

The importance of offering early genetic testing in everyone with amyotrophic lateral sclerosis

Amyotrophic lateral sclerosis (ALS) is a fatal and heterogeneous neurodegenerative disease, with only modestly effective therapies currently approved.^{1–4} While the underlying cause of the majority of ALS cases remains unknown, genetic variations linked to ALS have provided a tangible target for therapeutic development.⁵ As such, development of therapies aimed specifically at ALS-causing genes has been rapidly evolving in recent years. With clear evidence demonstrating incidence of ALS-associated pathological variants in isolated cases of ALS, it is becoming increasingly untenable to reserve testing for those with a family history alone.⁶ To ensure that all ALS patients with clinically actionable variants are identified, and can access treatments, there needs to be both broader access to and earlier offering of genetic testing for people newly diagnosed with ALS.

Genetic research has revealed genetic variations in more than 40 genes to be associated with causing ALS.^{7,8} Expansions of chromosome 9 open reading frame 72 (*C9orf72*), and variants in superoxide dismutase 1 (*SOD1*), TANK-binding kinase 1 (*TBK1*), fused in sarcoma (*FUS*), and TAR DNA-binding protein 43 (*TARDBP*) are the most prevalent globally.⁹ Regionally, prevalence varies, with *C9orf72* predominantly affecting European and North American populations, and *SOD1* being the most prevalent genetic variation in many parts of continental Asia.^{10,11} Mendelian dominant inheritance with high penetrance accounts for ~5–10% of ALS, often as familial cases; however, reduced disease penetrance, recessive inheritance, and pleiotropism (particularly for frontotemporal dementia; FTD) appear to be frequent, with pathological variants found in many cases diagnosed with apparently isolated disease (Table 1). In addition, it is expected that even variants with high penetrance will be found in people with no family history of disease¹² and the distinction between familial and apparently sporadic disease is outdated.¹³ *De novo* mutations and risk genes have also been associated with causing isolated ALS.¹⁴

There are currently different definitions of familial ALS versus sporadic ALS in the literature, without consensus within the scientific and clinical communities. From a clinical perspective, these entities are indistinguishable.^{7,15} While causative ALS genetic variants are largely associated with having a family history of the disease, incidence of known variants in the sporadic ALS population is

well reported in the literature (Table 1).^{7,8,16,17} Therefore, with underlying genetic causes being present in both familial ALS and sporadic ALS, as can be seen in Table 1, there is often no clinically useful difference between the two terms.

An underlying genetic cause of disease provides a more tangible target for drug development,¹⁸ and antisense oligonucleotide (ASO) and siRNA therapies have taken centre stage as being first out of the gate. Data from the clinical trial of tofersen, an ASO targeting *SOD1*—causing reduced expression of both the mutant and wild-type protein—were recently released.¹⁹ While the trial failed to meet the primary end point as was anticipated, neurofilament, a marker of neurodegeneration, was reduced, as were *SOD1* protein levels. Encouraging trends in the data indicate a need to treat early, and a clinical trial using tofersen in presymptomatic carriers, ATLAS (NCT04856982), is already underway. This emphasizes an urgent need to proactively address predictive testing in at-risk family members, while recognizing the added considerations regarding benefits and risks, as well as the psychosocial implications. While it took more than 20 years from the discovery of *SOD1* for this novel ASO to reach large scale human trials, the knowledge gained has laid the foundation for significantly decreasing drug development timelines in this area. For example, launch of a first-in-human trial of an ASO targeting the *C9orf72* repeat expansion (NCT03626012) was achieved in less than 7 years from its discovery in 2011.^{20,21} There are now active clinical trials targeting *SOD1* (NCT02623699), *FUS* (NCT04768972), *C9orf72* (NCT03626012, NCT04931862) and *ATXN2* (NCT04768972). While presence of the underlying genetic alteration is a requirement for participation in these clinical trials, presence of a family history is not. Genetically targeted therapies are likely to soon become mainstream in ALS, and confining testing to individuals with a family history is no longer tenable. There are numerous reasons why a family history of the disease could be missed.^{7,22} In particular, smaller family sizes and reduced or age-dependent gene penetrance results in an increased probability of ALS appearing sporadic, furthering the concept that distinguishing familial ALS and sporadic ALS is purely artificial.¹² As such, restricting clinical genetic testing to individuals reporting a family history will inevitably lead to patients with a genetic aetiology being unable to access emerging therapies, and it is imperative that all people living with ALS are offered genetic testing.

