Usher Syndrome Type 1C Research Update

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Outline

• **Usher syndrome review**
  • Characteristics and Prevalence
  • Types and subtypes
  • Clinical management and therapies under investigation

• **Lentz Lab Mission**
  • *USH1C* gene
  • Knock-in mouse model

• **Antisense Therapy for Acadian USH1C**
  • Targeting 216A mutation
  • Treatment of USH1C mice with ASOs

• **Gene Therapy for all USH1C**
  • USH1C gene therapy development
  • Treatment of USH1C mice with gene replacement therapy
Usher syndrome review – Prevalence and Types

- Usher syndrome (USH or US) is the leading genetic cause of concurrent hearing and vision impairment. Some individuals also have imbalance.

- Estimated 1 in ~20,000 individuals in the world have Usher

- Currently, there are 3 clinical types and 11 subtypes (genes):

<table>
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Usher syndrome review - Diagnosis

- Diagnosis is established with **clinical features** based on-
  - Severity of sensorineural hearing impairment (HI)
  - Presence of vestibular areflexia (imbalance)
  - Age of onset of retinitis pigmentosa (RP) – progressive visual loss that begins with night-blindness

**Type 1 (USH1)**
- Severe-profound HI
- Vestibular areflexia
- RP beginning in early adolescence

**Type 2 (USH2)**
- Mild - severe HI
- RP beginning in late adolescence

**Type 3 (USH3)**
- Post-lingual HI
- Variable Balance
- RP beginning in adulthood
**Usher syndrome review – Genetic Testing**

- Diagnosis is established with clinical features
- **Genetic testing** confirms diagnosis and determines the specific mutation that is the cause of the patient’s hearing and visual symptoms
- Multigene genetic testing panels are most commonly used-
  - Usher syndrome multigene panel
  - Inherited Retinal Dystrophy panel
  - Hereditary Hearing Loss panel

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Usher syndrome review – Clinical Management

- **Treatments**
  - USH1 – cochlear implants or sign language; occupational and physical therapy; low vision aids
  - USH2 – hearing aids, cochlear implants; low vision aids
  - USH3 – hearing aids, cochlear implants; low vision aids

- **Therapies under investigation**
  - UshStat retinal gene replacement of *MYO7A* for USH1B patients
    - Trial began in 2014 and has completed the dose escalation phase, however it is currently not recruiting
    - Clinicaltrials.gov (NIH) identifier: [NCT02065011](https://clinicaltrials.gov/ct2/show/NCT02065011)
  - QR-421a retinal antisense (ASO) treatment for USH2A patients with *USH2A* exon 13 mutations
    - ProQR Therapeutics
    - 3-month interim findings (March 2020) showed the ASO was safe and well tolerated in 8 USH2A patients, 2 patients also showed improvements in retinal sensitivity, retinal structure, and visual fields
    - More information:
      - [www.proqr.com](http://www.proqr.com) (website)
      - Clinicaltrials.gov (NIH) identifier: [NCT03780257](https://clinicaltrials.gov/ct2/show/NCT03780257)
    - Recruiting
Usher syndrome review – Clinical Management

• Therapies under investigation, cont.
  • CL-17-01 retinal antioxidant treatment for RP with Usher syndrome (Australia)
    • Nacuity Pharmaceuticals, Inc.
    • More information:
      • www.nacuity.com
      • Clinicaltrials.gov (NIH) identifier: NCT04355689
    • Not yet recruiting
  
  • NPI-001 oral antioxidant given to patients with RP (including all USH)
    • Nacuity Pharmaceuticals, Inc.
    • Completed a 30-patient study that showed it was well tolerated and improvements in retinal sensitivity
    • Currently conducting an extension study
    • More information: www.nacuity.com
NIH National Eye Institute

- Dr. Tiansen Li and Dr. Anand Swaroop
- Developed USH1C retinal organoid models from several USH1C patient fibroblast skin cells
- Currently screening known small molecule and other drug candidates
- Sponsored by Usher 2020 Foundation

