

Supplemental Material

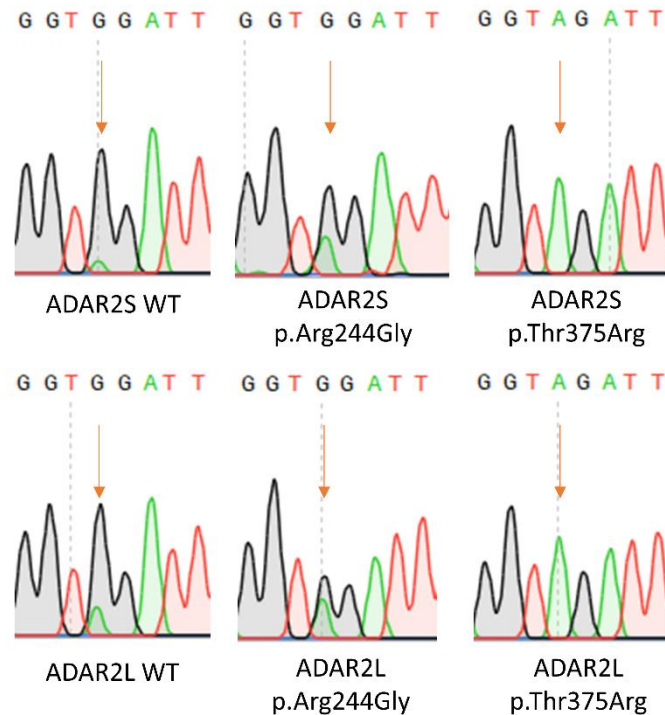
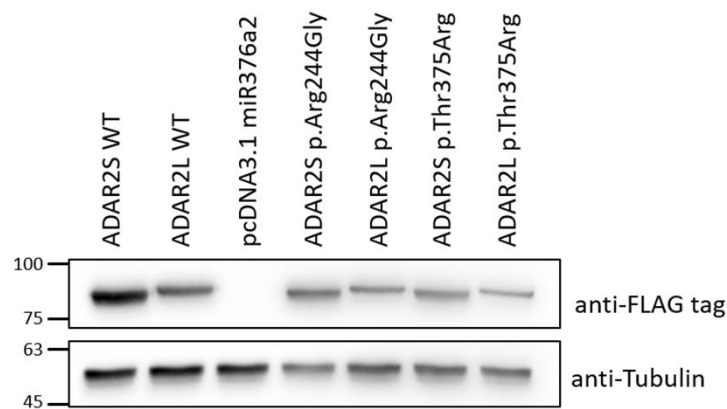
Novel ADAR2 variants in children with seizures, intellectual disability and motor delay have reduced RNA editing.

Qiupei Du^{1,2}, Anna Cherian^{1,2}, Raymond J. Louie³, Giulia Barcia^{4,5}, Natasha L. Rudy³, Rima Nabbout^{5,6}, Eugénie Sarda⁶, Maelle Charpié⁴, Wesley G. Patterson³, Liam P. Keegan^{*1} and Mary A. O'Connell^{*1}

¹Central European Institute for Technology, Masaryk University, Kamenice 5, Brno 62500, Czechia. ²National Centre for Biomolecular Research, Faculty of Science, Masaryk University, Kamenice 5, Brno 62500, Czechia. ³Greenwood Genetic Center, Greenwood, South Carolina USA. ⁴Department of Genetics, Necker Enfants Malades University Hospital, AP-HP, Université Paris Cité, Paris, France. ⁵Laboratory of Translational Research for Neurological Disorders, INSERM UMR 1163, Imagine Institute, Paris, France. ⁶Department of Pediatric Neurology, Reference Centre for Rare Epilepsies, Necker Enfants Malades University Hospital, AP-HP, Université Paris Cité, Paris, France.

* Corresponding authors

Email: Liam keegan@ceitec.muni.cz mary.oconnell@ceitec.muni.cz

A**B**

Supplemental Figure S1. Sanger sequencing and expression of ADAR2 variants p.Arg244Gly and p.Thr375Arg.

(A) Representative Sanger sequencing chromatograms showing RNA editing at the +4 site of *pri-mir-376a2* for ADAR2S and ADAR2L WT and mutant enzymes. The edited site is highlighted with an orange arrow. (B) Immunoblotting analysis demonstrating comparable expression levels of ADAR2 WT and mutant proteins. Anti-FLAG antibody detects ADAR2 proteins, and anti-Tubulin detection serves as a loading control.

Supplemental Table S1.

Oligonucleotide primers used for mutagenesis and RT-PCR

Primers for mutagenesis	Forward Primer	Reverse Primer
ADAR2 p.(ARG244GLY)	GAAGTGGGCCCAGGACTCAA GTATGACTTCCTCTCCGAGAGC GGGGA	TTGAGTCCTGGGCCCAGTTCGTT CAAGATCATCACGGGATTCTTCC CACTCGG
ADAR2 p.(THR375ARG)	ACAGGAAGAAAATGTATTAAT GGTGAATACATGAGT	ACATTTCTTCCTGTAGAAACAC T TATCACCTTG
RT-PCR primers		
RFP-GFP	GGTGGAGTTCAAGTCCATCTAC ATGG	TCGACCAGGATGGGCACCAC
RFP-Intron11	GGTGGAGTTCAAGTCCATCTAC ATGG	GCAGCTGCTGACATCTTTATAGT G
Control-RFP	CCCGGCTACTACTACGTGGA	ATCTCAGGAACAGGTGGTGG
pri-miR376a2	TGGGCTCCGTCGTCATTTT	CCATCTTTCCACTTACCCTGG
<i>Gria2</i> Q/R	CCTGGTCAGCAGATTTAGC	CTAACCTCGCCCATTTTCC