**List of Supplemental Material**

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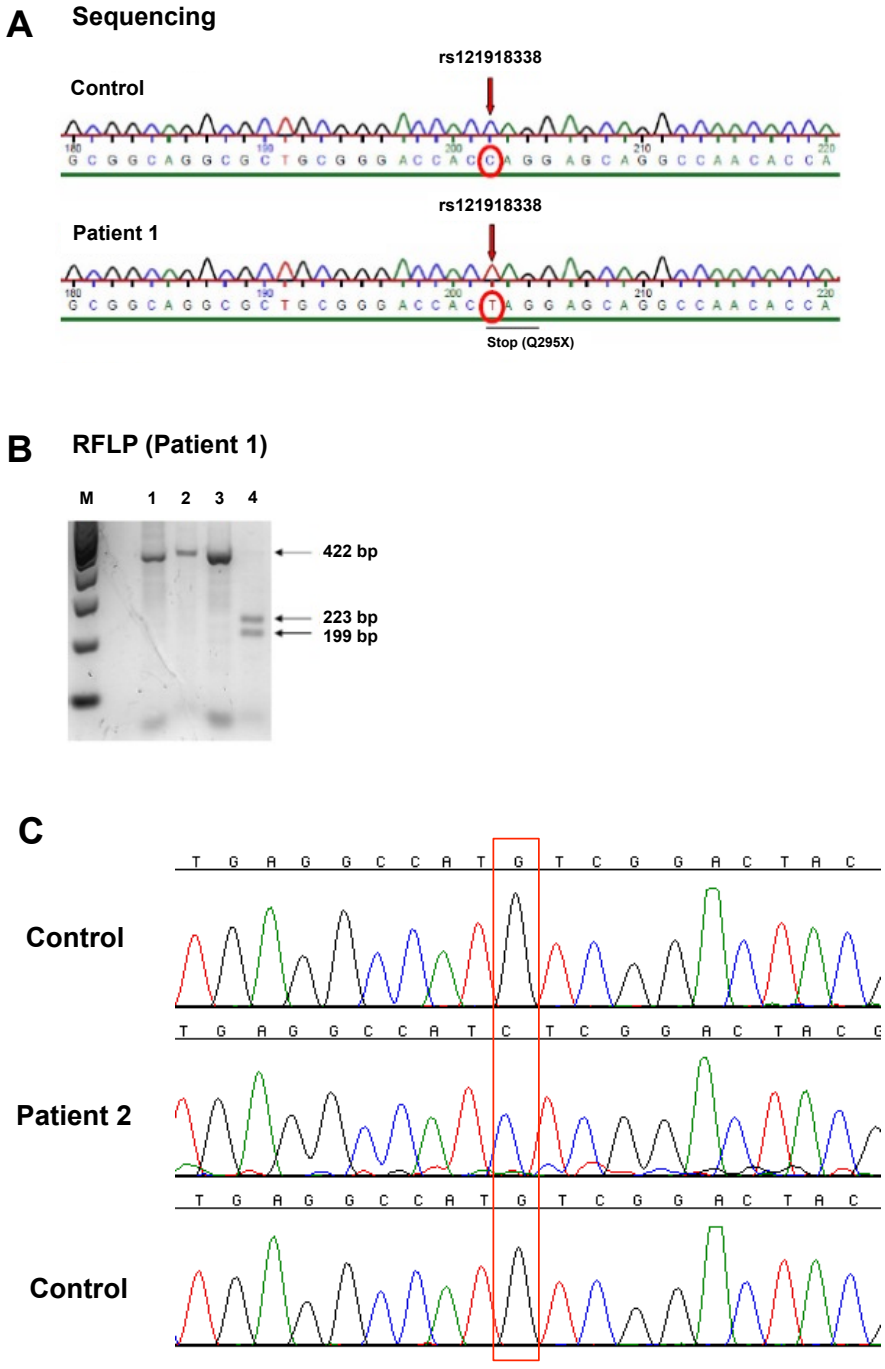
## SUPPLEMENTAL TABLES

### Supplemental Table 1. Amplification and sequencing primers used for the identification of the c.3G>C *CARD9* (Caspase Recruitment Domain Family Member 9) mutation in Patient 2.

