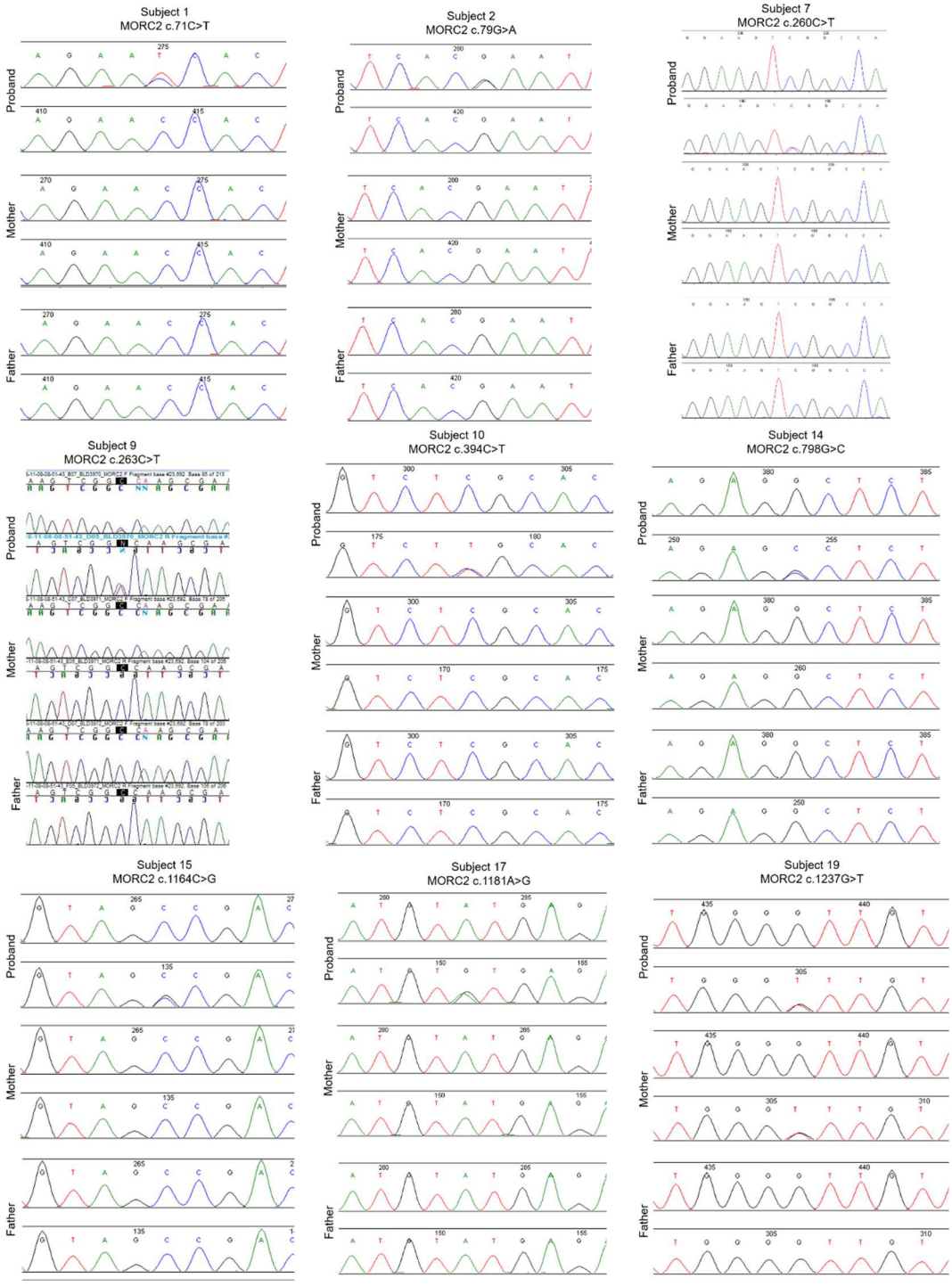
The American Journal of Human Genetics, Volume *107*

Supplemental Data

De Novo Variants in the ATPase Module of MORC2 Cause a Neurodevelopmental Disorder with Growth Retardation and Variable Craniofacial Dysmorphism

