Taylor & Francis
Taylor & Francis Group

Expert Opinion on Investigational Drugs

ISSN: (Print) (Online) Journal homepage: https://www.tandfonline.com/loi/ieid20

Risdiplam: an investigational survival motor neuron 2 (SMN2) splicing modifier for spinal muscular atrophy (SMA)

Theodora Markati, Gemma Fisher, Sithara Ramdas & Laurent Servais

To cite this article: Theodora Markati, Gemma Fisher, Sithara Ramdas & Laurent Servais (2022) Risdiplam: an investigational survival motor neuron 2 (SMN2) splicing modifier for spinal muscular atrophy (SMA), Expert Opinion on Investigational Drugs, 31:5, 451-461, DOI: 10.1080/13543784.2022.2056836

To link to this article: https://doi.org/10.1080/13543784.2022.2056836

EXPERT OPINION ON INVESTIGATIONAL DRUGS
2022, VOL. 31, NO. 5, 451–461
https://doi.org/10.1080/13543784.2022.2056836

Taylor & Francis
Taylor & Francis Group

DRUG EVALUATION

Risdiplam: an investigational *survival motor neuron 2 (SMN2)* splicing modifier for spinal muscular atrophy (SMA)

Theodora Markati [a,b], Gemma Fisher [a,b], Sithara Ramdas [a,b] and Laurent Servais [a,b,c]

[a]MDUK Oxford Neuromuscular Centre, Department of Paediatrics, University of Oxford, Oxford, UK; [b]Department of Paediatric Neurology, Oxford University Hospitals NHS Foundation Trust, Oxford, UK; [c]Division of Child Neurology, Centre de Références des Maladies Neuromusculaires, Department of Pediatrics, University Hospital Liège & University of Liège, Liege, Belgium

ABSTRACT

Introduction: Spinal muscular atrophy (SMA) is a rare autosomal recessive neuromuscular disease which is characterised by muscle atrophy and early death in most patients. Risdiplam is the third overall and first oral drug approved for SMA with disease-modifying potential. Risdiplam acts as a *survival motor neuron 2* (*SMN2*) pre-mRNA splicing modifier with satisfactory safety and efficacy profile. This review aims to critically appraise the place of risdiplam in the map of SMA therapeutics.

Areas covered: This review gives an overview of the current market for SMA and presents the mechanism of action and the pharmacological properties of risdiplam. It also outlines the development of risdiplam from early preclinical stages through to the most recently published results from phase 2/3 clinical trials. Risdiplam has proved its efficacy in pivotal trials for SMA Types 1, 2, and 3 with a satisfactory safety profile.

Expert opinion: In the absence of comparative data with the other two approved drugs, the role of risdiplam in the treatment algorithm of affected individuals is examined in three different patient populations based on the age and diagnosis method (newborn screening or clinical, symptom-driven diagnosis). Long-term data and real-world data will play a fundamental role in its future.

ARTICLE HISTORY
Received 17 December 2021
Accepted 20 March 2022

KEYWORDS
Central nervous system (CNS); neuromuscular disorders; risdiplam; RNA splicing modifier; SMN protein; SMN1 gene; SMN2 gene; spinal muscular atrophy (SMA)

1. Introduction

Spinal muscular atrophy (SMA) is a genetic neuromuscular disease that is characterised by progressive loss of motor neurons leading to progressive muscle weakness, atrophy, and motor and respiratory impairment. The most common form, called 5q-SMA, is caused by a homozygous deletion or loss-of-function mutations in the *survival of motor neuron 1* (*SMN1*) gene on locus 5q13 of chromosome 5, which codes for the homonymous survival motor neuron (SMN) protein [1,2]. 5q-SMA, henceforth called SMA, occurs in 1 in 10,000 live births and it is one of the leading genetic causes of childhood mortality. SMA is characterised by progressive initially proximal and axial muscle weakness, decreased or absent deep tendon reflexes, muscle atrophy, and – in the most severe forms without intervention – bulbar dysfunction and progressive respiratory failure as the cause of early death [3].

A paralogous gene in humans, *SMN2*, produces functional SMN protein, but at low and insufficient levels due to naturally occurring alternative splicing of its exon 7 that leads to a truncated transcript. The levels of SMN produced from *SMN2* can partially compensate for the loss of *SMN1*; therefore, increased *SMN2* copy numbers are associated with less severe clinical phenotypes, even though the correlation is not absolute [4]. SMA is characterised by progressive degeneration of α-motor neurons in the brain stem and spinal cord that leads to muscle atrophy and weakness. In preclinical studies, the need to restore SMN protein levels beyond the central nervous system to fully restore the normal phenotype and the association of the most severe clinical forms of SMA with cardiac and brain malformations suggest that SMA in its most severe forms is more than a motor neuron disease and can affect other cell types [5,6], as described in a recent review [7].

Risdiplam (market label: EVRYSDI™) (Box 1) is a small molecule *SMN2* pre-mRNA splicing modifier that promotes the inclusion of exon 7 and production of full-length *SMN2* mRNA, which can compensate for the loss of *SMN1* (Figure 1) [8]. The development of risdiplam has been led by a consortium comprised the SMA Foundation, a US non-profit organization, PTC Pharmaceuticals, a biotechnology company, and the pharmaceutical company F. Hoffmann La Roche. Risdiplam was granted Orphan Drug Designation by the US Food and Drug Administration (FDA) and the European Medicines Agency (EMA) in 2017 and 2019, respectively. Risdiplam gained approval by the FDA in August 2020, and in March 2021 the European Commission approved it for the treatment of patients affected by SMA who are older than 2 months of age [9]. This review describes the pathway to the clinical development of risdiplam (Figure 2) and critically appraises risdiplam in the map of the current therapeutic landscape for SMA.

CONTACT Laurent Servais laurent.servais@paediatrics.ox.ac.uk MDUK Oxford Neuromuscular Centre, Department of Paediatrics, University of Oxford, Level 2 Children Hospital, John Radcliffe Hospital, Headley Way, Headington, Oxford OX3 9DU, UK

Supplemental data for this article can be accessed here

Article highlights

- Spinal muscular atrophy (SMA) is a rare autosomal recessive neuromuscular disease caused by a homozygous deletion or loss-of-function mutations in the *survival of motor neuron 1* (*SMN1*) gene, which codes for the survival motor neuron protein (SMN).
- A paralogous gene in humans, *SMN2*, produces functional SMN protein, but at low and insufficient levels due to naturally occurring alternative splicing that leads to a truncated transcript.
- Risdiplam is a small molecule that acts as an *SMN2* pre-mRNA splicing modifier; it promotes the inclusion of exon 7.
- Risdiplam is delivered orally and has proved its efficacy in pivotal trials for SMA Types 1, 2, and 3 with a satisfactory safety profile. It was approved for clinical use by the US Food and Drug Administration in 2020.
- Currently, there are four ongoing clinical trials assessing risdiplam in different age groups and SMA types: the FIREFISH (NCT02913482), the SUNFISH (NCT02908685), the JEWELFISH (NCT03032172), and the RAINBOWFISH (NCT03779334) trials.
- Two other drugs (nusinersen and onasemnogene-abeparvovec-xioi), which aim to restore the SMN deficiency in motor neurons have been previously approved; all three approved drugs have different mechanisms and routes of administration.
- In the absence of comparative data between the approved drugs, three different patient populations need to be taken into consideration when trying to identify the role of risdiplam in the treatment algorithm of SMA.

