ELUCIDATING THE COMPLEX ROLE OF *ABCA7* IN LATE-ONSET ALZHEIMER'S DISEASE

MODEL-AD
Model Organism Development & Evaluation for Late-Onset Alzheimer's Disease

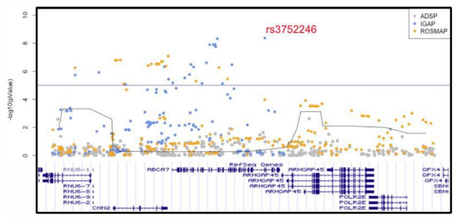
Gareth Howell[1], Kevin Kotredes[1], Ravi Pandey[3], Annat Haber[3], Greg Carter[1], Michael Sasner[1], Adrian Oblak[2], Paul Territo[2] for the IU/JAX/PITT MODEL-AD Center
[1]The Jackson Laboratory, Bar Harbor, Maine, USA; [2]Indiana University, Indianapolis, IN, USA;
[3]The Jackson Laboratory for Genomic Medicine, Farmington, CT, USA

ABSTRACT

Background: Genome-wide association studies (GWAS) identified a locus containing the ATP binding cassette subfamily A member 7 (*ABCA7*) gene as increasing risk for Alzheimer's disease (AD) (PANEL 1). ABC proteins transport various molecules across extra and intra-cellular membranes. *ABCA7* is part of the ABC1 subfamily and has high expression in hematopoietic lineage cells including natural killer cells and macrophages. More recently, ABCA7 expression has been shown in brain cells including neurons, astrocytes, microglia, endothelial cells and pericytes. However, the mechanisms by which variations in *ABCA7* increase risk for AD are not known.
Methods: The IU/JAX/PITT MODEL-AD Center prioritized the *A1527G* variant in *ABCA7* (*ABCA7*A1527G*) as a putative LOAD risk factor. CRISPR/CAS9 was used to introduce *Abca7*A1527G* variant on to B6.*APOE4.Trem2*R47H* mice (termed LOAD1, PANEL 2) and transcriptional profiling of brain hemispheres from mice at 4, 8 and 12 months old (mos) was used to evaluate molecular effects of *Abca7*A1527G* (PANEL 3). The *Abca7*A1527G* was then incorporated into B6.*APOE4.Trem2*R47H*.hAβ mice (termed LOAD2) to further evaluate the contribution of *Abca7.A1527G* to LOAD (PANEL 4). Female and male LOAD2.*Abca7*A1527G* and control mice are being characterized at 4-24 mos using an established phenotyping pipeline that includes behavior, PET/CT, multi-omics, fluid biomarkers and immunofluorescence.
Results: Brain transcriptional profiling at 12 mos showed *Abca7*A1527G* induced gene expression changes that are similar to some of those observed in human AD (PANEL 3). Enriched pathways included insulin receptor signaling, and granulocyte and neutrophil migration that include myeloid (*Trem2*, *Trem1*, *Csf1r*) and endothelial (*Pecam1*) related genes (PANEL 3). LOAD2.*Abca7*A1527G* mice showed no cognitive deficit at 12 mos but an uncoupling of brain glycolysis and regional perfusion was observed in a sex- and age-dependent manner (PANELS 4-5). Consistent with the uncoupled phenotype, IL6, IL10, and TNFα were elevated in plasma. Interestingly, TNFα, primarily expressed by microglia in the brain, was decreased. Also, by 12 mos, the density of neurons, astrocytes and microglia were reduced. Additional timepoints (18-24 mos), and assessment of LOAD2.*Abca7*A1527G* mice fed a high fat diet, are in progress.
Conclusions: Data support *ABCA7*A1527G* as a risk for LOAD that exerts its effect through interactions between cerebrovasculature, microglia and peripheral immune cells.

