ALS Drug Development: Reflections from the Past and a Way Forward

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Summary: Tremendous advances in our understanding of pathogenesis of amyotrophic lateral sclerosis (ALS) have provided a rich pipeline of drugs for clinical trialists. At least 32 unique compounds have been tested. Nevertheless, riluzole is currently the only treatment that prolongs survival. We present a critical overview of past clinical trials, how therapies are selected for testing in people, challenges with ALS clinical trial design and conduct, and ways to best move forward. **Key Words:** Amyotrophic lateral sclerosis, clinical trial, trial design, conduct, critical review.

INTRODUCTION

Amyotrophic lateral sclerosis (ALS) was first described in 1869 by Dr. Jean Martin Charcot.[1] ALS is a rare neurodegenerative disorder characterized by progressive muscle weakness, atrophy, and spasticity, reflecting loss of upper and lower motor neurons in the brain and spinal cord. Although symptoms and signs from motor neuron involvement predominate, cognitive and sensory abnormalities are also present in a subset of people with ALS.[2,3] The incidence is approximately 2 per 100,000 per annum and the prevalence is 6 per 100,000, respectively.[4,5] The burden of disease on patients, family members, and caregivers is substantial with high costs for assisted medical care.

Tremendous advances have occurred in understanding the genetics and pathogenesis of ALS. Symptom management has improved with studies supporting the benefit of early nutritional and respiratory support and exercise. At least 32 treatment trials have been conducted to develop therapies that slow disease progression. Nevertheless, riluzole is currently the only treatment that prolongs survival. The purpose of this article is to critically review past clinical trials in ALS and provide approaches on how to best move forward.

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PATHOGENESIS

Improved understanding of disease mechanisms underlying the selective degeneration of motor neurons has directly led to the identification of a wide range of potential targets for therapeutic intervention. These include toxicity from excess excitation of the motor neurons by transmitters such as glutamate, free radical-mediated oxidative cytotoxicity, mitochondrial dysfunction, protein aggregation, microglial activation, and cytoskeletal abnormalities.[6]

In approximately 90% of persons with ALS there is no apparent genetic linkage (sporadic ALS), but the remaining 5 to 10% of cases have a positive family history for ALS (familial ALS).[7] The discovery of several genes that cause familial motor neuron disease has provided new and exciting insights into disease mechanisms that are relevant to both familial and sporadic ALS. Mutations in the copper/zinc superoxide dismutase (SOD1) gene on chromosome 21 have been documented in 10 to 20% of families with ALS.[8] Mutations in alsin, senataxin, and vesicle-associated membrane protein B gene, are associated with motor neuron disease in a few families.[9] In addition, variations in several genes have been reported to alter the risk of developing sporadic ALS including apolipoprotein E (APOE),[10,11] ciliary neurotrophic factor,[12,13] the astrocytic glutamate transporter excitatory amino acid transporter 2/glutamate transporter 1 (EAAT2/GLT1),[14,15] vascular endothelial growth factor,[16] angiogenin,[17] paroxenases,[18] and amyotrophic lateral sclerosis 2 gene (ALS2).[19]

Table 1. *Summary of ALS Past Clinical Trials*

Proposed Mechanism of Action	ALS Past Clinical Trials*
1. Anti-excitotoxic agents	Riluzole, gabapentin, topiramate, lamotrigine, dextromethorphan, celecoxib
2. Antioxidant	Vitamin E, glutathione, N-acetylcysteine, coenzyme Q10, selegiline, topiramate
3. Immunomodulatory	Ganglioside, interferon beta-1a, cyclophosphamide, intravenous immunoglobulin, celecoxib, total lymphoid irradiation
4. Calcium regulation	Verapamil, nimodipine
5. Energy metabolism	Creatine monohydrate, coenzyme Q10, branched chain amino acids (BCAAs), L-threonine
6. Trophic factors	Ciliary neurotrophic factor, brain-derived neurotrophic factor (BDNF): intrathecal and subcutaneous, thyrotropin- releasing hormone (TRH)-intravenous and intrathecal, recombinant growth hormone, xaliproden, insulin-like growth factor 1 (IGF-1)
7. Anti-apoptotic	Omigapil (TCH346), minocycline, pentoxifylline
8. Anti-inflammatory	Celecoxib, minocycline, pentoxifylline
9. Parasympathomimetic	3,4-diaminopyridine, physostigmine

ALS = amyotrophic lateral sclerosis.
*Compounds could be listed in more than one category.

THERAPEUTIC DEVELOPMENT STRATEGIES

At least 32 unique compounds have been tested in phase II/III clinical trials in ALS during the last 15 years (Table 1). Currently there are seven active therapy trials in North America, Europe, and Asia, four upcoming trials (Table 2), and many more therapies are in early drug development. Symptomatic therapies for subjects with ALS are available, but most of these treatments have not been subjected to systematic investigation. There is evidence that early use of noninvasive ventilation[20] and percutaneous endoscopic gastrostomy[21] may prolong survival. Two small, randomized, controlled trials evaluating the effect of exercise suggest that a regular, moderate physical exercise program has a positive effect on short-term disability in ALS participants as measured by amyotrophic lateral sclerosis functional rating score (ALSFRS).[22,23] In general, high-quality, controlled studies are still very much needed to best guide symptom management in ALS.

There has been more focus on developing treatments that will slow the disease course. Using illustrative examples from previous trials, we will discuss how therapies are selected for testing in people and challenges with ALS clinical trial conduct and study designs.

