General overview of novel experimental therapeutic approaches in SMA

SMA stakeholder workshop

Organized by SMA Europe, TREAT-NMD and EMA

Francesco Muntoni

Dubowitz Neuromuscular Centre
UCL Institute of Child Health
& Great Ormond Street Hospital
London, UK

MRC Centre for Neuromuscular Diseases

NHS National Institute for Health Research

Francesco Muntoni: disclosures

Spinal Muscular Atrophy

- PI of Trophos SMA study (2013-14)
- PI of ISIS / Biogen antisense study (2015-16) (2 Biogen SAB meetings)
- PI of Roche drug trial (2015-16) (1 SAB in 2015)
- Avexis AAV Gene therapy (SAB 2015-2016)

Duchenne

- CI of three antisense trials with Sarepta Therapeutics (1 SAB meeting)
- PI of three Prosensa sponsored studies
- PI of two PTC124 sponsored trials (2 SAB meetings)
- CI of Summit Phase I and II studies on utrophin upregulation (1 SAB meeting)

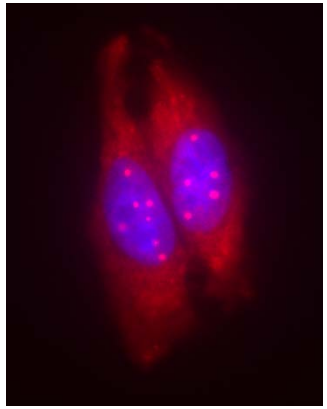
Others

- Member of Pfizer Rare Disease SAB since 2014

Topic discussed

- SMN protein localisation
- Role of SMN protein
- Translational research
  - increasing SMN protein levels
  - Dealing with secondary consequences

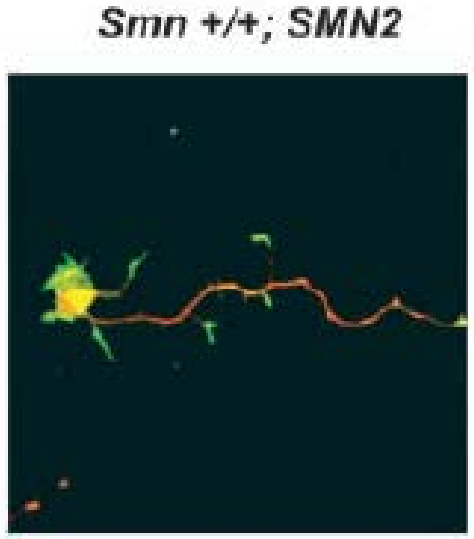
SMN protein



- Ubiquitously expressed protein that localizes to both the cytoplasm and the nucleus where it accumulates in structures known as Gems

- Gems: nuclear domains implicated in the assembly and modification of RNPs → SMN involvement in RNA regulation

- Also found in association with cytoskeletal elements in spinal dendrites and axons



The Journal of Cell Biology | Volume 163, Number 4, 2003

SMN plays multiple key roles in the post-transcriptional regulation of gene expression

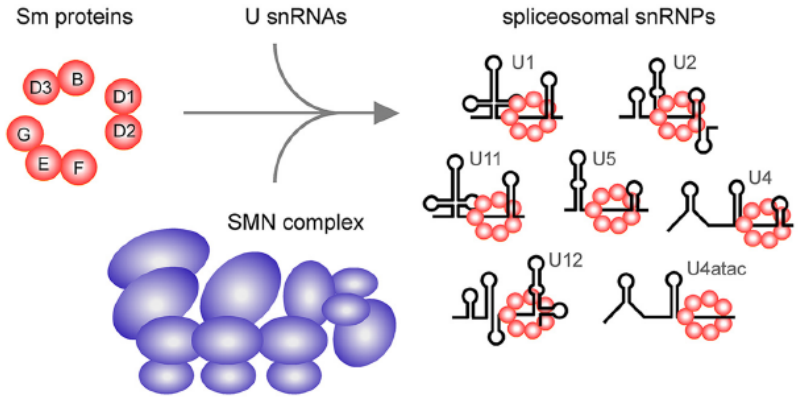
- Involved in:

