Degenerative Neurological and Neuromuscular Disease

Current and emerging treatment options for spinal muscular atrophy

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Abstract: Spinal muscular atrophy is one of the most common inherited neuromuscular conditions; our understanding of the genetic pathology and translational research coming from this insight has made significant progress over the past decade. This short review provides the background of the disease along with the bench to bedside progress of some promising treatment options to develop better understanding of the present state of the disease.

Keywords: SMN protein, neurodegenerative disease, orphan disease, therapeutics

Introduction

Spinal muscle atrophy (SMA) is an autosomal recessive neurodegenerative disease and a leading global genetic cause of infant death.1 It is characterized by the loss of α motor neurons from the anterior horn of the spinal cord resulting in muscle weakness, trunk paralysis, and muscle atrophy.2 With an estimated prevalence of 1 in 6,000–11,000 live births and a carrier frequency of 1 in 40 to 1 in 60 among different ethnic groups and geographical location,3–7 SMA is the most common monogenic disease fatal to infants and one of the most common forms of neuromuscular disorder in childhood.

SMA is classified into three major groups based on the age of onset and severity of the disease;8–11 Type I SMA (Werdnig–Hoffmann disease) is the most prevalent and severe form of the disease with postnatal onset within the first six months. These patients are never able to sit and usually succumb within 2–5 years of age due to respiratory failure.11–13 Patients with the intermediary form or type II SMA develop muscle weakness within 6–18 months of age. Although these patients can sit, due to progressive muscle weakness, they can never stand or walk. Type III SMA (Kugelberg–Welander disease) has an onset between 18 months and 30 years of age with patients able to walk on their own (with some assistance).11,12,14,15 The diagnosis of SMA is typically established through physical examination, patient history, electromyography followed by confirmatory genetic testing.3 Muscle biopsy may also be done in some cases16 although less frequently since the advent of genetic testing.

The lack of functional survival motor neuron (SMN) protein due to deletion or mutation in the SMN1 gene is the cause of SMA.17 SMN is an evolutionary conserved RNA-associated protein required for cellular viability; complete loss of functional full-length SMN protein is therefore embryonically lethal.18–20 Humans due to an evolutionary recent duplication event at chromosome 5q17,21 possess a nearly identical SMN paralog, SMN2 (Figure 1) and thus uniquely among all species can survive loss of SMN1.

Although, SMN2 has only few translationally insignificant nucleotide differences compared with SMN1, the C to T transition at position 6 of exon 7 results in the
production of an alternative splice variant. Consequently, 
*SMN2* produces only \( \sim 10\% \) of the full-length functional 
*SMN* protein produced by *SMN1*; the remaining mRNA 
lacks exon 7 and is translated into a truncated, unstable non-
functional protein called SMN\( \Delta 7 \) which, failing to oligomer-
ize, is quickly degraded (Figure 2).\(^{22,23}\)

Greater than 95% of all SMA patients have homozygous 
deletions of *SMN1* gene.\(^{17}\) All SMA patients have at least 
one copy of *SMN2*, which produces low levels of functional 
*SMN* protein and acts as a disease modifier. There is an 
inverse correlation between *SMN2* gene copy number and 
disease severity, ie, an increase in the *SMN2* gene copy 
number decreases the SMA severity.\(^{24,25}\) Typically, SMA 
type I patients have one or two copies of *SMN2* gene com-
pared with two to three copies in type II, and three to four 
copies in types III and IV. Individuals with more than four 
copies of the *SMN2* gene are completely asymptomatic not-
withstanding the deletion of *SMN1*.

A central function of 294 amino acid long SMN protein is 
the assembly of small nuclear ribonucleic proteins (snRNPs) 
which are essential for splicing.\(^{26–35}\) With mutations in the 
ubiquitously expressed SMN shown to cause SMA, we are 
left with a question posed in this disease gene cloning era; 
how do mutations in a gene which is expressed everywhere 
(ie, SMN) mainly impact neurons (despite several recent 
studies suggesting a role for SMN in other tissue types as well\(^{36–44}\)). Although the precise pathogenic molecular 
mechanism of SMA is not known, it is believed that the lack 
of motor neuron SMN protein may lead to a synaptopathy 
resulting in apoptotic death of motor neurons.\(^{20,45}\)

Presently, only multidisciplinary supportive care including, 
critically, respiratory support is available for most 
children with SMA.\(^{10}\) Although these interventions have 
improved both life expectancy and quality of the life, an 
effective therapy for SMA is eagerly awaited by patients, 
their families, researchers, clinicians, and support care staff 
alike. Several approaches are currently being pursued as the 
following section outlines.

