The American Journal of Human Genetics

Supplemental Data

Loss-of-Function Mutations in *CAST* Cause Peeling Skin, Leukonychia, Acral Punctate Keratoses, Cheilitis, and Knuckle Pads

Zhimiao Lin, Jiahui Zhao, Daniela Nitoiu, Claire A. Scott, Vincent Plagnol, Frances J. D. Smith, Neil J. Wilson, Christian Cole, Mary E. Schwartz, W. H. Irwin McLean, Huijun Wang, Cheng Feng, Lina Duo, Eray Yihui Zhou, Yali Ren, Lanlan Dai, Yulan Chen, Jianguo Zhang, Xun Xu, Edel A. O'Toole, David P. Kelsell and Yong Yang

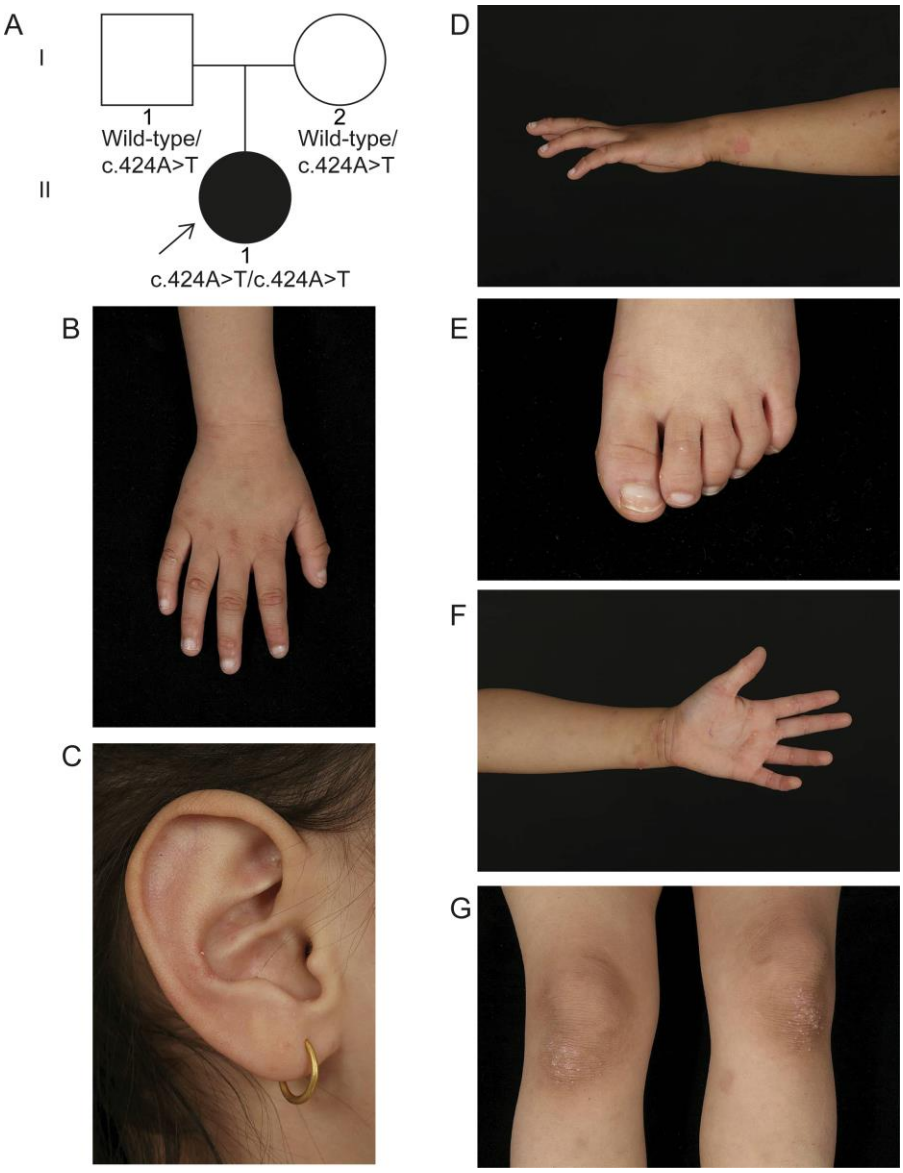

Figure S1. Further Details of the Clinical Phenotype of Individual 2.

(A) Pedigree of the family of individual 2 showing mutation segregation. The arrow indicates individual 2. (B) Overview of dorsum of hand showing knuckle pads on proximal and distal interphalangeal joints and leukonychia. (C) Scaly papule right ear antihelix. (D) Skin peeling with residual erythema left forearm. (E) Left foot showing leukonychia and papules with hyperkeratotic micropapules left second toe. (F) Punctate keratoses palm, most prominent along distal palmar creases. (G) Hyperkeratotic papules extensor surface of knees.

