Background

Duchenne muscular dystrophy (DMD) is a common inherited neuromuscular disorder characterized by progressive muscular degeneration.

Antisense oligonucleotide (ASO)-mediated exon skipping is the most promising way to express internally deleted dystrophin in DMD by correcting the reading frame of dystrophin mRNA.

DS-5141b is an ASO consisting of 2′-O,4′-C-ethylen-bridged nucleic acids (ENA†) and 2′-O-methyl RNA (2′-OMe) that induces dystrophin mRNA exon 45 skipping for the treatment of DMD. ENA® is a registered trademark of Daiichi Sankyo Co., Ltd.

Table 1. Properties of 2′-OMe, Morpholino (PMO) and ENA®

<table>
<thead>
<tr>
<th>Structure</th>
<th>2′-OMe</th>
<th>PMO</th>
<th>ENA®</th>
</tr>
</thead>
<tbody>
<tr>
<td>Charge</td>
<td>Negative</td>
<td>Neutral</td>
<td>Negative</td>
</tr>
<tr>
<td>Protein binding</td>
<td>High</td>
<td>Low</td>
<td>High</td>
</tr>
<tr>
<td>Permeability</td>
<td>Low</td>
<td>Very low</td>
<td>Low</td>
</tr>
<tr>
<td>Elimination from tissues</td>
<td>Slow</td>
<td>Fast</td>
<td>Slow</td>
</tr>
<tr>
<td>Affinity towards mRNA (ΔTm/modification)</td>
<td>+0.2 - 1.4°C*</td>
<td>+0.8°C*</td>
<td>+3.5 - 5.2°C**</td>
</tr>
<tr>
<td>Nuclease resistance</td>
<td>High</td>
<td>Very high</td>
<td>Very high</td>
</tr>
</tbody>
</table>

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Potent Exon 45 Skipping Activity of DS-5141b Compared with 2′-OMe ASO and PMO ASO in DMD Model Mice

Figure 1. Exon 45 skipping profiles of DS-5141b, 2′-OMe ASO and PMO ASO in mdx mice after weekly administration for 4 weeks

- DS-5141b showed clear exon 45 skipping in all tissues examined, including the heart, at doses over 3 mg/kg.
- PMO ASO did not induce any exon 45 skipping at doses up to 30 mg/kg; and 2′-OMe ASO had very slight exon skipping activity at a dose of 30 mg/kg.

Induction of Exon 45 Skipping by DS-5141b in Normal Mice

Figure 2. Exon 45 skipping profiles of DS-5141b in mdx mice with normal mice after a single s.c. administration

- At a dose of 30 mg/kg, DS-5141b showed clear exon 45 skipping in all tissues examined, including the heart and even in normal mice, after only a single administration.

Distribution of DS-5141b to Heart, Diaphragm and Skeletal Muscles in Normal Mice

Figure 3. Autoradiograms after single s.c. administration of [3H]DS-5141b (15 mg/kg)

- [3H]DS-5141b was distributed throughout the tissues of mice, with a long retention time in the heart, diaphragm and skeletal muscle.

Summary and Conclusion

- DS-5141b showed clear exon 45 skipping in all tissues examined (anterior tibial muscle, diaphragm, and heart) in mdx mice compared with 2′-OMe ASO and PMO ASO. Notably, exon skipping was detected in heart muscle at a low dose, suggesting cardiac improvement.
- In normal mice, DS-5141b induced exon skipping in all muscle examined, including the heart, which suggests potential benefits to uninjured tissues.
- Autoradiography study in mice revealed that DS-5141b is distributed not only in skeletal muscles, but also in the heart and diaphragm.
- DS-5141b is currently undergoing a Ph1/2 clinical trial (NCT02667483) in Japan. (Will be introduced in pre-record video and live panel presentation on Restoring Dystrophin at the Virtual Annual Conference on July 23rd.)