind the globe through binding of, e.g., a IGF1R-scFv gene product to the IGF1 receptor. Inhibition of HA production and adipogenesis will decrease the volume of the adipose tissue behind the globe causing retraction of the eyeballs back into the orbit (i.e., decreased bulging).

[0052] Definitions

[0053] To facilitate understanding of the disclosure, several terms and abbreviations as used herein are defined below as follows:

[0054] As used herein, the term “antibody” encompasses the various forms of antibodies including but not being limited to whole antibodies, monoclonal antibodies, antibody fragments, human antibodies, humanized antibodies, chimeric antibodies and genetically engineered antibodies as long as the characteristic properties such as specificity are retained.

[0055] As used herein, the terms “antigen-binding fragment,” “antigen-binding antibody fragment,” “fragment,” and “antibody fragment” are used interchangeably to refer to any fragment that comprises a portion of a full-length antibody and that binds to IGF1R, generally at least the antigen-binding portion (e.g., the heavy or light chain variable domain) or the variable region thereof (e.g., one or more CDRs) of a full-length antibody. Examples of antibody fragments include, but are not limited to, diabodies, single-chain antibody molecules, multispecific antibodies, Fab, Fab', F(ab')₂, Fv or scFv. Further, the term “antibody” as used herein includes both antibodies and antigen-binding fragments thereof. In addition, antibody fragments comprise single chain polypeptides having the characteristics of a VH chain, namely being able to assemble together with a VL chain or of a VL chain binding to IGF1R, namely being able to assemble together with a VH chain to a functional antigen-binding pocket and thereby providing the property of inhibiting the binding of IGF-1 and/or IGF-2 to IGF1R.

[0056] The terms “monoclonal antibody” or “monoclonal antibody composition,” as used herein refer to a preparation of antibody molecules of a single amino acid composition. Accordingly, the term “human monoclonal antibody” refers to antibodies displaying a single binding specificity which have variable and constant regions derived from human germline immunoglobulin sequences. In one embodiment, the human monoclonal antibodies are produced by a hybridoma which includes a B cell obtained from a transgenic non-human animal, e.g., a transgenic mouse, having a genome comprising a human heavy chain transgene and a light human chain transgene fused to an immortalized cell.

[0057] The term “expression cassette” refers to a polynucleotide comprising at least one polynucleotide sequence encoding a protein of interest (e.g., an anti-IGF1R antibody or an IGF1R-binding fragment thereof, such as a heavy chain, light chain, or scFv, or any other binding construct described herein). In some embodiments, (e.g., where the vector is an rAAV virion), the expression cassette may further comprise inverted terminal repeats (ITRs) flanking both sides of the at least one polynucleotide sequence encoding a protein of interest.

[0058] The term “human antibody” as used herein, is intended to include antibodies having variable and constant regions derived from human germline immunoglobulin sequences. The term “humanized antibody” as used herein refers to antibodies in which the framework or “complementarity determining regions” (CDR) have been modified to comprise the CDR of

an immunoglobulin of different specificity as compared to that of the parent immunoglobulin. In some embodiments, a murine CDR is grafted into the framework region of a human antibody to prepare the “humanized antibody.”

[0059] The term “recombinant human antibody,” as used herein, is intended to include all human antibodies that are prepared, expressed, created or isolated by recombinant means, such as antibodies isolated from a host cell such as an SP2-0, NS0, or CHO cell, or from an animal (e.g., a mouse) that is transgenic for human immunoglobulin genes or antibodies expressed using a recombinant expression vector transfected into a host cell. Such recombinant human antibodies have variable and constant regions derived from human germline immunoglobulin sequences in a rearranged form. The antigen-binding fragments and fusion proteins described herein can also be prepared by recombinant means.

[0060] The term “variable domain” (variable portion of a light chain (VL), and the variable portion of a heavy chain (VH)), as used herein, denotes each of the pair of light and heavy chains which is involved directly in binding the antibody to the antigen. The domains of variable human light and heavy chains have the same general structure and each domain comprises four framework (FR) regions whose sequences are widely conserved, connected by three “hypervariable regions” (also known as complementarity determining regions, or “CDRs”). The framework regions adopt a β -sheet conformation and the CDRs may form loops connecting the β -sheet structure. The CDRs in each chain are held in their three-dimensional structure by the framework regions and form together with the CDRs from the other chain the antigen binding site. The antibody heavy and light chain CDR3 regions play an important role in the binding specificity/affinity of antibodies.

[0061] The terms “complementarity determining region,” “CDR,” “hypervariable region,” or “antigen-binding portion of an antibody” are used interchangeably herein and refer to the amino acid residues of an antibody which are responsible for antigen-binding. The hypervariable region comprises amino acid residues from the complementarity determining regions or CDRs. “Framework” or “FR” regions are those variable domain regions other than the hypervariable region residues as herein defined. Therefore, the light and heavy chains of an antibody comprise from N- to C-terminus the domains FR1, CDR1, FR2, CDR2, FR3, CDR3, and FR4. Especially, CDR3 of the heavy chain is the region which contributes most to antigen binding. CDR and FR regions may be determined, e.g., according to the standard definition of Kabat et al., *Sequences of Proteins of Immunological Interest*, 5th Ed. Public Health Service, National Institutes of Health, Bethesda, Md. (1991)) and/or those residues from a “hypervariable loop.”

[0062] The term “fusion protein” as used herein relates to a protein or polypeptide comprising at least one first protein or polypeptide joined or linked to at least one second protein or polypeptide. In some aspects, the at least one protein or polypeptide is joined or linked to at least one second protein or polypeptide through one or more linker sequences (e.g., an intervening peptide sequence). An example of a fusion protein is a chimeric protein. In some aspects, the fusion protein is an scFv, a diabody, a triabody, or a tetrabody. A fusion protein can be created using routine techniques known in the art such as recombinant DNA technology, through joining or linking of two or more genes that originally coded for separate proteins. Thus, a fusion protein may comprise a multimer of different or identical binding proteins which are expressed as a single, linear polypeptide.

[0063] The terms “binding to IGF1R” or “specific binding to IGF1R” are used interchangeably herein and mean the binding of the antibody to IGF1R in an *in vitro* assay, preferably in a binding assay in which the antibody is bound to a surface, and binding of IGF1R is measured using Surface Plasmon Resonance (SPR). Binding means a binding affinity (K_D) of 10^{-8} M or less, preferably 10^{-13} to 10^{-9} M. Binding to IGF1R can be investigated by a BIAcore assay (Pharmacia Biosensor AB, Uppsala, Sweden). The affinity of the binding is defined by the terms k_a (rate constant for the association of the antibody from the antibody/antigen complex), k_d (dissociation constant), and K_D (k_d/k_a). The antibodies used in the methods disclose herein typically show a K_D of about 10^{-9} M or less.

[0064] The antibodies, or antigen-binding fragments thereof, used in the methods disclosed herein inhibit the binding of IGF-1 and/or IGF-2 to IGF1R. The inhibition is measured as IC_{50} in an assay for binding of IGF-1/IGF-2 to IGF-IR on cells. Such an assay is known to one of skill in the art and is described, for example, U.S. Pat. No. 7,579,157, which is incorporated herein in its entirety. The IC_{50} values of the antibodies used in the methods disclosed herein for the binding of IGF-1 and IGF-2 to IGF1R typically are no more than 2 nM. IC_{50} values are measured as average or median values of at least three independent measurements. Single IC_{50} values may be excluded from the scope.

[0065] The term “inhibiting the binding of IGF-1 and/or IGF-2 to IGF1R,” as used herein, refers to inhibiting the binding of I^{125} -labeled IGF-1 or IGF-2 to IGF-IR presented on the surface of cells in an *in vitro* assay. Inhibiting means an IC_{50} value of 2 nM or lower.

[0066] The phrase “therapeutically effective” is intended to qualify the amount of active ingredients used in the treatment of a disease or disorder or on the effecting of a clinical endpoint.

[0067] The term “therapeutically acceptable” refers to those compounds (or salts, prodrugs, tautomers, zwitterionic forms, etc.) which are suitable for use in contact with the tissues of patients without undue toxicity, irritation, and allergic response, are commensurate with a reasonable benefit/risk ratio, and are effective for their intended use.

[0068] As used herein, reference to “treatment” of a subject or patient is intended to include prevention, prophylaxis, attenuation, amelioration and therapy. Treatment may also include prevention of disease. Prevention of a disease may involve complete protection from disease, for example as in the case of prevention of infection with a pathogen, or may involve prevention of disease progression. For example, prevention of a disease may not mean complete foreclosure of any effect related to the diseases at any level, but instead may mean prevention of the symptoms of a disease to a clinically significant or detectable level. Prevention of diseases may also mean prevention of progression of a disease to a later stage of the disease.

[0069] The terms “subject” and “patient” are used interchangeably herein to mean all mammals including humans. Examples of subjects include, but are not limited to, humans, monkeys, dogs, cats, horses, cows, goats, sheep, pigs, and rabbits. In one embodiment, the subject or patient is a human.

[0070] The terms “affected with a disease or disorder,” “afflicted with a disease or disorder,” or “having a disease or disorder” are used interchangeably herein and refer to a subject or patient with any disease, disorder, syndrome or condition. No increased or decreased level of severity of the disorder is implied by the use of one the terms as compared to the other.

[0071] The term “disease” as used herein is intended to be generally synonymous, and is used interchangeably with, the terms “disorder,” “syndrome,” and “condition” (as in medical condition), in that all reflect an abnormal condition of the human or animal body or of one of its parts that impairs normal functioning, is typically manifested by distinguishing signs and symptoms, and causes the human or animal to have a reduced duration or quality of life.

[0072] When introducing elements of the present disclosure or the preferred embodiment(s) thereof, the articles “a,” “an,” “the” and “said” are intended to mean that there are one or more of the elements. The terms “comprising,” “including” and “having” are intended to be inclusive and mean that there may be additional elements other than the listed elements.

[0073] The term “and/or” when used in a list of two or more items, means that any one of the listed items can be employed by itself or in combination with any one or more of the listed items. For example, the expression “A and/or B” is intended to mean either or both of A and B, i.e., A alone, B alone or A and B in combination. The expression “A, B, and/or C” is intended

to mean A alone, B alone, C alone, A and B in combination, A and C in combination, B and C in combination or A, B, and C in combination.

[0074] When ranges of values are disclosed, and the notation “from n1 . . . to n2” or “between n1 . . . and n2” is used, where n1 and n2 are the numbers, then unless otherwise specified, this notation is intended to include the numbers themselves and the range between them. This range may be integral or continuous between and including the end values. By way of example, the range “from 2 to 6 carbons” is intended to include two, three, four, five, and six carbons, since carbons come in integer units. Compare, by way of example, the range “from 1 to 3 μM (micromolar),” which is intended to include 1 μM , 3 μM , and everything in between to any number of significant figures (e.g., 1.255 μM , 2.1 μM , 2.9999 μM , etc.).

[0075] The term “comprising” encompasses “including” as well as “consisting.” For example, a composition “comprising” X may consist exclusively of X or may include something additional e.g., X+Y.

[0076] As used herein, “Thyroid Eye Disease” (TED), “Thyroid-associated Ophthalmopathy” (TAO), “Thyroid Inflammatory Eye Disease (TIED),” “Graves’ Ophthalmopathy” (GO) or “Graves’ Orbitopathy” (GO) refer to the same disorder or condition and are used interchangeably. They all refer to the inflammatory orbital pathology associated with some autoimmune thyroid disorders, most commonly with “Graves’ Disease” (GD), but sometimes with other diseases, e.g., Hashimoto’s thyroiditis.

[0077] The terms “proptosis” and “exophthalmos” (also known as exophthalmus, exophthalmia, or exorbitism) refer to the forward projection, displacement, bulging, or protrusion of an organ. As used herein, the terms refer to the forward projection, displacement, bulging, or protrusion of the eye anteriorly out of the orbit. Proptosis and exophthalmos are considered by some of skill in the art to have the same meaning and are often used interchangeably, while others attribute subtle differences to their meanings. Exophthalmos is used by some to refer to severe proptosis; or to refer to endocrine-related proptosis. Yet others use the term exophthalmos when describing proptosis associated with the eye, in, for example, subjects with TED (TAO or GO).

[0078] As used herein, the terms “proptosis” and “exophthalmos” are used interchangeably and refer to the forward projection, displacement, bulging, or protrusion of the eye anteriorly out of the orbit. Owing to the rigid bony structure of the orbit with only anterior opening for expansion, any increase in orbital soft tissue contents taking place from the side or from behind will displace the eyeball forward. Proptosis or exophthalmos can be the result of several disease processes including infections, inflammations, tumors, trauma, metastases, endocrine lesions,

vascular diseases and extra orbital lesions. TED (TAO or GO) is currently recognized as the most common cause of proptosis in adults. Exophthalmos can be either bilateral, as is often seen in TED (TAO or GO), or unilateral (as is often seen in an orbital tumor).

[0079] Measurement of the degree of exophthalmos can be performed using an exophthalmometer, an instrument used for measuring the degree of forward displacement of the eye. The device allows measurement of the forward distance of the lateral orbital rim to the front of the cornea.

[0080] Computed tomography (CT) scanning and Magnetic resonance imaging (MRI) may also be used in evaluating the degree of exophthalmos or proptosis. CT scanning is an excellent imaging modality for the diagnosis of TED (TAO or GO). In addition to allowing visualization of the enlarged extraocular muscles, CT scans provide the surgeon or clinician with depictions of the bony anatomy of the orbit when an orbital decompression is required. MRI, with its multi-planar and inherent contrast capabilities, provides excellent imaging of the orbital contents without the radiation exposure associated with CT scan studies. MRI provides better imaging of the optic nerve, orbital fat, and extraocular muscle, but CT scans provide better views of the bony architecture of the orbit.

[0081] Orbital ultrasonography can also be used for the diagnosis and evaluation of TED (TAO or GO), because it can be performed quickly and with a high degree of confidence. High reflectivity and enlargement of the extraocular muscles are assessed easily, and serial ultrasonographic examinations can also be used to assess progression or stability of the ophthalmopathy.

[0082] Based on the technologies currently available, or that will become available in the future, one of skill in the art would be capable of determining the best modality for diagnosing and evaluating the extent of proptosis or exophthalmos.

[0083] Although it is generally accepted that the normal range of proptosis is 12-21 mm, it must be noted that the value for a normal person varies by age, gender and race. For example, in normal adult white males, the average distance of globe protrusion is 16.5 mm, with the upper limit of normal at 21.7 mm. In adult African Americans it averages 18.2 mm, with an upper normal limit of 24.1 mm in males and 22.7 mm in females. In Mexican adults, males averaged 15.2 mm and females averaged 14.8 mm and in Iran, for the age group of 20-70 years, the average was 14.7 mm. In Taiwanese adults, comparing normal subjects to those with Graves' Ophthalmopathy, the normal group had an average reading of 13.9 mm versus 18.3 mm for the TED group. Although the average and upper limits for exophthalmos or proptosis

vary widely, it is accepted in the field that a difference greater than 2 mm between the eyes is significant and not normal.

[0084] One of skill in the art, for example an ophthalmologist, surgeon or other clinician skilled in the knowledge and treatment of eye disorders would know what a normal value of proptosis is based on the age, gender and race of the subject and have the ability to diagnose or evaluate the presence or absence of proptosis as well as track its progression.

Activity Measures or Assessments

[0085] Several classification systems have been conceived to assess the clinical manifestations of TED (TAO or GO). In 1969, Werner reported the NOSPECS Classification (No physical signs or symptoms, Only signs, Soft tissue involvement, Proptosis, Extraocular muscle signs, Corneal involvement, and Sight loss) (Werner, S. C. *American Journal of Ophthalmology*, 1969, 68, no. 4, 646-648.)

[0086] The modified NOSPECS was also published by Werner in 1977 and has been broadly used since then (Werner, S. C. *American Journal of Ophthalmology*, 1977, 83, no. 5, 725-727). This classification grades for clinical severity and does not provide a means of distinguishing active TED (inflammatory progressive) from inactive TED (noninflammatory stationary). Therefore, the indication for treatments used to be based exclusively in the severity of symptoms without consideration whether the disease was active or inactive. In 1989, Mounts et al. described the Clinical Activity Score (CAS) (Mounts et al., *British Journal of Ophthalmology*, 1989, 73, no. 8, 639-644) as a way of assessing the degree of active disease. This score, based on the classical signs of acute inflammation (pain, redness, swelling, and impaired function) was proposed as a clinical classification to discriminate easily between active and inactive disease and was modified in 1997 (Mounts et al., *Clinical Endocrinology*, 1997, 47, no. 1, 9-14). This protocol is further described below.

[0087] As used herein, the term CAS refers to the protocol described and scored as disclosed below. According to this protocol, one point is given for the presence of each of the parameters assessed in the list below. The sum of all points defines clinical activity and provides the CAS. For patients assessed for the first time only items 1-7 are scored. A CAS $\geq 3/7$ indicates active TED. For patients that are assessed for the second or subsequent time (typically, 1-3 months later), items 8-10 are also scored; and a CAS $\geq 4/10$ indicates active disease. A ten-item CAS scale exists as well, but in clinical trials, the 7-item scale is generally used, being more amenable to longitudinal studies involving multiple assessments.

[0088] The CAS consists of seven components:

1. spontaneous retrobulbar pain,
2. pain on attempted eye movements (upward, side-to-side, and downward gazes),
3. conjunctival redness,
4. redness of the eyelids,
5. chemosis (conjunctival swelling/edema),
6. swelling of the caruncle/plica, and
7. swelling of the eyelids.

[0089] Each component is scored as present (1 point) or absent (0 points). The score at each efficacy assessment is the sum of all items present; giving a range of 0-7, where 0 or 1 constitutes inactive disease and 7 severe active ophthalmopathy. A change of >2 points is considered clinically meaningful.

[0090] Item 1, spontaneous orbital pain could be a painful, or oppressive feeling on, or behind, the globe. This pain may be caused by the rise in intraocular pressure, when the orbital tissues volume increases through excess synthesis of extracellular matrix, fluid accumulation, and cellular infiltration and expansion. Item 2, gaze evoked orbital pain, could be pain in the eyes when looking, or attempting to look, up, down or sideways, i.e., pain with upward, downward, or lateral eye movement, or when attempting upward, downward, or lateral gaze. This kind of pain could arise from the stretching of the inflamed muscle(s), especially on attempted up-gaze. The 'stretching pain' cannot be provoked by digital pressing on the eyeball, as would be expected if it were a manifestation of the raised intraorbital pressure. Both kinds of pain can be reduced after anti-inflammatory treatment. These kinds of pain are therefore considered to be directly related to autoimmune inflammation in the orbit and thus useful in assessing TED activity.

[0091] Swelling in TED (TAO or GO) is seen as chemosis (edema of the conjunctiva) and swelling of the caruncle and/or plica semilunaris. Both are signs of TED activity. Swollen eyelids can be caused by edema, fat prolapse through the orbital septum, or fibrotic degeneration. In addition to swelling, other symptoms indicative of active TED include redness and/or pain of the conjunctiva, eyelid, caruncle and/or plica semilunaris.

[0092] Other grading systems have also been developed for the assessment of TED (TAO or GO). The VISA Classification (vision, inflammation, strabismus, and appearance) (Dolman, P. J., and Rootman, J., *Ophthalmic Plastic and Reconstructive Surgery*, 2006, 22, no. 5, 319-324 and Dolman, P. J., *Best Practice & Research Clinical Endocrinology & Metabolism*, 2012, 26, no. 3, 229-248) and the European Group of Graves'

Orbitopathy (EUGOGO) Classification (Bartalena, L., et al., *European Journal of Endocrinology*, 2008, 158, no. 3, 273-285) are two such examples. Both systems are grounded in the NO SPECS and CAS classifications and use indicators to assess the signs of activity and the degree of severity. More importantly, they allow the clinician to guide the treatment of the patient with GO. VISA is more commonly used in North America and Canada while EUGOGO is in Europe. Since the VISA and EUGOGO protocols are not interchangeable, only one of them should be employed as a reference in a specific patient.

[0093] In some embodiments, an improvement to one or more symptoms of TED may be determined using metric and protocols known in the art. For example, an improvement as to any of the following symptoms may optionally be determined as follows:

(a) Proptosis (sometimes be referred to as exophthalmos): an improvement may be determined or measured with an exophthalmometer. The measurement is in millimeters and is consistent with eyelid retraction, Von Graefe's lid lag and lid edema.

(b) Double vision: an improvement may be determined or measured using an eye tracker and/or MRI scan.

(c) Pain: an improvement may be determined or measured via a patient assessment using a grading scale (e.g., an ocular pain assessment survey, "OPAS"), the VRS pain scale, etc.

(d) Redness: an improvement may be determined or measured via clinical grading or color image analysis, e.g., using a 0-4 scale in 0.5-unit increments.

(e) Functional vision: an improvement may be determined or measured by a visual function assessment via reading and/or simulators.

Thyroid Eye Disease/Graves Ophthalmopathy Quality of Life (GO-QoL)

[0094] In addition to proptosis (or exophthalmos) and CAS, quality of life (QoL) may also be evaluated with the use of the Graves' ophthalmopathy quality of life (GO-QoL) questionnaire. This questionnaire is designed to determine the improved quality of life after treatment. In some embodiments, questionnaire may determine the decreased or lack of side effects after being treated with an antibody, or an antigen-binding fragment, according to the methods disclosed herein, as compared to treatment with glucocorticoids.

[0095] The GO-QoL questionnaire has two self-assessment subscales. The first relates to the impact of visual function on daily activities, while the second relates to the impact of self-perceived appearance. Each subscale has 8 questions which are answered with: (i) yes—very much so; (ii) yes—a little; or (iii) no—not at all. Each question is scored 0-2, respectively, and

the total raw score is then mathematically transformed to a 0-100 scale, where 0 represents the most negative impact on quality of life, and 100 represents no impact. A change of >8 points on the 0-100 scale is considered to be clinically meaningful. The combined score takes raw scores from both subscales and again transforms them to a single 0-100 scale.

Severity Measures

[0096] For lid aperture, the distance between the lid margins are measured (in mm) with the patient looking in the primary position, sitting relaxed, and with distant fixation.

[0097] For swelling of the eyelids, the measure/evaluation is either “absent/equivocal,” “moderate,” or “severe.”

[0098] Redness of the eyelids is either absent or present.

[0099] Redness of the conjunctivae is either absent or present.

[0100] Conjunctival edema is either absent or present.

[0101] Inflammation of the caruncle or plica is either absent or present.

[0102] Exophthalmos was measured in millimeter using the same Hertel exophthalmometer and same intercanthal distance for an individual patient.

[0103] Subjective diplopia is scored from 0 to 3 (0 = no diplopia; 1 = intermittent, i.e., diplopia in primary position of gaze, when tired or when first awakening; 2 = inconstant, i.e., diplopia at extremes of gaze; 3 = constant, i.e., continuous diplopia in primary or reading position).

[0104] For eye muscle involvement, the ductions are measured in degrees.

[0105] Corneal involvement is either absent/punctate or keratopathy/ulcer.

[0106] For optic nerve involvement, i.e., best-corrected visual acuity, color vision, optic disc, relative afferent pupillary defect, the condition is either absent or present. In addition, visual fields are checked if optic nerve compression was suspected.

Severity Classification

[0107] Sight-threatening thyroid eye disease: Patients with dysthyroid optic neuropathy (DON) and/or corneal breakdown. This category warranted immediate intervention.

[0108] Moderate-to-severe thyroid eye disease: Patients without sight-threatening disease whose eye disease had sufficient impact on daily life to justify the risks of immunosuppression (if active) or surgical intervention (if inactive). Patients with moderate-to-severe thyroid eye disease usually had any one or more of the following: lid retraction ≥ 2 mm, moderate or severe soft tissue involvement, exophthalmos ≥ 3 mm above normal for race and gender, inconstant or constant diplopia.

[0109] Mild thyroid eye disease: Patients whose features of thyroid eye disease have only a minor impact on daily life insufficient to justify immunosuppressive or surgical treatment. They usually have only one or more of the following: minor lid retraction (<2 mm), mild soft tissue involvement, exophthalmos <3 mm above normal for race and gender, transient or no diplopia, and corneal exposure responsive to lubricants.

Assessment of Gorman Grading of Diplopia

[0110] The Gorman assessment of subjective diplopia includes four categories: no diplopia (absent), diplopia when the patient is tired or awakening (intermittent), diplopia at extremes of gaze (inconstant), and continuous diplopia in the primary or reading position (constant). Patients are scored according to which grade of diplopia they are experiencing. An improvement of ≥ 1 grade is considered clinically meaningful.

[0111] Additional testing, including clinical trial protocols and criteria which can be performed to determine efficacy for the treatment of TED can be found in U.S. Pre-Grant Pub. No. 2019/0225696A1, which is hereby incorporated by reference in its entirety.

IGF1R Antibodies and Antigen-binding Fragments Thereof

[0112] The sequences of the heavy chains and light chains of examples of antibodies that may be used in the methods disclosed herein, each comprising three CDRs on the heavy chain and three CDRs on the light chain are provided below. The sequences of the CDRs, heavy chains, light chains as well as the sequences of the nucleic acid molecules encoding the CDRs, heavy chains and light chains of the antibodies are disclosed in the sequence listing. The CDRs of the antibody heavy chains may be alternatively referred to as CDRH1 (or HCDR1), CDRH2 (or HCDR2) and CDRH3 (or HCDR3), respectively. Similarly, the CDRs of the antibody light chains may be alternatively referred to as CDRL1 (or LCDR1), CDRL2 (or LCDR2) and CDRL3 (or LCDR3), respectively.

[0113] Variant antibodies are also included within the scope of the disclosure. Thus, variants of the sequences recited in the application are also included within the scope of the disclosure. Such variants include natural variants generated by somatic mutation *in vivo* during the immune response or *in vitro* upon culture of immortalized B cell clones. Alternatively, variants may arise due to the degeneracy of the genetic code or may be produced due to errors in transcription or translation.

[0114] Further variants of the antibody sequences having improved affinity and/or potency may be obtained using methods known in the art and are included within the scope of the disclosure. For example, amino acid substitutions may be used to obtain antibodies with further

improved affinity. Alternatively, codon optimization of the nucleotide sequence may be used to improve the efficiency of translation in expression systems for the production of the antibody. Further, polynucleotides comprising a sequence optimized for antibody specificity or neutralizing activity by the application of a directed evolution method to any of the nucleic acid sequences disclosed herein are also within the scope of the disclosure.

[0115] In one embodiment variant antibody sequences may share 70% or more (i.e., 75%, 80%, 85%, 90%, 95%, 97%, 98%, 99% or more) amino acid sequence identity with the sequences recited in the application. In some embodiments such sequence identity is calculated with regard to the full length of the reference sequence (i.e., the sequence recited in the application). In some further embodiments, percentage identity, as referred to herein, is as determined using BLAST version 2.1.3 using the default parameters specified by the NCBI (the National Center for Biotechnology Information; <http://www.ncbi.nlm.nih.gov/>) [Blosum 62 matrix; gap open penalty = 11 and gap extension penalty = 1].

[0116] Antibodies, or antigen-binding fragments thereof, used with the methods disclosed herein can be of any isotype (e.g., IgA, IgG, IgM; i.e., an α , γ or μ heavy chain). In one embodiment the antibody is IgG. Within the IgG isotype, antibodies may be IgG1, IgG2, IgG3 or IgG4 subclass. The antibodies may have a κ or a λ light chain.

[0117] The antibodies or antigen-binding antibody fragments used with the methods disclosed herein can be administered by any route known to one of skill in the art.

[0118] In some embodiments the disclosure provides an antibody, or an antigen-binding fragment, that specifically binds to a human IGF1R protein, comprising a) a heavy chain variable domain, wherein the heavy chain variable domain comprises a CDR1 region having a polypeptide sequence of SEQ ID NO: 3, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 3, a CDR2 region having a polypeptide sequence of SEQ ID NO: 4, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 4, and/or a CDR3 region having a polypeptide sequence of SEQ ID NO: 5, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 5.

[0119] In some embodiments, the antibody or antigen-binding antibody fragment comprises (alone or in addition to a heavy chain variable domain, e.g., as described above), a light chain variable domain, comprising a CDR1 region having a polypeptide sequence of SEQ ID NO: 7, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 7, a CDR2 region having a polypeptide sequence of SEQ ID NO: 8, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 8, and/or a CDR3 region having a polypeptide

sequence of SEQ ID NO: 9 or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 9.

[0120] In some aspects, the antibody or antigen-binding antibody fragment comprises a heavy chain variable domain comprising a CDR1 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 3, a heavy chain variable domain comprising a CDR2 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 4, and/or a heavy chain variable domain comprising a CDR3 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 5; and/or a light chain variable domain comprising a CDR1 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 7, a light chain variable domain comprising a CDR2 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 8, and/or a light chain variable domain comprising a CDR3 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 9.

[0121] In some embodiments, the antigen-binding fragment may comprise a Fab, a Fab', a F(ab')₂, a variable fragment (Fv), a triabody, a tetrabody, a minibody, a bispecific F(ab')₂, a trispecific F(ab')₂, a diabody, a bispecific diabody, a single chain variable fragment (scFv), a scFv-Fc, a Fab-Fc, a nanobody (VHH), or a bispecific scFv.

[0122] In some embodiments, an antibody or antigen-binding fragment according to any aspect described herein may comprise one or more of the putative CDR sequences shown in Table 1 (e.g., any selection or combination of these sequences). In some aspects, the antibody or antigen-binding antibody fragment may comprise one or more CDR sequences that share each at least 75, 80, 85, 90, 95, or 100% sequence identity with any of the sequences shown in Table 1. In some aspects, the antibody or antigen-binding fragment may comprise one or more CDR sequences that have 1, 2, 3, 4, or 5 amino acid substitutions, deletions, or insertions as compared to any of the sequences shown in Table 1. To be clear, an antibody or antigen-binding fragment may comprise any selection or combination of the sequences shown in Table 1 (e.g., SEQ ID NO: 3; SEQ ID NOs: 3, 36, and 39; or SEQ ID NOs: 3, 5, 9, 36, 38, and 39), optionally with one or more substitutions, insertions or deletions.

	Chothia	North	IMGT
Putative Heavy Chain CDR1	GFTFSSYGMH (SEQ ID NO: 3)	AASGFTFSSYGMH (SEQ ID NO: 34)	GFTFSSYG (SEQ ID NO: 35)
Putative Heavy Chain CDR2	IIWFDGSSTYYADSVRG (SEQ ID NO: 4)	IIWFDGSSTY (SEQ ID NO: 36)	IWFDGSST (SEQ ID NO: 37)
Putative Heavy Chain CDR3	ELGRRYFDL (SEQ ID NO: 5)	-	-
Putative Light Chain CDR1	RASQSVSSYLA (SEQ ID NO: 7)	-	QSVSSY (SEQ ID NO: 38)
Putative Light Chain CDR2	DASKRAT (SEQ ID NO: 8)	YDASKRAT (SEQ ID NO: 39)	DAS (SEQ ID NO: 40)
Putative Light Chain CDR3	QQRSKWPPWT (SEQ ID NO: 9)	-	-

Table 1. Putative anti-IGF1R CDR sequences that may be incorporated into any of the antibodies or antibody-binding fragments thereof, described herein.

Nucleic Acid Molecules Encoding IGF1R Antibodies and Antigen-binding Fragments Thereof

[0123] In some embodiments, the expression cassette (e.g., of any rAAV vector described herein) comprises a polynucleotide encoding one or more anti-IGF1R antibodies, or antigen-binding fragments thereof, described herein (e.g., capable of binding to a human IGF1R protein). In some embodiments, the polynucleotide comprises an ORF encoding any anti-IGF1R antibody or antigen-binding fragment described herein.

