

RESEARCH MONOGRAPH . COMPOUND PROFILE

# Reldesemtiv

*A next-generation fast skeletal muscle troponin activator developed by Cytokinetics to amplify skeletal muscle force output through calcium sensitization of the sarcomere, evaluated in Phase 2 and Phase 3 clinical trials in amyotrophic lateral sclerosis and spinal muscular atrophy.*

|                |                                                                                                            |
|----------------|------------------------------------------------------------------------------------------------------------|
| DOCUMENT       | KDC-MN-1658                                                                                                |
| ISSUED         | 2026-05-22                                                                                                 |
| REVISION       | 1.0                                                                                                        |
| AUTHOR         | Kodi <b>ac</b> bio <b>l</b> abs Research                                                                   |
| COMPOUND CLASS | Second-generation fast skeletal muscle troponin activator (FSTA) selective for fast skeletal muscle fibers |

RESEARCH USE ONLY

## ABSTRACT

Reldesemtiv (CK-2127107) is a small-molecule fast skeletal muscle troponin activator (FSTA) that selectively binds the regulatory troponin complex in fast skeletal muscle fibers, slows the rate of calcium release from troponin C, and thereby sensitizes the sarcomere to calcium at submaximal stimulation frequencies. The compound was discovered at Cytokinetics, Inc. through property-based optimization of a high-throughput screening hit, yielding improved free exposure, in vivo muscle activation potency, and tolerability relative to the first-generation FSTA tirasemtiv, which had failed the Phase 3 VITALITY-ALS trial primarily because of dose-limiting tolerability (dizziness, nausea, weight loss, insomnia) and a 34.2 percent treatment discontinuation rate. Reldesemtiv does not activate slow skeletal or cardiac troponin complexes, providing a selectivity basis for its intended use in conditions characterized by fast skeletal muscle weakness secondary to attenuated neuronal input, including amyotrophic lateral sclerosis (ALS), spinal muscular atrophy (SMA), and chronic obstructive pulmonary disease. In a Phase 1 pharmacodynamic study in 16 healthy volunteers, reldesemtiv amplified the tibialis anterior force-frequency response by approximately 60 percent at 10 Hz nerve stimulation at the highest plasma concentrations tested, confirming the mechanism of action in human skeletal muscle. Pharmacokinetics across five Phase 1 studies demonstrated dose-proportional exposure with a terminal half-life of approximately 5 to 14 hours depending on dose, a time to peak concentration of 2 to 3 hours, and similar pharmacokinetic profiles in young and elderly subjects. The Phase 2 FORTITUDE-ALS trial (n=458; 12 weeks; placebo, 150, 300, or

450 mg twice daily) did not reach statistical significance on its primary endpoint of slow vital capacity change ( $p=0.11$ ), though trends favoring reldesemtiv were observed across all three endpoints and a post hoc analysis of the ALSFRS-R functional scale reached nominal significance ( $p=0.01$ ). The Phase 2 SMA study ( $n=70$ ; 8 weeks; 150 or 450 mg twice daily) reported statistically significant improvement in six-minute walk distance at week 4 (35.6 m,  $p=0.0037$ ) and maximum expiratory pressure at week 8 (13.2 cmH<sub>2</sub>O,  $p=0.03$ ) in the 450 mg group, with concentration-response relationships in the highest plasma concentration quartile reaching significance on both endpoints. The Phase 3 COURAGE-ALS trial ( $n=486$ ; 24 weeks; 300 mg twice daily versus placebo; 83 centers in 16 countries) was terminated for futility at the second planned interim analysis when conditional power for the primary endpoint (ALSFRS-R score change at 24 weeks) was 8.4 percent. The primary analysis showed a mean difference of negative 1.1 points (95 percent CI, negative 2.17 to negative 0.08;  $p=0.04$ ), numerically favoring placebo. No preplanned subgroup favored reldesemtiv. The compound is not approved by any regulatory authority for any indication. This monograph reviews the chemistry, discovery, molecular pharmacology, pharmacokinetics, preclinical and clinical evidence, sourcing, handling, adverse events, and a comparative assessment of reldesemtiv against tirasemtiv, tofersen, riluzole, edaravone, and risdiplam on five competency standards.

# 1. Compound Identification

## 1.1 Names, synonyms, and registry identifiers

**International Nonproprietary Name:** Reldesemtiv.

**Development codes:** CK-2127107 (Cytokinetics internal designation); CK-107 (abbreviated form). The CK-2127107 designation appears throughout the Phase 1 and early Phase 2 literature; the INN reldesemtiv was adopted for the FORTITUDE-ALS and COURAGE-ALS clinical programs.

**Chemical Abstracts Service registry number:** 1345410-31-2 [1].

**DrugBank identifier:** DB15256.

**World Anti-Doping Agency status:** Reldesemtiv is listed on the 2024 edition of the World Anti-Doping Agency Prohibited List as a non-approved substance capable of enhancing muscle performance [2].

## 1.2 Structural class and chemistry

Reldesemtiv is a pyrimidine-based small molecule featuring a cyclobutyl ring bearing two fluorine substituents (one on the cyclobutane ring, one on the adjacent pyridine) and a pyrrole carboxamide pharmacophore [1].

**International Union of Pure and Applied Chemistry name:** 1-[2-({[trans-3-fluoro-1-(3-fluoropyridin-2-yl)cyclobutyl]methyl}amino)pyrimidin-5-yl]-1H-pyrrole-3-carboxamide [1].

**Molecular formula:** C<sub>19</sub>H<sub>18</sub>F<sub>2</sub>N<sub>6</sub>O.

**Molecular weight:** 384.4 g/mol.

**Structural class:** The compound belongs to the fast skeletal muscle troponin activator (FSTA) class, a small family of direct sarcomere modulators that also includes tirasemtiv (CK-2017357, first-generation FSTA) and the related tool compound CK-2066260 [3, 4]. The FSTA class is structurally distinct from the cardiac sarcomere

modulators omecamtiv mecarbil (cardiac myosin activator) and the cardiac troponin activators, though all three classes were discovered at Cytokinetics through related high-throughput screening campaigns targeting sarcomere components.

**Stereochemistry:** The IUPAC name specifies the trans configuration at the 1,3-disubstituted cyclobutane ring, establishing the relative stereochemistry of the fluorine and the (3-fluoropyridin-2-yl) substituents. The compound contains no classical chiral centers but the trans-cyclobutane geometry is a defined stereochemical feature relevant to target binding and pharmacokinetic properties [1].

## 1.3 Synthesis

Reldesemtiv was discovered through property-based optimization of a high-throughput screening hit, designated compound 1 in the discovery publication [1]. The optimization campaign, conducted entirely at Cytokinetics by a 24-member medicinal chemistry and pharmacology team led by Scott E. Collibee and Bradley P. Morgan, targeted improvements in free plasma exposure and in vivo muscle activation potency relative to the first-generation FSTA tirasemtiv [1]. Synthetic routes are disclosed in patent WO-2019133605, involving cyclobutane carboxamide precursor formation, fluoropyridine coupling, and subsequent modifications to generate the final pyrimidine-pyrrole carboxamide scaffold [1]. The compound was advanced as the crystalline active pharmaceutical ingredient (API) or as an amorphous spray-dried dispersion (SDD) in tablet formulation for oral administration [5]. The marketed clinical trial formulations included oral suspension and tablet presentations at strengths appropriate for the 150, 300, and 450 mg twice-daily dosing regimens studied across the clinical program [5, 6].

## 2. Abstract

Reldesemtiv (CK-2127107) is a second-generation fast skeletal muscle troponin activator that selectively binds the troponin regulatory complex in fast skeletal muscle fibers, sensitizes the sarcomere to calcium, and increases force production at sub-maximal nerve stimulation frequencies. The compound was discovered at Cytokinetics, Inc. through property-based optimization of a high-throughput screening hit and was advanced into clinical development as a successor to tirasemtiv, which failed the Phase 3 VITALITY-ALS trial due to poor tolerability. Reldesemtiv demonstrated improved pharmacokinetic properties and tolerability in five Phase 1 studies and produced pharmacodynamic proof of mechanism in healthy volunteers (approximately 60 percent increase in tibialis anterior peak force at 10 Hz nerve stimulation). Phase 2 trials in ALS and SMA showed trends toward benefit but did not reach statistical significance on primary endpoints. The Phase 3 COURAGE-ALS trial (n=486) was terminated for futility with the primary analysis numerically favoring placebo. The compound is not approved for any indication. This monograph documents the chemistry, molecular pharmacology, pharmacokinetics, preclinical and clinical evidence base, and comparative positioning of reldesemtiv against five alternative agents in the ALS and SMA therapeutic landscape.

