



Unlocking protein production with **translational read-through** for **rare genetic** diseases

Oppenheimer 29th Annual Healthcare Conference
March 20, 2019

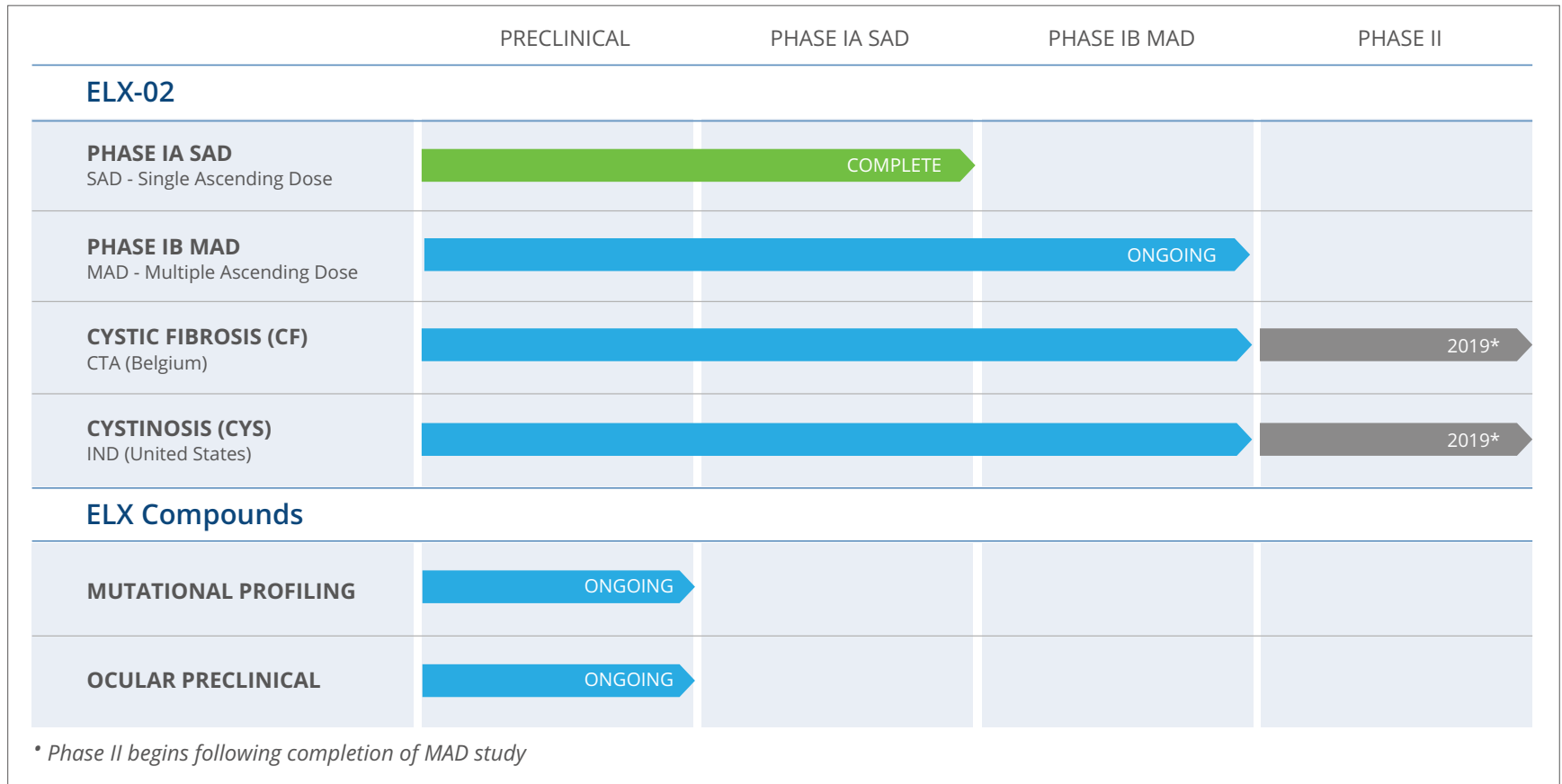
Forward-Looking Statements

This presentation contains forward-looking statements, which are generally statements that are not historical facts. Forward-looking statements can be identified by the words "expects," "anticipates," "believes," "intends," "estimates," "plans," "will," "outlook" and similar expressions. Forward-looking statements are based on management's current plans, estimates, assumptions and projections, and speak only as of the date they are made. We undertake no obligation to update any forward-looking statement in light of new information or future events, except as otherwise required by law. Forward-looking statements involve inherent risks and uncertainties, most of which are difficult to predict and are generally beyond our control. Actual results or outcomes may differ materially from those implied by the forward-looking statements as a result of the impact of a number of factors, including: the development of the Company's read-through technology; the approval of the Company's patent applications; the Company's ability to successfully defend its intellectual property or obtain necessary licenses at a cost acceptable to the Company, if at all; the successful implementation of the Company's research and development programs and collaborations; the Company's ability to obtain applicable regulatory approvals for its current and future product candidates; the acceptance by the market of the Company's products should they receive regulatory approval; the timing and success of the Company's preliminary studies, preclinical research, clinical trials, and related regulatory filings; the ability of the Company to consummate additional financings as needed; as well as those discussed in more detail in our Annual Report on Form 10-K and our other reports filed with the Securities and Exchange Commission.

Eloxx Pharmaceutical Highlights

- **Experienced Leadership Team**
 - Appointment of Dr. Susan Schneider SVP Ophthalmology
- **ELX-02 Clinical Progress**
 - EU Cystic Fibrosis Basic Science Meeting March 27th New Data
 - On Track for Completion of Phase 1b MAD 1H2019
 - Expect Topline Cystic Fibrosis Phase 2 in 2019
- **Building Ophthalmology Inherited Retinal Disorder Program**
 - Association for Research in Vision & Ophthalmology Meeting May New Data
 - IND Enabling Studies Focusing on Usher's Syndrome
 - Announced Partnership with Foundation Fighting Blindness
- **Actively Developing Opportunities for Collaboration to Advance Full Pipeline and Expand Therapeutic Programs**

Eloxx Pipeline



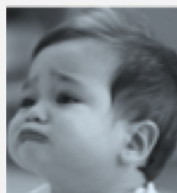
The Potential for Read-Through of Rare Genetic Diseases

>1,800

**Genetic
diseases
involve
nonsense
mutations**



**Cystic
Fibrosis**



Cystinosis



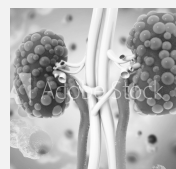
**Retinitis
Pigmentosa**



**Usher's
Syndrome**



**Primary
Ciliary
Dyskinesia**



**Polycystic
Kidney
Disease**

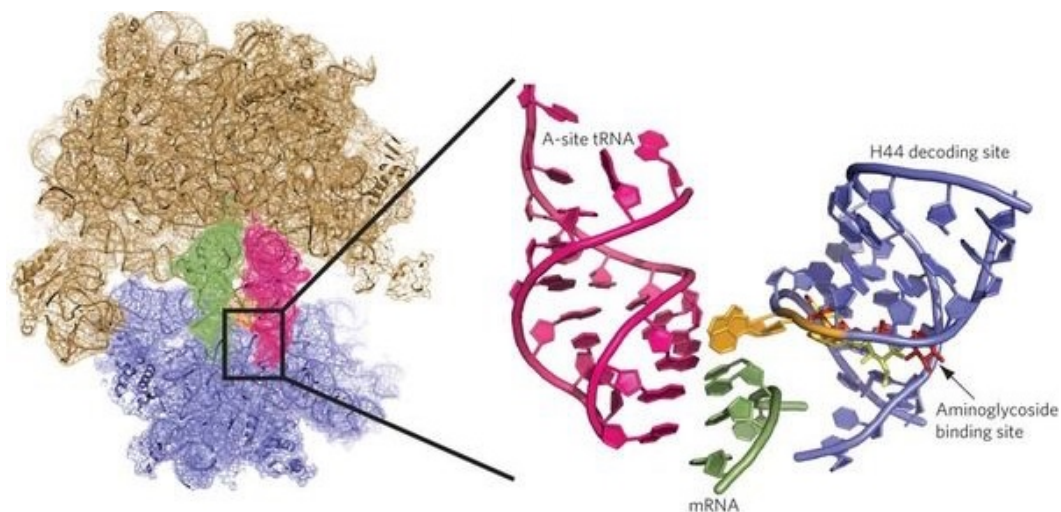
- In every genetic disease a subset of patients have nonsense mutations that impair the production of essential proteins
- Translational read through is directed at restoring the production of full length proteins by overcoming the premature stop codon and nonsense mediated decay