THERAPY SELECTION

The development of ALS therapeutics has followed a traditional discovery path. Targets are identified from a variety of *in vitro* and *in vivo* preclinical assays that often include models based on mutant SOD1 toxicity. Lead compounds are optimized and then brought to humans for testing. However, complexities of ALS still pose a major challenge in translating progress in understanding disease pathology and pathogenesis into novel therapies. One of the major obstacles arises from the lack of *in vitro* and *in vivo* models that faithfully reflect the disease in humans. *In vitro* systems include purified primary cultures of motor neurons from embryonic rat[24] and co-culture systems of motor neurons from embryonic mice plated on a glial feeder layer of cortical astrocytes,[25] and organotypic spinal cord slices from post-natal rats.[26] In

Table 2. *Current and Upcoming ALS Treatment Trials*

2008 ALS Active Clinical Trials	
Compound	Proposed Mechanisms of Action
Ceftriaxone	Increases EAAT2/GLT1 activity, antioxidant
ONO-2506	Prevents reactive astrocytosis; glutamate antagonism; COX2 inhibitor
Co-enzyme Q-10	Antioxidant; facilitates mitochondrial respiration
Memantine	N-methyl D-aspartate (NMDA) receptor antagonist
MCI-186	Free radical scavenger; blocks mitochondrial transition pore; up regulates bcl-2 expression
Diaphragm pacing	Provide respiratory support and muscle training?
Arimoclomol	Heat shock protein inducer
ALS Future Clinical Trials	
Antisense oligonucleotide SOD1	Decrease production of SOD1 protein
Talampanal	α-amino-3-hydroxy-5-methylisoxazole-4-propionic acid (AMPA) receptor modulator
TRO19622	Inhibits opening of mitochondrial transition pore; glutamate antagonist; anti-apoptotic
R+ pramipexol	Antioxidant

COX2 = cyclooxygenase 2; bcl-2 = B-cell lymphoma 2; ALS = amyotrophic lateral sclerosis; SOD1 = superoxide dismutase.

addition, cell lines expressing mutant SOD1 have been developed with motor neuron like cells (NSC34)[27] and HeLa cells.[28] The potential limitations of *in vitro* models include biochemical or structural differences between adult and embryonic cell types and inability to replicate metabolic interactions likely to be occurring *in vivo*.

Several experimentally induced SOD1 mutations (i.e., G93A, G37R, and G85R) have been developed in transgenic mouse models[29]; however, most potential therapies have been tested on the SOD1G93A model.[30] Riluzole, first demonstrated to be effective in humans, was later shown to be effective in the SOD1.[30] The divergence between improvement in the animal model *versus* no benefit in human sporadic ALS, as seen with vitamin E, gabapentin, topiramate, creatine, celecoxib, and minocycline suggests mechanistic differences between these types of ALS, inherent differences between the mouse and human disease, difficulties related to moving from mouse to humans, or inherent flaws in human trial designs. For example, the vacuolar degeneration seen in the SOD1 models is not a feature of the disease in humans. The pharmacokinetics of a drug may differ between the mouse and humans, and picking the appropriate dosage and route of administration in humans based on experience in the mouse is not simple. It is easier to measure the biological activity of therapy in the mouse by examining the brain tissue at autopsy, whereas markers of biological activity are often missing from human trials. Several of the clinical drug studies in people with ALS had significant trial design flaws that make it impossible to determine at this time whether the mouse model based on mutant SOD1 is a valid or invalid therapy screening tool. The animal model remains critically important tool to unravel the complex stages of motor neuron disease. It might be worthwhile to test potential therapies in models with less severe phenotypes (lower gene copy number), and in more than one model. It is critical to also test markers of biological effect of new therapies in these models as possible tools to use in early human studies.

POTENTIAL TARGETS

In 1994, the first randomized trial of riluzole demonstrated a modest increase in survival. Riluzole was tested in people based on data supporting a role of glutamate toxicity in ALS. Later on, the *in vitro* model (cultured motor neurons), SOD1 mouse model, and progressive motor neuronopathy mice have all revealed the protective effect of riluzole against glutamic acid. Two well-designed, pivotal trials[31,32] demonstrated the efficacy (prolongation of median survival by 2 to 3 months) and safety of riluzole. The dose-ranging study suggested that the 100 mg dose of riluzole has the best benefit-to-risk ratio. However, lamotrigine-glutamate release inhibitor[33] and other anti-excitotoxic agents, such as topiramate[34] and gabapentin,[35] have failed to show any efficacy in treatment of ALS. Riluzole has several pharmacodynamic properties beside presynaptic inhibition of the release of glutamate, such as inhibition of G-protein-dependent processes, modulation of N-methyl-D-aspartate ionotropic receptor, and the blockade of the voltage-gated sodium channels. Unfortunately, more than a decade later, we do not know how riluzole works in ALS.

Table 3. *Trial Design Challenges*

Trial Design Flaw	Clinical Trials
Dosage selection	Too high: topiramate, minocycline Too low: creatine, celecoxib Unknown (only one dosage tested): pentoxifylline
Drug delivery	CNTF, IGF-1, BDNF subcutaneous, celecoxib, creatine
Lack of pharmacodynamic marker	All clinical trials in ALS to date except for sodium phenylbutyrate and alpha-tocopherol
Inadequate sample size	Dextromethorphan, creatine (5 g), vitamin E, selegiline, nimodipine, verapamil, N-acetylcysteine

ALS = amyotrophic lateral sclerosis.

