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Cascade carrier testing after identification of late-onset spinal muscular atrophy in a pilot screening program in Karachi, Pakistan

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Abstract

Background

Spinal Muscular Atrophy (SMA) is an autosomal recessive neuromuscular disorder caused by homozygous deletion of the SMN1 gene. While classically presenting in infancy or early childhood, milder phenotypes with later onset are increasingly recognized, particularly in individuals carrying multiple copies of the SMN2 gene. In Pakistan, population-level data on SMA carrier frequency remains scarce, and systematic carrier screening programs are not yet routinely implemented.

Findings:

We report a 39-year-old Pakistani male with a homozygous deletion of SMN1 (exons 7 and 8) and four copies of SMN2, identified incidentally through a pilot screening program of 100 individuals at Aga Khan University Hospital (AKUH), Karachi. The patient had a slow progressive course, with normal motor development and independent ambulation during childhood followed by gradual motor decline in adulthood. Cascade screening of his eight siblings revealed one additional homozygous individual, six heterozygous carriers, and one unaffected sibling. Notably, one at-risk couple who are both heterozygous carriers, was identified, representing a 25% recurrence risk in any future pregnancy.

Conclusion

This case shows the clinical utility of cascade family testing following index case identification for autosomal recessive disorders like SMA in the Pakistani population.

1. Introduction

Spinal Muscular Atrophy (SMA) is an autosomal recessive (AR) neuromuscular disorder characterized by degeneration of the anterior horn cells of the spinal cord, resulting in progressive muscle weakness including respiratory muscles. It is caused by pathogenic variants in the survival motor neuron 1 (SMN1) gene. The SMN2 gene is a highly homologous copy of SMN1 and is located in cis with SMN1 on chromosome 5q11-q13 in an inverted orientation, SMN2 modifies disease severity through its production of limited amounts of functional SMN protein (Lefebvre et al., 1995).

SMA is one of the most common lethal genetic disorders in infancy, with a reported birth prevalence of approximately 1 in 10,000 live births and a carrier frequency of approximately 1 in 25–50 in most populations (Hendrickson et al., 2009; Prior et al., 2010). However, population-specific data from South Asia, and Pakistan in particular, remain limited. Given the high prevalence of consanguineous marriages in Pakistan (estimated at around 50% of all unions), the burden of AR disorders, including SMA, is expected to be substantial (Bittles and Black, 2010).

SMA is clinically categorized according to the age at symptom onset and the highest motor milestone achieved. Adult-onset SMA (Type 3 and Type 4) is a relatively rare phenotype that may present as slowly progressive proximal limb weakness, mimicking other neuromuscular conditions and is frequently diagnosed with significant delay. These individuals have four or more copies of SMN2, thus exhibit a particularly attenuated phenotype with preserved mobility into adulthood (Wirth et al., 2006).

Here, we present a case of SMA Type 4 who was incidentally identified through a previously conducted SMA carrier-screening study of 100 individuals (original study submitted for publication), following the detection of biallelic SMN1 deletion. This finding prompted cascade screening of the index case's first-degree relatives and their spouses

2. Findings

The index patient is a 39-year-old Pakistani male. He was identified as having a homozygous deletion of the *SMN1* gene (exons 7 and 8) with four copies of *SMN2*, confirmed by multiplex ligation-dependent probe amplification (MLPA) analysis.

The patient achieved normal motor milestones in childhood and walked without support during early life. At around 19-years of age, he began experiencing progressive difficulty with walking and ascending stairs, requiring external support. An electromyography and nerve conduction study (EMG/NCS) performed when the patient was approximately 21 years of age, was consistent with a chronic neurogenic process. Despite this, no definitive genetic diagnosis was established at that time.

At the time of evaluation, the patient was able to maintain a standing position with support for less than 10 minutes and unable to stand independently. He was able to take approximately 15–20 steps using a walking frame. He reported no respiratory difficulty.