This box summarizes key points contained in the article.

Box 1. Drug summary.

Drug name	Risdiplam (EVRYSDI™)
Indication	Spinal muscular atrophy (SMA)
Pharmacology description/ mechanism of action	*SMN2* pre-mRNA splicing modifier for the inclusion of exon 7
Route of administration	Oral
Chemical structure	$C_{22}H_{23}N_7O$
Clinical trials	FIREFISH (NCT02913482), SUNFISH (NCT02908685), JEWELFISH (NCT03032172), RAINBOWFISH (NCT03779334)

2. Methods

The current article is a scopic review for which two databases were used: PubMed/MEDLINE and Embase. Selected keywords were combined to create search strategies, adjusted for each screened database. Search terms included but were not limited to: 'spinal muscular atrophy,' 'sma,' '5q sma, 'risdiplam,' 'evrysdi,' 'RG7916,' 'RO7034067,' 'rna splicing,' 'survival motor neuron 2 splicing modifier' and 'smn2 splicing modifier.' For a full search strategy please refer to **Supplementary Material**. The search was limited to English language and to the last 10 years. References from relevant articles were searched for inclusion of additional papers which were not identified through the search strategy.

3. Results

3.1. Overview of the market

Prior to the approval of risdiplam, two additional drugs, targeting the root cause of SMA, were approved [10]. One is nusinersen (Spinraza®), an antisense oligonucleotide administered intrathecally that binds to *SMN2* pre-mRNA to modify splicing to increase SMN protein levels [11]. It gained approval in 2016. The other is onasemnogene abeparvovec-xioi (Zolgensma®), a self-complementary adeno-associated virus-based gene therapy that aims to provide a copy of the *SMN* gene to neurons; it is administered intravenously [12]. It was approved in 2019 for patients younger than 2 years [13]. Risdiplam is the third drug approved for the treatment of SMA and it is the only one which is administered systemically, targeting cells beyond motor neurons. A systematic approach by considering the evidence from clinical trials and the real-world evidence for efficacy, the safety profile, the route of administration and other factors is required to identify the role of each different drug in the treatment algorithm of patients based on their age and genotype. Many more candidate therapies are in the pipeline of SMA, including

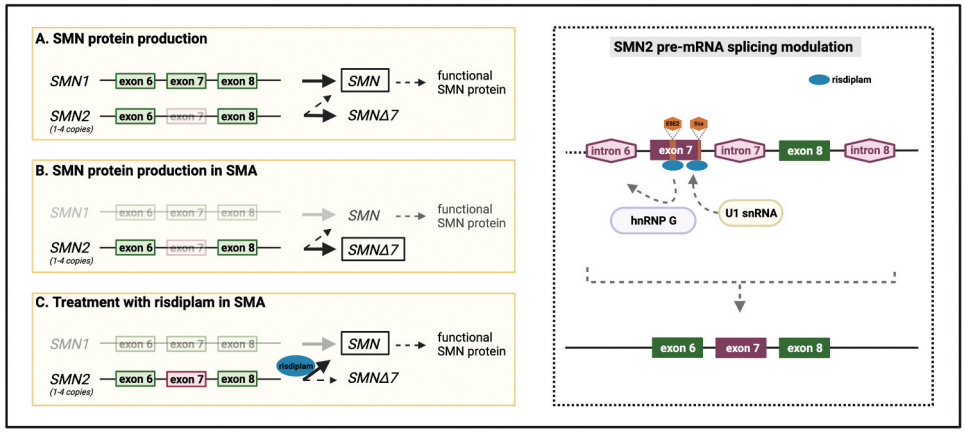

Figure 1. Risdiplam mechanism of action. The most common form, called 5q-SMA, is caused by a homozygous deletion or loss-of-function mutations in the *SMN1* gene on locus 5q13 of chromosome 5, which codes for the homonymous SMN protein A paralogous gene, *SMN2*, produces functional SMN protein, but at low and insufficient levels due to naturally occurring alternative splicing of its exon 7 that leads to atruncated transcript. The levels of SMN produced from *SMN2* can partially compensate for the loss of *SMN1*. Risdiplam is a small molecule, *SMN2* pre-mRNA splicing modifier that promotes the inclusion of exon 7 and production of a full-length *SMN2* mRNA, which can then compensate for the loss of *SMN1*. The working model resulting from studies on risdiplam-like *SMN2* splicing modifiers is that the compound binds on two sites within the exon 7 of the *SMN2* transcript, namely exonic splicing enhancer 2 (ESE2) and 5′ splice site (5′ss). Binding to the 5′ss enhances the binding of the U1 snRNA. The interaction with the ESE2 is believed to lead to dislocation of the hnRNP G allowing the binding of the U1 snRNP complex. These changes ultimately lead to the inclusion of exon 7 and the production of a full-length SMN2 mRNA. Abbreviations: SMA: spinal muscular atrophy, SMN: survival of motor neuron, ESE2: exonic splicing enhancer 2, 5′ss: 5′ splice site.

Created with BioRender under monthly paid subscription.

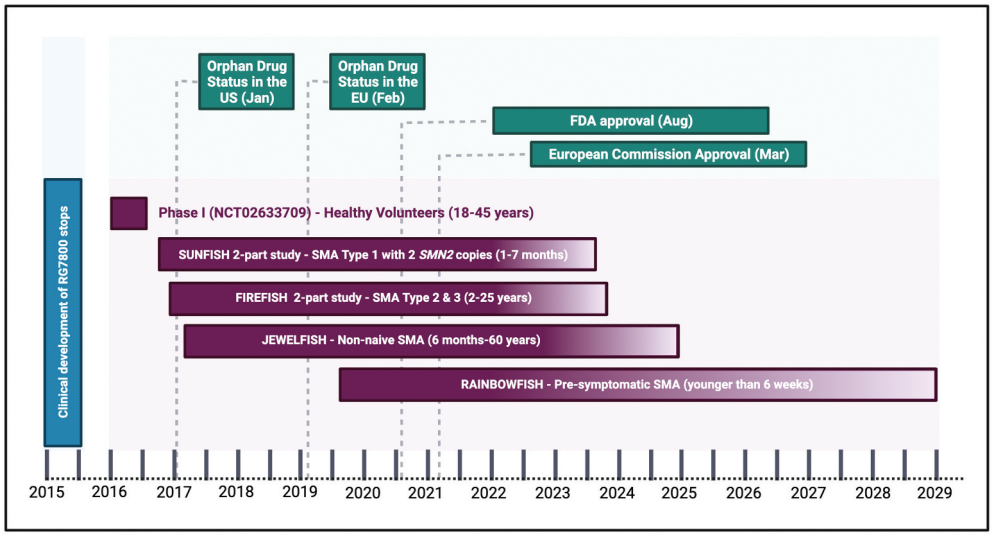

Figure 2. Timeline of risdiplam clinical development and key milestones of approval.
Created with BioRender under monthly paid subscription.

therapies that aim to improve the function of the neuromuscular junction or muscle contraction and size [14]. A combination therapy between a drug that targets the SMN deficiency and a drug that targets another molecular pathway could be expected in this context.