[0124] In some embodiments, the expression cassette provides expression of one or more anti-IGF1R antibodies or antigen-binding fragments in at least one eye, and/or lacrimal gland. In some embodiments, the expression cassette causes expression of one or more anti-IGF1R antibodies or antigen-binding fragments in at least one meibomian gland. In some embodiments, the expression cassette causes expression of one or more anti-IGF1R antibodies or antigen-binding fragment in the trabecular meshwork. In some embodiments, the expression cassette causes expression of one or more anti-IGF1R antibodies or antigen-binding fragments in the retina. As used herein, “subject” means any mammal, including mice, rabbits, non-human primates (NHP) and humans. In some embodiments, the subject is a human or NHP.

Moreover, “individual” or “patient” may be used interchangeably with “subject.” In some embodiments, expression of the one or more anti-IGF1R antibodies, or antigen-binding fragments thereof, is at any detectable level in the treated eye, whereas the one or more anti-IGF1R antibodies, or antigen-binding fragments thereof, may not be expressed in an untreated subject, or in the contralateral eye of a treated subject.

[0125] In some embodiments, the expression cassette comprises a polynucleotide encoding a protein that is capable of specifically binding to a human IGF1R protein. In some embodiments, the expression cassette comprises a polynucleotide comprising a nucleotide sequence encoding an amino acid sequence listed in Table 1. In some embodiments, the expression cassette comprises a polynucleotide comprising a nucleotide encoding an amino acid sequence identified in Table 1.

[0126] In some embodiments, the expression cassette comprises a polynucleotide comprising a nucleotide sequence encoding an antibody or an antigen-binding fragment having at least 80%, at least 85%, at least 90%, at least 95%, at least 96%, at least 97%, at least 98%, or at least 99% identity to any of the amino acid sequences set forth in SEQ ID NOs: 2-9 or 32-38. In some embodiments, the expression cassette comprises a polynucleotide comprising a nucleotide sequence encoding the amino acid sequence set forth in SEQ ID NOs: 2 or 6. In some embodiments, the expression cassette comprises a polynucleotide comprising a nucleotide sequence encoding a protein having at least 80%, at least 85%, at least 90%, at least 95%, at least 96%, at least 97%, at least 98%, or at least 99% identity to the amino acid sequences set forth in SEQ ID NOs: 2 or 6. In some embodiments, the expression cassette comprises a polynucleotide comprising a nucleotide sequence encoding the amino acid sequence set forth in one or more of SEQ ID NOs: 3-5, 7-9, or 34-40. In some embodiments, the expression cassette comprises a polynucleotide comprising a nucleotide sequence having at least 80%, at least 85%, at least 90%, at least 95%, at least 96%, at least 97%, at least 98%, or at least 99% identity with SEQ ID NOs: 21 or 22. In some embodiments, the expression cassette comprises a polynucleotide comprising a nucleotide sequence set forth in SEQ ID NO: 21. In some embodiments, the expression cassette comprises a polynucleotide comprising a nucleotide sequence set forth in SEQ ID NO: 22.

[0127] In some embodiments, the expression cassette comprises a polynucleotide comprising a nucleotide sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment, wherein the anti-IGF1R antibody, or an antigen-binding fragment, comprises or consists of an amino acid sequence having at least 80%, at least 85%, at least 90%, at least

95%, at least 96%, at least 97%, at least 98%, or at least 99% identity to SEQ ID NOs: 21 or 22.

[0128] Percent identity can be determined using any suitable method known in the art. For example, a method of determining percent identity of a nucleic acid sequence is genome sequencing. Methods for determining percent identity to a nucleic acid sequence or amino acid sequence are known to those of skill in the art.

Codon Optimization

[0129] In some embodiments, the expression cassette comprises a polynucleotide encoding an anti-IGF1R antibody or an antigen-binding fragment wherein the polynucleotide comprises or consists of a nucleotide sequence codon-optimized for expression in a target cell. In some embodiments, the target cell is a mammalian cell. In some embodiments, the target cell is a human cell, a murine cell, or a non-human primate (NHP) cell.

[0130] Codon optimization methods are known in the art and can be useful to achieve one or more desired results, e.g., to increase expression of a synthetic gene in a target cell. In some embodiments, an expression cassette comprises a nucleotide sequence that is sequence-optimized relative to a reference sequence using a method of sequence optimization. Methods of sequence optimization are known in the art, and include known sequence optimization tools, algorithms and services. Non-limiting examples include services from GeneArt (Life Technologies), DNA2.0 (Menlo Park CA), Geneious®, and GeneGPS® (Atum, Newark, CA).

[0131] In some embodiments, an expression cassette of the disclosure comprises a polynucleotide encoding an anti-IGF1R antibody, or an antigen-binding fragment, that is sequence-optimized relative to a reference sequence using a method of sequence optimization (e.g., GeneGPS®, e.g., Geneious®). In some embodiments, the reference sequence encoding the anti-IGF1R antibody or antigen-binding fragment is set forth in SEQ ID NO: 21 or 22. In some embodiments, the expression cassette comprises a polynucleotide encoding an anti-IGF1R antibody or antigen-binding fragment that is sequence-optimized relative to a reference sequence selected from SEQ ID NO: 21 or 22. In some embodiments, the reference sequence encoding the anti-IGF1R antibody or antigen-binding fragment is set forth in SEQ ID NO: 21. In some embodiments, the reference sequence encoding the anti-IGF1R antibody or antigen-binding fragment is set forth in SEQ ID NO: 22.

[0132] In some embodiments, the method of sequence optimization comprises a codon optimization algorithm described in US 7,561,972; US 7,561,973; US 8,126,653; and US 8,401,798, each of which is incorporated herein by reference in its entirety. In some

embodiments, the polynucleotide sequence is sequence-optimized based on codon usage bias in a host cell (e.g., mammalian cell, e.g., human cell, murine cell, non-human primate cell) relative to a reference sequence, using a method of sequence optimization known in the art (e.g., GeneGPS®, e.g., Geneious®). In some embodiments, the polynucleotide sequence comprises or consists of a nucleotide sequence that is codon-optimized relative to a reference sequence using a method of sequence optimization. In some embodiments, the polynucleotide sequence comprises or consists of a nucleotide sequence that is codon-optimized relative to a reference sequence for expression in a human host cell.

Expression Cassettes

[0133] The vectors of the disclosure comprise an expression cassette, i.e., a polynucleotide comprising at least one polynucleotide sequence encoding a protein of interest (e.g., an anti-IGF1R antibody or an anti-IGF1R antibody binding fragment, such as a heavy chain, light chain, or scFv, or any other binding construct described herein). In some embodiments the expression cassette encodes an anti-IGF1R antibody binding fragment. In some embodiments the expression cassette encodes an anti-IGF1R antibody binding fragment that is an scFv. In some embodiments, the expression cassette comprises other polynucleotide sequences, e.g., promoters, regulatory elements (e.g., one or more promoters), translation initiation sequences, coding sequences, and termination sequences.

[0134] In some embodiments, the expression cassette of the present disclosure comprises a polynucleotide sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment. In some embodiments, the expression cassette provides expression of an anti-IGF1R antibody, or an antigen-binding fragment, in at least one eye, the lacrimal gland, and/or the trabecular meshwork. In some embodiments, the eye, the lacrimal gland, the meibomian glands, and/or the trabecular meshwork to which a vector described herein is administered may express IGF1R at a lower abundance than in an eye, lacrimal gland, meibomian gland, and/or trabecular meshwork that has only endogenous (i.e., native) expression of IGF1R.

[0135] In some embodiments, an expression cassette provided herein is a bicistronic expression cassette configured to allow the simultaneous expression of two anti-IGF1R antibodies and/or antigen-binding fragments using a single mRNA transcript (e.g., via the incorporation of an internal ribosomal entry site, “IRES”). For example, a bicistronic expression cassette may encode a protein comprising both an anti-IGF1R antibody sequence and an anti-IGF1R antigen-binding fragment sequence (e.g., an scFv) selected from any of the sequences or embodiments described herein. In some embodiments, co-expression of two or

more different anti-IGF1R antibodies or antigen-binding fragments, in a cell of the eye, may provide synergistic therapeutic effects.

[0136] In some embodiments, the expression cassette of the present disclosure comprises a polynucleotide sequence encoding an anti-IGF1R scFv, comprising a light chain variable domain and a heavy chain variable domain of an immunoglobulin or a portion thereof. In some aspects, the light chain variable domain is N-terminal to the heavy chain variable domain, optionally separated by a polynucleotide sequence encoding a linker sequence. In other aspects, the heavy chain variable domain is N-terminal to the light chain variable domain, optionally separated by a polynucleotide sequence encoding a linker sequence. In some embodiments, the expression cassette provides expression of the scFv, in at least one eye, the lacrimal gland, and/or the trabecular meshwork. In some embodiments, the eye, the lacrimal gland, the meibomian glands, and/or the trabecular meshwork to which a vector described herein is administered may express IGF1R at a lower abundance than in an eye, lacrimal gland, meibomian gland, and/or trabecular meshwork that has only endogenous (*i.e.*, native) expression of IGF1R.

[0137] In some embodiments, an expression cassette provided herein is a bicistronic expression cassette configured to allow the simultaneous expression of two anti-IGF1R scFvs, each comprising a light chain variable domain and/or a heavy chain variable domain of an immunoglobulin or a portion thereof, using a single mRNA transcript (e.g., via the incorporation of an IRES). For example, a bicistronic expression cassette may encode a first protein comprising an anti-IGF1R scFv comprising the heavy chain variable domain of an immunoglobulin N-terminal to the light chain variable domain of an immunoglobulin, optionally separated by a polynucleotide sequence encoding a linker sequence; and a second protein comprising an anti-IGF1R scFv comprising the light chain variable domain of an immunoglobulin N-terminal to the heavy chain variable domain of an immunoglobulin, optionally separated by a polynucleotide sequence encoding a linker sequence. In some embodiments, co-expression of two or more different anti-IGF1R scFvs in a cell of the eye may provide synergistic therapeutic effects.

[0138] In some embodiments, the expression cassette of the present disclosure comprises a promoter. The term “promoter” as used herein refers to a DNA sequence that directs the binding of RNA polymerase and thereby promotes RNA synthesis, *i.e.*, a minimal sequence sufficient to direct transcription. Promoters and corresponding protein or polypeptide expression may be ubiquitous, meaning strongly active in a wide range of cells, tissues and species or cell-type specific, tissue-specific, or species specific. Promoters may be

“constitutive,” meaning continually active, or “inducible,” meaning the promoter can be activated or deactivated by the presence or absence of biotic or abiotic factors. Also included in the nucleic acid constructs or vectors of the disclosure are enhancer sequences that may or may not be contiguous with the promoter sequence. Enhancer sequences influence promoter-dependent gene expression and may be located in the 5′ or 3′ regions of the native gene.

[0139] Any suitable promoter region or promoter sequence therein can be used in the subject polynucleotide cassettes, so long as the promoter region promotes expression of a polynucleotide sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment, in at least one eye, the lacrimal gland, at least one meibomian gland, and/or the trabecular meshwork. In some embodiments, the promoter promotes expression of the gene in the mammalian eye, the lacrimal gland, the meibomian glands, and/or the trabecular meshwork. In some embodiments, the expression cassette comprises a cell-type specific promoter. The promoter may specifically promote transcription in the cells of the eye, the cells of the lacrimal gland, the cells of a meibomian gland, or the cells of the trabecular meshwork. For example, in some embodiments the promoter may comprise: a corneal stroma specific promoter such as a Keratocan promoter (Carlson EC, et. al., “In Vivo Gene Delivery and Visualization of Corneal Stromal Cells Using an Adenoviral Vector and Keratocyte-Specific Promoter.” *Investigative Ophthalmology & Visual Science* July 2004, Vol. 45, 2194-2200); a corneal epithelial/limbal stem cell specific promoter, such as a Keratin-12 or Pax-6 promoter (Wang, I., et al. “Cis-regulatory elements of the mouse Krt1.12 gene.” *Molecular vision*, 8, 94-101 (2002); Yoshihara, M., et al. “High-resolution promoter map of human limbal epithelial cells cultured with keratinocyte growth factor and rho kinase inhibitor.” *Sci Rep* 7, 2845 (2017); a Trabecular meshwork cell specific promoter such as a Chitinase 3-Like 1 Promoter (Liton PB, et al., “Specific targeting of gene expression to a subset of human trabecular meshwork cells using the chitinase 3-like 1 promoter.” *Invest Ophthalmol Vis Sci*. 2005 Jan;46(1):183-90); or an iris cell specific promoter such as a Phosphodiesterase 11A promoter (Janssen SF, et al., “Gene expression and functional annotation of the human ciliary body epithelia.” *PLoS One*. 2012;7(9):e44973).

[0140] In some embodiments, the promoter is a CAG promoter. In some embodiments, the promoter comprises a nucleotide sequence having at least 90%, 95%, 96%, 97%, 98%, or 99% identity with the nucleotide sequence set forth in SEQ ID NO: 10. In some embodiments, the promoter comprises SEQ ID NO: 10.

[0141] In some embodiments, the promoter is a CMV promoter. In some embodiments, the promoter comprises a nucleotide sequence having at least 95%, 96%, 97%, 98%, or 99%

identity to SEQ ID NO: 25. In some embodiments, the promoter comprises or consists of SEQ ID NO: 25.

[0142] In some embodiments, the enhancer is a CMV enhancer. In some embodiments, CMV enhancer comprises a nucleotide sequence having at least 95%, 96%, 97%, 98%, or 99% identity to SEQ ID NO: 26. In some embodiments, the enhancer comprises or consists of SEQ ID NO: 26.

[0143] In some embodiments, the expression cassette comprises a CMV enhancer and promoter. In some embodiments, the CMV enhancer and promoter comprises a nucleotide sequence having at least 95%, 96%, 97%, 98%, or 99% identity to SEQ ID NO: 27. In some embodiments, the enhancer and promoter comprises or consists of SEQ ID NO: 27.

[0144] The expression cassette may contain a polyadenylation (polyA) sequence. In some embodiments, the expression cassettes described herein comprise a transcription termination signal. Elements directing the efficient termination and polyadenylation of the heterologous nucleic acid transcripts increases heterologous gene expression. Transcription termination signals are generally found downstream of the polyadenylation signal. In some embodiments, vectors comprise a polyadenylation sequence 3' of a polynucleotide encoding a polypeptide to be expressed. The term "polyA site" or "polyA sequence" as used herein denotes a DNA sequence which directs both the termination and polyadenylation of the nascent RNA transcript by RNA polymerase II. Polyadenylation sequences can promote mRNA stability by addition of a polyA tail to the 3' end of the coding sequence and thus, contribute to increased translational efficiency. Cleavage and polyadenylation is directed by a poly(A) sequence in the RNA. The core poly(A) sequence for mammalian pre-mRNAs has two recognition elements flanking a cleavage-polyadenylation site. Typically, an almost invariant AAUAAA hexamer lies 20-50 nucleotides upstream of a more variable element rich in U or GU residues. Cleavage of the nascent transcript occurs between these two elements and is coupled to the addition of up to 250 adenosines to the 5' cleavage product. In some embodiments, the core poly(A) sequence is an ideal polyA sequence (*e.g.*, AATAAA, ATTAAA, AGTAAA). In some embodiments, the poly(A) sequence is an SV40 polyA sequence, a bovine growth hormone polyA sequence (BGH polyA), a rabbit β -globin polyA sequence (r β gpA), variants thereof, or another suitable heterologous or endogenous polyA sequence known in the art. In some embodiments, the expression cassette described herein comprises a polyA sequence. In some embodiments, the polyA sequence is a BGH polyA sequence. In some embodiments, the BGH polyA sequence comprises or consists of a nucleotide sequence having at least 95%, 96%, 97%,

98%, or 99% identity with SEQ ID NO: 28. In some embodiments, the BGH polyA sequence comprises or consists of SEQ ID NO: 28.

[0145] In some embodiments, the rAAV vectors of the disclosure comprise the Woodchuck Post-transcriptional Regulatory Element (WPRE). In some embodiments, the rAAV vector comprises a WPRE comprising SEQ ID NO: 29.

Recombinant Vectors

[0146] In some embodiments, expression cassettes are used to deliver an anti-IGF1R antibody, or an antigen-binding fragment, to at least one eye and/or lacrimal gland of a subject, *e.g.*, to treat an ocular disorder. In some embodiments, the subject expression cassettes are used to deliver an anti-IGF1R antibody, or an antigen-binding fragment, to at least one or more meibomian glands of a subject, *e.g.*, to treat an ocular disorder. Accordingly, in some embodiments, the composition that provides for the expression of an anti-IGF1R antibody, or an antigen-binding fragment, in at least one eye (or any cell or tissue) of a subject is a gene delivery vector, wherein the gene delivery vector comprises an expression cassette described herein.

[0147] In some embodiments, the gene delivery vector is an rAAV vector (*e.g.*, an rAAV virion). In some embodiments, the expression cassette is flanked on the 5' and 3' ends by functional AAV inverted terminal repeat (ITR) sequences. By “functional AAV ITR sequences” is meant that the ITR sequences function as intended for the rescue, replication and packaging of the AAV vector. Hence, AAV ITRs for use in the gene delivery vectors of the present disclosure need not have a wild-type nucleotide sequence, and may be altered by the insertion, deletion or substitution of nucleotides or the AAV ITRs may be derived from any of several AAV serotypes, *e.g.*, AAV1, AAV2, AAV3, AAV4, AAV5, AAV6, AAV7, AAV8, AAV9, or AAV10. In some embodiments, the AAV ITR is derived from AAV1. In some embodiments, the AAV ITR is derived from AAV2. In some embodiments, the AAV ITR is derived from AAV3. In some embodiments, the AAV ITR is derived from AAV4. In some embodiments, the AAV ITR is derived from AAV5. In some embodiments, the AAV ITR is derived from AAV6. In some embodiments, the AAV ITR is derived from AAV7. In some embodiments, the AAV ITR is derived from AAV8. In some embodiments, the AAV ITR is derived from AAV9. In some embodiments, the AAV ITR is derived from AAV10. In some embodiments, the rAAV vector has the wild-type rep and/or cap genes deleted in whole or in part. In some embodiments, the rAAV vector retains functional flanking ITR sequences. In some embodiments, the AAV ITR is one identified in [Table 2](#).

Name	Nucleotide Sequence	SEQ ID NO
AAV2 5' ITR	GCGCGCTCGCTCGCTCACTGAGGCCGCCCCGGGCAAAGCCCG GGCGTCGGGCGACCTTTGGTCGCCCGGCCTCAGTGAGCGAG CGAGCGCGCAGAGAGGGAGTGGCCAACCTCCATCACTAGGG GTTCTTGTAGTTAATGATTAAC	30
AAV2 3' ITR	GTTAATCATTAACTACAAGGAACCCCTAGTGATGGAGTTGG CCACTCCCTCTCTGCGCGCTCGCTCGCTCACTGAGGCCGGGC GACCAAAGGTCGCCCCGACGCCCGGGCTTTGCCCGGGCGGCC TCAGTGAGCGAGCGAGCGCGC	31
AAV5 5' ITR	CTCTCCCCCTGTGCGGTTGCTCGCTCGCTGGCTCGTTTGG GGGGGTGGCAGCTCAAAGAGCTGCCAGA	32
AAV5 3' ITR	CGACGGCCCTCTGGCCGTGCCCCCACAACGAGCCAGCGA GCGAGCGAACGCGACAGGGGGGAGAGTGCCACACTCTCAA GCAAGGGGGTTTTGTAAGCAGTGATGTCATAATGATG	33

Table 2. Exemplary ITR Sequences

[0148] In some embodiments, the rAAV vector comprises an AAV capsid derived from any adeno-associated virus serotype known in the art, or prospectively discovered, including without limitation, AAV1, AAV2, AAV3, AAV4, AAV5, AAV6, AAV7, AAV8, AAV9, AAV10, etc. For example, the AAV capsid may be a wild-type (or “native”) capsid. In some embodiments, the rAAV vector comprises an AAV capsid derived from AAV1. In some embodiments, the rAAV vector comprises an AAV capsid derived from AAV2. In some embodiments, the rAAV vector comprises an AAV capsid derived from AAV3. In some embodiments, the rAAV vector comprises an AAV capsid derived from AAV4. In some embodiments, the rAAV vector comprises an AAV capsid derived from AAV5. In some embodiments, the rAAV vector comprises an AAV capsid derived from AAV6. In some embodiments, the rAAV vector comprises an AAV capsid derived from AAV7. In some embodiments, the rAAV vector comprises an AAV capsid derived from AAV8. In some embodiments, the rAAV vector comprises an AAV capsid derived from AAV9. In some embodiments, the rAAV vector comprises an AAV capsid derived from AAV10. AAV capsids of particular interest include AAV2, AAV5, AAV8, and AAV9 (see Table 3). However, as with the ITRs, the polynucleotide encoding the capsid need not have a wild-type nucleotide sequence, but rather may be altered by the insertion, deletion, or substitution of nucleotides in the VP1, VP2 or VP3 sequence, so long as the polynucleotide encoding the capsid is able to transduce cells of the eye and/or lacrimal glands. Put another way, the AAV capsid may be a variant AAV capsid. In some embodiments, the rAAV vector is a “pseudotyped” AAV vector created by using the capsid (cap) gene of one AAV serotype and the rep gene from a different AAV serotype, *e.g.*, a pseudotyped AAV2 created by using rep from AAV2 and cap from

AAV1, AAV3, AAV4, AAV5, AAV6, AAV7, AAV8, or AAV9. For example, the rAAV vector may be rAAV2/1, rAAV2/3, rAAV2/4, rAAV2/5, rAAV2/6, rAAV2/7, rAAV2/8, rAAV2/9, etc., where the “X/Y” label at the end of each of the aforementioned rAAVs represents the serotype of the rep protein (X) and the serotype of the capsid (Y), used in the generation of each respective rAAV. In some embodiments, the rAAV is rAAV2/1. In some embodiments, the rAAV is rAAV2/3. In some embodiments, the rAAV is rAAV2/4. In some embodiments, the rAAV is rAAV2/5. In some embodiments, the rAAV is rAAV2/6. In some embodiments, the rAAV is rAAV2/7. In some embodiments, the rAAV is rAAV2/8. In some embodiments, the rAAV is rAAV2/9. In some aspects, the ITRs may also be selected from a particular serotype (e.g., ITRs from AAV2, AAV5, or any other AAV, may be incorporated into any of the plasmids or other vectors described herein.)

[0149] In some embodiments, the rAAV is replication defective, in that the rAAV vector cannot independently further replicate and package its genome. For example, with such replication defective embodiment, cells of a subject’s eye and/or lacrimal glands are transduced with a replication-deficient rAAV vector configured to express an anti-IGF1R antibody or an IGF1R antigen-binding fragment transgene in the transduced tissue, however, due to the fact that the rAAV vector lacks AAV rep and cap genes and accessory function genes, the rAAV is not able to replicate.

[0150] In some embodiments, rAAV vectors of the present disclosure encapsulating the expression cassettes as described herein can be produced using helper-free production. rAAVs normally require production by using components from a live helper virus, such as adenovirus, in a host cell for packaging of infectious rAAV vectors. rAAV helper-free production systems allow the production of infectious rAAV vectors without the use of a live helper virus. In the helper-free system, a host packaging cell line is co-transfected with three plasmids. A first plasmid contains adenovirus genes (*i.e.*, E2A, E4, and VA RNA genes) encoding products needed for the packaging of rAAV vectors. A second plasmid contains the required AAV genes (*i.e.*, rep and cap genes). A third plasmid contains the expression cassette, e.g., polynucleotide sequence encoding the anti-IGF1R antibody or IGF1R antigen-binding fragment and a promoter flanked by ITRs. A host packaging cell line can be, for example, AAV-293 host cells. Suitable host cells contain additional components required for packaging infectious rAAV vectors that are not supplied by the plasmids. In some embodiments, the cap genes can encode, for example, any of the AAV capsid proteins described herein. In some embodiments, the promoter is a promoter sequence as described herein. In some embodiments, the promoter sequence is a CAG sequence. In some embodiments, the polynucleotide encodes an anti-IGF1R

antibody or an IGF1R antigen-binding fragment which shares at least 80%, at least 85%, at least 90%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or 100% identity with SEQ ID NOs: 2 or 6. In some embodiments, the polynucleotide encodes a protein which shares at least 80%, at least 85%, at least 90%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or 100% identity with one or more of the CDRs identified as SEQ ID NOs: 3-5, 7-9, and/or 31-37.

[0151] AAV serotypes shown to infect the eye and/or lacrimal gland include AAV2, AAV5, AAV 5w8, and AAV9 (Rocha et al., *supra*). Exemplary amino acid and nucleotide sequences for several AAV capsid proteins are identified in Table 3.

[0152] In some embodiments, the AAV capsid protein shares at least 95%, 98%, or 100% identity to the AAV2 VP1 protein (SEQ ID NO: 11). In some embodiments, the polynucleotide sequence encoding the AAV2 VP1 protein shares at least 95%, 98%, or 100% identity with SEQ ID NO: 12. In some embodiments, the AAV capsid protein shares at least 95%, 98%, or 100% identity to the AAV2 VP3 protein (SEQ ID NO: 13). In some embodiments, the polynucleotide sequence encoding the AAV2 VP3 protein shares at least 95%, 98%, or 100% identity with SEQ ID NO: 14. In some embodiments, the AAV capsid protein shares at least 95%, 98%, or 100% identity to the AAV5 capsid protein (SEQ ID NO: 15). In some embodiments, the polynucleotide sequence encoding the AAV5 capsid protein shares at least 95%, 98%, or 100% identity with SEQ ID NO: 16. In some embodiments, the AAV capsid protein shares at least 95%, 98%, or 100% identity to the AAV8 capsid protein (SEQ ID NO: 17). In some embodiments, the polynucleotide sequence encoding the AAV8 capsid protein shares at least 95%, 98%, or 100% identity with SEQ ID NO: 18. In some embodiments, the AAV capsid protein shares at least 95%, 98%, or 100% identity to the AAV9 capsid protein (SEQ ID NO: 19). In some embodiments, the polynucleotide sequence encoding the AAV9 capsid protein shares at least 95%, 98%, or 100% identity with SEQ ID NO: 20.

Capsid Sequence	Protein	Nucleotide (Open Reading Frame)
AAV2 VP1	SEQ ID NO: 11	SEQ ID NO: 12
AAV2 VP3	SEQ ID NO: 13	SEQ ID NO: 14
AAV5 capsid protein	SEQ ID NO: 15	SEQ ID NO: 16
AAV8 capsid protein	SEQ ID NO: 17	SEQ ID NO: 18
AAV9 capsid protein	SEQ ID NO: 19	SEQ ID NO: 20

Table 3: AAV Capsid Sequences

Exemplary rAAV Vectors

[0153] In some embodiments, the rAAV vector comprises an AAV capsid. In some embodiments, an rAAV vector described herein comprises an expression cassette comprising

a polynucleotide comprising a nucleotide sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment. In some embodiments, an rAAV vector described herein comprises an expression cassette comprising a polynucleotide comprising a nucleotide sequence encoding an anti-IGF1R scFv (e.g., comprising SEQ ID NOs: 2 and/or 6). In some embodiments, an rAAV vector described herein comprises a bicistronic expression cassette comprising a polynucleotide comprising a nucleotide sequence encoding two anti-IGF1R antibodies, two anti-IGF1R antigen-binding fragments; or an anti-IGF1R antibody and an antigen-binding fragment. In some embodiments, the polynucleotide is operably linked to a promoter. In some embodiments, the rAAV vector comprises an AAV capsid and the expression cassette, wherein the expression cassette comprises a polynucleotide operatively linked to a promoter, wherein the polynucleotide comprises a nucleotide sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment. In some embodiments, the promoter is any described herein or known in the art. In some embodiments, the promoter is a CAG promoter. In some embodiments, the promoter is a CMV promoter. In some embodiments, the expression cassette further comprises a 5' ITR and/or a 3' ITR. In some embodiments, the 5' ITR is an AAV2 5' ITR. In some embodiments, the 3' ITR is a AAV2 3' ITR. In some embodiments, the expression cassette comprises a nucleotide sequence comprising from 5' to 3': a 5' ITR (e.g., an AAV2 5' ITR), a promoter (e.g., a CMV promoter), a 5' untranslated region (5' UTR), a polynucleotide sequence comprising a nucleotide sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment, a polyA sequence, and a 3' ITR (e.g., an AAV2 3' ITR). In some embodiments, the expression cassette comprises a nucleotide sequence comprising from 5' to 3': a 5' ITR (e.g., an AAV2 5' ITR), an enhancer (e.g., a CMV enhancer), a promoter (e.g., a CMV promoter), a 5' UTR, a polynucleotide sequence comprising a nucleotide sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment, a WPRE sequence, a polyA sequence, and a 3' ITR (e.g., an AAV2 3' ITR). In some embodiments, the 5' UTR comprises a Kozak sequence positioned immediately upstream of and adjacent to an initiation codon in the nucleotide sequence encoding the anti-IGF1R antibody, or the antigen-binding fragment.

[0154] In some embodiments, the rAAV vector comprises an AAV capsid and an expression cassette comprising a polynucleotide operatively linked to a promoter, wherein the polynucleotide comprises a nucleotide sequence encoding a humanized anti-IGF1R antibody, or an antigen-binding fragment described herein. In some embodiments, the rAAV vector comprises an AAV capsid and an expression cassette comprising a polynucleotide operatively linked to a CAG promoter or a CMV promoter, wherein the polynucleotide comprises a nucleotide sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment

described herein. In some embodiments, the polynucleotide comprises from 5' to 3' (i) a nucleotide sequence encoding a signal peptide, and (ii) a nucleotides sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment described herein.

[0155] In some embodiments, the rAAV vector comprises an AAV capsid and an expression cassette comprising a polynucleotide operatively linked to a promoter (e.g., a CAG promoter or CMV promoter), wherein the polynucleotide comprises from 5' to 3' (i) a nucleotide sequence encoding a signal peptide, and (ii) a nucleotide sequence encoding an anti-IGF1R antibody or antigen-binding fragment, wherein the anti-IGF1R antibody or antigen-binding fragment comprises or consists of an amino acid sequence that shares at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, or at least 99% identity to SEQ ID NOs: 2 and/or 6. In some embodiments, the anti-IGF1R antibody, or the antigen-binding fragment comprises or consists of the amino acid sequence set forth in SEQ ID NOs: 2 or 6.

[0156] In some embodiments, the rAAV vector comprises an AAV2 capsid and an expression cassette comprising a polynucleotide operatively linked to a promoter, wherein the polynucleotide comprises a nucleotide sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment described herein. In some embodiments, the rAAV vector comprises an AAV2 capsid and an expression cassette comprising a polynucleotide operatively linked to a CAG promoter or a CMV promoter, wherein the polynucleotide comprises from 5' to 3' (i) a nucleotide sequence encoding a signal peptide, and (ii) a nucleotide sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment described herein.

[0157] In some embodiments, the rAAV vector comprises an AAV2 capsid and an expression cassette comprising a polynucleotide operatively linked to a promoter (e.g., a CAG promoter or CMV promoter), wherein the polynucleotide comprises from 5' to 3' (i) a nucleotide sequence encoding a signal peptide, and (ii) a nucleotide sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment, wherein the anti-IGF1R antibody, or the antigen-binding fragment comprises or consists of an amino acid sequence that shares at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, or at least 99% identity to SEQ ID NOs: 2 or 6. In some embodiments, the anti-IGF1R antibody, or an antigen-binding fragment comprises or consists of an amino acid sequence set forth in SEQ ID NOs: 3-5, 7-9, or 34-40. In some embodiments, the rAAV vector comprises an AAV2 capsid and an expression cassette comprising a polynucleotide operatively linked to a promoter (e.g.,

a CAG promoter or CMV promoter), wherein the polynucleotide comprises from 5' to 3' (i) a nucleotide sequence encoding a signal peptide, and (ii) a nucleotide sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment, wherein the anti-IGF1R antibody, or the antigen-binding fragment comprises or consists of an amino acid sequence that shares at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, or at least 99% identity to SEQ ID NOs: 2 or 6.