| Amplification primers | Primer sequence | Annealing temperature | Product size |
|---|---|---|---|
| x1-2amp 662F | CTGAGATGCTGGCGAGGGCGAGGGTGTTCG | 68.5 | 2677 |
| x1-2amp 3338R | CGGGCCGGCTCCTTGCCTGTGACCTTCTTGTA | | |
| x3-7amp 2806F | GCCCCAGGCCCCAAACCCAACCAC | 67.5 | 2303 |
| x3-7amp 5108R | GCCGGAGGATTGCTTGAGGCCAGTAGTTTGA | | |
| x8-10amp 6616F | ACCGGGGGTGCAGGCTTGGGAGGAGAC | 67.8 | 1704 |
| x8-10amp 8319R | TCGCCCCATCTTCTAGCTTCATGCACCTGGTCGTT | | |
| x11-13amp 9249F | CTGGCATCTGGCGGCCTCCTGGAGCAATGA | 68.5 | 1741 |
| x11-13amp 10989R | CGCGCAGCGCAGTCCGCAGCATCC | | |

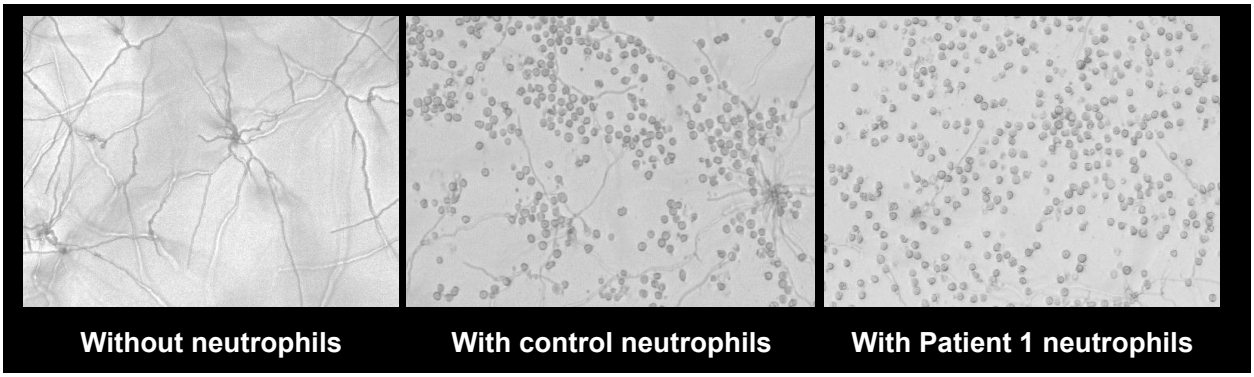
| Sequencing primers | Primer sequence |
|---|---|
| x1seq 931F | GGGGCCTCCTGGGCTGGGCGAATCAGAA |
| x1seq 1322R | AAATGAACACATCTCCGCCCAGGGGCCGTGAAAG |
| x2seq 2410F | TGGGGGATCCAGCGTGGGGATGGGACATG |
| x3seq 3507R | CGTGTTCCCCGGTGGCCCAGCTCCATCA |
| x4seq 3492F | GCCACCGGGGAACACGGCACCATCTGACAC |
| x5-6seq 3999F | AGCCTCATGAAGGCCGAGGACGACTGCAAGGTG |
| x5-6seq 4309R | AGCGCCTGCCGCCAGTCCTCCTCCAGTACCT |
| x7seq 5108R | GCCGGAGGATTGCTTGAGGCCAGTAGTTTGAG |
| x8seq 7305R | AGGCCCCCTCCATGCCCAGAACAACCTGCTAGG |
| x9seq 7384F | CTGCCCTCACAGCCCACCCACCCCATTTTCTATGA |
| x10seq 7969R | CATTGCGGCACGGGCTGCCTGTGCTGAA |
| x11-13seq 9388F | CGGGGAGGGGTGGGGCAGCCGTGCTATA |
| x11-13seq 10735R | GAGCGGGAATGCGGGTCACCCGTGCTGTTTATTTA |