Aminoglycosides' tolerability profile historically limited suitability for read-through treatment of serious genetic diseases

Aminoglycosides first showed read-through activity in nonsense mediated diseases

**Advances in our
understanding
of translational
read-through enables
design of novel
small molecules**

Defined Ribosomal Binding Site



- Well defined molecular interaction with helix 44
- Role in stabilization of tRNA binding at Site A
- Optimizing scaffold by altering interaction with prokaryotic and mitochondrial ribosomes
- Defined tissue penetration and tolerability profile for read-through applications

Aminoglycoside activity observed on single pre-translocation ribosome complexes. Feldman, MB; Terry DS; Altman RB; Blanchard SC. Nature Chemical Biology volume6, pages54–62 (2010)

Ex Vivo Treatment with a Novel Synthetic Aminoglycoside NB54 in Primary Fibroblasts from Rett Syndrome Patients Suppresses MECP2 Nonsense Mutations

Manuela Vecsler^{1,2,3}, Bruria Ben Zeev^{3,7}, Igor Nudel^{1,2,3}, Yair Anikster³, Amos J. Simon³, Nina Amariglio³, Gideon Rechavi^{2,3}, Timor Baasov³, Eva Gak^{1,2,3}

¹ Sagol Neuroscience Center, Sheba Medical Center, Tel Hashomer, Israel, ² Sackler Faculty of Medicine, Tel Aviv University, Tel Aviv, Israel, ³ Edmond Pediatric Hospital, Sheba Medical Center, Tel Hashomer, Israel, ⁴ The Edith and Joseph Fisher Enzyme Inhibitors Laboratory, Schulich Faculty of Chemistry, Technion-Israel Institute of Technology, Haifa, Israel, ⁵ Cancer Research Center, Sheba Medical Center, Tel Hashomer, Israel

Abstract

Background: Nonsense mutations in the X-linked methyl CpG-binding protein 2 (MECP2) comprise a significant proportion of cases of Rett Syndrome (RTT). Naturally occurring aminoglycosides, such as gentamicin, has shown promising suppression of nonsense mutations related to severe genetic diseases, however, their use in RTT is limited by toxicity. We have developed a novel synthetic aminoglycoside, NB54, which is designed to suppress MECP2 nonsense mutations in RTT.

Conclusions: Commercial aminoglycosides and NB54 induced significant read-through of the MECP2 p.R31X nonsense mutation. However, the observed read-through efficiency, along with its significantly reduced toxicity and good biocompatibility, indicate that the novel derivative NB54 represents a better choice than commercial aminoglycosides to read-through the p.R31X mutation in other ocular diseases. Our findings suggest that NB54 may be a promising treatment for RTT.

Novel Compound Library has Demonstrated Activity across Multiple Orphan Diseases

Physiology and Pharmacology

Beneficial Read-Through of a *USH1C* Nonsense Mutation by Designed Aminoglycoside NB30 in the Retina

Tobias Goldmann¹, Annie Rebibo-Sabbah², Nora Overlack¹, Igor Nudel^{1,2,3}, Valery Belakhov³, Timor Baasov³, Tamar Ben-Yosef², Uwe Wolfrum^{1,4}, and Kerstin Nagel-Wolfrum^{1,4}

Purpose: The human Usher syndrome (USH) is the most frequent cause of inherited combined deaf-blindness. USH is clinically and genetically heterogeneous, assigned to three clinical types. The most severe type is USH1, characterized by profound inner ear defects and retinitis pigmentosa. Thus far, no effective treatment for the ophthalmic component of USH exists. The p.R31X nonsense mutation in *USH1C* leads to a severe retinal degeneration. We investigated the effect of aminoglycosides on the read-through of the p.R31X mutation in cultured cells.

Conclusions: Commercial aminoglycosides and NB30 induced significant read-through of the *USH1C* p.R31X nonsense mutation. However, the observed read-through efficiency, along with its significantly reduced toxicity and good biocompatibility, indicate that the novel derivative NB30 represents a better choice than commercial aminoglycosides to read-through the p.R31X mutation in other ocular diseases. Our findings suggest that NB30 may be a promising treatment for USH1.

PLOS ONE | DOI:10.1371/journal.pone.0131279

RESEARCH REPORT

Aminoglycoside-Induced Premature Stop Codon Read-Through of Mucopolysaccharidosis Type I Patient Q70X and W402X Mutations in Cultured Cells

Makoto Kamei¹, Karina Kasperki¹, Maria Faller¹, Emma J. Parkison-Lawrence¹, Lisa Kangsgaard¹, Valery Belakhov², Timor Baasov², John A. Hopwood¹, Doug A. Brinks¹

Received: 19 April 2013 / Revised: 13 August 2013 / Accepted: 25 September 2013 / Published online: 4 November 2013
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Abstract: The premature stop codon mutations, Q70X and W402X, are the most common *α*-iduronidase gene mutations in mucopolysaccharidosis type I (MPS I). We investigated the effect of aminoglycosides on the read-through of the Q70X and W402X mutations in cultured cells. Aminoglycosides induced a significant increase in the read-through of the Q70X and W402X mutations in cultured cells.

Aminoglycoside antibiotics restore dystrophin function to skeletal muscles of *mdx* mice

Elisabeth R. Barton-Davis¹, Laurence Cordier¹, Daria I. Shoturma², Stuart E. Leland², and H. Lee Sweeney¹

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Address correspondence to: H. Lee Sweeney, A700 Richards Building, Department of Physiology, 3700 Hambo University of Pennsylvania School of Medicine, Philadelphia, Pennsylvania 19104-6085, USA. Phone: (215) 898-8988; Fax: (215) 898-0475; E-mail: Lwsweeney@mail.med.upenn.edu.

Received for publication July 14, 1999, and accepted July 16, 1999.

Duchenne muscular dystrophy (DMD) is caused by mutations in the dystrophin gene that result in the absence of the dystrophin protein in skeletal muscle. A significant number of these mutations are premature stop codons. On the basis of the observation that aminoglycoside antibiotics induce read-through of premature stop codons in cultured cells, we investigated the effect of aminoglycosides on the dystrophin protein in *mdx* mice.