Beside anti-excitotoxic agents, broad classes of drugs have been tested including vitamins, hormones, immunosuppressives, parasympathomimetics, antioxidants, anti-apoptotic, and neurotrophic factors. A summary of the past clinical trials is provided in Table 1. The results of most trials have been accepted as negative. However, several of these clinical trials had substantial design flaws; therefore, these may not represent failures of the therapies or the preclinical tools used to select them for testing. A summary of potential study design issues with past clinical trials is found in Table 3.

Recent approaches targeting therapies for people with familial ALS caused by mutations in SOD1 seem feasible and promising. These include the use of antisense oligonucleotides, small interfering RNA, and immunization. Intraventricular administration of antisense oligonucleotides to SOD1, significantly slowed disease progression in the mouse model of ALS caused by a mutation in SOD1.[36] Both SOD1 protein and messenger RNA levels were reduced throughout the brain and spinal cord. A human trial of antisense oligonucleotide is being planned for people with ALS and mutations in SOD1. Small interfering RNA can promote degradation of specific messenger RNA, and thus protein species. It may be useful in treatment of neurodegenerative diseases where accumulation of toxic protein drives pathogenesis.[37,38] A recent study explored immunization strategies

as potential avenues for treatment of familial ALS caused by SOD1 mutation. Repeated injections of bacterially purified recombinant SOD1 mutant protein before symptoms at 6 months of age were effective in delaying disease onset and extending the lifespan of G37R SOD1 mice.[39]

CHALLENGES IN ALS TRIAL CONDUCT

Enrollment

In any neurodegenerative disorder, it is important to try to enroll people early in their illness while avoiding the enrollment of people who do not have the disease under study. By the time people with ALS receive their diagnosis, 12 to 15 months have passed from their first symptom. In addition, inclusion criteria in ALS trials traditionally require presence of both upper and lower motor neuron dysfunction in multiple body areas to be confident about the diagnosis of ALS. This criterion excludes many people who have early ALS and delays involvement of people early in their illness. In a prospective population-based study, Traynor et al.[40] reported that 35% of the patients with ALS were considered trial ineligible at the time of diagnosis because they only met the possible El Escorial criteria (i.e., signs of upper motor neuron and lower motor neuron involvement in one region). Subjects diagnosed as having possible ALS (66%) became eligible during the follow-up period, 18% did not change diagnostic category from the time of diagnosis, and 16% died of their neurological illness without being considered trial-eligible based on El Escorial criteria of possible ALS.

Despite the devastating nature of the disease, enrollment is poor in clinical trials. Unpublished data from two major academic centers suggest that only 8% of people with ALS are enrolled in a clinical trial. On average, in both Europe and the North America, two people with ALS per trial site per month are enrolled in a clinical trial (standard deviation = 1.9; range, 0.1 to 7.5) (Richard Bedlack, personal communication). Possible reasons for low enrollment rates include overly rigorous inclusion criteria based on El Escorial criteria, stringent requirements for excellent respiratory function, frequent off-label use of medications that make people trial ineligible, lack of information among community physicians, participants, their families, and possibly investigators. The cost and burden of traveling to tertiary centers also adds to the complexity. Depending on the phase of drug development and the primary outcome measure of the trial, it would be prudent to design studies to improve enrollment. For example, most phase 2A safety studies are short in duration. It is not necessary to require excellent, high-respiratory function (>70% forced vital capacity predicted) for short-term studies. Including people early in their illness (possible ALS by El Escorial criteria) could both help enrollment and also allow earlier initia-

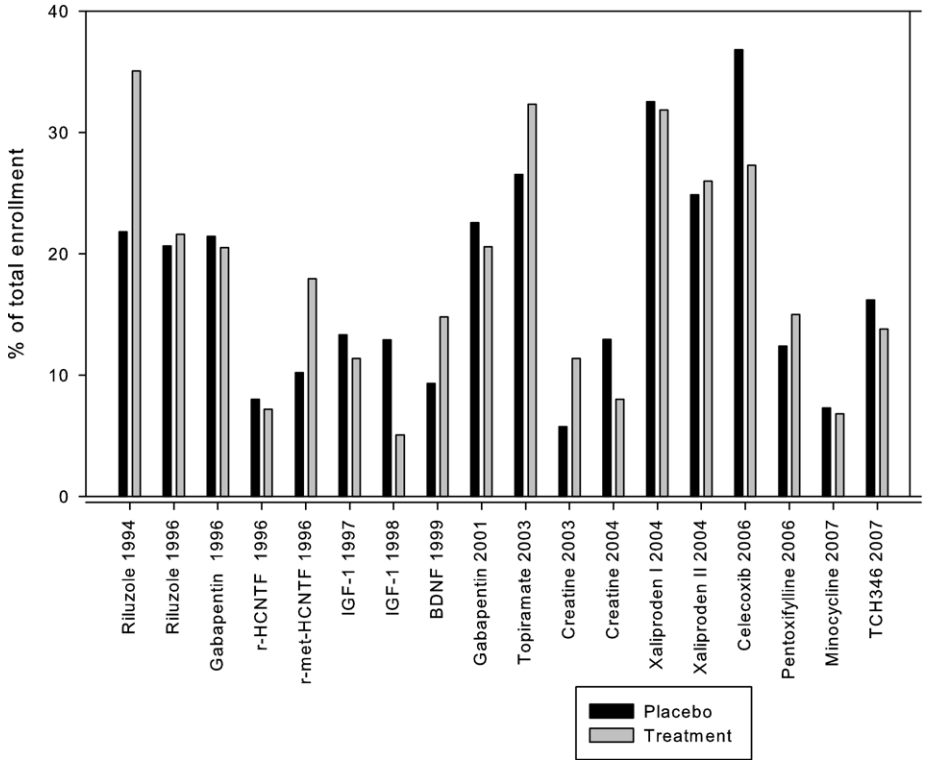

FIG. 1. Nondeath early discontinuation missing data from selected amyotrophic lateral sclerosis (ALS) clinical trials from 1994 to 2007. (Courtesy of Dr. Kevin Boylan, with permission.)

tion of potential therapies. Determining and eliminating the factors that limit enrollment will be critical as more therapeutic approaches become available for testing.

