Pharmacokinetics and target engagement of intravitreal administration of ANX007, an anti-C1q antibody fragment, in nonhuman primates

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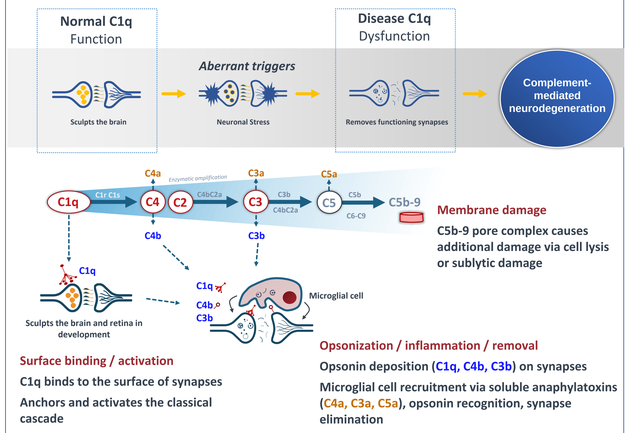
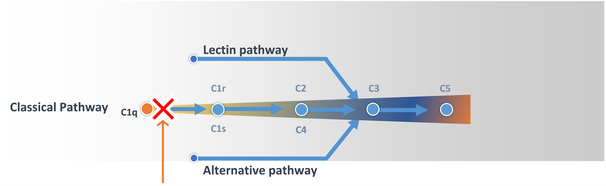
INTRODUCTION

- Geographic atrophy (GA), a neurodegenerative retinal disease, is a form of severe dry macular degeneration (AMD) and one of the leading causes of blindness with no treatment to stop its progression
- C1q and the classical complement cascade, which are key regulators of synaptic pruning in neuronal development, are aberrantly activated in neurodegenerative ophthalmic diseases of the retina, including GA and glaucoma[1,2,3,4,5]



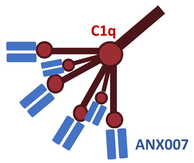
- C1q and its activating substrates are present in all layers of the outer retina, including retinal pigment epithelium and photoreceptor neurons and are positioned to drive chronic, aberrant, classical pathway activation
- Inhibition of C1q blocks initiation of the classical complement cascade
- Blocking C1q prevents activation of all downstream components of the classical pathway that drive a local immune response and tissue destruction in the retina, including C1q, C4, C3, and C5
- By selectively targeting C1q, the alternative and lectin pathways are left intact, which is potentially important for maintaining pathogen surveillance and retinal health



- ***Potential efficacy advantage:*** Shuts down all tissue-damaging components of classical cascade (e.g., C1q, C4, C3, C5)
- ***Potential safety advantage:*** Allows normal immune function of lectin and alternative pathways, which could be important for pathogen surveillance and maintenance of retinal health

ANX007

- ANX007 is a novel humanized IgG1 Fab antibody fragment that specifically recognizes the substrate-binding head groups of C1q and functionally inhibits the classical complement cascade, while leaving the lectin and alternative pathways intact



PURPOSE

- Nonclinical studies were completed to assess ANX007 biodistribution and C1q target engagement in the eye and serum following intravitreal (IVT) administration in cynomolgus monkeys

METHODS

- Nonclinical studies were conducted in compliance with all applicable sections of the Final Rules of the Animal Welfare Act regulations (Code of Federal Regulations, Title 9), the *Public Health Service Policy on Humane Care and Use of Laboratory Animals* from the Office of Laboratory Animal Welfare, and the *Guide for the Care and Use of Laboratory Animals* from the National Research Council
- IVT studies were performed using a single injection of 50 µl per eye (1 or 2.5 mg) or a double injection of 2 x 50 µl per eye (5 mg) administered ~10 minutes apart
- Injections were inserted through the sclera and pars plana approximately 4 mm posterior to the limbus. The needle was directed posterior to the lens into the mid-vitreous, and ANX007 was slowly injected
- Three studies were completed:

Study	Dosing	Collection Timepoints	Samples Collected	Pharmacokinetic (PK) and Pharmacodynamic (PD) Assay Summary
PK and ocular tolerability study	Single dose: 0, 1, or 5 mg/eye	Days 1 (6 hours post-dose), 3, 7, 10, 20, and 30; n=1-2/group/ timepoint	Serum, vitreous humor, retina (non-perfused), choroid (non-perfused)	Free ANX007 and free C1q: Qualified ELISA: serum, vitreous Exploratory ELISA: retina, choroid Total C1q: Exploratory ELISA: retina, choroid
Repeat dose ocular toxicology study with 4-week recovery	2 doses, 4 weeks apart (Day 1, Day 29): 0, 1, or 2.5 mg/eye	Days 44 and 59 (15 and 30 days following the second dose); n=2-6/group/ timepoint	Serum, aqueous humor, vitreous humor, optic nerve (non-perfused)	Free ANX007 and free C1q: Validated ELISA: serum, vitreous Exploratory ELISA: aqueous humor, optic nerve Total C1q: Exploratory ELISA: optic nerve
Repeat dose ocular toxicology study with 4-week recovery	2 doses, 4 weeks apart (Day 1, Day 29): 0 or 5 mg/eye	Days 44 and 59 (15 and 30 days following the second dose); n=4-6/group/ timepoint	Serum, aqueous humor, vitreous humor, retina (perfused), choroid (perfused), optic nerve (perfused)	Free ANX007 and free C1q: Validated ELISA: serum, vitreous Exploratory ELISA: aqueous humor, retina, choroid, optic nerve