3.2. The pathway to risdiplam identification

The development of risdiplam (RG7916/RO7034067) was preceded by clinical testing of RG7800/RO6885247, the first *SMN2* pre-mRNA splicing modifier to enter clinical development. Three classes of small molecules were identified via a high-throughput screening campaign aiming to identify small molecules from the PTC library. These molecules (coumarines, isocoumarines, and pyrido-pyrimidinone derivatives) were able to induce inclusion of exon 7 during the splicing of the *SMN2* pre-mRNA [15]. The screening was performed using a human embryonic kidney cell line that expressed an *SMN2* gene fragment from exon 6 to the 5′ area of exon 8 followed by the coding sequence of the firefly luciferase. The luciferase coding sequence was in frame only when exon 7 was included in the *SMN2* mRNA. All three classes exhibited high potency in both *in vitro* and *in vivo* studies [16]. However, coumarines and isocoumarines were associated with genotoxicity, phototoxicity, and chemical instability; therefore, only the pyrido-pyrimidinone series was pursued further. After an optimisation strategy, the compound RG7800 from this series was selected for clinical development [17].

RG7800 was assessed both in healthy individuals and in SMA patients, but its development was discontinued due to pre-clinical safety concerns. The first-in-human study to assess RG7800 for safety, tolerability, pharmacokinetics, and pharmacodynamics was performed in healthy males. The study was a single-ascending dose, placebo-controlled, double-blinded study in which RG7800 induced a dose- and exposure-dependent increase in full-length *SMN2* mRNA levels. These data supported the progress of RG7800 into a phase 2 clinical trial (MOONFISH; NCT03032172) for patients with SMA Types 2 and 3, in which it was shown that the compound caused a twofold increase in SMN protein levels after 12 weeks of treatment. However, in a long-term preclinical toxicity study on cynomolgus monkeys performed in parallel, it was observed that the animals developed non-reversible histological changes of the retina after daily doses for 39 weeks. Even though no adverse effects were observed in clinical studies and the exposure levels of the preclinical study were considerably higher, development of RG7800 was halted in 2015 [18].

After the MOONFISH trial, which was the first *in vivo* proof of mechanism for the *SMN2* splicing modifier, research focused on improving the safety and pharmacokinetic (PK)/pharmacodynamic (PD) profile. Non-clinical studies on the pharmacological properties and safety of RG7800 showed that specific properties of the compound accounted for off-target potential side effects. These included its interaction with the cardiac voltage sensitive potassium channel hERG, its large volume of distribution, and the histological findings of phospholipidosis as well as its phototoxic potential (even though it was smaller compared to other chemical classes with the ability to induce *SMN2* pre-mRNA splicing modification). Therefore, research focused on diminishing these adverse effects by targeting those properties of RG7800 that likely accounted for these effects such as its basicity, volume of distribution, half-life, and UV absorption [18].

Additionally, studies on the selectivity of the compound showed that RG7800 promoted the alternative splicing of other genes including the *forkhead box protein M1 (FOXM1)* and the *MAP Kinase Activating Death Domain (MADD)*, which are thought to be involved in cell cycle regulation and apoptosis, respectively. Further research with goals of increasing the selectivity for *SMN2* and decreasing 'off-target' splicing events led to the discovery of risdiplam. Even though risdiplam has similar 'off-target' effects as RG7800, it has enhanced target potency and an additional chemical modification (i.e. no N-dealkylation) that makes risdiplam safer by increasing its *in vivo* stability and decreasing the number of active metabolites. The off-target effects are associated with

consequences both *in vitro* and *in vivo* including micronucleation induction and degeneration of germ cells in the testes of monkeys and rats [18].

3.3. Mechanism of action and preclinical development of risdiplam

Risdiplam promotes the inclusion of exon 7 *in vitro* in SMA patient-derived fibroblasts and in motor neurons generated from induced pluripotent stem cells derived from patients with SMA Type 1. Full-length *SMN2* mRNA production and increased levels of SMN protein were observed in these cell models [18]. The exact mechanism of action of risdiplam is not yet completely understood.The working model resulting from studies on risdiplam-like *SMN2* splicing modifiers is that the compound binds on two sites within the exon 7 of the *SMN2* transcript, namely exonic splicing enhancer 2 (ESE2) and 5′ splice site (5′ss). Binding to the 5′ss enhances the binding of the U1 snRNA. The interaction with the ESE2 is believed to lead to dislocation of the hnRNP G allowing the binding of the U1 snRNP complex (Figure 1) [19,20].

Additionally, risdiplam was assessed in two SMA mouse models: the C/C-allele and the SMAΔ7. Adult C/C-allele mice, which have a mild form of SMA with normal life span but muscle weakness and reduced body weight, were treated orally for 10 days. SMAΔ7 mice, which have a severe form of SMA usually leading to death within the first 3 weeks of life, were treated with intraperitoneal injection of risdiplam from postnatal day 3 to postnatal day 23 and with oral gavage thereafter. In both studies, the SMN protein levels were increased in both the brain and in the quadriceps muscles [8,18]. Brain penetration and increases in SMN protein level have also been demonstrated with other oral SMN2 splicing modifiers with chemical similarities to risdiplam [16]. Treatment with risdiplam led to a dramatically significant prolongation of life as well as a gain in body weight in the SMAΔ7 mice model, showing that treatment with risdiplam could prevent the manifestations of an SMA phenotype in the severely affected mouse model. Risdiplam-treated SMAΔ7 mice also showed a dose-dependent improvement in neuromuscular architecture and increase of the motor neurons. Further animal studies have shown that risdiplam distributes in the central nervous system and other tissues, as expected based on its high passive permeability. This is possible as risdiplam is not a substrate of the human multidrug resistance protein 1, which would otherwise restrict blood-brain barrier penetration via ATP-dependent efflux. Total drug levels were similar in the plasma, muscle, and brain of 90 mice, 148 rats, and 24 monkeys used for the experiments [21].

3.4. Clinical development of risdiplam

3.4.1. General drug information

The timeline of risdiplam development is shown in Table 1. Risdiplam is produced as a powder that is dissolved in purified water for oral dosing. It is recommended that risdiplam be administered *per os* or via nasogastric/gastrostomy tube once daily after meals. The recommended dose is 0.2 mg/kg for patients between 2 months and 2 years. For patients older than 2 years, the recommended dose is 0.25 mg/kg for patients weighing less than 20 kg and 5 mg for patients weighing more than 20 kg. The time required for risdiplam to reach maximum plasma concentration after oral administration is 1 to 4 hours. At steady state, the apparent distribution volume is 6.3 L/kg, and risdiplam is mainly bound to serum albumin. After once daily oral administration in healthy subjects, steady-state exposures were reached after 7–14 days. Risdiplam is primarily metabolised by flavin monooxygenases FMO1 and FMO3 and by cytochrome P450 (CYP) proteins 1A1, 2J2, 3A4, and 3A7. The parent drug accounts for the majority (83%) of drug-related material in plasma, and the major metabolite is the pharmacologically inactive M1. Following a dose of 18 mg, approximately 53% of the dose (14% unchanged risdiplam) is excreted in feces and 28% in urine. In healthy adults, the half-life of risdiplam is approximately 50 hours. In preclinical studies, risdiplam was found to have adverse effects on reproductive organs, including germ cells, in males. Based on observations from animal studies, these effects are expected to be reversible upon discontinuation of risdiplam. The most common side effects of risdiplam are fever, diarrhea, and rash in at least 10% of treated patients with SMA Types 2 and 3. In SMA Type 1 patients, risdiplam-treated subjects presented with upper respiratory tract infection, constipation, pneumonia, and vomiting at an incidence of at least 10%, but these conditions are common in untreated SMA Type 1 patients and do not appear to be drug related. Less common adverse events observed during clinical trials in SMA Type 2 and 3 patients included mouth and aphthous ulcers, arthralgia, and urinary tract infection [9,13,22].

