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Supplemental Data

A Genome-wide In Vitro Bacterial-Infection Screen

Reveals Human Variation in the Host Response

Associated with Inflammatory Disease

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Figure S1. Absence of *CARD8* in a 24-mammal phylogenetic tree correlates with large group size. A mirror tree of 24 mammalian species displays colour-coded group size on the left and presence of *CARD8* on the right. Details regarding putative orthologues can be found in Table S1. From Pagel's test of independent evolution³¹, a p-value of 0.04 is obtained from 1000 simulations. This tree contains all mammalian species with genomes in UCSC and Ensembl, including those where low genome coverage makes calls of *CARD8* presence or absence less confident. Image was exported from Mesquite³².

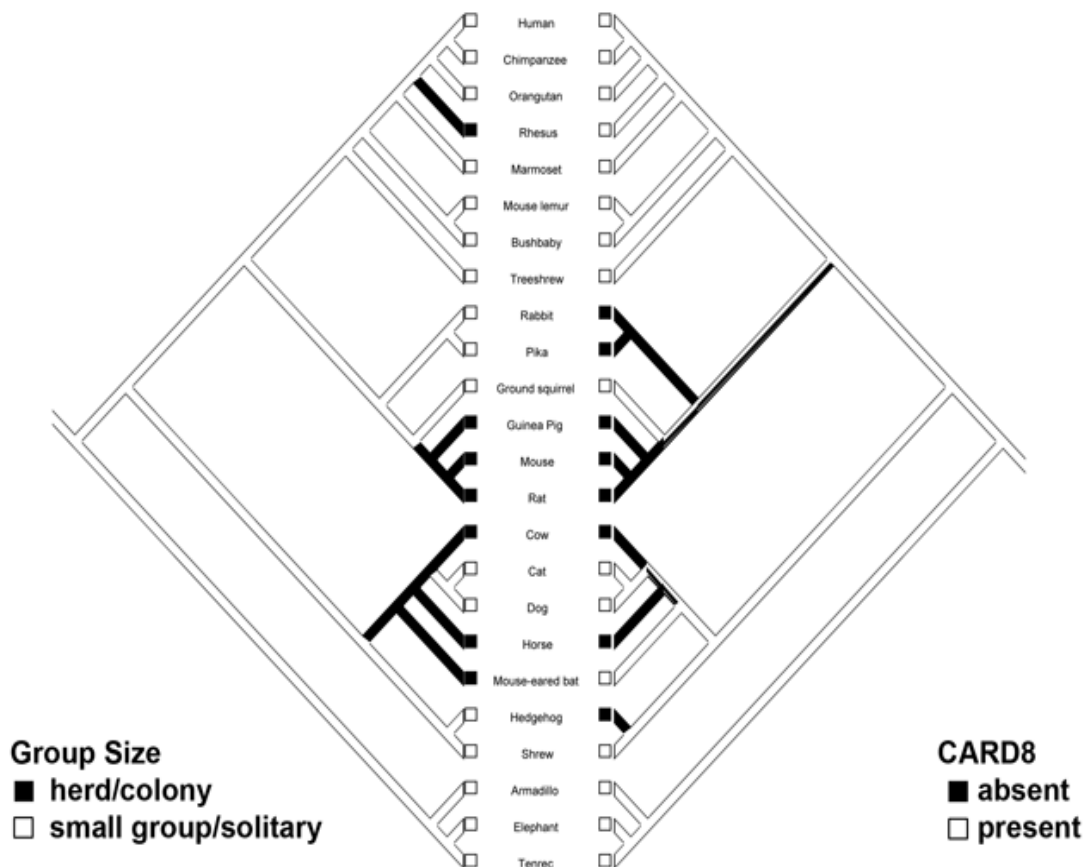


Table S1. CARD8 orthologues									
Species	Coverage ^a	Identification method	Ensembl genelink or UCSC prediction link	Chromosome or scaffold	Location	Ensembl BLAST e-value	Top Human BLAST hit ^b	Present?	Comments
<i>Primata</i>									
Homo sapiens	finished	UCSC query	ENSG00000105483	19	53493325-53450962			yes	complete; transcript is 1296bp
Pan troglodytes	6x	UCSC query	ENSPTRG00000023499	19	53825604-53932307	4.90E-222	CARD8	yes	complete
Pongo pygmaeus	6x	UCSC query	ENSPPYG00000010191	19	49661038-49778538	1.20E-203	CARD8	yes	complete
Macaca mulatta	5.1x	UCSC query	ENSMMUG00000017111	19	54486773-54499436	1.40E-173	CARD8	yes	complete
Callithrix jacchus	6x	UCSC query	Contig2244:73243-99350	Contig 2244	73243-99350	not in Ensembl	CARD8	yes	complete
Otolemur garnettii	1.5x	Ensembl query	ENSOGAG00000016636	Contig 436531	47623-78075	3.20E-136	CARD8	yes	N-terminus and three short internal stretches (~45 and ~151 bp) not yet found
Microcebus murinus	1.93x	Ensembl blast	GENSCAN00000000849	Contig 406220	534-11947	5.90E-26	NLRP1	likely yes	Predicted transcript extends 631 bp over 4 exons that align with CARD domain. Mouse lemur has a clear NLRP1 orthologue. This segment is either a part of a divergent CARD8 or a second NLRP1.