ORIGINAL RESEARCH

Synthetic Aminoglycosides Efficiently Suppress Cystic Fibrosis Transmembrane Conductance Regulator Nonsense Mutations and Are Enhanced by Ivacaftor

Xiaojiao Xue^{1,2}, Venkateshwar Mutyam³, Liping Tang³, Silpak Bilewicz³, Ming Du³, Laura A. Jackson³, Yanying Valery Belakhov³, Moran Shalev³, Jochen Schacht³, Robert J. Bridges³, Timor Baasov³, Jeong H. David M. Bedwell^{1,2,3,4,5,6}, and Steven M. Rowe^{1,2,3,4,5,6}

¹Departments of ¹Genetics, ²Medicine, ³Pediatrics, ⁴Cell Developmental and Integrative Biology, and ⁵Microbiology, and ⁶Gregory Fleming James Cystic Fibrosis Research Center, University of Alabama at Birmingham, Birmingham, Alabama; ⁷The Edith and Joseph Fisher Enzyme Inhibitors Laboratory, Schulich Faculty of Chemistry, Technion-Israel Institute of Technology, Haifa, Israel; ⁸Kresge Hearing Research Institute, Department of Otolaryngology, University of Michigan Medical School, Ann Arbor, Michigan; and ⁹Department of Physiology and Biophysics, Rosalind Franklin University, North Chicago, Illinois

Abstract

New drugs are needed to enhance premature termination codon (PTC) suppression in the treatment of cystic fibrosis (CF). We have developed a novel synthetic aminoglycoside, NB54, which is designed to suppress PTCs in CF. NB54 efficiently suppresses PTCs in CF cells and is enhanced by Ivacaftor.

synthetic aminoglycosides provide a 10-fold improvement in therapeutic index over gentamicin and other first-generation aminoglycosides. NB54 is a promising treatment for CF.

NIH Public Access

Author Manuscript
Mol Genet Metab. Author manuscript; available in PMC 2015 March 01.

Published in final edited form as:
Mol Genet Metab. 2014 March; 111(3): 374–381. doi:10.1016/j.ymgme.2013.12.007.

Long-Term Nonsense Suppression Therapy Moderates MPS I-H Disease Progression

Owen Gunn¹, Yanying Dai¹, Ming Du¹, Valery Belakhov², Jayakumar Kandasamy², Trenton R. Schoeb², Timor Baasov², David M. Bedwell¹, and Kim M. Keeling¹

¹Department of Microbiology, University of Alabama at Birmingham, Birmingham, AL, USA.
²The Edith and Joseph Enzyme Inhibitors Laboratory, Schulich Faculty of Chemistry, Technion-Israel Institute of Technology, Haifa, Israel
³Department of Genetics, University of Alabama at Birmingham, Birmingham, AL, USA

Abstract

Nonsense suppression therapy is a therapeutic approach to ameliorate the clinical course of genetic diseases caused by premature termination codons (PTC). We have developed a novel synthetic aminoglycoside, NB54, which is designed to suppress PTCs in genetic diseases.

Hum Genet (2013) 122:375–381
DOI 10.1007/s00430-013-0410-7

ORIGINAL INVESTIGATION

In vitro and ex vivo suppression by aminoglycosides of *PCDH15* nonsense mutations underlying type I Usher syndrome

Annie Rebibo-Sabbah¹, Igor Nudel¹,
Zohar M. Ahmed¹, Timor Baasov², Tamar Ben-Yosef²

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© Springer-Verlag 2013

Abstract Type I Usher syndrome (USH1) is a recessively inherited condition, characterized by profound prelingual deafness, vestibular ataxia, and progressive retinitis pigmentosa (RP). While the auditory component of USH1 can be treated by cochlear implants, there is no effective treatment for the retinal degeneration. We investigated the effect of aminoglycosides on the read-through of the p.R31X mutation in cultured cells.

such possible therapeutic approach is suppression of nonsense mutations by small molecules such as aminoglycosides. We decided to test this approach as a potential therapy for RP in USH1 patients due to the same mutation. We found that aminoglycosides induced a significant increase in the read-through of the p.R31X mutation in cultured cells.

NIH Public Access

Author Manuscript
Mol Genet Metab. Author manuscript; available in PMC 2013 January 1.

Published in final edited form as:
Mol Genet Metab. 2012 January; 105(1): 116–125. doi:10.1016/j.ymgme.2011.10.005.

The Designer Aminoglycoside NB84 Significantly Reduces Glycosaminoglycan Accumulation Associated with MPS I-H in the *Idua-W392X* Mouse

Dan Wang¹, Valery Belakhov², Jayakumar Kandasamy², Timor Baasov², Su-Chen Li³, Yu-Teh Li³, David M. Bedwell^{1,4}, and Kim M. Keeling^{1,4}

¹Department of Genetics, University of Alabama at Birmingham, Birmingham, AL 35294, USA
²The Edith and Joseph Enzyme Inhibitors Laboratory, Schulich Faculty of Chemistry, Technion-Israel Institute of Technology, Haifa, Israel
³Department of Microbiology, University of Alabama at Birmingham, Birmingham, AL 35294, USA
⁴Department of Pediatrics, University of Alabama at Birmingham, Birmingham, AL 35294, USA

Substantial Advantages for Orphan Drug Development

- **ELOXX Focus on High Unmet Medical Need**
 - Nonsense mutations represent important patient segments in over 1,800 diseases
 - Many of these diseases have no approved therapeutics
 - In some diseases the nonsense patient population is appropriate size for traditional clinical development
- **Developing Novel Therapeutics through Established Pathways**
 - Many orphan diseases have existing preclinical assays or animal models with correlations to clinical endpoints
 - Validated Phase 2 endpoints can guide phase transition and design of pivotal trials
- **Orphan Designation Confers Important Regulatory Considerations**
 - Potential for closer collaboration, accelerated development
 - Several economic or exclusivity incentives
 - In the US, Rare Pediatric Disease Priority Review Voucher Program
- **Ongoing Global Regulatory Interest in Accelerating Development for High Unmet Medical Need**

ELX-02 Clinical Development – Phase 1 Studies



January 16, 2019

Safety, Tolerability, and Pharmacokinetics of Single Ascending Doses of ELX 02, a Potential Treatment for Genetic Disorders Caused by Nonsense Mutations, in Healthy Volunteers

CLINICALTRIALS.GOV
Identifier: NCT03292302

COMPLETED



CLINICALTRIALS.GOV
Identifier: NCT03309605

A Phase 1, Randomized, Double-Blinded, Placebo-Controlled, Third Party Open, Multiple Dose Escalation, Single Center Study to Evaluate the Safety, Tolerability and Pharmacokinetics of Subcutaneously Administered ELX-02 in Independent Consecutive Cohorts of Healthy Subjects

ONGOING



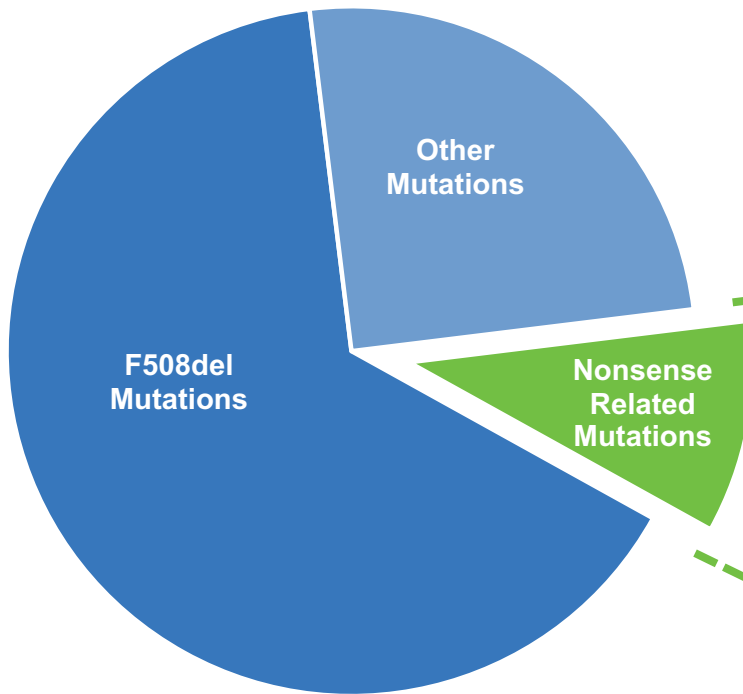
TO DATE:

