Amyotrophic lateral sclerosis: new genes, new models, and new mechanisms

Christine Vande Velde[1,3]*, Patrick A. Dion[2,3] and Guy A. Rouleau[1,3]

Addresses: [1]Department of Medicine, Université de Montréal, 1560 Sherbrooke East, Montreal, Canada, H2L 4W1; [2]Department of Pathology and Cell Biology, Université de Montréal, 1560 Sherbrooke East, Montreal, Canada, H2L 4W1; [3]CHUM Research Centre, 1560 Sherbrooke East, Montreal, Canada, H2L 4W1

* Corresponding author: Christine Vande Velde (c.vande.velde@umontreal.ca)

The electronic version of this article is the complete one and can be found at: http://f1000.com/reports/b/3/18

Abstract

Research aimed at understanding amyotrophic lateral sclerosis (ALS) has seen exceptional growth in the past few years. New genes, new models, and new mechanisms have not only improved our understanding, but also contributed to the increasing complexity of ALS pathogenesis. The focus of this piece is to highlight some of the more notable developments in the field and to encourage a re-appreciation for the superoxide dismutase 1 (SOD1) mouse models.

Introduction

In 1869, the French neurologist Jean-Martin Charcot described a fatal and progressive paralysis known as amyotrophic lateral sclerosis (ALS). This devastating neurodegenerative disease, also known as Lou Gehrig's disease (USA) or motor neuron disease (UK), has an incidence of 2–3 cases per 100,000 annually. In the United States, 25,000 patients are predicted to be affected at any one time, with only 50% of patients surviving 2–3 years after diagnosis. Patients experience muscle weakness and atrophy, leading to inevitable paralysis, due to the selective and progressive loss of motor neurons of the brain and spinal cord that is characteristic of the disease [1,2]. Most often, patients succumb to the disease because of denervation of muscles involved in respiration. Despite decades of research, we have yet to identify the "silver bullet" to cure, or even slow, the disease. There is only one therapeutic option for patients, riluzole, which offers a modest 3- to 6-month extension in survival [3,4]. The current lack of effective therapeutic options for patients is due, in part, to the general lack of understanding of how the disease initiates and progresses. However, in the past few years, there have been a significant number of advances that are expected to shed new light on this disease.

A total of 10% of all ALS cases are genetically inherited, while the remaining 90% have no known cause [1]. Of the familial cases, mutations in the gene encoding superoxide dismutase 1 (SOD1), one of the three superoxide dismutases responsible for destroying free superoxide radicals in the body, first described in 1993, are the most common [5]. So, it is not surprising that the bulk of research has focused on examining the consequences of pathogenic SOD1 mutations in a variety of models. Some researchers believe, maybe too cynically, that the inability to find an effective therapy is because the field relies too heavily on SOD1 mouse models; this cynicism stems from the fact that only a subset of ALS cases are due to mutations in this particular gene. Regardless, using SOD1 animal models, scientists have pushed the field forward by dissecting out a variety of molecular events in affected motor neurons [6].

New genes

In the past 3 years, two novel genes have emerged on the scene, bringing with them a new, previously little-considered mechanism of RNA metabolism and processing. The protein products of these new genes, TARDBP (encoding TAR DNA binding protein-43, or TDP-43) and FUS (also known as TLS, which encodes RNA-binding protein FUS), are known to participate in a multitude of

functions [7]. While these two genes appear (at first blush) to implicate very different mechanisms than those identified using the *SOD1* models, much work remains to be done to define the pathogenic mechanism(s) affected by these mutations. In the meantime, there has been a veritable stampede in the development of animal models to investigate these new genes. How, and if, these different genetic causes converge on the selective killing of motor neurons in ALS, yielding a more or less clinically indistinguishable phenotype, is an important question in the field. Identifying the common link between them is a major research focus as this intersection would be expected to be an ideal therapeutic target. Mutations in a third gene, *OPTN* (which encodes the protein optineurin, reported to be involved in protein trafficking, autophagy, and gene expression) are known to be causative for primary open-angle glaucoma, and have also been recently implicated in ALS. While the role for *OPTN* in ALS is not yet clear, several mutations have been found in both Japanese and Italian ALS patients, the latter cohort including both familial and sporadic cases [8-11]. For *TDP-43* and *FUS*, their discoveries have raised the very real possibility that RNA metabolism and processing is a central feature in neurodegeneration, including ALS. Of course, RNA metabolism and processing is a broad term that implies a wide range of cellular activities. To some extent, however, the field is expecting to see additional RNA-binding proteins surface in ALS, but the negative result of a recent mutational analysis for a *FUS/TLS* family member, *TAF15*, has dampened such expectations [12]. On the other hand, defects in the gene encoding the RNA-binding protein SMN1 (survival of motor neuron 1) have long been known in spinal muscular atrophy, a juvenile disease that is characterized by the loss of spinal cord motor neurons in young children [13].