Table 1 Reported incidence of some ALS-causing genetic variants in familial and sporadic ALS populations

Gene	Reported incidence	
	Familial ALS	Sporadic ALS
C9orf72 ⁷	40%	7–10%
SOD1 ⁷	20%	2–4%
TDP-43 ⁷	5%	1%
FUS ⁷	4%	<1%
ATXN2 ¹⁵	1%	1%
Total	70%	11–16%

Although numerous genes have been associated with ALS, this table includes those for which therapeutics are currently being developed. It should be noted that frequency of these variants vary greatly from region to region, with some countries having no significant incidence. Our knowledge is biased towards countries where epidemiology and population studies have been conducted, and this information remains unknown for numerous regions globally.

While genetic testing does not contribute to confirming or excluding a diagnosis of ALS, which is highlighted in the recent Gold Coast diagnostic criteria,²³ many current guidelines for clinical management of ALS are outdated or do not address genetic testing at all. The 2009 American Academy of Neurology ALS Practice Parameters,²⁴ the 2020 Canadian Best Practice Recommendations for the Management of ALS,²⁵ and the 2015 UK National Institute for Health and Care Excellence ALS Guideline²⁶ do not discuss genetic testing. The 2007 Clinical Guidelines from the European ALS Consortium Working Group,²⁷ and the 2011 European Federation of Neurological Societies Guidelines on Clinical Management of ALS²⁸ indicate that clinical genetic testing should be reserved for those patients with a family history of the disease or the D90A-SOD1 phenotype. In addition, all of these were published prior to the discovery of the C9orf72 repeat expansion, which confirmed the previously reported notion of phenotypic variance in ALS, furthering that practice of including frontotemporal dementia, and related disorders, in family history enquiries. The lack of guidelines for genetic testing in ALS has led to divergent testing practices globally.^{29–31} Guidelines on genetic testing for ALS are necessary given the scientific complexities, such as variant interpretation, and knowledge translation requirements, regardless of the specialist providing counselling, to ensure an informed decision is being made.

Recent studies have demonstrated that the previously conceived idea that ordering genetic testing on the basis of family history alone results in patients with a monogenetic aetiology to their disease not being identified. A study of apparently sporadic ALS showed that there was an increased risk to relatives because a proportion of people subsequently had a second relative affected.³² A UK study performed clinical genetic testing, using a 44-gene panel, in 100 new ALS diagnoses. Pathogenic variants were identified in 21 patients, with 15 cases meeting inclusion criteria for genetically-targeted therapy trials, despite only seven patients reporting a family history of ALS.³³ An Italian study using whole genome sequencing in a population-based cohort identified clinically significant pathogenic variants in almost 30% of their ALS cases.³⁴ Additionally, 13% of their ALS cases were found to have clinically actionable variants. Furthermore, genome-wide association studies of people with apparently sporadic ALS consistently find Mendelian disease gene variants.³⁵ Finally, studies have concluded that the number of SOD1 and C9orf72 ALS cases should be greater in the sporadic ALS population than in the familial ALS population.^{12,36}

Early identification of patients is also crucial. In familial ALS, it is unclear how much of the observed pathology seen in post-mortem studies is directly due to the gene defect, and how much damage may be attributed to secondary effects, collateral damage and compensatory mechanisms, particularly since the remaining cells are the ones that have survived the disease process so far.³⁷ Studies using transgenic rodent models indicate that the disease process begins a considerable amount of time prior to any physical symptoms. Therefore, targeting the initial upstream pathological mechanism is likely critical in arresting disease progression and limiting widespread pathology.³⁸










Advances in genetic technologies have undoubtedly contributed to identifying new variants.³⁹ Increased testing will contribute valuable data to resources, such as the ClinGen Variant Curation (CGVC), and guidelines, such as the American College of Medical Genetics and Genomics/Association for Molecular Pathology (ACMG/AMP), which can aid in variant interpretation and guide development of therapeutic strategies and precision medicine.