## SUPPLEMENTAL FIGURES

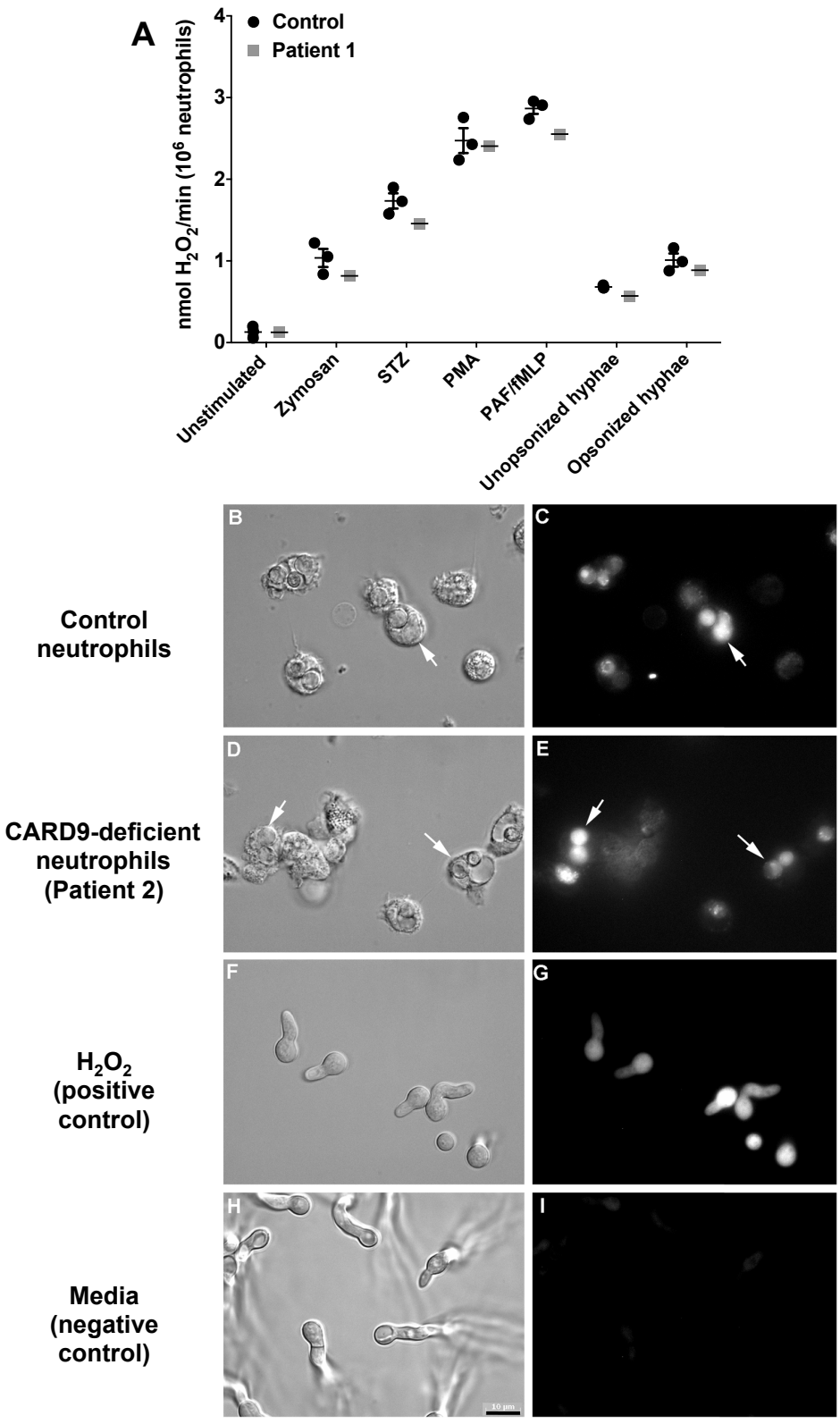



**Supplemental Figure 1. Biallelic *CARD9* (Caspase Recruitment Domain Family Member 9) mutations in the patients with extrapulmonary aspergillosis.** (A) Sequencing chromatogram of a healthy WT/WT individual (upper panel) and of Patient 1 (lower panel)

