Suprachoroidal delivery for ocular gene therapy: nonclinical experiments evaluating non-viral DNA nanoparticles.

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Financial Disclosures

None relevant to this presentation
DNA nanoparticles offer the potential for safe, efficacious, and repeat dosing of ocular gene therapy

Potential advantages:
• Unlike AAV (payload capacity of 5 kb), can transfer large genes (up to ~20 kb)
• Safety: Non-immunogenic, without viral capsid proteins or pre-existing immunity.
  • Potential for repeat and greater dosing
• Efficacy: in numerous ocular animal model, higher doses may be used to enhance transfection
• Manufacturing, Simpler than viral-based gene therapy

Potential disadvantages:
• Durability: May not represent one time therapy

DNA nanoparticle Gene Therapy: Well established literature

Comparative Analysis of DNA Nanoparticles and AAVs for Ocular Gene Delivery

DNA nanoparticles are safe and nontoxic in non-human primates

Lentiviral Vector Gene Transfer of Endostatin/Angiostatin for Macular Degeneration (GEM) Study

AAV8-antivegfr5 Ocular Gene Transfer for Neovascular Age-Related Macular Degeneration

Development of an inducible anti-VEGF:AAV gene therapy strategy for the treatment of wet AMD
Suprachoroidal (SC) injection offers the potential for safe, targeted, and efficient ocular gene therapy

- **Targeted treatment** of posterior tissues possible via SC injection
  - Spread of injectate flows circumferentially and posteriorly
- **Safety**
  - Avoids the risks of sub-retinal surgery
  - Does not require detachment of the photoreceptors from the RPEs, without associated risk of iatrogenic injection to already compromised disordered retina
  - SC injection procedure training is minimal
- **Access to care**
  - Does not require specialized gene therapy surgery treatment centers
  - In-office SC injection procedure is less expensive than surgical procedures
  - Procedure time is significantly less than standard sub-retinal procedure
Suprachoroidal Injection of DNPs in Non-Human Primates and Rabbits

**Study Objective**
- Evaluate the safety, tolerability, and retinal cell transfection following SC injection of DNPs

**Design**
- Ophthalmic examinations at Day 0, 1, and 7
  - Surface morphology, ocular inflammation, direct and indirect ophthalmoscopy, IOP, ERG
  - Eyes were enucleated at Day 7 and 21
    - Choroid and retina separated and processed for evaluation of luciferase activity

<table>
<thead>
<tr>
<th>Species</th>
<th>Group (n=4)</th>
<th>Test article</th>
<th>Route of Administration</th>
<th>Volume</th>
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<td>1</td>
<td>Vehicle</td>
<td>SC Injection</td>
<td>100 µL</td>
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<tr>
<td></td>
<td>2</td>
<td>Ellipsoid DNPs Luciferase</td>
<td>SC Injection</td>
<td>100 µL</td>
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<tr>
<td></td>
<td>3</td>
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<td>4</td>
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</tr>
</tbody>
</table>

Ellipsoids
- Colloidally stable DNPs Suspended in Saline
  - 8-10 nm in diameter

Rods
- Colloidally stable DNPs Suspended in Saline
  - 8-10 nm in diameter
NHP: DNA Nanoparticles Transfect RPE + Choroid and Retina

RPE-CHOROID

- Control (Saline)
- Ellipsoid
- Rod
- Ellipsoid
- Rod

Day 8
Day 22

Log (RLU/mg protein)

***
***
***

1-way ANOVA, p<0.0001.
Bonferroni's multiple comparison test: * p<0.05, ** p<0.01, *** p<0.001

RETINA

- Control (Saline)
- Ellipsoid
- Rod
- Ellipsoid
- Rod

Day 8
Day 22

Log (RLU/mg protein)

*

1-way ANOVA, p=0.0088.
Bonferroni's multiple comparison test: * p<0.05, ** p<0.01
Rabbit: DNA Nanoparticles Transfect the RPE + Choroid and Retina

Non Viral-Luciferase, Rabbit CHOROID

Non Viral-Luciferase, Rabbit RETINA
Study Summary

• Luciferase activity observed in the retina and RPE+choroid
• In rabbits, SC injection comparable to subretinal injections of luciferase DNPs produced activity
• SC injections of DNPs were generally well-tolerated across groups in both species

• Safety
  • SC injection of DNPs may address an unmet need in ocular gene delivery
  • Non-immunogenic, potential for repeat dosing

• Efficacy
  • Higher doses may be used to enhance transfection
  • DNPs can transfer large genes which may allow for gene therapy in the most common inherited retinal diseases (IRDs) such as Stargardt disease and Usher syndrome

• SC injections of DNPs offer the potential for a safer and efficient delivery method