[0158] In some embodiments, the rAAV vector comprises an AAV9 capsid and an expression cassette comprising a polynucleotide operatively linked to a promoter (e.g., a CAG promoter or CMV promoter), wherein the polynucleotide comprises from 5' to 3' (i) a nucleotide sequence encoding a signal peptide, and (ii) a nucleotide sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment, wherein the anti-IGF1R antibody, or the antigen-binding fragment comprises or consists of an amino acid sequence that shares at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, or at least 99% identity to SEQ ID NOs: 2 or 6. In some embodiments, the anti-IGF1R antibody, or an antigen-binding fragment comprises or consists of an amino acid sequence set forth in SEQ ID NOs: 3-5, 7-9, or 34-40. In some embodiments, the rAAV vector comprises an AAV9 capsid and an expression cassette comprising a polynucleotide operatively linked to a promoter (e.g., a CAG promoter or CMV promoter), wherein the polynucleotide comprises from 5' to 3' (i) a nucleotide sequence encoding a signal peptide, and (ii) a nucleotide sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment, wherein the anti-IGF1R antibody, or the antigen-binding fragment comprises or consists of an amino acid sequence that shares at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, or at least 99% identity to SEQ ID NOs: 2 and/or 6.

[0159] In some embodiments, the rAAV vector may comprise a nucleotide sequence encoding an anti-IGF1R scFv, wherein the anti-IGF1R scFv comprises a heavy chain variable domain of an immunoglobulin and a light chain variable domain of an immunoglobulin. In some embodiments, the heavy chain variable domain comprises or consists of an amino acid sequence that shares at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, or at least 99% identity to SEQ ID NOs: 2; and/or the light chain variable domain comprises or consists of an amino acid sequence that shares at least 85%, at least 86%,

at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, or at least 99% identity to SEQ ID NO: 6. In some embodiments, the nucleotide sequence encoding the heavy chain variable domain is positioned 5' to the nucleotide sequence encoding the light chain variable domain; in others, this orientation may be reversed. In some embodiments, the anti-IGF1R scFv may further comprise a nucleotide sequence encoding a signal peptide from 5' to the nucleotide sequence encoding the heavy chain variable domain or the light chain variable domain. In some embodiments, the rAAV vector comprises an AAV2, an AAV9 capsid, or a capsid derived from any other AAV serotype.

[0160] In some embodiments, the promoter is any described herein or known in the art. In some embodiments, the promoter is a CAG promoter. In some embodiments, the promoter is a CMV promoter. In some embodiments, the expression cassette further comprises a 5' ITR and/or a 3' ITR. In some embodiments, the 5' ITR is an AAV2 5' ITR. In some embodiments, the 3' ITR is a AAV2 3' ITR. In some embodiments, the expression cassette comprises a nucleotide sequence comprising from 5' to 3': a 5' ITR (*e.g.*, an AAV2 5' ITR), a promoter (*e.g.*, a CMV promoter), a 5' untranslated region (5' UTR), a polynucleotide sequence comprising a nucleotide sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment, a polyA sequence, and a 3' ITR (*e.g.*, an AAV2 3' ITR). In some embodiments, the expression cassette comprises a nucleotide sequence comprising from 5' to 3': a 5' ITR (*e.g.*, an AAV2 5' ITR), an enhancer (*e.g.*, a CMV enhancer), a promoter (*e.g.*, a CMV promoter), a 5' UTR, a polynucleotide sequence comprising a nucleotide sequence encoding an anti-IGF1R antibody, or an antigen-binding fragment, a WPRE sequence, a polyA sequence, and a 3' ITR (*e.g.*, an AAV2 3' ITR). In some embodiments, the 5' UTR comprises a Kozak sequence positioned immediately upstream of and adjacent to an initiation codon in the nucleotide sequence encoding the anti-IGF1R antibody, or an antigen-binding fragment.

Methods of Use

[0161] Methods and compositions described herein can be used to treat ocular conditions and reduce the associated symptoms of the ocular conditions. The terms "treatment", "treating" and the like are used herein to generally mean obtaining a desired pharmacologic and/or physiologic effect. The effect may be prophylactic in terms of completely or partially preventing a disease or symptom thereof, *e.g.*, reducing the likelihood that the disease or symptom thereof occurs in the subject, delaying the onset or progression of a disease or a symptom in a subject and/or may be therapeutic in terms of a partial or complete cure for a

disease and/or adverse effect attributable to the disease. “Treatment” as used herein covers any treatment of a disease in a mammal, and includes, without limitation: (a) inhibiting progress of the disease; (b) alleviating, reducing, or reducing an increase in one or more symptoms of the disease; (c) alleviating, reducing, or reducing an increase in one or more signs of the disease; (d) causing regression of the disease. The therapeutic agent may be administered before, during or after the onset of disease or injury. The treatment of ongoing disease, where the treatment stabilizes or reduces the undesirable clinical symptoms of the patient, is of particular interest. Such treatment is desirably performed prior to complete loss of function in the affected tissues. The subject therapy will desirably be administered during the symptomatic stage of the disease, and in some cases after the symptomatic stage of the disease.

[0162] As used herein, “administer,” “administering,” “administration” and the like refers to providing a substance (*e.g.*, a vector configured to express an anti-IGF1R antibody, or an antigen-binding fragment; or an anti-IGF1R antibody, or an antigen-binding fragment) to a subject in a manner that is pharmacologically useful (*e.g.*, to treat a disease, disorder, or condition in the subject).

[0163] Thus, in some embodiments, provided herein are methods of treating an ocular condition in a subject, the method comprising administering to the subject an rAAV vector provided herein or a pharmaceutical composition provided herein. As herein, the terms “subject” and “patient” are used interchangeably to refer to a mammal, including, but not limited to, human and non-human primates, including simians and humans; mammalian sport animals (*e.g.*, horses); mammalian farm animals (*e.g.*, sheep, goats, etc.); mammalian pets (dogs, cats, etc.); and rodents (*e.g.*, mice, rats, etc.). A subject can have, for example, any of the symptoms or ocular disorders described herein.

[0164] In some embodiments, the subject is a human adult, *e.g.*, a human adult over the age of 40 or a human adult 45 to 55 years of age. In some embodiments, the subject is a human adult over the age of 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, or 90.

[0165] In some embodiments, the ocular condition treated in accordance with the methods described herein is TED. In some embodiments, the ocular disease, disorder, or condition comprises: a) hypothyroid associated visual disturbance, b) a reduction in visual acuity due to lid retraction, c) lid wiper epitheliopathy, d) dry eye, e) aqueous deficiency, f) double vision, g) compressive optic neuropathy, h) ocular pain, i) proptosis, and/or j) diplopia. In some embodiments, the ocular disease, disorder, or condition is treated by administering 10^8 to 10^{14} vector genomes of any of the vectors described herein to one or more tissues of the subject

(e.g., to at least one eye, or to an accessory lacrimal gland or the trabecular meshwork of the subject).

[0166] In some embodiments, a method of treatment described herein comprises further administering to the subject one or more additional therapeutic agents. In some embodiments, the additional therapeutic agent is an agent that increases tear production. In some embodiments, the additional therapeutic agent that increases tear production is a cholinergic agonist. In some embodiments, the additional therapeutic agent is another AAV-based gene therapy construct.

[0167] In some embodiments, the treatment that increases tear production comprises administering an effective amount of a nicotinic acetylcholine receptor (nAChR) agonist, or a pharmaceutically acceptable salt thereof. In some embodiments, the nAChR agonist is one described herein. nAChRs are a class of pentameric ligand-gated ion channels that have high affinity and selectivity for both nicotine and acetylcholine (which resembles nicotine in its protonated form) and comprise combinations of alpha and beta subunits. Examples of nAChR subtypes include, but are not limited to, alpha3beta4, alpha4beta2, alpha3alpha5beta4, and alpha4alpha6beta2. An nAChR agonist may be characterized as a full or partial agonist as determined by its ability to activate a given receptor to produce a response as compared to the response at that receptor for acetylcholine (ACh). In general, a nAChR agonist is a full agonist if it evokes a response upon binding to a given receptor that is equal or greater to that of ACh. A nAChR agonist is a partial agonist if it evokes a lower response upon binding to the receptor as compared to the response generated from ACh.

[0168] nAChR agonist response, from which receptor activation can be determined can, for example, be generated using an appropriate cell-based assay. Cells designed to express a particular nAChR receptor subtype and generate an electrical current response when bound to and activated by a nAChR agonist can be used to characterize the agonist profile of a compound and the amount of receptor activation thus determined.

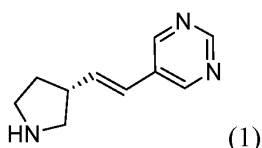
[0169] In some embodiments, the nAChR agonist, or a pharmaceutically acceptable salt thereof, selectively binds to at least one of the nAChR subtypes selected from alpha3beta4, alpha3alpha5beta4, alpha4beta2, and alpha4alpha6beta2. As used herein, “selectively binds” or “is selective for” means that a compound has a higher affinity for the nAChR subtype and/or a lower half-maximal effective concentration (EC50) for that nAChR subtype for at least one reference nAChR subtype. Selectivity may be associated with at least a 5-fold affinity difference in EC50 value, at least a 10-fold affinity difference in EC50 value, at least a 20-fold affinity difference in EC50 value, or at least a 50-fold affinity difference in EC50 value. In

some embodiments, the nAChR agonist, or a pharmaceutically acceptable salt thereof, selectively binds to nAChR subtype $\alpha 3\beta 4$. In some embodiments, the nAChR agonist, or a pharmaceutically acceptable salt thereof, selectively binds to nAChR subtype $\alpha 3\alpha 5\beta 4$. In some embodiments, the nAChR agonist, or a pharmaceutically acceptable salt thereof, selectively binds to nAChR subtype $\alpha 4\beta 2$. In some embodiments, the nAChR agonist, or a pharmaceutically acceptable salt thereof, selectively binds to nAChR subtype $\alpha 4\alpha 6\beta 2$. In some embodiments, the nAChR agonist, or a pharmaceutically acceptable salt thereof, selectively binds to nAChR subtype $\alpha 7$. In some embodiments, the nAChR agonist, or a pharmaceutically acceptable salt thereof, does not selectively bind to nAChR subtype $\alpha 7$.

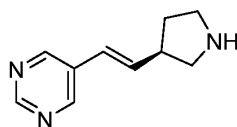
[0170] The nAChR agonists contemplated in this disclosure include varenicline, a pharmaceutically acceptable salt thereof, and compound 1, or a pharmaceutically acceptable salt thereof. In some embodiments the nAChR agonist is not varenicline.

[0171] Varenicline is characterized as a full agonist of the nAChR subtype $\alpha 7$ and a partial agonist of subtypes $\alpha 3\beta 4$, $\alpha 4\beta 2$, $\alpha 6\beta 2$, $\alpha 3\alpha 5\beta 4$, and $\alpha 4\alpha 6\beta 2$. In some embodiments, the nAChR agonist is varenicline, or a pharmaceutically acceptable salt thereof. Pharmaceutically acceptable salts of varenicline include varenicline tartrate. Additional related information for varenicline may be found in, for example, U.S. Patent 6,951,938, U.S. Patent 6,890,927, U.S. Patent 7,265,119, U.S. Patent 9,504,644, U.S. Patent 9,504,645, U.S. Patent 9,532,944, U.S. Patent 9,597,284, U.S. Patent 10,456,386, U.S. Patent 11,224,598, and U.S. Application Publication 2022/0233528.

[0172] Compound 1, as recited herein, refers to the structure:



[0173] An alternative structural representation of compound 1 is shown here:



[0174] Compound 1 may be also referred to by its chemical name. For instance, compound 1 is also referred to as (R)-5-((E)-2-pyrrolidin-3-ylvinyl)pyrimidine, or variations thereof including simpanicline, 5-{(E)-2-[(3R)-pyrrolidin-3-yl]vinyl}pyrimidine and (R,E)-5-((2-pyrrolidine-3-yl)vinyl)pyrimidine.

[0175] Compound 1 is a full agonist of nAChR subtypes $\alpha 4\beta 2$, $\alpha 3\beta 4$, $\alpha 3\alpha 5\beta 4$, and $\alpha 4\alpha 6\beta 2$. Compound 1 is a full agonist of nAChR subtypes $\alpha 4\beta 2$, and $\alpha 3\beta 4$.

[0176] Compound 1 is a partial agonist of subtype $\alpha 3\beta 2$.

[0177] Compound 1 is a weak partial agonist of subtype $\alpha 7$. In one example, a 300 micromolar concentration of compound 1 citrate evoked only 25% of the maximal ACh-evoked current.

[0178] In some embodiments, the nAChR agonist may be compound 1, or a pharmaceutically acceptable salt thereof. Pharmaceutically acceptable salts of compound 1 include galactarate (e.g., hemi-galactarate dihydrate) and citrate (e.g., mono-citrate). Patent related information for compound 1 may be found in *e.g.*, U.S. Patent 7,098,331, U.S. Patent 7,714,001, U.S. Patent 8,063,068, U.S. Patent 8,067,443, U.S. Patent 8,604,191, U.S. Patent 9,145,396, U.S. Patent 9,981,949, U.S. Patent 8,633,222, U.S. Patent 8,153,821, U.S. Patent 8,633,227, U.S. Patent 10,709,707, U.S. Patent Application Publication 2020-0345734, and PCT publication WO 2017/177024.

[0179] In some embodiments, the nAChR agonist is (R)-5-((E)-2-pyrrolidin-3-ylvinyl)pyrimidine, or a pharmaceutically acceptable salt thereof. In some embodiments, the nAChR agonist is (R)-5-((E)-2-pyrrolidin-3-ylvinyl)pyrimidine hemigalactarate dihydrate. In some embodiments, the nAChR agonist is (R)-5-((E)-2-pyrrolidin-3-ylvinyl)pyrimidine mono-citrate.

[0180] In some embodiments, the nAChR agonist increases tear production in a subject where tear secretion is compromised.

Methods of Treatment

[0181] In some embodiments, the disclosure provides a method of treating a subject with an ocular disease or disorder, comprising administering to the subject a vector (e.g., an rAAV) comprising a nucleotide encoding an anti-IGF1R antibody or antigen-binding fragment described herein.

[0182] In some embodiments, the disclosure provides rAAV vectors for use in a method of treating an ocular condition in a subject in need thereof, the method comprising administering a recombinant adeno-associated virus (rAAV) vector, the rAAV vector comprising an AAV capsid and an expression cassette comprising a polynucleotide encoding an anti-IGF1R antibody or antigen-binding fragment operatively linked to a promoter, or an anti-IGF1R antibody or antigen-binding fragment, to a tear film or an eye tissue of at least one

eye of the subject (“eye tissue” in the context of the present disclosure includes tissue associated with the anatomy or functioning of the eye, e.g., tissue of the orbit, such as the extraorbital fat pads, and/or tissue of glands associated with the eye). For example, administration to a least one eye tissue may include administration to at least one lacrimal gland (e.g., a main or an accessory lacrimal gland), or to at least one meibomian gland, and/or to the subject’s trabecular meshwork. In some embodiments, the rAAV vector is administered to a plurality of lacrimal glands of a subject. In some embodiments, the rAAV vector comprises an expression cassette encoding an anti-IGF1R antibody or an anti-IGF1R antigen-binding fragment.

[0183] Administration to at least one eye may be accomplished via topical administration, by injection, or by any other mode of administering a pharmaceutical agent to the eye, or any individual tissue(s) or associated glands, described herein or known in the art. For example, a pharmaceutical composition comprising any rAAV vector described herein, any anti-IGF1R antibody or antigen-binding fragment described herein, or a plasmid (or other) vector configured to express any anti-IGF1R antibody or antigen-binding fragment described herein, may be topically administered as a liquid formulation to the eye or to at least one tissue or associated gland thereof, by the subject or by a medical professional. In other embodiments, a pharmaceutical composition comprising any rAAV vector described herein, any plasmid (or other) vector configured to express an anti-IGF1R antibody or antigen-binding fragment described herein, or any anti-IGF1R antibody or antigen-binding fragment described herein, may be injected into a main or accessory lacrimal gland, a meibomian gland, or the trabecular meshwork, of at least one eye of the subject. Various other modes of administration are described herein, e.g., modes of administering the vectors and anti-IGF1R antibodies or fragments described herein that cause result in delivery of diffusion of anti-IGF1R antibodies or fragments, directly or indirectly, into the extraorbital fat pads of the subject’s eye, thereby blocking IGF-1 from binding to IGF1R and inhibiting IGF1R downstream signaling.

[0184] In some embodiments, the disclosure provides rAAV vector for use, or adaptable for use, to treat a subject with an ocular disease, disorder, or condition. In some embodiments, the rAAV vector for use or adaptable for use comprise an AAV capsid and an expression cassette comprising a polynucleotide encoding an anti-IGF1R antibody or antigen-binding fragment, wherein the polynucleotide is operatively linked to a promoter.

[0185] In some embodiments, the disclosure provides rAAV vectors for use in a method of treating an ocular condition in a subject in need thereof, the method comprising

administering a recombinant adeno-associated virus (rAAV) vector described herein to at least one eye of the subject or to at least one lacrimal gland of the eye of the subject.

[0186] In some embodiments, the disclosure provides rAAV vectors for use in a method of treating an ocular condition in a subject in need thereof, the method comprising administering a recombinant adeno-associated virus (rAAV) vector comprising an expression cassette comprising the nucleic acid sequence encoding SEQ ID NOs: 2 or 6 to at least one eye of the subject or to at least one lacrimal gland of the eye of the subject.

[0187] In some embodiments, expression of the anti-IGF1R antibody, or an antigen-binding fragment is maintained for about 1 day, 2 days, 3 days, 4 days, 5 days, 6 days, 7 days, 1 week, 2 weeks, 3 weeks, 4 weeks, 1 month, 2 months, 3 months, 4 months, 5 months, 6 months, 7 months, 8 months, 9 months, 10 months, 11 months, or 1 year.

[0188] As used herein, “approximately” or “about,” as applied to one or more values of interest, refers to a value that is similar to a stated reference value. In certain embodiments, “about” refers to a range of values that fall within 25%, 20%, 19%, 18%, 17%, 16%, 15%, 14%, 13%, 12%, 11%, 10%, 9%, 8%, 7%, 6%, 5%, 4%, 3%, 2%, 1% or less in either direction (greater than or less than) of the stated reference value unless otherwise stated or otherwise evident from the context (except where such number would exceed 100% of a possible value).

[0189] In some embodiments, the disclosure provides vectors (e.g., rAAV vectors) for use in a method of treating an ocular condition in a subject in need thereof. For example, a method may comprise administering a recombinant adeno-associated virus (rAAV) vector, the rAAV vector comprising an AAV capsid and an expression cassette comprising a polynucleotide encoding an anti-IGF1R antibody, or an antigen-binding fragment, operatively linked to a promoter, to at least one eye of the subject or to at least one lacrimal gland of the eye of the subject. In some embodiments, the rAAV vector for use is administered to the lacrimal gland of a subject. In some embodiments, the rAAV vector for use comprises an expression cassette encoding an anti-IGF1R antibody or an IGF1R antigen-binding antibody fragment.

[0190] In some embodiments, the disclosure provides rAAV vectors for use, or adaptable for use, to treat a subject with an ocular disease, disorder, or condition. In some embodiments, the rAAV vector for use or adaptable for use comprises an AAV capsid and an expression cassette comprising a polynucleotide encoding an anti-IGF1R antibody or antigen-binding fragment, wherein the polynucleotide is operatively linked to a promoter.

[0191] In some embodiments, the method comprises administering a dose of the rAAV to a subject having a condition associated with TED, wherein the condition is characterized by one or more symptoms described herein. In some embodiments, the dose is administered prior

to the onset of the one or more symptoms. In some embodiments, the administering prevents the onset of the one or more symptoms. In some embodiments, the administering reduces the severity of the one or more symptoms. For example, in some embodiments, the method comprises administering a dose of the rAAV to a subject experiencing one or more symptoms of TED, such as a) hypothyroid associated visual disturbance, b) a reduction in visual acuity due to lid retraction, c) lid wiper epitheliopathy, d) dry eye, e) aqueous deficiency, f) double vision, g) compressive optic neuropathy, h) ocular pain, i) proptosis, and/or j) diplopia, wherein the dose of the rAAV is administered and prevents or lessens the severity of the one or more symptoms. In some embodiments, the dose is administered following the onset of the one or more symptoms, wherein the administering reduces the severity of the one or more symptoms.

[0192] In some embodiments, the method comprises administering a dosing regimen of the rAAV to a subject having a condition associated with TED, wherein the condition is characterized by one or more symptoms described herein, and the dosing regimen comprises a first dose of the rAAV and at least one additional dose, wherein the first dose of the rAAV is administered to the subject prior to or subsequent to the onset of the one or more symptoms, and wherein the at least one additional dose is administered to the subject following the first dose, thereby preventing or reducing the severity of the one or more symptoms. In some embodiments, the dosing regimen comprises a frequency of dosing and/or dose amount that is selected based upon the pharmacokinetic parameters of the rAAV. In some embodiments, a clinician will administer the rAAV at a frequency and/or dose that achieves or maintains one or more desired effects (e.g., a clinically measurable reduction or improvement of any symptom of any condition described herein). In some embodiments, the clinically measurable reduction or improvement of any symptom is achieved at least or within 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, or 14 days following administration of the rAAV to the subject. In some embodiments, the clinically measurable reduction or improvement of any symptom is achieved at least or within 1, 2, 3, 4, 5, or 6 weeks following administration of the rAAV to the subject. In some aspects, the one or more desired effects comprise preventing one or more symptoms associated with the subject's condition. In some embodiments, the one or more desired effects is reducing the severity of one or more symptoms associated with the subject's condition. In some embodiments, the one or more desired effects comprises one or more clinically measurable changes achieved by or within 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, or 14 days following administering of a first dose of the rAAV, or by or within 1, 2, 3, 4, 5, or 6 weeks following administering of a first dose of the rAAV. In some embodiments, the one or more desired effects occurs at any time point following administering of the first dose of the rAAV. In some

embodiments, the one or more desired effects is achieved following administration of the at least one additional dose of the rAAV. In some embodiments, the one or more desired effects is achieved at any point during the dosing regimen. In some embodiments, the one or more desired effects is achieved following administration of the first dose of the rAAV and the subject is administered at least one additional dose of the rAAV to prevent reversal of the one or more desired effects. In some embodiments, the method comprises administering a first dose of the rAAV to the subject prior to, immediately following, or during the onset of one or more symptoms and an additional dose following a duration of about 1 week, about 2 weeks, about 3 weeks, about 1 month, about 2 months, about 3 months, about 4 months, about 5 months, about 6 months, about 7 months, about 8 months, about 9 months, about 10 months, about 11 months, about 1 year, about 1.5 years, or about 2 years after the first dose.

[0193] In some embodiments, administration of an rAAV described herein results in measurable expression of an antibody or fragment encoded by the rAAV within or by one or more cells, tissues, or organs of the subject within 1, 2, 3, 4, 5, 6, or 7 days of administration of the rAAV. In some embodiments, expression of the antibody or fragment encoded by the rAAV is detectable in one or more cells, tissues, or organs of the subject 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23 or 24 weeks post-administration.

[0194] In some embodiments, treatment of a condition described herein or any symptom thereof is achieved by administration of an anti-IGF1R antibody or fragment described herein (e.g., as a protein in a liquid carrier suitable for topical administration). For example, any of the antibodies or fragments described herein may be topically administered to a cell, tissue, or organ of a human subject (e.g., as a liquid administered to the eye). Administration of the antibodies or fragments described herein may provide a therapeutic effect (e.g., a measurable reduction in or improvement of one or more symptoms of any condition described herein).

[0195] In some embodiments, methods are provided for treating an ocular disease, disorder, or condition comprising a) a hypothyroid associated visual disturbance, b) a reduction in visual acuity due to lid retraction, c) lid wiper epitheliopathy, d) dry eye, e) aqueous deficiency, f) double vision, g) compressive optic neuropathy, h) ocular pain, i) proptosis, and/or j) diplopia. In some embodiments, the method comprises administering a dose of an anti-IGF1R antibody or an IGF1R-binding fragment thereof to a subject, wherein administration reverses or mitigates one or more symptoms of the condition described herein. In some embodiments, the dose is administered prior to the onset of the one or more symptoms. In some embodiments, the administering prevents the onset of at least one of the one or more symptoms. In some embodiments, the administering reduces the severity of at least one of the

one or more symptoms. For example, in some embodiments, the method comprises administering a dose of the rAAV to a subject having TED characterized by one or more symptoms (e.g., proptosis, or diplopia), wherein the dose of the rAAV is administered and prevents or lessens the severity of at least one of the one or more symptoms. In some embodiments, the dose is administered following the onset of the one or more symptoms, wherein the administering reduces the severity of at least one of the one or more symptoms.

[0196] In some embodiments, the method comprises administering a dosing regimen of the rAAV to a subject having a condition associated with TED, wherein the condition is characterized by one or more symptoms described herein, wherein the dosing regimen comprises a first dose of the rAAV and at least one additional dose, wherein the first dose of the rAAV is administered to the subject prior to or subsequent to the onset of the one or more symptoms, and wherein the at least one additional dose is administered to the subject following the first dose, thereby preventing or reducing the severity of at least one of the one or more symptoms. In some embodiments, the dosing regimen comprises a frequency of dosing and/or dose amount that is selected based upon the pharmacokinetic parameters of the vector (e.g., any rAAV vector described herein) or of anti-IGF1R antibody or antigen-binding fragment (if administered directly). In some embodiments, a clinician will administer the vector or the anti-IGF1R antibody or antigen-binding fragment at a frequency and/or dose that achieves or maintains one or more desired effects. In some embodiments, the one or more desired effects is preventing one or more symptoms associated with the subject's condition. In some embodiments, the one or more desired effects is reducing the severity of one or more symptoms associated with the subject's condition. In some embodiments, the severity of the one or more symptoms is measured using a method described herein or known in the art for evaluating a pathology associated with TED. In some embodiments, the one or more desired effects is achieved immediately following administering of the first dose of the vector. In some embodiments, the one or more desired effects occurs at any time point following administering of the first dose of the vector. In some embodiments, the one or more desired effects is achieved following administration of the at least one additional dose of vector (e.g., an rAAV vector). In some embodiments, the one or more desired effects is achieved at any point during the dosing regimen. In some embodiments, the one or more desired effects is achieved following administration of the first dose of the vector and the subject is administered at least one additional dose of the vector to prevent reversal of the one or more desired effects. In some embodiments, the method comprises administering a first dose of the vector to the subject prior to, immediately following, or during the onset of one or more symptoms and an additional dose

following a duration of about 1 week, about 2 weeks, about 3 weeks, about 1 month, about 2 months, about 3 months, about 4 months, about 5 months, about 6 months, about 7 months, about 8 months, about 9 months, about 10 months, about 11 months, about 1 year, about 1.5 years, or about 2 years after the first dose.

[0197] In some embodiments, the method comprises administering a dose of the vector to a subject having a condition associated with loss of accommodation, wherein the condition is characterized by one or more symptoms described herein. In some embodiments, the dose is administered prior to the onset of the one or more symptoms. In some embodiments, the administering prevents the onset of at least one of the one or more symptoms. In some embodiments, the administering reduces the severity of at least one of the one or more symptoms. For example, in some embodiments, the method comprises administering a dose of the vector to a subject having loss of accommodation characterized by one or more symptoms, wherein the dose of the vector is administered and prevents or lessens the severity of at least one of the one or more symptoms. In some embodiments, the dose is administered following the onset of the one or more symptoms, wherein the administering reduces the severity of at least one of the one or more symptoms.

[0198] In some embodiments, the method comprises administering a dosing regimen of the vector (e.g., an rAAV vector) to a subject having a condition associated with loss of accommodation, wherein the condition is characterized by one or more symptoms described herein and the dosing regimen comprises a first dose of the vector and at least one additional dose, wherein the first dose of the vector is administered to the subject prior to or subsequent to the onset of the one or more symptoms, and wherein the at least one additional dose is administered to the subject following the first dose, thereby preventing or reducing the severity of at least one of the one or more symptoms. In some embodiments, the dosing regimen comprises a frequency of dosing and/or dose amount that is selected based upon the pharmacokinetic parameters of the vector. In some embodiments, a clinician will administer the vector at a frequency and/or dose that achieves or maintains one or more desired effects. In some embodiments, the one or more desired effects is preventing one or more symptoms associated with the subject's condition. In some embodiments, the one or more desired effects is reducing the severity of one or more symptoms associated with the subject's condition. In some embodiments, the severity of the one or more symptoms is measured using a method described herein or known in the art for evaluating a pathology associated with loss of accommodation. In some embodiments, the one or more desired effects is achieved immediately following administering of the first dose of the vector. In some embodiments, the

one or more desired effects occurs at any time point following administering of the first dose of the vector. In some embodiments, the one or more desired effects is achieved following administration of the at least one additional dose of the vector. In some embodiments, the one or more desired effects is achieved at any point during the dosing regimen. In some embodiments, the one or more desired effects is achieved following administration of the first dose of the vector and the subject is administered at least one additional dose of the vector to prevent reversal of the one or more desired effects. In some embodiments, the method comprises administering a first dose of the vector to the subject prior to, immediately following, or during the onset of one or more symptoms and an additional dose following a duration of about 1 week, about 2 weeks, about 3 weeks, about 1 month, about 2 months, about 3 months, about 4 months, about 5 months, about 6 months, about 7 months, about 8 months, about 9 months, about 10 months, about 11 months, about 1 year, about 1.5 years, or about 2 years after the first dose.

[0199] In some embodiments, the method comprises administering a dose of the vector to a subject having a condition associated with TED, wherein the condition is characterized by one or more symptoms described herein. In some embodiments, the dose is administered prior to the onset of the one or more symptoms. In some embodiments, the administering prevents the onset of at least one of the one or more symptoms. In some embodiments, the administering reduces the severity of at least one of the one or more symptoms. For example, in some embodiments, the method comprises administering a dose of the vector to a subject having compressive optic neuropathy characterized by one or more symptoms, wherein the dose of the vector is administered and prevents or lessens the severity of at least one of the one or more symptoms. In some embodiments, the dose is administered following the onset of the one or more symptoms, wherein the administering reduces the severity of at least one of the one or more symptoms.

[0200] In some embodiments, the method comprises administering a dose of the vector (e.g., an rAAV vector) to a subject having a condition associated with meibomian gland dysfunction (MDI), wherein the condition is characterized by one or more symptoms described herein. In some embodiments, the dose is administered prior to the onset of the one or more symptoms. In some embodiments, the administering prevents the onset of the one or more symptoms. In some embodiments, the administering reduces the severity of the one or more symptoms. For example, in some embodiments, the method comprises administering a dose of the vector to a subject having MDI characterized by one or more symptoms (e.g., eye dryness, burning, itching, redness, discharge, blurry vision), wherein the dose of the vector is

administered and prevents or lessens the severity of at least one of the one or more symptoms. In some embodiments, the dose is administered following the onset of the one or more symptoms, wherein the administering reduces the severity of at least one of the one or more symptoms.

