The Occurrence of the Very Rare Autosomal Dominant Spinocerebellar Ataxia Subtypes SCA15, SCA31, and SCA36 in Poland and Its Implications for Clinical Practice

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ABSTRACT

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The autosomal dominant spinocerebellar ataxias (SCAs) are a genetically and clinically heterogeneous group of disorders characterized by degenerative changes in the brain and spinal cord, with disease onset ranging from infancy to adulthood. The most common SCAs are polyglutamine expansion SCAs, accounting for 45% of all autosomal dominant cerebellar ataxias. At the same time, SCA subtypes with rare or ultra-rare frequencies occur within the larger group. The molecular diagnostics of SCAs can be complicated and challenging due to the variability of genetic causes, including expansions of different repeats in coding or non-coding regions of genes, conventional mutations, and copy number variations. It is estimated that approximately 30% of patients with autosomal dominant cerebellar ataxia remain undiagnosed at the molecular level. In this study, we used polymerase chain reaction (PCR), repeat-primed PCR (RP-PCR), multiplex ligation-dependent probe amplification (MLPA), and Sanger sequencing to assess the occurrence of SCA15, SCA31, and SCA36 in Poland. Two of these rare SCA subtypes are caused by dynamic mutations, whereas SCA15 is caused by copy number variations, all exhibiting autosomal dominant inheritance.

STRESZCZENIE

Występowanie bardzo rzadkich autosomalnie dominujących postaci ataksji rdzeniowo-móżdżkowych SCA15, SCA31 i SCA36 w Polsce i jego implikacje dla praktyki klinicznej

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Autosomalnie dominujące ataksje rdzeniowo-móżdżkowe (SCA) stanowią genetycznie i klinicznie heterogenną grupę chorób, które charakteryzują się zmianami zwyrodnieniowymi mózgu i rdzenia kręgowego oraz zróżnicowanym początkiem choroby, tj. od niemowlęctwa do dorosłości. Do najczęstszych SCA zalicza się poliglutaminowe SCA związane z ekspansją powtórzeń, które stanowią 45% wszystkich autosomalnie dominujących ataksji móżdżkowych. Jednocześnie najliczniejszą grupą są pozostałe podtypy SCA z rzadką lub ultra rzadką częstością występowania. Diagnostyka molekularna SCA może być skomplikowana i stanowić duże wyzwanie ze względu na zróżnicowane przyczyny genetyczne, w tym ekspansje różnych powtórzeń w kodujących lub niekodujących regionach genów, konwencjonalne mutacje i zmiany liczby kopii. Zakłada się, że około 30% pacjentów z autosomalnie dominującą ataksją móżdżkową pozostaje niezdiagnozowanych na poziomie molekularnym. W niniejszym badaniu zastosowano reakcję łańcuchową polimerazy (PCR), zmodyfikowaną reakcję RP-PCR, zależną od ligacji multipleksową amplifikację sond (MLPA) i sekwencjonowanie metodą Sangera w celu oceny częstości występowania SCA15, SCA31 i SCA36 w Polsce.

The cohort consisted of patients from across Poland who were first clinically diagnosed with genetic ataxia, after which the most common SCA subtypes were excluded. None of the 350 tested ataxia patients had a large deletion in the ITPR1 gene, which causes SCA15. However, expansions of intronic hexanucleotide repeats in the NOP56 gene, causing SCA36, were identified in 11 individuals. These are the first Polish patients with a confirmed molecular diagnosis of SCA36. Moreover, distinct pentanucleotide repeats in the BEAN1 gene were identified in some Polish individuals. The identification of SCA36 among Polish patients indicates the need for genetic testing for SCA36 in the diagnostic setting in patients suspected of having a rare form of inherited ataxia.

Keywords: *Autosomal dominant cerebellar ataxia, SCA15, SCA31, SCA36, RP-PCR, MLPA*

Dwa spośród tych rzadkich podtypów SCA są spowodowane mutacjami dynamicznymi, podczas gdy SCA15 jest spowodowana zmianą liczby kopii, przy czym wszystkie są dziedziczone w sposób autosomalny dominujący. Badaną grupę stanowili pacjenci pochodzący z całej Polski, u których początkowo postawiono kliniczną diagnozę ataksji uwarunkowanej genetycznie, a następnie wykluczono inne, najczęstsze podtypy SCA. U żadnego z 350 badanych pacjentów z podejrzeniem ataksji nie wykryto dużej delecji w genie ITPR1, która jest przyczyną SCA15. Natomiast u 11 osób stwierdzono ekspansję intronowych heksanukleotydowych powtórzeń w genie NOP56, która jest przyczyną SCA36. Są to pierwsi polscy pacjenci z potwierdzoną diagnozą molekularną SCA36. Co więcej, u niektórych pacjentów polskiego pochodzenia wykryto różne pentanukleotydowe powtórzenia w genie BEAN1. Identyfikacja SCA36 wśród polskich pacjentów wskazuje na potrzebę przeprowadzania badań genetycznych w kierunku SCA36 w postępowaniu diagnostycznym w przypadku pacjentów z podejrzeniem rzadkiej postaci ataksji dziedzicznej.