- No SAE Observed
- No renal or otoacoustic SAE
- Generally well tolerated

INITIATED 6TH COHORT in Belgium
On track for final Cohort in US

Cystic Fibrosis Nonsense Mutation Patients Represent a Key Unmet Need Population

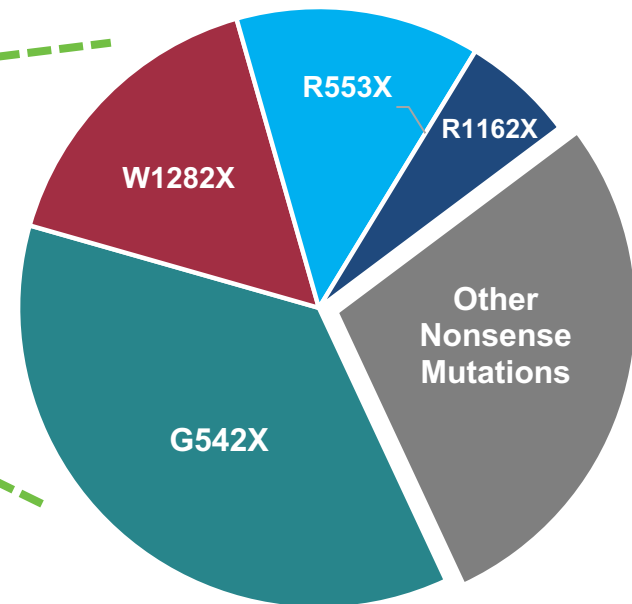
GLOBAL CYSTIC FIBROSIS PATIENTS
BY MUTATION/GENOTYPE



10 - 13%

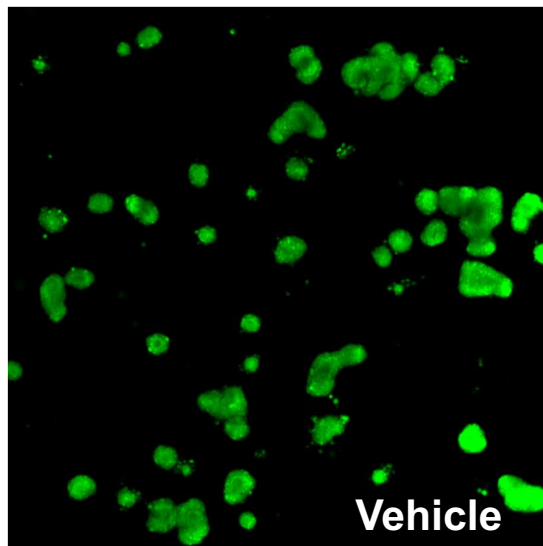
No Currently Approved Drugs
To Treat CFTR Nonsense Mutations

CFTR NONSENSE
MUTATION SUBTYPES

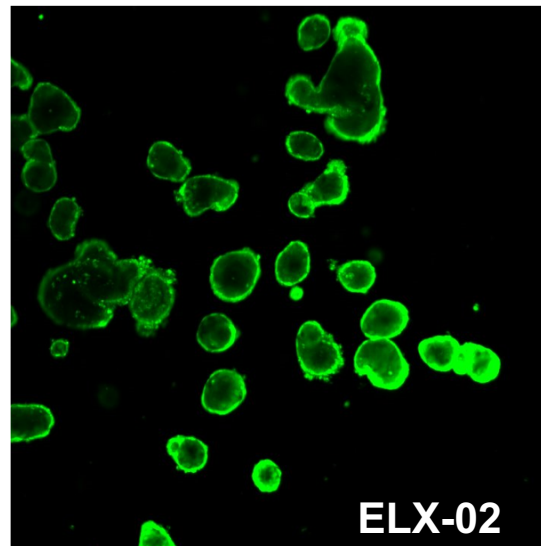


Organoid Swelling Assay Demonstrates CFTR Function

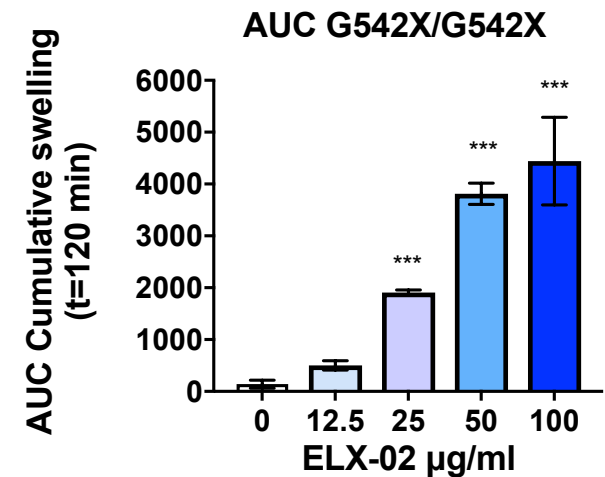
Cystic Fibrosis Organoid dose-responsive swelling assay response



Patient Organoid without drug treatment:
No Swelling of Organoids

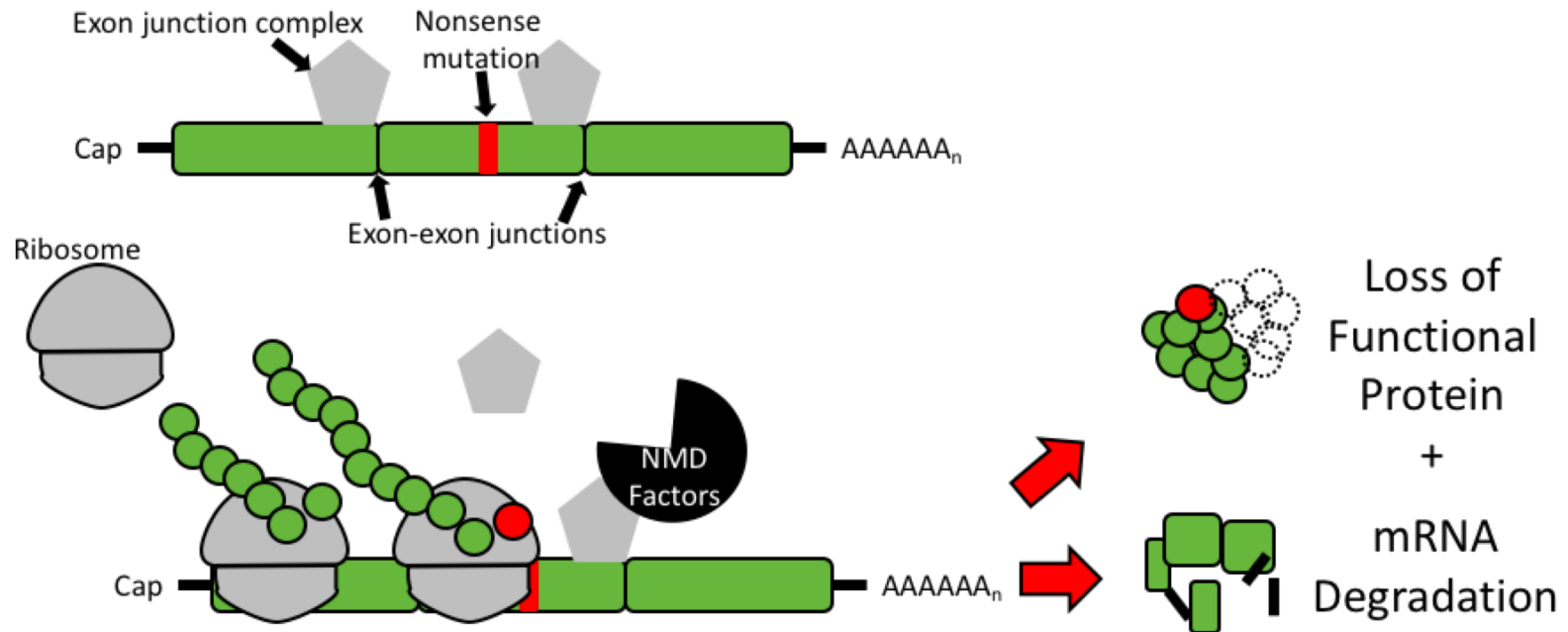


Patient Organoid with ELX-02 treatment:
Swelling of Organoids



Swelling quantification of patient organoid
with ELX-02 treatment.