3.4.2. A single ascending dose study in healthy subjects

Risdiplam was initially assessed in a randomised, placebo-controlled phase 1 trial (NCT02633709) in healthy individuals. The aim of the trial was to assess the safety and tolerability of single ascending oral risdiplam doses (ranging from 0.6 to 18.0 mg). Secondary objectives were to assess the effect of risdiplam on the *SMN2* mRNA, the effect of food as well as the effect of itraconazole, a CYP3A inhibitor, on risdiplam PK. The study included 33 healthy individuals (18–45 years), and it demonstrated that risdiplam was well-tolerated both in the fed and fasted state. With the use of Bayesian statistical methods, it was shown that risdiplam led to an increase of full-length *SMN2* mRNA in a dose-dependent manner with the 18.0 mg dose leading to 41% (confidence interval (CI) 95%: 27%, 55%) of the estimated maximum increase. Two drug-related adverse events reported by investigators were pollakiuria in the placebo group and headache with the highest risdiplam dose (18.0 mg) in the fasted cohort; these ultimately resolved. Other reported adverse events included headache, abdominal pain, diarrhea, and nasopharyngitis [23].

3.4.3. FIREFISH trial

FIREFISH (NCT02913482) is an ongoing, multicentre, open-label, two-part, phase 2/3 trial of risdiplam in infants (1–7 months) with SMA Type 1 and two copies of *SMN2*. Part 1 is an open-label, dose-finding study aiming to assess safety, tolerability, PK/PD of oral risdiplam as well as to define the

Table 1. Summary of clinical trials assessing risdiplam. Abbreviations: PK: pharmacokinetics, PD: pharmacodynamics, SMA: spinal muscular atrophy, SMN2: survival of motor neuron 2, OLE: open-label extension, BSID-III: Bayley Scale for Infant Development-III.

Name of trial/Phase (Identifier)	Participant characteristics	Design	Primary outcome	Status
Phase 1 (NCT02633709)	33 healthy individuals, ages 18–45	single-centre, randomised, single-ascending-dose, placebo-controlled	safety, tolerability, PK, PD	completed, [23]
FIREFISH Part I Phase 2/3 (NCT02913482)	21 participants, 1–7 months, SMA Type 1 with 2 *SMN2* copies	multicentre, open-label, dose-finding part, followed by OLE	safety, tolerability, PK, PD	ongoing, [24]
FIREFISH Part II Phase 2/3 (NCT02913482)	41 participants, 1–7 months, SMA Type 1 with 2 SMN2 copies	multicentre, open-label, confirmatory part, followed by OLE	percentage of infants who are sitting without support for at least 5 sec as assessed by BSID-III at Month 12	ongoing, [27]
SUNFISH Part I Phase 2/3 (NCT02908685)	51 participants, 2–25 years, ambulant and non-ambulant individuals with SMA Type 2 or SMA Type 3	multicentre, open-label, dose-finding part, followed by OLE	safety, tolerability, PK, PD	ongoing, [31]
SUNFISH Part II Phase 2/3 (NCT02908685)	180 participants, 2–25 years, non-ambulant individuals with SMA Type 2 or SMA Type 3	multicentre, open-label, confirmatory part, followed by OLE	change in total MFM32 score from baseline at Month 12	ongoing, [34]
JEWELFISH Phase 2 (NCT03032172)	174, non-naïve patients from 6 months to 60 years of age with any type (1–3) SMA	multicentre, open-label	safety, tolerability, PK, PD	ongoing, [35]
RAINBOWFISH Phase 2 (NCT03779334)	25 (target), pre-symptomatic SMA, up to 42 days of life, any number of *SMN2* copies	multicentre, open-label, confirmatory, followed by OLE	ability to sit independently for at least 5 sec, after 12 months of treatment as assessed by BSID-III	ongoing, [36]

dose to be used in Part 2. Part 2 is a confirmatory trial aiming to assess safety and efficacy of the risdiplam dose chosen at Part 1. After completion of 24 months in FIREFISH Part 1 or Part 2, participants are offered a choice to enter a 3-year open-label extension phase and continue to receive risdiplam. Study design allows for pooling of Part 1 and Part 2 data at 24 months as the eligibility criteria, dosing regimen, safety and efficacy assessments and schedule are the same. This trial began in December 2016 and the estimated completion date is in November 2023.

Part 1. Part 1 took place in seven centres across five countries. Part 1 recruited 21 infants who were divided into low-dose (0.08 mg/kg/day at 12 months, n = 4) and high-dose (0.2 mg/kg/day at 12 months, n = 17) cohorts. The median age at enrollment was 6.7 months (range: 3.3–6.9). Overall, risdiplam was safe, well-tolerated, and led to a median of 2.1-fold, compared to baseline levels, increase of SMN protein in blood after 4 weeks of treatment in the high-dose cohort. After 12 months of treatment, the survival rate was 90.5% (19/21), and none of the surviving infants lost their ability to swallow or required permanent ventilation (event-free survival). Permanent ventilation was defined as tracheostomy, equal or more than 16 hours of non-invasive ventilation per day, or intubation for more than 21 consecutive days in the absence of, or following the resolution of, an acute reversible event. Exploratory outcomes of efficacy were also assessed. In the high-dose cohort, 33% of participants (7/21) were able to sit independently for at least 5 seconds, as assessed by the Gross Motor Domain of Bayley Scale for Infant Development (BSID)-III at 12 months. Additionally, 11/21 (52%) participants reached a score of 40 or more in the Children's Hospital of Philadelphia Infant Test of Neuromuscular Disorders (CHOP-INTEND) [24]. These milestones were achieved by all participants who were in the high-dose cohort, and they are not normally seen within the natural history of SMA Type 1 [11,25,26]. After 12 months of risdiplam, one participant in the high-dose cohort was able to bear weight standing as assessed by the Hammersmith Infant Neurological Examination (HINE)-2. The survival after more than 23 months of treatment was 81% (17/21). The four participants who died suffered from respiratory complications, consistent with the natural progress of SMA. There were no drug-related adverse events that led to participants' withdrawal and the higher dose was chosen for Part 2 [24].