Johannesburg Gutenberg, University of Mainz & LMU Munich

- Dr. Uwe Wolfrum and Dr. Nikolai Klymiuk
- Developed a transgenic USH1C pig model with hearing, balance, and visual deficits
- Currently characterizing the pig model, studying mechanisms of USH1C disease, and creating a breeding herd to test new therapies
- Sponsored by Usher 2020 Foundation and FAUN Foundation
Usher syndrome review – USH1C Pre-clinical Therapeutic Development

**Oregon Health & Science University**
- Dr. John Brigande
- Developing an USH1C non-human primate model and genetic therapy approaches
- Sponsored by NIH/NIDCD

**Odylia Therapeutics**
- Founded by MEEI and Usher 2020 as a non-profit company to bring rare retinal disease therapies to the clinic
- In collaboration with Drs. Uwe Wolfrum, Kerstin Nagel-Wolfrum, Nikolai Klymiuk, Pigmod, and other experts, developing a comprehensive drug development plan to bring USH1C gene replacement therapy to clinical trials
- Currently testing AAV gene therapy in the USH1C pig model
- Sponsored by Usher 2020 and FAUN Foundation
# Usher syndrome Research - Lentz Lab Mission

- To understand **disease mechanisms** – how genetic changes cause hearing, balance, and vision impairments
- To **develop new therapies** for the treatment of hearing impairment, imbalance, and visual loss associated with **Usher syndrome**
- Focus on subtype **USH1C**

## Types:

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## Subtypes:
**Subtype USH1C is caused by mutations in the ****USH1C** gene

- **USH1C** gene is on chromosome 11 and contains 27 exons that are used to encode 3 families of Harmonin proteins (Harmonin-a, -b, -c)
- Harmonin proteins are found in the ear and eye
- ~47 mutations known* to cause USH1C

* Listed as “Pathogenic” in at least 1 US or European Database: ClinVar; HGMD; UMD-USHbases; LOVD (2020)
Splicing is the process by which introns are removed to produce a mature messenger RNA that only contains exons used to make a protein.

216G>A splicing mutation is a founder mutation in the Acadian populations.

The G → A change at position 216 in the USH1C gene causes aberrant splicing that results in a truncated mRNA and protein; and no functional protein in the eye and ear.

**Correct Splicing**

- exon 1
- exon 2
- exon 3
- exon 4

**Aberrant Splicing**

- exon 1
- exon 2
- exon 3
- exon 4

216A

Lentz et al. 2005
USH1C mouse model in the Lentz Lab

- Knock-in mouse model of the Acadian USH1C c.216G>A
- Similar symptoms as patients
  - Severe-profound hearing loss  →  abnormal/no auditory brainstem response (ABR)
  - Balance problems  →  circling in cage and head tossing
  - Mild vision loss  →  reduced electroretinogram (ERG) and slow photoreceptor loss

USH1C mouse model exhibits hearing impairment, circling behavior, & visual dysfunction.

Lentz et al. 2007, 2010
Antisense Therapy for Acadian USH1C

- Designed an Antisense Oligonucleotide (ASO) to target the 216A mutation in the pre-spliced RNA and correct splicing
- ASOs are short pieces of nucleic acids (DNA/RNA) that bind to their target
- **216A-targeted ASO** is designed to bind to the 216A mutation which blocks splicing proteins from cutting at the wrong place, and forces correct splicing

![ASO-corrected 216A Splicing](image)
ASOs restore hearing in USH1C mice

- ASOs injected systemically restore hearing thresholds (ABRs) in USH1C mice
- Treatment must be given before hearing begins-

Single Systemic ASO Treatment given on post-natal day (P)1

![Hearing Thresholds (ABRs)](image)

Lentz et al 2013, 2017
ASOs restore balance in USH1C mice

• ASOs restore balance behavior in USH1C mice
• Treatment timing?
ASOs restore vision in USH1C mice

- ASOs injected one time directly into the eye by intravitreal injection restore visual function in USH1C mice for 3 months
- Continued treatments maintain visual benefits for up to 1 year

**Multiple Treatments**

4 Doses – P21, 3, 6, 9 months

*Lentz unpublished data*
Development of ASO therapy for Acadian USH1C

- Next steps to develop the ASO as a treatment for visual loss in USH1C
- NIH funded grant to
  1) Optimize the ASO drug
  2) Identify USH1C patients and determine clinical outcomes measures
Optimization of ASOs for Acadian USH1C – Aim 1