showing homozygous c.883C>T *CARD9* mutation (rs121918338). (B) The CARD9 stop codon polymorphism (rs121918338) of Patient 1 was confirmed by restriction-fragment length polymorphism (RFLP) analysis. (M) 100 bp marker, (1) undigested PCR product of a healthy WT/WT individual, (2) digested PCR product of a healthy WT/WT individual, (3) undigested PCR product of Patient 1, (4) digested PCR product of Patient 1. Digestion with BfaI (restriction site: C**T**AG). (C) Sequencing chromatogram of Patient 2 (middle panel) and two healthy WT/WT individuals (upper and lower panels) showing homozygous c.3G>C *CARD9* mutation.

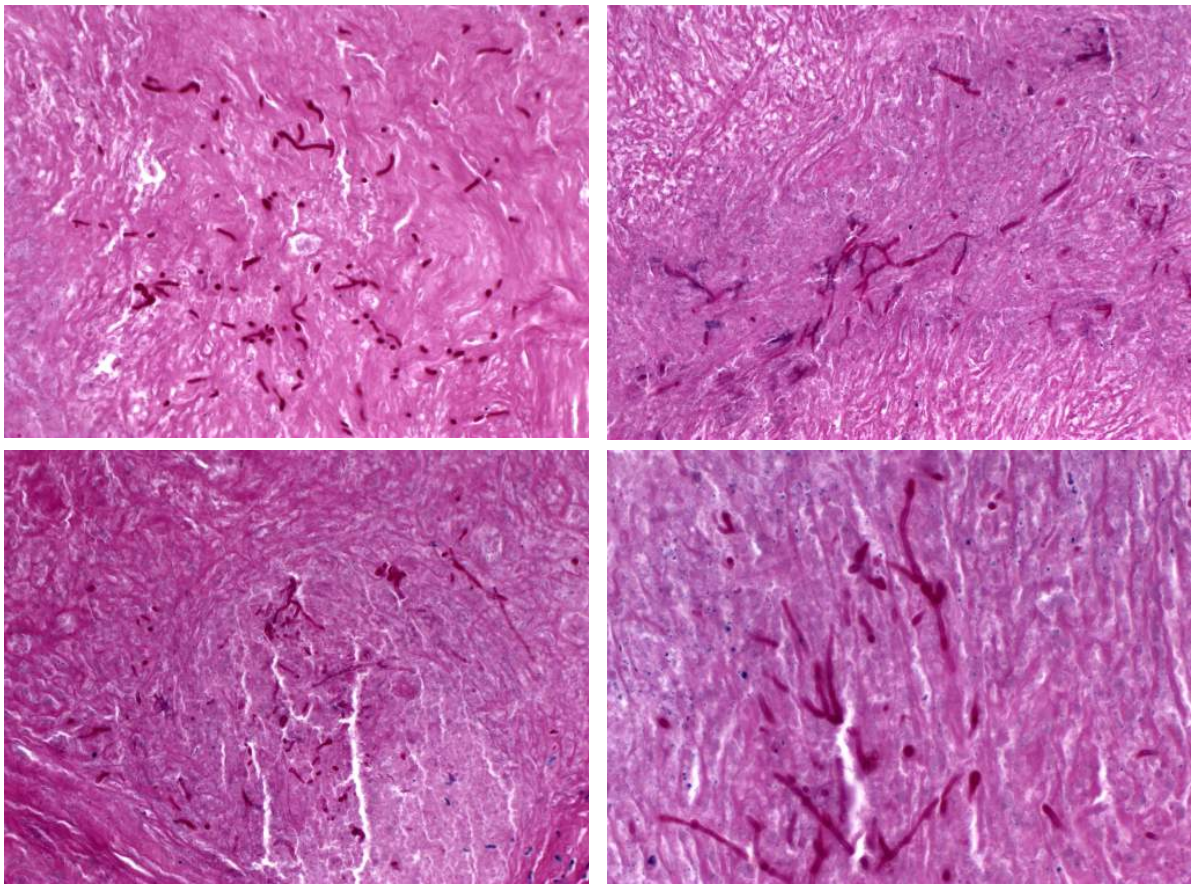



**Supplemental Figure 2. CARD9 (Caspase Recruitment Domain Family Member 9) deficiency does not impair the ability of neutrophils to prevent *Aspergillus* conidial germination into hyphae.** Representative images of *Aspergillus* hyphal germination 20 hours after *ex vivo* co-incubation of conidia with control neutrophils (middle panel), neutrophils from Patient 1 (right panel) or no exposure to neutrophils (left panel).