## 3. Discovery and Development History

### 3.1 Origin of the fast skeletal muscle troponin activator class

The FSTA class was established at Cytokinetics, Inc. (South San Francisco, California) through a high-throughput screen measuring the rate of ATP hydrolysis in rabbit fast skeletal muscle myofibrils as a surrogate for force generation [3]. The screen identified an initial hit compound (CK-1303249) with AC40 potency of 7.0 micromolar. Systematic optimization of substituents yielded tirasemtiv (CK-2017357, compound 25 in the discovery series), with AC40 of 0.1 micromolar, representing approximately 70-fold potency improvement [3]. The mechanism was confirmed as direct activation of the fast skeletal muscle troponin complex through mixed-sarcomere reconstitution experiments: activation was observed only in combinations containing fast skeletal thin filaments, confirming selectivity over slow skeletal, cardiac, and smooth muscle isoforms [3].

### 3.2 Tirasemtiv clinical development and failure

Cytokinetics advanced tirasemtiv through preclinical studies in the B6SJL-SOD1(G93A) transgenic mouse model of ALS, where the compound significantly increased submaximal isometric force, forelimb grip strength, grid hang time, and rotarod performance, and notably increased diaphragm force and tidal volume in treated female mice [7]. These data supported advancement to clinical trials in ALS.

The Phase 3 VITALITY-ALS trial (NCT02496767) enrolled 744 participants with ALS, of whom 565 tolerated a 2-week open-label lead-in of tirasemtiv 125 mg twice daily and received randomized treatment (3:2:2:2 to placebo or three escalating tirasemtiv dose levels) for 24 weeks [8]. The trial did not meet its primary endpoint: slow vital capacity (SVC) declined by 14.4 percent (95% CI, negative 16.8 to negative 11.9) in the placebo group and 13.4 percent (95% CI, negative 15.3 to negative 11.6) in the tirasemtiv group ( $p=0.56$ ) [8]. Secondary endpoints likewise showed no significant differences. The critical limitation was tolerability: 34.2 percent of participants randomized to tirasemtiv discontinued treatment by 24 weeks, compared to 12.2 percent on placebo, driven by dizziness, fatigue, nausea, weight loss, and in-

somnia [8]. However, participants who tolerated their intended dose showed a numeric trend toward dose-related SVC slowing ( $p=0.11$ ), suggesting the underlying mechanism might hold therapeutic potential if tolerability could be improved [8].

### 3.3 Discovery of reldesemtiv

The failure of tirasemtiv on tolerability grounds drove a second-generation optimization campaign at Cytokinetics. Reldesemtiv (CK-2127107) was discovered through property-based optimization of the same high-throughput screening lineage, yielding a compound with improved free plasma exposure and in vivo muscle activation potency relative to tirasemtiv [1]. The compound was also designed to have less potential for drug-drug interactions than tirasemtiv [4]. The discovery was published in the Journal of Medicinal Chemistry in 2021, with all 24 authors affiliated with Cytokinetics [1].

### 3.4 Phase 1 clinical program

Five Phase 1 studies were completed in healthy subjects: a single ascending dose study (CY 5011;  $n=35$  healthy males, doses 30 to 4000 mg), a multiple ascending dose study in young and elderly subjects (CY 5012;  $n=59$ , doses 300 to 500 mg for 10 to 17 days), a pharmacokinetic/pharmacodynamic study (CY 5013;  $n=16$  healthy males, pharmacodynamic assessment of tibialis anterior force-frequency response), a formulation study (CY 5014), and a food effect study (CY 5015) [4]. The results of CY 5011, CY 5012, and CY 5013 were published in a single report by Andrews et al. (2018) in Muscle and Nerve [4].

### 3.5 Phase 2 clinical program

Following Phase 1, Cytokinetics advanced reldesemtiv into three Phase 2 programs: FORTITUDE-ALS (NCT03160898) in ALS patients (458 enrolled, reported 2021), a hypothesis-generating study in SMA patients (NCT02644668; 70 enrolled, reported 2021), and a study of physical function in COPD patients (NCT02662582) [5, 6, 9]. An additional study evaluated anti-fatigability effects in elderly subjects with mobility limitations (NCT03065959) [10]. The compound received orphan drug designation for SMA.

### 3.6 Phase 3 and program termination

Based on trends observed in FORTITUDE-ALS, particularly in a post hoc enriched subgroup of intermediate- and fast-progressing patients (ALSFRS-R difference versus placebo of 1.84 points [SE 0.49] at 12 weeks versus 0.87 points [SE 0.35] in the overall population), Cytokinetics designed and initiated COURAGE-ALS (NCT04944784), a Phase 3 trial enrolling 486 patients at 83 centers in 16 countries from August 2021 to July 2023 [9, 11]. The trial employed an enrichment strategy requiring symptom onset within 24 months and ALSFRS-R total score of 44 or below at baseline [9]. COURAGE-ALS was terminated for futility at the second planned interim analysis, with conditional power of 8.4 percent for the primary endpoint and less than 1 percent for the combined assessment of function and survival [11]. The primary analysis showed the reldesemtiv group declining faster than placebo (mean difference, negative 1.1 points; 95% CI, negative 2.17 to negative 0.08;  $p=0.04$ ) [11]. The results were published in JAMA Neurology in March 2025. Cytokinetics subsequently discontinued the ALS development program. As of the date of this monograph, reldesemtiv is not approved by any regulatory authority for any indication and is not in active clinical development.

## 4. Molecular Pharmacology

### 4.1 Target: the fast skeletal muscle troponin regulatory complex

The troponin complex is the calcium-sensitive regulatory switch of striated muscle contraction. It comprises three subunits: troponin C (TnC; the calcium-binding subunit containing four EF-hand motifs, of which two in the C-terminal domain, sites III and IV, bind calcium constitutively and two in the N-terminal regulatory domain, sites I and II, bind calcium transiently during excitation-contraction coupling), troponin I (TnI; the inhibitory subunit that blocks actin-myosin interaction in the absence of calcium), and troponin T (TnT; the tropomyosin-binding subunit that anchors the complex to the thin filament) [3, 12]. In fast skeletal muscle fibers, these subunits are encoded by the fast skeletal isoform genes (TNNC2, TNNI2, TNNT3), which are distinct from the slow skeletal (TNNC1, TNNI1, TNNT1) and cardiac (TNNC1, TNNI3, TNNT2) isoforms. The sequence homology between fast and slow skeletal troponin isoforms is approximately 50 percent, providing a structural basis for the selectivity of compounds that bind at the isoform-specific interface [3].

During normal contraction, calcium released from the sarcoplasmic reticulum binds to the N-terminal regulatory sites of TnC, inducing a conformational change that exposes a hydrophobic pocket on TnC. This pocket engages the switch region of TnI (approximately residues 104 to 131 of fast skeletal TnI), releasing TnI from actin and thereby permitting tropomyosin to shift, exposing myosin-binding sites on actin and enabling cross-bridge cycling [12]. Force production is therefore directly determined by the fractional occupancy of TnC regulatory sites by calcium and by the kinetics of calcium binding and release.

### 4.2 Mechanism of reldesemtiv: calcium sensitization through troponin C compacting

Reldesemtiv binds to the fast skeletal muscle troponin complex at the interface between TnC and the TnI switch region. Molecular dynamics simulations (150 ns, AMBER18 force field) combined with molecular docking (AutoDock Vina 4.2) and MM/PBSA free energy calculations indicate that reldesemtiv engages the TnI switch

region through interactions with residues Arg113, Met116, Val114, and Met121, forming conventional hydrogen bonds, carbon-hydrogen bonds, pi-alkyl, pi-sulfur, and halogen interactions [12]. The calculated total binding free energy is negative 41.30 plus or minus 3.68 kcal/mol, with van der Waals forces contributing negative 48.95 plus or minus 3.69 kcal/mol and electrostatic interactions contributing negative 25.15 plus or minus 7.90 kcal/mol [12]. Per-residue energy decomposition analysis identified Arg113 (negative 3.96 kcal/mol), Met116 (negative 2.23 kcal/mol), Val114 (negative 1.28 kcal/mol), and Met121 (negative 0.63 kcal/mol) as the principal contributors to total binding energy [12].

The mechanistic consequence of reldesemtiv binding is troponin C compacting. Upon reldesemtiv binding, molecular dynamics simulations show decreased root-mean-square deviation of TnC (from 2.82 plus or minus 0.33 angstroms to 2.73 plus or minus 0.37 angstroms), decreased radius of gyration (from 23.77 plus or minus 0.75 to 23.41 plus or minus 0.32 angstroms), and decreased solvent-accessible surface area (from 8684.63 plus or minus 275.87 to 8430.20 plus or minus 248.30 square angstroms) [12]. The compacting effect is more pronounced at the C-terminal domain containing EF-hands III and IV than at the N-terminal regulatory domain. Issahaku et al. (2023) interpreted this conformational tightening as creating structural conditions that favor slow release of calcium ions from TnC, resulting in sensitization of the sarcomere to calcium at submaximal concentrations and prolonged force generation at physiological stimulation frequencies [12].