The offer of genetic testing to all people newly diagnosed with ALS does not come without the need for reflection. Appropriate genetic counselling, ensuring an informed decision based on an evaluation of both risks and benefits, and consideration for challenging psychosocial scenarios, such as a variant identified in an individual with no previous family history, need to be addressed. Much like our understanding of genetics in ALS and the advent of genetic therapies, this discussion is an evolution that anticipates a, hopefully, not so distant future when a therapy is approved.

In conclusion, drug development for genetically targeted therapies in ALS is rapidly advancing, and all people with ALS who could potentially benefit from these emerging therapies need to be identified. There is an increasing body of evidence for a genetic component to all ALS. Given that familial ALS and sporadic ALS are clinically indistinguishable, and the lack of relevance for therapeutic access, there is a need to retire this distinction with regards to access to genetic testing, and treat all new ALS diagnoses equal. ALS-associated genetic variations in seemingly isolated ALS are expected, well reported in the literature, and not infrequent, and recent studies have demonstrated that widespread genetic testing identifies patients living with ALS who have clinically actionable variants. All patients diagnosed with ALS, regardless of clinical presentation or family history, need to be offered appropriate genetic counselling, and clinical genetic testing for ALS-associated genetic variations.^{40,41} Genetic testing needs to be offered early to ensure timely access to therapeutic intervention.

Current clinical management guidelines are lacking when it comes to genetic testing in ALS. Recognizing the need to change practices, and introduce widespread genetic counselling and testing in ALS, needs to start occurring now, and cannot wait for these publications to be updated.

Finally, there are numerous regions, globally, that do not have access to genetic testing for ALS. There is a need for further engagement with ALS clinicians practicing in under-represented geographies to better understand how these practices would fit into diverse cultural and socioeconomic populations. It is of the utmost importance that this disparity be addressed, as it is clear from the literature that our current understanding of genetic ALS is predominantly biased towards populations and countries that have had the privilege of conducting such research.^{42,43} Broader access to genetic testing will not only provide access to novel therapies but increase our overall understanding of ALS.

Kristiana Salmon,¹  Matthew C. Kiernan,^{2,3,†}  Seung H. Kim,^{4,†}  Peter M. Andersen,^{5,†}  Adriano Chio,^{5,7,†}  Leonard H. van den Berg,^{8,†}  Philip Van Damme,^{9,10,†}  Ammar Al-Chalabi,^{11,†}  Patricia Lillo,^{12,†}  Jinsy A. Andrews,^{13,†} and Angela Genge,^{1,†}

1 Department of Neurology and Neurosurgery, Montreal Neurological Institute-Hospital, McGill University, Montreal, Quebec, Canada

2 Brain and Mind Centre, University of Sydney, Sydney, New South Wales, Australia

3 Department of Neurology, Royal Prince Alfred Hospital, Sydney, New South Wales, Australia

4 Department of Neurology, College of Medicine, Hanyang University, Seoul, South Korea

5 Department of Clinical Science, Neurosciences, Umeå University, Umeå, Sweden

6 ALS Centre, Department of Neuroscience ‘Rita Levi Montalcini’, University of Torino, Turin, Italy

7 SC Neurologia 1U, AOU Città della Salute e della Scienza of Torino, Turin, Italy

8 Department of Neurology, UMC Utrecht Brain Center, University Medical Center Utrecht, Utrecht, The Netherlands

9 Department of Neurosciences, Laboratory for Neurobiology, KU Leuven and Center for Brain and Disease Research, VIB, Leuven Brain Institute, Leuven, Belgium

10 Department of Neurology, University Hospital Leuven, Leuven, Belgium

11 Department of Basic and Clinical Neuroscience, King’s College London, Maurice Wohl Clinical Neuroscience Institute, London, UK

12 Departamento de Neurología Sur, Facultad de Medicina, Universidad de Chile and Geroscience Center for Brain Health and Metabolism, Santiago, Chile

13 The Eleanor and Lou Gehrig ALS Center, The Neurological Institute, New York, NY, USA

[†]These authors are listed geographically, from east to west.