In the case of *TDP-43* mutations, a recent major advance is the identification of the target transcripts bound by TDP-43 [14-17]. However, despite the plethora of potential TDP-43 regulatable transcripts that these approaches have yielded, the field is a long way from understanding the biology. And surprisingly, despite widespread prediction, as yet there has been no convincing demonstration that either *TDP-43* or *FUS* mutants alter the splicing pattern of any known transcripts. However, both have been localized to, and implicated in, the formation and regulation of stress granules, which is a normal cellular response to exogenous stress stimuli [18-24], and there is significant speculation that this pathway, not previously implicated in ALS, may be a factor in the disease. Another piece of evidence for this comes from studies showing that recognized markers of stress granules colocalize with

TDP-43 and FUS inclusions in postmortem patient material [20,25].

Likely one of the most exciting recent developments in ALS research is the identification of genes that can modify aspects of the disease. While disease gene modifiers, alleles/genes that either are associated with a higher than normal risk of developing the disease in the population or modify the onset and/or duration of the disease, have been reported in other disease contexts, few such examples were known to exist for ALS. Some years back, an association was reported between a marker influencing *KIFAP3* expression and patient survival; KIFAP3 (kinesin-associated protein 3) participates with the motor protein kinesin in the movement of organelles and molecules towards the neuromuscular junction [26]. However, an independent follow-up study did not confirm this association [27]. Conversely and most recently, *ATXN2* (encoding ataxin-2), known to cause spinocerebellar ataxia type 2, was linked to ALS in several different studies [28-32]. Specifically, intermediate length expansions of ataxin-2's polyglutamine repeats (27-33 glutamines) are found in ALS patients. Intriguingly, the possibility that this locus could be implicated in ALS was presented 3 years ago using a very different genetic approach [33]. What is exciting here is that the recent emergence of *ATXN2* is akin to the identification of *APOE* (encoding apolipoprotein E) allele ε4, which confers an increased risk in Alzheimer's disease [34]. And as we have seen for *APOE* in Alzheimer's disease, the biology behind the *ATXN2* association in ALS is certain to be an area of intense investigation.

The implication of *ATXN2* in ALS, as well as in spinocerebellar ataxia type 2, highlights the recent general acknowledgement that there is genetic overlap between neurodegenerative diseases. Most recently, mutations in the gene encoding valosin-containing protein (*VCP*), previously identified as causative for both frontotemporal dementia [35] and the rare syndrome IBMPFD (inclusion body myopathy with early-onset Paget disease of the bone and frontotemporal dementia) [36], were described in a cohort of ALS patients using exome sequencing [37]. The idea that ALS and frontotemporal dementia might share some common pathways was raised in 2006, when TDP-43 was identified as a major component of neuronal aggregates that figure significantly in the pathology of patients with ALS or frontotemporal dementia [38] and, clinically, the association between ALS and frontotemporal dementia has been recognized for more than a decade [39]. This overlap was further reinforced by the identification of *TDP-43* mutations in frontotemporal dementia patients without ALS features [40,41]. Thus, it is now widely

believed that ALS and frontotemporal dementia are part of a disease spectrum. It is evident that genes such as *ALS2*, *SETX*, and *VAPB*, which are thought to cause familial forms of ALS, may be allelic for, or implicated in, other neurological diseases. As our genetic understanding of ALS advances, what has become very apparent is that *SOD1* mutations have never been reported in any other disease. So is the SOD1 mouse model really so bad? Wouldn't a purely ALS-causing gene be the ideal choice with which to dissect mechanism(s) and develop therapeutics?