  A. assembly of snRNPs spliceosomes

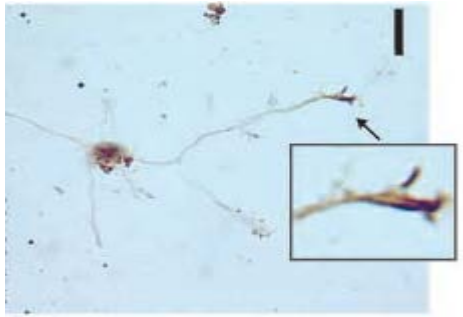
  B. unique 3' end processing of replication dependent histone mRNAs

  C. axonal transport and local translation of mRNAs at the distal end of developing neurons

A



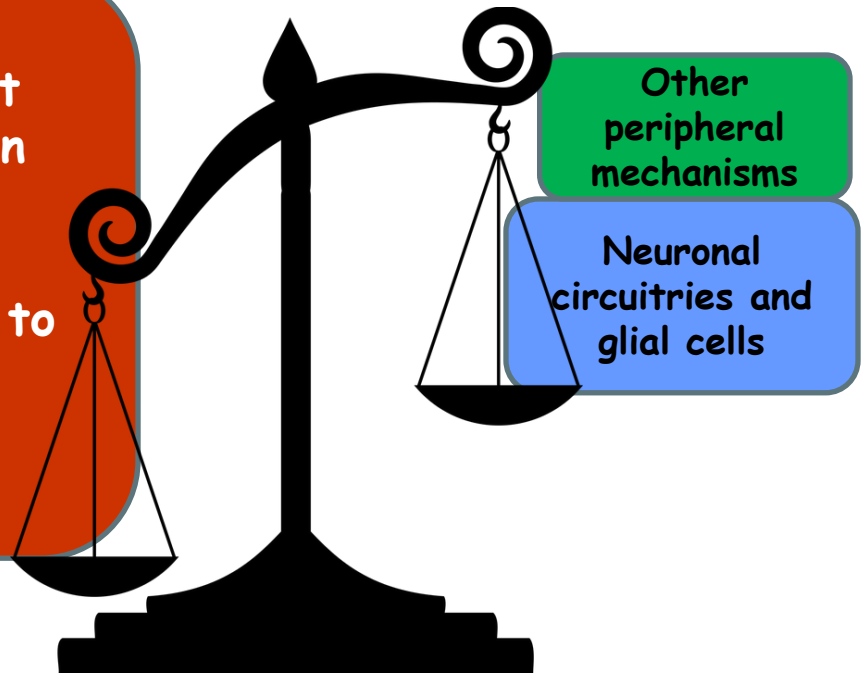
C



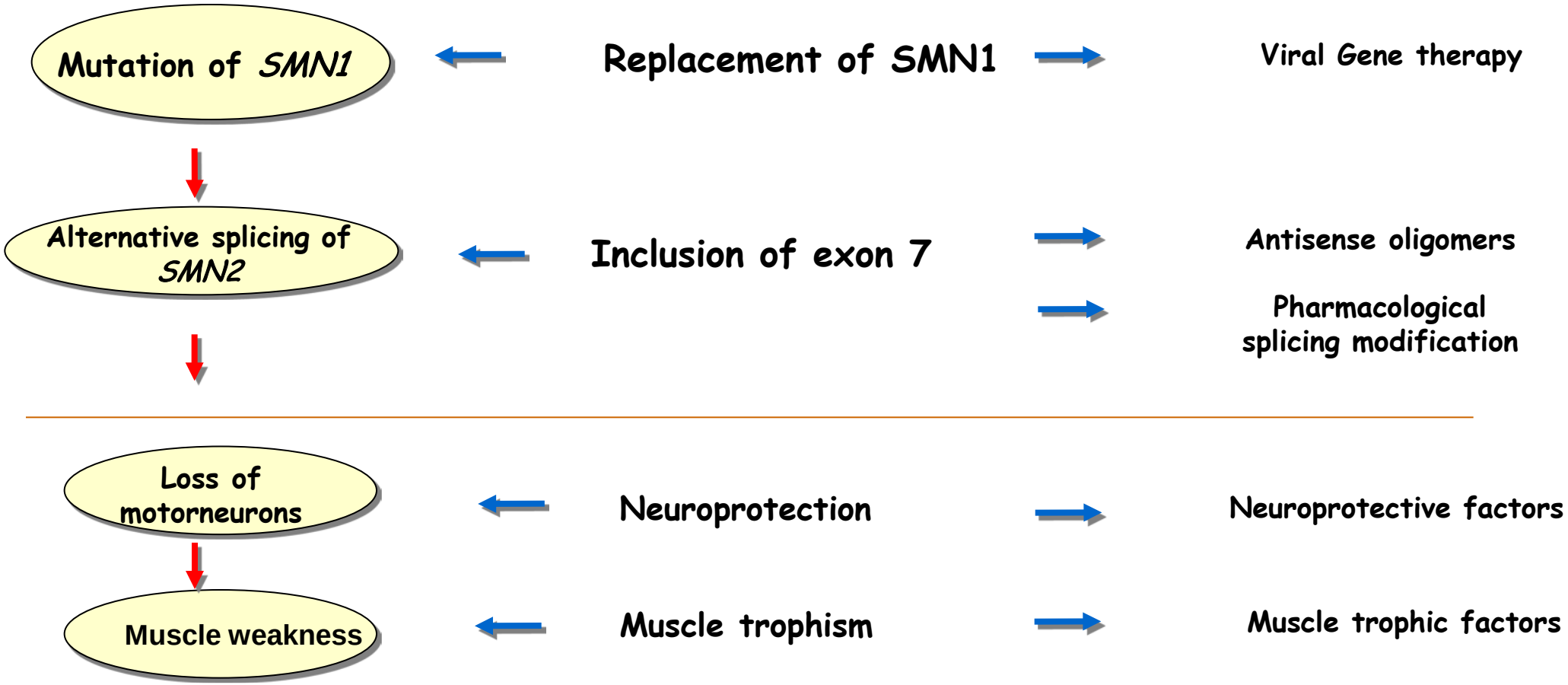
What determines the motor neuron pathology?

- Motor neuron death → cell autonomous event
- Motor neuron SMN replacement necessary in SMN protein upregulation therapies

- SMN deficiency elsewhere might contribute to motor neuron pathology, but the extent of this is not clear in the human