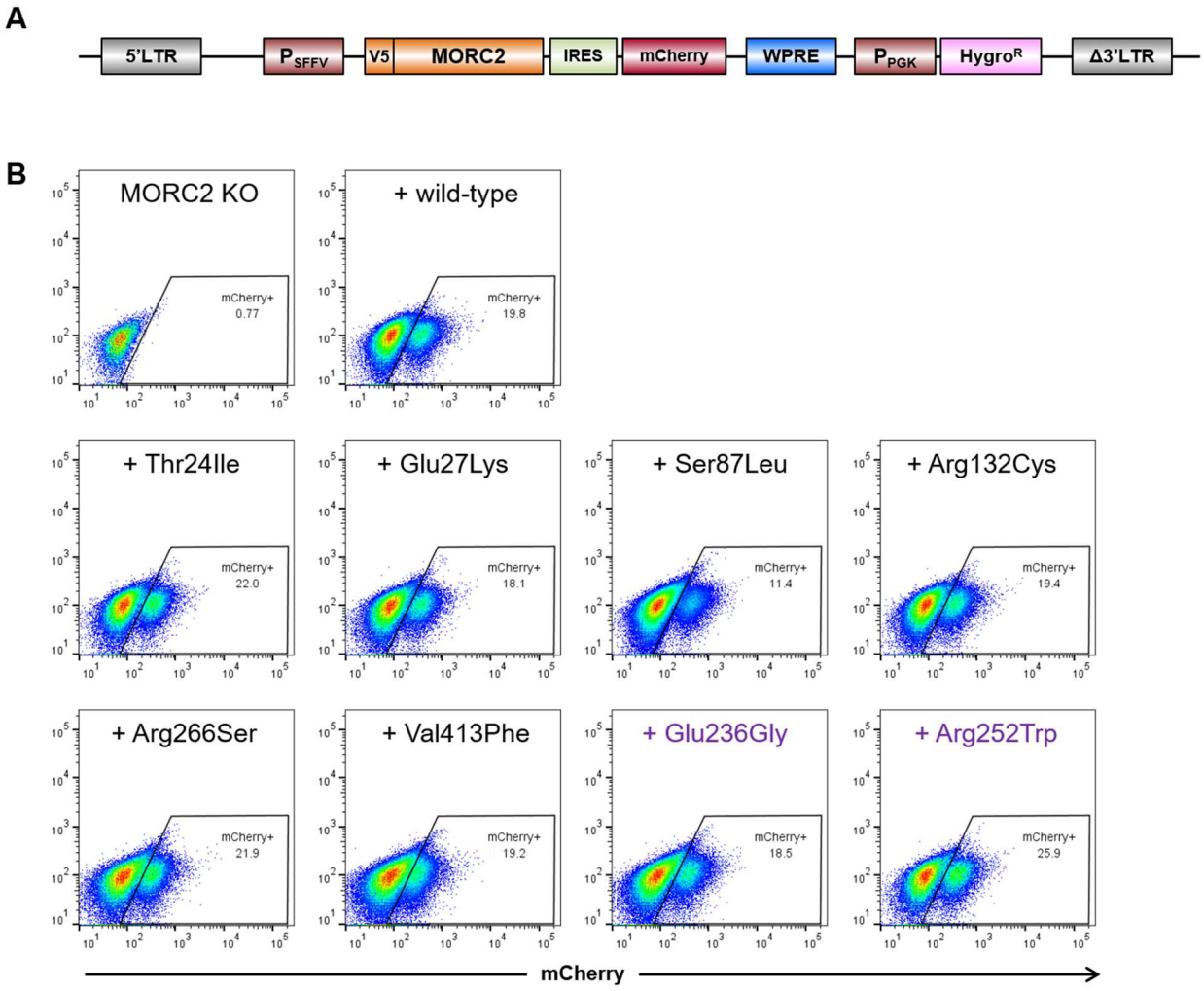
Maria J. Guillen Sacoto, Iva A. Tchasovnikarova, Erin Torti, Cara Forster, E. Hallie Andrew, Irina Anselm, Kristin W. Baranano, Lauren C. Briere, Julie S. Cohen, William J. Craigen, Cheryl Cytrynbaum, Nina Ekhilevitch, Matthew J. Elrick, Ali Fatemi, Jamie L. Fraser, Renata C. Gallagher, Andrea Guerin, Devon Haynes, Frances A. High, Cara N. Inglese, Courtney Kiss, Mary Kay Koenig, Joel Krier, Kristin Lindstrom, Michael Marble, Hannah Meddaugh, Ellen S. Moran, Chantal F. Morel, Weiyi Mu, Eric A. Muller II, Jessica Nance, Marvin R. Natowicz, Adam L. Numis, Bridget Ostrem, John Pappas, Carl E. Stafstrom, Haley Streff, David A. Sweetser, Marta Szybowska, Undiagnosed Diseases Network, Melissa A. Walker, Wei Wang, Karin Weiss, Rosanna Weksberg, Patricia G. Wheeler, Grace Yoon, Robert E. Kingston, and Jane Juusola

Figure S1. Sequence traces of reported variants in *MORC2*.