Part 2. The single-arm Part 2 recruited 41 participants in 14 centres across 10 countries. Median age at enrollment was 5.3 months (range: 2.2 to 6.9). The primary outcome measure was the ability to sit without support for at least 5 seconds after 12 months of treatment (for the participant recruited last) as assessed by the Gross Motor Domain of BSID-III. After 12 months of treatment, 29% of infants (12/41) were able to sit without support for at least 5 seconds, which was significantly higher compared to the 5% performance criterion, which was chosen based on natural history data [27]. This is a milestone which is not achieved within the natural history of individuals with SMA Type 1 [25,28]. For the secondary outcome measures, performance was compared with predefined performance criteria, which were based on the upper boundaries of the CI around the percentage of historical controls who achieved the same performance criterion or milestone. Data was derived from historical controls of untreated patients with similar characteristics to the trial population [26,28,29]. After 12 months of treatment, 56% (23/41) *vs*. 17% (performance criterion) of participants achieved a score of 40 or higher in the CHOP-INTEND, and 90% (37/41) *vs*. 17% had an increase of at least four points. Overall, 78% (32/41) *vs*. 12% of participants were considered to have a HINE-2 motor-milestone response with two of them being able to stand with support. Additionally, 85% (35/41) *vs*. 42% had an event-free survival at 12 months. Three participants died within the first 3 months following enrollment from respiratory complications typical of SMA Type 1. At 24 months of treatment, the percentage of participants who were able to sit without support for 5 seconds (primary outcome) increased to 61% (25/41), and 44% (18/41) were able to sit without support for 30 seconds. More importantly, all participants who acquired the milestone at 12 months of treatment (n = 12) continued to maintain this ability. Even though no infant was able to walk at 24 months, six were able to stand with support, two were able to crawl on hands and knees, one infant was able to bounce, and one was able to walk holding onto an object. The percentage of children who were able to achieve a score of 40 or more in the CHOP-INTEND score increased to 76%. At 24 months 83% (34/41) had an event-free survival, and no further deaths were reported from Month 12. The most common adverse events reported during this trial included upper respiratory tract infections, pneumonia, pyrexia, constipation, nasopharyngitis, bronchitis, diarrhea, and rhinitis [24,27,30].

3.4.4. SUNFISH trial

SUNFISH (NCT02908685) is an ongoing, two-part multicentre, randomised, placebo-controlled phase 2/3 trial in SMA patients aged 2–25 years, who were diagnosed with late onset SMA Type 2 or Type 3. Part 1 was the exploratory, dose-finding component of the trial, assessing safety, tolerability, and PK/PD of different risdiplam dose levels in ambulant and non-ambulant individuals with SMA Type 2 and SMA Type 3. Part 2 assesses the safety and efficacy of the risdiplam dose, which was selected in Part 1, in non-ambulant individuals with SMA Type 2 and 3. After completion of 24 months in Part 2 participants are offered the opportunity to continue in an open-label extension phase with regular monitoring of safety, tolerability, and efficacy. This trial began in October 2016 and estimated completion date is in September 2023.

Part 1. Part 1 included 51 participants in four countries who were divided into two age groups with at least two dose levels each and were randomised (2:1) to receive either risdiplam or placebo; the risdiplam doses tested were 0.02, 0.05, 0.15, or 0.25 mg/kg for patients aged 2–11 years (n = 31), and 3 and 5 mg for patients aged 12–25 years (n = 20). Following a minimum 12-week, double-blind treatment period, placebo participants were switched to risdiplam at the dose tested in their cohort. Median age at screening was 7 years (range: 2–24 years). Of all the participants, 73% had SMA Type 2 and 27% had SMA Type 3. The functional characteristics of

participants in Part 1 were variable and ranged from strong ambulant to weak non-ambulant including sitters, non-sitters, and walkers. Risdiplam was well-tolerated in the assessed doses, and there were no drug-related adverse events that led to withdrawal of participants. A median twofold increase in SMN protein was seen from week 4 of treatment at the pivotal dose, as compared to baseline [31]. Part 1 included exploratory efficacy outcomes 24-month data was available on 50 patients, and there was an improvement or stabilisation in Motor Function Measure-32 (MFM32), Revised Upper Limb Module (RULM), and Hammersmith Functional Motor Scale Expanded for SMA (HFMSE) total scores, as compared to natural history data and a placebo-arm from a previous clinical trial with similar population characteristics [32,33]. A greater improvement in motor function was observed in patients aged 2–11 years than in patients aged 12–25 years. There were no clinically significant changes in respiratory function over 24 months in patients aged 2–11 and 12–25 years. Part 1 informed the dose selection of 0.25 mg/kg (weight <20 kg) and 5 mg for (weight >20 kg) for Part 2 [31].

Part 2. In the confirmatory Part 2, 180 individuals were randomised (2:1) to receive the risdiplam dose selected at Part 1 or placebo for 12 months in 42 sites across 14 countries. After the first 12-month period, all participants were switched to risdiplam for another 12 months. After completion of the 24-month treatment period, individuals could continue in the open-label extension for 3 years. The median age at recruitment was 9 years (range: 2–25 for the risdiplam group, 2–24 for the placebo group). The primary endpoint of efficacy was the change in MFM32 score at 12 months of treatment as compared to baseline. The 12-month analysis revealed that the study met its primary endpoint and two out of six key secondary endpoints. The least squares mean change from baseline in MFM32 was 1.36 (95% CI: 0.61, 2.11) in the risdiplam group and – 0.19 (95% CI: – 1.22, 0.84) in the placebo group, with a statistically significant difference in favor of risdiplam. Additionally, 70% of the risdiplam-treated participants showed stabilization or improvement on the MFM32 (score ≥0) from baseline. The domains with largest improvements on the MFM32 were the proximal (D2) and the distal (D3), which are of particular importance to the non-ambulant population. Additionally, risdiplam-treated participants showed statistically significant improvement on the RULM score, one of the key secondary endpoints. The most common adverse events reported during this trial included pneumonia, pyrexia, diarrhea, rash, mouth and aphthous ulcers, urinary tract infection, and arthralgias. There were no treatment withdrawals due to drug-related adverse events [34].

3.4.5. JEWELFISH trial

JEWELFISH (NCT03032172) is a multicentre, open-label trial primarily evaluating the safety, tolerability, and PK/PD of daily risdiplam in non-naïve patients from 6 months to 60 years with any type of SMA. Participants could have been previously enrolled in the MOONFISH trial or could have previously received treatment with nusinersen, olesoxime, or onasemnogene abeparvovec-xioi. This study began in March 2017, and the estimated completion date is in December 2024. After 24 months, participants are offered the chance to enter an extension phase. The primary endpoints are safety and PK. The key secondary endpoint is the PK/PD relationship. PD investigations include analyses of *SMN2* mRNA splice forms and SMN protein. Key exploratory endpoints relate to efficacy with measures of respiratory function, motor function, and milestones. Upon completion of enrollment, 174 participants had been recruited, and one participant had withdrawn from the study at baseline. Of the remaining 173 patients, 13 have previously received RG7800, 76 nusinersen, 14 onasemnogene abeparvovec-xioi, and 70 olesoxime. At the 12-month interim analysis, no treatment-related safety findings leading to withdrawal were reported. The discontinuation rate at 12 months was 5% (9 patients). A sustained increase in median blood SMN protein concentration was shown to be more than twofold compared to baseline levels, irrespective of previous treatment. This is consistent with PD data from the other clinical trials in treatment-naïve patients. Interim exploratory efficacy data using the MFM32 total score showed that overall, motor function remained stable at 12 months of treatment. JEWELFISH is ongoing and will provide further data on the long-term safety [35].