Therapeutic targets for SMA



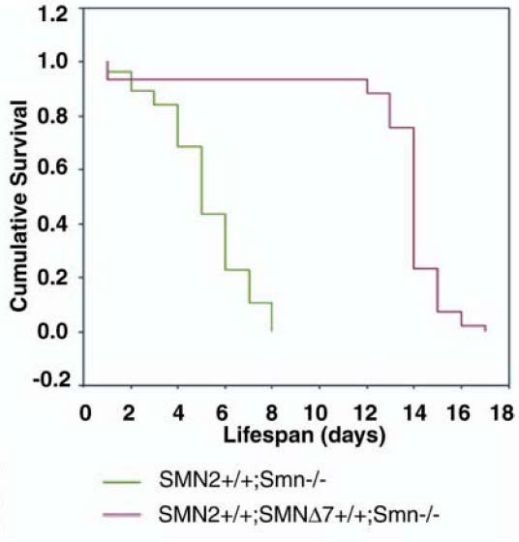
There are 2 commonly used human SMN2 transgenic SMA mouse models

- SMN2+/+; SMNΔ7; Smn−/−, referred to as SMNΔ7 SMA mice (Le et al, HMG 2005)
- (SMN2)2 +/−; Smn−/−, referred to as SMN2 mice or Taiwanese SMA mice (Hsieh-Li et al, Nature, 2000)

Both models have a short lifespan of ~ 15 or 10 days.

Very helpful for the preclinical developments

Narrow therapeutic window, with maximal benefit when treatment administered pre-symptomatically