[0201] In some embodiments, the method comprises administering a dosing regimen of the vector to a subject having a condition associated with TED, wherein the condition is characterized by one or more symptoms described herein, wherein the dosing regimen comprises a first dose of the vector and at least one additional dose, wherein the first dose of the vector is administered to the subject prior to or subsequent to the onset of the one or more symptoms, and wherein the at least one additional dose is administered to the subject following the first dose, thereby preventing or reducing the severity of at least one of the one or more symptoms. In some embodiments, the dosing regimen comprises a frequency of dosing and/or dose amount that is selected based upon the pharmacokinetic parameters of the vector. In some embodiments, a clinician will administer the vector at a frequency and/or dose that achieves or maintains one or more desired effects. In some embodiments, the one or more desired effects is preventing one or more symptoms associated with the subject's condition. In some embodiments, the one or more desired effects is reducing the severity of one or more symptoms associated with the subject's condition. In some embodiments, the severity of the one or more symptoms is measured using a method described herein or known in the art for evaluating a pathology associated with TED. In some embodiments, the one or more desired effects is achieved immediately following administering of the first dose of the vector. In some embodiments, the one or more desired effects occurs at any time point following administering of the first dose of the vector. In some embodiments, the one or more desired effects is achieved following administration of the at least one additional dose of the vector. In some embodiments, the one or more desired effects is achieved at any point during the dosing regimen. In some embodiments, the one or more desired effects is achieved following administration of the first dose of the vector and the subject is administered at least one additional dose of the vector to prevent reversal of at least one of the one or more desired effects. In some embodiments, the method comprises administering a first dose of the vector to the subject prior to, immediately following, or during the onset of one or more symptoms and an additional dose following a duration of about 1 week, about 2 weeks, about 3 weeks, about 1 month, about 2 months, about 3 months, about 4 months, about 5 months, about 6 months, about 7 months, about 8 months, about 9 months, about 10 months, about 11 months, about 1 year, about 1.5 years, or about 2 years after the first dose.

[0202] In some embodiments, the disclosure provides a method for treating a) hypothyroid associated visual disturbance, b) a reduction in visual acuity due to lid retraction, c) lid wiper epitheliopathy, d) dry eye, e) aqueous deficiency, f) double vision, g) compressive optic neuropathy, h) ocular pain, i) proptosis, and/or j) diplopia, in a subject in need thereof, the method comprising administering to the subject one or more therapeutically effective doses of a vector described herein.

[0203] In some embodiments, the disclosure provides a vector (e.g., an rAAV vector) described herein for use in the manufacture of a medicament for treating a) hypothyroid associated visual disturbance, b) a reduction in visual acuity due to lid retraction, c) lid wiper epitheliopathy, d) dry eye, e) aqueous deficiency, f) double vision, g) compressive optic neuropathy, h) ocular pain, i) proptosis, and/or j) diplopia, in a subject in need thereof.

[0204] In some embodiments, the disclosure provides a vector (e.g., an rAAV vector) described herein for use in a method of treating a) hypothyroid associated visual disturbance, b) a reduction in visual acuity due to lid retraction, c) lid wiper epitheliopathy, d) dry eye, e) aqueous deficiency, f) double vision, g) compressive optic neuropathy, and/or h) ocular pain in a subject in need thereof, the method comprising administering to the subject an effective amount of the vector.

[0205] In some embodiments, the disclosure provides a vector described herein for use in the manufacture of a medicament for treating TED in a subject in need thereof.

[0206] In some embodiments, the disclosure provides a vector described herein for use in a method of treating TED in a subject in need thereof, the method comprising administering to the subject an effective amount of any vector described herein, or an effective amount of any anti-IGF1R antibody, or an antigen-binding fragment, described herein.

Modes of Administration

[0207] In some embodiments, the disclosure provides methods comprising administering a vector, e.g., an rAAV vector comprising an AAV capsid and an expression cassette comprising a polynucleotide encoding an anti-IGF1R antibody or antigen-binding fragment operatively linked to a promoter, or an anti-IGF1R antibody or antigen-binding fragment, to the eye of the subject. Administration may be to a tear film or an eye tissue of the subject (e.g., to a main or accessory lacrimal gland of an eye of the subject, to a meibomian gland of an eye of the subject, or to the trabecular meshwork of an eye of a subject).

[0208] The lacrimal functional unit is composed of main and accessory lacrimal glands, the ocular surface, and the interconnecting innervation. For each eye, a main lacrimal gland is

situated superotemporally in the orbital within the lacrimal fossa of the frontal bone. The accessory glands, known as the Wolfring's glands and Krause's glands are located in the eyelid. In the upper eyelid there are about 2 to 5 Wolfring's glands and about forty Krause's glands. In the lower eyelid there are about 6 to 8 Krause's glands. Specific location and anatomy of the lacrimal functional unit is well-known (Conrady *et al.*, *J. Ophthalmol.*; 2016: 7542929 (2016)). Together, the lacrimal glands secrete tear film onto the ocular surface through lacrimal ducts. Lacrimal glands also express and secrete proteins and products necessary for corneal regeneration and promoting transparency into the tear film, such as transforming growth factor- β and retinol (Conrady *et al.*, *J. Ophthalmol.*; 2016: 7542929 (2016); Pan *et al. Optom Vis Sci.*; 95:27-31 (2018)). In addition to secreting tears into the eye, the lacrimal duct drains the lacrimal fluid into the nasal cavity.

[0209] Administration of a vector (e.g., an rAAV vector) to the lacrimal gland may be accomplished by topical administration to the ocular surface, direct injection into the lacrimal gland, and/or topical administration to the lacrimal gland. The lacrimal gland may be accessed surgically or by manipulation of the eyelid. Manipulation of the eyelid provides access to the tissue for administration topically (e.g., by lavage of the tissue with a pharmaceutical composition comprising the vector). Direct injection into the lacrimal gland may be done by penetrating the skin over the lacrimal gland (**FIG. 2A**) or by manipulating the eyelid to access the lacrimal gland (**FIG. 2B**). Administration of a vector to the lacrimal gland may be accomplished by transconjunctival injection. In some embodiments, the vector is administered to the lacrimal gland by direct injection as depicted in **FIG. 2A**. In some embodiments, the vector is administered to the lacrimal gland by manipulating the eyelid as depicted in **FIG. 2B**.

[0210] In some embodiments, cells within the eye, the lacrimal gland, and/or the nasolacrimal duct are transduced or transfected by the vector. Cells within the eye, the lacrimal gland and/or the nasolacrimal duct include, without limitation, acinar cells, ductal cells, and/or myoepithelial cells, as well as cells of the iris and ciliary body ("ICB"), lens epithelial cells, cells of the meibomian glands and the trabecular meshwork. In some embodiments, the transduced or transfected cells within the eye, the lacrimal gland, and/or the nasolacrimal duct express a therapeutically effective amount of an anti-IGF1R antibody or antigen-binding fragment. In some embodiments, the transduced or transfected cells in the eye, the lacrimal gland, and/or the nasolacrimal duct secrete a therapeutically effective amount of an anti-IGF1R antibody or antigen-binding fragment into the tear film. In some embodiments, cells within the meibomian glands are transduced or transfected by the vector. In some embodiments, cells within the trabecular meshwork are transduced or transfected by the vector. In some

embodiments, a therapeutically effective amount of an anti-IGF1R antibody or antigen-binding fragment is secreted into the nasal cavity of the subject. In some embodiments, a therapeutically effective amount of an anti-IGF1R antibody or antigen-binding fragment is secreted onto the ocular surface of the subject.

[0211] Delivery of rAAV vectors to the eye and/or lacrimal gland to express a transgene into tear film has been demonstrated *in vivo*. The main lacrimal glands of mice were directly injected with rAAV vectors encoding a luciferase transgene with serotypes AAV2, AAV4, AAV5, AAV 5w8, AAV x5, AAV 9, AAV12, and bovine AAV (BAAV). AAV9, AAV 5w8, AAV5, and AAV2 are each able to transduce the lacrimal ductal and acinar cells of the lacrimal gland (Rocha et al., *supra*).

[0212] In some embodiments, the vector is administered to a lacrimal gland of the subject. In some embodiments, the lacrimal gland is the main lacrimal gland. In some embodiments, the lacrimal gland is any of the Wolfring's glands or the Krause's glands of the subject.

[0213] Compositions and vectors according to the disclosure may be administered to the lacrimal gland of the subject by any suitable method. For example, the subject composition may be administered by direct injection to the main or accessory lacrimal glands.

[0214] Access to the lacrimal gland in human subjects can be achieved, for example, by manually elevating the upper eyelid to expose the palpebral lobe of the lacrimal gland and delivering the therapeutic agent using a syringe, *e.g.*, with a 30G needle.

[0215] In some embodiments, the vector is administered to the trabecular meshwork of an eye of the subject.

[0216] In some embodiments, administration of the vector, or any antigen-binding fragment described herein, is performed via injection, using a liquid carrier (*e.g.*, one or more lipid nanoparticles or liposomes), or using an erodible insert (*e.g.*, a polymeric gel containing the vector to be administered).

[0217] In some embodiments, the vector, or an antigen-binding fragment described herein, is administered to the endothelial cells of the cornea of a subject. This could be achieved by injection into an adjacent or appropriate tissue such as an intracorneal injection, for example. In some embodiments, the vector is administered to other cells and/or tissues of the subject. For example, administration may be performed by conjunctival injection, injection into corneal stroma, suprachoroidal injection, subtenon injection, intravitreal injection, intracameral injection, injection into a meibomian gland, or subdermal eyelid injection. For some tissues, administration of the vector may be performed via injection, using a liquid carrier (*e.g.*, one or

more lipid nanoparticles), or using an erodible insert (e.g., a polymeric gel containing the vector to be administered).

[0218] The vectors of the disclosure are generally delivered to the subject as a pharmaceutical composition. Pharmaceutical compositions comprise a pharmaceutically acceptable solvent (*e.g.*, water, etc.) and one or more excipients. In some embodiments, the pharmaceutical compositions comprise a buffer at about neutral pH (pH 5, 6, 7, 8, or 9). In some embodiments, the pharmaceutical composition comprises phosphate buffered saline (*e.g.*, PBS at pH of about 7). The pharmaceutical compositions may comprise a pharmaceutically acceptable salt. The concentration of the salt may be selected to ensure that the pharmaceutical composition is isotonic to, or nearly isotonic to, the target tissue.

[0219] In various embodiments, the pharmaceutical compositions of the disclosure comprise about 1×10^8 genome copies per milliliter (GC/mL), about 5×10^8 GC/mL, about 1×10^9 GC/mL, about 5×10^9 GC/mL, about 1×10^{10} GC/mL, about 5×10^{10} GC/mL, about 1×10^{11} GC/mL, about 5×10^{11} GC/mL, about 1×10^{12} GC/mL, about 5×10^{12} GC/mL, about 5×10^{13} GC/mL, or about 1×10^{14} GC/mL of the vector. In various embodiments, the pharmaceutical compositions of the disclosure comprise about 1×10^8 genome copies per milliliter (GC/mL), about 5×10^8 GC/mL to about 1×10^9 GC/mL, about 1×10^9 GC/mL to about 5×10^9 GC/mL, about 5×10^9 GC/mL to about 1×10^{10} GC/mL, about 1×10^{10} GC/mL to about 5×10^{10} GC/mL, about 5×10^{10} GC/mL to about 1×10^{11} GC/mL, about 1×10^{11} GC/mL to about 5×10^{11} GC/mL, about 5×10^{11} GC/mL to about 1×10^{12} GC/mL, about 1×10^{12} GC/mL to about 5×10^{12} GC/mL, about 5×10^{12} GC/mL to about 5×10^{13} GC/mL, or about 5×10^{13} GC/mL to about 1×10^{14} GC/mL of the vector. In various further embodiments, the pharmaceutical compositions of the disclosure comprise about 1×10^8 genome copies per milliliter (GC/mL), about 5×10^8 GC/mL to about 5×10^9 GC/mL, about 5×10^9 GC/mL to about 5×10^{10} GC/mL, about 5×10^{10} GC/mL to about 5×10^{11} GC/mL, about 5×10^{11} GC/mL to about 5×10^{12} GC/mL, or about 5×10^{12} GC/mL to about 1×10^{14} GC/mL of the vector. In some embodiments, the pharmaceutical compositions of the disclosure comprise about 5×10^8 GC/mL to about 5×10^{10} GC/mL, about 5×10^{10} GC/mL to about 5×10^{12} GC/mL, or about 5×10^{12} GC/mL to about 1×10^{14} GC/mL of the vector.

[0220] In some embodiments, the pharmaceutical compositions of the disclosure comprise about 1×10^{12} GC/mL to about 6.2×10^{12} GC/mL of the vector. In some embodiments, the pharmaceutical compositions of the disclosure comprise about 1×10^{12} GC/mL or about 6.2×10^{12} GC/mL of the vector.

[0221] In some embodiments, the pharmaceutical compositions of the disclosure are administered in a total volume of about 10 μL , about 20 μL , about 30 μL , about 40 μL , about 50 μL , about 60 μL , about 70 μL , about 80 μL , about 90 μL , about 100 μL , 110 μL , about 120 μL , about 130 μL , about 140 μL , about 150 μL , about 160 μL , about 170 μL , about 180 μL , about 190 μL , or about 200 μL . In some embodiments, the pharmaceutical compositions of the disclosure are administered in a total volume of about 10 μL to about 20 μL , about 20 μL to about 30 μL , about 30 μL to about 40 μL , about 40 μL to about 50 μL , about 50 μL to about 60 μL , about 60 μL to about 70 μL , about 70 μL to about 80 μL , about 80 μL to about 90 μL , about 90 μL to about 100 μL , about 100 μL to 110 μL , 110 μL to about 120 μL , about 120 μL to about 130 μL , about 130 μL to about 140 μL , about 140 μL to about 150 μL , about 150 μL to about 160 μL , about 160 μL to about 170 μL , about 170 μL to about 180 μL , about 180 μL to about 190 μL , or about 190 μL to about 200 μL .

[0222] Genome copies per milliliter can be determined by quantitative polymerase chain reaction (qPCR) using a standard curve generated with a reference sample having a known concentration of the polynucleotide genome of the vector (or virus, in the case of a viral vector). For AAV, the reference sample used is often the transfer plasmid used in generation of the rAAV vector but other reference samples may be used.

[0223] Alternatively, or in addition, the concentration of a viral vector can be determined by measuring the titer of the vector on a cell line. Viral titer is typically expressed as viral particles (vp) per unit volume (*e.g.*, vp/mL). In various embodiments, the pharmaceutical compositions of the disclosure comprise about 1×10^8 viral particles per milliliter (vp/mL), about 5×10^8 vp/mL, about 1×10^9 vp/mL, about 5×10^9 vp/mL, about 1×10^{10} vp/mL, about 5×10^{10} vp/mL, about 1×10^{11} vp/mL, about 5×10^{11} vp/mL, about 1×10^{12} vp/mL, about 5×10^{12} vp/mL, about 5×10^{13} vp/mL, or about 1×10^{14} vp/mL of the rAAV vector. In various further embodiments, the pharmaceutical compositions of the disclosure comprise about 1×10^8 viral particles per milliliter (vp/mL) to about 5×10^8 vp/mL, about 5×10^8 vp/mL to about 1×10^9 vp/mL, about 1×10^9 vp/mL to about 5×10^9 vp/mL, about 5×10^9 vp/mL to about 1×10^{10} vp/mL, about 1×10^{10} vp/mL to about 5×10^{10} vp/mL, about 5×10^{10} vp/mL to about 1×10^{11} vp/mL, about 1×10^{11} vp/mL to about 5×10^{11} vp/mL, about 5×10^{11} vp/mL to about 1×10^{12} vp/mL, about 1×10^{12} vp/mL to about 5×10^{12} vp/mL, about 5×10^{12} vp/mL to about 5×10^{13} vp/mL, or about 5×10^{13} vp/mL to about 1×10^{14} vp/mL of the rAAV vector.

Assessment of Efficacy

[0224] A variety of tests are available to evaluate ocular conditions in a subject before, during, and after treatment with any of the methods or compositions disclosed herein. In some of the embodiments disclosed herein, the effective treatment of the subject is indicated by one or more of the tests that can be, for example, a) Eye Dryness score test on a visual analog scale, b) Schirmer's test, c) Corneal Fluorescein Staining test, and d) Ocular Surface Disease Index test. In some embodiments, the effective treatment of the subject is indicated by one or more of the tests that can be, for example Symptom Assessment in Dry Eye Questionnaire (SANDE), meibomian gland atrophy/drop-out, expression of glands, meibography/meibometry, tear film lipid layer thickness, corneal fluorescein staining, tear break up time (TBUT), distance-corrected near visual activity (DCNVA), intraocular pressure measurement (e.g., tonometry), visual acuity, slit lamp examination, or ophthalmoscope examination. Tests to evaluate the signs and symptoms of an ocular condition may be administered under standardized or reproducible conditions in order to obtain a subject's test score. Conditions include exposing the subject to an environment artificially created to adversely challenge the subject or where the environment (temperature, humidity, air flow) is monitored and carefully controlled.

[0225] For example, in some embodiments, an improvement to one or more symptoms of TED may be determined using metric and protocols known in the art. For example, an improvement as to any of the following symptoms may optionally be determined as follows:

(a) Proptosis (sometimes be referred to as exophthalmos): an improvement may be determined or measured with an exophthalmometer. The measurement is in millimeters and is consistent with eyelid retraction, Von Graefe's lid lag and lid edema.

(b) Double vision: an improvement may be determined or measured using an eye tracker and/or MRI scan.

(c) Pain: an improvement may be determined or measured via a patient assessment using a grading scale (e.g., an ocular pain assessment survey, "OPAS"), the VRS pain scale, etc.

(d) Redness: an improvement may be determined or measured via clinical grading or color image analysis, e.g., using a 0-4 scale in 0.5-unit increments.

(e) Functional vision: an improvement may be determined or measured by a visual function assessment via reading and/or simulators.

[0226] The efficacy of a method described herein can be assessed using any suitable method known in the art.

[0227] In some embodiments, the methods described herein result in one or more symptoms of the ocular condition being reduced compared to the symptoms of the ocular condition before administration of the vector. As used herein, “symptoms” include any of the diagnostic criteria or symptoms associated with a given ocular condition, including those described herein. Non-limiting examples of symptoms that may be alleviated by treatment in accordance with the methods described herein include, for example, a worsening of visual acuity, as well as the requirement for corrective lenses (e.g., spectacles or contact lenses) and/or surgery.

[0228] In some embodiments, a method of treatment described herein results in a delay of the onset of an ocular condition. In some embodiments, the onset of an ocular condition is delayed relative to the onset of the ocular condition in a control subject. In some embodiments, the onset of the ocular condition is delayed relative to the onset of the ocular condition in a contralateral eye. In some embodiments, a method of treatment described herein results in a delay of the progression of an ocular condition. In some embodiments, the progression of an ocular condition is delayed relative to the progression of the ocular condition in a control subject. In some embodiments, the progression of the ocular condition is delayed compared to the progression of the ocular condition in a contralateral eye. A “control subject” may be, for example, an untreated control subject. In some embodiments, the control subject is an age-matched subject who is not treated with a vector comprising an expression cassette, the expression cassette comprising a polynucleotide encoding an anti-IGF1R antibody, or an antigen-binding fragment. In some embodiments, a control subject is an age-matched untreated subject. By “contralateral eye” it is meant the eye of the subject that is opposite from the eye that has been treated with a composition according to the present disclosure. The contralateral eye can be used as a control for treatment, so long as the subject suffers from bilateral disease, or in the case of a model animal, has been subjected to the experimental protocol leading up to treatment in both eyes.

[0229] A method of treatment described herein may result in the slowing of the progression of an ocular condition. In some embodiments, the progression of the condition in the subject is slowed by about 10% to about 20%, about 20% to about 30%, about 30% to about 40%, about 40% to about 50%, about 50% to about 60%, about 60% to about 70%, about 70% to about 80%, about 80% to about 90%, about 90% to about 95% or by more than about 95% compared to a control subject.

[0230] In some embodiments, a method of treatment described herein results in the onset of the condition in a subject being delayed by about 6 months to about 9 months, about 9

months to about 12 months, about 12 months to about 15 months, about 15 months to about 18 months, about 18 months to about 21 months, about 21 months to about 24 months, to about 2 years to about 3 years, about 3 years to about 4 years, about 4 years to about 5 years, about 5 years to about 6 years, about 6 years to about 7 years, about 7 years to about 8 years, about 8 years to about 9 years, about 9 years to about 10 years, about 10 years to about 15 years, about 15 years to about 20 years, or by more than 20 years compared to a control subject.

[0231] In some embodiments, a method of treatment described herein results in an increase in visual acuity in a subject. In some embodiments, a method of treatment described herein results in a delay in the decrease in visual acuity in a subject, e.g., relative to a contralateral eye or relative to a control subject. Visual acuity can be measured using charts on which a subject is to identify letters from a certain distance, including, for example, the Snellen acuity chart, and the Early Treatment of Diabetic retinopathy Study (ETDRS) chart (Bailey and Kitchin, Vision Research 90 (2013) 2–9; Bennet *et al.*, Semin Pediatr Neurol. 2019 October; 31: 30–40). Visual acuity may also be assessed using binocular distance corrected near visual acuity (DCNVA).

[0232] In some embodiments, the visual acuity of a subject treated in accordance with a method described herein remains unchanged for at least about 6 months to about 9 months, about 9 months to about 12 months, about 12 months to about 15 months, about 15 months to about 18 months, about 18 months to about 21 months, about 21 months to about 24 months, to about 2 years to about 3 years, about 3 years to about 4 years, about 4 years to about 5 years, about 5 years to about 6 years, about 6 years to about 7 years, about 7 years to about 8 years, about 8 years to about 9 years, about 9 years to about 10 years, about 10 years to about 15 years, about 15 years to about 20 years, or for more than 20 years after the administration of the rAAV vector.

[0233] In some embodiments, the visual acuity of a subject treated in accordance with a method described herein does not decrease by more than 5% for at least about 6 months to about 9 months, about 9 months to about 12 months, about 12 months to about 15 months, about 15 months to about 18 months, about 18 months to about 21 months, about 21 months to about 24 months, to about 2 years to about 3 years, about 3 years to about 4 years, about 4 years to about 5 years, about 5 years to about 6 years, about 6 years to about 7 years, about 7 years to about 8 years, about 8 years to about 9 years, about 9 years to about 10 years, about 10 years to about 15 years, about 15 years to about 20 years, or for more than 20 years after the administration of the rAAV vector. In some embodiments, the visual acuity of a subject treated in accordance with a method described herein does not decrease by more than 10% for at least

about 6 months to about 9 months, about 9 months to about 12 months, about 12 months to about 15 months, about 15 months to about 18 months, about 18 months to about 21 months, about 21 months to about 24 months, to about 2 years to about 3 years, about 3 years to about 4 years, about 4 years to about 5 years, about 5 years to about 6 years, about 6 years to about 7 years, about 7 years to about 8 years, about 8 years to about 9 years, about 9 years to about 10 years, about 10 years to about 15 years, about 15 years to about 20 years, or for more than 20 years after the administration of the rAAV vector.

[0234] Another indicator of visual acuity may be the requirement for corrective lenses (e.g., contact lenses or spectacles), usually increasing in strength as a condition progresses. In some embodiments, a subject treated in accordance with a method described herein requires corrective lenses prior to the administration of the rAAV vector and the method results in an unchanged requirement for the strength of corrective lenses for at least about 6 months to about 9 months, about 9 months to about 12 months, about 12 months to about 15 months, about 15 months to about 18 months, about 18 months to about 21 months, about 21 months to about 24 months, to about 2 years to about 3 years, about 3 years to about 4 years, about 4 years to about 5 years, about 5 years to about 6 years, about 6 years to about 7 years, about 7 years to about 8 years, about 8 years to about 9 years, about 9 years to about 10 years, about 10 years to about 15 years, about 15 years to about 20 years, or for more than 20 years after the administration of the rAAV vector.

[0235] In some embodiments, the effective treatment of the subject is indicated by a symptom assessment in dry eye questionnaire (SANDE). In some embodiments, the effective treatment of the subject is indicated by meibomian gland atrophy/drop-out. In some embodiments, the effective treatment of the subject is indicated by expression of glands. In some embodiments, the effective treatment of the subject is indicated by meibography/meibometry. In some embodiments, the effective treatment of the subject is indicated by tear film lipid layer thickness. In some embodiments, the effective treatment of the subject is indicated by corneal fluorescein staining. In some embodiments, the effective treatment of the subject is indicated by tear break up time (TBUT). In some embodiments, the effective treatment of the subject is indicated by distance-corrected near visual acuity (DCNVA). In some embodiments, the effective treatment of the subject is indicated by intraocular pressure measurement. In some embodiments, the effective treatment of the subject is indicated by visual acuity. In some embodiments, the effective treatment of the subject is indicated by slit lamp examination. In some embodiments, the effective treatment of the subject

is indicated by ophthalmoscope examination. The methods described herein for assessing effective treatment of a subject are known to those of skill in the art.

Maintenance Of Effective Treatment Over Time

[0236] The efficacy of a method of treatment described herein may be evaluated at any suitable time point after administration, for example, at about 1 month, about 2 months, about 3 months, about 4 months, about 5 months, about 6 months, about 7 months, about 8 months, about 9 months, about 10 months, about 11 months, about 12 months, about 18 months, about 24 months, about 3 years, about 4 years or about 5 years after the administration of a vector described herein. In some embodiments, the efficacy is measured at two or more time points after the administration of an rAAV vector described herein, for example, every 3 months, every 6 months or every 12 months after the administration of the vector.

[0237] The present disclosure may provide for effective treatment over a period of time where a measure of efficacy (e.g., visual acuity) is maintained. The term “maintained” as used in the present disclosure and as it relates to the maintenance of a measure of efficacy (e.g., visual acuity) in a subject’s score refers to the statistically significant improvement not diminishing below a certain threshold over time. The efficacy measure after treatment according to the disclosed methods can be maintained without additional dosing or after one or more subsequent doses.

Dosing Timing And Methods Of Administration

[0238] The schedule of doses administered to a subject depends on various considerations including the duration of effectiveness of each dose, the transduction (or transfection) efficiency of the rAAV vector, and the effect of the dose on the body. For example, wherein the patient’s condition does not improve, upon the health provider’s discretion, the method of treating an ocular condition as described herein, may be adjusted in dose or administered repeatedly in order to ameliorate or otherwise control or limit the symptoms of the subject’s ocular condition. For instance, the period of time between administrations of one or more doses is extended, or the period of time between days the subject is administered one or more doses is extended. As a non-limiting example, administration of one or more doses is modified to administration of one or more doses after measuring symptoms of the ocular condition.

[0239] The term “dose”, as used herein, may refer to a dose of a pharmaceutical composition of the disclosure, or a dose of the treatment that reduces symptoms in an ocular condition.

[0240] In some of the embodiments described herein, a dose of the vector is a dose of a rAAV vector carrying an expression cassette. In such cases, delivery of an appropriate dose (*e.g.*, effective amount) of the gene product is achieved by administering an appropriate amount/titer of the vector to the target site which allows expression of an effective amount of the gene product over a period of time. An “effective amount,” as used herein, refers to an amount or dose of a vector, treatment, or composition described herein that is sufficient to reduce the symptoms and or signs of an ocular condition described herein. The term “amount” as used herein refers to an absolute amount (*e.g.*, an absolute amount of protein, or of a non-viral vector, or of rAAV vector particles) or concentration (*e.g.*, a concentration of protein in a solution), whether the amount referred to in a given instance refers to an absolute amount, concentration, or both, will be clear to the skilled artisan based on the context provided herein.

[0241] In some embodiments, the vector is an rAAV vector (*e.g.*, an rAAV virion). In some embodiments, the rAAV vector is administered to the lacrimal gland. In some embodiments, the rAAV vector is administered to the lacrimal gland by topical administration. In some embodiments, the rAAV vector is administered to the lacrimal gland by direct injection. In some embodiments, the rAAV vector is administered to the trabecular meshwork. In some embodiments, a dose of the rAAV vector results in the stable production of the gene product for a period of time (*e.g.*, about 1 day, about 2 days, about 4 days, about 1 week, about 2 weeks, about 3 weeks, about 4 weeks, about 1 month, about 2 months, about 3 months, about 4 months, about 5 months, about 6 months, about 9 months, about 12 months, or longer). In some embodiments, a dose of the rAAV vector results in the stable production of the gene product for about 1 week. In some embodiments, a dose of the rAAV vector results in the stable production of the gene product for about 2 weeks. In some embodiments, a dose of the rAAV vector results in the stable production of the gene product for about 3 weeks. In some embodiments, a dose of the rAAV vector results in the stable production of the gene product for about 4 weeks. In some embodiments, a dose of the rAAV vector results in the stable production of the gene product for about 1 month. In some embodiments, a dose of the rAAV vector results in the stable production of the gene product for about 2 months. In some embodiments, a dose of the rAAV vector results in the stable production of the gene product for about 3 months. In some embodiments, a dose of the rAAV vector results in the stable production of the gene product for about 4 months. In some embodiments, a dose of the rAAV vector results in the stable production of the gene product for about 5 months. In some embodiments, a dose of the rAAV vector results in the stable production of the gene product for about 6 months. In some embodiments, a dose of the rAAV vector results in the stable

production of the gene product for about 9 months. In some embodiments, a dose of the rAAV vector results in the stable production of the gene product for about 12 months.

[0242] In some embodiments, a method described herein comprises administering an effective amount of an rAAV vector described herein to a subject, wherein the rAAV vector comprises a polynucleotide encoding at least one gene product (e.g., an anti-IGF1R antibody, or an antigen-binding fragment). In some embodiments, the method comprises delivering a first dose and one or more subsequent doses of the rAAV vector. The one or more subsequent doses are administered after a period of time after the first dose. In some embodiments, this period of time between the first dose and the next subsequent dose is at least 1 day, at least 3 days, at least 1 week, at least 2 weeks, at least 3 weeks, at least 4 weeks, at least 1 month, at least 2 months, at least 4 months, at least 6 months, at least 9 months, at least 12 months, or longer. In some embodiments, this period of time between the first dose and the next subsequent dose is between 1-7 days, between 1-4 weeks, between 2-6 weeks, between 4-8 weeks, between 1-3 months, between 2-4 months, between 3-6 months, between 4-12 months, between 6-24 months. In some embodiments, the period of time between the one or more subsequent doses is at least 1 day, at least 3 days, at least 1 week, at least 2 weeks, at least 3 weeks, at least 4 weeks, at least 1 month, at least 2 months, at least 4 months, at least 6 months, at least 9 months, at least 12 months, or longer. In some embodiments, the period of time between the one or more subsequent doses is between 1-7 days, between 1-4 weeks, between 2-6 weeks, between 4-8 weeks, between 1-3 months, between 2-4 months, between 3-6 months, between 4-12 months, between 6-24 months.

[0243] In some embodiments, the method comprises a first dose and one or more subsequent doses of the rAAV vector. In some embodiments, the one or more subsequent doses are administered after a period of time after the first dose. This period of time between the first dose and the next subsequent dose is at least 1 hour, at least 2 hours, at least 3 hours, at least 4 hours, at least 5 hours, at least 6 hours, at least 7 hours, or at least 8 hours. The period of time between the first dose and the next subsequent dose is between 1-3 hours, 2-4 hours, 3-6 hours, or 4-8 hours. The period of time between the one or more subsequent doses is at least 1 hour, at least 2 hours, at least 3 hours, at least 4 hours, at least 5 hours, at least 6 hours, at least 7 hours, or at least 8 hours. The period of time between the one or more subsequent doses is between 1-3 hours, 2-4 hours, 3-6 hours, or 4-8 hours.

Pharmaceutical Compositions and Kits

[0244] In some embodiments, the disclosure provides a pharmaceutical composition comprising a vector (e.g., an rAAV vector) described herein, or an anti-IGF1R antibody, or an antigen-binding fragment. In some embodiments, the pharmaceutical composition comprises a vector described herein or an anti-IGF1R antibody, or an antigen-binding fragment, and a pharmaceutically acceptable carrier, delivery agent, or excipient.

[0245] In some embodiments, the disclosure provides use of a vector or pharmaceutical composition described herein, in the manufacture of a medicament for treatment of an ocular disease, disorder, or condition. In some embodiments, the disclosure provides use of a vector or of an anti-IGF1R antibody, or an antigen-binding fragment, or a pharmaceutical composition comprising any of these components, for use, or adaptable for use, in the treatment of an ocular disease, disorder, or condition.