3.4.6. RAINBOWFISH trial

The RAINBOWFISH (NCT03779334) is an ongoing, multicentre, open-label, single-arm trial in pre-symptomatic SMA infants (regardless of *SMN2* copy number) which aims to assess efficacy, safety, and PK/PD of risdiplam. In this trial, which began in August 2019, participants receive risdiplam once daily for 24 months and then they enter an open-label extension phase of at least 36 months. For inclusion, participants had to be younger than 6 weeks of age (42 days of age) at the first dose; the target recruitment is 25 participants. The primary endpoint is the ability to sit without support for at least 5 seconds as assessed by BSID-III at 12 months of treatment. Data was analysed when 12 patients had been included (n = 5 with two copies of *SMN2*, n = 7 with more than two copies of *SMN2*). Of these 12 participants, eight were identified via newborn screening (NBS) and the rest via family history. The median duration of treatment at the data cutoff point was 7.4 months (range: 1.1–18), and five patients had received more than 12 months of treatment. Available data for the five patients who completed treatment for more than 12 months showed that four infants achieved the maximal HINE-2 score of 26, including an infant with two *SMN2* copies. The remaining infant, who had two copies of *SMN2*, was able to stand with support and had a score of 23 on the HINE. These five infants achieved the near maximum CHOP-INTEND scores (>60 (n = 4), 58 (n = 1)) and were able to continue to be exclusively orally fed without any swallowing concerns. Three treatment-related adverse events were reported in three participants, including elevated alanine aminotransferase, elevated aspartate aminotransferase, skin discolouration, and diarrhea. At the reported data cutoff, these adverse events

were resolved or were resolving with ongoing risdiplam treatment. The primary analysis for the RAINBWOFISH trial will be completed once the last enrolled patient has completed month 12 of treatment [36].

3.5. Safety and tolerability

Safety data integrated from the different ongoing studies has been presented [37]. Briefly, when considering the FIREFISH, SUNFISH, and JEWELFISH trial data, no treatment-related safety findings leading to withdrawal from risdiplam treatment were reported for up to 38.9 months in 465 patients. Adverse events that can be reliably related to risdiplam are rash and diarrhea. Importantly, risdiplam treatment has not led to retinal toxicity in clinical studies [38].

4. Conclusion

Risdiplam is a small molecule which has proved its efficacy in pivotal trials for SMA Types 1, 2, and 3. Its PK/PD profile is reliable with daily oral risdiplam doses leading to twofold SMN protein increases. No treatment-related serious side effects leading to drug withdrawal are associated with its use so far despite observations of 'off-target' splicing modification in animal models. Serious adverse events associated with histological changes in the retina or renal toxicity have not been seen so far in humans.

5. Expert opinion

Risdiplam is the third disease-modifying drug approved for SMA, and as such, its place in the treatment algorithm needs to be defined. For this to be achieved, three independent groups of patients based on the manner and timing of diagnosis need to be taken into consideration: pre-symptomatic patients identified via NBS, newly diagnosed, symptomatic patients under 2 years, and the rest of prevalent cases.

In several countries, the time lag to obtain onasemnogene abeparvovec-xioi approval for patients identified via NBS has led to the need of a 'bridging' therapy, usually achieved with one or more doses of nusinersen. Risdiplam could constitute a valuable alternative to nusinersen in this context not only because of the oral (*versus* the intrathecal) route of administration, but also because the steady state is achieved more rapidly (1 week for risdiplam *vs*. 2 months for nusinersen). A different scheme of nusinersen administration is currently in a phase 2/3 clinical trial (clinicaltrials.gov identifier: NCT04089566) that may result in more rapid attainment of steady state levels. Additionally, risdiplam is more considerably more cost-effective than nusinersen. The loading dose of nusinersen currently costs 353,200 euros, whereas 3 months of risdiplam will cost 18,083 euros for a 5-kg baby [39]. However, risdiplam is not yet approved in infants younger than 2 months. The aim of the RAINBOWFISH trial, which has recently completed recruitment, is to gather data on the safety profile and on pharmacodynamics in this population. Risdiplam could theoretically become the first choice for those identified via NBS with four *SMN2* copies, as onasemnogene abeparvovec-xioi is not approved for this population. There is no established clinical opinion of whether pre-symptomatic patients with four copies should be treated, but current consensus is leaning toward treatment [40,41].

For SMA, the timing of administration is key to obtain optimal drug efficacy for a disease-modifying treatment with nusinersen and onasemnogene abeparvovec-xioi [42]. This was confirmed by preliminary data from the RAINBOWFISH trial. Identifying patients via NBS, where possible, or raising awareness for the importance of early diagnosis where NBS is not available is key in avoiding a long diagnostic journey and loss of precious time [43]. Currently, across the world, only 2% of the newborns are screened for SMA, although this number is expected to steadily increase up to 20% in the next 5 years [44,45].

Pre-symptomatic patients with three *SMN2* copies treated with onasemnogene abeparvovec-xioi achieve normal development, and about half of those with two copies do. Although no sign of active denervation or motor regression has been observed so far in patients treated with onasemnogene abeparvovec-xioi, the expression of the transgene for the entire life of the patient is not guaranteed. Risdiplam could constitute a potential follow-up treatment if the effect of gene therapy is limited in time.

For newly diagnosed, symptomatic patients, onasemnogene abeparvovec-xioi appears to be an attractive option for families because of the 'off-medication' period, which follows the single-shot injection [46]. For these patients under the age of 2 years in the US, or below 23 kg in the EU, risdiplam could become a valuable alternative to onasemnogene abeparvovec-xioi, in two cases: in those with high titres of AAV-neutralising antibodies and in those who are unable to access appropriate funding for gene therapy. Both onasemnogene abeparvovec-xioi and risdiplam have demonstrated positive results on the bulbar function of patients with SMA Type 1 and could, therefore, be considered as an attractive first choice in this population [12,24,27].

Most of the prevalent SMA cases, including older patients, are currently treated with nusinersen [47]. For this population, there are both advantages and disadvantages when comparing the two drugs. From the one side, the oral administration of risdiplam makes it an attractive choice, not only for the ease of administration, but also for its potential to address the systemic impact of the disease; in contrast, nusinersen which is delivered intrathecally. However, the oral route can become a drawback when there is suspicion of poor compliance, in teenagers for instance. The rationale of systemic distribution in SMA Type 2 or 3 remains controversial as clinically significant symptoms from other systems have been reported only in humans with the most severe forms of SMA [48]. Dyslipidaemia or other metabolic abnormalities have been reported in late onset forms, but at present are not really considered an unmet need in this population [49]. In addition, these findings were always reported in comparison with a control population and not with a population with a similar level of disability and lack of mobility. In this context, it remains unclear if the systemic distribution of risdiplam gives added value in comparison

with nusinersen. Concerns with regard to potential long-term toxicity, not seen yet during the first 3 years of follow-up, or effects on male fertility and teratogenicity could constitute rationale to keep patients on nusinersen.

Considering that the efficacy of the three different disease-modifying drugs in SMA is not dramatically different and in the absence of any comparative studies, it is very likely that families and healthcare professionals will make a choice largely driven by factors as the safety, the route and frequency of administration, and the price [50]. Currently, there is no clear rationale of why one drug choice should be made over the other for any of the patient groups discussed above. Several other drugs are in development for spinal muscular atrophy [14]. Risdiplam is due to be tested with an antibody against myostatin in ambulant patients (clinicaltrials.gov identifier NCT05115110). The potential of a combination therapy with agents acting beyond SMN restoration is promising.