Study retention

In past ALS trials, the dropout rate was high, particularly in trials of longer duration or studies with significant toxicity (FIG. 1). With intent-to-treat (ITT) analyses, high dropout rates result in the dilution of the observed benefits of the new therapy. The real-life clinical improvement typically exceeds ITT improvement observed in a clinical trial because treatment compliance is better once the drug has been approved. For example, in a clinical trial with 1-year follow-up, a 60% improvement in primary outcome measure on treatment is needed to show a 50% improvement in the ITT analysis to account for nonadhering participants. The variation in improvement in the primary outcome measure on treatment required to show a 50% improvement in the ITT analysis increases with increasing duration of follow-up. Thus, 75% and 90% improvement in primary outcome measure on treatment are required for a trial with 2 and 3 years of follow-up to detect 50% improvement in the ITT analysis.

This critical effect of early drug discontinuation was demonstrated with riluzole. Two well-designed, pivotal trials[31,32] of riluzole demonstrated the prolongation of median survival by 2 to 3 months. Forty-four of 155 participants (i.e., 27 in the riluzole group and 17 in the placebo group) and 205 of the 959 participants (i.e., 50 in the placebo group, 48 in the 50 mg/day, 54 in the 100 mg/day, and 53 in the 200 mg/day riluzole group) discontinued treatment early in each of the trials, respectively. However, data from three observational databases[41–43] suggested that survival advantage in patients with ALS who take riluzole may be far greater than that reported in randomized, controlled studies. Imputing the missing outcomes using baseline values, or the worst case value, regression predictions, last observation carried forward, and sensitivity analysis are some of the ways the missing data has been handled, although no approach has been ideal.

Analyses of trials conducted by the Northeast ALS Consortium (NEALS)[44–46] suggest that the most common reasons for early discontinuation include subject's choice followed by mortality, adverse events, disease progression, and difficulty traveling. Trials should try to address these by making visits less difficult, as short as is reasonable, and most importantly by communicating and educating the participants at every level.

Off-label use of study medications

Off-label use of study medication significantly delayed enrollment for topiramate, celecoxib, and minocycline clinical trials. In two of these trials, participants given study medication (topiramate and minocycline) did worse than those on a placebo, emphasizing the importance of avoiding off-label use of unproven treatments. Another potential problem with a commercially available drug is that participants in the placebo group may opt to take the drug, and the ones in the treatment group may opt to take higher dosages than assigned. Unaccounted use of study medication has the potential to reduce the power of the study and increase frequency of adverse events.

CHALLENGES IN ALS TRIAL DESIGN

Dosage selection

Picking the right dosage is one of the most challenging aspects of therapy development. In many ALS trials, only a single dosage of a study drug was tested, not allowing for dosage response comparisons. The phase II/III randomized trial of Omigapil (TCH346)[47] in participants with ALS and the second study of riluzole[32] provide examples of well-conducted clinical trials that explored a broad range of dosages. Only two clinical trials to date in ALS have included a pharmacodynamic marker demonstrating that the therapy being tested has the desired biological effect. The placebo-controlled trial of alpha-tocopherol[48] and phase II study of sodium phenylbutyrate (a histone de-acetylation inhibitor) clearly demonstrated that at dosages starting at 9 g/day, histone acetylation was increased compared to untreated values (phase II study of sodium phenylbutyrate in ALS; submitted, under review). This study effectively identified a dosage that had the desired biological effect in the population of interest.

In the clinical trials of minocycline[49] and topiramate[44] in ALS, the highest tolerated dosage was selected for testing. In a randomized, phase III, placebo-controlled trial of 400 mg of minocycline daily, Gordon et al.[49] reported that participants' rate of decline, measured by the ALSFRS revised, was faster in the minocycline-treated group (-1.30 *versus* -1.04 units/month; $p = 0.005$). Survival duration and measures of breathing and strength did not differ significantly in the two groups. A phase II safety study by the same authors had suggested accelerated decline in ALSFRS revised in subjects treated with up to 400 mg/day ($p = 0.047$).[50] In hindsight, a pilot safety study had predicted the adverse effect of minocycline seen in the larger efficacy study.

Topiramate use, at dosages up to 800 mg/day, in a randomized, placebo-controlled study in people with ALS was associated with a more rapid decrease in arm strength ($p = 0.017$).[44] Topiramate use was associated with more adverse events than placebo. No preliminary studies assessing dosage were conducted. A dosage of 800 mg/day was chosen based on the therapeutic range defined by *in vitro* studies and data from its use in epilepsy. In choosing the dosage for a trial, the maximum

tolerated dose may not be the optimum dosage. Developing a biological marker of drug effect that can be used in humans would greatly assist dosage selection in early phase studies.