Słowa kluczowe: *autosomalnie dominująca ataksja móżdżkowa, SCA15, SCA31, SCA36, RP-PCR, MLPA*

Introduction

Spinocerebellar ataxias (SCAs) belong to a group of neurodegenerative disorders that are heterogeneous in terms of clinical manifestations, inheritance patterns, and molecular backgrounds. The known and potential causes of these diseases may comprise various types of mutations, including single-base substitutions, deletions, insertions, duplications, or dynamic mutations resulting in the multiplication of microsatellite repeats of variable length and sequence [1]. Both point mutations and microsatellite repeats can be located in different regions of genes, such as the 5'UTR of specific mRNAs (e.g., SCA12), exons (e.g., SCA27), introns (e.g., SCA10), or the 3'UTR (e.g., SCA8) [2]. To date, more than 40 subtypes of SCAs have been described, including 17 caused by dynamic mutations (SCA1–3, 6–8, 10, 12, 17, 27B, 31, 36, 37, DRPLA (dentatorubral-pallidoluysian atrophy), FRDA (Friedreich ataxia), FXTAS (fragile X tremor/ataxia syndrome), and CANVAS (cerebellar ataxia, neuropathy, and vestibular areflexia syndrome)) [3–6]. The remaining types are caused predominantly by point mutations. This study focuses on selected SCA subtypes with dominant inheritance mode and characterized by sequence length changes in the corresponding genes presenting as deletions in *ITPR1* (MIM #147265) gene or insertions/expansions of microsatellite repeats in the *BEAN1* (MIM #612051) gene and expansion of the intronic repeats in the *NOP56* (MIM #614154) gene, responsible for SCA15 (MIM #606658), SCA31 (MIM #117210) and SCA36 (MIM #614153), respectively [7–9].

Until now, the clinical diagnosis of inherited ataxias has been based on criteria such as clinical features, age at symptom onset, family history, and brain magnetic resonance imaging (MRI), which are subsequently confirmed or excluded by genetic testing. The current classification system correlates mutations with the most frequent gene-disease correlation (one mutation = one disease). However, mutations within the same gene and causing different diseases were described in previous reports; for example, mutations in the *ITPR1* or *CACNA1A* genes are associated with SCA15, SCA16, and SCA29 [4] or SCA6, EA2 (episodic ataxia type 2), and FHM1 (familial hemiplegic migraine type 1) [10], respectively. Recent research suggests there may be additional dependencies and modifying factors related to gene interactions [11]. Given the genetic heterogeneity of SCAs, gene testing is complex and requires various molecular techniques tailored to the specific genetic defect. Currently, genotyping based on the polymerase chain reaction (PCR) can detect small expansions in coding or non-coding regions of the genome, confirming diagnoses of conditions such as SCA1, SCA2, SCA12, and SCA17. However, detecting large expansions requires additional methods such as RP-PCR (e.g., for SCA8, SCA36, FRDA, and FXTAS) or hybridization techniques. The subsequent method, essential for determining the copy number of DNA sequences, is multiplex ligation-dependent probe amplification (MLPA), which is useful for detecting deletions (e.g., SCA15, SCA16) or duplications (e.g., SCA20) [12].

This study aimed to identify ultra-rare SCA subtypes caused by dynamic and copy number mutations using various molecular techniques in a cohort of patients clinically diagnosed with inherited ataxia.

Materials and methods

DNA samples

Retrospective genetic analyses of SCA15, SCA31, and SCA36 were performed on 350 unrelated Polish patients (probands) representing 350 different families who exhibited signs of cerebellar ataxia and tested negative for the repeat expansions causing SCA1–3, 6, 7, 8, 12, and 17. Molecular analysis of SCA31 was performed on 331 of these patients, while 350 individuals were examined for SCA15 and SCA36. Most of the index patients had a positive family history. For SCA31 analysis, a control group comprised 65 elderly individuals without neurological symptoms (healthy individuals aged 60+). Blood samples were collected after obtaining informed consent from all participants under protocols approved by the Bioethical Commission of the Institute of Psychiatry and Neurology in Warsaw (resolution number: 30/2021, dated 17.11.2021). Genomic DNA was extracted from peripheral blood leukocytes using either the standard phenol/chloroform method or automated isolation on a MagNA Pure Compact Nucleic Acid Isolation System (Roche Instrument Center AG, Rotkreuz, Japan).