Several gaps remain in our scientific understanding of the place of risdiplam in the treatment map of SMA. First is its safety and pharmacokinetic profile in infants less than 2 months, which should be addressed by the RAINBOWFISH study. Second, long-term safety remains unknown, even if the data so far are reassuring. Efficacy in adults constitutes another gap in our current knowledge. The added value of risdiplam in conjunction with other approved medications is still not proven using a methodologically designed study. On these three last points, real-world data should progressively bring evidence. Nevertheless, the efficacy in adults or the potential add-on value of using risdiplam on top of onasemnogene abeparvovec-xioi or nusinersen will remain very challenging to determine. Indeed, older patients have a limited potential for improvement, and current clinical outcome measures are not able to capture minimal changes, especially in a heterogenous population with no standardization of the evaluation across centres, as it is the case in real-world evidence.

Finally, key lessons learned from the clinical development of risdiplam could be used in future developments for SMA or for other rare diseases of childhood. For example, the design of a prospective natural history study with inclusion and exclusion criteria which match those of the phase III trial not only allows a rapid and efficient clinical trial inclusion but also enables comparison with the data of the open-label part 1 of the SUNFISH trial [51]. A similar approach has been used recently in centronuclear myopathy and in Duchenne muscular dystrophy [52,53]. Another key challenge relates to the difficulty of conducting a clinical trial in a lethal disease of infants for which there are approved drugs. The FIREFISH trial could only be conducted with the relocation of patients from countries where nusinersen was not available. This constituted a challenge at trial conclusion, when parents had to return to their home country, where standards of care were not the same. Moreover, the SUNFISH trial provided additional evidence of the sensitivity of MFM32 in comparison to HFMSE [54]. It also allowed the first deployment of a new patient-reported outcome measure, the SMA independence scale, which demonstrates good sensitivity to change [55].

Declaration of interest

L Servais has been a consultant for F. Hoffmann-La Roche Ltd. He is a member of the Steering committee of MANATEE and RAINBOWFISH and coordinating investigator of Newborn Screening (NBS) for SMA in the UK and in Belgium, of which F. Hoffmann-La Roche Ltd. is a co-funder. S Ramdas is a member of the advisory boards for F. Hoffmann-La Roche Ltd., Novartis, and Sarepta. The authors have no other relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript. This includes employment, consultancies, honoraria, stock ownership or options, expert testimony, grants or patents received or pending, or royalties.

Reviewer disclosures

One reviewer received consultant honoraria from Roche and Biogen and speakers fees (as well as research support) from Biogen, Roche and Novartis Peer reviewers on this manuscript have no other relevant financial or other relationships to disclose

Funding

The work was supported by the Onassis Foundation, of which T Markati is a Scholar (Scholarship ID: F ZQ 040-1/2020-2021).

ORCID

Theodora Markati http://orcid.org/0000-0003-0115-264X
Gemma Fisher http://orcid.org/0000-0001-5351-7327
Sithara Ramdas http://orcid.org/0000-0003-3790-2190
Laurent Servais http://orcid.org/0000-0001-9270-4061

References

Papers of special note have been highlighted as either of interest (•) or of considerable interest (••) to readers.

1. Lefebvre S, Bürglen L, Reboullet S, et al. Identification and characterization of a spinal muscular atrophy-determining gene. Cell. 1995;80:155–165.
2. Verhaart IEC, Robertson A, Wilson IJ, et al. Prevalence, incidence and carrier frequency of 5q–linked spinal muscular atrophy – a literature review. Orphanet J Rare Dis. 2017;12:124.
3. Crawford TO, Pardo CA. The neurobiology of childhood spinal muscular atrophy. Neurobiol Dis. 1996;3:97–110.
4. Mercuri E, Bertini E, Iannaccone ST. Childhood spinal muscular atrophy: controversies and challenges. Lancet Neurol. 2012;11:443–452.
5. Tisdale S, Pellizzoni L. Disease mechanisms and therapeutic approaches in spinal muscular atrophy. J Neurosci. 2015;35:8691–8700.
6. Shababi M, Lorson CL, Rudnik-Schöneborn SS. Spinal muscular atrophy: a motor neuron disorder or a multi-organ disease? J Anat. 2014;224:15–28.
7. Yeo CJJ, Darras BT. Overturning the paradigm of spinal muscular atrophy as just a motor neuron disease. Pediatr Neurol. 2020;109:12–19.
8. Singh RN, Ottesen EW, Singh NN. The first orally deliverable small molecule for the treatment of spinal muscular atrophy. J Exp Neurosci. 2020;15:263310552097398.
9. Dhillon S. Risdiplam: first approval. Drugs. 2020;80:1853–1858.