The selected dosage might have been too low for creatine[45] and celecoxib[46] in ALS. No beneficial effect of creatine 5 g/day[45] or 10 g/day[51] was demonstrated in ALS. However, in other neurodegenerative diseases, such as Huntington's disease, creatine 5 g was ineffective, but at 10 g and higher there was some evidence of benefit.[52] It is not certain if the dosages for both creatine and celecoxib were adequate to produce central nervous system delivery of the drug. In the $SOD1^{G93A}$ mouse model, prostaglandin E2 formation is markedly increased, and oral administration of chow containing celecoxib effectively inhibited spinal cord prostaglandin E2 levels, delayed the onset of weakness and weight loss, and prolonged survival by 25%.[53] However, in a trial of celecoxib in people with ALS, prostaglandin E2 levels in CSF were not elevated at baseline and did not decline with treatment.

Therefore, it is possible that some drug treatments and, more importantly, the scientific hypothesis behind their use in ALS participants may have been erroneously rejected. In all these studies (topiramate, minocycline, Celebrex [celecoxib, Pfizer Inc., Groton, CT] and creatine), it is not clear if the drug or the dosage tested failed, reminding us of the need to conduct traditional dosage ranging and pharmacodynamic studies before embarking in phase III trials.

Inadequate sample size

Undoubtedly, many past ALS trials were insufficiently powered in terms of participant numbers to achieve statistical significance. Trials of dextromethorphan (n = 45),[54] vitamin E (n = 160),[55] selegiline (n = 133),[56] creatine, 5 gm (n = 104),[45] nimodipine (n = 87),[57] and verapamil (n = 72)[58] were all underpowered to detect any reasonable effect size. Sample size requirements vary based on the primary outcome (Tables 4 and 5). For example, a study in which the primary outcome measure is survival, 1200 participants are needed to demonstrate a 50% change in median survival during 1 year of follow-up (90% power and alpha level of 0.05). Increasing the length of follow-up can reduce the number of participants needed. However, longer trials are associated with higher dropout rates. Sample size of 90% power and alpha level of 5%, based on the ALSFRS as the primary outcome, requires 356 patients to show a 30% change in rate of decline and 200 patients for a 40% change in rate of decline of ALSFRS. However, large changes in ALSFRS may not mean large changes in survival. For example, a decrease in the rate of change of ALSFRS by 30% decreases the 12-month hazard of death by only 4%, which translates to a 6-month increase in median survival.

Table 4. *Sample Size for Survival (90% Power, Alpha 5%, 2 Arms)*

Change	Change in Median	Change in 1-Year Survival Rate	Sample Size
100%	2.5 yrs	11%	400/(yrs of follow-up)
50%	1.3 yrs	7.5%	1200/(yrs of follow-up)
33%	10 months	5.5%	2300/(yrs of follow-up)

Table 5. *Sample Size for ALSFRS (90% Power, Alpha 5%, 1-Year, 2 Arms)*

Change in Drop	Sample Size (Total N)
25% (1.0→0.75)	512
30% (1.0→0.7)	356
40% (1.0→0.6)	200

ALSFRS = amyotrophic lateral sclerosis functional rating score.

To try to minimize sample size, a lead-in phase was used in the trials of Omigapil (TCH346)[47] and minocycline.[49] These studies took advantage of the ability to decrease variation by comparing each person's rate of decline to their own pretreatment slope. However, in both studies, a nonlinear decline in ALSFRS scores was found. This potentially invalidates some of the statistical assumptions.

Drug interactions

Early on in therapy development, it is very important to know of any interactions of study medication with riluzole. The phase III efficacy trial of xaliproden showed beneficial effect of xaliproden (2 mg/day) without riluzole on vital capacity (p = 0.009), but no effect with background riluzole therapy.[59] In mice, minocycline inhibits the efflux protein p-glycoprotein for which riluzole is a substrate. This may lead to increased riluzole CNS levels. The pharmacokinetic and toxicokinetic interactions of riluzole and minocycline were not fully explored prior to the phase 3 clinical trial.[49,60] It is possible that minocycline altered the metabolism and blood levels of riluzole, thus exacerbating the CNS side effects of riluzole. Minocycline-treated participants were more likely to have non-serious gastrointestinal and neurological adverse events; dizziness (54 *versus* 19; $p < 0.0001$) and fatigue (32 *versus* 10; $p = 0.003$).[49] It is possible that ALSFRS is very sensitive to such side effects and the difference in the ALSFRS revised slope observed in the study were related to side effects rather than a true worsening of disease pathology.

Phase 2 design challenges

Taking a drug from "bench-to-bedside" is a complex and expensive process. Given the poor success rate of

phase III trials in ALS and the relative rarity of this disorder, new initiatives are necessary to optimize phase II clinical trials designed to minimize resources on drugs that are likely to fail in later development. Traditional phase II studies in ALS are toxicity focused and have not been efficiency focused. This may be the most important reason for the large number of ALS trial failures. It may be preferable to design phase II studies incorporating predictive markers for therapeutic response (proof-of-concept) and multiple dosages to address dose-response relationships. We need to learn what dosage of the drug gives us a better response rate, can be safely administered, and has the desired biological effect prior to proceeding to phase 3 studies.

Statistical modifications to early phase II trials can also help screen drugs in the ALS pipeline more efficiently. One approach is to use a different significance level for screening each drug and doing a second trial only if one-sided $p < 0.3$. Suppose the tenth drug in our pipeline is effective, then we will decrease the sample size to find this drug by 11%. An alternative approach is selection design (i.e., using multiple drugs simultaneously that have been shown to be efficacious in models [*in vitro* and *in vivo*] for the treatment of ALS patients to screen against each other and picking the winner for testing against placebo at $p = 0.05$. This will reduce the sample size requirement to find an effective drug by 50%.[61] Regardless of approach, it remains critical to test the pharmacokinetics and pharmacodynamics of the new treatment in phase II studies.