The net functional consequence is that reldesemtiv increases the calcium sensitivity of the fast skeletal sarcomere without altering peak force at maximal (tetanic) stimulation. Force is amplified at submaximal nerve stimulation frequencies (the 5 to 15 Hz range corresponding to normal daily motor activity) where calcium transients are incomplete and TnC occupancy is partial [4]. At supramaximal frequencies where TnC is near-fully occupied, the compound has minimal additional effect [4].

## 4.3 Selectivity for fast skeletal muscle

Reldesemtiv is highly selective for the fast skeletal muscle troponin complex and does not activate the slow skeletal or cardiac troponin complexes [1, 4]. This selectivity was established through the original tirasemtiv work: mixed-sarcomere reconstitution assays demonstrated that activation was observed only in combinations containing fast skeletal thin filaments, not slow skeletal or cardiac thin fila-

ments [3]. The selectivity window derives from the approximately 50 percent sequence divergence between fast and slow skeletal troponin isoforms, particularly in the TnI switch region where reldesemtiv binds [3, 12]. The absence of cardiac troponin activation distinguishes the FSTA class from cardiac sarcomere modulators and is the pharmacological basis for the absence of direct cardiac effects at therapeutic concentrations.

## 4.4 Pharmacodynamic proof of mechanism in humans

In the Phase 1 pharmacodynamic study (CY 5013), 16 healthy male volunteers received placebo, 300, 1000, or 3000 mg of reldesemtiv in a randomized, double-blind, four-period crossover design with seven-day minimum washout between doses [4]. The force-frequency response of foot dorsiflexion following transcutaneous electrical stimulation of the deep fibular nerve to activate the tibialis anterior muscle was assessed at baseline and at 1, 3, 5, and 7 hours post-dose, with concurrent blood sampling for plasma concentration measurement [4].

Reldesemtiv significantly increased the tibialis anterior peak force response in a dose-dependent and concentration-dependent manner. The largest increase in peak force was approximately 60 percent at 10 Hz stimulation at the highest plasma concentrations (greater than 7 micrograms per milliliter) [4]. The effect was frequency-dependent: maximal amplification occurred at intermediate stimulation frequencies (5 to 15 Hz) characteristic of normal human motor function, with effects diminishing at higher frequencies approaching tetanic fusion [4]. Andrews et al. (2018) reported that reldesemtiv "appears more potent and produced larger increases in force than tirasemtiv, a first-generation FSTA, in a similar pharmacodynamic study," supporting its advancement as a more potent successor compound [4].

## 4.5 Comparison to tirasemtiv at the molecular level

Both tirasemtiv and reldesemtiv activate the fast skeletal troponin complex through calcium sensitization, and both exhibit equivalent fiber-type selectivity. Tirasemtiv was reported in separate literature to potentiate GABA-A receptor activity at concentrations near its therapeutic range, a potential off-target interaction implicated in the dizziness and central nervous system adverse events that limited its tolerability in VITALITY-ALS [12]. Reldesemtiv was designed to avoid this off-target activity, and the improved tolerability profile observed across clinical studies (no excess

central nervous system adverse events relative to placebo) is consistent with reduced off-target binding [1, 11]. Additionally, reldesemtiv was designed to have less potential for drug-drug interactions than tirasemtiv [4].

## 5. Pharmacokinetics and Routes of Administration

### 5.1 Absorption and dose proportionality

Reldesemtiv is orally bioavailable; all clinical studies employed oral administration. In the single ascending dose study (CY 5011), 35 healthy male volunteers received doses from 30 to 4000 mg, with dose-proportional increases in area under the plasma concentration-time curve (AUC) observed across the studied range [4]. Maximum plasma concentration (C<sub>max</sub>) ranged from 0.15 micrograms per milliliter at 30 mg to 10.02 micrograms per milliliter at 4000 mg [4]. The compound was generally well tolerated at all single doses up to 4000 mg with no serious adverse events reported [4].

In the pharmacodynamic study (CY 5013), the following single-dose pharmacokinetic parameters were observed (mean values): at 300 mg, C<sub>max</sub> 1.51 micrograms per milliliter, AUC(0-infinity) 11.58 microgram-hours per milliliter, and half-life 4.76 hours; at 1000 mg, C<sub>max</sub> 3.68, AUC 37.80, half-life 6.47 hours; at 3000 mg, C<sub>max</sub> 6.43, AUC 87.61, half-life 7.04 hours [4]. Time to peak plasma concentration was approximately 2 to 3 hours across the dose range [4].

### 5.2 Multiple-dose pharmacokinetics

In the multiple ascending dose study (CY 5012), 59 healthy young (18 to 55 years) and elderly (65 to 85 years) volunteers received 300 or 500 mg once or twice daily for 10 to 17 days [4]. Pharmacokinetic profiles were similar between young and elderly participants, and steady state was achieved within the 10 to 17 day dosing period [4]. AUC increased somewhat more than dose-proportionally with repeated dosing, consistent with modest accumulation [4].

In the Phase 2 SMA study, steady-state pharmacokinetic parameters at week 8 were: for 150 mg twice daily, C<sub>max</sub> 1.40 micrograms per milliliter (coefficient of variation 43.18 percent), trough concentration (C<sub>trough</sub>) 0.34 micrograms per milliliter (CV 88.27 percent), and AUC(0-12) 9.00 microgram-hours per milliliter (CV 49.38 percent); for 450 mg twice daily, C<sub>max</sub> 3.34 micrograms per milliliter (CV

48.77 percent), C<sub>trough</sub> 0.94 micrograms per milliliter (CV 98.06 percent), and AUC(0-12) 25 microgram-hours per milliliter (CV 52.19 percent) [6]. Plasma concentrations were stable between weeks 2 and 8, confirming steady-state attainment [6]. A linear relationship between C<sub>max</sub> and improvement in six-minute walk distance was observed ( $p=0.0086$ ), supporting a concentration-response relationship [6].

## 5.3 Terminal half-life

The terminal half-life of rel-desemtiv was dose-dependent in single-dose studies, ranging from 2.96 hours (median) at the lowest doses to 14.24 hours at the highest doses in CY 5011 [4]. The terminal half-life was generally reported as approximately 12 hours in the Cytokinetics clinical development summaries, consistent with the twice-daily dosing regimen selected for Phase 2 and Phase 3 studies [5].

## 5.4 Metabolism

Eleven metabolites of rel-desemtiv were identified through in vitro incubation with pooled human liver microsomes and three-dimensionally cultivated HepaRG human hepatocytes by Euler et al. (2025) [2]. The principal metabolic pathways include hydroxylation and glucuronide conjugation; hydroxylated 3-fluoro-2-(3-fluoro-1-methylcyclobutyl)pyridine and its glucuronide were identified as suitable urinary target analytes for doping control detection [2]. The specific cytochrome P450 enzymes responsible for the primary metabolic steps have not been published in the peer-reviewed literature as of the date of this monograph.

## 5.5 Protein binding, volume of distribution, and clearance

Formal pharmacokinetic parameters for protein binding, volume of distribution, and clearance have not been published in the peer-reviewed literature. The compound was designed to have improved free plasma exposure relative to tirasemtiv [1]; tirasemtiv had free fractions of 6.9 percent in rat plasma and 1.9 percent in human plasma [3]. Investigators requiring formal pharmacokinetic parameters for rel-desemtiv should consult the clinical study reports filed with regulatory authorities or contact the sponsor.

## 5.6 Detection and elimination

In a microdosing study (50 micrograms oral dose), reldesemtiv and its metabolites were detectable in human urine for 24 to 72 hours using an optimized HPLC-MS/MS method [2]. This detection window, while established for doping control purposes, indicates relatively rapid renal elimination of metabolites following single low-dose administration.

## 5.7 Routes of administration and formulation

All clinical administration has been oral. The compound was formulated as the crystalline active pharmaceutical ingredient (API) or as an amorphous spray-dried dispersion (SDD) in tablet form [5]. Clinical trial formulations included oral suspension and tablets at 150, 300, and 450 mg strengths for the twice-daily dosing regimens [5, 6, 11].

## 5.8 Special populations

In the Phase 1 multiple ascending dose study, pharmacokinetics were similar in young (18 to 55 years) and elderly (65 to 85 years) participants [4]. No formal pharmacokinetic studies in hepatic impairment, renal impairment, or pediatric populations have been published in the peer-reviewed literature. The Phase 2 SMA study included patients aged 12 years and older [6].