New models and mechanisms

This brings us back to the utility of the SOD1 mouse models. As alluded to earlier, these models have uncovered several biological mechanisms thought to actively participate in the selective motor neuron degeneration characteristic of ALS (please see [6] for a recent comprehensive review). Of these possibilities, there has been a considerable focus on mitochondria, specialized organelles that are primarily responsible for meeting the energy demands of the power-hungry motor neurons, and how SOD1 might affect their normal function. This focus on mitochondria is due in large part to the observation that SOD1 preferentially associates with mitochondria from affected spinal cords but less so, if at all, with mitochondria from other tissues [42-44]. This work has developed in parallel with a series of groups developing tools designed to specifically recognize a disease-specific conformation of SOD1 [45-47]. Using these tools, it is now recognized that a misfolded form of SOD1 does exist and it is found to be bound to the mitochondrial surface [44,48]. Thus, it fits well that further investigation has found that mitochondrial functions at the surface, such as protein import [49] and metabolite conductance [48], are disrupted by the presence of misfolded SOD1. Other aspects of mitochondrial function may also be affected, as significant changes in axonal mitochondrial morphology have recently been reported in two different mutant SOD1 models [50]. What remains to be clarified is the sequence of these events in disease development. Regardless, it is significant (and exciting) that a misfolded form of SOD1 can be detected in the motor neurons of some sporadic ALS patients who lack a genetic mutation in *SOD1* [47,51]. One possible explanation for this may come from recent studies showing that the wild type SOD1 protein can become mutant-like *in vitro* through oxidation [52,53]. Together, these findings firmly link SOD1 to sporadic disease and reinforce the utility of the SOD1 animal models.

However, it is surely true that other genes might exist that modify the disease in humans. Using the power of mouse genetics, a recent report using the well-characterized ALS model of mice expressing *SOD1* with a glycine to alanine mutation (G93A) bred into multiple different genetic backgrounds yielded several disease modifying loci [54]. The timing of disease onset and the progression/duration of the disease was variable in the different backgrounds. While much additional work needs to be done, an exciting list of candidate genes will undoubtedly emerge from these studies.

The ultimate goal of every ALS research team is to define how the disease starts and progresses so that we can craft a therapeutic to halt the disease. To do this, we need to have confidence in our animal models. While the *TARDBP* and *FUS/TLS* mice are ways away from demonstrating their true utility in this regard, this is not true for the SOD1 models. In fact, of all the neurodegenerative disease models, the mutant SOD1 mice are some of the most faithful mimics of human disease. So why have the dozens (if not hundreds) of clinical trials in ALS not yet yielded our "silver bullet"? Is it poor preclinical trial design? Unrealistic expectations/ outcome measures for patients in clinical trials? Both of these have recently been addressed by the field in the revamped way SOD1 mouse models are to be used in the preclinical evaluation of potential therapeutics. This is summarized nicely in the recently developed ALS mouse handbook [55]. The implementation of these consensus guidelines should hopefully avoid erroneous interpretations and thus rationalize the choice of therapeutics pushed forward into Phase I/II human trials. At the patient level, there has been a significant paradigm shift with regards to trial design. Improvements to patient quality of life are now considered to be more important and/or indicative of success than time of survival. Such new designs are also more attractive to potential patients enrolling in the trial [56].

Conclusion

In a relatively short period of time, the ALS field has acquired a remarkable amount of information about the potential cellular mechanisms underlying the disease. The challenge lying ahead is how to integrate and assimilate the recent and emerging data to identify common mechanisms as these will likely represent the best potential targets for future therapies. However, let's suppose for a moment that we don't get that. What if we find a handful of targets without finding the unifier? Perhaps we should start the discussion now about personalized patient care? Given the stochastic nature of ALS, the variability in age and site of onset, the inherited and noninherited forms, perhaps it is naïve to think that a "one size fits all" approach will benefit every ALS patient. Perhaps customized, combination therapies

should be our goal? Maybe we should be thinking in terms of finding the "silver bullets", not "bullet".

Abbreviations

ALS, amyotrophic lateral sclerosis; ALS2, alsin; APOE, apolipoprotein E; ATXN2, ataxin-2; FUS/TLS, fused in sarcoma/translocated in liposarcoma; KIFAP3, kinesin-associated protein 3; OPTN, optineurin; SETX, senataxin; SOD1, superoxide dismutase 1; TARDBP and TDP-43, TAR DNA binding protein-43; VAPB, vesicle-associated membrane protein-associated protein B..

Competing interests

The authors declare that they have no competing interests.

References

1. Dion PA, Daoud H, Rouleau GA: **Genetics of motor neuron disorders: new insights into pathogenic mechanisms.** *Nat Rev Genet* 2009, **10**:769-82.

2. Boillee S, Vande Velde C, Cleveland DW: **ALS: A disease of motor neurons and their nonneuronal neighbors.** *Neuron* 2006, **52**:1-21.