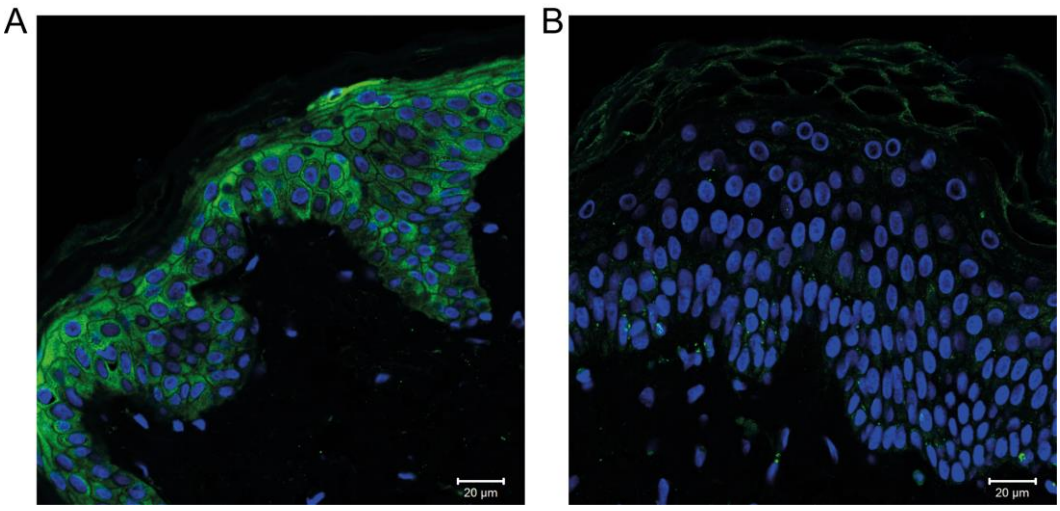

Figure S2. Immunohistochemical Staining of Calpastatin in the Epidermis of Individual 2 and the Normal Control

(A, B) Immunofluorescent staining of calpastatin (green) showed that calpastatin labeling is reduced in individual 2 (B) compared to normal control (A). DAPI was used to stain the nuclei (blue).

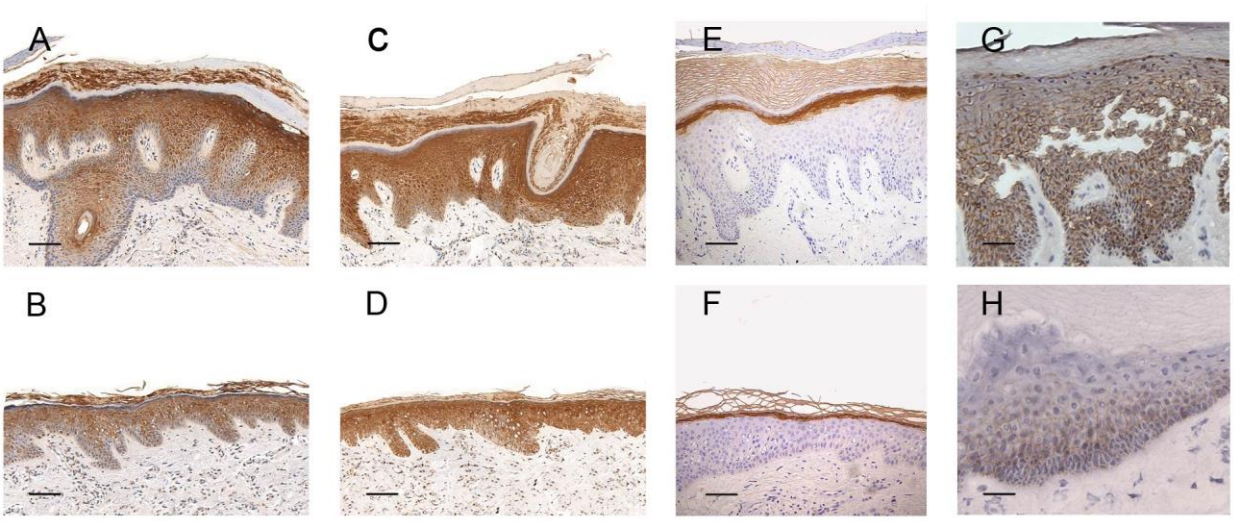

Figure S3. Immunohistochemical Staining of Terminal Differentiation Markers and Desmoplakin in the Epidermis of Individual 1 and the Normal Control

Keratin 1 (A), keratin 10 (C) and loricrin (E) of individual 1 showed normal distribution with slightly increased staining in the upper epidermis compared with the normal control (B, D, F). Desmoplakin abundance was upregulated in individual 1 with an abnormal cytoplasmic localization (G) compared to the normal control (H). The scale bars on all images represent 50 µm.

