

SHORT REPORT

A Novel *TAF1C* Missense Variant Causes Neurodevelopmental Regression via Disrupted Nucleolar Localization and Nucleoplasmic Aggregation

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Received: 13 April 2025 | **Revised:** 5 May 2025 | **Accepted:** 7 May 2025

Keywords: cerebellar atrophy | neurodevelopmental regression | nucleolar dysfunction | protein mislocalization | *TAF1C* variant

ABSTRACT

TAF1C (TATA box-binding protein-associated factor, RNA polymerase I subunit C) is an essential component of the RNA polymerase I transcription machinery responsible for ribosomal RNA synthesis and nucleolar function. Variants in *TAF1C* have recently emerged as rare genetic causes of early-onset neurological syndromes characterized by nucleolar stress and impaired ribosome biogenesis, leading to developmental delay and brain atrophy. Here, we report a novel homozygous missense variant (c.1766C>T; p.Ser589Leu) in *TAF1C* in a 3-year 8-month-old boy who exhibited normal development until age two, followed by generalized seizures and progressive neurodevelopmental regression, spasticity, microcephaly, and cerebellar atrophy. MRI revealed asymmetric diffusion restriction and diffuse cerebellar atrophy. Although the mutant *TAF1C* transcript and protein were expressed at normal levels in peripheral blood cells, immunofluorescence analysis revealed a loss of nucleolar localization and the formation of abnormal thread-like aggregates within the nucleoplasm. These findings suggest that the p.Ser589Leu variant causes disease through functional mislocalization rather than loss of expression. Our case expands the phenotypic and mechanistic spectrum of *TAF1C*-related disorders and highlights the importance of proper subnuclear localization for maintaining neuronal function and development.

1 | Introduction

Transcriptional regulations are important for neuron function in various stages of neurodevelopment [1]. Increasing evidence links mutations in transcription factors to neurodevelopmental disorders, particularly those regulating RNA metabolism and gene expression during early brain development [2]. Transcription factors such as *TAF1C* and *UBTF* are essential components of the basal transcriptional machinery, and their disruption has been associated with severe early-onset neurological syndromes [3, 4]. Mutations in these genes impair core cellular processes such as ribosomal RNA synthesis, chromatin remodeling, and subnuclear organization, leading to neuronal dysfunction and degeneration [3, 4].

TAF1C (TATA box-binding protein-associated factor RNA polymerase I subunit C) is a critical subunit to initiate transcription of ribosomal DNA [5]. The *TAF1C* protein contains structural motifs such as the beta barrel domain and compositionally biased domain, which are involved in protein-protein interactions and nucleolar localization [6]. Functionally, *TAF1C* ensures proper assembly of the pre-initiation complex and maintenance of nucleolar integrity [6]. Disruptions in this machinery compromise rRNA synthesis, impair ribosome biogenesis, and trigger nucleolar stress responses, notably via activation of p53-dependent apoptotic pathways [3]. A recent study has identified biallelic missense variants in *TAF1C* as a genetic cause of severe early-onset neurodevelopmental disorders [3].

In the current study, we describe a novel homozygous missense variant in *TAF1C* (c.1766C>T; p.Ser589Leu) in a young male presenting with neurodevelopmental regression. The mutant protein failed to localize to the nucleolus and instead formed thread-like aggregates in the nucleoplasm of white blood cells. Clinically, the child exhibited a distinct phenotype marked by preserved early development, followed by generalized seizures, spasticity, microcephaly, cerebellar atrophy, and splenomegaly.

2 | Methods

The detailed methods and materials are given in the [Supporting Information](#).