Increasing SMN protein levels

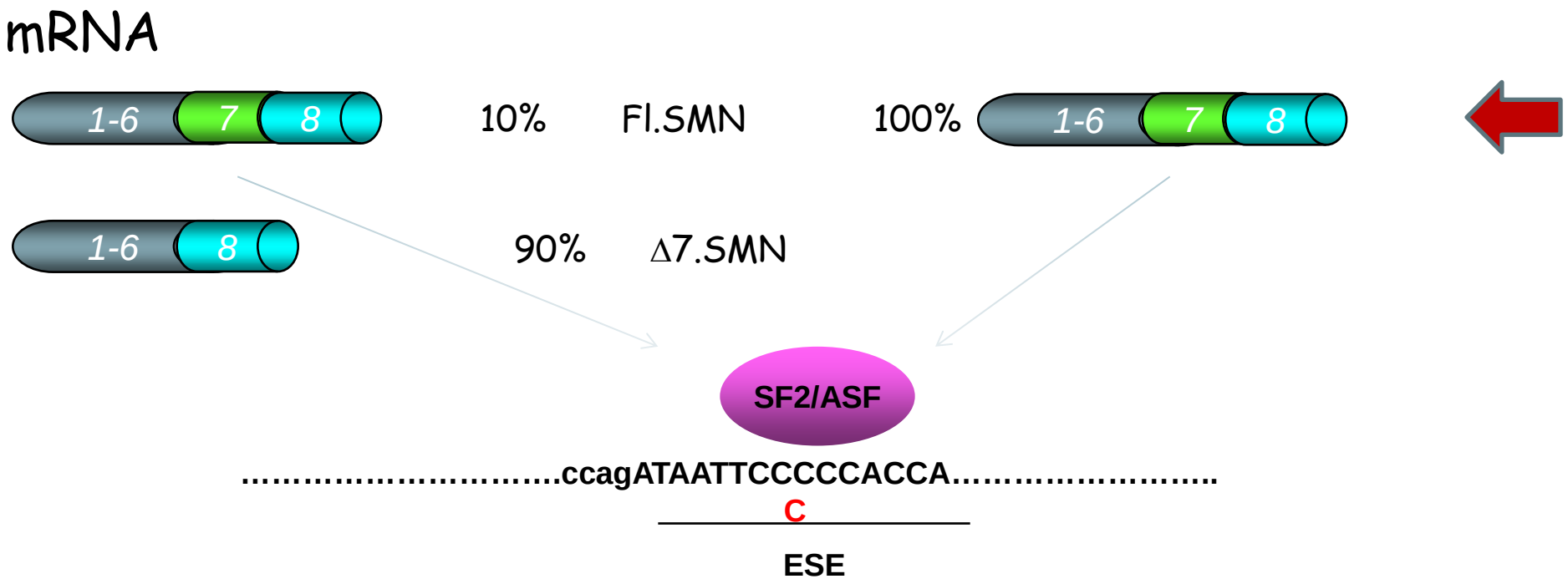
a. Alter SMN2 splicing

b. Replace SMN1

Small molecules

Antisense oligonucleotides

Viral gene therapy



Antisense oligomers for splice switching

- Different backbone chemistries
- The one in current clinical trials is 2'O(2-methoxyethyl) modified AO drug designed to target an hnRNP-A1/A2-dependent splicing silencer, ISS-N1
- AOs do not cross the blood brain barrier
- Need to be administered via repeated lumbar punctures in SMA patients
- Long half life once in the CNS ($\geq$ 6 months)

Nusinersen Clinical Program Overview

Study Number	Study Name Logo	Protocol Title	Population Characteristics	SMN2 Copy #	Study Design & Treatment Period
ISIS 396443-CS3b	ENDEAR	A Phase 3, Randomized, Double-blind, Sham-Procedure Controlled Study to Assess the Clinical Efficacy and Safety of ISIS 396443 Administered Intrathecally in Patients with Infantile-onset Spinal Muscular Atrophy	• Symptomatic • ≤7 months of age at time of screening • Onset of signs and symptoms ≤6 months	2	• Global • Randomized, double-blind, sham-procedure controlled • 13 months
ISIS 396443-CS4	CHERISH	A Phase 3, Randomized, Double-blind, Sham-Procedure Controlled Study to Assess the Clinical Efficacy and Safety of ISIS 396443 Administered Intrathecally in Patients with Later-onset Spinal Muscular Atrophy	• Symptomatic • Age 2-12 years at time of screening • Onset of signs and symptoms >6 months • Can sit, but has never walked	Any	• Global • Randomized, double-blind, sham-procedure controlled • 15 months
2325M201	NURTURE	A Phase 2, Open-Label Study to Assess the Efficacy, Safety, Tolerability and Pharmacokinetics of Multiple Doses of ISIS 396443 Delivered Intrathecally to Subjects with Genetically Diagnosed and Presymptomatic Spinal Muscular Atrophy	• Presymptomatic • ≤6 weeks of age at time of first dose	2 or 3	• Global • Open label • 2 years
2325M202	EMBRACE	A phase 2, randomized, double-blind, sham-procedure controlled study to assess the safety and tolerability and explore the efficacy of ISIS 396443 administered intrathecally in subjects with spinal muscular atrophy who are not eligible to participate in the clinical studies ISIS 396443-CS3B or ISIS 396443-CS4	• Symptomatic • Symptom onset > 6 months *(age 6 -28 months)* • Symptom onset ≤ 6 months *(2 copies: age >7 months; 3 copies: no upper age limit)*	2 or 3	• US, UK, & Germany • Randomized, double-blind, sham-procedure controlled • 12 months
ISIS 396443-CS11	SHINE	An Open-label Extension study for patients with Spinal Muscular Atrophy who previously participated in Investigational Studies of ISIS 396443	• Previously participated in ENDEAR and CHERISH	Any	• Global • Open Label with Sham control blinded loading period (lead in) • 2 years