10. Ramdas S, Servais L. New treatments in spinal muscular atrophy: an overview of currently available data. Expert Opin Pharmacother. 2020;21:307–315.
11. Finkel RS, Mercuri E, Darras BT, et al. Nusinersen versus Sham control in infantile-onset spinal muscular atrophy. N Engl J Med. 2017;377:1723–1732.
12. Mercuri E, Muntoni F, Baranello G, et al. Onasemnogene abeparvovec gene therapy for symptomatic infantile-onset spinal muscular atrophy type 1 (STR1VE-EU): an open-label, single-arm, multicentre, phase 3 trial. Lancet Neurol. 2021;20:832–841.
13. Kakazu J, Walker NL, Babin KC, et al. Risdiplam for the use of spinal muscular atrophy. Orthop Rev (Pavia). 2021;13:25579.
14. Servais L, Baranello G, Scoto M, et al. Therapeutic interventions for spinal muscular atrophy: preclinical and early clinical development opportunities. Expert Opin Investig Drugs. 2021;30:519–527.
15. Pinard E, Green L, Reutlinger M, et al. Discovery of a novel class of survival motor neuron 2 splicing modifiers for the treatment of spinal muscular atrophy. J Med Chem. 2017;60:4444–4457.
16. Naryshkin NA, Weetall M, Dakka A, et al. SMN2 splicing modifiers improve motor function and longevity in mice with spinal muscular atrophy. Science. 2014;345:688–693.
17. Ratni H, Karp GM, Weetall M, et al. Specific correction of alternative survival motor neuron 2 splicing by small molecules: discovery of a potential novel medicine to treat spinal muscular atrophy. J Med Chem. 2016;59:6086–6100.
• **The pathway to risdiplam identification.**
18. Ratni H, Ebeling M, Baird J, et al. Discovery of risdiplam, a selective survival of motor neuron-2 (SMN2) gene splicing modifier for the treatment of spinal muscular atrophy (SMA). J Med Chem. 2018;61:6501–6517.
• **The pathway to risdiplam identification.**
19. Campagne S, Boigner S, Rüdisser S, et al. Structural basis of a small molecule targeting RNA for a specific splicing correction. Nat Chem Biol. 2019;15:1191–1198.
20. Sivaramakrishnan M, McCarthy KD, Campagne S, et al. Binding to SMN2 pre-mRNA-protein complex elicits specificity for small molecule splicing modifiers. Nat Commun. 2017;8:1476.
21. Poirier A, Weetall M, Heinig K, et al. Risdiplam distributes and increases SMN protein in both the central nervous system and peripheral organs. Pharmacol Res Perspect. 2018;6:e00447.
22. EVRYSDI: prescribing information [Internet] Last accessed March 2022. Available from: https://www.gene.com/download/pdf/evrysdi_prescribing.pdf
23. Sturm S, Günther A, Jaber B, et al. A phase 1 healthy male volunteer single escalating dose study of the pharmacokinetics and pharmacodynamics of risdiplam (RG7916, RO7034067), a SMN2 splicing modifier. Br J Clin Pharmacol. 2019;85:181–193.
24. Baranello G, Darras BT, Day JW, et al. Risdiplam in Type 1 spinal muscular atrophy. N Engl J Med. 2021;384:915–923.
•• **Results from the FIREFISH trial.**
25. Finkel RS, McDermott MP, Kaufmann P, et al. Observational study of spinal muscular atrophy type I and implications for clinical trials. Neurology. 2014;83:810–817.
• **Natural history study for spinal muscular atrophy.**
26. Kolb SJ, Coffey CS, Yankey JW, et al. Natural history of infantile-onset spinal muscular atrophy. Ann Neurol. 2017;82:883–891.
• **Natural history study for spinal muscular atrophy.**
27. Darras BT, Masson R, Mazurkiewicz-Bełdzińska M, et al. Risdiplam-treated infants with Type 1 spinal muscular atrophy versus historical controls. N Engl J Med. 2021;385:427–435.
•• **Results from the FIREFISH trial.**
28. De Sanctis R, Coratti G, Pasternak A, et al. Developmental milestones in type I spinal muscular atrophy. Neuromuscul Disord. 2016;26:754–759.
29. Kolb SJ, Coffey CS, Yankey JW, et al. Baseline results of the Neuro NEXT spinal muscular atrophy infant biomarker study. Ann Clin Transl Neurol. 2016;3:132–145.
30. Servais L, Baranello G, Masson R, et al. FIREFISH Part 2: efficacy and safety of risdiplam (RG7916) in infants with Type 1 spinal muscular atrophy (SMA) (1302). Neurology. 2020;94:1302.
31. Mercuri E, Baranello G, Boespflug-Tanguy O, et al. Risdiplam in Types 2 and 3 Spinal Muscular Atrophy: a randomised, placebo-controlled, dose-finding trial followed by 24 months of treatment at the pivotal dose (submitted).
32. Muntoni F, Bertini E, Comi G, et al. Long-term follow-up of patients with type 2 and non-ambulant type 3 spinal muscular atrophy (SMA) treated with olesoxime in the OLEOS trial. Neuromuscul Disord. 2020;30:959–969.
33. Annoussamy M, Seferian AM, Daron A, et al. Natural history of Type 2 and 3 spinal muscular atrophy: 2-year NatHis-SMA study. Ann Clin Transl Neurol. 2021;8:359–373.
• **Natural history study for spinal muscular atrophy.**
34. Mercuri E, Deconinck N, Mazzone ES, et al. Safety and efficacy of once-daily risdiplam in type 2 and non-ambulant type 3 spinal muscular atrophy (SUNFISH part 2): a phase 3, double-blind, randomised, placebo-controlled trial. Lancet Neurol. 2022;21:42–52.
•• **Results from the SUNFISH trial.**
35. Chiriboga C, Bruno C, Duong T, et al. EP.279 JEWELFISH: safety, pharmacodynamic and exploratory efficacy data in non-naïve patients with spinal muscular atrophy (SMA) receiving risdiplam. Neuromuscul Disord. 2021;31:S134–S135.
36. Finkel RS, Farrar MA, Vlodavets D, et al. RAINBOWFISH: preliminary efficacy and safety data in risdiplam-treated infants with presymptomatic SMA. Muscular Dystrophy Association Clinical and Scientific Conference; 2022 Mar 13-16; Nashville, USA
37. Baranello G, Servais L, Bertini E, et al. Pooled safety data from the risdiplam clinical trial development program. British Paediatric Neurology Association; 2022 Jan; Virtual Congress.
38. Sergott RC, Amorelli GM, Baranello G, et al. Risdiplam treatment has not led to retinal toxicity in patients with spinal muscular atrophy. Ann Clin Transl Neurol. 2021;8:54–65.
39. Dangouloff T, Botty C, Beaudart C, et al. Systematic literature review of the economic burden of spinal muscular atrophy and economic evaluations of treatments. Orphanet J Rare Dis. 2021;16:47.
40. Boemer F, Caberg J-H, Beckers P, et al. Three years pilot of spinal muscular atrophy newborn screening turned into official program in Southern Belgium. Sci Rep. 2021;11(1):19922.
41. Glascock J, Sampson J, Connolly AM, et al. Revised recommendations for the treatment of infants diagnosed with spinal muscular atrophy via newborn screening who have 4 copies of SMN2. J Neuromuscul Dis. 2020;7:97–100.
42. Dangouloff T, Servais L. Clinical evidence supporting early treatment o patients with spinal muscular atrophy: current perspectives. Tcrm. 2019;15:1153–1161.
43. Pera MC, Coratti G, Berti B, et al. Diagnostic journey in spinal muscular atrophy: is it still an odyssey? PLoS One. 2020;15:e0230677.
44. Dangouloff T, Vrščaj E, Servais L, et al. Newborn screening programs for spinal muscular atrophy worldwide: where we stand and where to go. Neuromuscul Disord. 2021;31:574–582.
45. Dangouloff T, Burghes A, Tizzano EF, et al. 244th ENMC international workshop: newborn screening in spinal muscular atrophy May 10-12, 2019, Hoofdorp, The Netherlands. Neuromuscul Disord. 2020;30:93–103.
46. Deng S, Lee BH, Ciafaloni E. Parent perceptions in choosing treatment for infants with spinal muscular atrophy diagnosed through newborn screening. J Child Neurol. 2021 37 ;43–49.
47. Hagenacker T, Wurster CD, Günther R, et al. Nusinersen in adults with 5q spinal muscular atrophy: a non-interventional, multicentre, observational cohort study. Lancet Neurol. 2020;19:317–325.

48. Rudnik-Schöneborn S, Vogelgesang S, Armbrust S, et al. Digital necroses and vascular thrombosis in severe spinal muscular atrophy. Muscle Nerve. 2010;42:144–147.
49. Deguise M-O, Chehade L, Kothary R. Metabolic dysfunction in spinal muscular atrophy. Int J Mol Sci. 2021;22:5913.
50. Carey KA, Farrar MA, Kasparian NA, et al. F amily, healthcare professional, and societal preferences for the treatment of infantile spinal muscular atrophy: a discrete choice experiment. Dev Med Child Neurol. 2021. (Online ahead of printing). DOI:10.1111/dmcn.15135.
51. Chabanon A, Seferian AM, Daron A, et al. Prospective and longitudinal natural history study of patients with Type 2 and 3 spinal muscular atrophy: baseline data NatHis-SMA study. PLoS One. 2018;13:e0201004.
52. Lilien C, Reyngoudt H, Seferian AM, et al. Upper limb disease evolution in exon 53 skipping eligible patients with Duchenne muscular dystrophy. Ann Clin Transl Neurol. 2021;8:1938–1950.
53. Fouarge E, Monseur A, Boulanger B, et al. Hierarchical Bayesian modelling of disease progression to inform clinical trial design in centronuclear myopathy. Orphanet J Rare Dis. 2021;16:3.
54. Bertini E, Dessaud E, Mercuri E, et al. Safety and efficacy of olesoxime in patients with type 2 or non-ambulatory type 3 spinal muscular atrophy: a randomised, double-blind, placebo-controlled phase 2 trial. Lancet Neurol. 2017;16:513–522.
55. Trundell D, Skalicky A, Staunton H, et al. Development of the SMA Independence scale-upper limb module (SMAIS-ULM): a novel scale for individuals with Type 2 and non-ambulant Type 3 SMA. J Neurol Sci. 2022;432:120059.