3 | Results

3.1 | Case Report

We report a case of a 3-year 8-month-old boy with progressive neurodevelopmental regression. The child was born to non-consanguineous parents (Figure 1A) following an uneventful pregnancy and had normal early development until the age of two. By this time, he could stand, walk, and construct two- to three-word sentences. However, at the age of three, he experienced a generalized tonic-clonic seizure, followed by a progressive decline in motor functions, spasticity, and increased limb tone. Over the next few months, he developed tremors, chronic constipation, feeding difficulties, and an inability to accept

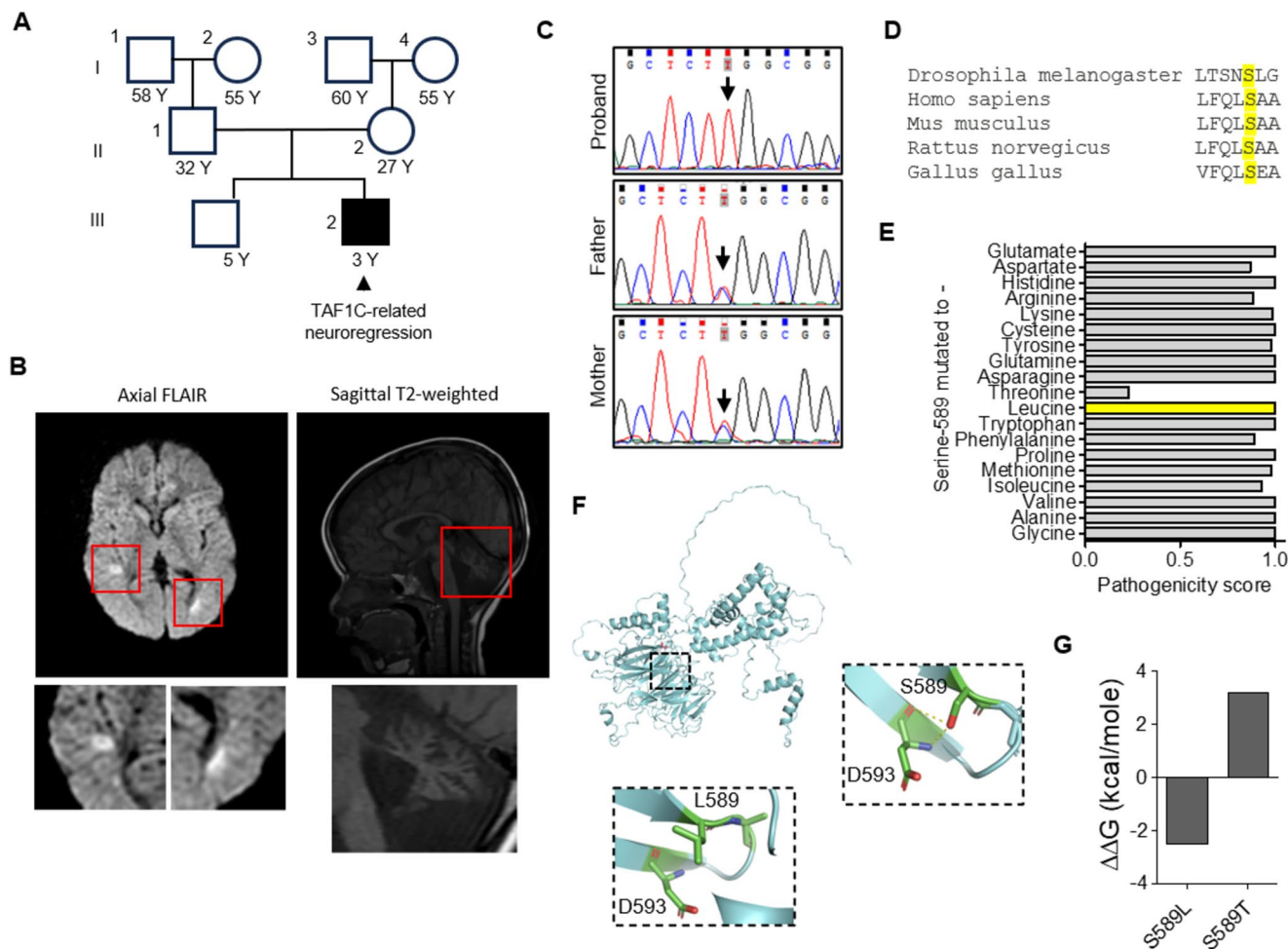


FIGURE 1 | Clinical, genetic, evolutionary, and structural characterization of the *TAF1C*-p.Ser589Leu variant in the proband. (A) Three-generational pedigree of the family showing autosomal recessive inheritance of the *TAF1C* c.1766C>T (p.Ser589Leu) variant. (B) Brain MRI of the proband at age 3 years shows asymmetrical diffusion restriction in the left parietooccipital and right temporal lobes along with subtle T2/FLAIR hyperintensities and diffuse cerebellar atrophy. (C) Sanger sequencing chromatograms confirm the homozygous c.1766C>T variant in the proband and heterozygosity in both parents. (D) Multiple sequence alignment of *TAF1C* orthologs across species reveals evolutionary conservation of the serine at position 589. (E) PolyPhen-2.0 analysis of all possible substitutions at residue S589 demonstrates that most substitutions, including S589L, are predicted to be probably damaging. (F) Left: Structural model of *TAF1C* from AlphaFold2, highlighting the position of Ser589 located at the edge of a β -sheet within a β -barrel domain. Right: Interaction map showing native contacts between S589 and D593 in wild-type *TAF1C*, which are lost in the S589L mutant. (G) Predicted change in folding free energy ($\Delta\Delta G$) of *TAF1C* due to the S589L substitution, as computed using FoldX. [Colour figure can be viewed at [wileyonlinelibrary.com](#)]

solid food. Despite motor regression, cognitive functions remained relatively preserved. The proband's cognitive function was assessed through a combination of clinical observation and developmental milestone tracking, as well as standardized cognitive and developmental screening tools. These included age-appropriate neurodevelopmental assessments such as the Bayley Scales of Infant and Toddler Development (Bayley-III) or the Mullen Scales of Early Learning (MSEL), which were used to evaluate cognitive, motor, language, and adaptive behavior. On clinical examination, the