Molecular genetic analyses

PCR (polymerase chain reaction) – amplification of gene-specific regions containing microsatellite motifs was performed using fluorescently labeled primers, followed by electrophoresis of the PCR products on a capillary sequencer (ABI PRISM 3130 Genetic Analyzer, Applied Biosystems/Hitachi, Tokyo, Japan) with the POP7 polymer. The GS500-ROX internal size standard was used to determine the number of microsatellite repeats in *BEAN1* (SCA31) and *NOP56* (SCA36). Primer sequences used for PCR SCA31 test: Reverse 5' 6-FAM-CAGCGGGGTGAGAGAGTTACTG 3', Forward 5' GGCATAGTGGCACATGCATG 3'. Primers for PCR SCA36: Reverse 5' AACGCAACCTCAGCGTCT 3', Forward 5' 6-FAM-CGACGGTGGGGGTTTC 3'. Primers were designed using the Primer3Plus tool (https://www.primer3plus.com/index.html).

RP-PCR (repeat-primed PCR) – for SCA36 amplification of gene-specific regions containing microsatellite repeats to exclude or confirm large expansions. This method employed three primers, including a fluorescently labeled locus-specific primer flanking the repeat and paired primers amplifying multiple priming sites within the repeat. A characteristic product ladder enabled indirect, qualitative, and rapid identification of large pathogenic variants with the hexanucleotide $(GGCCTG)_n$ expansion in the *NOP56* gene. Primers for RP-PCR SCA36: Second reverse primer 5' TACGCATCCCAGTTTGAGACG 3', Forward primer 5' 6FAM-TTTCGGCCTGCGTTCGGG 3', First reverse primer 5' TACGCATCCCAGTTTGAGACGCAGGCCCAGGCCCAGGCCCAGGCC 3' [9]. Capillary electrophoresis was used to assess the length and structure of the products.

MLPA (Multiplex ligation-dependent probe amplification) – was used to detect deletions within the *ITPR1* gene (SCA15). The reagents contained probes for exons and control probes annealing to complementary regions on autosomal chromosomes. These probes were designed by Dr. Christian Beetz (Institute of Clinical Chemistry, Jena, Germany). Heterozygous deletions of target sequences resulted in a 35–50% reduction in the relative peak area of the amplification product for the affected probe.

Sanger Sequencing based on the fluorescently labeled dideoxynucleotides enabled verification of the number of repeats obtained by capillary electrophoresis and determination of the pentanucleotide repeats structure in the *BEAN1* gene. The sequencing products were separated by electrophoresis in a denaturing polyacrylamide slab gel. Primer sequences for Sanger sequencing SCA31 were the same as those used for the PCR SCA31 test, but without labeling the reverse primer with 6-FAM. Using a UV transilluminator, proper single alleles in heterozygotes were excised from a 2.0% agarose gel and purified with the Zymoclean™ Gel DNA Recovery Kit (Zymo Research, Irvine, CA, USA) following the manufacturer's protocol. DNA concentration for each purified sample was measured using a NanoDrop™ 2000 Spectrophotometer (Thermo Scientific). Purified PCR samples were subsequently sequenced using the BigDye XTerminator® Purification Kit (Applied Biosystems), and the BigDye® Terminator v3.1 Cycle Sequencing Kit (Applied Biosystems).

Results

Deletion analysis in SCA15

None of the 350 ataxia patients screened for copy number variations (CNVs) using MLPA had a deletion in the *ITPR1* gene (Figure 1).

Number of microsatellite repeats in the BEAN1 gene

The range of pentanucleotide repeats (SCA31) was 9–59 in the control group and 10–76 in the patient group (Figure 2). Homozygosity in the control group was 27.4%. The most common allele in both the control group and patients was the allele with 17 repeats, occurring in 26.2% and 23.9% of cases, respectively. The next most common alleles in the control group

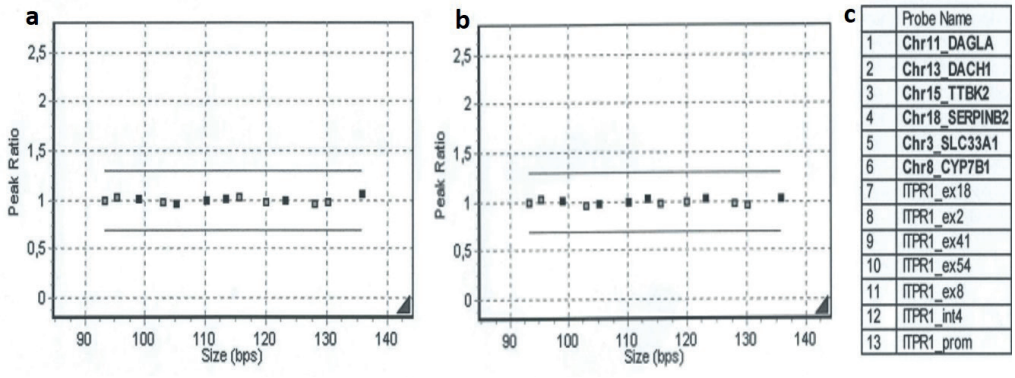

Figure 1. MLPA analysis of the *ITPR1* gene showing normal copy number status for patients 1 (a) and 2 (b), with the names of 13 designed probes (c)