His wife carries no SMN1 deletion. The couple has four children; all are obligate heterozygous carriers. The couple is not at risk of having a child with homozygous SMN1 deletion.

Mutational analysis of the SMN1 gene was performed for deletions in exons 7 and 8 using the MLPA method. The SALSA MLPA P060-B2 SMA kit was used for this purpose (*P060 SMA Carrier - MRC Holland*, no date) .

Following identification of the index case, cascade screening was offered to all available first-degree relatives and their spouses, if available and consented. The index patient has eight siblings. The screening results are summarized in Fig. 1.

3. Discussion

The index case demonstrates a phenotype consistent with SMA Type 4, defined by onset of symptoms after 18 years of age and a mildly progressive clinical course. This classification is supported by his preserved mobility in childhood, symptom onset in early adulthood, absence of respiratory or bulbar

involvement, and the presence of four *SMN2* copies, a genotype most strongly associated with the adult-onset subtype (Wirth et al., 2006) .

A significant diagnostic delay is evident in this case: the patient underwent EMG/NCS at age 21, which suggested a chronic neurogenic process, yet confirmatory genetic testing was not performed until the pilot screening program. Across all SMA subtypes, diagnostic delay worsens as phenotype severity decreases (Lin, Kalb and Yeh, 2015). For SMA Type 4, delays are expected to be far greater, with many patients likely being misdiagnosed with conditions such as limb-girdle muscular dystrophy, multiple sclerosis, or other neurological disorders (Mustafa et al., 2025) .

Genetic confirmation is essential not only for diagnosis but for prognosis, treatment planning, eligibility assessment for emerging disease-modifying therapies and, as this case shows, for enabling targeted cascade family screening (Mercuri et al., 2018; Wirth et al., 2020). The identification of a second affected sibling in this case, highlights the value of proactive cascade screening. Cascade testing enables carriers to make informed reproductive decisions, thus has been demonstrated to be a cost-effective approach for identifying at-risk individuals across many disease types (Schmidlen et al., 2022). As in this case, it identified one high-risk couple who faces a 25% risk of having an affected child with each pregnancy. Without cascade screening, this couple would have remained unaware of their carrier status, potentially resulting in the birth of an affected child. This finding signifies the clinical and ethical need for systematic cascade screening following identification of an index SMA case, consistent with recommendations from both ACMG and the European Reference Network for Neuromuscular Diseases (EURO-NMD).

Under the ACMG 2021-tiered carrier screening framework, SMA is classified as a Tier 1 condition: the highest priority tier, alongside Cystic Fibrosis (CF). Tier 1 conditions are recommended for universal pre-conception screening to all individuals, regardless of ethnicity or family history. This reflects both the severity of the condition and the multiethnic distribution of SMN1 deletion carriers (Edwards et al., 2015; *Obstetrics and Gynecology*, 2017).

The advent of gene therapy with Onasemnogene abeparvovec and disease-modifying agents such as Nusinersen has transformed the therapeutic landscape of SMA, with early diagnosis and treatment offering the greatest potential for improved outcomes (Hoy, 2019). Nevertheless, the substantial financial burden of these therapies limits their availability and uptake in LMICs. This underlines the importance of preventive strategies, including cascade screening in at-risk families to offer carrier screening and premarital counseling, aimed at reducing disease incidence for AR disorders like SMA.

4. Conclusion

Cascade testing after a late-onset SMA diagnosis identified a second affected relative and an at-risk couple in this family. Wider access to carrier screening and cascade testing could help families in Pakistan make informed reproductive decisions.

5. Limitations

Carrier status was confirmed using the MLPA method. As MLPA detects copy number variations, only SMN1 deletions and duplications were evaluated. Point mutations in SMN1 are not identifiable by this method; therefore, carriers harboring pathogenic point mutations may not have been detected in our study

6. Compliance with Ethics Guidelines

Mustafa Mohsin Pishori, Zeeshan Ansar Ahmed and Bushra Afroze declare that they have no conflicts of interest.