Correspondence to: Angela Genge

E-mail: Angela.genge@mcgill.ca

Funding

M.C.K. was supported by funding from the National Health and Medical Research Council of Australia Program Grant (#1037746), NHMRC Practitioner Fellowship (#1156093) and NHMRC Partnership Project (#1153439). A.C. was supported by the Italian Ministry of Health (Ministero della Salute, Ricerca Sanitaria Finalizzata, grant RF-2016-02362405); the Progetti di Rilevante Interesse Nazionale program of the Ministry of Education, University and Research (grant 2017SNW5MB); and the EU Joint Programme–Neurodegenerative Disease Research (Strength, ALS-Care and Brain-Mend projects), granted by Italian Ministry of Education, University and Research. His work was performed under the Department of Excellence grant of the Italian Ministry of Education, University and Research to the ‘Rita Levi Montalcini’ Department of Neuroscience, University of Torino, Italy. L.H.v.d.B. is supported by The Netherlands ALS Foundation. P.V.D. holds a senior clinical investigatorship of FWO-Vlaanderen and is supported by the E. von Behring Chair for Neuromuscular and Neurodegenerative Disorders, the ALS Liga België and the KU Leuven funds ‘Een Hart voor ALS’, ‘Laeversfonds voor ALS Onderzoek’ and the ‘Valéry Perrier Race against ALS Fund’. A.A.C. is an NIHR Senior Investigator (NIHR202421), supported by the NIHR Biomedical Research Centre at South London and

Maudsley NHS Foundation Trust and King’s College London, and part-funded by an EU Joint Programme–Neurodegenerative Disease Research (JPND) project. He is supported through the following funding organisations under the aegis of JPND–www.jpnd.eu [United Kingdom, Medical Research Council (MR/L501529/1; MR/R024804/1) and Economic and Social Research Council (ES/L008238/1)] and through the Motor Neurone Disease Association.

Competing interests

K.S., S.H.K., P.M.A. and P.L. report no competing interests relevant to the manuscript. M.C.K. is the Editor in Chief of the Journal of Neurology, Neurosurgery and Psychiatry (BMJ Publishers, UK). A.C. reports serving on advisory boards for Amylyx, Biogen, Cytokinetics, Denali, Eli Lilly, Mitsubishi Tanabe, Roche, and has received a research grant from Biogen. L.H.v.d.B. serves on advisory boards of Amylyx, argenx, Biogen, Corcept, Cytokinetics, Ferrer, QurAlis, Orion, Orphazyme, Sanofi. P.V.D. reports serving on advisory boards for Alector, Alexion, argenx, Biogen, Ferrer, Muna Therapeutics, QurAlis, UCB, and has received a research grant from CSL Behring. A.A.C. reports consultancies or advisory boards for Amylyx, Apellis, Biogen, Brainstorm, Cytokinetics, GSK, Eli Lilly, Mitsubishi Tanabe Pharma, Novartis, Orion Pharma, QurAlis, and Wave Life Sciences. J.A.A. reports consultancies for AL-S Pharma, Amylyx, Biogen, Cytokinetics, Denali, Orphazyme and Wave Life Sciences, and has received research funding from Alexion, Biogen, Cytokinetics, MGH/Healey Center for ALS and NIH/NeuroNEXT. A.G. reports consultancies or advisory boards for Alexion, AL-S Pharma, Amylyx, Anelixis, Anexon, Apellis, Atlantic Research Group, Biogen, Calico, Cytokinetics, Eli Lilly, Ionis, Mitsubishi Tanabe Pharma, Orion Pharma, QurAlis, Roche, Sanofi, UCB, and Wave Life Sciences, and has received research funding from Biogen and Novartis. Several authors of this publication are member of the European Reference Network for Rare Neuromuscular Diseases (ERN-NMD).

References

1. Kiernan MC, Vucic S, Cheah BC, et al. Amyotrophic lateral sclerosis. *Lancet*. 2011;377(9769):942–955.
2. Miller RG, Mitchell JD, Lyon M, Moore DH. Riluzole for amyotrophic lateral sclerosis (ALS)/motor neuron disease (MND). *Cochrane Database Syst Rev*. 2002;(2):CD001447.
3. Takei K, Watanabe K, Yuki S, Akimoto M, Sakata T, Palumbo J. Edaravone and its clinical development for amyotrophic lateral sclerosis. *Amyotroph Lateral Scler Frontotemporal Degener*. 2017;18(sup1):5–10.
4. Brown RH, Al-Chalabi A. Amyotrophic lateral sclerosis. *N Engl J Med*. 2017;377(2):162–172.
5. Hardiman O, van den Berg LH. The beginning of genomic therapies for ALS. *N Engl J Med*. 2020;383(2):180–181.
6. Turner MR, Al-Chalabi A, Chio A, et al. Genetic screening in sporadic ALS and FTD. *J Neurol Neurosurg Psychiatry*. 2017;88(12):1042–1044.
7. Andersen PM, Al-Chalabi A. Clinical genetics of amyotrophic lateral sclerosis: what do we really know? *Nat Rev Neurol*. 2011;7(11):603–615.
8. Perrone B, Conforti FL. Common mutations of interest in the diagnosis of amyotrophic lateral sclerosis: how common are common mutations in ALS genes? *Expert Rev Mol Diagn*. 2020;20(7):703–714.