## 6. Preclinical Pharmacology

### 6.1 In vitro pharmacology: troponin complex activation

The FSTA mechanism was established through the tirasemtiv discovery work and confirmed for the reldesemtiv chemotype. In reconstituted sarcomere systems containing rabbit fast skeletal myofibrils, the FSTAs increase the rate of ATP hydrolysis (a surrogate for force generation) at submaximal calcium concentrations, with tirasemtiv exhibiting AC40 of 0.1 micromolar [3]. Mixed-sarcomere experiments demonstrated that activation occurred exclusively in preparations containing fast skeletal thin filaments (actin-tropomyosin-fast troponin); slow skeletal and cardiac thin filament preparations were not activated, confirming the molecular selectivity [3].

Reldesemtiv was identified through property-based optimization as having improved free exposure and in vivo muscle activation potency compared to tirasemtiv [1]. The precise in vitro AC40 or EC50 for reldesemtiv against the reconstituted troponin complex has not been published in the peer-reviewed literature separately from the discovery patent (WO-2019133605); the medicinal chemistry publication states that the optimization produced improved potency but does not disclose the specific numeric value in the publicly accessible abstract [1].

### 6.2 Intact muscle pharmacology in rodents

In intact rat skeletal muscle in vivo, reldesemtiv increases muscle force at submaximal nerve stimulation frequencies, increases muscle power, and decreases muscle fatigability [1, 4]. The compound increased running performance in healthy rats and in rats with heart failure produced by myocardial infarction under slowly accelerating conditions on a rotarod apparatus, demonstrating that the mechanism can improve exercise tolerance in both healthy and disease-impaired muscle [4].

The compound also restored the reduced contractile force of muscle fibers obtained from human subjects on mechanical ventilation to the force levels of control (non-ventilated) muscle fibers, providing an ex vivo demonstration of the mechanism in human tissue [4].

## 6.3 Preclinical pharmacology of tirasemtiv in the SOD1 ALS model

The most extensive preclinical data in an ALS-relevant model were generated with tirasemtiv rather than reldesemtiv, but the shared mechanism permits translational inference. Hwee et al. (2014) reported that tirasemtiv significantly increased submaximal isometric force, forelimb grip strength, grid hang time, and rotarod performance in the B6SJL-SOD1(G93A) transgenic mouse model of ALS [7]. Importantly, tirasemtiv also increased diaphragm force and tidal volume in treated female mice, supporting the hypothesis that FSTA-mediated muscle activation could preserve respiratory function in ALS [7]. These preclinical data were the principal mechanistic basis for advancing FSTAs into ALS clinical trials.

## 6.4 Related tool compound CK-2066260

CK-2066260 is a structurally related FSTA tool compound used in mechanistic studies. Cheng et al. (2019) reported that CK-2066260 increases fatigue resistance by reducing the energetic cost of muscle contraction in skeletal muscle preparations [13]. Cheng et al. (2020) demonstrated that CK-2066260 mitigates skeletal muscle weakness independently of the underlying cause, supporting the hypothesis that FSTA-mediated calcium sensitization may have broad applicability across conditions with skeletal muscle weakness [14]. These findings with the tool compound provided pharmacological support for the clinical evaluation of reldesemtiv in diverse conditions including ALS, SMA, COPD, and age-related muscle weakness.

## 6.5 Translational validity assessment

The FSTA mechanism has robust in vitro and in vivo validation at the level of skeletal muscle physiology: calcium sensitization reliably increases force output at submaximal stimulation in multiple muscle preparations and species. The disconnect between the preclinical pharmacology and the Phase 3 clinical outcome in ALS reflects the fundamental challenge that improving peripheral skeletal muscle force does not arrest or slow the upstream motor neuron degeneration that drives ALS disease progression [11, 15]. The mechanism addresses a downstream consequence (muscle weakness) rather than the upstream pathology (motor neuron loss), and the COURAGE-ALS result suggests that force amplification alone is insufficient to

produce measurable clinical benefit in ALS when motor neuron degeneration continues unabated. This translational gap was acknowledged in the Shefner and Cudkowicz (2024) editorial on failures in ALS Phase 3 trials [15].

## 7. Clinical Evidence Base

The following review of clinical evidence is organized by indication and trial phase. Reldesemtiv has been evaluated in clinical trials for amyotrophic lateral sclerosis, spinal muscular atrophy, chronic obstructive pulmonary disease, and age-related mobility limitation. The compound is not approved for any indication. Investigators should treat all reported clinical findings as research data, not as evidence of therapeutic efficacy for clinical use.

### 7.1 Phase 1 in healthy volunteers

Three Phase 1 studies published by Andrews et al. (2018) established safety, pharmacokinetics, and pharmacodynamic proof of mechanism [4]. In CY 5011 (single ascending dose; n=35), oral doses up to 4000 mg were well tolerated with dose-proportional exposure [4]. In CY 5012 (multiple ascending dose; n=59), 300 to 500 mg given once or twice daily for 10 to 17 days produced similar pharmacokinetics in young and elderly subjects with steady-state achievement [4]. In CY 5013 (pharmacodynamic study; n=16), reldesemtiv amplified the tibialis anterior force-frequency response by approximately 60 percent at 10 Hz at plasma concentrations greater than 7 micrograms per milliliter, with maximal effects at the 5 to 15 Hz range corresponding to normal motor activity [4].

### 7.2 Phase 2 in ALS: FORTITUDE-ALS

FORTITUDE-ALS (NCT03160898) was a Phase 2, double-blind, randomized, placebo-controlled, dose-ranging trial enrolling 458 patients with ALS, randomized 1:1:1:1 to placebo, reldesemtiv 150, 300, or 450 mg twice daily for 12 weeks, with a 4-week follow-up period [5]. Approximately 85 percent of patients completed 12 weeks of treatment [5].

The primary endpoint was change in percent predicted slow upright vital capacity (SVC) at 12 weeks. The primary analysis did not reach statistical significance ( $p=0.11$ ) [5]. Secondary endpoints were the ALS Functional Rating Scale-Revised (ALSFRS-R;  $p=0.09$ ) and a muscle strength mega-score ( $p=0.31$ ), neither of which reached significance [5].

Post hoc analyses pooling all reldesemtiv doses versus placebo showed nominal significance on ALSFRS-R ( $p=0.01$ ) [5]. The trial investigators concluded that "trends favoring reldesemtiv for all three endpoints, with effect sizes generally regarded as clinically important" supported evaluation in a Phase 3 trial [5].

A post hoc analysis of durable medical equipment (DME) prescription and acceptance showed that DME acceptance was lower in the reldesemtiv groups (150 mg: 17.0 percent; 300 mg: 21.2 percent; 450 mg: 24.8 percent) compared to placebo (28.9 percent), with a hazard ratio of 0.61 for all reldesemtiv doses combined [16]. A separate health utility analysis using EQ-5D-5L reported a statistically significant but modest utility difference of 0.03 ( $p=0.0008$ ) favoring reldesemtiv at week 12, corresponding to an incremental QALY gain of 0.004 ( $p=0.0058$ ) [17].

## 7.3 Phase 2 in SMA

A Phase 2, double-blind, placebo-controlled, hypothesis-generating study (NCT02644668) enrolled 70 patients aged 12 years or older with type II, type III, or type IV SMA (91.4 percent type III; 8.6 percent type II) into two sequential ascending-dose cohorts: cohort 1 at 150 mg twice daily (2:1 reldesemtiv to placebo) and cohort 2 at 450 mg twice daily (2:1 reldesemtiv to placebo) for 8 weeks [6].

The study evaluated eight pharmacodynamic outcome measures. The primary findings were on six-minute walk distance (6MWD) and maximum expiratory pressure (MEP). For 6MWD, the 450 mg group showed a least-squares mean difference of 35.6 meters versus placebo at week 4 ( $p=0.0037$ ) and 24.9 meters at week 8 ( $p=0.058$ ) [6]. For MEP, both dose levels showed significant improvement at week 8: 150 mg, 11.7 cmH<sub>2</sub>O versus placebo ( $p=0.038$ ); 450 mg, 13.2 cmH<sub>2</sub>O versus placebo ( $p=0.03$ ) [6].

Concentration-response analyses revealed significant effects in the highest C<sub>max</sub> quartile (greater than 3.29 micrograms per milliliter): 6MWD difference of 43.3 meters versus placebo ( $p=0.010$ ) and MEP difference of 28.8 cmH<sub>2</sub>O ( $p=0.0002$ ) [6]. Notably, a 6MWD benefit of 30.8 meters ( $p=0.038$ ) persisted 4 weeks after cessation of dosing [6].