3. Lacomblez L, Bensimon G, Leigh PN, Guillet P, Meininger V: **Dose-ranging study of riluzole in amyotrophic lateral sclerosis. Amyotrophic Lateral Sclerosis/Riluzole Study Group II.** *Lancet* 1996, **347**:1425-31.

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

4. Bensimon G, Lacomblez L, Meininger V: **A controlled trial of riluzole in amyotrophic lateral sclerosis. ALS/Riluzole Study Group.** *N Engl J Med* 1994, **330**:585-91.

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

5. Rosen DR, Siddique T, Patterson D, Figlewicz DA, Sapp P, Hentati A, Donaldson D, Goto J, O'Regan JP, Deng HX: **Mutations in Cu/Zn superoxide dismutase gene are associated with familial amyotrophic lateral sclerosis.** *Nature* 1993, **362**:59-62.

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

6. Ilieva H, Polymenidou M, Cleveland DW: **Non-cell autonomous toxicity in neurodegenerative disorders: ALS and beyond.** *J Cell Biol* 2009, **187**:761-72.

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

7. Lagier-Tourenne C and Cleveland DW: **Rethinking ALS: the FUS about TDP-43.** *Cell* 2009, **136**:1001-4.

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

8. Maruyama H, Morino H, Ito H, Izumi Y, Kato H, Watanabe Y, Kinoshita Y, Kamada M, Nodera H, Suzuki H, Komure O, Matsuura S, Kobatake K, Morimoto N, Abe K, Suzuki N, Aoki M, Kawata A, Hirai T, Kato T, Ogasawara K, Hirano A, Takumi T, Kusaka H, Hagiwara K, Kaji R, Kawakami H: **Mutations of optineurin in amyotrophic lateral sclerosis.** *Nature* 2010, **465**:223-6.

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

9. Millecamps S, Boillee S, Chabrol E, Camu W, Cazeneuve C, Salachas F, Pradat PF, Danel-Brunaud V, Vandenberghe N, Corcia P, Le FN, Lacomblez L, Bruneteau G, Seilhean D, Brice A, Feingold J, Meininger V, Leguern E: **Screening of OPTN in French familial amyotrophic lateral sclerosis.** *Neurobiol Aging* 2011, **32**:557-3.

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

10. Sugihara K, Maruyama H, Kamada M, Morino H, Kawakami H: **Screening for OPTN mutations in amyotrophic lateral sclerosis in a mainly Caucasian population.** *Neurobiol Aging* 2011, **32**:1923.e9-10.

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

11. Del BR, Tiloca C, Pensato V, Corrado L, Ratti A, Ticozzi N, Corti S, Castellotti B, Mazzini L, Soraru G, Cereda C, D'Alfonso S, Gellera C, Comi GP, Silani V: **Novel optineurin mutations in patients with familial and sporadic amyotrophic lateral sclerosis.** *J Neurol Neurosurg Psychiatry* 2011, [Epub ahead of print].

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

12. Ticozzi N, Vance C, Leclerc AL, Keagle P, Glass JD, McKenna-Yasek D, Sapp PC, Silani V, Bosco DA, Shaw CE, Brown RH Jr., Landers JE: **Mutational analysis reveals the FUS homolog TAF15 as a candidate gene for familial amyotrophic lateral sclerosis.** *Am J Med Genet B Neuropsychiatr Genet* 2011, **156**:285-90.

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

13. Lefebvre S, Burglen L, Reboullet S, Clermont O, Burlet P, Viollet L, Benichou B, Cruaud C, Millasseau P, Zeviani M: **Identification and characterization of a spinal muscular atrophy-determining gene.** *Cell* 1995, **80**:155-65.

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

14. Sephton CF, Cenik C, Kucukural A, Dammer EB, Cenik B, Han Y, Dewey CM, Roth FP, Herz J, Peng J, Moore MJ, Yu G: **Identification of neuronal RNA targets of TDP-43-containing ribonucleo-protein complexes.** *J Biol Chem* 2011, **286**:1204-15.

    F1000 Factor 8
    Evaluated by Guy Rouleau 17 Aug 2011

15. Tollervey JR, Curk T, Rogelj B, Briese M, Cereda M, Kayikci M, Konig J, Hortobagyi T, Nishimura AL, Zupunski V, Patani R, Chandran S, Rot G, Zupan B, Shaw CE, Ule J: **Characterizing the RNA targets and position-dependent splicing regulation by TDP-43.** *Nat Neurosci* 2011, **14**:452-8.