Tupaia belangeri	2x	Ensembl query	ENSTBEG00000012091	Contig 528017	57915-84614	9.10E-73	CARD8	yes	Three internal stretches (~141, ~134, ~126 bp) not yet found

Myotis lucifugus	1.7x	Ensembl blast	GENSCAN00000295729	Contig 106043	6058-15577	5.00E-51	NLRP1	likely yes	Has ~8 genscan predictions that are similar to CARD8 and NLRP1, making it difficult to assign orthology
Erinaceus europaeus	1.86x	CARD8 not present						likely no	low coverage genome
Sorex araneus	1.9x	Ensembl blast	GENSCAN00000317472	Contig 239443	4668-19938	1.10E-46	NLRP1	likely yes	1167 bp transcript, but missing N-term. Already has a clear NLRP1 orthologue.
<i>Afrotheria</i>									
Loxodonta africana	2x	Ensembl query	ENSLAFG00000003556	Contig 307228	458-43593	4.90E-131	CARD8	yes	Two internal stretches (~192 and ~45bp) not yet found
Echinops telfairi	2x	Ensembl blast	GENSCAN00000132977	conting 734181	581-752	2.50E-14	CARD8	likely yes	Only 172 bp found so far, corresponding to C-term of protein. 64% protein identity with CARD8
<i>Xenarthra</i>									
Dasyus novemcinctus	2x	Ensembl blast	ENSDNOG00000007165	Scaffold 7797	6069-29001	1.20E-63	CARD8	yes	One internal stretch of ~427bp not yet found from middle of transcript

^aDue to the low coverage in some of these genomes (2x coverage means that ~14% of the genome is likely to have been missed), calls for absence of CARD8 in these species may not prove to be accurate.

^bThe top BLAST hit using the putative CARD8 orthologue against all human proteins. NLRP1 was likely acquired as a duplication of CARD8 followed by acquisition of NACHT and LRR domains.⁵⁶

Table S2. No association between <i>CARD8</i> rs2043211 and risk of sepsis in trauma patients				
<i>Trauma patients with or without sepsis</i>	AA	AT	TT	p-value ^a
Trauma with sepsis (n=669)	317 (47.4%)	289 (43.2%)	63 (9.4%)	0.53
Trauma with SIRS without infection (n=759)	349 (46.0%)	330 (43.5%)	80 (10.5%)	

Genotypes are relative to the + strand of chromosome 19. *CARD8* coding sequence is on the - strand.

^ap-value is for the Cochran-Armitage trend test using a genotypic model with scores of 0, 0.75, and 1 for the 3 genotypes.