Other outcome measures (Hammersmith Functional Motor Scale Expanded, Revised Upper Limb Module, Timed Up and Go, maximum inspiratory pressure, forced vital capacity, handheld dynamometry) showed no significant differences

from placebo at week 8 [6]. The patient-reported Spinal Muscular Atrophy Health Index did not differ between reldesemtiv 450 mg and placebo ( $p=0.83$ ) [6].

## 7.4 Phase 3 in ALS: COURAGE-ALS

COURAGE-ALS (NCT04944784) was a Phase 3, double-blind, randomized, placebo-controlled trial evaluating reldesemtiv 300 mg twice daily versus placebo in patients with ALS, conducted at 83 centers in 16 countries from August 2021 to July 2023 [11]. The trial enrolled 486 participants (mean age 59.4 years; 63.6 percent male), randomized 2:1 to reldesemtiv ( $n=325$ ) or placebo ( $n=161$ ) [11]. The design was a hybrid decentralized trial with approximately half of study visits performed remotely via video telemedicine [11]. Enrollment was enriched for patients with symptom onset within 24 months and ALSFRS-R total score of 44 or below at baseline, based on post hoc findings from FORTITUDE-ALS indicating larger treatment effects in intermediate- and fast-progressing patients [9].

The primary endpoint was change in ALSFRS-R total score from baseline to week 24. Secondary endpoints included the combined assessment of function and survival (CAFS), forced vital capacity, ALSAQ-40 quality of life, and handgrip strength [11].

The Data Monitoring Committee recommended termination for futility at the second planned interim analysis when conditional power for the primary endpoint was 8.4 percent and conditional power for CAFS was less than 1 percent [11]. The primary analysis showed a mean group difference of negative 1.1 points (SE 0.53; 95% CI, negative 2.17 to negative 0.08;  $p=0.04$ ), numerically favoring placebo [11]. The CAFS analysis showed a win probability of 0.44 for reldesemtiv versus 0.49 for placebo (win ratio 0.91; 95% CI, 0.77 to 1.10;  $p=0.11$ ) [11]. Forced vital capacity, ALSAQ-40, and handgrip strength similarly failed to show any slowing of disease progression [11].

Preplanned subgroup analyses comparing clinical and demographic characteristics failed to identify any subgroup that favored reldesemtiv [11]. A post hoc analysis by baseline creatinine as a marker of muscle mass also showed no benefit [11]. Plasma pharmacokinetic analysis through week 24 confirmed appropriate treatment assignment and expected plasma levels of reldesemtiv [11].

## 7.5 COPD and other indications

A Phase 2 study (NCT02662582) evaluated CK-2127107 on physical function in subjects with COPD, utilizing cycle ergometer exercise tolerance as the primary outcome measure [10]. Results from this study have not been published in the peer-reviewed literature as of the date of this monograph. An additional Phase 2 study (NCT03065959) assessed anti-fatigability effects in elderly subjects with mobility limitations [10]. These programs appear to have been deprioritized following the COURAGE-ALS futility determination.

## 7.6 Open-label extension

An open-label extension study (NCT05442775) permitted COURAGE-ALS participants to continue receiving reldesemtiv after completion of the Phase 3 double-blind period [11]. All participants received active treatment during the 24-week open-label period [11]. Analysis of durable medical equipment utilization in the open-label extension has been reported by Rudnicki et al. (2026) [18].

## 8. Sourcing and Quality Verification

Reldesemtiv is not commercially available as a registered pharmaceutical product in any jurisdiction. It has no approved indication and no marketed formulation.

Research-grade reldesemtiv is available from chemical and pharmaceutical research suppliers (MedChemExpress, Selleckchem, Cayman Chemical, TargetMol, and other vendors of investigational compound libraries) at purities typically greater than 98 percent by HPLC. Supply is in milligram to low-gram quantities as the free base.

Investigators should confirm compound identity and purity on every research-grade lot through:

- High-performance liquid chromatography (HPLC) area-percent purity assessment, targeting greater than 98 percent
- Mass spectrometry confirmation of molecular weight (expected  $[M+H]^+ = 385.4$  for the protonated free base)
- Nuclear magnetic resonance spectroscopy ( $^1H$ -NMR and  $^{19}F$ -NMR) for structural confirmation, with the two fluorine atoms providing distinctive  $^{19}F$  chemical shifts for identity verification
- Assessment of the trans-cyclobutane geometry by NMR or crystallographic methods if stereochemical purity is critical to the investigation

Third-party analytical laboratories provide independent confirmation of vendor certificates of analysis. Given the research-only status of the compound and the limited commercial sourcing, lot-to-lot variability may be observed, and on-receipt analytical confirmation is appropriate for any investigation requiring quantitative pharmacological assessment.

The World Anti-Doping Agency classification of reldesemtiv as a prohibited substance should be noted by investigators in sports medicine contexts [2].

## 9. Reconstitution and Handling

Reldesemtiv is supplied as a white to off-white solid by research-grade chemical suppliers. The compound is orally bioavailable as a solid oral formulation (tablet or spray-dried dispersion) in clinical applications [5].

For in vitro research, the compound should be dissolved in dimethyl sulfoxide (DMSO) for stock solution preparation. Given the molecular weight of 384.4 g/mol, a 10 millimolar stock solution requires 3.84 milligrams per milliliter of DMSO. Working concentrations for in vitro sarcomere activation assays are typically in the low-micromolar range based on the mechanism of action.

For in vivo rodent studies, the compound has been administered orally as a suspension or solution, consistent with the oral bioavailability demonstrated across clinical studies [1, 4].

Storage at negative 20 degrees Celsius in a desiccated container is recommended for long-term storage of the solid compound. Stock solutions in DMSO should be stored at negative 20 degrees Celsius in single-use aliquots to minimize freeze-thaw degradation. Light protection is reasonable practice. Aqueous working dilutions should be prepared fresh before use, as aqueous stability data have not been published for the free base.

The compound contains two fluorine atoms (one on the cyclobutane ring, one on the pyridine ring) that may be relevant to metabolic tracing studies using <sup>19</sup>F-NMR or radiolabeled analogs [2].

# 10. Stack Interaction Considerations

## 10.1 Metabolic interactions

The specific cytochrome P450 enzymes responsible for reldesemtiv metabolism have not been published in the peer-reviewed literature, limiting assessment of metabolic drug-drug interactions. The compound was designed to have less potential for drug-drug interactions than tirasemtiv [4], suggesting that the development team identified and avoided specific CYP liabilities during the optimization campaign.

Eleven metabolites have been identified, principally through hydroxylation and glucuronidation pathways [2]. Investigators should exercise caution when combining reldesemtiv with strong inhibitors or inducers of hepatic CYP enzymes pending publication of the formal in vitro CYP characterization. Co-administration with other investigational compounds metabolized through unknown or overlapping pathways should be approached with appropriate pharmacokinetic monitoring.

## 10.2 Hepatic function interactions

The observation of dose-dependent transaminase elevations (ALT and AST) across Phase 2 and Phase 3 studies [5, 11] raises a precaution against concurrent administration with other hepatotoxic compounds. In COURAGE-ALS, ALT elevation occurred in 6.5 percent of reldesemtiv-treated patients versus 1.9 percent of placebo patients, and AST elevation occurred in 5.5 percent versus 0.6 percent [11]. These elevations generally resolved on drug discontinuation [5] but indicate that investigators combining reldesemtiv with other compounds should monitor hepatic function.

## 10.3 Renal function interactions

A dose-dependent decrease in estimated glomerular filtration rate was observed in FORTITUDE-ALS, trending toward resolution after study drug discontinuation [5]. The mechanism is not characterized; it may reflect inhibition of tubular creatinine secretion (an artifact of eGFR measurement) rather than true nephrotoxicity.

Investigators combining reldesemtiv with other compounds that affect renal function or creatinine handling should monitor renal function and consider whether eGFR changes represent pharmacological or measurement artifacts.

## 10.4 Pharmacodynamic interactions

**Riluzole and edaravone:** The COURAGE-ALS trial stratified randomization by concurrent riluzole and edaravone use [11], permitting co-administration with these standard-of-care ALS treatments. No specific pharmacokinetic interaction signal was reported, and the combination was tolerated throughout the study [11]. This provides practical evidence that reldesemtiv can be combined with riluzole and edaravone without unexpected pharmacodynamic or safety concerns.

**SMN-targeted therapies:** In the SMA context, reldesemtiv addresses skeletal muscle force output downstream of the motor neuron, while nusinersen, risdiplam, and onasemnogene abeparvovec address SMN protein deficiency at the genetic level. The mechanisms are non-overlapping and potentially synergistic. The Phase 2 SMA study did not formally evaluate combinations with SMN-targeted therapies [6].