9. Renton AE, Chiò A, Traynor BJ. State of play in amyotrophic lateral sclerosis genetics. *Nat Neurosci.* 2014;17(1):17–23.
10. Zou Z-Y, Zhou Z-R, Che C-H, Liu C-Y, He R-L, Huang H-P. Genetic epidemiology of amyotrophic lateral sclerosis: a systematic review and meta-analysis. *J Neurol Neurosurg Psychiatry.* 2017;88(7):540–549.
11. Shahrzaila N, Sobue G, Kuwabara S, et al. Amyotrophic lateral sclerosis and motor neuron syndromes in Asia. *J Neurol Neurosurg Psychiatry.* 2016;87(8):821–830.
12. Al-Chalabi A, Lewis CM. Modelling the effects of penetrance and family size on rates of sporadic and familial disease. *Hum Hered.* 2011;71(4):281–288.
13. Al-Chalabi A. Perspective: Don't keep it in the family. *Nature.* 2017;550(7676):S112.
14. Al-Chalabi A, van den Berg LH, Veldink J. Gene discovery in amyotrophic lateral sclerosis: implications for clinical management. *Nat Rev Neurol.* 2017;13(2):96–104.
15. Byrne S, Bede P, Elamin M, et al. Proposed criteria for familial amyotrophic lateral sclerosis. *Amyotroph Lateral Scler.* 2011;12(3):157–159.
16. van Es MA, Dahlberg C, Birve A, Veldink JH, van den Berg LH, Andersen PM. Large-scale SOD1 mutation screening provides evidence for genetic heterogeneity in amyotrophic lateral sclerosis. *J Neurol Neurosurg Psychiatry.* 2010;81(5):562–566.
17. Williams KL, Fifta JA, Vucic S, et al. Pathophysiological insights into ALS with C9ORF72 expansions. *J Neurol Neurosurg Psychiatry.* 2013;84(8):931–935.
18. Kiernan MC, Vucic S, Talbot K, et al. Improving clinical trial outcomes in amyotrophic lateral sclerosis. *Nat Rev Neurol.* 2021;17(2):104–118.
19. Biogen Announces Topline Results from the Tofersen Phase 3 Study and its Open-Label Extension in SOD1-ALS. Accessed 20 October 2021. <https://investors.biogen.com/news-releases/news-release-details/biogen-announces-topline-results-tofersen-phase-3-study-and-its>
20. Renton AE, Majounie E, Waite A, et al. A hexanucleotide repeat expansion in C9ORF72 is the cause of chromosome 9p21-linked ALS-FTD. *Neuron.* 2011;72(2):257–268.
21. DeJesus-Hernandez M, Mackenzie IR, Boeve BF, et al. Expanded GGGGCC hexanucleotide repeat in noncoding region of C9ORF72 causes chromosome 9p-linked FTD and ALS. *Neuron.* 2011;72(2):245–256.
22. Chio A, Battistini S, Calvo A, et al. Genetic counselling in ALS: facts, uncertainties and clinical suggestions. *J Neurol Neurosurg Psychiatry.* 2014;85(5):478–485.
23. Shefner JM, Al-Chalabi A, Baker MR, et al. A proposal for new diagnostic criteria for ALS. *Clin Neurophysiol.* 2020;131(8):1975–1978.
24. Miller RG, Jackson CE, Kasarskis EJ, et al. Practice parameter update: the care of the patient with amyotrophic lateral sclerosis: drug, nutritional, and respiratory therapies (an evidence-based review): report of the Quality Standards Subcommittee of the American Academy of Neurology. *Neurology.* 2009;73(15):1218–1226.
25. Shoesmith C, Abrahao A, Benstead T, et al. Canadian best practice recommendations for the management of amyotrophic lateral sclerosis. *CMAJ.* 2020;192(46):E1453–E1468.
26. Motor Neuron Disease: assessment and management—Clinical Guideline. Accessed 20 October 2021. <https://www.nice.org.uk/guidance/ng42/documents/motor-neurone-disease-full-guideline2>