child had microcephaly (head circumference 48cm), low body weight (10.1 kg), increased muscle tone in all four limbs, and spasticity. Ultrasound examination revealed splenomegaly but no hepatomegaly. MRI of the brain revealed asymmetrical areas of diffusion restriction in the left parietooccipital and right temporal lobes, along with subtle T2/FLAIR hyperintensities, suggesting encephalopathy (Figure 1B). Diffuse cerebellar atrophy was also noted (Figure 1B), while basal ganglia, thalami, and major vascular structures appeared normal. The combination of neurodevelopmental regression, microcephaly, cerebellar atrophy, and splenomegaly suggests a possible neurodegenerative disorder. The proband underwent electroencephalography (EEG) at the age of 3 years and 8 months, which revealed normal findings.

3.2 | Genetic Findings

Singleton exome sequencing [7] of the proband identified a novel homozygous variant in the *TAF1C* gene, corresponding to c.1766C>T, located at exon 16 of the *TAF1C* (transcript: NM_005679) (Figure 1C). This is a missense variant, potentially leading to p.Ser589Leu in the protein. Notably, the c.1766C>T variant is absent from population databases such as gnomAD and clinical repositories such as ClinVar, suggesting that it is a

novel variant not previously reported in the context of *TAF1C*-related diseases. The proband's parents were found to be heterozygous carriers of this variant (Figure 1C), consistent with an autosomal recessive inheritance pattern.

3.3 | Pathogenicity Prediction of the p.Ser589Leu in TAF1C

The missense variant p.Ser589Leu in *TAF1C* affects a highly conserved serine residue (Figure 1D), predicted to disrupt the protein's structure and function. To evaluate its pathogenic potential, multiple computational tools were utilized. PolyPhen-2.0 predicted the S589L substitution to be damaging, highlighting that replacing Ser589 with any amino acid other than threonine would likely cause significant structural and functional alterations (Figure 1E). Additional *in silico* analyses further supported the pathogenicity of the p.Ser589Leu. The Combined Annotation Dependent Depletion (CADD) score classified it as deleterious, while MutationTaster identified it as disease-causing. Similarly, ClinPred predicted the variant to be pathogenic. The S589 is located at the edge of a beta strand of the beta barrel domain. S589 interacts with D593 in wild-type *TAF1C*, thereby stabilizing the beta strand (Figure 1F). L589 in the mutant *TAF1C* loses this interaction (Figure 1F). The S589L variant also destabilizes the folding of the mutant protein (Figure 1G).