[0246] In some embodiments, the pharmaceutically acceptable carrier comprises phosphate buffered saline. In some embodiments, the pharmaceutical composition is formulated to be compatible with its intended route of administration (e.g., intralacrimal). In some embodiments, the pharmaceutical composition is formulated for administration into the lacrimal gland. In some embodiments, the pharmaceutical composition is formulated for administration onto the ocular surface.

[0247] In some embodiments, the disclosure provides a pharmaceutical composition for use in treating an ocular condition in a subject, wherein the pharmaceutical composition comprises a vector (e.g., an rAAV vector), according to any aspect described herein, encoding an anti-IGF1R antibody, or an antigen-binding fragment, and at least one pharmaceutically acceptable carrier. Any concentration of the vector that is suitable to effectively transduce or transfect cells of the eye, the lacrimal gland, the meibomian glands, and/or the trabecular meshwork cells can be prepared for contacting cells of the eye, the lacrimal gland, the meibomian glands, and/or the trabecular meshwork *in vitro* or *in vivo*. For example, the vector may be formulated at a concentration of 10^8 vector genomes per ml or more, for example, 5×10^8 vector genomes per mL; 10^9 vector genomes per mL; 5×10^9 vector genomes per mL; 10^{10} vector genomes per mL; 5×10^{10} vector genomes per mL; 10^{11} vector genomes per mL; 5×10^{11} vector genomes per mL; 10^{12} vector genomes per mL; 5×10^{12} vector genomes per mL; 10^{13} vector genomes per mL; 1.5×10^{13} vector genomes per mL; 3×10^{13} vector genomes per mL; 5×10^{13} vector genomes per mL; 7.5×10^{13} vector genomes per mL; 9×10^{13} vector genomes per mL; 1×10^{14} vector genomes per mL, 5×10^{14} vector genomes per mL or more, but typically

not more than 1×10^{15} vector genomes per mL. Similarly, any total number of vectors (e.g., rAAV vectors) suitable to provide appropriate transduction or transfection of cells of the eye, the lacrimal gland, the meibomian glands, and/or the trabecular meshwork cells to confer the desired effect or treat the disease can be administered to the mammal or to the primate's eye. In various embodiments, at least 10^5 ; 2.5×10^5 ; 5×10^5 ; 7.5×10^5 ; 10^6 ; 2.5×10^6 ; 5×10^6 ; 7.5×10^6 ; 10^7 ; 2.5×10^7 ; 5×10^7 ; 7.5×10^7 ; 10^8 ; 2.5×10^8 ; 5×10^8 ; 7.5×10^8 ; 10^9 ; 2.5×10^9 ; 5×10^9 ; 7.5×10^9 ; 10^{10} ; 2.5×10^{10} ; 5×10^{10} ; 7.5×10^{10} ; 10^{11} ; 2.5×10^{11} ; 5×10^{11} ; 7.5×10^{11} ; 10^{12} ; 2.5×10^{12} ; 5×10^{12} ; 7.5×10^{12} ; 10^{13} ; 2.5×10^{13} ; 5×10^{13} ; 7.5×10^{13} ; 10^{14} ; 2.5×10^{14} ; 5×10^{14} ; 7.5×10^{14} ; 10^{15} ; 2.5×10^{15} ; 5×10^{15} ; or 7.5×10^{15} ; 10^{16} ; 2.5×10^{16} ; 5×10^{16} ; or 7.5×10^{16} vectors, or more, but typically not more than 1×10^{15} vectors are injected per eye. For example, in some embodiments, about 1×10^9 to about 1×10^{10} , about 1×10^{10} to about 1×10^{11} , about 1×10^{11} to about 1×10^{12} , about 1×10^{12} to about 1×10^{13} , or about 1×10^{13} to about 1×10^{15} genome copies of the vector are administered per eye. In some aspects, the total number of vectors administered to the eye of the human subject or animal being treated may comprise a concentration within a range defined by any pair of concentrations described in this paragraph. Any suitable number of administrations of the vectors to the mammal or the primate eye can be made. In some embodiments, the methods comprise a single administration; in other embodiments, multiple administrations are made over time as deemed appropriate by an attending clinician.

[0248] In some embodiments, a suitable amount or concentration of the vector (or any other expression construct configured to express an anti-IGF1R antibody or antigen-binding fragment) in a therapeutic formulation may be a concentration effective to express from 100 pg/mL to 50 μ g/mL of the anti-IGF1R antibody or antigen-binding fragment, in a tear film of a subject subsequent to administration of the composition to the subject. For example, the amount or concentration of the vector may be one that results in the expression of 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49 or 50 μ g/mL of anti-IGF1R antibody or antigen-binding fragment (or a concentration within a range defined by any pair of the foregoing values) in a tear film of a subject. Expression may be measured, e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23 hours after administration, or after a longer duration of time, such as after 1, 2, 3, 4, or 5 days.

[0249] The vector may be formulated into any suitable unit dosage, including, without limitation, 1×10^8 vector genomes or more, for example, 1×10^9 , 1×10^{10} , 1×10^{11} , 1×10^{12} , or 1×10^{13} vector genomes or more, in certain instances, 1×10^{14} vector genomes, but usually no more than 4×10^{15} vector genomes. In some embodiments, the viral vector is formulated into

any suitable unit dosage, including, without limitation, 1×10^7 , 1×10^8 , 1×10^9 , 1×10^{10} , 1×10^{11} , 1×10^{12} , or 1×10^{13} vector genomes or more. In some embodiments, the unit dosage is at most about 5×10^{15} vector genomes, *e.g.*, 1×10^{14} vector genomes or less, for example 1×10^{13} , 1×10^{12} , 1×10^{11} , 1×10^{10} , or 1×10^9 vector genomes or less, in certain instances 1×10^8 vector genomes or less, and typically no less than 1×10^8 vector genomes. In some embodiments, the unit dosage is 1×10^{10} to 1×10^{11} vector genomes. In some cases, the unit dosage is 1×10^{10} to 3×10^{12} vector genomes. In some embodiments, the unit dosage is 1×10^9 to 3×10^{13} vector genomes. In some embodiments, the unit dosage is 1×10^8 to 3×10^{14} vector genomes. In some embodiments, the vector is an rAAV vector comprising an AAV capsid and an expression cassette configured to express an anti-IGF1R antibody or antigen-binding fragment and a unit dosage is formulated at a concentration of at least, at most, exactly, or about 1×10^5 , 1×10^6 , 1×10^7 , 1×10^8 , 1×10^9 , 1×10^{10} , 1×10^{11} , 1×10^{12} , 1×10^{13} , 1×10^{14} , 1×10^{15} , or 1×10^{16} of the rAAV vector, or at a concentration within a range defined by any pair of concentrations described in this paragraph.

[0250] In some embodiments, the disclosure provides a vector comprising a plasmid configured to express anti-IGF1R antibody or antigen-binding fragment, and a unit dosage is formulated at a concentration of at least, at most, exactly, or about 1×10^7 , 5×10^7 , 1×10^8 , 5×10^8 , 1×10^9 , 5×10^9 , 1×10^{10} , 5×10^{10} , 1×10^{11} , 5×10^{11} , 1×10^{12} , 5×10^{12} , 1×10^{13} , 5×10^{13} , 1×10^{14} , 5×10^{14} , or 1×10^{15} of the plasmid (or at a concentration within a range defined by any pair of the foregoing concentrations).

[0251] In some embodiments, the unit dosage of pharmaceutical composition may be measured using multiplicity of infection (MOI). By MOI it is meant the ratio, or multiple, of vector or viral genomes of the rAAV vectors provided herein to the cells to which the nucleic acid may be delivered. In some embodiments, the MOI may be 1×10^6 . In some embodiments, the MOI may be 1×10^5 - 1×10^7 . In some cases, the MOI may be 1×10^4 - 1×10^8 . In some embodiments, recombinant viruses of the disclosure are at least about 1×10^1 , 1×10^2 , 1×10^3 , 1×10^4 , 1×10^5 , 1×10^6 , 1×10^7 , 1×10^8 , 1×10^9 , 1×10^{10} , 1×10^{11} , 1×10^{12} , 1×10^{13} , 1×10^{14} , 1×10^{15} , 1×10^{16} , 1×10^{17} , and 1×10^{18} MOI. In some embodiments, recombinant viruses of this disclosure are 1×10^8 to 3×10^{14} MOI. In some embodiments, recombinant viruses of the disclosure are at most about 1×10^1 , 1×10^2 , 1×10^3 , 1×10^4 , 1×10^5 , 1×10^6 , 1×10^7 , 1×10^8 , 1×10^9 , 1×10^{10} , 1×10^{11} , 1×10^{12} , 1×10^{13} , 1×10^{14} , 1×10^{15} , 1×10^{16} , 1×10^{17} , and 1×10^{18} MOI.

[0252] In some embodiments, the amount of pharmaceutical composition comprises about 1×10^8 to about 1×10^{15} rAAV vectors, about 1×10^9 to about 1×10^{14} rAAV vectors, about 1×10^{10} to about 1×10^{13} rAAV vectors, or about 1×10^{11} to about 3×10^{12} rAAV vectors.

[0253] In preparing the subject rAAV compositions, any host cells for producing rAAV vectors may be employed, including, for example, mammalian cells (*e.g.* 293 cells), insect cells (*e.g.* SF9 cells), microorganisms and yeast. Host cells can also be packaging cells in which the AAV rep and cap genes are stably maintained in the host cell or producer cells in which the rAAV vector genome is stably maintained and packaged. Exemplary packaging and producer cells are derived from SF-9, 293, A549 or HeLa cells. rAAV vectors are purified and formulated using standard techniques known in the art.

[0254] In some embodiments, the disclosure provides for use of an rAAV vector described herein in the manufacture of a medicament. In some embodiments, the disclosure provides for use of an rAAV vector described herein in the manufacture of a medicament for use in a method described herein.

[0255] In some embodiments, the disclosure provides a kit comprising an rAAV herein, and instructions for use. In some embodiments, the kit comprises an rAAV herein, and a package insert containing instructions for use of the kit. In some embodiments, the kit comprises an rAAV herein, and a pharmaceutically acceptable carrier, or a pharmaceutical composition comprising the rAAV and instructions for treating or delaying progression of a disease, disorder, or condition described herein in a subject in need thereof.

EXAMPLES

[0256] The following specific examples are to be construed as merely illustrative, and not limitative of the remainder of the disclosure in any way whatsoever.

Example 1: Preparation of an rAAV Vector for Expressing an Anti-IGF1R Construct

[0257] rAAV vector nucleotide comprising of an expression cassette encoding an antigen-binding fragment of an anti-IGF1R antibody was prepared. A vector map of a plasmid for this rAAV vector is depicted in **FIG. 1** and the nucleotide sequence of this vector is provided (SEQ ID NO: 23). In this case, the antigen-binding fragment of an anti-IGF1R antibody was an scFv construct comprising a heavy chain variable domain (SEQ ID NO: 2) connected to a light chain variable domain (SEQ ID NO: 6) via a GS linker sequence (SEQ ID NO: 42), as shown by **FIG. 1**. Putative CDR regions in both domains were predicted using the Chothia, North, and IMGT CDR definitions, as shown in Table 1. The “Chothia”, “North” and “IMGT” columns listed in Table 1 refer to alternative numbering systems. *See* Dondelinger, *M*, et al. “Understanding the significance and implications of antibody numbering and antigen-binding surface/residue definition.” *Frontiers in immunology* 9 (2018): 2278. As shown by **FIG. 1**, this

plasmid for this rAAV vector further illustrates the use of additional components described in the present application, e.g., a CMV enhancer and promoter (SEQ ID NO: 27), a WPRE sequence (SEQ ID NO: 29), a BGH polyA sequence (SEQ ID NO: 28), a 5' and 3' ITRs (SEQ ID NOs: 30 and 31, respectively).

[0258] This plasmid, similar plasmids, or other vectors described herein, may be used to transfect or transduce one or more cells, tissues, or organs of a human subject (e.g., an eye or an accessory lacrimal gland) in order to generate expression of anti-IGF1R antibodies, or antigen-binding fragments thereof, such as the exemplary scFv used in this example. Furthermore, this plasmid and others similar to it as described herein may be used to manufacture rAAV vectors which can be used to transduce cells, tissues, or organs of a human subject.

Example 2: IGF1R ELISA Assay

[0259] Exemplary scFv constructs based on the heavy chain variable domain and light chain variable domain sequences described herein were expressed from plasmid constructs in order to evaluate the binding ability and other properties of such constructs.

[0260] First, plasmid DNA for the VH-VL and the VL-VH scFv constructs were transfected into HeLa cells. The VH-VL construct comprised SEQ ID NO: 44 and the VL-VH construct comprised SEQ ID NO: 43. Both of these constructs further included a 6x His-tag sequence on the C-terminus for purification. Cells were cultured and conditioned media was collected to conduct an ELISA for human IGF1R (Abcam: ab275102). Transgene products from conditioned media were run as samples in a human IGF1R ELISA to assess the ability of the transgene product to bind the intended target, IGF1R.

[0261] The results of this IGF1R ELISA experiment are shown in **FIG. 3** and defined in Table 4 below. In **FIG. 3**, the wells associated with the two test constructs are annotated (blue rectangle); and the wells A-H in the left column represent a standard curve. These results demonstrate that both conformations of the scFv transgene (VH-VL and VL-VH) are able to bind to IGF1R. However, this analysis is not indicative of the level of binding or binding affinity.

Row	Standard Value	Test Samples
A	0.838-20,000 pg/mL	0.858-VH/VL
B	0.794-10,000 pg/mL	0.676-VL/VH
C	0.648-5,000 pg/mL	-
D	0.451-2,500 pg/mL	-
E	0.276-1,250 pg/mL	-
F	0.120-625 pg/mL	-
G	0.104-312.5 pg/mL	-
H	0.083-Blank	-

Table 4. Summary of the ELISA results shown in **FIG. 3**.

Example 3: Binding Affinity/Inhibition and Kinetics Studies

[0262] Conditioned media containing the VH-VL and VL-VH scFv constructs was sent to Gifford Bioscience Limited (Birmingham, United Kingdom) to conduct a Biacore® assay to determine the kinetic parameters of the transgene products binding to IGF1R (R&D Systems 305-GR-050). A Biacore T200® instrument was utilized to develop a method for determining the association rate (“ k_{on} ”) and affinity (“ k_D ”) for the binding of the native IGF1 ligand (R&D Systems® 291-G1-01M), the VH-VL and VL-VH scFv transgene products, and teprotumumab (Proteogenix® PX-TA1242-100), a commercially available human IGF1R monoclonal antibody. Two off-target receptors, Insulin Receptor A (“IR-A”) and Insulin Receptor B (“IR-B”), were utilized as experimental controls. Additionally, an assessment would be made as to the ability of these reagents to block IGF1 ligand from binding its endogenous receptor, IGF1R. Briefly, the Biacore T200 Surface Plasmon Resonance (“SPR”) sensor was prepared by desorbing, pre-cleaning the CM5 Series S sensor (Cytiva® 100530) and immobilizing the ligand (IGF1R, at 20 ug/mL in 10 mM sodium acetate pH 4.5). IGF1R proteins were immobilized on the sensor surface by amine coupling: The CM5 sensor surface was activated with 1-ethyl-3-(3-dimethylaminopropyl)-carbodiimide (“EDC”) - N-hydroxy-succinimide (“NHS”) followed by coupling of the IGF1R protein to the surface and then all unreacted EDC-NHS groups were inactivated with ethanolamine.

[0263] For binding and reproducibility studies, IGF1 and teprotumumab (supplied/reconstituted in PBS) were prepared at a single concentration in running buffer (HBS-T or PBS-T). Samples were injected in quadruplicate, then regenerated back to baseline, before commencing the next binding cycle. Buffer blank injections were included before and after each analyte quadruplicate set for blank cycle subtraction.

[0264] For kinetic affinity studies, IGF1 and teprotumumab were prepared as a 2-fold dilution series, at 10 concentrations, in PBS-T. Samples were injected in ascending concentration order, then regenerated back to baseline. Buffer blank injections were included before and after each analyte concentration series, for blank cycle subtraction to correct for any systematic sensor drift.

[0265] For the binding inhibition study, teprotumumab was prepared at a single concentration and IGF1 at two concentrations in PBS-T. For each binding cycle, teprotumumab was injected first, as a “capture” step, to allow the mAb to bind to the immobilized IGF1R surface. With the mAb still bound (i.e., without a regeneration step) IGF1 was then injected during a second “binding” step to bind to any remaining IGF1R that was not bound by the mAb.

The surface was then regenerated back to baseline. Buffer blank injections during the capture step were used instead of teprotumumab to measure the maximal IGF1 binding response in the absence of teprotumumab (100% binding or 0% inhibition). With teprotumumab injected and captured during the first “capture” step, buffer blank injections during the second “binding” step were used to provide a reference cycle without IGF1 binding for subtraction of the mAb dissociation rate. All sample combinations were repeated in duplicate.

[0266] For the calculation of kinetic affinities: IGF1 and teprotumumab kinetic affinity concentration series binding responses to IGF1R (Fc2), IR-A (Fc3) and IR-B (Fc4) were reference channel subtracted, i.e., Fc2-1, Fc3-1 and Fc4-1, to remove any bulk sample effect. Data were then buffer-blank cycle subtracted to remove any sensor drift. IGF1 binding data were fitted to a 2-state binding, with the refractive index (“RI”) correction factor set to a local constant value of “0.” Teprotumumab binding data were similarly reference channel subtracted then blank buffer cycle subtracted. Data were then fitted to a single site binding model, with the RI correction factor set to a local constant value of “0.”

[0267] For calculation of binding inhibition: IGF1 binding responses in the absence of teprotumumab (cycles 7-12) were “zeroed” in the Y-axis to the “baseline” report point averaged over 5 seconds before the start of the IGF1 injection. Data were reference channel subtracted, then buffer + buffer blank subtracted using cycle 7 as the blank. Data were then exported as a .csv file.

[0268] IGF1 binding responses in the presence of teprotumumab (cycles 13-18) were also zeroed to the “baseline” report point, then reference channel subtracted, then teprotumumab + buffer subtracted, using cycle 13 as the blank (to correct for the slow teprotumumab dissociation rate). Data were then exported as a .csv file.

[0269] The two exported datasets for IGF1 binding in the absence and presence of teprotumumab (using different blank cycles for correcting the different baseline drifts) were imported into Excel for further analysis. Data were plotted for the time interval of the IGF1 injection and dissociation (400 – 1200 seconds). Maximal binding response amplitudes for both duplicates of IGF1 at 100 nM and 10 nM, binding in the absence or presence of teprotumumab at 50 nM, were averaged over the final 5 seconds of the association phase. Inhibition of IGF1 binding in the presence of teprotumumab was calculated as the percentage reduction in binding response.

[0270] Results: The target receptor (IGF1R) and the related two off-target receptors (IR-A and IR-B) were immobilized to very similar levels on Fc2-4, ensuring equivalence of receptor density on the sensor surface. The reproducibility studies in HBS-T and PBS-T

demonstrated that binding responses for IGF1 and teprotumumab were more consistent using PBS-T as the running buffer. Additionally, there was less bulk effect by eliminating the HBS-T running buffer vs PBS sample buffer mismatch. Extending the stabilization period to 180 seconds also improved the recovery to baseline and hence binding response consistency. Determining the kinetic binding affinities for IGF1 binding to IGF1-R, IR-A and IR-B generated KDs of 5.1 nM, 89.6 nM and 272 nM, respectively. The binding response data fitted best to a two-state model. Without being bound to a theory, this may indicate negative cooperativity for IGF1 binding to all three receptors, similar to that known for insulin binding to IR-A and IR-B.

[0271] Kinetic affinity determination for teprotumumab fitted best to a single site model, with a KD of 96 pM (0.096 nM). Teprotumumab showed negligible binding to IR-A and IR-B. In the binding competition study, blocking the IGF1-R surface with teprotumumab at 50 nM reduced binding of IGF1 at 10 nM by 59% and at 100 nM by 54%, compared to IGF1 binding at the same concentrations in the absence of teprotumumab. However, blockade of the IGF1R surface by teprotumumab at 50 nM had not reached an equilibrium plateau even though the concentration was sufficiently high to do so (50 nM = 500 x KD, equivalent to >99% binding).

[0272] The results of this study are provided in **FIGs. 4-10**, and in Table 5 (summarizing the results shown in **FIG. 10**).

[0273] As illustrated by this data, IGF1 binding to IGF1R in PBS-T shows very similar reproducible consecutive responses, and these responses also settle to the original baseline after regeneration (**FIGS. 6-7**). Teprotumumab binding to IGF1R in PBS-T shows slightly more variable consecutive responses (**FIG. 7**). The responses quickly settle to the original baseline after regeneration. **FIGs. 8-9** show kinetic activity results for IGF-1 (**FIG. 8**) and for teprotumumab® (**FIG. 9**), and **FIG. 10** shows kinetic activity data for IGF-1 binding to IGF-1R with and without teprotumumab. The addition of teprotumumab resulted in 59% inhibition with IGF-1 at 10 nM and 54% inhibition with IGF-1 at 100 nM, as summarized by Table 5 below.

Analyte	Concentration (nM)	Response (RUs)	% Inhibition
IGF1 (no Teprotumumab)	10	40.1	-
IGF1 (no Teprotumumab)	100	147.8	-
IGF1 (+ Teprotumumab)	10	16.5	59%
IGF1 (+ Teprotumumab)	100	67.7	54%

Table 5. Inhibition of IGF1 binding by Teprotumumab.

Example 4: Binding Assays Using VL-VH and VH-VL scFv Constructs

[0274] The next set of studies were conducted to test the scFv transgene products (VL-VH and VH-VL) from unpurified conditioned media. Plasmid DNA encoding the respective VH-VL and VL-VH constructs was transfected into HeLa cells and cultured for 72 hours. The conditioned media was collected, frozen and shipped on dry ice for sample analysis. No purification or enrichment was performed on the conditioned media. Serial dilution of the experimental samples (VH-VL and VL-VH) was performed with Advanced DMEM, 5% fetal bovine serum (FBS), 1x penicillin-streptomycin and 1x Glutamax. Analytes were tested as described in Table 6 below.

Analytes (in Test Order)	Stock Solution	Final Concentration	Dilution	Serial Dilution	Concentrations (nM) (in Test Order)
IGF1	26.32 μ M	25 nM in PBS-T	1052.8-fold	2-fold (in PBS-T)	0.2, 0.39, 0.78, 1.56, 3.13, 6.25, 12.5, 25
Teprotumumab	2.03 μ M	50 nM in PBS-T	40.54-fold	2-fold (in PBS-T)	0.1, 0.2, 0.39, 0.78, 1.56, 3.13, 6.25, 12.5, 25, 50
VH-VL	4 nM	4 nM in media	None	2-fold (in media)	0.25, 0.5, 1, 2, 4
VL-VH	4 nM	4 nM in media	None	2-fold (in media)	0.25, 0.5, 1, 2, 4

Table 6. Concentration parameters for the analytes used in this experiment.

[0275] The target receptor (IGF1R) and the related two off-target receptors (IR-A and IR-B) were immobilized to similar levels to the study described in Example 3 on Fc2-4, ensuring equivalence of receptor density on the sensor surface. For this stage of the study, the receptors were immobilized at a lower density (< 4,000 RUs) than in Stage 1 (~ 10,000 RUs) to minimize re-binding of the samples during the dissociation phase of the binding cycle. This provides a more reliable measurement of the sample off-rate and subsequent calculation of the KD value.

[0276] The recombinant small protein VL-VH scFv (“VL-VH”) was selective for IGF1R. No binding responses were observed at the related receptors IR-A or IR-B. The concentration of the His-tagged proteins of interest in the VL-VH and VH-VL conditioned media samples was estimated to be 4 nM (based upon ELISA assay data from conditioned media samples). VL-VH binding responses fitted to a single-site binding model gave a KD value of 7.53 nM.

[0277] The unpurified second small protein, VH-VL scFv (“VH-VL”), did not bind to any of the three receptors.

[0278] With the lower level of receptor proteins immobilized on the surface in comparison to Stage 1, kinetic affinity values were also determined for binding of the endogenous ligand IGF1 and the mAb teprotumumab. The response for IGF1 binding to IGF1R is smaller in amplitude than that observed in Stage 1 but the KD value of 8.74 nM is similar to that determined previously (5.1 nM). Similarly, the teprotumumab binding response at IGF1R was smaller in amplitude but with a similar high affinity KD value (0.42 nM) compared to Stage 1 (0.096 nM).

These data suggest that unpurified transgene products present in conditioned media for the VL-VH scFv construct demonstrated a binding affinity of 8.74 nM to IGF-1R; whereas, the alternative scFv conformation (VH-VL), did not. Moreover, the association constant of the transgene product present in conditioned media demonstrated an association constant similar to the purified commercial product, teprotumumab. This data is described in Table 7 below.

Ligand	Model	K _a (1/Ms)	k _d (1/s)	k _{a2} (1/Ms)	k _{d2} (1/s)	K _D (M)	R _{max} (RU)	Chi ² (RU ²)
VL-VH	Single-site	2.46E+05	1.85E-03			7.53E-09	844.2	1.06
VH-VL						No binding		
IGF1	Two-state	1.25E+06	2.31E-02	3.90E-03	3.47E-03	8.74E-09	36.74	5.83
Teprotumumab	Single-site	3.93E+05	1.65E-04			4.19E-10	1536	144

Table 7. Binding data for unpurified samples of the VL-VH and VH-VL scFv constructs with respect to the IGF1R target receptor, in comparison to the IGF1 ligand and teprotumumab.

[0279] Additional results of this study are shown in **FIGs. 11-12**. Briefly, IGF1R (Fc2) was used as a ligand (flow channel), unpurified samples of the VL-VH and VH-VL constructs were tested at a concentration of 0.25 to 4 nM, and the maximum binding response observed was 107 RUs at 4 nM for the VL-VH construct (**FIG. 11**). No binding response was detected for the VH-VL construct (**FIG. 12**). Kinetic affinity data for the VL-VH and VH-VL constructs is summarized in Table 8 below.

Kinetic Affinity Fits								
Ligand	Model	K _a (1/Ms)	k _d (1/s)	k _{a2} (1/Ms)	k _{d2} (1/s)	K _D (M)	R _{max} (RU)	Chi ² (RU ²)
VL-VH	Single-site	2.46E+05	1.85E-03			7.53E-09	844.2	1.06
VH-VL						No binding		

Table 8. Kinetic affinity fits for unpurified samples of the VL-VH and VH-VL scFv constructs.

Example 5: Analysis of Purified VH-VL and VL-VH scFv Constructs

[0280] Purified VH-VL and VL-VH scFv constructs were subsequently produced in order to further study these constructs. In brief, plasmid DNA encoding VL-VH and VH-VL including the 6x His-tag was utilized to transfect 296-6E human embryonic kidney cells. Cells were cultured in Freestyle F17 (ThermoFisher® A1383501) supplemented with 0.1% Pluronic F-68 (Gibco® 24040-032), 4 nM GlutaMax (Gibco 35050-061), 25 ug/mL Geneticin (Gibco® 10131-027). Cells densities were maintained at 0.3-3.0 x 10⁶ cells/mL in shake flasks incubated

at 37 degrees Celsius at 5% CO₂ while shaking at 135 rpm. VL-VH plasmid was utilized to transfect 296-6E cells at a 600 mL scale and VH-VL plasmid was transfected at a 200 mL scale using PolyPlus PEIPro at a 1:1 w/v DNA:PEI ratio. A higher quantity of VL-VH was prepared due to the previous findings with unpurified protein suggesting additional studies would need to be undertaken with this transgene product given the previous superior binding data. Cell cultures were harvested 4-6 days post-transfection via centrifugation. The conditioned culture media supernatants were clarified by centrifugation at 9,300 x g for 30 minutes.

[0281] In order to purify these transgene products via the 6x His-tag, a 1 cm diameter Bio-Rad® Econo-column was loaded with Ni-NTA His-Bind (EMD Millipore) resin according to the resin manufacturer's recommendations. Each conditioned media supernatant was adjusted to 20 mM imidazole and top loaded into the Ni-NTA resin columns at 4 °C over 3-4 hours via gravity flow. The resin was washed with 20 column volumes of Buffer A (1x PBS (2.7 mM KCl, 1.7 mM KH₂PO₄, 136 mM NaCl, 10.1 mM Na₂HPO₄) 20 mM imidazole pH 7.4) and then elution of the His-tagged protein was performed with Buffer B (Buffer A + 280 mM imidazole pH 7.4). Fractions were collected and absorbance was measured on a spectrophotometer at a 280 nm wavelength prior to pooling. Select fractions were analyzed via SDS-PAGE and stained with Instant Blue. **FIG. 13** shows SDS-PAGE results for the affinity chromatography fraction for the VL-VH construct, and **FIG. 14** shows SDS-PAGE results for the affinity chromatography fraction for the VH-VL construct. A summary of the purification results is also provided in Tables 9 and 10. As depicted in lane 4 of the SDS-PAGE gels shown in **FIGs. 13-14**, the eluted fraction (IMAC Pool) has a single protein band at around 25 kDa. Eluted protein samples were then dialyzed in 1x PBS (2.7 mM KCl, 1.7 mM KH₂PO₄, 136 mM NaCl, 10.1 mM Na₂HPO₄) pH 7.4 to clear the imidazole.

VL-VH	Volume (mL)	Concentration (mg/mL)*	Total protein (mg)
Affinity pool	3.5	2.7	9.5
Post-dialysis pool	3.6	3.0	7.2
Final/Delivered protein	3.09	1.9	5.9

* Extinction coefficient A₂₈₀ of 2.1 = 1 mg/mL

Table 9. Analysis of the purified VL-VH scFv product generated in this experiment.

VH-VL	Volume (mL)	Concentration (mg/mL)*	Total protein (mg)
Affinity pool	0.9	0.6	0.5
Post-dialysis pool	1.0	0.5	0.5
Final/Delivered protein	0.2	0.8	0.2

* Extinction coefficient A₂₈₀ of 2.1 = 1 mg/mL

Table 10. Analysis of the purified VH-VL scFv product generated in this experiment.

[0282] Protein samples were flash frozen in liquid nitrogen and stored at -80°C and then shipped on dry ice for further Biacore analysis. Purified VL-VH and VH-VL samples were then run again in the Biacore analyses to assess binding affinities and the ability to block the IGF1 ligand from binding to its native receptor, IGF1R. The results demonstrated greater levels of binding and inhibition of IGF-1 from binding to IGF1R, with both VL-VH and VH-VL constructs displaying improved performance compared to commercially available teprotumumab. The results of these studies are provided in **FIGs. 15-23**, and in Tables 11-13. These results show that the VL-VH construct demonstrated significantly greater levels of binding and inhibition of IGF1 from binding to IGF1R, exceeding that of the commercially available and marketed teprotumumab control. While both VH-VL and VL-VH bound to IGF1R and inhibited IGF1 binding, there was a significant difference between the two, illustrating the importance of the cloning sequence and transgene product conformation.

[0283] **FIGs. 15-16** illustrate the kinetic affinity of the VL-VH (**FIG. 15**) and VH-VL (**FIG. 16**) constructs for IGF1R, respectively. For the assay shown in **FIGs. 15-16**, IGF1R (Fc4) was used as the immobilized ligand, the VL-VH and VH-VL constructs were tested at a concentration of 0.1 to 50 nM, and the maximum binding response observed was 430 RUs at 50 nM for the VL-VH construct and 359 RUs at 50 nM for the VH-VL construct. The kinetic affinity fits are described in Table 11 below.

Kinetic Affinity Fits								
	Model	k_a ($M^{-1}s^{-1}$)	k_d (s^{-1})	k_{a2} (s^{-1})	k_{d2} (s^{-1})	K_D (M)	Rmax (RU)	Chi ² (RU ²)
VL-VH	Two-State	9.26E+05	3.28E-03	2.16E-03	6.14E-04	7.85E-10	451.4	17.5
VH-VL	Two-State	8.03E+05	5.37E-03	1.13E-03	5.86E-04	2.29E-09	390.1	6.21

Table 11. Kinetic affinity fits for the purified VL-VH and VH-VL scFv constructs.