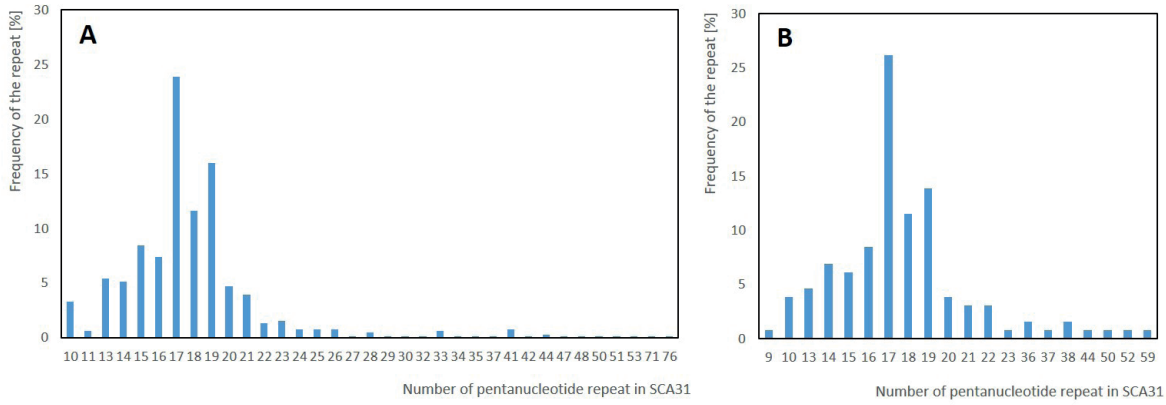

Figure 2. The frequency of microsatellite repeats in the *BEAN1* gene associated with SCA31 among 331 patients (A) and 65 control individuals (B)

and patients were those with 19 repeats (13.9% and 16.0%, respectively) and 18 repeats (11.5% and 11.6%, respectively). For alleles with more than 23 repeats, the incidence in the control samples ranged from 0.8% to 1.5%. However, the occurrence of alleles with more than 26 repeats among patients was lower, ranging from 0.2% to 0.8%.

The structure of the microsatellite repeats in the BEAN1 gene

Sanger sequencing of the *BEAN1* gene regions revealed that the common element in most of the 12 sequenced alleles was the $(TAAAA)_n$ motif, occurring in a range of 1–24 repeats. These 12 sequenced alleles included seven homozygotes, two normal (smaller) alleles, and three large alleles, all of which were from the control group. Additionally, one large allele with 50 pentanucleotide repeats was identified in a patient. Other identified pentameric repeat tracts in these alleles were: $(TAACA)_n$ in the range of 26–34 repeats, and $(AAAAA)_n$, where n=1 (Table 1, Figure 3).

The smallest sequenced allele contained 10 pentanucleotide repeats: $(TAAAA)_8(TAA)_1(TAAAA)_2$. The largest sequenced allele contained 57 repeats: $(TAACA)_{34}(T)_1(TAAAA)_{23}(AAA)_1$. Both alleles were detected in heterozygotes from the control group.

Expansions in the NOP56 gene

The normal range of alleles observed in Polish patients contained 3 to 10 $(GGCCTG)_n$ hexanucleotide repeats. RP-PCR analysis identified the expansion of intronic hexanucleotide repeat regions in the *NOP56* gene, which causes SCA36, in seven families. This expansion was confirmed in 11 affected individuals, and all exhibited the characteristic ladder pattern. However, detailed clinical characteristics will be the subject of further study. RP-PCR enables the presence or absence of an expansion to be confirmed or excluded by displaying or not displaying the characteristic ladder pattern, respectively. However, it does not allow for the exact determination of repeat numbers in expanded alleles.

