DUX4 siRNA Optimization for the Development of an Antibody-Oligonucleotide Conjugate (AOC™) for the Treatment of Facioscapulohumeral Muscular Dystrophy (FSHD)

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FSHD is Often Diagnosed in Young Adults with Few Treatment Options

There are no approved therapeutics for FSHD

AFFECTS

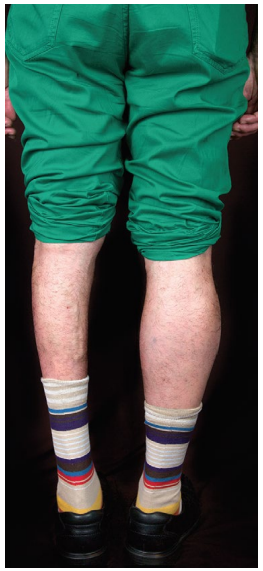
~16,000 - 38,000

PEOPLE IN THE US[1,2]

0

APPROVED THERAPIES[3]

- Facioscapulohumeral Dystrophy (FSHD)
- One of the most common forms of muscular dystrophy[1]
- Autosomal dominant disease caused by abnormal expression of double homeobox 4 (DUX4)[4,5]
- Characterized by progressive, asymmetric skeletal muscle loss with onset often in teenage and adult years[6]
- About 20% of patients will end up using a wheelchair[6]



DeSimone et al. 2017

1. Deenen JCW, et al. *Neurology*. 2014;83(12):1056-1059; 2. US Census Bureau. Quick Facts. https://www.census.gov/quickfacts/fact/table/US/ [Last Accessed February 2022]; 3. Cohen J, et al. *Trends Mol Med.* 2021;27(2):123-137; 4. Lemmers RJLF, et al. *Science.* 2010;329(5999):1650–1653; 5. Snider L, et al. *PLoS Genet*. 2010;6(10):e1001181; 6. Tawil R and Van Der Maarel SM. *Muscle Nerve.* 2006;34(1):1–15; 7. DeSimone AM, et al. *Compr Physiol*. 2017;7(4):1229-1279.

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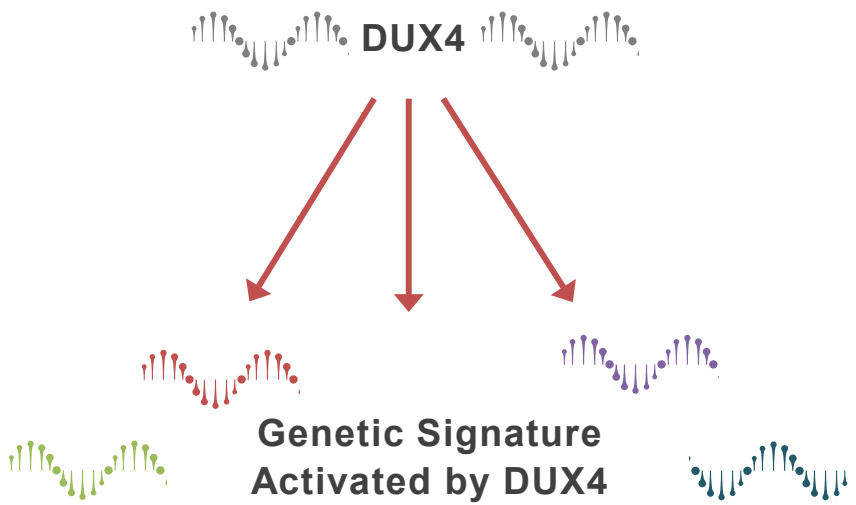
FSHD is Caused by Aberrant Expression of DUX4 in Muscle