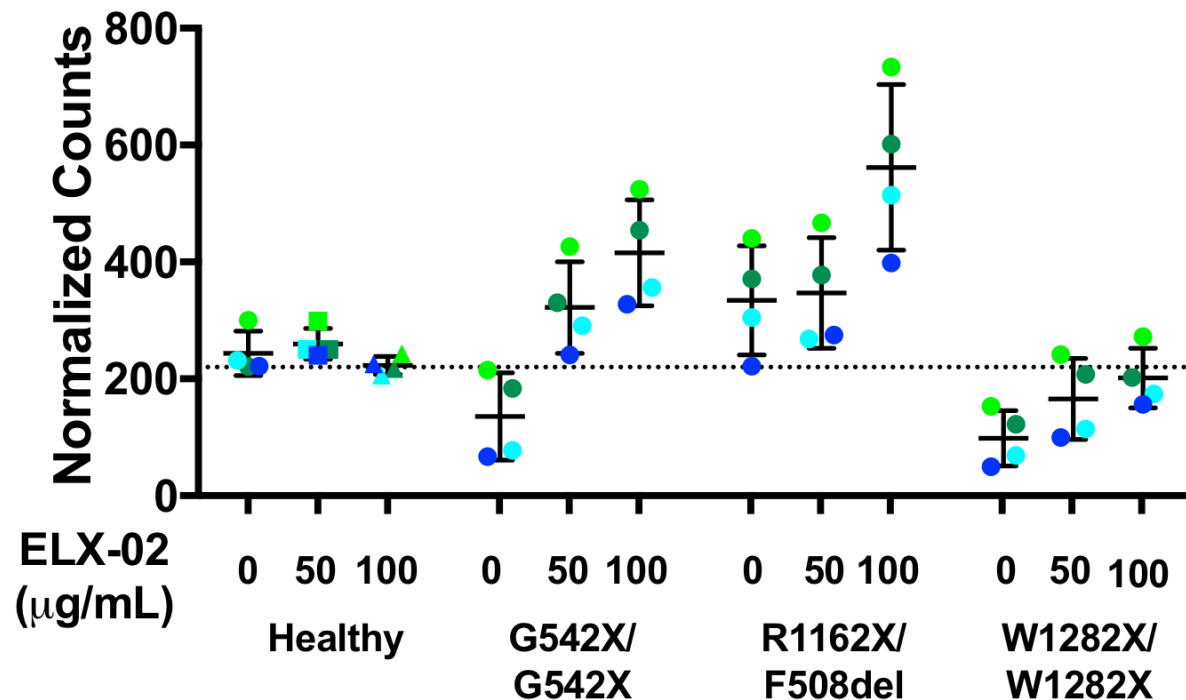
Nonsense Mutations Can Cause a “Double-Hit”, Loss of mRNA and Functional Protein



For more information: Kurosaki & L.E. Maquat. Nonsense-mediated mRNA decay in humans at a glance. *J. Cell Sci.* 1;129(3):461-7 (2016).

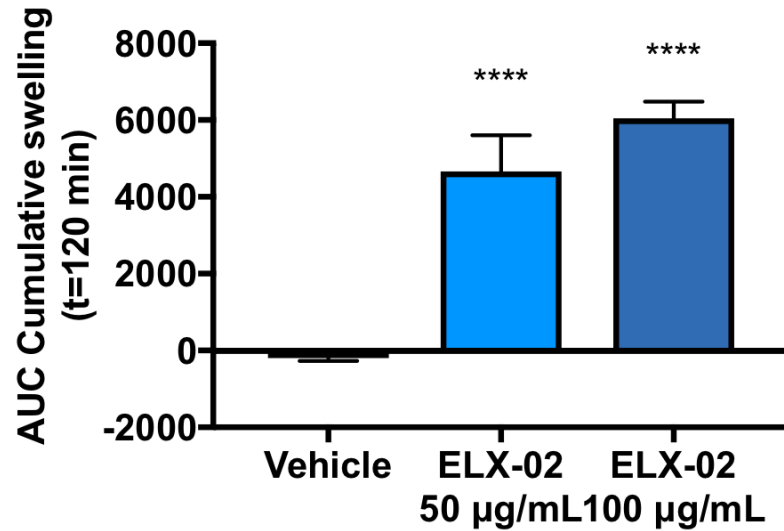
CFTR mRNA Comparisons to Healthy, Wild-type Control

Cystic Fibrosis Organoid Responsive to ELX-02 demonstrates elevations above wild-type

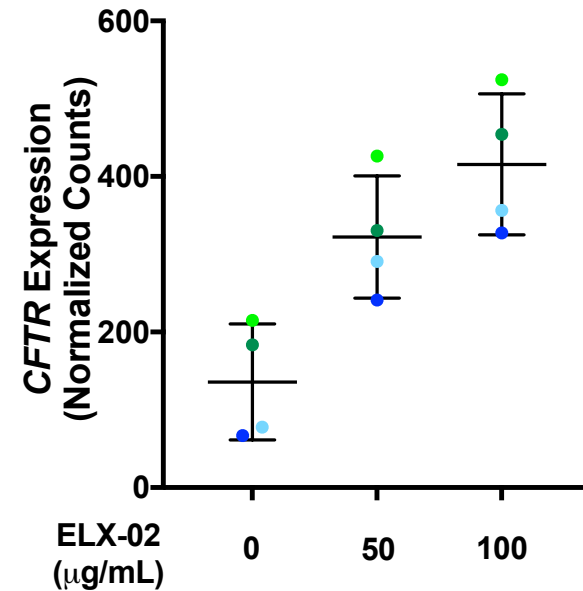


Homozygous Nonsense Mutation (G542X)