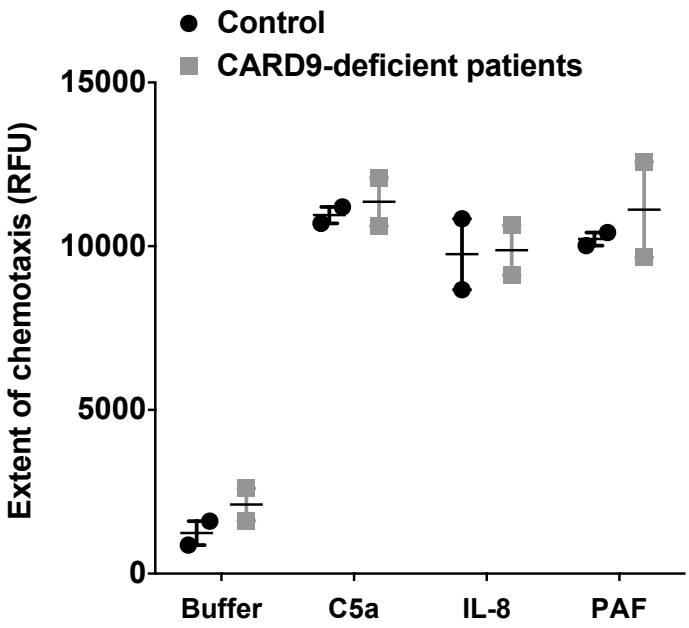



**Supplemental Figure 3. CARD9 (Caspase Recruitment Domain Family Member 9) deficiency does not impair neutrophil-mediated oxidative cytotoxicity against**

***Aspergillus fumigatus*.** (A) Neutrophils from Patient 1 do not exhibit impaired release of hydrogen peroxide toward various fungal and non-fungal stimuli *ex vivo*, determined by the Amplex Red method (n = 3 healthy donors; n = 1 experiment with Patient 1 cells). STZ, serum-treated zymosan; PMA, phorbol-12-myristate-13-acetate; PAF, platelet-activating factor; fMLP, formyl-Met-Leu-Phe. (B-I) Neutrophils from healthy donors (B and C) and Patient 2 (D and E) induce similar degrees of GFP fluorescence in the *A. fumigatus* B-5233/GFP strain upon oxidative stress (white arrows). GFP-fluorescence is induced in the *A. fumigatus* B-5233/GFP strain by 375 μM hydrogen peroxide (positive control; F and G) but not by R25 media alone (negative control; H and I). (B, D, F, and H), light microscopy. (C, E, G and I), fluorescence microscopy. All quantitative data represent mean ± SEM.



**Supplemental Figure 4. The c.883C>T *CARD9* (Caspase Recruitment Domain Family Member 9) loss-of-function mutation is associated with impaired neutrophil accumulation in the infected tissue.** Periodic acid–Schiff (PAS) stain of the mesenteric lymph node biopsy of Patient 1 shows necrotic tissue with invasion by branching hyphae, consistent with *Aspergillus* species. No neutrophil infiltration is seen. Magnification, 400× (upper left and lower right panels); 200× (upper right and lower left panels).




**Supplemental Figure 5. CARD9 (Caspase Recruitment Domain Family Member 9) deficiency does not impair neutrophil chemotaxis.** Neutrophils from Patient 1 and another CARD9-deficient patient with the c.883C>T (p.Q295X) mutation do not exhibit impaired chemotaxis toward complement component C5a ($10^{-8}$ M), IL-8 ($10^{-8}$ M) or platelet-activating factor (PAF; $10^{-7}$ M) *ex vivo*. Two healthy donors were used as controls on two independent experiments.