**Therapeutic strategies for SMA**

**Gene therapy**

Gene therapy is one of the most promising therapeutic 
advances for SMA. In the past 6 years, several groups 
have used self-complementary adeno-associated virus 
serotypes 8 and 9 carrying human *SMN1* cDNA to treat 
mouse models of SMA. The most encouraging results (ame-
lioration of disease phenotype and dramatic extension in the 
life span) were observed with pre-symptomatic treatment of 
SMA mice.\(^{46–49}\)

These promising results helped scientists at Nationwide 
Children’s Hospital to receive a fast track designation from the 
US Food and Drug Administration (FDA) for ChariSMA\(^{TM}\) 
(gene therapy product) to be tested as a SMA therapeutic. 
At the time of writing, a phase I clinical trial at Nationwide 
Children’s Hospital in collaboration with AveXis Inc. and 
The Sophia’s Cure Foundation is underway to evaluate safety

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**Figure 1** Human SMN locus on chromosome 5q showing inverted duplication of 
*SMN1* and *NAIP* genes. 
**Abbreviations**: NAIP, neuronal apoptosis inhibitory protein; SMN, survival motor 
neuron.

**Figure 2** Schematic diagram showing *SMN1* (A) and *SMN2* (B) genes along with their corresponding mRNA and protein products. 
**Abbreviation**: SMN, survival motor neuron.
and efficacy of gene transfer (systemic AAV9-delivered
SMN1 gene) in SMA type I patients. The major challenges
to bring this treatment into clinics are clinical safety, the cost
of virus and the possibility of an immune response neutral-
izing AAV.50

**SMN2-dependent therapies**

Given the role of SMN2 as a modifier gene for SMA, the
inverse correlation between the SMN2 gene copy number
and disease severity, the gene itself has become a drug
target. Strategies includes a) inducing the expression of
SMN2, b) modulating splicing of SMN2-derived transcript,
and c) stabilizing the full-length SMN2-derived mRNA and/
or protein (Figure 3).51,52

**Activation of SMN2 promoter**

Histone deacetylases (HDACs) through chromatin condensa-
tion are known to repress transcription of genes such as
SMN2. Several HDAC inhibitors including sodium butyrate,
valproic acid (VPA), and phenyl butyrate have been assessed
for SMN2 induction in cellular and animal models of the
disease as well as in clinical trials as potential therapeutic for
SMA.53-57 Although they showed promise in cell culture and
SMA mouse models, no significant clinical improvement has
been observed in SMA patients with HDAC inhibitors.2,58,59

The STAT5 pathway has been implicated in the activa-
tion of SMN2 promoter.60-62 The human lactation hormone
prolactin (PRL) and human growth hormone (HGH) have
been shown to activate the STAT5 pathway which results in
an increase in both SMN2 gene transcription and full-length
SMN protein both in vitro and in vivo, resulting in attenuation
of the SMA mouse model severity. PRL treatment resulted in
significant survival and attenuation of disease phenotype in
the SMA mouse model, a possible reflection of the signifi-
cant SMN induction observed with PRL treatment in vivo.61

Although, PRL has been proven safe and was successfully
tested in humans for the treatment of lactation-deficient
mothers,63 the absence of clinical grade PRL is delaying its
further assessment as a potential SMA therapeutic in the
patient population. A Phase II trial in SMA type II/III patients
showed no improvement in muscle function or strength after
3 months of treatment with HGH.64 However, it should be
noted that only a single low dose of HGH was used and
peripheral white blood cell SMN levels were not assessed
in the patient population. Before crossing HGH off the list
of SMA therapeutics, it may be beneficial to do a HGH dose
escalation study especially in younger SMA patient popula-
tion assessing muscle strength and monitoring changes in
lymphocyte SMN levels.

The deCODE project initiated by Families of SMA (now
cureSMA) identified C5-substituted quinazoline activity in
increasing SMN2 promoter activity and thus SMN protein
in cell-based assays. The DcpS inhibitor RG3039, a deriva-
tive of quinazolines, has since been tested and reported to
mildly improve both survival and motor function in two
mouse models of SMA through an increase in full-length

![Figure 3](https://www.dovepress.com/)

**Figure 3** Current SMN2 gene-derived therapeutic approaches to SMA.

**Abbreviations:** SMN, survival motor neuron; SMA, spinal muscular atrophy.
SMN levels. The compound was initially out-licensed to Repligen Corporation (Waltham, MA, USA) which in 2012 out-licensed to Pfizer Pharmaceuticals (New York, NY, USA); however, the program has now been halted.

**Correction of splicing**

The significant majority of transcripts arising from SMN2 gene lack exon 7; agents that suppress exon 7 skipping have thus become the goal of a number of laboratories. Several small compounds including HDAC inhibitors such as VPA, TSA, and sodium butyrate have been assessed for their capacity to increase full-length SMN transcript by altering the splicing process in vitro. However, regardless of any putative effect observed in vitro, no beneficial effect has carried over to clinical trials.