    F1000 Factor 8
    Evaluated by Guy Rouleau 17 Aug 2011

16. Polymenidou M, Lagier-Tourenne C, Hutt KR, Huelga SC, Moran J, Liang TY, Ling SC, Sun E, Wancewicz E, Mazur C, Kordasiewicz H, Sedaghat Y, Donohue JP, Shiue L, Bennett CF, Yeo GW, Cleveland DW: **Long pre-mRNA depletion and RNA**

missplicing contribute to neuronal vulnerability from loss of TDP-43. *Nat Neurosci* 2011, **14**:459-68.

> F1000 Factor 9
> Evaluated by Valina Dawson 04 May 2011, Guy Rouleau 17 Aug 2011

17. Xiao S, Sanelli T, Dib S, Sheps D, Findlater J, Bilbao J, Keith J, Zinman L, Rogaeva E, Robertson J: **RNA targets of TDP-43 identified by UV-CLIP are deregulated in ALS.** *Mol Cell Neurosci* 2011, **47**:167-80.

> F1000 Factor 8
> Evaluated by Guy Rouleau 17 Aug 2011

18. McDonald KK, Aulas A, Destroismaisons L, Pickles S, Beleac E, Camu W, Rouleau GA, Vande Velde C: **TAR DNA-binding protein 43 (TDP-43) regulates stress granule dynamics via differential regulation of G3BP and TIA-1.** *Hum Mol Genet* 2011, **20**:1400-10.

19. Freibaum BD, Chitta RK, High AA, Taylor JP: **Global analysis of TDP-43 interacting proteins reveals strong association with RNA splicing and translation machinery.** *J Proteome Res* 2010, **9**:1104-20.

> F1000 Factor 6
> Evaluated by Guy Rouleau 17 Aug 2011

20. Liu-Yesucevitz L, Bilgutay A, Zhang YJ, Vanderwyde T, Citro A, Mehta T, Zaarur N, McKee A, Bowser R, Sherman M, Petrucelli L, Wolozin B: **Tar DNA binding protein-43 (TDP-43) associates with stress granules: analysis of cultured cells and pathological brain tissue.** *PLoS One* 2010, **5**:e13250.

> F1000 Factor 6
> Evaluated by Guy Rouleau 17 Aug 2011

21. Colombrita C, Zennaro E, Fallini C, Weber M, Sommacal A, Buratti E, Silani V, Ratti A: **TDP-43 is recruited to stress granules in conditions of oxidative insult.** *J Neurochem* 2009, **111**:1051-61.

> F1000 Factor 6
> Evaluated by Guy Rouleau 17 Aug 2011

22. Bosco DA, Lemay N, Ko HK, Zhou H, Burke C, Kwiatkowski TJ Jr., Sapp P, McKenna-Yasek D, Brown RH Jr., Hayward LJ: **Mutant FUS proteins that cause amyotrophic lateral sclerosis incorporate into stress granules.** *Hum Mol Genet* 2010, **19**:4160-75.

> F1000 Factor 6
> Evaluated by Guy Rouleau 17 Aug 2011

23. Dormann D, Rodde R, Edbauer D, Bentmann E, Fischer I, Hruscha A, Than ME, Mackenzie IR, Capell A, Schmid B, Neumann M, Haass C: **ALS-associated fused in sarcoma (FUS) mutations disrupt Transportin-mediated nuclear import.** *EMBO J* 2010, **29**:2841-57.

> F1000 Factor 7
> Evaluated by Yuh Min Chook 09 Nov 2010, Guy Rouleau 17 Aug 2011

24. Gal J, Zhang J, Kwinter DM, Zhai J, Jia H, Jia J, Zhu H: **Nuclear localization sequence of FUS and induction of stress granules by ALS mutants.** *Neurobiol Aging* 2010, [Epub ahead of print].

> F1000 Factor 6
> Evaluated by Guy Rouleau 17 Aug 2011

25. Moisse K, Volkening K, Leystra-Lantz C, Welch I, Hill T, Strong MJ: **Divergent patterns of cytosolic TDP-43 and neuronal progranulin expression following axotomy: implications for TDP-43 in the physiological response to neuronal injury.** *Brain Res* 2009, **1249**:202-11.

