# Molecular pathology selected abstracts

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## *TCEAL1* loss of function: cause of a rare X-linked dominant neurodevelopmental syndrome

January 2023—An international group of scientists and clinicians identified the molecular cause of a rare neurodevelopmental syndrome affecting children worldwide. This discovery was made possible through such publicly available online databases as MyGene2, GeneMatcher, and Matchmaker Exchange, which match genotypic profiles with phenotypic profiles of rare diseases. The causative gene underlying this novel neurodevelopmental syndrome is *TCEAL1* (transcription elongation factor A-like 1), a single coding-exon gene that encodes a nuclear phosphoprotein, TCEAL1, involved in transcriptional regulation. *TCEAL1* is located on the X chromosome in a region known as the Xq22.2 sub-band, which is approximately 1.2 Mb. Disruptions in this sub-band have been associated with other neurological diseases. In particular, a span within the Xq22.2 sub-band containing six contiguous genes, including *TCEAL1*, has been associated with emerging early onset neurological disease trait (EONDT), which affects 46,XX females. EONDT consists of hypotonia at birth, neurobehavioral abnormalities, severe intellectual disability, and mild dysmorphic facial features. The authors searched online databases to determine if a single gene within the aforementioned region of the X chromosome was responsible for an undefined monogenic neurodevelopmental disorder. In the study, four males and three females with de novo loss-of-function *TCEAL1* variants were identified. Their clinical features resembled those of EONDT patients harboring contiguous deletions involving *TCEAL1* and the adjacent gene *PLP1*, including developmental delay/intellectual disability, neurobehavior abnormalities, and dysmorphic craniofacial features. Additional features that characterize the novel *TCEAL1* loss-of-function disorder include hypotonia, stereotypic movement disorder, and abnormal gait or nonambulatory status. A subset of patients showed abnormal myelination, structural brain anomalies, or seizures. Furthermore, some patients had ocular, gastrointestinal, or immune system abnormalities. Overall, the female patients had milder symptoms than the male patients, which supports a role for *TCEAL1* gene dosage in the observed variation in disease severity. An eighth patient, who was male, had a maternally inherited *TCEAL1* missense variant and presented with a different set of clinical features, which included neuromuscular features in the absence of developmental delay/intellectual disability or dysmorphic craniofacial features, potentially suggesting an alternate molecular mechanism. Molecular mechanisms involving the Xq22.1-Xq22.2 region may contribute to the multiple, though rare, neurological disorders associated with this chromosomal region. Genomic instability of the Xq22.1-Xq22.2 region may be due to such factors as increased repeat sequences, copy number variants (CNV), intergenic microdeletions, and the effect of X-chromosome inactivation in females. Unfortunately, standard screening methods may not detect microdeletions of *TCEAL1*. Given the genomic instability in Xq22.1-Xq22.2, the authors propose applying DNA sequencing and CNV assessment to this region.

Hijazi H, Reis LM, Pehlivan D, et al. *TCEAL1* loss-of-function results in an X-linked dominant neurodevelopmental syndrome and drives the neurological disease trait in Xq22.2 deletions. *Am J Hum Genet*. 2022. https://doi.org/10.1016/j.ajhg.2022.10.007

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## Single-cell multiomics of human clonal hematopoiesis: revelations about mutations

As people age, a subset of blood cells will acquire cancer-associated mutations without a diagnosis of blood cancer. Because this condition, known as clonal hematopoiesis, can be a precursor of hematologic malignancy, it is of increasing clinical and scientific interest. Clonal hematopoiesis, in turn, can be subclassified based on peripheral blood cell counts and the characteristics of precursor bone marrow cells. The clonal stem cells harboring somatic alterations are often of the myeloid lineage and produce erythrocytes, granulocytes, or megakaryocytes as they mature. There is significant overlap between the mutations seen in clonal hematopoiesis and those seen in acute myeloid leukemia (AML). However, patients with frank malignancy often harbor additional acquired mutations or genomic aberrations that lead to cells becoming cancerous. Consequently, while patients with clonal hematopoiesis have an increased risk of progressing to frank malignancy, this may or may not occur, and the role these low-level mutations play in tumorigenesis is unclear. The authors performed a detailed analysis of the genomic and epigenomic changes involved in clonal hematopoiesis by applying multiomics to single-cell analysis and comparing clonal and normal background stem cells from the same patients. Stem cells were obtained from patients with clonal hematopoiesis harboring a mutation in codon R882 of the epigenetic modifying gene DNA methyltransferase 3α (*DNMT3A*). This is a common and well-characterized mutation seen in AML and clonal hematopoiesis. In gene-expression profiling, clonal and nonclonal stem cells demonstrated a similar degree of gene expression. Interestingly, stem cells harboring the *DNMT3A* R882 mutation were enriched in the megakaryocytic-erythroid progenitor cell populations. Compared to nonclonal stem cells, *DNMT3A* R882 clonal stem cells showed increased expression of genes previously associated with leukemia stem cells, genes involved in proinflammatory signaling, and genes associated with megakaryocytic-erythroid hematopoiesis, consistent with dysregulation of megakaryocytic-erythroid stem-cell lineages. To further understand the implications of the *DNMT3A* R882 alteration, the authors used a single-cell multimodal approach, evaluating DNA methylation, RNA sequencing, and targeted somatic genotyping, to assess the molecular landscape of the abnormal cells. *DNMT3A* R882-mutated stem cells, unlike nonmutated stem cells, showed an overall decrease in methylation status across the genome, similar to previous findings about genomic hypomethylation in AML. A large subset of the hypomethylated genes in *DNMT3A* R882 stem cells are associated with cell-lineage differentiation and are regulated by a different epigenetic regulator, PRC2, which is suggestive of orchestrated, multilayered epigenetic dysregulation. Single-cell gene-expression profiling of *DNMT3A* R882 stem cells showed increased expression of key hematopoietic transcription factors, including MYC/MAX, HIF1A/ARNT, USF1/2, and KLF1. Notably, the increased expression correlated with selective hypomethylation of sequence-specific CpG motifs in *DNMT3A* R882 stem cells, suggesting an underlying mechanism, which parallels findings for AML. Overall, this elegant study demonstrates the utility of a single-cell multiomics approach to investigating the cellular dynamics of gene control in stem cells with cancer-associated mutations.

Nam AS, Dusaj N, Izzo F, et al. Single-cell multi-omics of human clonal hematopoiesis reveals that *DNMT3A* R882 mutations perturb early progenitor states through selective hypomethylation. *Nat Genet*. 2022;54:1514–1526.

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