[0284] **FIGs. 17-20** provide binding response data illustrating the inhibition of IGF1 binding to IGF1R by purified VL-VH (**FIGs. 17-18**) and VH-VL (**FIGs. 19-20**) constructs. Tables 12-13 summarize the findings of these studies. As illustrated by this data, the commercially available human monoclonal antibody teprotumumab demonstrated 59% inhibition of IGF1 binding to IGF1R; whereas the VL-VH construct demonstrated 78.9% inhibition (**FIG. 18**) and the VH-VL construct demonstrated 54.6% inhibition (**FIG. 19**).

Inhibition of IGF1 Binding by VL-VH		
VL-VH Concentration (nM)	IGF1 Response (RUs)	% Inhibition
0	50.3	
1.56	45.9	8.7
3.13	43.0	14.4
6.25	36.3	27.9
12.5	24.9	50.4
25	10.6	78.9

Table 12. VL-VH inhibition of IGF1 binding to IGF1R at different test concentrations.

Inhibition of IGF1 Binding by VH-VL		
VH-VL Concentration (nM)	IGF1 Response (RUs)	% Inhibition
0	50.2	
3.13	44.9	10.7
6.25	42.0	16.4
12.5	35.7	29.0
25	33.3	33.8
50	22.8	54.6

Table 13. VH-VL inhibition of IGF1 binding to IGF1R at different test concentrations.

[0285] As illustrated by the studies discussed above, purified samples of the VH-VL and VL-VH scFv constructs performed better than unpurified equivalent samples. Without being bound by a theory, crude protein and potential competition from non-specific proteins likely interfered with the interaction between the VH-VL and VL-VH scFv constructs in the crude sample and so the results of that study (Example 4) are not inconsistent with the findings based on purified samples of the VH-VL and VL-VH scFv constructs (Example 5).

Example 6: Comparison of Purified VH-VL and VL-VH scFv Constructs Against Teprotumumab

[0286] The purified VH-VL and VL-VH constructs produced in Example 5 were evaluated against the commercially available and marketed product teprotumumab as a baseline. This study demonstrated that the purified VL-VH scFv transgene product has a significantly lower IC₅₀ and nominally lower EC₅₀ with respect to blocking IGF1 from binding to IGF1R and percent bound protein to IGF1R, respectively. As demonstrated by **FIGs. 21-23**, the VL-VH IC₅₀ was 11.49 nM in blocking IGF1 binding, the VH-VL IC₅₀ was 22.26 nM, and the teprotumumab IC₅₀ was 31.85 nM. Thus, both the VL-VH and VH-HL constructs displayed enhanced inhibition of IGF1 binding to IGF1R compared to commercially available teprotumumab. Furthermore, the EC₅₀ of the VL-VH construct binding to IGF1R was 21.74 nM, compared to teprotumumab's EC₅₀ of 24.09 nM.

[0287] Collectively these results show that the VL-VH scFv construct displayed superior results compared against teprotumumab (the human monoclonal antibody that is the active biological therapeutic in Tepezza[®] and approved by FDA for the treatment of Thyroid Eye Disease), while the alternative VH-VL scFv construct also demonstrated better results compared to teprotumumab, though not as good as VL-VH. Furthermore, VL-VH displayed superior results to VH-VL, demonstrating that the conformation of the variable domains is significant.

* * * *

[0288] In closing, it is to be understood that although aspects of the present specification are highlighted by referring to specific embodiments, one skilled in the art will readily appreciate that these disclosed embodiments are only illustrative of the principles of the subject matter disclosed herein. Therefore, it should be understood that the subject matter described herein is in no way limited to a particular compound, composition, article, apparatus, methodology, protocol, and/or reagent, etc., described herein, unless expressly stated as such. In addition, those of ordinary skill in the art will recognize that certain changes, modifications, permutations, alterations, additions, subtractions and sub-combinations thereof can be made in accordance with the teachings herein without departing from the spirit of the present specification.

[0289] Use of the terms “may” or “can” in reference to an embodiment or aspect of an embodiment also carries with it the alternative meaning of “may not” or “cannot.” As such, if the present specification discloses that an embodiment or an aspect of an embodiment may be or can be included as part of the inventive subject matter, then the negative limitation or exclusionary proviso is also explicitly meant, meaning that an embodiment or an aspect of an embodiment may not be or cannot be included as part of the inventive subject matter. In a similar manner, use of the term “optionally” in reference to an embodiment or aspect of an embodiment means that such embodiment or aspect of the embodiment may be included as part of the inventive subject matter or may not be included as part of the inventive subject matter. Whether such a negative limitation or exclusionary proviso applies will be based on whether the negative limitation or exclusionary proviso is recited in the claimed subject matter.

[0290] Notwithstanding that the numerical ranges and values setting forth the broad scope of the invention are approximations, the numerical ranges and values set forth in the specific examples are reported as precisely as possible. Any numerical range or value, however, inherently contains certain errors necessarily resulting from the standard deviation found in their respective testing measurements. Recitation of numerical ranges of values herein is

merely intended to serve as a shorthand method of referring individually to each separate numerical value falling within the range. Unless otherwise indicated herein, each individual value of a numerical range is incorporated into the present specification as if it were individually recited herein (e.g., any disclosure of a range with integer endpoints should be interpreted as also describing subranges defined by any pair of integers within the broader range).

[0291] The terms “a,” “an,” “the” and similar references used in the context of describing the present invention (especially in the context of the following claims) are to be construed to cover both the singular and the plural, unless otherwise indicated herein or clearly contradicted by context. Further, ordinal indicators—such as “first,” “second,” “third,” etc.—for identified elements are used to distinguish between the elements, and do not indicate or imply a required or limited number of such elements, and do not indicate a particular position or order of such elements unless otherwise specifically stated. All methods described herein can be performed in any suitable order unless otherwise indicated herein or otherwise clearly contradicted by context. The use of any and all examples, or exemplary language (e.g., “such as”) provided herein is intended merely to better illuminate the present invention and does not pose a limitation on the scope of the invention otherwise claimed. No language in the present specification should be construed as indicating any non-claimed element essential to the practice of the invention.

[0292] When used in the claims, whether as filed or added per amendment, the open-ended transitional term “comprising” (and equivalent open-ended transitional phrases thereof like including, containing and having) encompasses all the expressly recited elements, limitations, steps and/or features alone or in combination with unrecited subject matter; the named elements, limitations and/or features are essential, but other unnamed elements, limitations and/or features may be added and still form a construct within the scope of the claim. Specific embodiments disclosed herein may be further limited in the claims using the closed-ended transitional phrases “consisting of” or “consisting essentially of” in lieu of or as an amended for “comprising.” When used in the claims, whether as filed or added per amendment, the closed-ended transitional phrase “consisting of” excludes any element, limitation, step, or feature not expressly recited in the claims. The closed-ended transitional phrase “consisting essentially of” limits the scope of a claim to the expressly recited elements, limitations, steps and/or features and any other elements, limitations, steps and/or features that do not materially affect the basic and novel characteristic(s) of the claimed subject matter. Thus, the meaning of the open-ended transitional phrase “comprising” is being defined as encompassing all the

specifically recited elements, limitations, steps and/or features as well as any optional, additional unspecified ones. The meaning of the closed-ended transitional phrase “consisting of” is being defined as only including those elements, limitations, steps and/or features specifically recited in the claim whereas the meaning of the closed-ended transitional phrase “consisting essentially of” is being defined as only including those elements, limitations, steps and/or features specifically recited in the claim and those elements, limitations, steps and/or features that do not materially affect the basic and novel characteristic(s) of the claimed subject matter. Therefore, the open-ended transitional phrase “comprising” (and equivalent open-ended transitional phrases thereof) includes within its meaning, as a limiting case, claimed subject matter specified by the closed-ended transitional phrases “consisting of” or “consisting essentially of.” As such embodiments described herein or so claimed with the phrase “comprising” are expressly or inherently unambiguously described, enabled and supported herein for the phrases “consisting essentially of” and “consisting of.”

[0293] All patents, patent publications, and other publications referenced and identified in the present specification are individually and expressly incorporated herein by reference in their entirety for the purpose of describing and disclosing, for example, the compositions and methodologies described in such publications that might be used in connection with the present invention. These publications are provided solely for their disclosure prior to the filing date of the present application. Nothing in this regard should be construed as an admission that the inventors are not entitled to antedate such disclosure by virtue of prior invention or for any other reason. All statements as to the date or representation as to the contents of these documents is based on the information available to the applicants and does not constitute any admission as to the correctness of the dates or contents of these documents.

[0294] Lastly, the terminology used herein is for the purpose of describing particular embodiments only, and is not intended to limit the scope of the present invention, which is defined solely by the claims. Accordingly, the present invention is not limited to that precisely as shown and described.

SEQUENCE LISTING

SEQ ID NO:	Name	Sequence
1	Human Insulin-like growth factor 1 receptor isoform 1 precursor (GenBank Accession No. NP_000866.1, 1,367 aa; encoded by GenBank Accession No. NM_000875.5, 12,235 nt)	MKSGSGGGSPSTSLWGLLFLSAALSLWPTSGEICGPGIDIRNDYQQ KRLNCTVIEGYLHILLISKAEDYRSYRFPKLTVITEYLLLFVAG LESLGDLFPNLTVIRGWKLFYNYALVIFEMTNLKDIGLYNLRNITR GAIRIEKNADLCYLSTVDWSLILDVSNYYIVGNKPPKECGDLC TMEEKPMCEKTTINNEYNYRCWTTNRCQKMCPTCGKRACTENNEC CHPECLGSCSAPDNDTACVACRHYYYAGVCPACPPNTYRFE GWRCVDRDFCANILSAESSDSEGFVIHDGECMQECPSGFIRNGS QSMYCI PCEGPCPKVCEEEKKTKTIDSVTSAQMLQGCTIFKGNLL INIRRGNNIASELENFMGLIEVVTGYVKIRHSHALVSLSFLKNLRL ILGEEQL EGNYSFYVLDNQNLQQLDWDHRLNLTIKAGKMYFAFN PKLCVSEIY RMEEVGTGKGRQSKGDINTRNNGERASCESDVLHFT STTTSKNRIITWHRYRPPDYRDLSFTVYYKEAPFKNVTEYDGGD ACGSNSWNMVDVDLPPNKDVEPGILLHGLKPWTQYAVYVKA VTLTMVENDHIRGAKSEILYIRTNASVPSIPLDVL SASNSSSQLIVKWNPPSLPNGNSYIYIVRWQRQPDG YLYRHNYCSKDKIPIRKYADGTIDIEEVTENPKTE VCGGEKGPCCACPKTEAEKQAEKEEA EYRKVFENFLHNSIFVPRPERKRRDVMQVANT TMSRSRNTTAADTYNITDPEELETEY PFFESRV DNKERTVISNLRPFTLYRIDIHSCN HEAEKLGCSASN FVFARTMPAEGADDIPGPV TWEPRPENSIFLKWPEPENPNGLILMYEIKYGSQVE DQRECVSRQEYRKYGGA KLNRLNPGNYTARIQATSLSGNGSWTDPVFFYVQ AKTGYENFIHLIIALPVA VLLIVGGLVIMLYVFHRKRNNSRLGNGVLYASV NPEYFSAADVVPDEWEVAREKITMSRELGGSGFM VYEGVAKGVVKDEPETRVAIKTVNEAASMRERIE FLNEASVMKEFNCHHVVRLLGVVSQGGQPTLV IMELMTRGDLKSYLRSRPEMENN PVL APPSLSKMIQMA GEIADGMAYLNANKFVHRDLAARNCMVAEDFTVK IGDFGMTRDIYETDYYRKGGKLLPVRWMSPE SLKDGVTFTYSDVWSFGVVLWEIATLAEQPYQ GLSNEQVLR FVMEGGLLDKPDNCPDMLF ELMRMCWQYNPKMRPSFLEIISSIKEEMEPGF REVSYFYSEENKLPEPEELDLEPENMESVPLD PSASSSSLPLPDRHSGHKAENGPGPGVL VLRASFDERQPYAHMNGGRKNERALPLPQSSTC
2	Exemplary Anti-IGF1R Antibody Heavy Chain Variable Domain (Putative CDR1, CDR2, and CDR3 shown with emphasis, respectively)	QVELVESGGGVVQPGRSQRLS CAAS GFTFSSYGMH WVRQAPGKGLE WVA I IWF DGSSTYYADSVRG RFTISR DNSKNTLYLQMN SLRAEDTAVYFCAR ELGRRYFDL WGRGTLVSVSS
3	Exemplary Anti-IGF1R Antibody Heavy Chain, Putative CDR1	GFTFSSYGMH
4	Exemplary Anti-IGF1R Antibody Heavy Chain, Putative CDR2	IWF
5	Exemplary Anti-IGF1R Antibody Heavy Chain, Putative CDR3	ELGRRYFDL
6	Exemplary Anti-IGF1R Antibody Light Chain Variable Domain	EIVLTQSPATLSLSPGERATLSC RASQSVSSYLA WYQQKPGQAPRL LIY DASKRAT GIPARFSGSGGTDFTLTIS SLEPEDFAVYYC QQRS KWPEWT FGQGTKVESKRT

	(Putative CDR1, CDR2, and CDR3 shown with emphasis, respectively)	
7	Exemplary Anti-IGF1R Antibody Light Chain, Putative CDR1	RASQSVSSYLA
8	Exemplary Anti-IGF1R Antibody Light Chain, Putative CDR2	DASKRAT
9	Exemplary Anti-IGF1R Antibody Light Chain, Putative CDR3	QQRSKWPPWT
10	CAG promoter	ataacttacggtaaataatggcccgccctggctgaccgccaacgacccc cgccattgacgtcaataatgacgtatgttcccatagtaacgcaa tagggactttccattgacgtcaatgggtggagattttacggtaa tgcccacttggcagtacatcaagtgtatcatatgccaaagtacgccc cctattgacgtcaatgacggtaaataatggcccgccctggcattatgccc agtacatgaccttatgggactttcctacttggcagtacatctactc gagggcacgttctgcttactctccccatctccccccccctcccccac cccaattttgtattttattttttttaattttttgtgcagcgat gggggcggggggggggggggggcgcgccaggcggggcggggcgg ggcgagggggcggggcggggcggagcgagaggtgcggcggcagcca atcagagcggcgcgctccgaaagtctcttttatggcgaggcggcg gcggcggcgccctataaaaaagcgaagcgcgcgggcggggcgggagcg ggatcagccaccgcggtggcgccctagagtcgacgaggaactgaaa aaccagaaagttaactg
11	AAV2 VP1 (protein)	MAADGYLPDWLEDTLSEGIQWWKLKPGPPPKPAERHKDDSRGLV LPGYKYLGPFNGLDKGEPVNEADAAALEHDKAYDRQLDSGDNPYLK YNHADAEFQERLKEDTSFGGNLGRAVFQAKKRVLEPLGLVEEPVKT APGKKRPVEHSPVEPDSSSGTGKAGQQPARKRLNFGQTDADSVDP PQPLGQPPAAPSGLGTNTMATGSGAPMADNNEGADGVGNSSGNWHC DSTWMGDRVITTTSTRTWALPTYNNHLYKQISSQSGASNDNHYFGYS TPWGYFDENRFHCHFSPRDWQRLINNNWGFPRKRLNFKLFNIQVKE VTQNDGTTTIANNLSTVQVFTDSEYQLPYVLGSAHQGCLPPFPAD VFMVPQYGYLTLNNGSQAVGRSSFYCLEYFPSQMLRTGNNTFSYT FEDVPFHSSYAHSQSLDRLMNPLIDQYLYLSRTNTPSGTTTQSRL QFSQAGASDIRDQSRNWLPGPCYRQQRVSKTSADNNNSEYSWTGAT KYHLNGRDSLVPNGPAMASHKDDEEKFFPQSGVLI FGKQGSEKTNV DIEKVMITDEEEIIRTTNPVATEQYGSVSTNLQRGNRQAATADVNTQ GVLPGMVWQDRDVYLQGPIWAKIPHDTDGHFHPSPLMGGFGLKHPPP QILIKNTPVPANPSTTFSAAKFASFITQYSTGQVSVEIEWELQKEN SKRWNPFIQYTSNYNKSVNVDFTVDTNGVYSEPRPIGTRYLTRNL
12	AAV2 VP1 (nucleotide)	atggctgccgatgggttatcttccagattggctcgaggacactctct ctgaaggaataagacagtggtggaagctcaaacctggccaccacc accaaagcccgagagcggcagataaggacgacagcaggggtcttg ctctcctgggtacaagtacctcggacccttaacggactcgacaagg gagagccggtcaacgaggcagacgcccggccctcgagcagcagaaa agcctacgaccggcagctcgacagcggagacaacctgtacctcaag tacaaccacgcccagcggaggtttcaggagcgccttaaagaagata cgtcttttgggggcaacctcggacgagcagctctccaggcgaaaaa gaggggtcttgaaacctctgggcctggttgaggaacctgttaagacg gctccgggaaaaaagaggcggtagagcactctcctgtggagccag actcctcctcggaacgggaaaggcgggccagcagcctgcaagaaa aagattgaatttttggtcagactggagacgcagactcagtacctgac

		ccccagcctctcggacagccaccagcagccccctctggctctgggaa ctaatac gatggctacaggcagtgggcgaccaatggcagacaataa cgagggcgccgacggagtggttaattcctcgggaaattggcattgc gattccacatggatggcgacagagtcattaccaccagcaccgaa cctgggccctgccacctaacaaccacctctacaaacaaatttc cagccaatcaggagcctcgaacgacaatcactactttggctacagc accccttgggggtattttgacttcaacagattccactgccactttt caccacgtgactggcaaagactcatcaacaacaactggggattccg acccaagagactcaacttcaagctctttaacattcaagtcaaagag gtcacgcagaatgacggtagcagcagcattgccaataaccttacca gcacggttcagggtgtttactgactcggagtaccagctcccgtacgt cctcggctcggcgcatcaaggatgcctcccgcttcccagcagac gtcttcatgggtgccacagtatggatacctcacctgaacaacggga gtcaggcagtaggacgctcttcattttactgcctggagtactttcc ttctcagatgctgctaccggaaacaactttaccttcagctacact tttgaggacgttctttccacagcagctacgctcacagccagagtc tggaccgtctcatgaatcctctcatcgaccagtacgtgtattactt gagcagaacaacactccaagtgaaccaccacgcagtcaggctt cagttttctcaggccggagcagtgacattcgggaccagtc tagga actggcttctcggaccctgttaccgccagcagcagatcaaaagac atctgcgataacaacaacagtgaatactcgtggactggagctacc aagtaccacctcaatggcagagactctctggtgaatccgggcccgg ccatggcaaggccacaaggacgatgaagaaaagtttttctcagag cggggttctcatctttgggaagcaaggctcagagaaaacaaatgtg gacattgaaaaggctcatgattacagacgaagaggaaatcaggacaa ccaatcccgtggctacggagcagtatggttctgtatctaccaacct ccagagaggcaacagacaagcagctaccgcagatgtcaacacacaa ggcgttcttccaggcatggtctggcaggacagagatgtgtaccttc aggggcccctctgggcaaagattccacacacggacggacattttca cccctctcccctcatgggtggattcggacttaaacacctcctcca cagattctcatcaagaacaccccggtacctgcgaatccttcgacca ccttcagtgcggcaaagtttgcttctctcatcacacagtactccac gggacaggtcagcgtggagatcgagtgggagctgcagaaggaaaac agcaaacgctggaatccgaaattcagtacacttccaactacaaca agtctgttaatgtggactttactgtggacactaatgctgtattc agagcctcgccccattggcaccagatacctgactcgtaatctgttaa
13	AAV2 VP3 (protein)	MATGSGAPMADNNEGADGVGNSSGNWHDSTWMGDRVITTTSTRTWA LPTYNNHLYKQISSQSGASNDNHYFGYSTPWGYFDFNRFHCHFSPR DWQRLINNNWGFPRKRLNFKLFNIQVKEVTQNDGTTTIANNLSTV QVFTDSEYQLPYVLGSAHQGCLPPFPADVFMVPQYGYLTNLNGSQA VGRSSFYCLEYFPSQMLRTGNNFTFSYTFEDVPFHSSYAHQSILDR LMNPLIDQYLYLSRTNTPSGTTTQSRLQFSQAGASDIRDQSRNWL PGPCYRQQRVSKTSADNNNSEYSWTGATKYHLNGRDSLVPNPGPAMA SHKDDEEKFFPQSGVLI FGKQGSEKTNVDIEKVMITDEEIRTTNP VATEQYGSVSTNLQRGNRQAATADVNTQGVLPGMVWQDRDVYLQGP IWAKIPHTDGHFHPSPLMGGFGLKHPPPQILIKNTVPANPSTTFS AAKFASFITQYSTGQVSVEIEWELQKENS KRWNPEIQYTSNKNKSV NVDFTVDTNGVYSEPRPIGTRYLTRNL
14	AAV2 VP3 (nucleotide)	atggctacaggcagtgggcgaccaatggcagacaataacgagggcg ccgacggagtggttaattcctcgggaaattggcattgcgattccac atggatggcgacagagtcattaccaccagcaccgaaacctgggcc ctgccacctaacaaccacctctacaaacaaatttcagccaat caggagcctcgaacgacaatcactactttggctacagcacccttg ggggtattttgacttcaacagattccactgccacttttcaccacgt gactggcaaagactcatcaacaacaactggggattccgaccaaga

		gactcaacttcaagctctttaacattcaagtcaaagaggtcacgca gaatgacggtacgacgagattgccaataaccttaccagcaggtt caggtgtttactgactcggagtagcagctcccgtacgtcctcggt cggcgcatcaaggatgcctcccgcgttcccagcagacgtcttcat ggtgccacagtatggatacctcaccctgaacaacgggagtcaggca gtaggacgctcttcattttactgcctggagtactttccttctcaga tgctgctgacccggaaacaactttaccttcagctacacttttgagga cgttcctttccacagcagctacgctcacagccagagtctggaccgt ctcatgaatcctctcatcgaccagtacctgtattacttgagcagaa caaacactccaagtgggaaccaccacgcagtcagggttcagttttc tcaggccggagcgagtgacattcgggaccagtcaggaactggcctt cctggaccctgttaccgcccagcagcagtagtatcaaagacatctgcgg ataacaacaacagtgataactcgtggactggagctaccaagtacca cctcaatggcagagactctctggtgaatccgggcccggccatggca agccacaaggacgatgaagaaaagttttttctcagagcgggttc tcattcttgggaagcaaggctcagagaaaacaaatgtggacattga aaaggtcatgattacagacgaagaggaaatcaggacaaccaatccc gtggctacggagcagtagtggttctgtatctaccaacctccagagag gcaacagacaagcagctaccgcagatgtcaacacacaaggcgttct tccaggcatggtctggcaggacagagatgtgtaccttcaggggccc atctgggcaaagattccacacacggacggacattttcaccctctc ccctcatgggtgattcggacttaaacaccctcctccacagattct catcaagaacaccccgtacctgcgaatccttcgacccttcagtg gcggcaaagtttgcttctctcatcacacagtactccacgggacagg tcagcgtggagatcgagtgaggagctgcagaaggaaaacagcaaacg ctggaatcccgaattcagtagacttccaactacaacaagtctgtt aatgtggactttactgtggacactaatggcgtgtattcagagcctc gccccattggcaccagatacctgactcgtaatctgtaa
15	AAV5 (protein)	MSFVDHPPDWLEEVGEGLEFLGLEAGPPKPKPNQQHQDQARGLVL PGYNYLGPGNGLDRGEPVNRADDEVAREHDISYNEQLEAGDNPLYKY NHADAEFQEKLADDTSFGGNLGKAVFQAKKRVLFPGLVEEGAKTA PTGKRIDDHFPRKKARTEEDSKPSTSSDAEAGPSGSQQLQIPAPQ ASSLGADTMSAGGGPLGDNNQGADGVGNASGDWHCDSTWMGDRVV TKSTRTWVLPSPYNNHQYREIKSGSVDGSNANAYFGYSTPWGYFDVN RFHSHWSPRDWQRLINNYWGFPRSLRVKIFNIQVKEVTQDSTTT IANNLTSTVQVFTDDDYQLPYVVGNGTEGCLPAFPQPQVFTLPQYGY ATLNRDNTENPTERSFFCLEYFPSKMLRTGNNFETYNFEEVPFH SSFAPSQNLFKLANPLVDQYLYRFVSTNNTGGVQFNKNLAGRYANT YKNWFPGPMGRTQGWNLGSGVNRASVSATFATNRMELEGASYQVPP QPNGMTNNLQGSNTYALENTMIFNSQPANPGTTATYLEGNMLITSE SETQPVNRVAYNVGGQMATNNQSSTTAPATGTYNLQEIIVPGSVWME RDVYLQGPWAKIPETGAHFHPSAMGGFGLKHPPMMLIKNTPVP GNITSFSDVPVSSFITQYSTGQVTVMEMEWELKKENSKRWNPEIQYT NNYNDPQFVDFAPDSTGEYRTRTRPIGTRYLTRPL
16	AAV5 (nucleotide)	atgtcttttgttgatcaccctccagattggttggaagaagttggtg aaggcttcgcgaggttttttgggccttgaagcggggccaccgaaacc aaaacccaatcagcagcatcaagatcaagcccgtggtcttctgtctg cctgggtataactatctcggaccggaaacggctctcgatcgagagag agcctgtcaacagggcagacgaggtcgcgcgagagcacgacatctc gtacaacgagcagcttgaggcgggagacaacccctacctaagtac aaccacgcgagcgcgaggtttcaggagaagctcgccgacgacacat ccttcgggggaaacctcggaaggcagtccttcaggccaagaaaag ggttctcgaaccttttggcctggttgaagaggggtgctaagacggcc cctaccggaaagcggatagacgaccactttccaaaagaaaagagg ctcggaccgaagaggactccaagccttcacctcgtcagacgcccga

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17	AAV8 (protein)	MAADGYLPDWLEDNLSEGIREWALKPGAPKPKANQQKQDDGRGLV LPGYKYLGFENGLDKGEPVNAADAAALEHDKAYDQQLQAGDNPYLR YNHADAEFQERLQEDTSFGGNLGRAVFQAKKRVLEPLGLVEEGAKT APGKKRPVEPSPQRSPTSSTGIGKKGQPPARKRLNFGQTDSESV DPQPLGEPFAAPSGVGPNTMAAGGGAPMADNNEGADGVSSSGNWH CDSTWLGDRVITSTRTWALPTYNNHLYKQISNGTSGGATNDNTYF GYSTPWGYFDFNRFHCHFSRQWRLINNNWGFPRKLSFKLFNIQ VKEVTQNEGTKTIANNLTSTIQVFTDSEYQLPYVLGSAHQGCLPPF PADVFMIPQYGYLTLNNGSQAVGRSSFYCLEYFPSQMLRTGNFQF TYTFEDVPFHSSYAHSQSLDRLMNPLIDQYLYLSRTQTTGGTANT QTLGFSQGGPNTMANQAKNWLPGPCYRQQRVSTTTGQNNNSNFAWT AGTKYHLNGRNSLANPGIAMATHKDDEERFFPSNGILIFGKQNAAR DNADYSDVMLTSEEEIKTTNPVATEEYGIVADNLQQQNTAPQIGTV NSQALPGMVWQNRDVYLQGPWAKIPHTDGNFHPSPMLMGFGGLKH PPQILIKNTVPADPPTTFNQSKLNSFITQYSTGQVSVEIEWELQ KENSKRWNPFIQYTSNYYKSTSVDFAVNTEGVYSEPRPIGTRYLTR NL

18	AAV8 (nucleotide)	atggctgccgatgggttatcttccagattggctcgaggacaacctct ctgagggcatttcgagagtggtggcgctgaaacctggagccccgaa gccccaaagccaaccagcaaaagcaggacgacggccggggtctggtg cttcctggctacaagtacctcgacccttcaacggactcgacaagg gggagcccgctcaacgcggcgacgcagcgccctcgagcacgacaa ggcctacgaccagcagctgcaggcgggtgacaatccgtacctgcgg tataaccacgcccagcgcgagtttcaggagcgtctgcaagaagata cgtcttttgggggcaacctcgggcgagcagtcctccaggccaagaa gcggggttctcgaacctctcgggtctggttgaggaaggcgctaagacg gctcctggaaagaagagaccggtagagccatcacccagcgttctc cagactcctctacgggcatcggaagaaaggccaacagcccgccag aaaaagactcaattttgggtcagactggcgactcagagtcagttcca gacctcaacctctcggagaacctccagcagcgccctctggtgtgg gacctaatacaatggctgcaggcgggtggcgaccaatggcagacaa taacgaaggcgccgacggagtggttagttcctcgggaaattggcat tgcgattccacatggctgggcgacagagtcaccaccagcacc gaacctgggcccctgccacctacaacaaccacctctacaagcaaat ctccaacgggacatcgggaggagccaccaacgacaacacctacttc ggctacagcaccctcgggggtattttgactttaacagattccact gccacttttcaccacgtgactggcagcgactcatcaacaacaactg gggattccggcccaagagactcagcttcaagctcttcaacatccag gtcaaggaggtcacgcagaatgaaggcaccagaccatcgccaata acctcaccagcaccatccaggtgtttacggactcggagtcaccagct gccgtacgttctcggctctgcccaccagggtgcctgcctccgttc ccggcggacgtgttcattgattccccagtagcgtacctaactca acaacggtagtcaggccgtgggacgctcctccttctactgcctgga atactttccttcgcagatgctgagaaccggcaacaacttccagttt acttacaccttcgaggacgtgcctttccacagcagctacgcccaca gccagagcttgagaccggctgatgaatcctctgattgaccagtacct gtactacttgtctcggactcaaacaacaggaggcacggcaaatacg cagactctgggttcagccaagggtgggcctaatacatggccaatc aggcaagaactggctgccaggaccctgttaccgccaacaacgcgt ctcaacgacaaccgggcaaaacaacaatagcaactttgcctggact gctgggaccaaataccatctgaatggaagaaattcattggttaatc ctggcatcgctatggcaacacacaaagacgagagcggtttttt tcccagtaacgggatcctgatttttggcaaaaaatgctgccaga gacaatgaggattacagcgatgtcatgctcaccagcgaggaagaaa tcaaaaccactaaccctgtggctacagaggaatacgggtatcggtggc agataacttgcagcagcaaaacacggctcctcaaattggaactgtc aacagccagggggccttaccgggtatggtctggcagaaccgggacg tgtacctgcaggggtcccatctgggccaagattcctcacacggacgg caacttccacccgtctcggctgatgggcggcctttggcctgaaacat cctccgcctcagatcctgatcaagaacacgcctgtacctgcggatc ctccgaccaccttcaaccagtcaaagctgaactctttcatcacgca atacagcaccggacaggtcagcgtggaattgaatgggagctgcag aaggaaaaacagcaagcgtggaaccccgagatccagtacacctcca actactacaaatctacaagtgtggactttgctgttaatacagaagg cgtgtactctgaaccccgccccattggcaccggttacctcaccgt aatctg
19	AAV9 (protein)	MAADGYLPDWLEDNLSEGIREWALKPGAPQPKANQQHQDNARGLV LPGYKYLPGNGLDKGEPVNAADAAALEHDKAYDQQLKAGDNPYLK YNHADAEFQERLKEDTSFGGNLGRAVFQAKRRLLEPLGLVEEAAKT APGKKRPVEQSPQEPDSSAGIGKSGAQPAKKRLNFGQTGDTESEVPD PQPIGEPPAAPSGVGSMTMASGGGAPVADNNEGADGVSSSSGNWHC DSQWLGRVITTTSTRTWALPTYNNHLYKQISNSTSGGSSNDNAYFG