Table 1. The structure of pentanucleotide repeats in the *BEAN1* gene among the control group (C1–C11) and the patient (P1)

	Structure of pentanucleotide repeats in BEAN1 gene	Number of pentanucleotide repeats
C1	$(TAAAA)_8(TAA)_1(TAAAA)_2$	10
C2	$(TAAAA)_{13}(TAA)_1(TAAAA)_2$	15
C3-C5	$(TAAAA)_{15}(TAA)_1(TAAAA)_2$	17
C6-C8	$(TAAAA)_{16}(TAA)_1(TAAAA)_2$	18
C9	$(TAACA)_{31}(TAAAA)_5(TAA)_1$	36
C10	$(TAACA)_{26}(T)_1(TAAAA)_{24}(AAA)_1$	50
P1	$(TAACA)_{28}(TAAAA)_1(AAAAA)_1(TAAAA)_{20}(AAA)_1$	50
C11	$(TAACA)_{34}(T)_1(TAAAA)_{23}(AAA)_1$	57

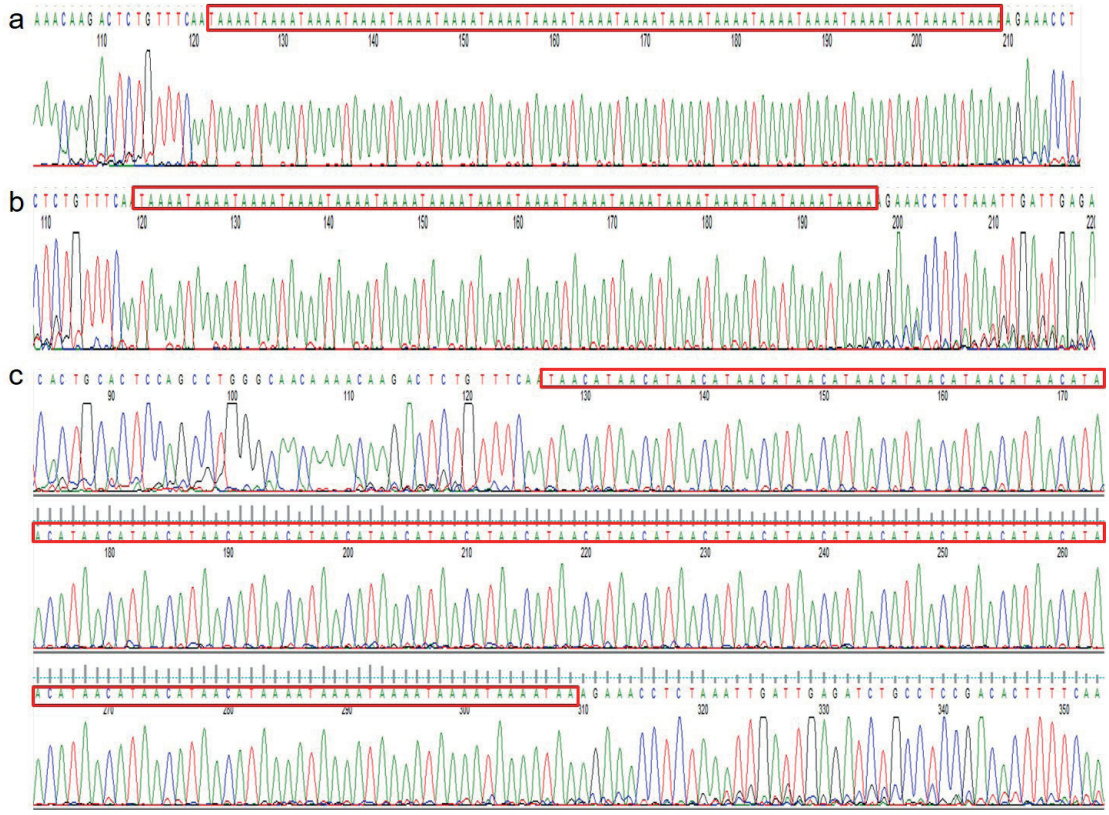

Figure 3. Sanger sequencing results for the sample of (a) case C3, who has 17 (TAAAA) repeats, (b) case C2, who has 15 (TAAAA) repeats, (c) case C9, who has 31 (TAACA) and 5 (TAAAA) repeats

The exact size of repeats can be determined by Southern blot analysis, but due to its time-consuming and costly nature, it is not used in routine genetic diagnosis of SCA36. In four presymptomatic cases, only normal alleles were detected.

Discussion

It has long been known that SCAs caused by dynamic mutations occur with varying frequencies depending on the population, a phenomenon associated with the founder effect [13,14]. Spinocerebellar ataxia type 15 (SCA15), characterized by very slow progression and mostly pure cerebellar ataxia, has been reported worldwide, including in Australia [7,12,15,16], England [16,17], Japan [18–22], Italy [23,24], Germany [25], and predominantly in Western European families [26]. Most SCA15 patients present with classic signs of cerebellar ataxia, including gait and limb/truncal ataxia, dysphagia, and titubation; some may also exhibit pyramidal signs [18,23]. Despite the widespread occurrence of SCA15 in Europe, we did not detect any cases among 350 Polish individuals affected by SCAs.