27. Andersen PM, Borasio GD, Dengler R, et al. Good practice in the management of amyotrophic lateral sclerosis: clinical guidelines. An evidence-based review with good practice points. EALSC Working Group. *Amyotroph Lateral Scler.* 2007;8(4):195–213.
28. EFNS Task Force on Diagnosis and Management of Amyotrophic Lateral Sclerosis, Andersen PM, Abrahams S, et al. EFNS guidelines on the clinical management of amyotrophic lateral sclerosis (MALS)—revised report of an EFNS task force. *Eur J Neurol.* 2012;19(3):360–375.
29. Vajda A, McLaughlin RL, Heverin M, et al. Genetic testing in ALS: A survey of current practices. *Neurology.* 2017;88(10):991–999.
30. Klepek H, Nagaraja H, Goutman SA, Quick A, Kolb SJ, Roggenbuck J. Lack of consensus in ALS genetic testing practices and divergent views between ALS clinicians and patients. *Amyotroph Lateral Scler Frontotemporal Degener.* 2019;20(3–4):216–221.
31. Arthur KC, Doyle C, Chio A, Traynor BJ. Use of genetic testing in amyotrophic lateral sclerosis by neurologists. *JAMA Neurol.* 2017;74(1):125–126.
32. Hanby MF, Scott KM, Scotton W, et al. The risk to relatives of patients with sporadic amyotrophic lateral sclerosis. *Brain.* 2011;134(Pt 12):3454–3457.
33. Sheppard SR, Parker MD, Cooper-Knock J, et al. Value of systematic genetic screening of patients with amyotrophic lateral sclerosis. *J Neurol Neurosurg Psychiatry.* 2021;92(5):510–518.
34. Grassano M, Calvo A, Moglia C, et al. Mutational analysis of known ALS genes in an Italian population-based cohort. *Neurology.* 2021;96(4):e600–e609.
35. van Rheenen W, Shatunov A, Dekker AM, et al. Genome-wide association analyses identify new risk variants and the genetic architecture of amyotrophic lateral sclerosis. *Nat Genet.* 2016;48(9):1043–1048.
36. Brown CA, Lally C, Kupelian V, Flanders WD. Estimated prevalence and incidence of amyotrophic lateral sclerosis and SOD1 and C9orf72 genetic variants. *Neuroepidemiology.* 2021;55(5):342–353.
37. Jonsson PA, Ernhill K, Andersen PM, et al. Minute quantities of misfolded mutant superoxide dismutase-1 cause amyotrophic lateral sclerosis. *Brain.* 2004;127(Pt 1):73–88.
38. Ekhtiari Bidhendi E, Bergh J, Zetterstrom P, et al. Mutant superoxide dismutase aggregates from human spinal cord transmit amyotrophic lateral sclerosis. *Acta Neuropathol.* 2018;136(6):939–953.
39. Lek M, Karczewski KJ, Minikel EV, et al. Analysis of protein-coding genetic variation in 60,706 humans. *Nature.* 2016;536(7616):285–291.
40. Müller K, Oh KW, Nordin A, et al. De novo mutations in SOD1 are a cause of ALS. *J Neurol Neurosurg Psychiatry.* 2022;93(2):201–206.
41. Cooper-Knock J. Implications of confirmed de novo pathogenic SOD1 mutations. *J Neurol Neurosurg Psychiatry.* 2022;93(2):118.
42. Veldink JH. ALS genetic epidemiology 'How simple is the genetic epidemiology of ALS?'. *J Neurol Neurosurg Psychiatry.* 2017;88(7):537.
43. Erazo D, Luna J, Preux PM, Boumediene F, Couratier P. Epidemiological and genetic features of amyotrophic lateral sclerosis in Latin America and the Caribbean: a systematic review. *Amyotroph Lateral Scler Frontotemporal Degener.* 2022;23(1-2):4–15.