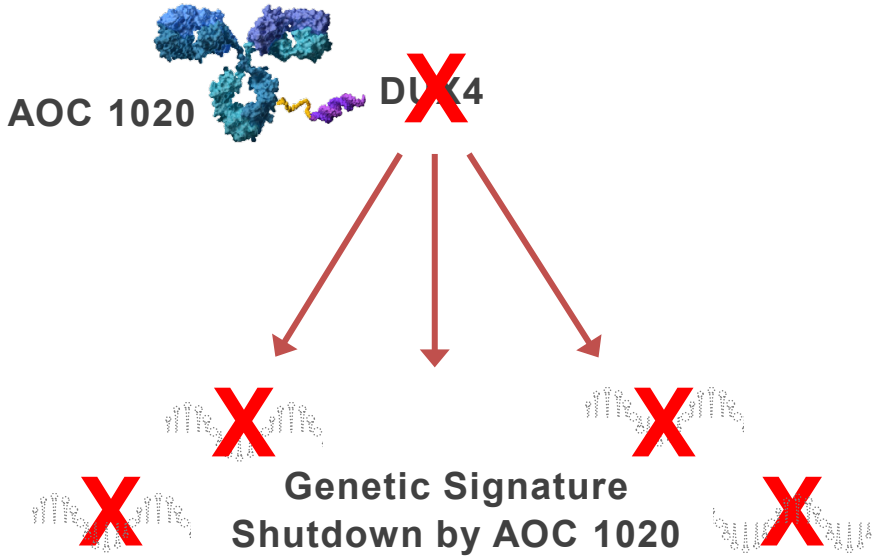
DUX4 activates genes that are toxic to muscle cells

MECHANISM OF DISEASE[1,2]



Apoptosis, immune signaling altered
myogenesis inhibited

THERAPEUTIC APPROACH[3,4]



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1. Lemmers RJLF, et al. *Science.* 2010;329(5999):1650–1653; 2. Snider L, et al. *PLoS Genet.* 2010;6(10):e1001181;
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AOC 1020 FSHD Development Candidate is Designed to be a Potent and Specific Inhibitor of DUX4 Expression

Transferrin mAb

Screen DUX4 siRNAs in FSHD donor cells using gene signature

Off-target analysis by RNA seq

Select potent siRNAs with minimal off-target profile

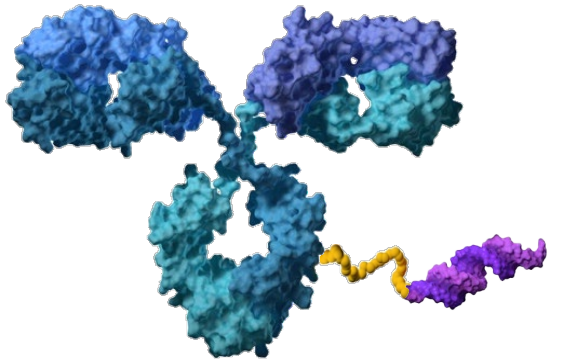
Target candidate profile met

Evaluate in FSHD mouse model & GLP studies

COMPLETED

ONGOING

AOC 1020



- Antibody directed to human transferrin receptor 1 (TfR1)
- Non-cleavable linker designed to facilitate siRNA delivery to muscle
- Stabilized siRNA targeting DUX4 mRNA (siDUX4.6)