Ionis/ Biogen sponsored clinical trials

Small molecules to modify splicing of SMN2

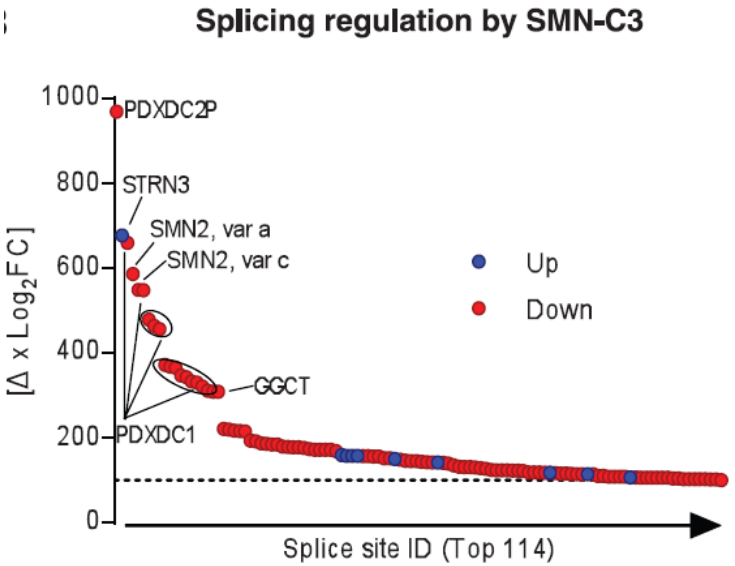
RESEARCH | REPORTS

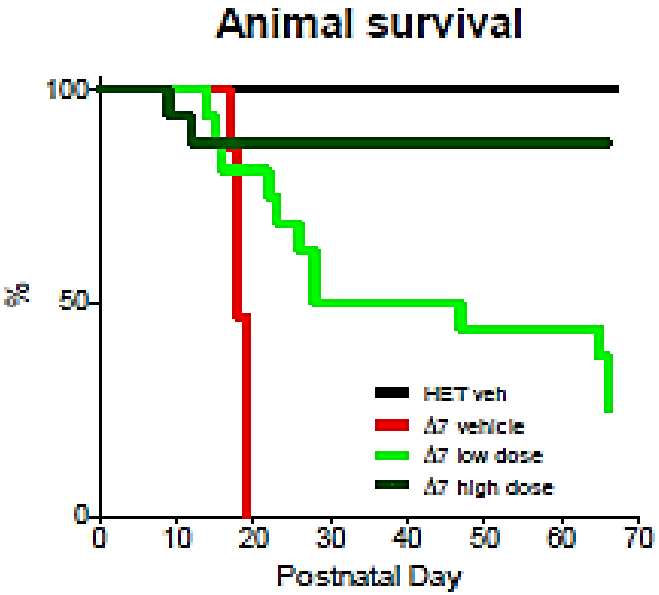
MOTOR NEURON DISEASE

SMN2 splicing modifiers improve motor function and longevity in mice with spinal muscular atrophy