Recently, RO6885247 from PTC Therapeutics (South Plainfield, NJ, USA) and Hoffmann-LaRoche (Nutley, NJ, USA) has been shown to profoundly affect the splicing of SMN2 gene to include more exon 7 showing a dramatic positive impact on SMA model mice.

A Phase I clinical trial is currently underway to assess the safety profile along with tolerability of this compound.

Antisense oligos (ASOs) are increasingly being used for therapeutic/experimental purposes to treat a number of diseases including SMA, duchenne muscular dystrophy, and myotonic dystrophy. A bifunctional ASO complementary to SMN2 exon 7 pre-mRNA sequences has been designed and tested to promote the inclusion of exon 7 and at the same time inhibit binding of negative splicing factors which ultimately results into increased production of full-length SMN mRNA.

ASOs are not blood brain barrier (BBB) penetrant, which initially posed a hurdle in their use as a SMA therapeutics. Several reports have shown that intrathecal injections of ASOs results into improvement in survival and disease phenotype in SMA mice. However, in 2011, Hua et al reported an increase in SMN levels along with attenuation of SMA phenotype in SMA mice with systemic delivery of ASO. ISIS pharmaceutical has successfully completed early phase trials to show safety, tolerability, and pharmacokinetics of intrathecal ISIS-SMN Rx (ASOs) in SMA patients. A Phase III has been initiated to test the safety and efficacy of intrathecal administration of ISIS-SMN Rx in patients with infantile-onset and later onset of SMA. This represents a significant breakthrough in the field of SMA as this is the first compound which has reached Phase III clinical trial. The results from these trials are eagerly awaited by the SMA community.

**Full-length SMN transcript and protein stabilization**

In addition to the upregulation of SMN2 transcription and the modulation of splicing, the stabilization of either mRNA or protein is an often overlooked but nonetheless viable potential therapeutic approach. In this regard, SMN mRNA has a specific AU-rich element region in its 3′-untranslated region which marks the mRNA for degradation. In silico mining of gene expression data sets was used to identify the p38 pathway as a means of enhancing SMN2 levels. In the initial study, it was shown that treatment of neuronal cells with the p38 activator anisomycin results in the transllocation of protein HuR to the cytoplasm where it binds the 3′-untranslated region of SMN mRNA and stabilizes the transcript inhibiting its degradation which in turn increases the SMN protein level.

Since then several p38-activating compounds have been identified and reported to increase the SMN protein level in vitro as well as in vivo.

Celecoxib, a safe, well-tolerated prescription medication used widely for arthritis and in some pediatric rheumatologic diseases, was identified as a BBB penetrant, p38-activating compound. Treatment with low dose of celecoxib increased SMN protein levels in both human and mouse nerve cell cultures, as well as in patient fibroblasts. In a severe SMA mouse model, mice treated with celecoxib showed an increase in the SMN protein in central nervous system (CNS) tissues. In addition, SMA mice also showed an improved motor function and a statistically significant 40% extension of survival as compared to mice treated with placebo. A Phase I/II clinical trial is planned for this FDA approved agent in 2015; it may be with the hope that celecoxib may serve as an adjunctive therapy for SMA, particularly given the low safe doses are required for SMN induction.

Aminoglycoside antibiotics, such as tobramycin and amikacin which have been shown to mask premature stop codon mutations in some genes, have been used to increase SMN protein levels in patient fibroblasts. However, there efficacy and safety has yet to be tested successfully in animal models of the disease. An alternative potential therapeutic approach involves targeting the ubiquitin–proteasome pathway which targets many proteins including SMN for degradation. The FDA-approved proteasome inhibitor bortezomib has been shown to increase SMN levels both in vitro and in vivo. However, the major obstacle to the use of bortezomib as a therapeutic is its inability to cross the BBB.

Finally, through the experience gained from treatments in animal models of the disease, it is the general consensus that, as with most disorders, early timing of the treatment is...
critical for maximum benefit; optimally, this would involve pre-symptomatic identification of infants with SMA. Newborn screening is therefore an important step in the most effective use of novel therapies of SMA allowing intervention before the clinical course is set.51,84,85 Hopefully through early intervention along with the promising therapeutic candidates described in Table 1, there will be an effective therapy and possibly a cure for SMA in the near future.

**Disclosure**

The authors report no conflicts of interest in this work.

**References**


**Table 1 Current therapeutic pipeline for SMA**

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<th>Therapeutic approach</th>
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<td>Phase I clinical trial</td>
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**Abbreviations:** ASO, Antisense oligos; SMA, spinal muscle atrophy.


Emerging treatment options for SMA