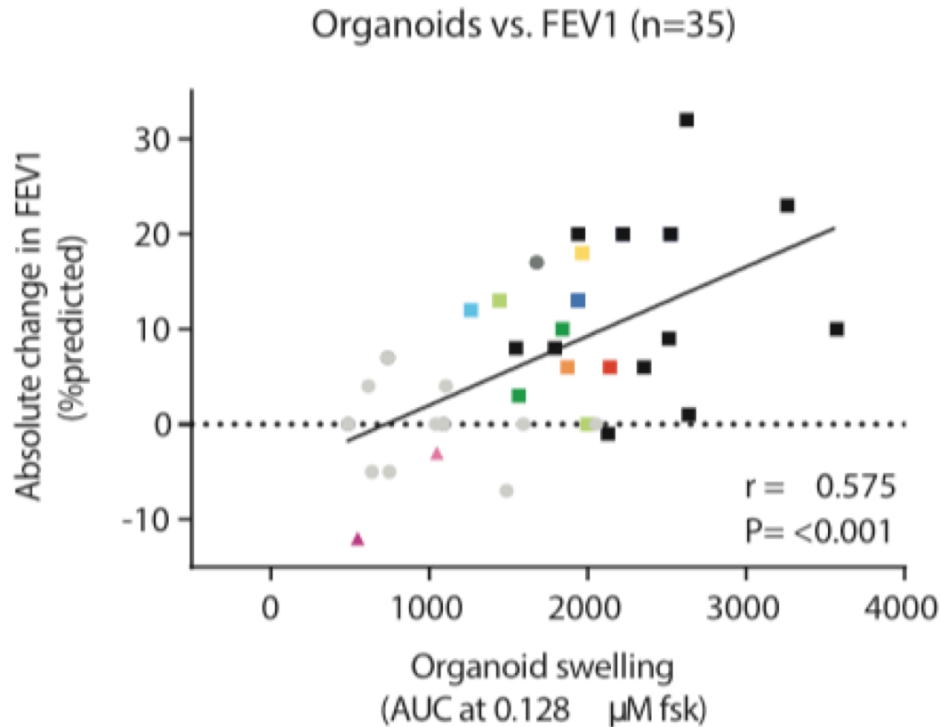
SWELLING (FUNCTION) ASSAY



NANOSTRING (MRNA) ASSAY



New HUB Organoid Data Correlates with Clinical Response



✕ FEV1 response correlates with organoid swelling in samples collected across multiple clinical trials.

✕ Genistein & Curcumin (circles)

✕ Ivacaftor (squares)

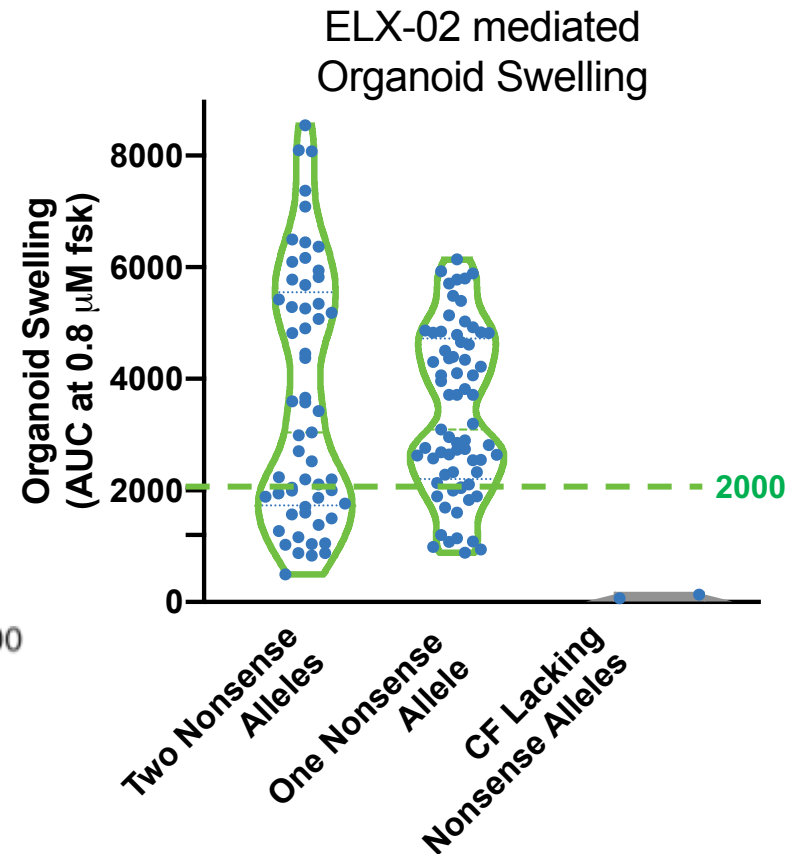
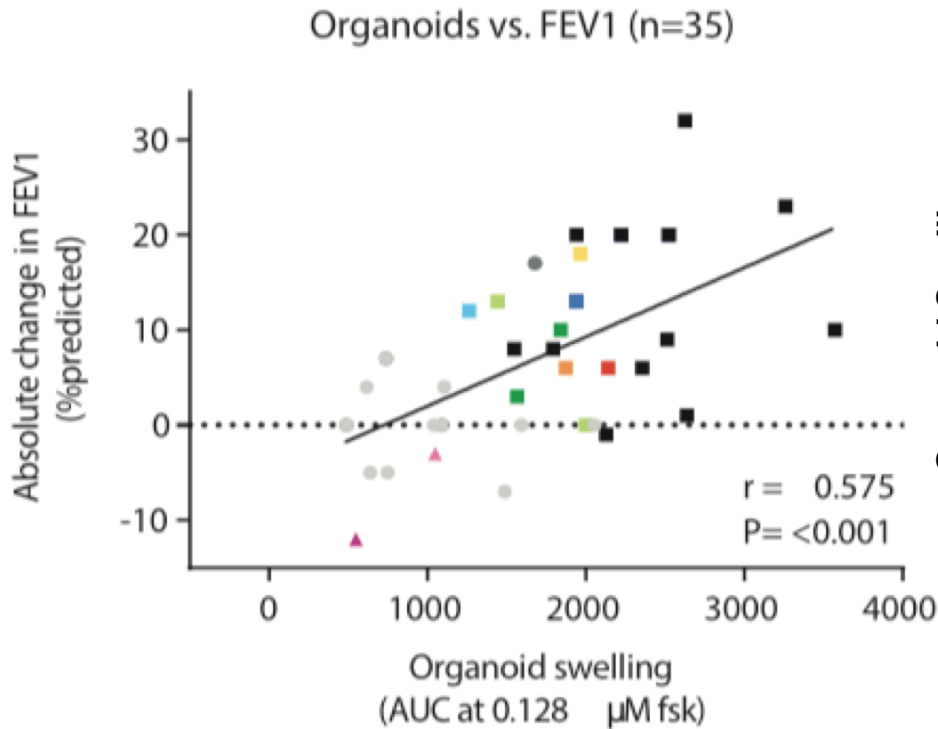
✕ Lumacaftor & Ivacaftor (triangles)

✕ Organoid results correlate with sweat chloride change

✕ ($r = -0.708$, $p < 0.001$)

Berkers et al. Cell Reports (2019) Rectal Organoids Enable Personalized Treatment of Cystic Fibrosis.