> F1000 Factor 6
> Evaluated by Guy Rouleau 17 Aug 2011

26. Landers JE, Melki J, Meininger V, Glass JD, van den Berg LH, van Es MA, Sapp PC, van Vught PW, McKenna-Yasek DM, Blauw HM, Cho TJ, Polak M, Shi L, Wills AM, Broom WJ, Ticozzi N, Silani V, Ozoguz A, Rodriguez-Leyva I, Veldink JH, Ivinson AJ, Saris CG, Hosler BA, Barnes-Nessa A, Couture N, Wokke JH, Kwiatkowski TJ Jr., Ophoff RA, Cronin S, Hardiman O, et al.: **Reduced expression of the Kinesin-Associated Protein 3 (KIFAP3) gene increases survival in sporadic amyotrophic lateral sclerosis.** *Proc Natl Acad Sci U S A* 2009, **106**:9004-9.

> F1000 Factor 11
> Evaluated by Stanley Appel 09 Jun 2009, Guy Rouleau 17 Aug 2011

27. Traynor BJ, Nalls M, Lai SL, Gibbs RJ, Schymick JC, Arepalli S, Hernandez D, van der Brug MP, Johnson JO, Dillman A, Cookson M, Moglia C, Calvo A, Restagno G, Mora G, Chio A: **Kinesin-associated protein 3 (KIFAP3) has no effect on survival in a population-based cohort of ALS patients.** *Proc Natl Acad Sci U S A* 2010, **107**:12335-8.

> F1000 Factor 6
> Evaluated by Guy Rouleau 17 Aug 2011

28. Lee T, Li YR, Ingre C, Weber M, Grehl T, Gredal O, de CM, Meyer T, Tysnes OB, Auburger G, Gispert S, Bonini NM, Andersen PM, Gitler AD: **Ataxin-2 intermediate-length polyglutamine expansions in European ALS patients.** *Hum Mol Genet* 2011, **20**:1697-700.

> F1000 Factor 6
> Evaluated by Guy Rouleau 17 Aug 2011

29. Elden AC, Kim HJ, Hart MP, Chen-Plotkin AS, Johnson BS, Fang X, Armakola M, Geser F, Greene R, Lu MM, Padmanabhan A, Clay-Falcone D, McCluskey L, Elman L, Juhr D, Gruber PJ, Rub U, Auburger G, Trojanowski JQ, Lee VM, Van Deerlin VM, Bonini NM, Gitler AD: **Ataxin-2 intermediate-length polyglutamine expansions are associated with increased risk for ALS.** *Nature* 2010, **466**:1069-75.

> F1000 Factor 6
> Evaluated by Guy Rouleau 17 Aug 2011

30. Yu Z, Zhu Y, Chen-Plotkin AS, Clay-Falcone D, McCluskey L, Elman L, Kalb RG, Trojanowski JQ, Lee VM, Van Deerlin VM, Gitler AD, Bonini NM: **PolyQ Repeat Expansions in ATXN2 Associated with ALS Are CAA Interrupted Repeats.** *PLoS One* 2011, **6**:e17951.

> F1000 Factor 6
> Evaluated by Guy Rouleau 17 Aug 2011

31. Ross OA, Rutherford NJ, Baker M, Soto-Ortolaza AI, Carrasquillo MM, Dejesus-Hernandez M, Adamson J, Li M, Volkening K, Finger E, Seeley WW, Hatanpaa KJ, Lomen-Hoerth C, Kertesz A, Bigio EH, Lippa C, Woodruff BK, Knopman DS, White CL III, Van Gerpen JA, Meschia JF, Mackenzie IR, Boylan K, Boeve BF, Miller BL, Strong MJ, Uitti RJ, Younkin SG, Graff-Radford NR, et al.: **Ataxin-2 repeat-length variation and neurodegeneration.** *Hum Mol Genet* 2011, **20**:3207-12.

> F1000 Factor 6
> Evaluated by Guy Rouleau 17 Aug 2011

32. Daoud H, Belzil V, Martins S, Sabbagh M, Provencher P, Lacomblez L, Meininger V, Camu W, Dupre N, Dion PA, Rouleau GA: **Association of Long ATXN2 CAG Repeat Sizes With Increased Risk of Amyotrophic Lateral Sclerosis.** *Arch Neurol* 2011, **68**:739-42.

33. Cronin S, Blauw HM, Veldink JH, van Es MA, Ophoff RA, Bradley DG, van den Berg LH, Hardiman O: **Analysis of genome-wide copy number variation in Irish and Dutch ALS populations.** *Hum Mol Genet* 2008, **17**:3392-8.