PANEL1: GENOME WIDE ASSOCIATION STUDIES IDENTIFY ASSOCIATION BETWEEN *ABCA7* LOCUS AND ALZHEIMER'S DISEASE

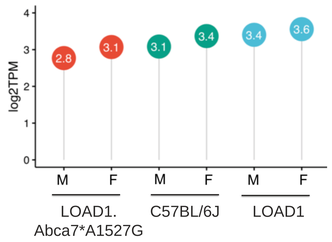
A missense variant (rs3752246, p.Gly1527Ala, OR = 1.2) was identified in a large-scale GWAS at a genome-wide significance level (p-value= 5.0×10−7) [2]. This eQTL (https://gtexportal.org/home/snp/rs3752246) variant encodes a glycine to alanine substitution at amino acid position 1527 in exon 32 of the canonical transcript and is associated with decreased expression of ABCA7. transcripts in multiple brain regions.



PANEL 2: CREATION AND VALIDATION OF THE *Abca7*A1527G* ALLELE

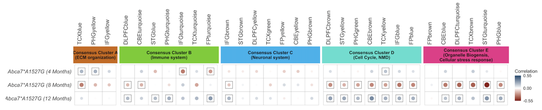
The ABCA7*A1527G allele was introduced into the LOAD1 mouse strain (B6.*APOE4.Trem2*R47H*; **LOAD1.*Abca7*A1527G***, available as JAX Stock No. 028709) by utilizing CRISPR/cas9 endonuclease mediated genome editing. Based on human to mouse RNA and protein alignments, the equivalent amino acid in the mouse is 1541 in exon 32. For consistency and ease of comparison, we refer to this variant as *Abca7*A1527G*.

Cohorts of male and female LOAD1.*Abca7*A1527G,* LOAD1 and B6 controls mice were aged to 3 months and *Abca7* expression levels in brains assessed by RNA-seq.

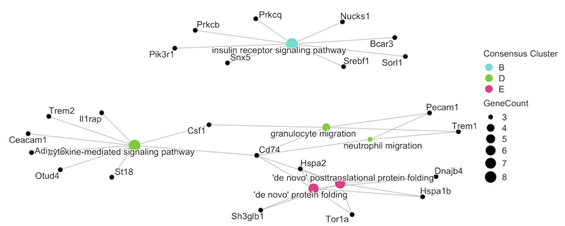


PANEL 3: *Abca7*A1527G* MODIFIES GENES RELEVANT TO LOAD AT 12 MONTHS

Cohorts of male and female LOAD1.*Abca7*A1527G* mice were aged to 4, 8 and 12 months and LOAD-relevant genes assessed in the brains usinf a Nanostring panel. Alignment to human AD data show *Abca7*A1527G* causes changes in AMP-AD clusters relating to the immune system, cell cycle, organelle biogenesis and cellular stress response.

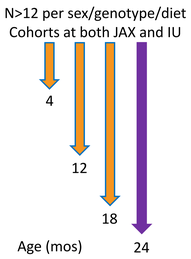


Gene set enrichment and network analysis predicts perturbations in insulin signaling, granulocyte and neutrophil migration, and cytokine mediated signaling.



PANEL 4: PHENOTYPING LOAD2.*ABCA7*A1527G*

To further evaluate the role of *Abca7*A1527G* in LOAD, we created a quadruple homozygous strain B6.*APOE4.Trem2*R47H.hAbeta.Abca7*A1527G* (**LOAD2.*Abca7*A1527G***, available as JAX Stock No. 036243). Cohorts of male and female LOAD2.*Abca7*A1527G* and LOAD2 controls are being characterized using a combination of a cross-sectional and longitudinal design.