ELX-02 Response in Organoids Compares Favorably to Published Results

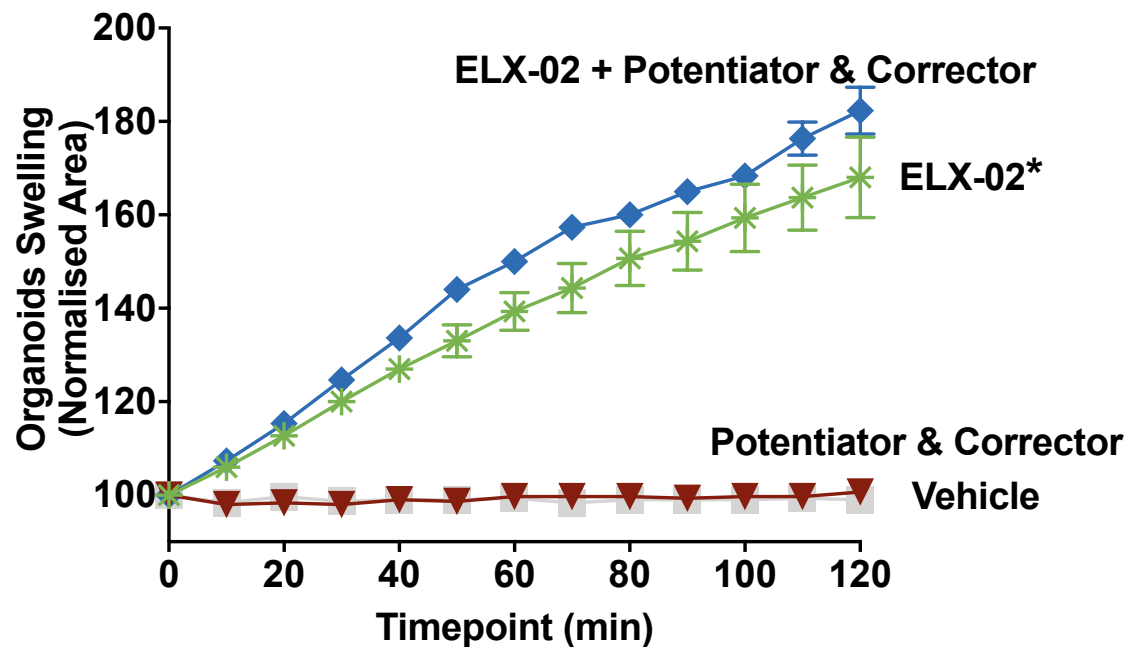


Berkers et al. Cell Reports (2019) Rectal Organoids Enable Personalized Treatment of Cystic Fibrosis.

Eloxx data on file.

Homozygous Nonsense Mutation (G542X)

Cystic Fibrosis Organoid Responsive to ELX-02, Enhanced by Combination



* Source: European Cystic Fibrosis Society 41st Conference, Belgrade, Serbia, June 2018

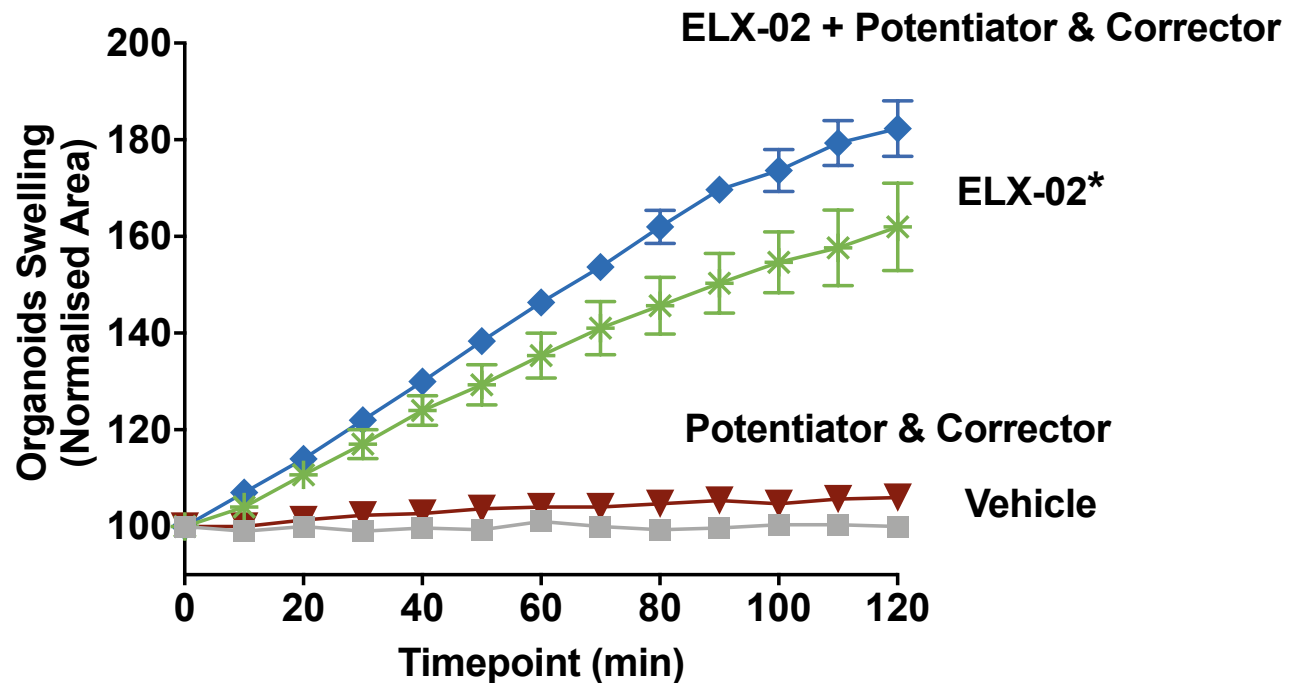
- 100 µg/mL ELX-02

- Error Bars represent SEM

- $p < 0.0001$ vs Vehicle, Potentiator & Corrector. N.S. vs. ELX-02 + Potentiator & Corrector by Ordinary One Way ANOVA With Sidak's Multiple Comparison Testing on AUC transformations

Complex Heterozygous Nonsense Mutation (G542X:R1066C missense)

Cystic Fibrosis Organoid Responsive to ELX-02, Enhanced by Combination

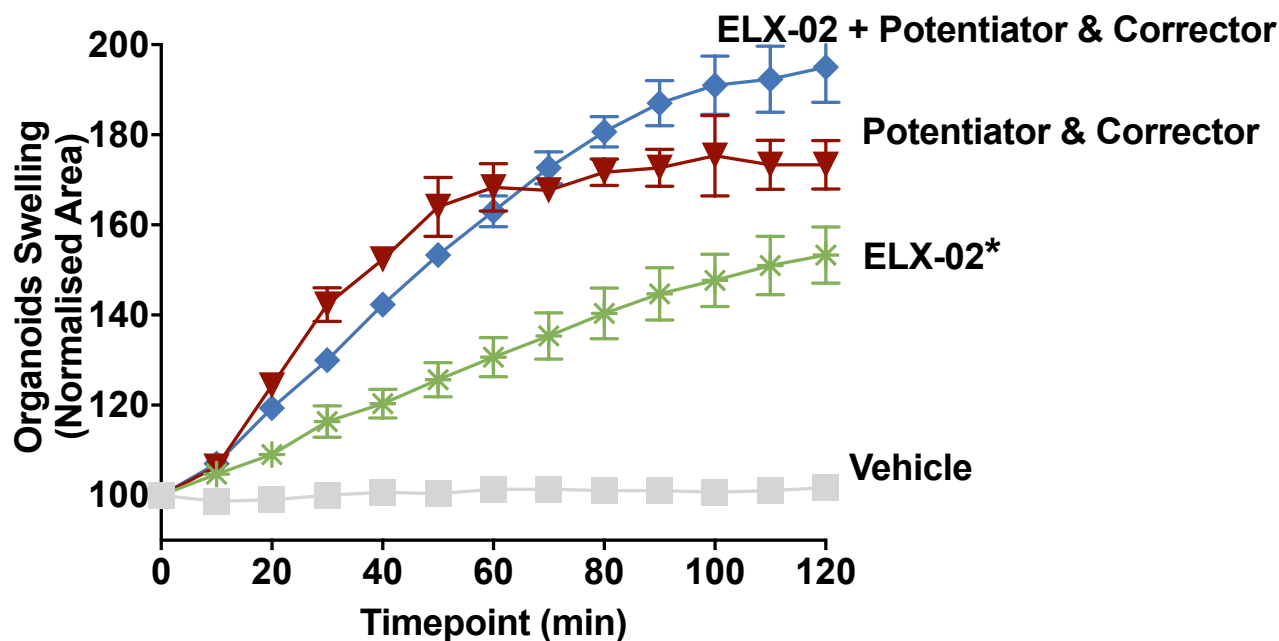


*Source: European Cystic Fibrosis Society 41st Conference, Belgrade, Serbia, June 2018

- 100 µg/mL ELX-02
- Error bars represent SEM
- $p < 0.0001$ ELX-02 vs Potentiator & Corrector, Vehicle, $p < 0.05$ ELX-02 vs Triple treatment by One Way Anova with Sidak's Multiple Comparison Testing on AUC transformations.