> F1000 Factor 6
> Evaluated by Guy Rouleau 17 Aug 2011

34. Strittmatter WJ, Saunders AM, Schmechel D, Pericak-Vance M, Enghild J, Salvesen GS, Roses AD: **Apolipoprotein E: high-avidity binding to beta-amyloid and increased frequency of type 4 allele in late-onset familial Alzheimer disease.** *Proc Natl Acad Sci U S A* 1993, **90**:1977-81.

35. Forman MS, Mackenzie IR, Cairns NJ, Swanson E, Boyer PJ, Drachman DA, Jhaveri BS, Karlawish JH, Pestronk A, Smith TW, Tu PH, Watts GD, Markesbery WR, Smith CD, Kimonis VE: **Novel ubiquitin neuropathology in frontotemporal dementia with valosin-containing protein gene mutations.** *J Neuropathol Exp Neurol* 2006, **65:**571-81.

36. Watts GD, Wymer J, Kovach MJ, Mehta SG, Mumm S, Darvish D, Pestronk A, Whyte MP, Kimonis VE: **Inclusion body myopathy associated with Paget disease of bone and frontotemporal dementia is caused by mutant valosin-containing protein.** *Nat Genet* 2004, **36:**377-81.

37. Johnson JO, Mandrioli J, Benatar M, Abramzon Y, Van Deerlin VM, Trojanowski JQ, Gibbs JR, Brunetti M, Gronka S, Wuu J, Ding J, McCluskey L, Martinez-Lage M, Falcone D, Hernandez DG, Arepalli S, Chong S, Schymick JC, Rothstein J, Landi F, Wang YD, Calvo A, Mora G, Sabatelli M, Monsurro MR, Battistini S, Salvi F, Spataro R, Sola P, Borghero G, et al.: **Exome sequencing reveals VCP mutations as a cause of familial ALS.** *Neuron* 2010, **68:**857-64.

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

38. Neumann M, Sampathu DM, Kwong LK, Truax AC, Micsenyi MC, Chou TT, Bruce J, Schuck T, Grossman M, Clark CM, McCluskey LF, Miller BL, Masliah E, Mackenzie IR, Feldman H, Feiden W, Kretzschmar HA, Trojanowski JQ, Lee VM: **Ubiquitinated TDP-43 in frontotemporal lobar degeneration and amyotrophic lateral sclerosis.** *Science* 2006, **314:**130-33.

    F1000 Factor 11
    Evaluated by Stanley Appel 13 Feb 2007, Guy Rouleau 17 Aug 2011

39. Neary D, Snowden JS, Mann DM: **Cognitive change in motor neurone disease/amyotrophic lateral sclerosis (MND/ALS).** *J Neurol Sci* 2000, **180:**15-20.

40. Borroni B, Bonvicini C, Alberici A, Buratti E, Agosti C, Archetti S, Papetti A, Stuani C, Di LM, Gennarelli M, Padovani A: **Mutation within TARDBP leads to frontotemporal dementia without motor neuron disease.** *Hum Mutat* 2009, **30:**E974-83.

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

41. Borroni B, Archetti S, Del BR, Papetti A, Buratti E, Bonvicini C, Agosti C, Cosseddu M, Turla M, Di LD, Pietro CG, Gennarelli M, Padovani A: **TARDBP mutations in frontotemporal lobar degeneration: frequency, clinical features, and disease course.** *Rejuvenation Res* 2010, **13:**509-17.

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

42. Liu J, Lillo C, Jonsson PA, Vande Velde C, Ward CM, Miller TM, Subramaniam JR, Rothstein JD, Marklund S, Andersen PM, Brannstrom T, Gredal O, Wong PC, Williams DS, Cleveland DW: **Toxicity of Familial ALS-Linked SOD1 Mutants from Selective Recruitment to Spinal Mitochondria.** *Neuron* 2004, **43:**5-17.

    F1000 Factor 6
    Evaluated by Albert La Spada 17 Nov 2004

43. Pasinelli P, Belford ME, Lennon N, Bacskai BJ, Hyman BT, Trotti D, Brown RH, Jr.: **Amyotrophic Lateral Sclerosis-Associated SOD1 Mutant Proteins Bind and Aggregate with Bcl-2 in Spinal Cord Mitochondria.** *Neuron* 2004, **43:**19-30.