3.4 | Cellular and Functional Impact of TAF1C Variant

We investigated the molecular consequences of the S589L (c.1766C>T) missense variant in the *TAF1C* gene in a male proband presenting with neurodevelopmental abnormalities.

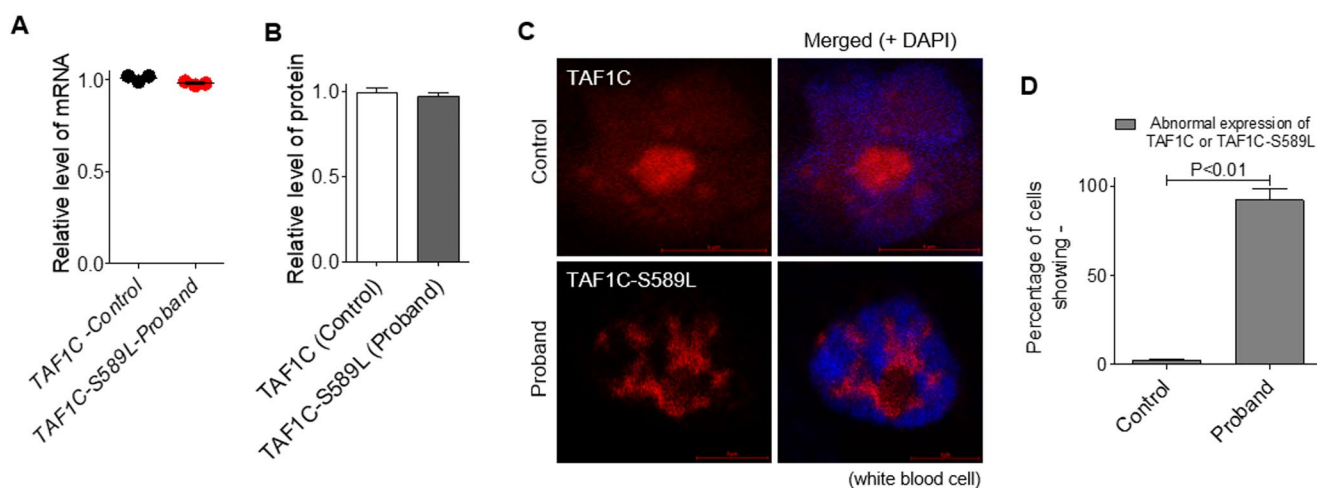


FIGURE 2 | Functional characterization of *TAF1C* variant (p.S589L) in white blood cells of the proband. (A) Quantitative PCR analysis of *TAF1C* transcript levels in white blood cells of the control and proband carrying the c.1766C>T variant. No significant difference in mRNA levels is observed between control and proband samples. (B) ELISA analysis of *TAF1C* protein levels in white blood cells. No significant reduction in *TAF1C* protein levels is observed in the proband compared to the control. (C) Immunofluorescence staining of *TAF1C* in white blood cells. In control cells, wild-type *TAF1C* localizes to both the nucleolus and nucleoplasm. In contrast, cells from the proband expressing the mutant *TAF1C*-p.S589L exhibit disrupted subcellular distribution with loss of nucleolar localization and the presence of abnormal thread-like aggregates within the nucleus. (D) Quantification of *TAF1C* immunofluorescence patterns from panel C, showing a significant increase in the proportion of cells displaying mislocalized or aggregated *TAF1C* in the proband compared to the control. [Colour figure can be viewed at wileyonlinelibrary.com]

TABLE 1 | Clinical and genetic characteristics of the probands.