Complex Heterozygous Nonsense Mutation (G542X:F508del)

Cystic Fibrosis Organoid Responsive to ELX-02, Enhanced by Combination



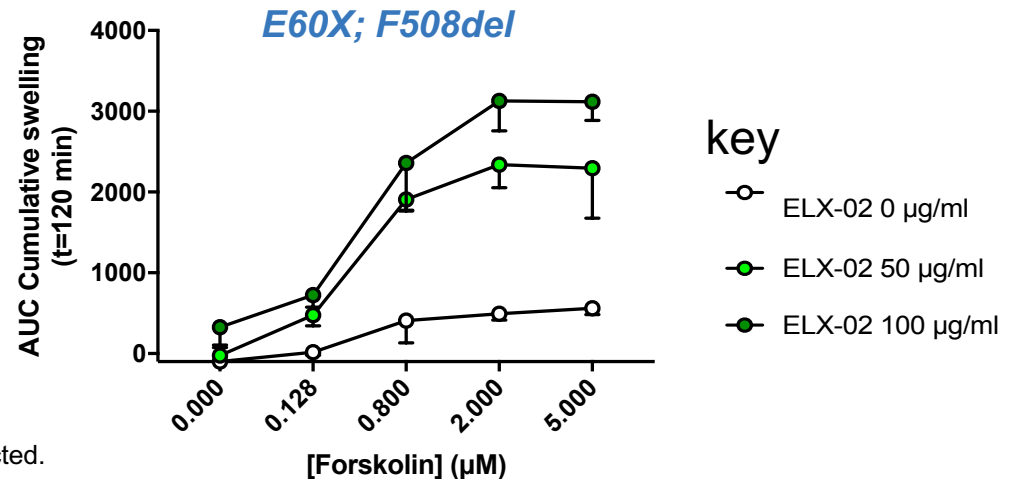
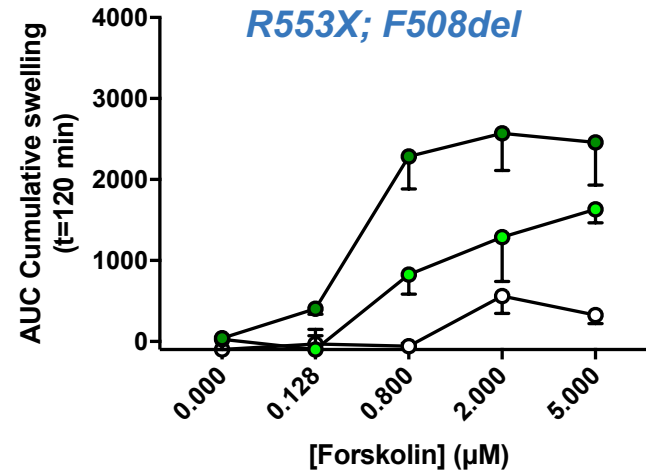
*Source: European Cystic Fibrosis Society 41st Conference, Belgrade, Serbia, June 2018

- 100 µg/mL ELX-02
- Error bars represent SEM
- $p=0.00012$ ELX-02 vs Potentiator & Corrector, $p=0.0006$ ELX-02 vs Vehicle, ELX-02 vs ELX-02+ Potentiator & Corrector by One Way Anova with Sidak's Multiple Comparison Testing on AUC transformations.

ELX-02 Organoid Response is Observed Across a Range of CF Genotypes

- ✕ *G542X; G542X*
- ✕ *G542X; F508del*
- ✕ *G542X; R1066C*
- ✕ *R1162X; F508del*
- ✕ *G542X; W1282X*
- ✕ *W1282X; W1282X*
- ✕ *R553X; F508del*
- ✕ *E60X; F508del*

New Genotypes

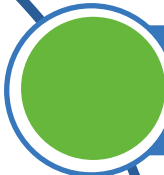
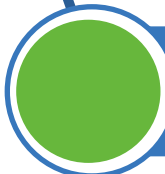
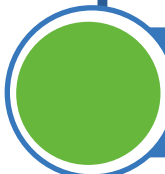



Eloxx data on file
Data from multiple individuals with the same genotypes also collected.

Clinical Update for ELX-02 Phase 2 in Cystic Fibrosis

- ✓ Orphan Drug Designation granted by EMA
- ✓ Clinical Trial Application (CTA) for Phase 2 Study received final approved by the FAMHP in Belgium
- Expanding MAD Study
 - ✓ 6th cohort initiated in Belgium
 - Completion in 1H2019 with final cohort in US
- Engaging with investigators on a protocol for Phase 2 to insure rapid execution
 - No more than 24 patients
 - Will evaluate changes in sweat chloride at ascending doses
 - Planned enrollment will focus on patients with G542X nonsense mutation on one (complex heterozygote) or both alleles (homozygote)
- Expect Topline Cystic Fibrosis Phase 2 data in 2019
- EU Basic Science Meeting March 27th New Data

Substantial Opportunities in Inherited Retinal Diseases

		Incidence	US Prevalence	Nonsense Proportion
	Usher Syndrome	3.2-6.2/ 100K	16.3K	20%
	Retinitis Pigmentosa	1-5/10K	67K	15-50%
	Choroideremia	1-9/100K	6.5K	36%
	Stickler's Syndrome	1-9/ 100K	66K	20%

Population values from National Organization for Rare Disorders (NORD) and Orphanet

Han et al, Korean J Pediatr 2016.; Wilkin et al, Am J Med Genet 2000; Freund et al, Mol Genet Genomic Med 2016; Aparisi et al, Orphan J Rare Dis 2014

Inherited Retinal Disease Program Initiated

- **Advancing Several Compounds from our Library**
- **Currently in IND-Enabling Studies**
 - Demonstrated positive activity on nonsense mutations across a variety of inherited retinal disorders
 - Favorable tolerability profile
 - Support the use for intravitreal injection
- **Development Focus on Ushers Syndrome, Leber's Congenital Amaurosis, or Other Forms of Retinitis Pigmentosa Caused by Nonsense Mutations**
- **Wide Ranging Partnership with the Foundation Fighting Blindness**
 - Partnership includes broad scientific engagement to support Eloxx's ocular portfolio development
- **ARVO Meeting May 2, 2019 New Data**

Usher Syndrome

- **No drugs approved or in late-stage development for nonsense variants**
- **Significant unmet medical need for nonsense forms**
 - **Over 4,000 patients in North America alone**
- **Academic collaborations have demonstrated activity with Eloxx Novel Library Compounds**
 - **Read through protein expression In vitro retinal compatibility**
- **Encouraging IND enabling studies supporting preservation of electroretinogram (ERG) and retinal histology**

Eloxx Pharmaceutical Highlights

- **Experienced Leadership Team**
 - Appointment of Dr. Susan Schneider SVP Ophthalmology
- **ELX-02 Clinical Progress**
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 - On Track for Completion of Phase 1b MAD 1H2019
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- **Building Ophthalmology Inherited Retinal Disorder Program**
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- **Actively Developing Opportunities for Collaboration to Advance Full Pipeline and Expand Therapeutic Programs**



Thank you.

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March 20, 2019