Phase III design challenges

Outcome measures (function *versus* survival). ALS is a heterogeneous disorder. There is progressive decline in strength, but the rate of decline varies considerably from one individual to the other. Survival is the gold standard, primary endpoint for ALS trials. However, it brings with it the requirement for adequate length of study (usually 18 months). There are multiple outcome measures commonly used in various ALS clinical trials including the Tufts quantitative neuromuscular exam, hand-held dynamometry, and manual muscle testing as measures of rate of decline in muscle strength, ALSFRS revised, forced vital capacity, and motor unit number estimates. It is not known yet whether any of these outcome measures are a valid surrogate marker for survival. Muscle strength is clearly related to disease progression in ALS. However, in recent years there have been concerns that rate of decline in muscle strength dose not relate to survival. The potentially significant limitation of forced vital capacity is that a large group of patients with marked bulbar involvement often can not make a good seal around the mouthpiece, and therefore produce spuriously poor or inconsistent results. Also, variation in progression according to ALS phenotype

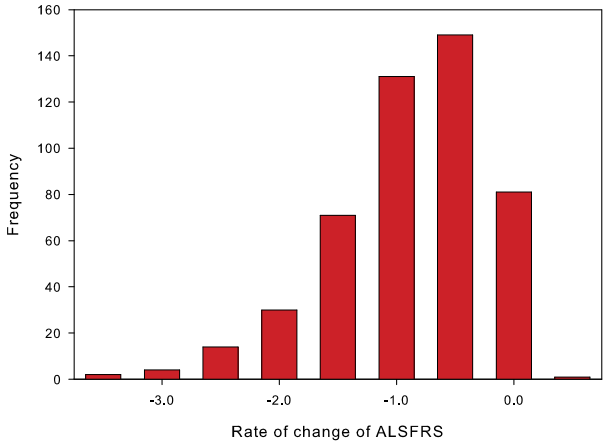

FIG. 2. Variability in rate of decline of amyotrophic lateral sclerosis functional rating score (ALSFRS) revised. Cohort with topiramate placebo, creatine (all), Celebrex (all), CoQ10 (all), but excluding subjects who had <3 visits of ALSFRS values (n = 483).

(e.g., bulbar patients have earlier respiratory failure) may make it less representative of overall disease course. The development of sniff nasal pressure test may represent a more useful marker of respiratory decline.[62,63]

The ALSFRS revised is widely used primary outcome measure in recent clinical trials as it can be quickly administered in person or on phone, so there is minimal loss of data on follow-up. However, there are several potential problems with ALSFRS. It declines at an average of 1 unit per month, but there is considerable variation in the rate of decline (FIG. 2). However, a subset of participants with ALS in past trials did not change in their ALSFRS score over 12 months. It is not clear how to handle ALSFRS score when the subject dies. If we consider ALSFRS as zero at the time of death, then the curve is nonlinear. However, ignoring deaths altogether might introduce bias in the results. Analysis of our data reveals a huge variability in the ALSFRS score prior to death, with a mean of 29 ± 8 (minimum, 11; maximum, 47).

Effects of treatment may be different for survival *versus* function. Riluzole, the only approved Food and Drug Administration medication for ALS improved survival modestly but had no effect on strength.[32] Xaliproden (without riluzole) had an effect on vital capacity ($p = 0.009$) but no effect on survival.[59] Topiramate[44] and minocycline[49] had no effect on survival, but had a negative effect on functional measures. Pentoxifylline had a negative effect on survival (unadjusted risk of 1.28 with treatment) but no effect on rate of deterioration of ALSFRS revised or in manual muscle testing.[64] Differences in study design may account for some of the observed variations, but differences in drug activity can not be excluded.

Changing natural history. As symptom management changes in ALS with earlier introduction of supportive measures, such as percutaneous gastrostomy and noninvasive ventilation, it is possible that the natural history of

Table 6. *Natural History of ALS in Clinical Trials*

Clinical Trial	Number of Subjects on Placebo	Treatment Period (months)	Mean Rate of Decline MVIC Arm (month)*	Mean Rate of Decline FVC/VC (month)*	Mean Rate of Decline ALSFRS (month)*
Gabapentin, 1996[67]	70	6	−0.132	−2.04%	Not done
BDNF, 1999[68]	387	9	Not done	−2.28%	−0.7
Gabapentin, 2001[69]	102	9	−0.088	−2.56%	Not given
Creatine, 2003[51]	87	12	−0.076	−2.07%	Not done
Topiramate, 2003[44]	98	12	−0.075	−2.46%	−0.92
Creatine, 2004[45]	54	6	−0.067	Not done	−1.005
Celecoxib, 2006[46]	99	12	−0.090	−2.19%	−1.078
Pentoxifylline, 2006[64]	201	18	Not done	Not done	−0.86
Omigapil (TCH346), 2007[47]	108	9	Not done	Not given	−0.771 (lead-in) −0.942 (double blind phase)
Minocycline, 2007[49]	206	13	Not done	−2.31 (lead-in) −3.01 (double blind)	−0.81 (lead-in) −1.04 (double blind)

BDNF = brain-derived neurotrophic factor; MVIC = maximum voluntary isometric contraction; FVC/VC = forced vital capacity/vital capacity; MVIC = xxxxx.
*Placebo group only.

the illness has changed. Significant increase in survival in 793 Italian patients with ALS was observed over a 28-year interval.[65] A recent study also suggested that ALS is becoming less aggressive over time.[66] In this study, the authors noted improved survival with slower disease progression independent of specific outcome-modifying therapies in ALS subjects diagnosed from 1999 to 2004, compared with subjects diagnosed from 1984 to 1999. This variability in natural history invalidates the use of placebo groups as historic controls for future ALS clinical trials. Data from three trials conducted by the Northeast ALS consortium from 1999 to 2006 suggest that survival has improved, while the rates of change in strength, ALSFRS, and forced vital capacity have not. A summary of outcome measures data from the placebo groups from several clinical trials is found in Table 6 and FIGs. 3, 4, 5, and 6. Although improved survival is good, it necessitates even larger or longer phase III studies when the primary outcome measure is survival.