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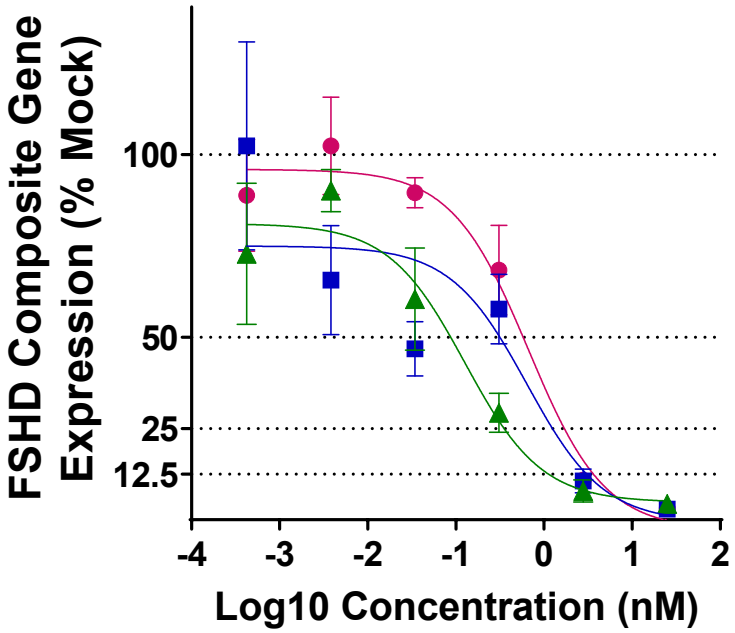
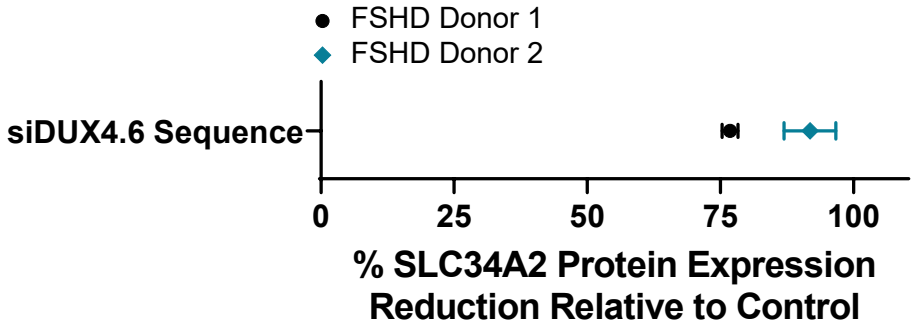
Lead DUX4 siRNA Sequence siDUX4.6 Inhibits DUX4 Regulated Genes in FSHD Patient-Derived Muscle Cells

Sub-Nanomolar Potency of the siDUX4.6 Sequence *In Vitro* in FSHD Primary Patient-Derived Myotubes



	IC50 (nM)	Emax (%)
MB02	0.639	100
MB05	0.127	95
MB06	0.665	100

Activity of the siDUX4.6 Sequence in FSHD Donor Myotubes Monitored by Downregulation of SLC34A2 Protein



- Robust downregulation of DUX4-regulated genes was observed with the lead siDUX4.6 siRNAs in FSHD donor myotubes *in vitro*
- FSHD Composite is a mean expression of DUX4-regulated genes KHDC1L, LEUTX, MBD3L2, ZSCAN4