Spinocerebellar ataxia type 31 (SCA31) is caused by a ≥2.5-kb insertion containing complex pentanucleotide repeats, including $(TGGAA)_n$ within the introns of the brain-expressed associated with NEDD4 (*BEAN1*) and thymidine kinase 2 (*TK2*) genes, which are transcribed in opposite directions [8]. Most SCA31 patients present with cerebellar syndrome and late-onset [27]

ataxia of the trunk and limbs, dysarthria [28,29], decreased muscle tone, and pyramidal signs. A distinctive feature described in many SCA31 patients is hearing loss [30,31]. Studies by Sato et al. (2009) [8] and Ishikawa et al. (2011) [32] have shown that the mutation site in SCA31 varies greatly depending on the population and consists of different combinations of $(TGGAA)_n$, $(TAGAA)_n$, $(TACAA)_n$, $(GAAAA)_n$, $(TAACA)_n$, $(TGAAA)_n$, and $(TAAAA)_n$. Among these pentanucleotide repeats, only $(TGGAA)_n$ segregates with the disease and is abundant in the centromeres of several human chromosomes [8,32,33]. In Polish controls, the SCA31 mutation site includes the pentanucleotide repeats of either $(TAAAA)_n$ or $(TAAAA)_n$ in combination with $(TAACA)_n$, whereas $(TAACA)_n$ and $(TAAAA)_n$, together with $(AAAAA)_1$, were found in a single Polish patient. Notably, pentanucleotide polymorphic repeats of $(TAAAA)_n$ have been observed in both Japanese [8] and European Caucasian [32] populations, while $(TAACA)_n$ has only been reported in European Caucasian [32] populations. Moreover, similar to findings in the European Caucasian population [32], the stretch of $(TGGAA)_n$ was not detected in the Polish population. SCA31 is common and widely distributed in the Japanese population, with a strong founder effect [8,33]. In contrast, it is rare or absent in Chinese [29,34] and European Caucasian [32] populations. Furthermore, the SCA31 mutation site is associated with pentanucleotide repeat sequences of variable composition (pure in the European Caucasian [32] population and complex in the Japanese [8] population) and varying numbers of pentamers, complicating the interpretation of diagnostic testing for SCA31. Additional cases must be studied to comprehensively investigate the structure of pentanucleotide repeats in the *BEAN1* gene across other European countries. Our study has certain limitations, including a small sample size for SCA31 testing, the lack of an RP-PCR assay, and the absence of Southern hybridization. Future research will be necessary to address these gaps.

Another presented disease is spinocerebellar ataxia type 36 (SCA36) also described as late-onset ataxia [35]. It is caused by the expansion of $(GGCCTG)_n$ hexanucleotide repeats in the first intron of the nucleolar protein 56 gene (*NOP56*) located on chromosome 20 [9]. SCA36 has been identified in Japanese [9,35,36], Spanish [35,37,38], French [35], Portuguese [35], German [35], Chinese [35,39], and U.S. [40] populations.

SCA36 patients primarily exhibit progressive cerebellar ataxia, gait disturbances, truncal instability, dysarthria, and postural tremor [9]. Other symptoms include hearing impairment [35,37], cognitive impairment, reduced sensory action potentials [35], and, in some cases, motor neuron involvement, including tongue atrophy [9,35], skeletal muscle atrophy, and fasciculations [9].

In Polish patients, the mean age at onset was 47.75 years, indicating a definitive late-onset condition, with ataxia as the first symptom. Cerebellar atrophy was present in all confirmed cases. Tongue fasciculation and atrophy were observed, while muscle atrophy was infrequently noted in one patient. Cognitive decline and mood disturbances were absent in all cases.

Several studies have shown that the pathogenic expanded allele in SCA36 is generally large, ranging from approximately 650 to 2,500 repeats [9,37]. However, the identification of shorter expansions, containing 25 to 31 hexanucleotide repeats in the *NOP56* gene in three affected ataxia patients [35], suggests that SCA36 can be caused by both large and short expansions [9,35,37].

The prevalence of SCA36 varies across populations. Previous studies have shown the highest occurrence of SCA36 in Spain (Costa da Morte, Galicia region), accounting for 6.3% of cases (n=160) [37]. In contrast, SCA36 is uncommon or ultra-rare in U.S. or German populations and accounts for 0.7% (n=577) [40] or 0% (n=175) [35], respectively. Furthermore, prevalence of SCA36 varies depending on the region, e.g., in the Chinese population it ranges from 0.6% (n=512) in Han Chinese [39] to 2.3% (n=601) in Mainland China [41]. Interestingly, the presence of SCA36 in Poland is unexpected given its absence in neighboring Germany [35]. Its prevalence in the Polish population (3.1%, n=350) is comparable to that in the Japanese population (3.6%, n=251) [9]. This indicates the need for molecular testing for SCA36 in Polish patients with suspected hereditary ataxia of unknown etiology.