RESULTS

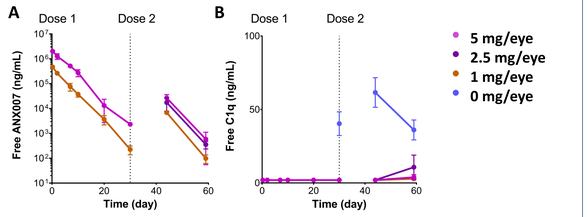
Tolerability

- All doses were well-tolerated
  - No ANX007-related changes were noted in body weights, food consumption, electroretinography, tonometry, and clinical pathology
  - No adverse ANX007-related changes were noted in clinical observations, ophthalmic examinations, organ weights, and macroscopic or microscopic examination of the eyes

Ocular Fluid PK and PD

- Vitreous free ANX007 levels were measurable up to 30 days following single dose or repeat doses of ANX007 at all dose levels studied (**Figure 1A**). Free C1q levels were inhibited at these timepoints in treated animals (**Figure 1B**)
- Free ANX007 half-life in the vitreous was ~3 days
- Aqueous and vitreous levels were highly correlated, and aqueous levels free ANX007 were ~4-fold lower than in the vitreous (data not shown)

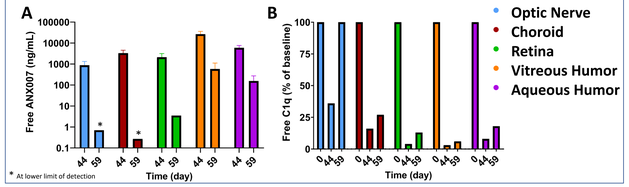
Figure 1: Mean Vitreous PK and PD



Ocular Tissue PK and PD

- No changes in total C1q levels were observed (data not shown)
- Following two 5 mg/eye doses at a one-month interval, free ANX007 was detected in all sampled tissues and fluids (**Figure 2A**). Correspondingly, in perfused tissue, C1q suppression was observed through 30 days post-last dose (Day 59) in the retina and choroid and through 15 days post-last dose (Day 44) in the optic nerve samples, confirming drug distribution to the back of the eye (**Figure 2B**, control animals indicated as Day 0/baseline)
- Baseline optic nerve C1q levels are the highest of the tissues assessed (non-normalized data not shown) and may result in faster recovery of C1q levels in the optic nerve as compared to other tissues and fluids

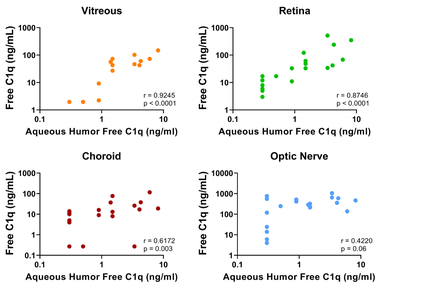
Figure 2: Mean Ocular Tissue and Fluid PK and PD



RESULTS (continued)

- Presence of free ANX007 was associated with a complete reduction of free C1q (full target engagement) in ocular fluids and perfused ocular tissues
- Aqueous humor free C1q suppression reflects C1q suppression in perfused tissues, particularly the vitreous and retina, as shown in **Figure 3**

Figure 3: Correlation of Aqueous Humor C1q Suppression to Vitreous and Ocular Tissues



CONCLUSIONS

- **Following IVT administration, ANX007, a Fab antibody fragment targeted against C1q, distributes to relevant sites of neurodegenerative ophthalmic disease within the retina, with clear evidence of C1q target engagement**
- **All dose levels were well-tolerated**
- **Engagement of C1q in the aqueous humor reflects inhibition in the vitreous and retinal tissue, and it is hypothesized to be sufficient to mitigate classical complement activation in neurodegenerative ophthalmic disease**
- **These results support further clinical evaluation of ANX007 for the treatment of such diseases; the ARCHER study, a Phase 2 study of ANX007 in GA, is currently enrolling (NCT04656561)**

REFERENCES [1]Stasi et al., Invest Ophthalmol Vis Sci. 2006;47(3):1024-1029; [2]Stevens et al., Cell. 2007;131:1164-1178; [3]Tezel et al., Invest Ophthalmol Vis Sci. 2010;51(10):5071-5082; [4]Howell et al., J Clin Invest. 2011;121:1429-1444; [5]Silverman et al., Mol Neurodegener. 2016;11:24.

AUTHOR DISCLOSURES At the time the work was completed, all authors were employees of Annexon Biosciences (Code E, Employment), except for Kirsten Mease who is a paid consultant from ToxStrategies, Inc. (Code C, Consultant)

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