Nikolai A. Naryshkin,[1] Marla Weetall,[1] Amal Dakka,[1] Jana Narasimhan,[1] Xin Zhao,[1] Zhihua Feng,[2] Karen K. Y. Ling,[2] Gary M. Karp,[1] Hongyan Qi,[1] Matthew G. Woll,[1] Guangming Chen,[1] Nanjing Zhang,[1] Vijayalakshmi Gabbeta,[1] Priya Vazirani,[1] Anuradha Bhattacharyya,[1] Bansri Furia,[1] Nicole Risher,[1] Josephine Sheedy,[1] Young-Choon Moon,[1] Panayiota Trifillis,[1] Ellen M. Welch,[1] Joseph M. Colacino,[1] John Babiak,[1] Neil G. Almstead,[1] Stuart W. Peltz,[1]* Loren A. Eng,[2] Karen S. Chen,[2] Jesse L. Mull,[4] Maureen S. Lynes,[4] Lee L. Rubin,[4] Paulo Fontoura,[2] Luca Santarelli,[5] Daniel Haehnke,[5] Kathleen D. McCarthy,[5] Roland Schmucki,[5] Martin Ebeling,[5] Manaswini Sivaramakrishnan,[5] Chien-Ping Ko,[2] Sergey V. Paushkin,[5] Hasane Ratni,[5] Irene Gerlach,[5] Anirvan Ghosh,[5] Friedrich Metzger[5]*

8 AUGUST 2014 • VOL 345 ISSUE 6197 [illegible] SCIENCE





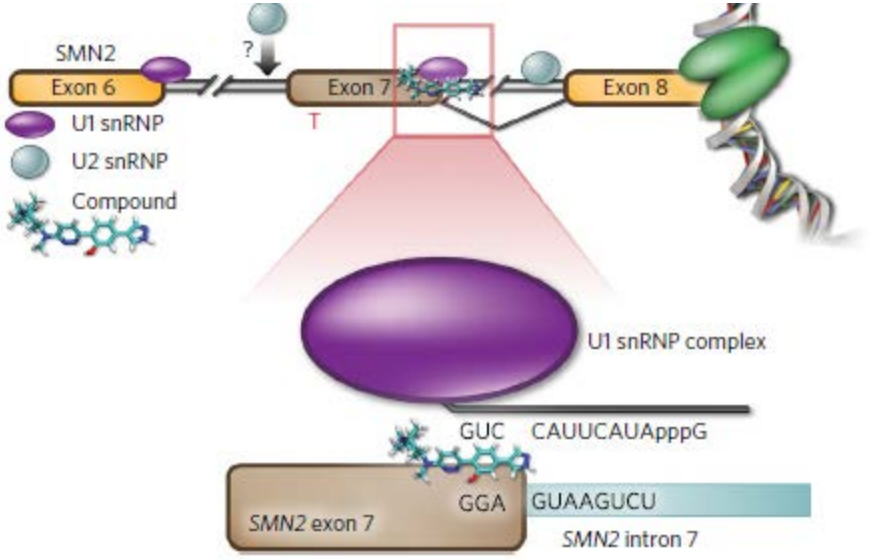
Roche sponsored clinical trial

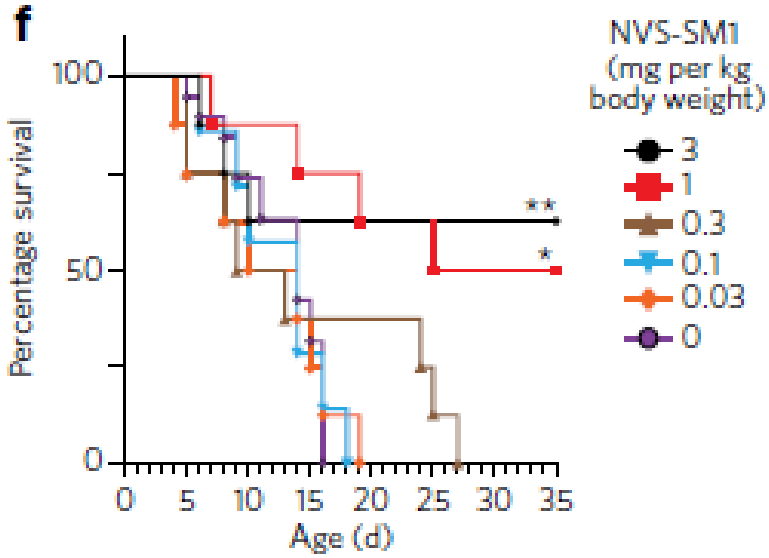
nature chemical biology
ARTICLE
PUBLISHED ONLINE: 1 JUNE 2015 | DOI: 10.1038/NCHEMBIO.1837