	Knuutinen et al. [3]	This study
Age of proband	7 and 8 Years	3 Years 8 Months
Gender	Female (both probands)	Male
Consanguinity	Yes (for both probands)	No
Global developmental delay	Severe	No global delay in early development; normal milestones until age 2
Neuroregression	Yes	Yes – Progressive motor regression following a generalized seizure at age 3
Age of onset of regression	Neonatal, 4 months	3 Years
MRI observations	Mild cerebral, cerebellar, and brainstem atrophy, thinning of the corpus callosum, atrophy of the cerebellar peduncles, frontal periventricular white matter hyperintensities on T2/FLAIR sequences, progressive atrophy was most pronounced in pontocerebellar structures, while cerebral atrophy remained stable	Asymmetrical areas of diffusion restriction in the left parietooccipital and right temporal lobes, along with subtle T2/FLAIR hyperintensities
EEG observations	Intermittent hypsarrhythmia and a slow background pattern	Normal
Seizure characteristics	Epileptic spasms, refractory to multiple antiepileptic drugs	Generalized tonic–clonic seizure
Clinical observations	Proband 1: hypsarrhythmia, precocious puberty, microcephaly (−2.3 SD), central hypotonia, spasticity. Proband 2: Spastic paraplegia, hypotonia, hyporeflexia, dysmorphic features, strabismus, nystagmus, microcephaly (−1.1 SD)	Generalized tonic–clonic seizures, progressive spasticity, increased muscle tone in all limbs, tremors, chronic constipation, feeding difficulties, inability to accept solid food, preserved cognitive functions
Additional features	Proband 1: Gastrostomy tube, delayed dentition, cardiac conduction abnormality, transient liver findings. Proband 2: Facial dysmorphism, no endocrine or cardiac findings	Microcephaly (head circumference 48 cm), low body weight (10.1 kg), splenomegaly, recurrent febrile episodes
Variant in TAF1C	c.1165C>T; p.Arg389Cys and c.1213C>T; p.Arg405Cys	c.1766C>T; p.Ser589Leu
Zygosity	Homozygous for both	Homozygous

Transcript levels of *TAF1C* in the proband’s white blood cells were quantified using quantitative RT-PCR. The mutant *TAF1C* transcript (c.1766C>T) was expressed at levels comparable to those of the wild-type *TAF1C* transcript in the control individual (Figure 2A), indicating that the S589L mutation does not affect mRNA stability or expression. At the protein level, ELISA-based quantification revealed that the mutant TAF1C-S589L protein was abundantly expressed in the proband’s white blood cells, at levels similar to wild-type TAF1C protein observed in the control (Figure 2B). These results suggest that the S589L variant does not significantly impact the expression or overall stability of the TAF1C protein.

However, subcellular localization studies using immunofluorescence microscopy uncovered critical differences. In healthy control cells, wild-type TAF1C displayed a characteristic nuclear and nucleolar localization pattern, consistent with its known function in transcriptional regulation. In contrast, the mutant

TAF1C-S589L protein in the proband’s white blood cells failed to localize to the nucleolus (Figure 2C). Instead, the protein was retained within the nucleoplasm, where it formed abnormal thread-like aggregates, suggestive of a loss of proper subnuclear compartmentalization and potential aggregation (Figure 2C,D).

4 | Discussion

Mutations in *TAF1C* that regulate Pol I activity represent a rare and emerging subgroup of inherited conditions associated with neurodevelopmental regression, highlighting a novel pathomechanism involving nucleolar dysfunction.

The variant identified in this study, p.Ser589Leu, is novel, and this missense variant affects a highly conserved serine residue within the TAF1C protein. The S589L substitution likely alters protein structure or impairs its normal function. Given the

importance of TAF1C in maintaining nucleolar integrity and promoting rRNA synthesis, structural disturbances in this protein can lead to significant disruptions in cellular homeostasis, particularly in metabolically active neuronal populations.