The occurrence of rare spinocerebellar ataxia subtypes varies depending on the population. Molecular testing for these inherited ataxias should take into account the specific distribution of SCA subtypes within each population.

References

1. Durr A.: Autosomal dominant cerebellar ataxias: polyglutamine expansions and beyond. Lancet Neurol 2010; 9(9): 885-894.
2. Pearson C.E., Edamura K.N., Cleary J.D.: Repeat instability: mechanisms of dynamic mutations. Nat Rev Genet 2005; 6(10): 729-742.
3. Epplen J.T.: Repeat Expansion Diseases, Dynamic Mutations Cause (Neurological) Model Disorders. In: Encyclopedic Reference of Genomics and Proteomics in Molecular Medicine. Berlin: Springer; 2005. 1627-1632.
4. Sandford E., Burmeister M.: Genes and genetic testing in hereditary ataxias. Genes 2014; 5(3): 586-603.
5. Seixas A.I., Loureiro J.R., Costa C. et al.: A Pentanucleotide ATTTC Repeat Insertion in the Non-coding Region of *DAB1*, Mapping to SCA37, Causes Spinocerebellar Ataxia. Am J Hum Genet 2017; 101(1): 87-103.
6. Cui Z.T., Mao Z.T., Yang R. et al.: Spinocerebellar ataxias: from pathogenesis to recent therapeutic advances. Front Neurosci 2024; 18: 1422442.