		YSTPWGYFDENRFHCHFSRQRLINNNWGRPKRLNFKLFNIQV KEVTDNNGVKTIANNLSTVQVFTDSYQLPYVLGSAHEGCLPPFP ADVFMIPQYGYLTLNDGSQAVGRSSFYCLEYFPSQMLRTGNNFQFS YEFENVFHHSSYAHSQSLDRLMNPLIDQYLYYLSKTINGSGQNQQT LKFSVAGPSNMAVQGRNYIPGPSYRQQRVSTTVTQNNNSEFAWPGA SSWALNGRNSLMNPGPAMASHKEGEDRFFPLSGSLIFGKQGTGRDN VDADKVMITNEEEIKTTNPVATESYQVATNHQSAQAQAQTGWVQN QGILPGMVWQDRDVYLQGPWAKIPHTDGNFHPSPLMGGFGMKHPP PQILIKNTFPVPADPPTAFNKKDLNSFITQYSTGQVSVEIEWELQKE NSKRWNPEIQYTSNYYKSNNVEFAVNTEGVYSEPRPIGTRYLTRNL
20	AAV9 (nucleotide)	atggctgccgatgggtatcttccagattggctcgaggacaacctta gtgaagggaattcgcgagtggtgggctttgaaacctggagccctca acccaaggcaaatcaacaacatcaagacaacgctcgaggtctgtg cttccgggttacaaataccttgaccggcaacggactcgacaagg gggagccggtcaacgcagcagacgcggcgccctcgagcagacaaa ggcctacgaccagcagctcaaggccggagacaacccgtacctcaag tacaaccacgcccagcgcgagttccaggagcggctcaaagaagata cgtcttttgggggcaacctcgggagcagctcttccaggccaaaaa gaggcttcttgaacctcttggctctggttgaggaagcggctaagacg gctcctggaaagaagaggcctgtagagcagctctcctcaggaaccgg actcctccgcggtattggcaaatcgggtgcacagcccgtaaaaa gagactcaatttcgggtcagactggcgacacagagtcagctccagac cctcaaccaatcggagaacctcccgcagccccctcaggtgtgggat ctcttacaatggcttcaggtgggtggcgaccagtggcagacaataa cgaagggtgccgatggagtggttagttcctcgggaaattggcattgc gattcccaatggctgggggacagagtcataccaccagcaccggaa cctgggcccctgccacctacaacaatcacctctacaagcaaatctc caacagcacatctggaggatcttcaaatgacaacgcctacttcggc tacagcaccctcgggggtattttgacttcaacagattccactgcc acttctcaccacgtgactggcagcagactcatcaacaacaactgggg attccggcctaagcgactcaacttcaagctcttcaacattcaggtc aaagagggttacggacaacaatggagtcagaccatcgccaataacc ttaccagcaggtccaggtcttcacggactcagactatcagctccc gtacgtgctcgggtcgggtcacgagggctgcctcccgccgttccca gcggacgttttcatgattcctcagtaggggtatctgacgtctaatg atggaagccaggccgtgggtcggttcgtccttttactgcctggaata tttcccgctcgaaatgctaagaacgggtaacaacttccagttcagc tacgagtttgagaacgtacctttccatagcagctacgctcacagcc aaagcctggaccgactaatgaatccactcatcgaccaatacttgta ctatctctcaaagactattaacggttctggacagaatcaacaaacg ctaaaattcagtggtggcgaccagcaacatggctgtccaggga gaaactacatacctggaccagctaccgacaacaacgtgtctcaac cactgtgactcaaaacaacaacagcgaatttgcttgccctggagct tcttcttgggtctcaatggacgtaataagcttgatgaatcctggac ctgctatggccagccacaagaaggagaggttcttcttcttctt gtctggtatctttaaatttttggcaacaaggaaactggaagagacaac gtggatgcggacaaagtcatgataaccaacgaagaagaattaaaa ctactaaccggtagcaacggagtcctatggacaagtggccacaaa ccaccagagtgcccaagcacaggcgagaccggctgggttcaaaac caagggaatacttccgggtatgggttggcaggacagagatgtgtacc tgcaaggacccatttggggccaaaattcctcacacggacggcaactt tcacccttctccgctgatgggaggggttggaatgaagacccgcct cctcagatcctcatcaaaaacacacctgtacctgcggatcctcaa cggccttcaacaaggacaagctgaactctttcatcaccagtatctc tactggccaagtcagcgtggagatcgagtgggagctgcagaaggaa

		aacagcaagcgctggaacccggagatccagtacacttccaactatt acaagtctataaatggtgaatttgctgttaataactgaaggtgtata tagtgaaccccgccccattggcaccagatacctgactcgtaatctg t
21	Exemplary Anti-IGF1R Antibody Heavy Chain cDNA	CAGGTGGAACCTGGTTCGAGAGCGGAGGCGGCGTGGTGCAGCCTGGCA GAAGCCAGAGACTGAGCTGCGCCGCTAGCGGCTTCACCTTCAGCAG CTACGGCATGCACTGGGTTCAGACAGGCCCTGGCAAGGGCTGGAA TGGGTGGCCATCATCTGGTTCGACGGCAGCTCAACATACTACGCCG ACAGCGTGCAGGGGCCGTTTACCATCAGCAGAGATAATAGCAAGAA CACCTGTACCTGCAGATGAACAGCCTTAGAGCCGAGGACACCGCC GTGTACTTCTGCGCCCGGAACTGGGCAGGCGGTACTTTGACCTGT GGGCAGAGGCACACTGGTGTCCGTGTCCAGC
22	Exemplary Anti-IGF1R Antibody Light Chain cDNA	GAGATCGTGCTGACCCAGAGCCCTGCTACACTGAGCCTGAGCCCCG GCGAGCGGGCTACCCTGTCTTGTAGAGCCAGCCAAAGCGTGTCTTC TTATCTGGCCTGGTACCAGCAAAAGCCTGGACAGGCTCCTAGACTG CTGATCTACGACGCCAGCAAAAGAGCCACCGGCATCCCCGCCAGAT TCTCTGGCAGCGGCAGCGGCACCGACTTCACACTGACAATTAGCTC CCTCGAGCCCCGAGGATTTGCGCCGTGTACTACTGCCAGCAGCGCTCC AAATGGCCACCATGGACCTTTGGCCAGGGCACCAAGGTGGAATCTA AGCGGACC
23	pAAV-VH-VL-Stuffer- 2 Plasmid Sequence (as shown in FIG.1) (7191 bp) Exemplary anti-IGF1R antibody heavy chain shown with bold emphasis; exemplary anti-IGF1R antibody light chain shown underlined; exemplary linker sequence shown lower-case and italicized.	TCGCGCGTTTCGGTGATGACGGTGAAAACCTCTGACACATGCAGCT CCCGGAGACGGTCACAGCTTGTCTGTAAGCGGATGCCGGGAGCAGA CAAGCCCGTCAGGGCGCGTCAGCGGGTGTGGCGGGTGTCCGGGCT GGCTTAACATATGCGGCATCAGAGCAGATTGTACTGAGAGTGCACCA TATGCGGTGTGAAATACCGCACAGATGCGTAAGGAGAAAAATACCGC ATCAGGCGCCATTTCGCCATTTCAGGCTGCGCAACTGTTGGGAAGGGC GATCGGTGCGGGCCTCTTCGCTATTACGCCAGCTGGCGAAAGGGGG ATGTGCTGCAAGGCGATTAAAGTTGGTAACGCCAGGGTTTTCCAG TCACGACGTTGTAAAACGACGGCCAGTGAATTGACGCGTATTGGGA TTGACCTAACGAAAAGTGAACCTTCATAATACGTGCTGTCCACGCA CATGGTAGATTTGGACAAAATTGAATGGAGTCTGATCAACCTTCAC ACCGATCTAGATATCGCGCGCTCGCTCGCTCACTGAGGCCGCCCCG GCAAAGCCCCGGGCGTCGGGCGACCTTTGGTCGCCCCGGCTCAGTGA GCGAGCGAGCGCGCAGAGAGGGAGTGGCCAACTCCATCACTAGGGG TTCCTTGTAGTTAATGATTAAACGATCTTTACAAATTCAGCCAGG TGATTTCAACAAATTTTGTGACGATTTAGGCGCACTATCCCCATA ACTACAAATTAGAAAATAGCGTTTCCTTGACACTAGTCTAGTTATTA ATAGTAATCAATTACGGGGTCATTAGTTCATAGCCCATATATGGAG TTCCGCGTTACATAACTTACGGTAAATGGCCCGCTGGCTGACCGC CCAACGACCCCCGCCCATTGACGTCAATAATGACGTATGTTCCCAT AGTAACGCCAATAGGGACTTTCCATTGACGTCAATGGGTGGACTAT TTACGGTAAACTGCCCACTTGGCAGTACATCAAGTGTATCATATGC CAAGTACGCCCCCTATTGACGTCAATGACGGTAAATGGCCCGCCTG GCATTATGCCAGTACATGACCTTATGGGACTTTCCCTACTTGGCAG TACATCTACGTATTAGTCATCGCTATTACCATGGTGATGCGGTTTT GGCAGTACATCAATGGGCGTGGATAGCGGTTTGACTCACGGGGATT TCCAAGTCTCCACCCCATTTGACGTCAATGGGAGTTTTGTTTTGGCAC CAAAATCAACGGGACTTTCCAAAATGTCGTAACAACCTCCGCCCCAT TGACGCAAAATGGGCGGTAGGCGGTGACGGTGGGAGGTCTATATAAG CAGAGCTCTCTGGCTAACTAGAGAACCCACTGCTTACTGGCTTATC GAAATAAGCTTTCTCAGGGGAGATCTCGTTTAGTGAACCGTCAGAT CCTCACTCTCTTCCGCATCGCTGTCTGCGAGGGCCAGCTGTTGGGC TCGCGGTTGAGGACAAACTCTTCGCGGTCTTTCCAGTACTCTTGGG TCGGAAACCCGTCGGCCTCCGAACGGTACTCCGCCACCGAGGGACC TGAGCGAGTCCGCATCGACCGGATCGGAAAACCTCTCGAGAAAGGC

		GTCTAACCAGTCACAGTCGCAAGGTAGGCTGAGCACCGTGGCGGGC GGCAGCGGGTGGCGGTGCGGGTTGTTTCTGGCGGAGGTGCTGCTGA TGATGTAATTAAAGTAGGCGGTCTTGAGACGGCGGATGGTCGAGGT GAGGTGTGGCAGGCTTGAGATCCAGCTGTTGGGGTGAGTACTCCCT CTCAAAGCGGGCATTACTTCTGCGCTAAGATTGTCAGTTTCCAAA AACGAGGAGGATTTGATATTCACCTGGCCCCGATCTGGCCATACACT TGAGTGACAATGACATCCACTTTGCCTTTCTCTCCACAGGTGTCCA CTCCCAGGTCCAAGTTTAAACGCCGCCGCCATGAGCGTTCTTACAC AGGTGCTGGGACTGCTGCTGCTGTGGCTGACCGATGCCAGATGCCA GGTGGAACTGGTTCGAGAGCGGAGGCGGCGTGGTCAGCCTGGCAGA AGCCAGAGACTGAGCTGCGCCGCTAGCGGCTTACCTTCAGCAGCT ACGGCATGCACCTGGGTTCAGACAGGCCCTGGCAAGGGCTGGAAATG GGTGGCCATCATCTGGTTCGACGGCAGCTCAACATACTACGCCGAC AGCGTGCAGGGGCCGGTTTACCATCAGCAGAGATAATAGCAAGAACA CCCTGTACCTGCAGATGAACAGCCTTAGAGCCGAGGACACCGCCGT GTACTTCTGCGCCCCGGAACTGGGCAGGCGGTACTTTGACCTGTGG GGCAGAGGCACACTGGTGTCCGTGTCCAGC<i>ggcggaggcggttctg</i> <i>gcggcgagggtccggcgaggaggcg</i>GAGATCGTGCTGACCCAGAG CCTGTCTACACTGAGCCTGAGCCCCGCGCAGCGGGCTACCTGTCT TGTAGAGCCAGCCAAAGCGTGTCTTCTTATCTGGCCTGGTACCAGC <u>AAAAGCCTGGACAGGCTCCTAGACTGCTGATCTACGACGCCAGCAA</u> <u>AAGAGCCACCGGCATCCCCGCCAGATTCTCTGGCAGCGGCAGCGGC</u> <u>ACCGACTTCACACTGACAATTAGCTCCCTCGAGCCCCGAGGATTTTCG</u> <u>CCGTGTACTACTGCCAGCAGCGCTCCAAATGGCCACCATGGACCTT</u> <u>TGGCCAGGGCACCAAGGTGGAATCTAAGCGGACCTAGTCCGTGTAA</u> TCAACCTCTGGATTACAAAATTTGTGAAAGATTGACTGATATTCTT AACTATGTTGCTCCTTTTACGCTGTGTGGATATGCTGCTTTAATGC CTCTGTATCATGCTATTGCTTCCCGTACGGCTTTTCGTTTTCTCCTC CTGTATATAATCCTGGTTGCTGTCTCTTTATGAGGAGTTGTGGCCC GTTGTCCGTCAACGTGGCGTGGTGTGCTCTGTGTTTGTGACGCAA CCCCCACTGGCTGGGGCATTGCCACCACCTGTCAACTCCTTTCTGG GACTTTCGCTTTCCCCCTCCCTATCGCCACGGCAGAACTCATCGCC GCCTGCCTTGCCCGCTGCTGGACAGGGGCTAGGTTGCTGGGCACTG ATAATTCCGTGGTGTGTGCGGGGAAGCTGACGTCCGACTGTGCCCTT CTAGTTGCCAGCCATCTGTTGTTTGGCCCTCCCCCGTGCCTTCCTT GACCCTGGAAGGTGCCACTCCCCTGTCTTTCCCTAATAAAATGAG GAAATTGCATCGCATTGTCTGAGTAGGTGTCATTCTATTCTGGGGG GTGGGGTGGGGCAGGACAGCAAGGGGGAGGATTGGGAAGACAATAG CAGGCATGCTGGGGATGCGGTGGGCTCTATGGAAGACAACCAGCAT CTCGCGTCTTGCCCTAACCCCCCTACATGCTGTTATAGAGAAACAGT GGAAACCCGGTGCCAGGCGATGGAATGTCCTTAACTCTGGCAGGAA ATTAAAGGGAACGTATATACAACGCAAAGAAGCTGGAAAATTGGCG AGGGAATCCTGTTTCTGTCTATCCAAGAATGGGCATGAGGTGGCAA CCGTCGTGCTAGCGTACAGGGTGCACTTTGTAACCATTTGGGACAC CGGACACTCGCTGTTTTTCGAAATTACCCTTTAAGCGCGGGTATTGA ACCAGGCTTATGCCCAAGATCGTAGCAAGCAGACTCAAACAAGATA TATTTTGGCCGCCTTACAGACGAACTAGTTGGAGGTTATGGAGCA TACTATCACGTGGGCGGCCACTGGTGAGTTACTACACCCAGGGGC AACGTTGATGCTCCTAAAAAATCTGGCTGGACGCAAGCCGTAACA CCCGTGTCACTTCATAATCGTTTGCAATTCAGGGCTTGATCTACAC TGGATTGCCATTCTCTCAAAGTATTATGCAGGACGGCGTGCGCGTT CCATGTAAACCTGTCTAACTTACCTGAGACTAGTTGGAAGTGTGG CTAGATCTTTGCTCACGCATCTAGTCGGTCCACGTTTGGTTTTTAA GATCCAATGATCTTCAAAACGCTGCAAGATTCTCAACCTGCTTTAC TAAGCGCTGGGTCCCTACTCCAGCGGGATTTTTTATCTAAAGACGAT
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	GAGAGGAGTATTCGTGACACCACATAGCTTTTCATGTCCTGATCGGA AGGATCGTTGGCGCCCGACCCCTCAGACTCTGTAGTGAGTTCTATGT CCGAGCCATTGCATGCGAGATCGGTAGATTGATAGGGGATACAGAA TATCCCTGGATGCAATAGACGGACAGCTTGGTATCCTAAGCGTAGT CGCGCGTCCGAACCCAGCTCTACTTTAGAGGCCCTCGGATTCTGGTG CCCGCAGGCCCGCAGAACCGATTAGGGGCATGTACAACAATATTTAT TAGTCACCTTTGAGACACGATCTCCACCTCAGTGGAAATTTAGTTC CTGCTATAATTAGCCTTCCTCATAAGTTGCATTACTTCAGCGTCCC AACTGCACCCCTTACCACGAAGACAGGTTTGTCCATTTCCCATACTGC GGCGTTGGCAGGGGGTTTCGCATGTCCCACGCGAAACGTTGCTGAAG GCTCAGGTTTCTGAGCGACAAAAGCGCTGGCTAGAATTACCTACC GGCCTCCACCATAACCTTCGATATTCGCGCCCACTCTCCCATTATC CGCACAAGTGATGTGATGCGATTGCCCGCTAAGATAGTTAATCAT TAACTACAAGGAACCCCTAGTGATGGAGTTGGCCACTCCCTCTCTG CGCGCTCGCTCGCTCACTGAGGCCGGGCGACCAAAGTCCGCCGAC GCCCGGGCTTTGCCCGGGCGGCCTCAGTGAGCGAGCGAGCGCGCA TATCGAATCGAATGCAGTGATCTCCAGGTGCGAATCAAAAATACT AGGTAAGTAGAGGATTTGCGACGTTCTAAGCGTTGGTCCATGTGAA TCGCCATCCAGGATCCCAATGGCGCGCCGAGCTTGGCTCGAGCATG GTCATAGCTGTTTCTGTGTGAAATTGTTATCCGCTCACAATTCCA CACAACATACGAGCCGGAAGCATAAAGTGTAAGCCCTGGGGTGCCCT AATGAGTGAGCTAACTCACATTAATTGCGTTGCGCTCACTGCCCGC TTTCCAGTCGGGAAACCTGTCGTGCCAGCTGCATTAATGAATCGGC CAACGCGCGGGGAGAGGCGGTTTTCGTATTGGGCGCTCTTCCGCTT CCTCGCTCACTGACTCGCTGCGCTCGGTCGTTCGGCTGCGGCGAGC GGTATCAGCTCACTCAAAGGCGGTAATACGGTTATCCACAGAATCA GGGGATAACGCAGGAAAGAACATGTGAGCAAAAGGCCAGCAAAAGG CCAGGAACCGTAAAAAGGCCGCGTTGCTGGCGTTTTTCCATAGGCT CCGCCCCCTGACGAGCATCACAAAATCGACGCTCAAGTCAGAGG TGGCGAAAACCCGACAGGACTATAAAGATACCAGGCGTTTCCCCCTG GAAGCTCCCTCGTGCGCTCTCCTGTTCCGACCTGCGCGTTACCGG ATACCTGTCCGCCTTTCTCCCTTCGGGAAGCGTGGCGCTTTCTCAT AGCTCAGCTGTAGGTATCTCAGTTCGGTGTAGGTCGTTCGCTCCA AGCTGGGCTGTGTGCACGAACCCCGTTTCAGCCGACCGCTGCGC CTTATCCGGTAAGTATCGTCTTGAGTCCAACCCGGTAAGACACGAC TTATCGCCACTGGCAGCAGCCACTGGTAACAGGATTAGCAGAGCGA GGTATGTAGGCGGTGCTACAGAGTTCTTGAAGTGGTGGCCTAACTA CGGCTACACTAGAAGAACAGTATTTGGTATCTGCGCTCTGCTGAAG CCAGTTACCTTCGGAAAAAGAGTTGGTAGCTCTTGATCCGGCAAAC AAACCACCGCTGGTAGCGGTGGTTTTTTTTGTTTGCAAGCAGCAGAT TACGCGCAGAAAAAAGGATCTCAAGAAGATCCTTTGATCTTTTCT ACGGGGTCTGACGCTCAGTGGAACGAAACTCACGTTAAGGGATTT TGGTCATGAGATTATCAAAAAGGATCTTCACCTAGATCCTTTTAAA TTAAAAATGAAGTTTAAATCAATCTAAAGTATATATGAGTAACT TGGTCTGACAGTTAGAAAAACTCATCGAGCATCAATGAAACTGCA ATTTATTATCATATCAGGATTATCAATACCATATTTTTGAAAAAGCCG TTTCTGTAATGAAGGAGAAAACTCACCGAGGCAGTTCCATAGGATG GCAAGATCCTGGTATCGGTCTGCGATTCCGACTCGTCCAACATCAA TACAACCTATTAATTTCCCCTCGTCAAAAATAAGGTTATCAAGTGA GAAATCACCATGAGTGACGACTGAATCCGGTGAGAATGGCAAAAGT TTATGCATTTCTTTCCAGACTTGTTCAACAGGCCAGCCATTACGCT CGTCATCAAAATCACTCGCATCAACCAACCGTTATTCAATTCGTGA TTGCGCCTGAGCGAGACGAAATACGCGATCGCTGTTAAAGGACAA TTACAAACAGGAATCGAATGCAACCGGCGCAGGAACACTGCCAGCG CATCAACAATATTTTACCTGAATCAGGATATTCTTCTAATACCTG
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		GAATGCTGTTTTCCAGGGATCGCAGTGGTGAGTAACCATGCATCA TCAGGAGTACGGATAAAATGCTTGATGGTCGGAAGAGGCATAAAAT CCGTCAGCCAGTTTAGTCTGACCATCTCATCTGTAACATCATTGGC AACGCTACCTTTGCCATGTTTCAGAAACAACCTCTGGCGCATCGGGC TTCCCATACAATCGATAGATTGTCGCACCTGATTGCCCCGACATTAT CGCGAGCCCATTTATACCCATATAAAATCAGCATCCATGTTGGAATT TAATCGCGGCCTAGAGCAAGACGTTTTCCCGTTGAATATGGCTCATA CTCTTCCTTTTTCAATATTATTGAAGCATTTATCAGGGTTATTGTC TCATGAGCGGATACATATTTGAATGTATTTAGAAAAATAAACAAAT AGGGGTTCGCGGCACATTTCCCCGAAAAGTGCCACCTGACGTCTAA GAAACCATTATTATCATGACATTAACCTATAAAAAATAGGCGTATCA CGAGGCCCTTTCGTC
24	pAAV-VL-VH-Stuffer- 2 Plasmid Sequence (7191 bp) Exemplary anti-IGF1R antibody heavy chain shown with bold emphasis; exemplary anti-IGF1R antibody light chain shown underlined; exemplary linker sequence shown lower-case and italicized.	TCGCGCGTTTTCGGTGATGACGGTGAAAACCTCTGACACATGCAGCT CCCGGAGACGGTCACAGCTTGTCTGTAAGCGGATGCCGGGAGCAGA CAAGCCCGTCAGGGCGCGTCAGCGGGTGTGGCGGGTGTGGGGCT GGCTTAACATATGCGGCATCAGAGCAGATTGTACTGAGAGTGCACCA TATGCGGTGTGAAATACCGCACAGATGCGTAAGGAGAAAAATACCGC ATCAGGCGCCATTGCGCCATTGAGGCTGCGCAACTGTTGGGAAGGGC GATCGGTGCGGGCCTCTTCGCTATTACGCCAGCTGGCGAAAGGGG ATGTGCTGCAAGGCGATTAAGTTGGGTAACGCCAGGGTTTTCCAG TCACGACGTTGTAAAACGACGGCCAGTGAATTGACGCGTATTGGGA TTGACCTAACGAAAAGTGAACCTTCATAATACGTGCTGTCCCACGCA CATGGTAGATTTGGACAAAATTGAATGGAGTCTGATCAACCTTCAC ACCGATCTAGATATCGCGCGCTCGCTCGCTCACTGAGGCCCGCCCG GCAAAGCCCGGGCGTCGGGCGACCTTTGGTCGCCCCGGCTCAGTGA GCGAGCGAGCGCGCAGAGAGGGAGTGGCCAACCTCCATCACTAGGGG TTCCTTGTAGTTAATGATTAACGGATCTTTACAAATTCAAGCCAGG TGATTTCAACAAATTTTGCTGACGATTTAGGCGCACTATCCCTTAA ACTACAAATTAGAAAATAGCGTTTCCTTGACACTAGTCTAGTTATTA ATAGTAATCAATTACGGGGTCATTAGTTTCATAGCCCATATATGGAG TTCCGCGTTACATAACTTACGGTAAATGGCCCGCTGGCTGACCGC CCAACGACCCCCGCCCATTGACGTCAATAATGACGTATGTTCCCAT AGTAACGCCAATAGGGACTTTCCATTGACGTCAATGGGTGGACTAT TTACGGTAAACTGCCCACTTGGCAGTACATCAAGTGTATCATATGC CAAGTACGCCCCCTATTGACGTCAATGACGGTAAATGAGCCCGCTG GCATTATGCCCAGTACATGACCTTATGGGACTTTCTTACTTGGCAG TACATCTACGTATTAGTCATCGCTATTACCATGGTGATGCGGTTTT GGCAGTACATCAATGGGCGTGGATAGCGGTTTGACTCACGGGGATT TCCAAGTCTCCACCCCATTTGACGTCAATGGGAGTTTGTTTTGGCAC CAAAATCAACGGGACTTTCCAAAATGTCGTAACAACCTCCGCCCCAT TGACGCAAAATGGGCGGTAGGCGGTGACGGTGGGAGGTCTATATAAG CAGAGCTCTCTGGCTAACTAGAGAACCCACTGCTTACTGGCTTATC GAAATAAGCTTTCTCAGGGGAGATCTCGTTTAGTGAACCGTCAGAT CCTCACTCTCTTCCGCATCGCTGTCTGCGAGGGCCAGCTGTTGGGC TCGCGGTTGAGGACAAACTCTTCGCGGTCTTTCCAGTACTCTTGGA TCGGAAACCCGTCGGCCCTCCGAACGGTACTCCGCCACCGGGGACC TGAGCGAGTCCGCATCGACCGGATCGGAAAACCTCTCGAGAAAGGC GTCTAACCAAGTCACAGTCGCAAGGTAGGCTGAGCACCGTGGCGGGC GGCAGCGGGTGGCGGTGGGGTTGTTTCTGGCGGAGGTGCTGCTGA TGATGTAATTAAAGTAGGCGGTCTTGAGACGGCGGATGGTCGAGGT GAGGTGTGGCAGGCTTGAGATCCAGCTGTTGGGGTGAGTACTCCCT CTCAAAAGCGGGCATTACTTCTGCGCTAAGATTGTCAAGTTTCCAAA AACGAGGAGGATTTGATATTCACCTGGCCCCGATCTGGCCATACACT TGAGTGACAATGACATCCACTTTGCCTTTCTCTCCACAGGTGTCCA CTCCAGGTCCAAGTTTAAACGCCGCCGCCATGAGCGTTCTTACAC