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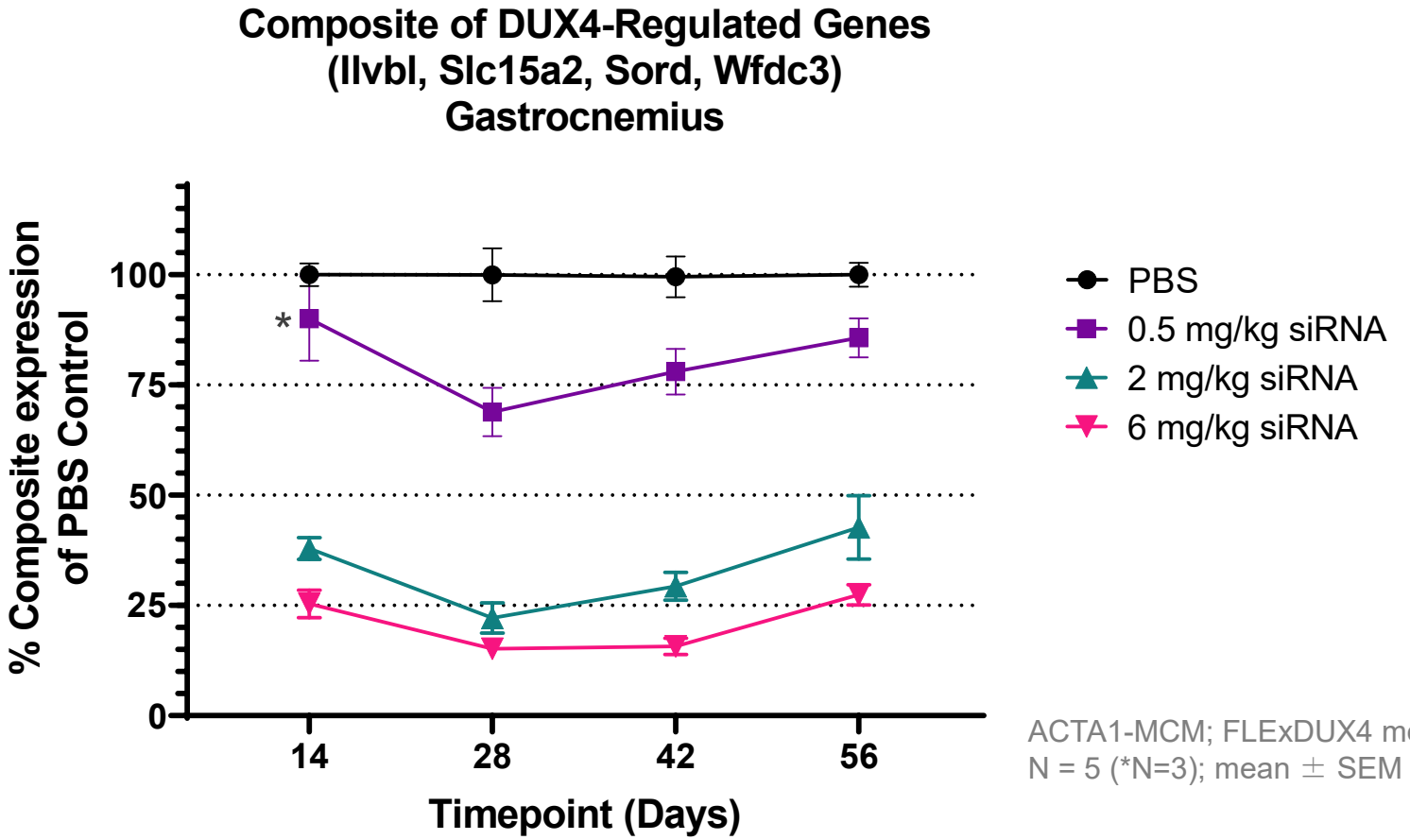
siDUX4.6 Shows Potent Inhibition of DUX4 Regulated Genes in Transgenic DUX4 Mouse Model of FSHD for 8 Weeks

Dose-dependent inhibition of DUX4-regulated genes in skeletal muscles



- The siRNA clinical candidate siDUX4.6 demonstrated activity *in vivo* towards the human DUX4 mRNA
- Approximately a 75% reduction in DUX4 responsive genes was induced after a single systemic IV administration of 6 mg/kg of siRNA within the AOC

