

CLAIMS

WHAT IS CLAIMED IS:

1. A small interfering RNA (siRNA)-antibody conjugate comprising an anti-transferrin receptor antibody or antigen binding fragment thereof conjugated to a siRNA molecule comprising a passenger strand and a guide strand, wherein the guide strand is from 19 to 30 nucleotides in length and comprises a sequence selected from a group consisting of SEQ ID NOs: 72, 76, 126, 131, 132, 134, 135, 136, 212, 216, 266, 271, 272, 274, 275, and 276, and wherein the siRNA molecule mediates RNA interference against DUX4 mRNA and downregulation of DUX4 biomarker mRNA levels.
2. The siRNA-antibody conjugate of claim 1, wherein the anti-transferrin receptor antibody or antigen binding fragment thereof binds to a transferrin receptor on a cell surface of a muscle cell.
3. The siRNA-antibody conjugate of claim 1 or 2, wherein the siRNA molecule comprises at least one 2' modified nucleotide, at least one modified internucleotide linkage, or at least one inverted abasic moiety.
4. The siRNA-antibody conjugate of any one of claims 1-3, wherein the passenger strand and the guide strand each independently comprises at least one 2' modified nucleotide, at least one modified internucleotide linkage, or at least one inverted abasic moiety.
5. The siRNA-antibody conjugate of any one of claims 1-4, wherein the siRNA molecule hybridizes to at least 8 contiguous bases of the target sequence of a human DUX4 mRNA.
6. The siRNA-antibody conjugate of any one of claims 1-5, wherein the guide strand is from 19 to 25 nucleotides in length.
7. The siRNA-antibody conjugate of any one of claims 1-6, wherein the siRNA-antibody conjugate comprises a linker connecting the anti-transferrin receptor antibody or antigen binding fragment thereof to the siRNA molecule.
8. The siRNA-antibody conjugate of any one of claims 3-7, wherein the at least one 2' modified nucleotide comprises 2'-O-methyl, 2'-O-methoxyethyl (2'-O-MOE), 2'-O-aminopropyl, 2'-deoxy, 2'-deoxy-2'-fluoro, 2'-O-aminopropyl (2'-O-AP), 2'-O-dimethylaminoethyl (2'-O-DMAOE), 2'-O-dimethylaminopropyl (2'-O-DMAP), 2'-O-dimethylaminoethoxyethyl (2'-O-

DMAEOE), 2'-O-N-methylacetamido (2'-O-NMA) modified nucleotide, locked nucleic acid (LNA), ethylene nucleic acid (ENA), or a combination thereof.

9. The siRNA-antibody conjugate of any one of claims 3-8, wherein the at least one modified internucleotide linkage comprises a phosphorothioate linkage or a phosphorodithioate linkage.

10. The siRNA-antibody conjugate of any one of claims 3-9, wherein the siRNA molecule comprises 3 or more 2' modified nucleotides selected from 2'-O-methyl modified nucleotide and 2'-deoxy-2'-fluoro modified nucleotide.

11. The siRNA-antibody conjugate of any one of claims 1-10, wherein the siRNA molecule conjugate has a siRNA to antibody ratio from 1 to 4.

12. The siRNA-antibody conjugate of any one of claims 1-11, wherein the siRNA molecule comprises a 5'-terminal vinylphosphonate modified nucleotide.

13. The siRNA-antibody conjugate of any one of claims 1-12, wherein the siRNA molecule conjugate is formulated for parenteral administration.

14. The siRNA-antibody conjugate of any one of claims 1-13, wherein the anti-transferrin receptor antibody or antigen binding fragment thereof is conjugated to the passenger strand of the siRNA molecule.

15. The siRNA-antibody conjugate of any one of claims 1-14, wherein the passenger strand of the siRNA molecule comprises a sequence selected from a group consisting of SEQ ID NOs: 2, 6, 56, 61, 62, 64, 65, 66, 142, 146, 196, 201, 202, 204, 205, and 206.

16. The siRNA-antibody conjugate of any one of claims 1-15, wherein the downregulation of DUX4 biomarker mRNA levels is evaluated by expression level of one or more DUX4 biomarker genes selected from a group consisting of MBD3L2, TRIM43, PRAMEF1, ZSCAN4, KHDC1L, and LEUTX.

17. The siRNA-antibody conjugate of any one of claims 1-16, wherein the anti-transferrin receptor antibody or antigen binding fragment thereof is conjugated to the guide strand of the siRNA molecule.

18. Use of the siRNA-antibody conjugate of any one of claims 1-17 in the manufacture of a medicament for treating facioscapulohumeral muscular dystrophy (FSHD) in a subject.

19. The use of claim 18, wherein the RNA interference against the human DUX4 mRNA prevents or prevents progress of muscle atrophy in the subject.

20. The use of claim 18 or 19, the FSHD is caused by genetic and epigenetic changes in D4Z4 DNA region of long arm of chromosome 4 in muscle cells of the subject or associated with muscle atrophy of the subject.
21. The siRNA-antibody conjugate of any one of claims 1-17 for use in therapy.