SMN2 splice modulators enhance U1–pre-mRNA association and rescue SMA mice

James Palacino[1,3], Susanne E Swalley[1,3]*, Cheng Song[1,3], Atwood K Cheung[1], Lei Shu[1], Xiaolu Zhang[1], Mailin Van Hoosear[1], Youngah Shin[1], Donovan N Chin[1], Caroline Gubser Keller[2], Martin Beibel[2], Nicole A Renaud[1], Thomas M Smith[1], Michael Salcius[1], Xiaoying Shi[1], Marc Hild[1], Rebecca Servais[1], Monish Jain[1], Lin Deng[1], Caroline Bullock[1], Michael McLellan[1], Sven Schuierer[2], Leo Murphy[1], Marcel J J Blommers[2], Cecile Blaustein[1], Frada Berenshteyn[1], Arnaud Lacoste[1], Jason R Thomas[1], Guglielmo Roma[2], Gregory A Michaud[1], Brian S Tseng[1], Jeffery A Porter[1], Vic E Myer[1], John A Tallarico[1], Lawrence G Hamann[1], Daniel Curtis[1], Mark C Fishman[1], William F Dietrich[1], Natalie A Dales[1] & Rajeev Sivasankaran[1]*

Small molecules to modify splicing of SMN2





Novartis sponsored clinical trial

Gene therapy:
scAAV9 improves survival in SMA mice

Human Molecular Genetics, 2011, Vol. 20, No. 4 **681–693**
doi:10.1093/hmg/ddq514
Advance Access published on November 30, 2010

Intravenous scAAV9 delivery of a codon-optimized *SMN1* sequence rescues SMA mice

Elisa Dominguez[1,†], Thibaut Marais[1,†], Nicolas Chatauret[1,2], Sofia Benkhelifa-Ziyyat[1], Sandra Duque[1], Philippe Ravassard[3,4,5], Romain Carcenac[1], Stéphanie Astord[1], Aurélie Pereira de Moura[1], Thomas Voit[1] and Martine Barkats[1,*]

Intramuscular scAAV9-SMN Injection Mediates Widespread Gene Delivery to the Spinal Cord and Decreases Disease Severity in SMA Mice

Sofia Benkhelifa-Ziyyat[1], Aurore Besse[1], Marianne Roda[1], Sandra Duque[1], Stéphanie Astord[1], Romain Carcenac[1], Thibaut Marais[1] and Martine Barkats[1]

Improving Single Injection CSF Delivery of AAV9-mediated Gene Therapy for SMA: A Dose–response Study in Mice and Nonhuman Primates

Kathrin Meyer[1], Laura Ferraiuolo[1], Leah Schmelzer[1], Lyndsey Braun[1], Vicki McGovern[2], Shibi Likhite[1,3], Olivia Michels[1], Alessandra Govoni[1], Julie Fitzgerald[5], Pablo Morales[4], Kevin D Foust[5], Jerry R Mendell[1,3,5], Arthur HM Burghes[2] and Brian K Kaspar[1,3,5]

Molecular Therapy vol. 23 no. 3, 477–487 mar. 2015

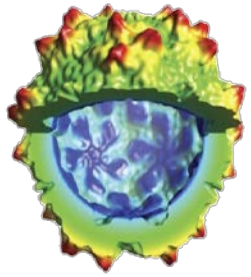
scAAV9, self-complementary adeno-associated virus serotype 9.