One of the key findings of our study is that the S589L variant does not reduce the transcript or overall protein level of TAF1C. These results clearly distinguish the current case from previously reported *TAF1C* variants, such as those described by Knuutinen et al. (2020), where TAF1C variants (p.Arg389Cys and p.Arg405Cys) led to substantial reductions in both mRNA and protein levels. In contrast, our findings suggest that the S589L variant does not cause disease via a loss-of-expression mechanism, but rather through a disruption of subcellular localization and protein function [3]. This hypothesis is supported by our immunofluorescence-based subcellular localization studies. In control cells, wild-type TAF1C exhibited a characteristic nucleolar and nucleoplasmic distribution, in line with its role in nucleolar transcription of rRNA. In stark contrast, the TAF1C-S589L mutant protein failed to localize to the nucleolus and instead formed abnormal thread-like aggregates in the nucleoplasm. These aggregates may reflect misfolded protein species or unsuccessful assembly into the transcriptional machinery, although further studies are needed to confirm their precise nature. Such mislocalization can be highly detrimental [8], as it may not only impair the essential process of ribosome biogenesis but also contribute to nucleolar stress. Nucleolar dysfunction is a well-known trigger for p53 activation, leading to cell cycle arrest or apoptosis, particularly in neuronal tissues [9]. Thus, the S589L variant appears to cause a functional defect despite normal expression, supporting the idea that correct localization is essential for TAF1C function and neuronal integrity. Clinically, the phenotype of our proband shows both overlapping and distinct features compared to the two patients reported in the study by Knuutinen et al. [3] (Table 1).

Taken together, this case study adds to the emerging literature on TAF1C-associated neurodevelopmental disorders and expands the known variant spectrum.

Author Contributions

S.R.A.: conceived and designed the study, contributed to the experimental work, performed data analysis, and drafted the manuscript. **N.V.:** study analysis, contributed to the interpretation of the genetic findings. **N.N.:** clinical evaluation, diagnosis, methodology, and data analysis.

Acknowledgments

The authors sincerely thank the proband and his parents for their valuable participation in this study.

Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability Statement

The data generated and analyzed in this study are available from the corresponding author upon reasonable request.

References

1. A. E. West, E. C. Griffith, and M. E. Greenberg, "Regulation of Transcription Factors by Neuronal Activity," *Nature Reviews. Neuroscience* 3 (2002): 921–931.
2. J. Santos-Terra, I. Deckmann, M. Fontes-Dutra, G. B. Schwingel, V. Bambini-Junior, and C. Gottfried, "Transcription Factors in Neurodevelopmental and Associated Psychiatric Disorders: A Potential Convergence for Genetic and Environmental Risk Factors," *International Journal of Developmental Neuroscience* 81 (2021): 545–578.
3. O. Knuutinen, A. Pyle, M. Suo-Palosaari, et al., "Homozygous TAF1C Variants Are Associated With a Novel Childhood-Onset Neurological Phenotype," *Clinical Genetics* 98 (2020): 493–498.
4. R. J. Tinker, T. Guess, D. C. Rinker, et al., "A Novel, Likely Pathogenic Variant in UBTF-Related Neurodegeneration With Brain Atrophy Is Associated With a Severe Divergent Neurodevelopmental Phenotype," *Molecular Genetics & Genomic Medicine* 10 (2022): e2054.
5. E. M. Crombie, K. Cleverley, H. T. M. Timmers, and E. M. C. Fisher, "The Roles of TAF1 in Neuroscience and Beyond," *Royal Society Open Science* 11 (2024): 240790.
6. A. Bernardini, P. Mukherjee, E. Scheer, et al., "Hierarchical TAF1-Dependent Co-Translational Assembly of the Basal Transcription Factor TFIID," *Nature Structural & Molecular Biology* 30 (2023): 1141–1152.
7. P. Udupa and D. K. Ghosh, "Implementation of Exome Sequencing to Identify Rare Genetic Diseases," *Methods in Molecular Biology* 2719 (2024): 79–98.
8. M. A. Mensah, H. Niskanen, A. P. Magalhaes, et al., "Aberrant Phase Separation and Nucleolar Dysfunction in Rare Genetic Diseases," *Nature* 614 (2023): 564–571.
9. K. Holmberg Olausson, M. Nister, and M. S. Lindstrom, "p53-Dependent and -Independent Nucleolar Stress Responses," *Cells* 1 (2012): 774–798.

Supporting Information

Additional supporting information can be found online in the Supporting Information section.