The study was approved by the Ethical Review Committee (ERC) of Aga Khan University Hospital, Karachi, Pakistan (Approval No. 2024-9044-30991). All research activities were conducted in accordance with the ethical principles of the Declaration of Helsinki. Informed consent was obtained from the index patient and participating family members for genetic testing and publication of the findings. All identifying information was anonymized to maintain confidentiality.

Declarations

Author Contribution

Author ContributionsM.P: Conceptualization, investigation, data curation, formal analysis, visualization, and writing - original draft.B.A: Patient identification and clinical care, clinical data acquisition, supervision, and writing - review and editing.Z.A: Pathological expertise, validation, and writing - review and editing.All authors reviewed and approved the final version of the manuscript.

Data Availability

The data generated and analysed during this study are not publicly available because they consist of individual-level genetic results from members of a single family, and sharing them could compromise participants' privacy. De-identified summary data are provided in Figure 1.

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Figures

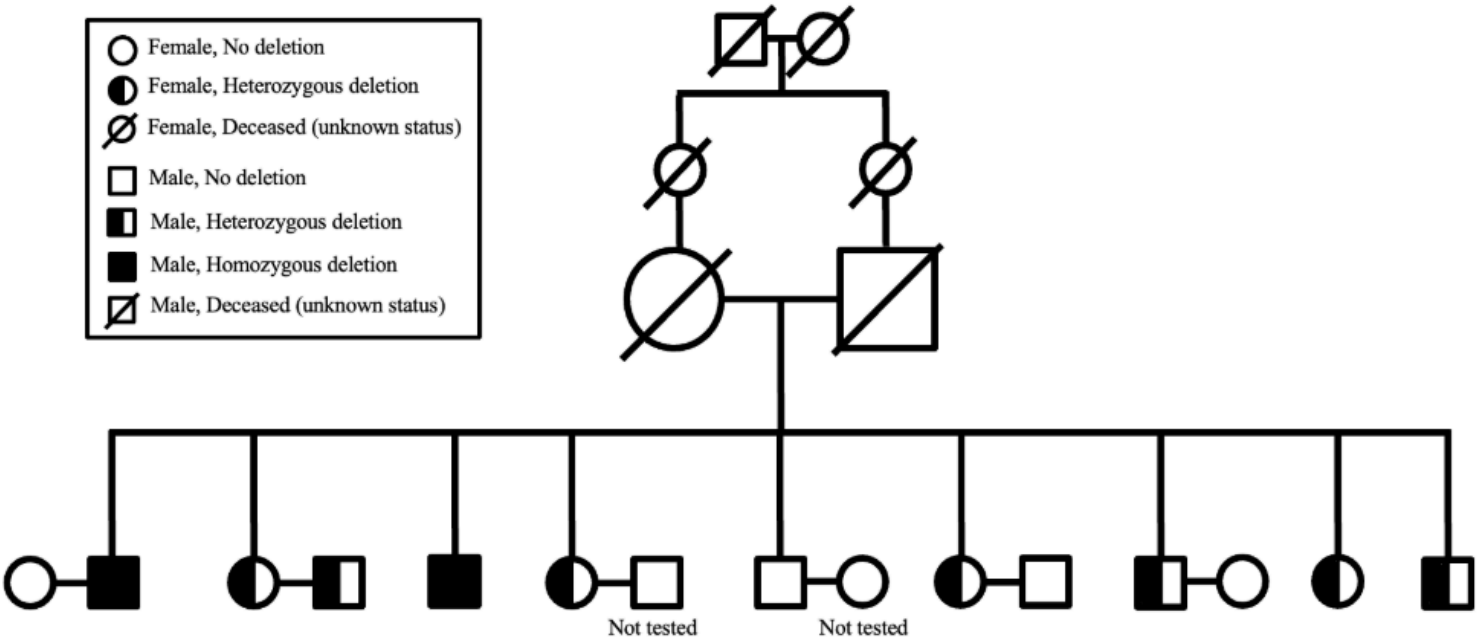


Figure 1

Pedigree of the family