ACTA1-MCM; FLExDUX4 mouse model of FSHD
N = 5 (*N=3); mean ± SEM

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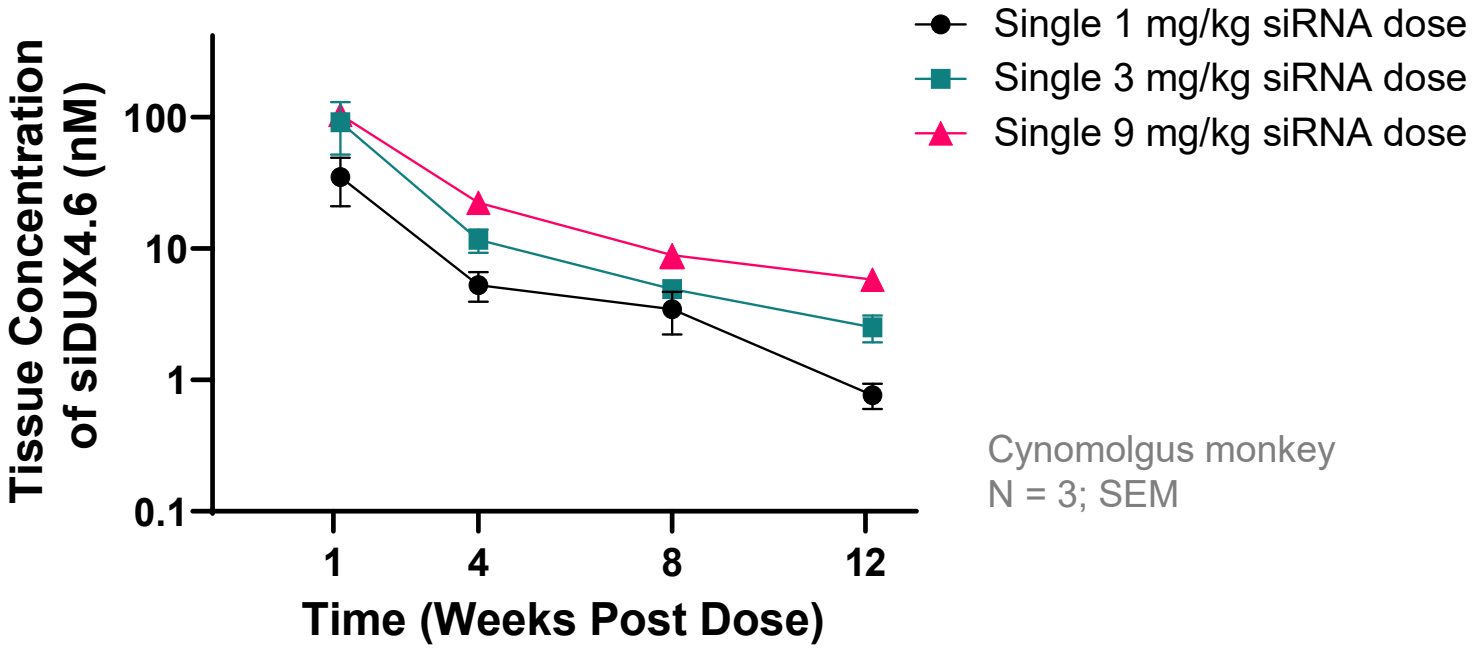
AOC 1020 PK Results in NHP Muscle Tissue Support an Infrequent Dosing Regimen for FSHD Patients



Cynomolgus monkey
N = 3; SEM

- AOC 1020 produced dose-dependent increase in siRNA tissue exposure in skeletal muscle tissues following single systemic IV doses
- The muscle tissue concentration for siDUX4.6 in NHP at therapeutically relevant doses is above IC50 values that we typically observed for other TfR1-based AOCs
- Based on our data, we anticipate this will allow for an infrequent dose schedule in the clinic

AOC 1020 is On-Track to be in the Clinic by the End of 2022

- siDUX4.6 was selected as the clinical candidate siRNA targeting DUX4, having an activity across variety of FSHD patient derived muscle cells, with a sub-nanomolar potency *in vitro*
- siDUX4.6 shows no concerning seed-mediated off-target profile in human muscle cells
- siDUX4.6 demonstrated potent, specific inhibition of DUX4 regulated genes in an FSHD mouse model for 8 weeks after single systemic dose
- AOC 1020 is well-tolerated in cynomolgus monkey preclinical studies at all doses and dose frequencies tested
- AOC 1020 is currently in GLP toxicology studies
- Avidity is planning to enter the clinic with AOC 1020 for treatment of FSHD by end of 2022

Authors and Acknowledgements

Authors

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