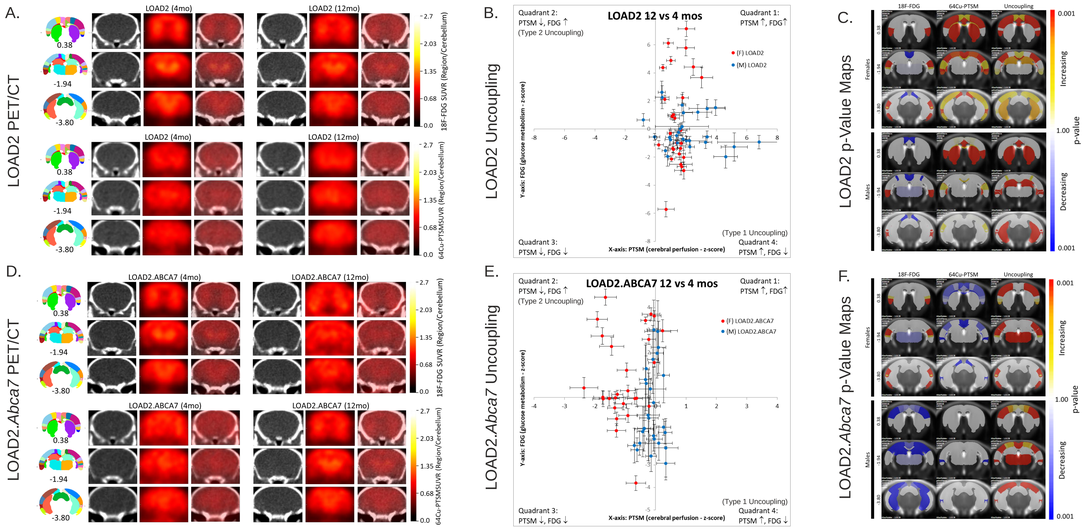


Longitudinal
Neuroimaging (MRI)
T2-weighted imaging
Diffusion MRI
Behavior and Cognition
Frailty, Y maze, activity
Biomarkers
Blood and CSF

Cross-sectional
Neuroimaging (PET/CT)
64Cu-PTSM (vascular), 18F-FDG (glucose uptake)
Multi-Omics
Transcriptomics, Proteomics, Metabolomics
Neuropathology
Amyloid and Tau, Neurodegeneration, Neuroinflammation

PANEL 5: PET/CT REVEALS *Abca7*A1527G* DRIVES UNCOUPLING OF GLUCOSE UPTAKE AND BLOOD FLOW

Glucose uptake (18F-FDG) and tissue perfusion (64Cu-PTSM) were determined in 4mo and 12mo female (red) and male (blue) LOAD2 (**A-C**) and LOAD2.*Abca7*A1527G* (**D-F**) for all brain regions. Uncoupling analysis (**B,C,E,F**) of 28 brain (56 bilateral) regions in male (blue) and female (red) mice in 12mo vs 4mo for LOAD2 (B) and LOAD2.*Abca7*A1527G* (E). Univariate statistical modeling of the regions to the 4 mo controls are shown at 3 bregma targets for LOAD2 (C) and LOAD2.*Abca7*A1527G* (F). Data are presented as mean±StDev, and show Type 1 uncoupling (quadrant 4) for LOAD2 and Type 2 uncoupling (quadrant 2) for LOAD2.*Abca7*A1527G*.



CONCLUSIONS

- Transcriptional profiling predicts the *Abca7*1527G* variant increases risk for LOAD through perturbations in a variety of processes including immune, vascular, cell stress responses, insulin signaling, and granulocyte function.
- The combination of *APOE4*, Trem2*R47H, hAβ, and *Abca7*A1527G* variants induced sex-, age-, and genotype-dependent LOAD-relevant neurovascular uncoupling which is accelerated relative the LOAD2 base strain.
- See P4-003 (Rizzo *et al*) for more details on LOAD2

FURTHER INFORMATION

- gareth.howell@jax.org and pterrito@iu.edu
- MODEL AD: model-ad.org
- AD Knowledge portal: adknowledgeportal.org
- AD Data Explorer: modeladexplorer.org

ACKNOWLEDGEMENT

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