**Other sarcomere modulators:** Concurrent use with cardiac sarcomere modulators (omecamtiv mecarbil, mavacamten) is not expected to produce pharmacodynamic overlap given the selectivity of reldesemtiv for fast skeletal troponin and the selectivity of cardiac modulators for cardiac sarcomere proteins. However, no formal interaction studies have been conducted.

# 11. Adverse Events and Safety Signal

## 11.1 Phase 1 safety

In the single ascending dose study (CY 5011; n=35), the most common adverse events were mild dizziness (6 of 35 participants; dose-related) and headache (5 of 35; dose-related), with cough, nausea, and diarrhea also reported [4]. All events were mild to moderate; no serious adverse events or treatment discontinuations occurred [4]. In the multiple ascending dose study (CY 5012; n=59), headache and diarrhea were the most frequent adverse events, and four participants showed ALT/AST elevation at 2 to 5 times the upper limit of normal; none met Hy's law criteria, and all resolved within 6 to 17 days [4]. In the pharmacodynamic study (CY 5013; n=16), adverse events were mild in 56.3 percent and moderate in 12.5 percent of subjects, with headache and dizziness (both dose-related) most common [4].

## 11.2 Phase 2 safety in SMA

In the SMA Phase 2 study (n=70), the overall adverse event incidence was 92.3 percent in placebo versus 83.3 percent (150 mg) and 85.0 percent (450 mg) in reldesemtiv groups [6]. The most common adverse events in the combined reldesemtiv groups versus placebo were headache (25.0 versus 19.2 percent), constipation (11.4 versus 0 percent), nausea (11.4 versus 19.2 percent), and fatigue (9.1 versus 15.4 percent) [6]. Three serious adverse events occurred in reldesemtiv groups (gastroenteritis, myocarditis, traumatic fracture with elevated liver enzymes and creatine kinase); all resolved and none were assessed as drug-related by investigators [6]. Treatment discontinuation due to adverse events occurred in 7.7 percent of placebo patients, 0 percent of 150 mg patients, and 5.0 percent of 450 mg patients [6].

## 11.3 Phase 2 safety in ALS

In FORTITUDE-ALS (n=458), nausea and fatigue were the most commonly reported adverse events [5]. A dose-dependent decrease in estimated glomerular filtration rate and transaminase elevations in approximately 5 percent of patients were ob-

served; both hepatic and renal abnormalities trended toward resolution after study drug discontinuation [5]. The compound was described as "well tolerated" by the investigators [5].

## 11.4 Phase 3 safety in ALS

In COURAGE-ALS (n=486), treatment-emergent adverse events occurred in 79.4 percent (reldesemtiv) versus 77.6 percent (placebo) of participants during the double-blind period [11]. Serious adverse events occurred in 12.6 percent versus 15.5 percent, respectively [11].

The principal safety signals compared to placebo were:

- ALT elevation: 6.5 percent versus 1.9 percent [11]
- AST elevation: 5.5 percent versus 0.6 percent [11]
- Falls: 17.8 percent versus 14.3 percent [11]
- Dose interruptions for transaminase elevation: 1.5 percent versus 0 percent [11]
- Permanent discontinuation for adverse events: 1.8 percent versus 0.6 percent [11]

Hospitalization rates were similar between groups (18.0 percent reldesemtiv versus 17.6 percent placebo) [19].

The transaminase elevation signal is the most consistent safety finding across clinical studies and represents a compound-specific hepatic effect that, while generally reversible, warrants monitoring in any future research applications.

## 11.5 Tolerability comparison to tirasemtiv

The critical tolerability improvement of reldesemtiv over tirasemtiv was the absence of the dose-limiting central nervous system effects (dizziness, insomnia) and gastrointestinal effects (nausea, weight loss) that drove 34.2 percent treatment discontinuation in VITALITY-ALS [8]. In COURAGE-ALS, the discontinuation rate for adverse events was 1.8 percent on reldesemtiv versus 0.6 percent on placebo [11], confirming that the tolerability objective of the second-generation optimization

was achieved. The improved tolerability ensured adequate drug exposure throughout the trial, removing the confounding tolerability limitation that complicated interpretation of the tirasemtiv data.

## 12. Comparable Compounds Assessment

This section presents a structured comparative assessment of five compounds against reldesemtiv, evaluated on five competency standards: novelty (1 to 5), effect size (1 to 5), promising potential (1 to 5), side-effect profile (1 to 5), and overall validation (1 to 5). Each comparator was selected for its relevance to the ALS, SMA, or skeletal muscle activation therapeutic space.

### 12.1 Tirasemtiv (CK-2017357)

**Mechanism:** First-generation fast skeletal muscle troponin activator, identical mechanism to reldesemtiv (calcium sensitization of the fast skeletal troponin complex). AC40 of 0.1 micromolar against rabbit fast skeletal myofibrils [3]. Selectivity for fast skeletal over slow skeletal and cardiac troponin is equivalent to reldesemtiv [3].

**Development trajectory:** Cytokinetics advanced tirasemtiv through extensive preclinical work in the SOD1(G93A) ALS mouse model, demonstrating significant improvements in muscle force, grip strength, and respiratory function [7]. The Phase 3 VITALITY-ALS trial (NCT02496767) enrolled 744 participants, of whom 565 completed the open-label lead-in and were randomized [8]. The trial failed: SVC decline was 14.4 percent on placebo versus 13.4 percent on tirasemtiv ( $p=0.56$ ) [8]. Treatment discontinuation was 34.2 percent on tirasemtiv versus 12.2 percent on placebo, driven by dizziness, fatigue, nausea, weight loss, and insomnia [8].

**Comparison to reldesemtiv:** Reldesemtiv was designed to address tirasemtiv's tolerability limitations, and it succeeded: discontinuation for adverse events was 1.8 percent versus 34.2 percent. However, both compounds failed Phase 3 in ALS, suggesting the FSTA mechanism itself, rather than tolerability alone, may be insufficient for clinical benefit in ALS.

**Scores:** Novelty 3 (established mechanism); Effect size 1 (no significant clinical effect); Promising potential 1 (development terminated); Side-effect profile 2 (significant tolerability issues); Overall validation 2 (preclinical validation but Phase 3 failure).

## 12.2 Tofersen (QALSODY)

**Mechanism:** Antisense oligonucleotide (ASO) targeting SOD1 mRNA, delivered intrathecally. Tofersen directly binds and mediates degradation of SOD1 mRNA produced from mutated SOD1 genes, reducing synthesis of the toxic mutant SOD1 protein that causes motor neuron degeneration [20]. The mechanism addresses the upstream genetic cause of SOD1-ALS (approximately 2 percent of all ALS cases).

**Development trajectory:** The Phase 3 VALOR trial (NCT02623699) enrolled 108 adults with SOD1-ALS and administered tofersen 100 mg intrathecally at 14-day intervals (loading) then monthly (maintenance) [20]. The trial did not meet its primary endpoint of ALSFRS-R change at 28 weeks. However, tofersen produced a 33 percent reduction in CSF SOD1 protein levels and decreased plasma neurofilament light chain (NfL), a biomarker of neurodegeneration [20]. Based on the NfL biomarker evidence, the FDA granted accelerated approval in April 2023; clinical benefit has not been conclusively demonstrated, and confirmatory trials are required [20]. Open-label extension data over 12 months showed that earlier initiation of tofersen led to deceleration in clinical and respiratory decline [20]. Adverse events include pleocytosis (42 percent versus 8 percent placebo), headache, injection-site pain, and rare severe events including myelitis and papilledema [20].

**Comparison to reldesemtiv:** Tofersen addresses the upstream cause (SOD1 gene) in a specific ALS subpopulation (SOD1-mutant), whereas reldesemtiv addresses a downstream consequence (muscle weakness) across all ALS patients. Tofersen's disease-modifying mechanism is fundamentally more targeted but applicable to approximately 2 percent of ALS patients. Reeldesemtiv's mechanism was applicable to all ALS patients but proved clinically ineffective.

**Scores:** Novelty 5 (first-in-class gene-targeted ALS therapy); Effect size 2 (biomarker effect significant, clinical effect not proven); Promising potential 4 (accelerated FDA approval, confirmatory data maturing); Side-effect profile 2 (intrathecal administration, pleocytosis, rare severe neuroinflammation); Overall validation 4 (FDA approval on biomarker basis, long-term data accumulating).

## 12.3 Riluzole

**Mechanism:** Benzothiazole with multiple neuromodulatory actions including inhibition of persistent sodium current (IC50 approximately 1.8 micromolar in spinal motor neurons), reduction of presynaptic glutamate release, and suppression of

repetitive neuronal firing [21]. The mechanism is neuroprotective rather than muscle-directed: riluzole reduces excitotoxic motor neuron injury rather than amplifying muscle force output.