A WAY FORWARD

Riluzole is the only therapy that has shown the ability to slow disease progression in patients with ALS. Un-

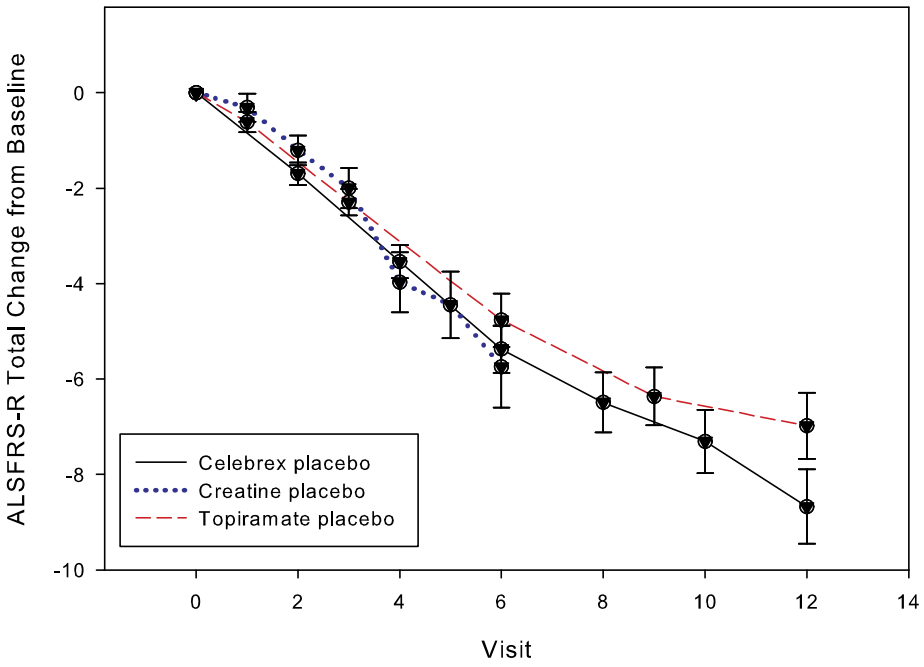

FIG. 3. Mean (± SE) change from baseline in amyotrophic lateral sclerosis functional rating score (ALSFRS) revised total at each visit for placebo groups. (Northeast Amyotrophic Lateral Sclerosis Consortium [NEALS] trials conducted from 1999 to 2006.)

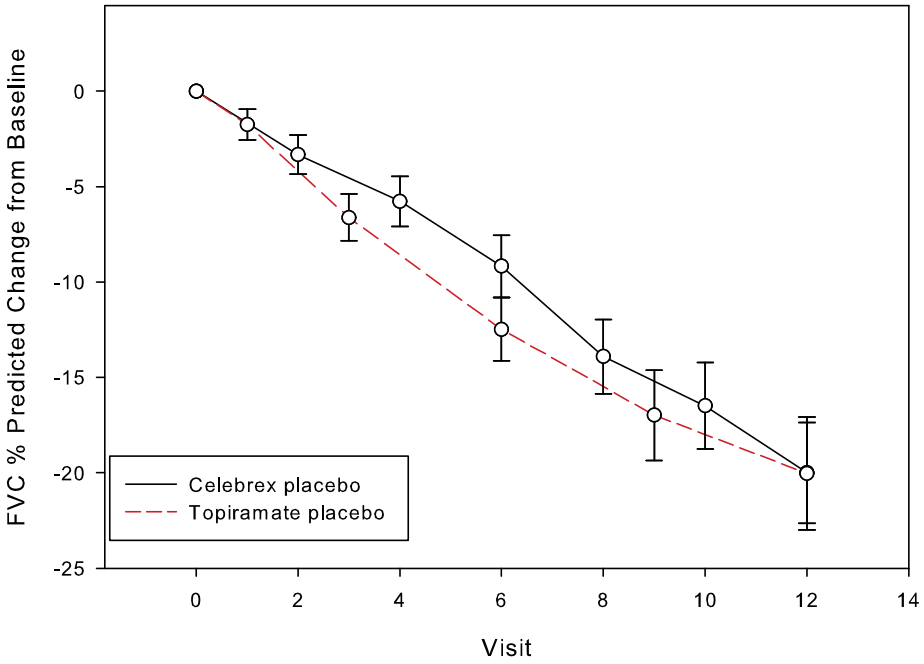

FIG. 4. Mean (± SE) change from baseline in percent predicted forced vital capacity (FVC) at each visit for placebo groups. (Northeast Amyotrophic Lateral Sclerosis Consortium [NEALS] trials conducted from 1999 to 2006.)

derstanding the biological actions of riluzole and the mechanisms by which it improves survival in ALS could help develop better treatments. Development of more potent riluzole analogs should be considered.

There needs to be more extensive preclinical testing of therapies, including understanding dosage, drug interactions, mechanisms of action, and pharmacodynamic markers. Focused, early phase II studies defining dosage, pharmacodynamics, and drug interactions will improve the likelihood of success for phase III trials.