	AGGTGCTGGGACTGCTGCTGCTGTGGCTGACCGATGCCAGATGCGA GATCGTGCTGACCCAGAGCCCTGCTACACTGAGCCTGAGCCCCGGC GAGCGGGCTACCCTGTCTTGTAGAGCCAGCCAAAGCGTGTCTTCTT ATCTGGCCTGGTACCAGCAAAAGCCTGGACAGGCTCCTAGACTGCT GATCTACGACGCCAGCAAAAGAGCCACCGGCATCCCCGCCAGATTC TCTGGCAGCGGCAGCGGCACCGACTTCACACTGACAATTAGCTCCC TCGAGCCCCGAGGATTTTCGCCGTGTACTACTGCCAGCAGCGCTCCAA ATGGCCACCATGGACCTTTGGCCAGGGCACCAAGGTGGAATCTAAG CGGACCggcggagggcggttctggcggcgaggctccggcgaggag gcCAGGTGGAACTGGTTCGAGAGCGGAGGCGCGTGGTGCAGCCTGG CAGAAGCCAGAGCTGAGCTGCGCCGCTAGCGGCTTACCTTCAGC AGCTACGGCATGCACTGGGTTCAGACAGGCCCTGGCAAGGGCCTGG AATGGGTGGCCATCATCTGGTTCGACGGCAGCTCAACATACACGC CGACAGCGTGCGGGGCCGGTTCACCATCAGCAGAGATAATAGCAAG AACACCCGTGTACCTGCAGATGAACAGCCTTAGAGCCGAGGACACCG CCGTGTACTTCTGCGCCCGGAACTGGGCAGGCGGTACTTTGACCT GTGGGGCAGAGGCACACTGGTGTCCGTGTCCAGCTAGTCCTGTAA TCAACCTCTGGATTACAAAATTTGTGAAAGATTGACTGATATTCTT AACTATGTTGCTCCTTTTACGCTGTGTGGATATGCTGCTTTAATGC CTCTGTATCATGCTATTGCTTCCCGTACGGCTTTCGTTTTCTCCTC CTTGTATAAATCCTGGTTGCTGTCTCTTTATGAGGAGTTGTGGCC GTTGTCCGTCAACGTGGCGTGGTGTGCTCTGTGTTGCTGACGCAA CCCCACTGGCTGGGGCATTGCCACCACCTGTCAACTCCTTTCTGG GACTTTTCGCTTTCCCCCTCCCTATCGCCACGGCAGAACTCATCGCC GCCTGCCTTGCCCGCTGCTGGACAGGGGCTAGGTTGCTGGGCACTG ATAATTCCGTGGTGTGTGCGGGAAGCTGACGTCCGACTGTGCCTT CTAGTTGCCAGCCATCTGTTGTTTGCCCTCCCCCGTGCCTTCCTT GACCTTGAAGGTGCCACTCCCACTGTCTTTCTTAATAAAATGAG GAAATTGCATCGCATTGTCTGAGTAGGTGTCATTCTATTCTGGGGG GTGGGGTGGGGCAGGACAGCAAGGGGGAGGATTGGGAAGACAATAG CAGGCATGCTGGGGATGCGGTGGGCTCTATGGAAGACAACCAGCAT CTCGCGTCTTGCCTAACCCCCCTACATGCTGTTATAGAGAATCAGT GGAAACCCGGTGCCAGGCGATGGAATGTCCTTAACCTGGCAGGAA ATTAAAGGGAACGTATATACAACGCAAGAAGCTGGAAATTTGGCG AGGGAATCCTGTTTCTGTCTATCCAAGAATGGGCATGAGGTGGCAA CCGTCTGTGCTAGCGTACAGGGTGCCTTTGTAACCATTTGGGACAC CGGACACTCGCTGTTTTCGAAATTACCCTTTAAGCGCGGGTATTGA ACCAGGCTTATGCCCAAGATCGTAGCAAGCAGACTCAAACAAGATA TATTTTGCCCGCCTTACAGACGAACTAGTTGGAGGTTATGGAGCA TACTATCACGTGGGCGGCCACTGGTGAGTTACTACACCCCAAGGGC AACGTTGATGCTCCTAAAAAACTCTGGCTGGACGCAAGCCGTAAACA CCCGTGCACTTCATAATCGTTTGCAATTCAGGGCTTGATCTACAC TGGATTGCCATTCTCTCAAAGTATTATGCAGGACGGCGTGCGCGTT CCATGTAAACCTGTCTAACTTACCTGAGACTAGTTGGAAGTGTGG CTAGATCTTTTGTCTACGCATCTAGTCGGTCCACGTTTGGTTTTTAA GATCCAATGATCTTCAAAACGCTGCAAGATTCTCAACCTGCTTTAC TAAGCGCTGGGTCTACTCCAGCGGGATTTTTTATCTAAAGACGAT GAGAGGAGTATTCTGTCAGACCACATAGCTTTCATGTCTGTATCGGA AGGATCGTTGGCGCCCGACCCCTCAGACTCTGTAGTGAGTTCTATGT CCGAGCCATTGCATGCGAGATCGGTAGATTGATAGGGGATACAGAA TATCCCTGGATGCAATAGACGGACAGCTTGGTATCCTAAGCGTAGT CGCGCGTCCGAACCCAGCTCTACTTTAGAGGCCTCGGATTCGGTG CCCGCAGGCCCGCAGAACCGATTAGGGGCATGTACAACAATATTTAT TAGTCACCTTTGAGACACGATCTCCACCTCACTGGAATTTAGTTC CTGCTATAATTAGCCTTCCTCATAAGTTGCATTACTTCAGCGTCCC
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		AACTGCACCCTTACCACGAAGACAGGTTTGTCCATTCCCATACTGC GGCGTTGGCAGGGGGTTTCGCATGTCCCACGCGAAACGTTGCTGAAG GCTCAGGTTTCTGAGCGACAAAAAGCGCTGGCTAGAATTACCTACC GGCTCCACCATAACCTTCGATATTTCGCGCCCACTCTCCCATTAATC CGCACAAAGTGGATGTGATGCGATTGCCCCGCTAAGATAGTTAATCAT TAACTACAAGGAACCCCTAGTGATGGAGTTGGCCACTCCCTCTCTG CGCGCTCGCTCGCTCACTGAGGCCGGGCGACCAAAGGTCGCCCCGAC GCCCCGGGCTTTGCCCCGGGCGGCCTCAGTGAGCGAGCGAGCGCGCA TATCGAATCGAATGCAGTGATCTCCCAGGTGCGAATCAAAAATACT AGGTAAGTACAGGATTTCGCGACGTTCTAAGCGTTGGTCCATGTGAA TCGCCATCCAGGATCCCAATGGCGCGCCGAGCTTGGCTCGAGCATG GTCATAGCTGTTTCTGTGTGAAATTGTTATCCGCTCACAAATTTCCA CACAAACATACGAGCCGGAAGCATAAAGTGTAAGCCCTGGGGTGCCCT AATGAGTGAGCTAACTCACATTAATTGCGTTGCGCTCACTGCCCCGC TTTCCAGTCGGGAAACCTGTCGTGCCAGCTGCATTAATGAATCGGC CAACGCGCGGGGAGAGGCGGTTTTCGTATTGGGCGCTCTCCGCTT CCTCGCTCACTGACTCGCTGCGCTCGGTCTCGGCTGCGGCGAGC GGTATCAGCTCACTCAAAGGCGGTAATACGGTTATCCACAGAAATCA GGGGATAACGCAGGAAAGAACATGTGAGCAAAAGGCCAGCAAAAGG CCAGGAACCGTAAAAAGGCCGCGTTGCTGGCGTTTTTCCATAGGCT CCGCCCCCTGACGAGCATCACAAAAATCGACGCTCAAGTCAGAGG TGGCGAAACCCGACAGGACTATAAAGATACCAAGCGTTTTCCCCTG GAAGCTCCCTCGTGCGCTCTCCTGTTCCGACCCCTGCCGCTTACCGG ATACCTGTCCGCCTTTCTCCCTTCGGGAAGCGTGCGCTTTCTCAT AGCTCACGCTGTAGGTATCTCAGTTTCGGTGTAGGTGTTTCGCTCCA AGCTGGGCTGTGTGCACGAACCCCCCGTTACGCCCCGACCGTGCGC CTTATCCGGTAACATATCGTCTTGAGTCCAACCCGGTAAGACACGAC TTATCGCCACTGGCAGCAGCCACTGGTAACAGGATTAGCAGAGCGA GGTATGTAGGCGGTGCTACAGAGTTCTTGAAGTGGTGGCTAACTA CGGCTACACTAGAAGAACAGTATTTGGTATCTGCGCTCTGCTGAAG CCAGTTACCTTCGGAAGAGAGTTGGTAGCTCTTGATCCGGCAAAAC AAACCACCGCTGGTAGCGGTGGTTTTTTTGTGTTGCAAGCAGCAGAT TACGCGCAGAAAAAAGGATCTCAAGAAGATCCTTTGATCTTTCT ACGGGTCTGACGCTCAGTGGAACGAAAACACGTTAAGGGATT TGGTCATGAGATTATCAAAAAGGATCTTACCTAGATCTTTTAAA TTAAAAATGAAGTTTTAAATCAATCTAAAGTATATATGAGTAACT TGGTCTGACAGTTAGAAAAACTCATCGAGCATCAATGAACTGCA ATTTATTATATCAGGATTATCAATACCATATTTTTTGAAAAAGCCG TTTCTGTAATGAAGGAGAAAACTCACCAGGCGAGTTCCATAGGATG GCAAGATCCTGGTATCGGTCTGCGATTCCGACTCGTCCAACATCAA TACAACCTATTAATTTCCCCTCGTCAAAAATAAGGTTATCAAGTGA GAAATCACCATGAGTGACGACTGAATCCGGTGAGAATGGCAAAAGT TTATGCATTTCTTTCCAGACTTGTTCAACAGGCCAGCCATTACGCT CGTCATCAAAATCACTCGCATCAACCAAACCGTTATTATTCTGTGA TTGCGCTTGAGCGAGACGAAATACGCGATCGTGTAAAAAGGACAA TTACAAACAGGAATCGAATGCAACCGGCGCAGGAACACTGCCAGCG CATCAACAATATTTTACCTGAATCAGGATATTCTTCTAATACCTG GAATGCTGTTTTCCAGGGATCGCAGTGGTGAGTAACCATGCATCA TCAGGAGTACGGATAAAATGCTTGATGGTCGGAAGAGGCATAAATT CCGTCAGCCAGTTTAGTCTGACCATCTCATCTGTAACATCATGGC AACGCTACCTTTGCGATGTTTCAGAAACAACCTTGCGCGCATCGGGC TTCCCATACAATCGATAGATTGTGCGACCTGATTGCCCCACATTAT CGCGAGCCCATTTATACCCATATAAATCAGCATCCATGTTGGAATT TAATCGCGGCTAGAGCAAGACGTTTCCCGTTGAATATGGCTCATA CTCTTCCTTTTTCAATATTATTGAAGCATTTATCAGGGTTATTGTC
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		TCATGAGCGGATACATATTTGAATGTATTTAGAAAAATAAACAAAT AGGGGTTCCGCGCACATTTCCCCGAAAAGTGCCACCTGACGTCTAA GAAACCATTATTATCATGACATTAACCTATAAAAAATAGGCGTATCA CGAGGCCCTTTCGTC
25	CMV Promoter	GTGATGCGGTTTTTGGCAGTACATCAATGGGCGTGGATAGCGGTTTG ACTCACGGGGATTTCCAAGTCTCCACCCCATTTGACGTCAATGGGAG TTTGTTTTGGCACCAAAATCAACGGGACTTTCCAAAATGTCGTAAC AACTCCGCCCCATTGACGCAAATGGGCGGTAGGCGTGTACGGTGGG AGGTCTATATAAGCAGAGCTCTCTGGCTAACTAGAGAACCCACTGC TTACTGGCTTATCGAAAT
26	CMV enhancer	CTAGTTATTAATAGTAATCAATTACGGGGTCATTAGTTCATAGCCC ATATATGGAGTTCCGCGTTACATAACTTACGGTAAATGGCCCGCCT GGCTGACCGCCCAACGACCCCGCCCATTTGACGTCAATAATGACGT ATGTTCCCATAGTAACGCCAATAGGGACTTTCCATTGACGTCAATG GGTGGACTATTTACGGTAAACTGCCCACTTGGCAGTACATCAAGTG TATCATATGCCAAGTACGCCCCCTATTGACGTCAATGACGGTAAAT GGCCCGCTGGCATTATGCCCAGTACATGACCTTATGGGACTTTCC TACTTGGCAGTACATCTACGTATTAGTCATCGCTATTACCATG
27	CMV enhancer and promoter	CTAGTTATTAATAGTAATCAATTACGGGGTCATTAGTTCATAGCCC ATATATGGAGTTCCGCGTTACATAACTTACGGTAAATGGCCCGCCT GGCTGACCGCCCAACGACCCCGCCCATTTGACGTCAATAATGACGT ATGTTCCCATAGTAACGCCAATAGGGACTTTCCATTGACGTCAATG GGTGGACTATTTACGGTAAACTGCCCACTTGGCAGTACATCAAGTG TATCATATGCCAAGTACGCCCCCTATTGACGTCAATGACGGTAAAT GGCCCGCTGGCATTATGCCCAGTACATGACCTTATGGGACTTTCC TACTTGGCAGTACATCTACGTATTAGTCATCGCTATTACCATGGTG ATGCGGTTTTTGGCAGTACATCAATGGGCGTGGATAGCGGTTTTGACT CACGGGGATTTCCAAGTCTCCACCCCATTTGACGTCAATGGGAGTTT GTTTTGGCACCAAAATCAACGGGACTTTCCAAAATGTCGTAACAAC TCCGCCCCCATTTGACGCAAATGGGCGGTAGGCGTGTACGGTGGGAGG TCTATATAAGCAGAGCTCTCTGGCTAACTAGAGAACCCACTGCTTA CTGGCTTATCGAAAT
28	BGH polyA Sequence	CGACTGTGCCTTCTAGTTGCCAGCCATCTGTTGTTTTGCCCCTCCCC CGTGCTTCTCTTGACCCTGGAAGGTGCCACTCCCCTGTCCTTTCC TAATAAAATGAGGAAATTGCATCGCATTGTCTGAGTAGGTGTCATT CTATTCTGGGGGGTGGGGTGGGGCAGGACAGCAAGGGGGAGGATTG GGAAGACAATAGCAGGCATGCTGGGGATGCGGTG
29	WPRE sequence	TCCTGTTAATCAACCTCTGGATTACAAAATTTGTGAAAGATTGACT GATATTCTTAATATGTTGCTCCTTTTACGCTGTGTGGATATGCTG CTTTAATGCCTCTGTATCATGCTATTGCTTCCCGTACGGCTTTCTG TTTCTCCTCCTTGATATAAATCCTGGTTGCTGTCTCTTTATGAGGAG TTGTGGCCCGTTGTCCGTCAACGTGGCGTGGTGTGCTCTGTGTTTG CTGACGCAACCCCCACTGGCTGGGGCATTGCCACCACCTGTCAACT CCTTTCTGGGACTTTTCGCTTTCCCCCTCCCTATCGCCACGGCAGAA CTCATCGCCGCTGCCTTGCCCGCTGCTGGACAGGGGCTAGGTTGC TGGGCACTGATAATTCCGTGGTGTGTGCGGGGAAGCTGACGTC
30	AAV2 5' ITR	GCGCGCTCGCTCGCTCACTGAGGCCGCGCGGGCAAAGCCGGGCGT CGGGCGACCTTTGGTCGCCCAGCCTCAGTGAGCGAGCGAGCGCGCA GAGAGGGAGTGGCCAATCCATCACTAGGGGTTCTTGTAGTTAAT GATTAAC
31	AAV2 3' ITR	GTTAATCATTAATACTACAAGGAACCCCTAGTGATGGAGTTGGCCACT CCCTCTCTGCGCGCTCGCTCGCTCACTGAGGCCGGGCGACCAAAGG TCGCCCCGACGCCCGGGCTTTGCCCGGGCGGCCCTCAGTGAGCGAGCG AGCGCGC

32	AAV5 5' ITR	CTCTCCCCCTGTGCGTTTCGCTCGCTCGCTGGCTCGTTTGGGGGG GTGGCAGCTCAAAGAGCTGCCAGA
33	AAV5 3' ITR	CGACGGCCCTCTGGCCGTGCCCCCCCCAAACGAGCCAGCGAGCGAG CGAACGCGACAGGGGGGAGAGTGCCACACTCTCAAGCAAGGGGGTT TTGTAAGCAGTGATGTCATAATGATG
34	Exemplary Anti-IGF1R Antibody Heavy Chain, Putative CDR1	AASGFTFSSYGMH
35	Exemplary Anti-IGF1R Antibody Heavy Chain, Putative CDR1	GFTFSSYG
36	Exemplary Anti-IGF1R Antibody Heavy Chain, Putative CDR2	IIWFDGSSTY
37	Exemplary Anti-IGF1R Antibody Heavy Chain, Putative CDR2	IWFDGSST
38	Exemplary Anti-IGF1R Antibody Light Chain, Putative CDR1	QSVSSY
39	Exemplary Anti-IGF1R Antibody Light Chain, Putative CDR2	YDASKRAT
40	Exemplary Anti-IGF1R Antibody Light Chain, Putative CDR2	DAS
41	Exemplary Linker Sequence cDNA	GGCGGAGGCGGTTCTGGCGGCGGAGGCTCCGGCGGAGGAGGC
42	Exemplary Linker Sequence	GGGGSGGGSGGGG
43	VL-VH construct produced in Example 2 (Linker sequence shown lower-case and italicized.)	EIVLTQSPATLSLSPGERATLSCRASQSVSSYLAWYQQKPGQAPRL LIYDASKRATGIPARFSGSGGTDFTLTISSLEPEDFAVYYCQQRS KWPPWTFGQGTKVESKRT <i>ggggsgggsgggg</i> QVELVESGGGVVQP GRSQRLSCAASGFTFSSYGMHWVRQAPGKGLEWVAIIWFDGSSTYY ADSVRGRFTISRDN SKNTLYLQMNSLRAEDTAVYFCARELGRRYFD LWGRGTLVSVSS
44	VH-VL construct produced in Example 2 (Linker sequence shown lower-case and italicized.)	QVELVESGGGVVQPGRSQRLSCAASGFTFSSYGMHWVRQAPGKGLE WVAIIWFDGSSTYYADSVRGRFTISRDN SKNTLYLQMNSLRAEDTA VYFCARELGRRYFDLWGRGTLVSVSS <i>ggggsgggsgggg</i> EIVLTQ SPATLSLSPGERATLSCRASQSVSSYLAWYQQKPGQAPRLLIYDAS KRATGIPARFSGSGGTDFTLTISSLEPEDFAVYYCQQRKWPWTF FGQGTKVESKRT

CLAIMS

We claim:

1. An antibody or an antigen-binding fragment that specifically binds to a human Insulin-like Growth Factor 1 Receptor (“IGF1R”) protein, comprising:

a) a heavy chain variable domain, comprising

a CDR1 region having a polypeptide sequence of SEQ ID NO: 3, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 3,

a CDR2 region having a polypeptide sequence of SEQ ID NO: 4, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 4, and/or

a CDR3 region having a polypeptide sequence of SEQ ID NO: 5, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 5; and/or

b) a light chain variable domain, comprising

a CDR1 region having a polypeptide sequence of SEQ ID NO: 7, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 7,

a CDR2 region having a polypeptide sequence of SEQ ID NO: 8, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 8, and/or

a CDR3 region having a polypeptide sequence of SEQ ID NO: 9 or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 9.

2. The antibody or antigen-binding fragment according to claim 1, wherein

the heavy chain variable domain comprises a CDR1 region having a polypeptide sequence of SEQ ID NO: 3,

the heavy chain variable domain comprises a CDR2 region having a polypeptide sequence of SEQ ID NO: 4, and/or

the heavy chain variable domain comprises a CDR3 region having a polypeptide sequence of SEQ ID NO: 5; and/or

the light chain variable domain comprises a CDR1 region having a polypeptide sequence of SEQ ID NO: 7,

the light chain variable domain comprises a CDR2 region having a polypeptide sequence of SEQ ID NO: 8, and/or

the light chain variable domain comprises a CDR3 region having a polypeptide sequence of SEQ ID NO: 9.

3. The antibody or antigen-binding fragment according to claim 1, wherein

the heavy chain variable domain comprises a CDR1 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 3,

the heavy chain variable domain comprises a CDR2 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 4, and/or

the heavy chain variable domain comprises a CDR3 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 5; and/or

the light chain variable domain comprises a CDR1 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 7,

the light chain variable domain comprises a CDR2 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 8, and/or

the light chain variable domain comprises a CDR3 region having a polypeptide sequence having 1, 2, 3, 4 or 5 insertions, substitutions, or deletions as compared to SEQ ID NO: 9.

4. The antibody or antigen-binding fragment according to claim 1, wherein

the heavy chain variable domain comprises a polypeptide sequence of SEQ ID NO: 2; and/or

the light chain variable domain comprises a polypeptide sequence of SEQ ID NO: 6.

5. The antibody or antigen-binding fragment according to any one of claims 1-4, wherein the antibody or antigen-binding fragment specifically binds to:

a) an IGF1R protein having a polypeptide sequence of SEQ ID NO: 1, or a fragment comprising at least 20 contiguous amino acids thereof;

b) an alpha chain of an IGF1R protein having a polypeptide sequence comprising the sequence spanning positions 31-736 of SEQ ID NO: 1, or a fragment comprising at least 20 contiguous amino acids thereof; and/or

c) a beta chain of an IGF1R protein having a polypeptide sequence comprising the sequence spanning positions 741-1367 of SEQ ID NO: 1, or a fragment comprising at least 20 contiguous amino acids thereof.

6. The antibody or antigen-binding fragment according to any one of claims 1-5, wherein the antibody or antigen-binding fragment is a rabbit antibody or antigen-binding fragment or a mouse antibody or antigen-binding fragment.

7. The antibody or antigen-binding fragment according to any one of claims 1-6, wherein the antibody or antigen-binding fragment is a monoclonal antibody or antigen-binding fragment.

8. The antibody or antigen-binding fragment according to any one of claims 1-6, wherein the antibody or antigen-binding fragment comprises a monoclonal antibody or antigen-binding fragment, a grafted antibody or antigen-binding fragment, a chimeric antibody or antigen-binding fragment, a human antibody or antigen-binding fragment, or a humanized antibody or antigen-binding fragment.

9. The antibody or antigen-binding fragment according to any one of claims 1-6, being an antigen-binding fragment comprising a Fab, a Fab', a F(ab')₂, a variable fragment (Fv), a triabody, a tetrabody, a minibody, a bispecific F(ab')₂, a trispecific F(ab')₂, a diabody, a bispecific diabody, a single chain variable fragment (scFv), a scFv-Fc, a Fab-Fc, a VHH, or a bispecific scFv.

9. The antibody or antigen-binding fragment according to any one of claims 1-8, being an antigen-binding fragment comprising an scFv.

10. The antibody or antigen-binding fragment according to any one of claims 1-9, being an antigen-binding fragment comprising an scFv, the scFv comprising a light chain variable domain N-terminal to a heavy chain variable domain, optionally wherein the light chain variable domain and the heavy chain variable domain are connected by a linker sequence.

11. The antibody or antigen-binding fragment according to any one of claims 1-9, being an antigen-binding fragment comprising an scFv, the scFv comprising a heavy chain variable

domain N-terminal to a light chain variable domain, optionally wherein the heavy chain variable domain and the light chain variable domain are connected by a linker sequence.

12. The antibody or antigen-binding fragment according to any one of claims 9-11, wherein the light chain variable domain comprises a polypeptide sequence of SEQ ID NO: 6, a polypeptide sequence having 1, 2, 3, 4, or 5 amino acid substitutions as compared to SEQ ID NO: 6, or a polypeptide sequence having at least 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, or 99% sequence identity compared to SEQ ID NO: 6; and/or

the heavy chain variable domain comprises a polypeptide sequence of SEQ ID NO: 2, a polypeptide sequence having 1, 2, 3, 4, or 5 amino acid substitutions as compared to SEQ ID NO: 2, or a polypeptide sequence having at least 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, or 99% sequence identity compared to SEQ ID NO: 2.

13. The antibody or antigen-binding fragment according to any one of claims 9-11, being an antigen-binding fragment which is an scFv comprising

a polypeptide sequence of SEQ ID NO: 43, a polypeptide sequence having 1, 2, 3, 4, or 5 amino acid substitutions as compared to SEQ ID NO: 43, or a polypeptide sequence having at least 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, or 99% sequence identity compared to SEQ ID NO: 43; and/or

a polypeptide sequence of SEQ ID NO: 44, a polypeptide sequence having 1, 2, 3, 4, or 5 amino acid substitutions as compared to SEQ ID NO: 44, or a polypeptide sequence having at least 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, or 99% sequence identity compared to SEQ ID NO: 44.

14. The antibody or antigen-binding fragment according to any one of claims 9-13, wherein the light chain variable domain and the heavy chain variable domain are covalently linked by a linker sequence, wherein the linker sequence comprises a polypeptide sequence of SEQ ID NO: 42, or a sequence having 1, 2, 3, 4, or 5 amino acid substitutions compared to SEQ ID NO: 42.

15. The antibody or antigen-binding fragment according to any one of claims 1-14, operably linked to a signal peptide.

16. The antibody or antigen-binding fragment according to any one of claims 1-15, operably linked to a signal peptide configured to result in secretion of the antibody or antigen-binding fragment from a human cell.
17. A nucleic acid molecule encoding the antibody or antigen-binding fragment according to any one of claims 1-16.
18. The nucleic acid molecule of claim 17, wherein the nucleic acid molecule further comprises a promoter or enhancer sequence operably linked to the encoded antibody or antigen-binding fragment.
19. The nucleic acid molecule of claims 17 or 18, wherein the nucleic acid molecule further encodes a signal peptide sequence operably linked to the encoded antibody or antigen-binding fragment.
20. A vector comprising the nucleic acid molecule of any one of claims 17-19.
21. The vector of claim 20, wherein the vector comprises an adenoviral vector, an HIV vector, a lentiviral vector, an adeno-associated virus vector ("AAV"), a plasmid vector, or a C³DNA vector.
22. The vector of claim 21, wherein the AAV comprises AAV2, AAV4, AAV8, or AAV9.
23. A liposome comprising the nucleic acid molecule of any one of claims 17-19.
24. A recombinant adeno-associated virus ("rAAV") vector comprising an AAV capsid and an expression cassette, the expression cassette comprising a polynucleotide encoding the antibody or antigen-binding fragment according to any one of claims 1-16, operatively linked to a promoter.
25. The rAAV vector of claim 24, wherein the antibody or antigen-binding fragment is an scFv.
26. The rAAV vector of claim 24, wherein the scFv comprises:

a heavy chain variable domain comprising a polypeptide sequence of SEQ ID NO: 2;
and/or

a light chain variable domain comprising a polypeptide sequence of SEQ ID NO: 6.

27. The rAAV vector of claim 24, wherein the scFv comprises:

a) a heavy chain variable domain, comprising

a CDR1 region having a polypeptide sequence of SEQ ID NO: 3, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 3,

a CDR2 region having a polypeptide sequence of SEQ ID NO: 4, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 4, and/or

a CDR3 region having a polypeptide sequence of SEQ ID NO: 5, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 5; and/or

b) a light chain variable domain, comprising

a CDR1 region having a polypeptide sequence of SEQ ID NO: 7, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 7,

a CDR2 region having a polypeptide sequence of SEQ ID NO: 8, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 8, and/or

a CDR3 region having a polypeptide sequence of SEQ ID NO: 9 or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 9.

28. The rAAV vector of any one of claims 24-27, wherein the expression cassette is flanked by two inverted terminal repeats ("ITRs").

29. The rAAV vector of claim 28, wherein the ITRs are AAV2 ITRs.

30. The rAAV vector or the composition of any one of claims 24-29, wherein the AAV capsid comprises a viral protein that shares at least 85%, 90%, 95%, 98%, or 100% identity with SEQ ID NOS: 13, 15, 17, or 19.

31. The rAAV vector of any one of claims 24-29, wherein the AAV capsid comprises a viral protein that shares at least 85%, 90%, 95%, 98%, or 100% identity with SEQ ID NO: 19.

32. A composition comprising an rAAV vector, wherein the rAAV vector comprises:
- (a) an AAV2, AAV5, AAV8, or AAV9 capsid, and
 - (b) an expression cassette, wherein the expression cassette comprises a polynucleotide comprising a nucleotide sequence sharing at least 90 or 95% identity to a nucleotide sequence comprising SEQ ID NOs: 21 or 22, and wherein the polynucleotide is linked to a promoter.
33. A composition comprising an rAAV vector, wherein the rAAV vector comprises:
- (a) an AAV2, AAV5, AAV8, or AAV9 capsid, and
 - (b) an expression cassette, wherein the expression cassette comprises a polynucleotide sequence sharing at least 90 or 95% identity with SEQ ID NOs: 21 or 22.
34. The composition of claims 32 or 33, wherein the AAV capsid is AAV2.
35. The composition of claims 32 or 33, wherein the AAV capsid is AAV5.
36. The composition of claims 32 or 33, wherein the AAV capsid is AAV8.
37. The composition of claims 32 or 33, wherein the AAV capsid is AAV9.
38. A composition comprising an rAAV vector, wherein the rAAV vector comprises:
- (a) an AAV capsid, and
 - (b) an expression cassette, wherein the expression cassette comprises a polynucleotide encoding a polypeptide sequence sharing at least 90 or 95% sequence identity with SEQ ID NOs: 2 or 6, and wherein the polynucleotide is linked to a promoter.
39. A pharmaceutical composition comprising the antibody or antigen-binding fragment according to any one of claims 1-16, the nucleic acid molecule of any one of claims 17-19, the rAAV vector of any one of claims 24-31, or the composition of any one of claims 32-38, and a pharmaceutically acceptable carrier.
40. The pharmaceutical composition of claim 39, wherein the composition comprises about 1×10^7 to about 1×10^{14} genome copies per milliliter of the rAAV vector.

41. The pharmaceutical composition of claim 39, wherein the composition comprises about 1×10^{12} to about 6.5×10^{12} genome copies per milliliter of the rAAV vector.

42. A kit comprising the antibody or antigen-binding fragment of any one of claims 1-16, the rAAV of any one of claims 24-31, the composition of any one of claims 32-38, or the pharmaceutical composition of any one of claims 39-41, and a pharmaceutically acceptable carrier, and instructions for use in treating an ocular disease, disorder, or condition in a subject, comprising administering the antibody, antigen-binding fragment, composition, or pharmaceutical composition to the eye of the subject.

43. The kit of claim 42, wherein the ocular disease, disorder, or condition is Graves' Orbitopathy or Thyroid Eye Disease ("TED").

44. A method of treating an ocular disease, disorder, or condition in a subject in need thereof, the method comprising administering an effective amount of the antibody or antigen-binding fragment of any one of claims 1-16, the rAAV of any one of claims 24-31, the composition of any one of claims 32-38, or the pharmaceutical composition of any one of claims 39-41, to a tear film or an eye tissue of the subject,

optionally, wherein (i) the eye tissue comprises tissue of a main or at least one accessory lacrimal gland, a meibomian gland, or a trabecular meshwork, of the subject; and (ii) the administration results in expression of the antibody or antigen-binding fragment of any one of claims 1-16 by one or more cells of the subject, or delivery or diffusion of the antibody or antigen-binding fragment of any one of claims 1-16 into at least one extraorbital fat pad of an eye of the subject.

45. The method of claim 44, wherein the administration to the tear film or the eye tissue of the subject comprises administering the antibody, antigen-binding fragment, rAAV, composition, or pharmaceutical composition to the at least one accessory lacrimal gland of the subject.

46. The method of claim 44 or 45, wherein the administration to the tear film or the eye tissue of the subject comprises administering the antibody, antigen-binding fragment, rAAV, composition, or pharmaceutical composition to the meibomian gland.

47. The method of any one of claims 44-46, wherein the administration to the tear film or the eye tissue of the subject comprises administering the antibody, antigen-binding fragment, rAAV, composition, or pharmaceutical composition to the subject's trabecular meshwork.

48. The method of any one of claims 44-47, wherein the ocular disease, disorder, or condition is Thyroid eye disease ("TED").

49. The method of any one of claims 44-48, wherein the method results in:
a) an improvement of at least one symptom of the ocular condition; and/or
b) a delay of the progression of the ocular condition.

50. The method of claim 49, wherein the at least one symptom of the ocular condition comprises:
a) swelling of one or more eyelids; and/or
b) redness and/or pain of the conjunctiva, eyelid, caruncle and/or plica semilunaris of at least one eye of the subject.

51. The method of any one of claims 44-49, wherein the subject is a human subject.

52. A cell transfected or transduced with the vector of any one of claims 20-22.

53. A kit comprising the pharmaceutical composition of any one of claims 39-41 and instructions for use in treating a condition in a human subject, wherein the instructions comprise administering the pharmaceutical composition to a tear film or an eye tissue of the human subject, optionally wherein the eye tissue comprises tissue of a main or accessory lacrimal gland, a meibomian gland, or the subject's trabecular meshwork.

54. The pharmaceutical composition of any one of claims 39-41 for use in the manufacture of a medicament for treating a condition in a human subject in need thereof.

55. A fusion protein, comprising a first variable domain and a second variable domain, connected by a linker sequence, that specifically binds to a human Insulin-like Growth Factor 1 Receptor ("IGF1R") protein, wherein
each of the first and the second variable domains comprise either:

- (i) a CDR1 region having a polypeptide sequence of SEQ ID NO: 3, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 3,
a CDR2 region having a polypeptide sequence of SEQ ID NO: 4, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 4, and/or
a CDR3 region having a polypeptide sequence of SEQ ID NO: 5, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 5; or
 - (ii) a CDR1 region having a polypeptide sequence of SEQ ID NO: 7, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 7,
a CDR2 region having a polypeptide sequence of SEQ ID NO: 8, or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 8, and/or
a CDR3 region having a polypeptide sequence of SEQ ID NO: 9 or having at least 50, 60, 70, 80, 85, 90, 95 or 100% sequence identity with SEQ ID NO: 9.
56. The fusion protein of claim 55, wherein the first variable domain is different from the second variable domain.
57. The fusion protein of claim 55, wherein the first variable domain is a heavy chain variable domain comprising (i) and the second variable domain is a light chain variable domain comprising (ii).
58. The fusion protein of any one of claims 55-57, wherein the fusion protein is an scFv.
59. The fusion protein of any one of claims 55-58, wherein the fusion protein further comprises an N-terminal signal peptide sequence configured to cause secretion of the fusion protein from a human cell.
60. The fusion protein of any one of claims 57-59, wherein the first variable domain is N-terminal to the second variable domain.
61. The fusion protein of any one of claims 57-59, wherein the second variable domain is N-terminal to the first variable domain.
62. A nucleic acid molecule encoding the fusion protein of any one of claims 55-61.

63. A vector comprising the nucleic acid molecule of claim 62.
64. The vector of claim 63, wherein the vector comprises an adenoviral vector, an HIV vector, a lentiviral vector, an adeno-associated virus vector ("AAV"), a plasmid vector, or a C³DNA vector.
65. A pharmaceutical composition, comprising the fusion protein of any one of claims 55-61, the nucleic acid molecule of claim 62, or the vector of claims 63 or 64, and at least one pharmaceutically acceptable carrier.
66. A method of treating an ocular disease, disorder, or condition in a subject, comprising administering the fusion protein of any one of claims 55-61, the nucleic acid molecule of claim 62, the vector of claims 63 or 64, or the pharmaceutical composition of claim 65, to the eye of the subject.
67. The method of claim 66, wherein the ocular disease, disorder, or condition is Graves' Orbitopathy or Thyroid Eye Disease ("TED"), and treating comprising administering an effective amount of the fusion protein of any one of claims 55-61, the nucleic acid molecule of claim 62, the vector of claims 63 or 64, or the pharmaceutical composition of claim 65, thereby improving at least one symptom of the ocular condition and/or delaying the progression of the ocular condition.
68. The method of claims 66 or 67, wherein administration is to a tear film or an eye tissue of at least one eye of the subject.
69. The method of any one of claims 66-68, wherein the fusion protein of any one of claims 55-61 is administered, and administration results in the fusion protein being delivered to or diffusing into at least one extraorbital fat pad of the eye of the subject.
70. The method of any one of claims 66-68, wherein the nucleic acid molecule of claim 62 or the vector of claims 63 or 64 is administered, and administration results in expression of the fusion protein of any one of claims 55-61 by one or more cells of the human subject, and delivery or diffusion of the fusion protein into at least one extraorbital fat pad of the eye of the subject.

71. The method of any one of claims 66-68, wherein the pharmaceutical composition of claim 65 is administered, and administration results in (1) expression of the fusion protein of any one of claims 55-61 by one or more cells of the human subject, and/or (2) delivery or diffusion of the fusion protein of any one of claims 55-61 into at least one extraorbital fat pad of the eye of the subject.