AAV9 for SMA gene therapy

Jerry Mendell (Columbus, Ohio) is pursuing AAV gene therapy in infants with SMA I

Single IV injection

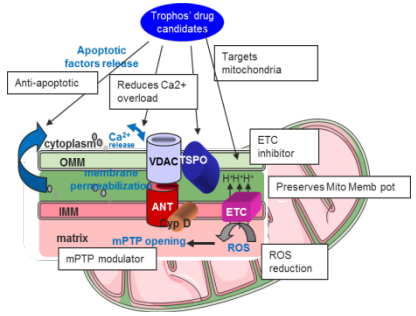
Two doses used:
A. Cohort 1: 6.7×10^{13} vg/kg
B. Cohort 2: 2.0×10^{14} vg/kg

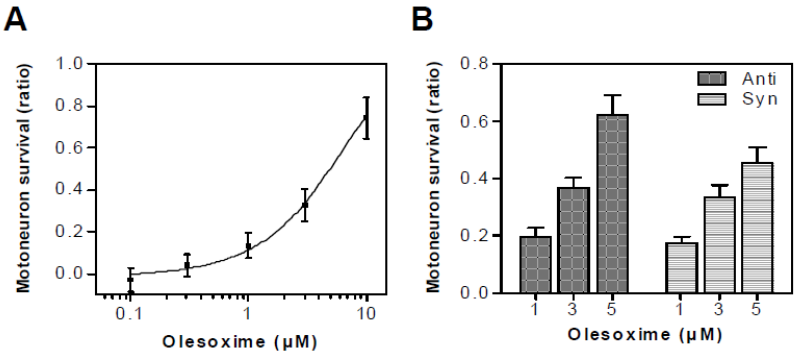


2 SMN2 copy numbers

Avexis sponsored clinical trial

Dealing with secondary consequences of SMN deficiency: mitochondrial dysfunction





Cholesterol-oximes → survival-promoting activity on motor neurons deprived of neurotrophic factors.

Prevents permeability transition pore opening mediated by oxidative stress

Randomized, double-blind, placebo-controlled, Phase 2 study was performed on 165 patients aged 3–25 years with Type 2 or non-ambulatory Type 3 SMA

Additional studies underway

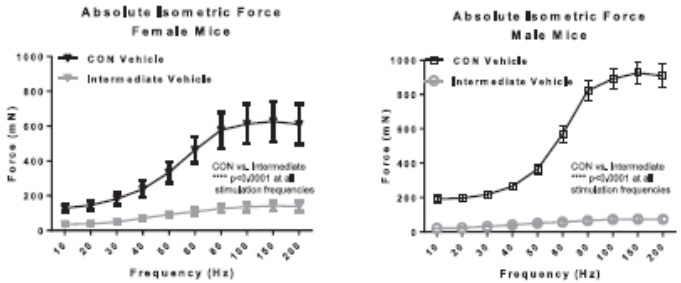
Roche sponsored clinical trial

Fast Skeletal Muscle Troponin Activator *tirasemtiv* Increases Muscle Function Performance in Mouse Models of Spinal Muscular Atrophy

Darren T. Hwee, Ph.D.[1], Leo Kim, B.S.[1], Emily Reedich, B.S.[2], W. David Arnold, M.D.[3], Nancy L. Kuntz, M.D.[2], Christine J. DiDonato, Ph.D.[2], Fady I. Malik, M.D., Ph.D.[1] and Jeffrey R. Jasper, Ph.D.[1]
[1]Cytokinetics, Inc., South San Francisco, CA, [2]Lurie Children's Hospital of Chicago, Chicago, IL., [3]Department of Neurology and Physical Medicine, The Ohio State University, Columbus, OH
DISCLOSURE OF INTERESTS: DTH, LK, FIM, and JRJ are currently employees of **Cytokinetics, Inc.** and were compensated financially for their work.

2016 Annual SMA Conference. Orlando, FL, June 2016.

Tirasemtiv : specific fast skeletal muscle troponin activator
Sensitizes the sarcomere to calcium
Increased muscle force *in situ* in response to submaximal rates of nerve stimulation



Clinical trial
A Phase 2, Double-Blind, Randomized, Placebo-Controlled, Multiple Dose Study of CK-2127107 in Two Ascending Dose Cohorts of Patients With Spinal Muscular Atrophy (SMA), currently recruiting

Cytokinetic sponsored clinical trial

Conclusion

- Different therapeutic strategies focus on increasing SMN protein levels
- Multiple studies on infants with type I SMA (and 2 *SMN2* copy number) underway
- Uncharacteristic functional improvement and acquisition of novel gross milestones such as sitting, standing and, in some patients, taking a few steps, reported.
- This might suggest that there is a window for therapeutic response also for SMA I infants
- Studies focused on different downstream targets underway