7. Storey E., Gardner R.J., Knight M.A. et al.: A new autosomal dominant pure cerebellar ataxia. Neurology 2001; 57(10): 1913-1915.
8. Sato N., Amino T., Kobayashi K. et al.: Spinocerebellar ataxia type 31 is associated with "inserted" penta-nucleotide repeats containing $(TGGAA)_n$. Am J Hum Genet 2009; 85(5): 544-557.
9. Kobayashi H., Abe K., Matsuura T. et al.: Expansion of intronic GGCCTG hexanucleotide repeat in *NOP56* causes SCA36, a type of spinocerebellar ataxia accompanied by motor neuron involvement. Am J Hum Genet 2011; 89(1): 121-130.
10. Jayadev S., Bird T.D.: Hereditary ataxias: overview. Genet Med 2013; 15(9): 673-683.
11. Pang S.Y-Y., Teo K.-C., Hsu J.S. et al.: The role of gene variants in the pathogenesis of neurodegenerative disorders as revealed by next generation sequencing studies: a review. Transl Neurodegener 2017; 6: 27.
12. Ganesamoorthy D., Bruno D.L., Schoumans J. et al.: Development of a multiplex ligation dependent probe amplification assay for diagnosis and estimation of the frequency of spinocerebellar ataxia type 15. Clin Chem 2009; 55(7): 1415-1418.
13. Krysa W., Sulek A., Rakowicz M. et al.: High relative frequency of SCA1 in Poland reflecting a potential founder effect. Neurol Sci 2016; 37(8): 1319-1325.
14. Scott S.S.O., Pedroso J.L., Barsottini O.G.P. et al.: Natural history and epidemiology of the spinocerebellar ataxias: Insights from the first description to nowadays. J Neurol Sci 2020; 417: 117082.
15. Knight M.A., Kennerson M.L., Anney R.J. et al.: Spinocerebellar ataxia type 15 (SCA15) maps to 3p24.2-3pter: exclusion of the *ITPR1* gene, the human orthologue of an ataxic mouse mutant. Neurobiol Dis 2003; 13(2): 147-157.
16. van de Leemput J., Chandran J., Knight M.E. et al.: Deletion at *ITPR1* Underlies Ataxia in Mice and Spinocerebellar Ataxia 15 in Humans. PLoS Genet 2007; 3(6): e108.
17. Novak M.J., Sweeney M.G., Li A. et al.: An *ITPR1* gene deletion causes spinocerebellar ataxia 15/16: a genetic, clinical and radiological description. Mov Disord 2010; 25(13): 2176-2182.
18. Hara K., Fukushima T., Suzuki T. et al.: Japanese SCA families with an unusual phenotype linked to a locus overlapping with SCA15 locus. Neurology 2004; 62(4): 648-651.
19. Miura S., Shibata H., Furuya H. et al.: The contactin 4 gene locus at 3p26 is a candidate gene of SCA16. Neurology 2006; 67(7): 1236-1241.
20. Hara K., Shiga A., Nozaki H. et al.: Total deletion and a missense mutation of *ITPR1* in Japanese SCA15 families. Neurology 2008; 71(8): 547-551.
21. Iwaki A., Kawano Y., Miura S. et al.: Heterozygous deletion of *ITPR1*, but not SUMF1, in spinocerebellar ataxia type 16. J Med Genet 2008; 45(1): 32-35.
22. Obayashi M., Ishikawa K., Izumi Y. et al.: Prevalence of inositol 1,4,5-triphosphate receptor type 1 gene deletion, the mutation for spinocerebellar ataxia type 15, in Japan screened by gene dosage. J Hum Genet 2012; 57(3): 202-206.
23. Di Gregorio E., Orsi L., Godani M. et al.: Two Italian families with *ITPR1* gene deletion presenting a broader phenotype of SCA15. Cerebellum 2010; 9(1): 115-123.
24. Castrioto A., Prontera P., Di Gregorio E. et al.: A novel spinocerebellar ataxia type 15 family with involuntary movements and cognitive decline. Eur J Neurol 2011; 18(10): 1263-1265.
25. Synofzik M., Beetz C., Bauer C. et al.: Spinocerebellar ataxia type 15: diagnostic assessment, frequency, and phenotypic features. J Med Genet 2011; 48(6): 407-412.
26. Marelli C., van de Leemput J., Johnson J.O. et al.: SCA15 Due to Large *ITPR1* Deletions in a Cohort of 333 White Families with Dominant Ataxia. Arch Neurol 2011; 68(5): 637-643.
27. Owada K., Ishikawa K., Toru S. et al.: A clinical, genetic, and neuropathologic study in a family with 16q-linked ADCA type III. Neurology 2005; 65(4): 629-632.
28. Fujioka S., Sundal C., Wszolek Z.K.: Autosomal dominant cerebellar ataxia type III: a review of the phenotypic and genotypic characteristics. Orphanet J Rare Dis 2013; 8: 14.
29. Ouyang Y., He Z., Li L. et al.: Spinocerebellar ataxia type 31 exists in Northeast China. J Neurol Sci 2012; 316(1-2): 164-167.
30. Ouyang Y., Sakoe K., Shimazaki H. et al.: 16q-linked autosomal dominant cerebellar ataxia: a clinical and genetic study. J Neurol Sci 2006; 247(2): 180-186.
31. Hirano R., Takashima H., Okubo R. et al.: Clinical and genetic characterization of 16q-linked autosomal dominant spinocerebellar ataxia in South Kyushu, Japan. J Hum Genet 2009; 54(7): 377-381.
32. Ishikawa K., Durr A., Klopstock T. et al.: Pentanucleotide repeats at the spinocerebellar ataxia type 31 (SCA31) locus in Caucasians. Neurology 2011; 77(20): 1853-1855.
33. Ishikawa K.: Spinocerebellar ataxia type 31 (SCA31). J Hum Genet 2023; 68(3): 153-156.
34. Lee Y.C., Liu C.S., Lee T.Y. et al.: SCA31 is rare in the Chinese population on Taiwan. Neurobiol Aging 2012; 33(2).
35. Obayashi M., Stevanin G., Synofzik M. et al.: Spinocerebellar ataxia type 36 exists in diverse populations and can be caused by a short hexanucleotide GGCCTG repeat expansion. J Neurol Neurosurg Psychiatry 2015; 86(9): 986-995.
36. Sugihara K., Maruyama H., Morino H. et al.: The clinical characteristics of spinocerebellar ataxia 36: a study of 2121 Japanese ataxia patients. Mov Disord 2012; 27(9): 1158-1163.
37. García-Murias M., Quintáns B., Arias M. et al.: 'Costa da Morte' ataxia is spinocerebellar ataxia 36: clinical and genetic characterization. Brain 2012; 135(Pt 5): 1423-1435.
38. Arias M., García-Murias M., Sobrido M.J.: Spinocerebellar ataxia 36 (SCA36): «Costa da Morte ataxia». Neurologia 2017; 32(6): 386-393.
39. Lee Y.C., Tsai P.C., Guo Y.C. et al.: Spinocerebellar ataxia type 36 in the Han Chinese. Neurol Genet 2016; 2(3): e68.
40. Valera J.M., Diaz T., Petty L.E. et al.: Prevalence of spinocerebellar ataxia 36 in a US population. Neurol Genet 2017; 3(4): e174.
41. Zeng S., Zeng J., He M. et al.: Genetic and clinical analysis of spinocerebellar ataxia type 36 in Mainland China. Clin Genet 2016; 90(2): 141-148.

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The results of the proposed statutory project may contribute to a better understanding of the epidemiology of selected rare spinocerebellar ataxias in Poland. Upon completing this research, it will be possible to incorporate these genetic tests into the diagnostic offerings of the Department of Genetics at the Institute of Psychiatry and Neurology in Warsaw. Additionally, the findings will be published as reports and articles in scientific journals dedicated to neurological diseases.

The authors declare no conflict of interest.

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