**Development trajectory:** Riluzole received FDA approval in 1995 as the first pharmacological therapy for ALS based on two randomized controlled trials demonstrating a modest survival benefit of approximately 2 to 3 months in meta-analyses [21]. Brain tissue concentrations at the therapeutic dose (100 mg per day, producing plasma concentrations of 0.5 to 2 micromolar) are three to four times higher than plasma concentrations [21]. The compound is metabolized primarily by CYP1A2 [21]. The most common adverse events are nausea, asthenia, and elevated serum alanine transaminase [21].

**Comparison to reldesemtiv:** Riluzole is the longstanding standard of care in ALS with a proven (though modest) survival benefit, while reldesemtiv failed to demonstrate any clinical benefit. The mechanisms are completely non-overlapping: riluzole is neuroprotective and reldesemtiv is muscle-activating. COURAGE-ALS permitted concurrent riluzole use, and no interaction was observed [11].

**Scores:** Novelty 2 (established for 30 years); Effect size 2 (modest, 2 to 3 month survival benefit); Promising potential 3 (standard of care, no expansion); Side-effect profile 4 (well tolerated at approved dose); Overall validation 5 (FDA approved, replicated survival benefit, standard of care).

## 12.4 Edaravone (Radicava)

**Mechanism:** Free radical scavenger that reduces oxidative stress. The pivotal Phase 3 study in a selected ALS population demonstrated a 33 percent slowing of ALSFRS-R decline compared to placebo [22]. The oral suspension formulation has a bioavailability of approximately 57.3 percent relative to intravenous administration [22].

**Development trajectory:** Edaravone was approved by the FDA for ALS (initially as intravenous formulation, subsequently as oral suspension). The original approval was based on a Japanese pivotal Phase 3 trial in a narrowly defined early-stage ALS population. A global Phase 3b confirmatory study (MT-1186-A02) was conducted to further evaluate efficacy in a broader ALS population; the March 2024 results reportedly did not confirm the original findings in the broader population [15]. Long-term oral safety data over 48 weeks showed a favorable safety profile with no new safety concerns [22].

**Comparison to reldesemtiv:** Both compounds attempted to slow ALS progression through non-neuroprotective mechanisms (edaravone via antioxidant; reldesemtiv via muscle activation). Edaravone achieved regulatory approval based on a selected population, though confirmatory data in broader populations have been questioned. Reldesemtiv failed definitively in a broad ALS population with no subgroup showing benefit.

**Scores:** Novelty 3 (antioxidant mechanism in ALS); Effect size 2 (33 percent slowing in selected population, confirmatory data questioned); Promising potential 3 (approved but confirmatory trials uncertain); Side-effect profile 4 (well tolerated); Overall validation 3 (FDA approved on limited data, ongoing questions about efficacy).

## 12.5 Risdiplam (Evrysdi)

**Mechanism:** Orally administered small molecule that modifies SMN2 pre-mRNA splicing to increase inclusion of exon 7 and production of functional survival motor neuron (SMN) protein [23]. The mechanism directly addresses the genetic cause of SMA (loss of SMN1 gene function, compensated by the hypomorphic SMN2 gene). Risdiplam produces a twofold increase in SMN protein levels [23].

**Development trajectory:** The FIREFISH trial (type 1 SMA) demonstrated significant event-free survival and motor milestone achievement. The SUNFISH Part 2 trial (Phase 3, type 2 and non-ambulant type 3 SMA) demonstrated significant motor function improvement compared to placebo [23]. Risdiplam received FDA approval for SMA in August 2020 as the first oral SMN-targeted therapy.

**Comparison to reldesemtiv:** In the SMA therapeutic landscape, risdiplam addresses the upstream genetic cause (SMN protein deficiency), while reldesemtiv addresses a downstream consequence (reduced skeletal muscle force). The reldesemtiv Phase 2 SMA data showed improvements in 6MWD and MEP [6], but these functional gains were smaller and less durable than the motor function improvements reported in the SMN-targeted therapy trials. The two mechanisms are potentially complementary (upstream gene correction plus downstream muscle activation), but this combination has not been tested clinically.

**Scores:** Novelty 4 (first oral SMN splicing modifier); Effect size 4 (significant motor function improvement in Phase 3); Promising potential 5 (FDA approved, transformative for SMA); Side-effect profile 4 (well tolerated); Overall validation 5 (FDA

approved across SMA types, replicated efficacy).

## 12.6 Comparator summary

| COMPOUND   | NOVELTY | EFFECT SIZE | PROMISING POTENTIAL | SIDE-EFFECT PROFILE | OVERALL VALIDATION |
|------------|---------|-------------|---------------------|---------------------|--------------------|
| Tirasemtiv | 3       | 1           | 1                   | 2                   | 2                  |
| Tofersen   | 5       | 2           | 4                   | 2                   | 4                  |
| Riluzole   | 2       | 2           | 3                   | 4                   | 5                  |
| Edaravone  | 3       | 2           | 3                   | 4                   | 3                  |
| Risdiplam  | 4       | 4           | 5                   | 4                   | 5                  |

Reldesemtiv occupies a distinctive but ultimately unsuccessful position in the ALS and SMA therapeutic landscape. It represents the most pharmacologically refined approach to skeletal muscle activation (a clear improvement over tirasemtiv in tolerability and drug exposure), but the COURAGE-ALS result demonstrates that the FSTA mechanism alone is insufficient to produce measurable clinical benefit in ALS when upstream motor neuron degeneration continues. The compound's strongest clinical signal was in SMA, where the Phase 2 data on 6MWD and MEP suggest that muscle activation can produce functional gains in a condition where residual motor neurons are present but delivering suboptimal drive. However, the SMA program has not advanced to Phase 3, and the approved SMN-targeted therapies (risdiplam, nusinersen, onasemnogene abeparvovec) have established a high efficacy bar.

The principal scientific legacy of the reldesemtiv program is the demonstration that pharmacodynamic proof of mechanism (skeletal muscle force amplification in healthy volunteers) does not predict clinical efficacy in a neurodegenerative context, reinforcing the translational challenge that improving muscle output cannot compensate for progressive motor neuron loss. For investigators studying skeletal muscle physiology, calcium sensitization, or neuromuscular disease mechanisms, reldesemtiv remains a well-characterized research tool with defined pharmacodynamics, pharmacokinetics, and clinical safety data.

## 13. References

1. Collibee SE, Bergnes G, Muci AR, et al. Discovery of Reldesemtiv, a Fast Skeletal Muscle Troponin Activator for the Treatment of Impaired Muscle Function. *J Med Chem*. 2021;64(20):14930-14941. doi:10.1021/acs.jmedchem.1c01067. PMID 34636234.
2. Euler L, Deinert K, Wagener F, Walpurgis K, Thevis M. Identification of human metabolites of fast skeletal troponin activators Tirasemtiv and Reldesemtiv for doping control purposes. *Drug Test Anal*. 2025;17(6):812-824. doi:10.1002/dta.3786. PMID 39138120.
3. Collibee SE, Bergnes G, Muci AR, et al. Discovery of Tirasemtiv, the First Direct Fast Skeletal Muscle Troponin Activator. *ACS Med Chem Lett*. 2018;9(4):354-358. doi:10.1021/acsmedchemlett.7b00546. PMC5900333.
4. Andrews JA, Miller TM, Vijayakumar V, et al. CK-2127107 amplifies skeletal muscle response to nerve activation in humans. *Muscle Nerve*. 2018;57(5):729-734. doi:10.1002/mus.26017. PMID 29150952. PMC6681065.
5. Shefner JM, Andrews JA, Genge A, et al. A Phase 2, Double-Blind, Randomized, Dose-Ranging Trial Of Reldesemtiv In Patients With ALS. *Amyotroph Lateral Scler Frontotemporal Degener*. 2021;22(3-4):287-299. doi:10.1080/21678421.2020.1822410. PMID 32969758.
6. Rudnicki SA, Andrews JA, Duong T, et al. Reldesemtiv in Patients with Spinal Muscular Atrophy: a Phase 2 Hypothesis-Generating Study. *Neurotherapeutics*. 2021;18(2):1127-1136. doi:10.1007/s13311-020-01004-3. PMID 33624184. PMC8423982.
7. Hwee DT, Kennedy AR, Hartman JJ, et al. Fast Skeletal Muscle Troponin Activator tirasemtiv Increases Muscle Function and Performance in the B6SJL-SOD1G93A ALS Mouse Model. *PLoS ONE*. 2014;9(5):e96921. doi:10.1371/journal.pone.0096921. PMC4013064.
8. Shefner JM, Cudkowicz ME, Hardiman O, et al. A phase III trial of tirasemtiv as a potential treatment for amyotrophic lateral sclerosis. *Amyotroph Lateral Scler Frontotemporal Degener*. 2019;20(5-6):285-296. doi:10.1080/21678421.2019.1612922. PMID 31081694.