Wild-type alleles from both parents and the heterozygous *de* novo variant from Subjects 1, 2, 7, 9, 10, 14, 15, and 17. Subject 19 inherited the heterozygous variant from her affected mother (Subject 20).

Figure S2. Mutations in MORC2 hyperactivate HUSH-mediated silencing in a reporter re-repression assay.



(**A**) Schematic representation of the MORC2 lentiviral expression vector used. In addition to V5-tagged MORC2, this vector also expresses mCherry from an internal ribosome entry site (IRES) to enable an accurate assessment of the multiplicity of infection achieved.

(**B**) Mutations in MORC2 result in enhanced transgene repression by the HUSH complex. MORC2 knockout (KO) HeLa cells harboring a derepressed GFP reporter construct were transduced with expression vectors encoding either wild-type or mutant MORC2. Fewer than 30% of the cells were transduced (mCherry^{+}) in each case, thereby ensuring that the vast majority of cells (>95%) expressed just a single copy of the exogenous MORC2 construct. The restoration of repression of the GFP transgene among the mCherry^{+} populations was then followed over the course of 12 days (Figure 3).

Table S1. Assertion criteria for variant classification

	Thr24Ile	Glu27Lys[a]	Ser87Leu[a]	Ala88Val	Arg132Cys[a]	Arg266Ser	Ser388Arg	Tyr394Cys	Val413Phe
GRCh37/hg19	chr22:31354678	chr22:31354670	chr22:31345795	chr22:31345792	chr22:31342360	chr22:31337446	chr22:31334102	chr22:31334085	chr22:31333934
coding DNA[b]	c.71C>T	c.79G>A	c.260C>T	c.263C>T	c.394C>T	c.798G>C	c.1164C>G	c.1181A>G	c.1237G>T
ClinVar submission	SCV001134972.1	SCV001134974.1	SCV000618293.2	SCV000999384.1	SCV000571490.3	SCV000573276.4	SCV001134979.1	SCV000589765.2	SCV001134978.1
gnomAD	absent	absent	absent	absent	absent	absent	absent	absent	absent
Provean	damaging	benign	damaging	benign	damaging	damaging	damaging	damaging	damaging
CADD	17.9	19.5	27	25.4	26.3	26.3	25.9	26.1	25.2
MutationTaster2	damaging	damaging	damaging	damaging	damaging	damaging	damaging	damaging	damaging
Domain[c]	GHKL domain	GHKL domain	GHKL domain, ATP lid	GHKL domain	GHKL domain	transducer-like domain	transducer-like domain	transducer-like domain	transducer-like domain
ACMG criteria applied[2]	PS2, PS3[c], PM2, PP3, PP4	PS2 (x4), PS3[c], PM2, PP4, BP4	PS2 (x3), PS3[c], PM2, PP3, PP4	PS2, PM2, PP3, PP4	PS2 (x3), PS3[c], PM2, PP3, PP4	PS2, PS3[c], PM2, PP3, PP4	PS2, PM2, PP3, PP4	PS2 (x2), PM2, PP3, PP4	PS3[c], PM2, PP3, PP4
ACMG class	PATH	PATH	PATH	LPATH	PATH	PATH	LPATH	PATH	LPATH

a DDD Study (2017)[1] reported *de novo* variants in individuals with developmental delay but no detailed clinical information was provided. Those cases were not considered for variant classification.
b NM_001303256.2 transcript
c Functional assays presented in this paper. Please note that the ClinVar variant class and/or evidence details at the time of this submission (April, 2020) does not reflect the use of this criteria, pending publication.

Supplemental References

1. Deciphering Developmental Disorders Study. (2017). Prevalence and architecture of de novo mutations in developmental disorders. *Nature* 433-438.

2. Richards S., Aziz N., Bale S., Bick D., Das S., Gastier-Foster J., Grody W.W., Hegde M., Lyon E., Spector E., et al. (2015). Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology. *Genet. Med.* 405-24.