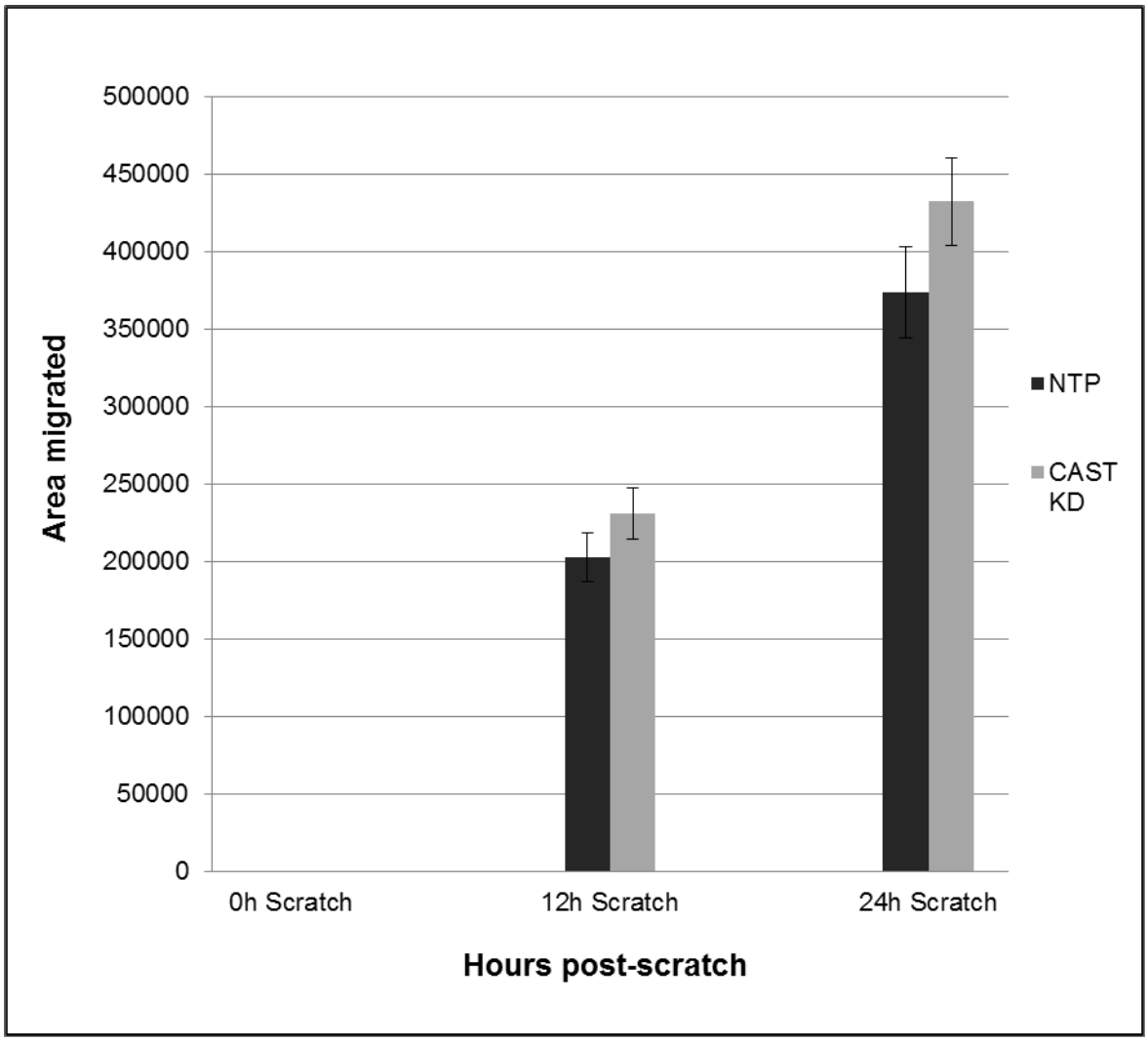

Figure S4. Scratch Assay

No significant difference was observed between NTP siRNA and *CAST* siRNA treated cells suggesting that there is no significant increase in cell migration and scratch closure (n=8 from 3 independent experiments).

Table S1. Primer Pairs for Confirmation of *CAST* Mutations

	Forward Primers (5'-3')	**Reverse Primers (5'-3')**
c.607dup	GCTTCTTGCCTGAATGTGGC	CCATGGCCTTATTTGCTCTCC
c.424A>T	AATTTTGGGGGAAGGATTTG	ATTGCTGGGCAGTAGGAGAA
c.1750delG	AGTTAAGTGATGGCATTGTGC	CATCTCGCTAAATCATCAGTC

Table S2. Primer Pair for qRT-PCR

	Forward Primers (5’-3’)	**Reverse Primers (5’-3’)**
CAST	CACAGTGCCAGATGATGCT	TCCTCAGACAAAGCATCCAGA