9. Shefner JM, Al-Chalabi A, Andrews JA, et al. COURAGE-ALS: a randomized, double-blind phase 3 study designed to improve participant experience and increase the probability of success. *Amyotroph Lateral Scler Frontotemporal Degener.* 2023;24(5-6):523-534. doi:10.1080/21678421.2023.2215836. PMID 37254449.
10. ClinicalTrials.gov. NCT02662582. A Study to Assess the Effect of CK-2127107 on Physical Function in Subjects With Chronic Obstructive Pulmonary Disease. NCT03065959. A Study to Assess the Anti-fatigability Effect of CK-2127107 in Elderly Male and Female Subjects With Limitations in Mobility.
11. Shefner JM, Cudkowicz ME, Genge A, et al. Reldesemtiv in Amyotrophic Lateral Sclerosis: Results From the COURAGE-ALS Randomized Clinical Trial. *JAMA Neurol.* 2025;82(5):477-485. doi:10.1001/jamaneurol.2025.0241. PMID 40126464. PMC11933997.
12. Issahaku AR, Ibrahim MAA, Mukelabai N, Soliman MES. Intermolecular And Dynamic Investigation of The Mechanism of Action of Reldesemtiv on Fast Skeletal Muscle Troponin Complex Toward the Treatment of Impaired Muscle Function. *Protein J.* 2023;42(4):263-275. doi:10.1007/s10930-023-10091-y. PMC10352438.
13. Cheng AJ, Hwee DT, Kim LH, et al. Fast skeletal muscle troponin activator CK-2066260 increases fatigue resistance by reducing the energetic cost of muscle contraction. *J Physiol.* 2019;597(17):4615-4625. doi:10.1113/JP278235.
14. Cheng AJ, Hwee DT, Kim LH, et al. Fast skeletal muscle troponin activator CK-2066260 mitigates skeletal muscle weakness independently of the underlying cause. *J Cachexia Sarcopenia Muscle.* 2020;11(6):1639-1652. doi:10.1002/jcsm.12624.
15. Shefner JM, Cudkowicz ME. Failures to Replicate: What Recent Negative Phase 3 Trials Have Taught Us about Amyotrophic Lateral Sclerosis Clinical Research. *Ann Neurol.* 2024;96(2):211-215. PMID 38888068.
16. Rudnicki SA, Andrews JA, Genge A, et al. Prescription and acceptance of durable medical equipment in FORTITUDE-ALS, a study of reldesemtiv in ALS: post hoc analyses of a randomized, double-blind, placebo-controlled clinical trial. *Amyotroph Lateral Scler Frontotemporal Degener.* 2022;23(3-4):263-270. PMID 34218726.

17. Gebrehiwet P, Meng L, Rudnicki SA, et al. Health utilities and quality-adjusted life years for patients with amyotrophic lateral sclerosis receiving reldesemtiv or placebo in FORTITUDE-ALS. *J Med Econ.* 2023;26(1):488-493. PMID 36930042.
18. Rudnicki SA, et al. The Use of Durable Medical Equipment in COURAGE-ALS, a Phase 3, Multicentre, Randomized Clinical Trial for ALS. *Muscle Nerve.* 2026. doi:10.1002/mus.70150. PMID 41553108.
19. Shefner JM, et al. Hospitalizations as an outcome measure in COURAGE-ALS. *Amyotroph Lateral Scler Frontotemporal Degener.* 2025. PMID 40503807.
20. Tofersen. StatPearls. NCBI Bookshelf NBK594270. Accessed May 2026. See also: Miller T, Cudkowicz ME, Genge A, et al. Trial of Antisense Oligonucleotide Tofersen for SOD1 ALS. *N Engl J Med.* 2022;387(12):1099-1110.
21. Bellingham MC. A Review of the Neural Mechanisms of Action and Clinical Efficiency of Riluzole in Treating Amyotrophic Lateral Sclerosis: What have we Learned in the Last Decade? *CNS Neurosci Ther.* 2011;17(1):4-31. doi:10.1111/j.1755-5949.2009.00116.x.
22. Shimizu H, Nishimura Y, Shiide Y, et al. Oral edaravone demonstrated a favorable safety profile in patients with amyotrophic lateral sclerosis after 48 weeks of treatment. *Muscle Nerve.* 2023;67(5):401-409. PMID 36504406. PMC10107134.
23. Darras BT, Masson R, Mazurkiewicz-Beldzinska M, et al. Risdiplam-Treated Infants with Type 1 Spinal Muscular Atrophy versus Historical Controls. *N Engl J Med.* 2021;385(5):427-435. See also: 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(1):42-52.
24. VITALITY-ALS study design: Shefner JM, Wolff AA, Meng L, et al. VITALITY-ALS, a phase III trial of tirasemtiv. *Amyotroph Lateral Scler Frontotemporal Degener.* 2018;19(5-6):321-331. PMID 29402141.
25. Patent WO-2019133605. Cytokinetics, Inc. Reldesemtiv synthesis and formulation.
26. NCT04944784. ClinicalTrials.gov. A Study to Evaluate the Efficacy and Safety of Reldesemtiv in Patients With Amyotrophic Lateral Sclerosis (ALS). COURAGE-ALS.

27. NCT03160898. ClinicalTrials.gov. A Study to Evaluate Efficacy, Safety and Tolerability of CK-2127107 in Patients With Amyotrophic Lateral Sclerosis (ALS). FORTITUDE-ALS.
28. NCT02644668. ClinicalTrials.gov. A Study of CK-2127107 in Patients With Spinal Muscular Atrophy.
29. NCT05442775. ClinicalTrials.gov. A Phase 3, Open-Label Extension of COURAGE-ALS (CY 5031).
30. NCT02662582. ClinicalTrials.gov. A Study to Assess the Effect of CK-2127107 on Physical Function in Subjects With Chronic Obstructive Pulmonary Disease.
31. NCT03065959. ClinicalTrials.gov. A Study to Assess the Anti-fatigability Effect of CK-2127107 in Elderly Male and Female Subjects With Limitations in Mobility.
32. Bora RS, Kaur A, Nishi R. Risdiplam, the First Approved Small Molecule Splicing Modifier Drug as a Blueprint for Future Transformative Medicines. *ACS Pharmacol Transl Sci.* 2021;4(3):913-924. PMC8201486.
33. Miller TM, Cudkowicz ME, Genge A, et al. Trial of Antisense Oligonucleotide Tofersen for SOD1 ALS. *N Engl J Med.* 2022;387(12):1099-1110.
34. Abe K, Aoki M, Tsuji S, et al. Safety and efficacy of edaravone in well defined patients with amyotrophic lateral sclerosis: a randomised, double-blind, placebo-controlled trial. *Lancet Neurol.* 2017;16(7):505-512.
35. Miller TM, Cudkowicz ME, Shaw PJ, et al. Phase 1-2 Trial of Antisense Oligonucleotide Tofersen for SOD1 ALS. *N Engl J Med.* 2020;383(2):109-119.
36. 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(1):42-52.
37. Day JW, Darras BT, De Vivo DC, et al. Risdiplam-Treated Infants with Type 1 Spinal Muscular Atrophy versus Historical Controls (FIREFISH). *N Engl J Med.* 2021;385(5):427-435.
38. DrugBank. Reldesemtiv. DB15256. Accessed May 2026.
39. World Anti-Doping Agency. The 2024 Prohibited List. International Standard. Reldesemtiv listed as S0 (non-approved substance).

40. Shefner JM, Wolff AA, Meng L, et al. VITALITY-ALS, a phase III trial of tirasemtiv, a selective fast skeletal muscle troponin activator, as a potential treatment for patients with amyotrophic lateral sclerosis: study design and baseline characteristics. *Amyotroph Lateral Scler Frontotemporal Degener.* 2018;19(5-6):321-331. PMID 29402141.
  41. Shefner JM, Andrews JA, Genge A, et al. A Study to Evaluate the Efficacy, Safety and Tolerability of Reldesemtiv (CK-2127107) in Patients with ALS. FORTITUDE-ALS study protocol. ClinicalTrials.gov NCT03160898.
- 

*This monograph is provided for research use only. Not for medical, diagnostic, or therapeutic purposes. Not for human consumption. Reldesemtiv is not approved by any regulatory authority for any indication and is not in active clinical development as of the date of this monograph. The compound's Phase 3 trial in ALS (COURAGE-ALS) was terminated for futility with the primary analysis numerically favoring placebo. Investigators should weight the evidence accordingly. Compound class generalizations are noted where compound-specific data are sparse; investigators should verify the applicability of such generalizations to their specific research context.*