Additionally, understand barriers to a subject's participation and retention in studies will improve study conduct and speed development of better treatments. A more sensitive ALS outcome measure that can detect small but

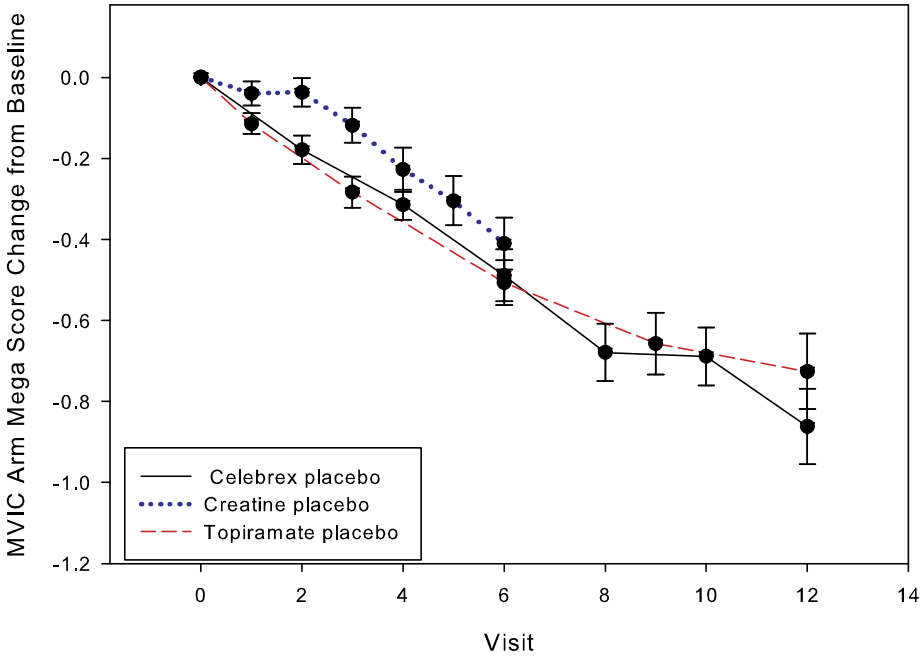

FIG. 5. Mean (± SE) change from baseline in maximum voluntary isometric contraction (MVIC) arm mega score total at each visit for placebo groups. (Northeast Amyotrophic Lateral Sclerosis Consortium [NEALS] trials conducted from 1999 to 2006.)

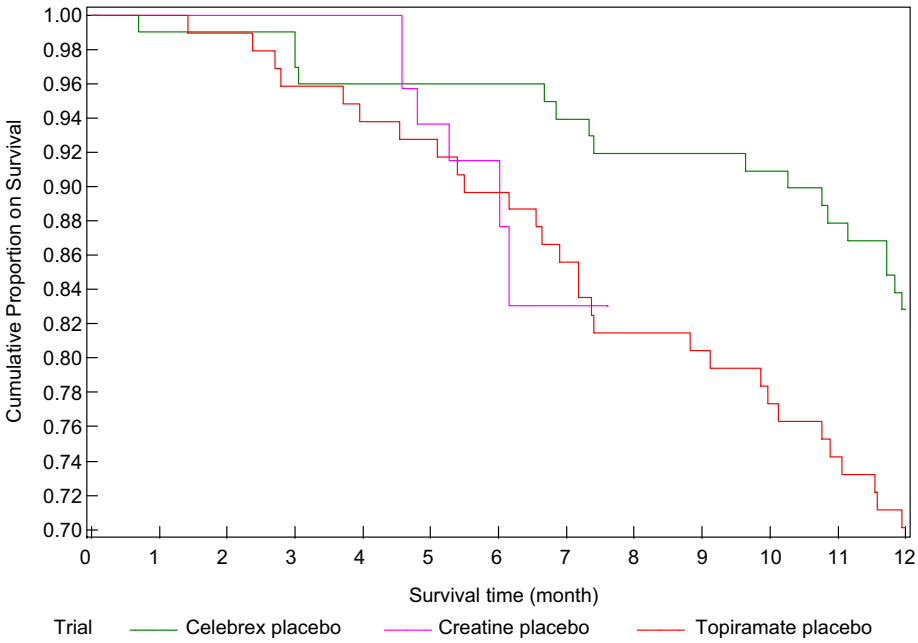

FIG. 6. Kaplan-Meier survival probabilities through 12 months for placebo groups (Northeast Amyotrophic Lateral Sclerosis Consortium [NEALS] trials conducted from 1999 to 2006). The participants in topiramate and creatine trials had forced vital capacity ≥ 50%, whereas participants in the Celebrex trial had forced vital capacity ≥ 60% predicted.

important responses could revolutionize therapy development.

CONCLUSION

During the past decade, significant progress has been made in our understanding of the pathogenesis of ALS. We have gained tremendous experience in trials in terms of trained sites, biostatisticians, academic coordination centers, natural history datasets, and large repositories of well-phenotyped samples. However, there remains an unmet need to find treatments that effectively slow the progression of ALS or cure it completely. Preclinical studies that help identify dosage, pathways, and pharmacodynamic markers will allow design of better early phase II studies in humans. These will result in better-designed phase III studies. To accomplish this, more investment is needed by industry, foundations, and government funding agencies in early therapy development in animals and humans. With coordinated effort combined with increased knowledge of the disease pathology and the therapeutic challenges, effective therapies are now, more than ever, within reach.

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