• Current best performing ASO:
  • Single treatment improves vision by ~ 20 - 40% for 3 months

• Can we increase ASO activity or duration of effect?
  • Test 100-200 new ASOs with slight modifications in sequence and chemistry

• Currently, we have completed ~65% of the testing and are waiting for long-term studies for some of them (duration of effect)

• Once the testing is complete and have the ASO with the highest activity and/or longest effect, the next steps are to prove it’s safe
Prospective Natural History of Visual Loss in USH1C – Aim 2

• Identify Acadian USH1C patients
  • *Retrospective NHS*, enrolling:
    • Louisiana residents – All USH
    • Non-Louisiana residents – All USH1C

• Prospective NHS to determine clinical outcomes measures that could be used to guide a clinical trial
  • Enrolling USH1C patients age 12 – 65 years
  • 4 clinic visits – 1 visit every 6 months for 2 years
  • Clinics –
    • Dr. Maria Reinoso, LSUHSC, New Orleans, Louisiana
    • Dr. Robert Koenekoop, MUHC, Montreal, Canada
    • Dr. Wadih Zein, NEI, Bethesda, Maryland

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**Retrospective Natural History Study of Usher Syndrome in Louisiana (2014-present) Patient Population**

<table>
<thead>
<tr>
<th>Total Enrolled</th>
<th>103</th>
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<tbody>
<tr>
<td>Louisiana</td>
<td>75</td>
</tr>
<tr>
<td>Canada</td>
<td>12</td>
</tr>
<tr>
<td>Other</td>
<td>16</td>
</tr>
<tr>
<td>% Males</td>
<td>50% (52/103)</td>
</tr>
<tr>
<td>Age range</td>
<td>18 mo – 93 years</td>
</tr>
<tr>
<td>With genetic confirmation</td>
<td>70% (72/103)</td>
</tr>
<tr>
<td>USH1</td>
<td>90/103</td>
</tr>
<tr>
<td>USH1B</td>
<td>1/67</td>
</tr>
<tr>
<td><strong>USH1C</strong></td>
<td>65/67</td>
</tr>
<tr>
<td>USH1D</td>
<td>1/67</td>
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<tr>
<td>USH2</td>
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<tr>
<td>USH2A</td>
<td>2/2</td>
</tr>
<tr>
<td>USH3</td>
<td>3/103</td>
</tr>
<tr>
<td>USH3A</td>
<td>3/3</td>
</tr>
<tr>
<td>Atypical-USH</td>
<td>1/103</td>
</tr>
<tr>
<td>Other</td>
<td>1/103</td>
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Gene therapy for USH1C

- **Gene of interest**
  - *Normal* USH1C gene delivery to restore functional harmonin protein

- **Viral vector**
  - Adeno-associated virus (AAV)
    - Replication deficient (cannot reproduce without a *helper virus* present)
    - Not known to cause disease in humans
Gene therapy restores hearing in USH1C mice

- Gene therapy is injected directly into the ear
- Gene therapy (AAV-Ush1c-b) restores hearing and balance in USH1C mice
- Treatment must be given before hearing begins

Hearing thresholds (ABRs)

Balanced Behavior (Open-field)

Pan et al. 2017
Does gene therapy restore vision in USH1C mice?

- **Gene therapy** is injected directly into the eye by subretinal injection
- Expression of **full-length** *Ush1c* in the AAV treated eye, but not the untreated eye, of USH1C mice
- Improves vision in USH1C mice?
**USH1C Research Summary and Next steps**

- **ASO therapy** restores hearing, balance, and vision in USH1C mice
- Currently **optimizing the ASO drug** to improve its activity and/or duration of effect as a **treatment for visual loss in Acadian USH1C patients**
- **Gene replacement therapy** restores hearing and balance in USH1C mice
- Currently testing gene replacement therapy for visual loss in USH1C mice
- **Natural History Studies for patients**-
  1) *retrospective* NHS to improve our understanding of the natural clinical history of Usher syndrome in Louisiana
  2) *Prospective* NHS of visual loss in USH1C
  3) *Prospective* NHS of imbalance in USH1C
  - Contact Dr. Lentz for more information about participating – jlentz@lsuhsc.edu
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