    F1000 Factor 7
    Evaluated by Albert La Spada 17 Nov 2004, Guy Rouleau 17 Aug 2011

44. Vande Velde C, Miller TM, Cashman NR, Cleveland DW: **Selective association of misfolded ALS-linked mutant SOD1 with the cytoplasmic face of mitochondria.** *Proc Natl Acad Sci U S A* 2008, **105:**4022-7.

45. Rakhit R, Robertson J, Vande Velde C, Horne P, Ruth DM, Griffin J, Cleveland DW, Cashman NR, Chakrabartty A: **An immunological epitope selective for pathological monomer-misfolded SOD1 in ALS.** *Nat Med* 2007, **13:**754-9.

    F1000 Factor 8
    Evaluated by Nereo Bresolin 20 Jun 2007, Guy Rouleau 17 Aug 2011

46. Gros-Louis F, Soucy G, Lariviere R, Julien JP: **Intracerebroventricular infusion of monoclonal antibody or its derived Fab fragment against misfolded forms of SOD1 mutant delays mortality in a mouse model of ALS.** *J Neurochem* 2010, **113:**1188-99.

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

47. Forsberg K, Jonsson PA, Andersen PM, Bergemalm D, Graffmo KS, Hultdin M, Jacobsson J, Rosquist R, Marklund SL, Brannstrom T: **Novel antibodies reveal inclusions containing non-native SOD1 in sporadic ALS patients.** *PLoS One* 2010, **5:**e11552.

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

48. Israelson A, Arbel N, Da Cruz S, Ilieva H, Yamanaka K, Shoshan-Barmatz V, Cleveland DW: **Misfolded mutant SOD1 directly inhibits VDAC1 conductance in a mouse model of inherited ALS.** *Neuron* 2010, **67:**575-87.

    F1000 Factor 6
    Evaluated by Guy Rouleau 17 Aug 2011

49. Li Q, Vande Velde C, Israelson A, Xie J, Bailey AO, Dong MQ, Chun SJ, Roy T, Winer L, Yates JR, Capaldi RA, Cleveland DW, Miller TM: **ALS-linked mutant superoxide dismutase 1 (SOD1) alters mitochondrial protein composition and decreases protein import.** *Proc Natl Acad Sci U S A* 2010, **107:**21146-51.

    F1000 Factor 6
    Evaluated by Nereo Bresolin 15 Dec 2010

50. Vande Velde C, McDonald KK, Boukhedimi Y, McAlonis-Downes M, Lobsiger CS, Bel Hadj S, Zandona A, Julien JP, Shah SB, Cleveland DW: **Misfolded SOD1 associated with motor neuron mitochondria alters mitochondrial shape and distribution prior to clinical onset.** *PLoS One* 2011, **6:**e22031.

51. Bosco DA, Morfini G, Karabacak NM, Song Y, Gros-Louis F, Pasinelli P, Goolsby H, Fontaine BA, Lemay N, McKenna-Yasek D, Frosch MP, Agar JN, Julien JP, Brady ST, Brown RH, Jr.: **Wild-type and mutant SOD1 share an aberrant conformation and a common pathogenic pathway in ALS.** *Nat Neurosci* 2010, **13:**1396-403.

    F1000 Factor 9
    Evaluated by Stanley Appel 19 Nov 2010, Guy Rouleau 17 Aug 2011

52. Banci L, Bertini I, Boca M, Girotto S, Martinelli M, Valentine JS, Vieru M: **SOD1 and amyotrophic lateral sclerosis: mutations and oligomerization.** *PLoS One* 2008, **3:**e1677.

53. Ezzi SA, Urushitani M, Julien JP: **Wild-type superoxide dismutase acquires binding and toxic properties of ALS-linked mutant forms through oxidation.** *J Neurochem* 2007, **102:**170-8.

54. Heiman-Patterson TD, Sher RB, Blankenhorn EA, Alexander G, Deitch JS, Kunst CB, Maragakis N, Cox G: **Effect of genetic background on phenotype variability in transgenic mouse models of amyotrophic lateral sclerosis: a window of opportunity in the search for genetic modifiers.** *Amyotroph Lateral Scler* 2011, **12:**79-86.

55. **The Jackson Laboratory: 'Manuals'.** [http://jaxmice.jax.org/manual/index.html#als]

56. Zinman L and Cudkowicz M: **Emerging targets and treatments in amyotrophic lateral sclerosis.** *Lancet Neurol* 2011, **10:**481-90.