Genome-Wide Association Study of Alzheimer Disease Related Endophenotypes across Clinical Stages of Disease
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**Introduction:** Previous genome-wide association (GWA) studies have considered endophenotypes to understand neuropathological mechanisms of Alzheimer’s disease (AD). However, comparison of the influence of genetic susceptibility factors at pre-clinical and clinical stages of AD has not been fully investigated.

**Methods:** We conducted GWA studies for cerebrospinal fluid (CSF) levels of amyloid-beta-42 (Aβ_{42}) and tau, brain MRI-measured hippocampal volume (HPV), and the logical memory test score of immediate recall (LMT) in samples of AD (n=190), mild cognitively impaired (MCI, n=581), and cognitively normal (CN, n=305) participants of the Alzheimer’s Disease Neuroimaging Initiative (ADNI). Top ranked SNPs (p<5x10^{-6}) were examined for potential function and pathway analysis seeded with genes encoded by the significant SNPs was performed using IPA software. Top-ranked GWA findings in or near coding regions were mapped on 3D protein structures obtained from the protein data bank (PDB). We used Kaplan-Meier (KM) survival analysis to estimate the AD conversion rate according to genotypes of the top-ranked SNPs. Also, we predicted the levels of Aβ_{42} and tau on age using linear mixed effects modeling including all available longitudinal CSF measures in order to see different rates of endophenotypic changes over time by genotypes of the top ranked SNPs.
**Results:** Gnome-wide significant (GWS, p<5x10^{-8}) association was observed with LOC286114 and APOE for Aβ_{42} level in the total sample. GWS association was observed for Aβ_{42} with SNPs in GRIN2B rs74442473 (p-value in meta-analysis [p] =3x10^{-8}) and NACA2 rs2378873 (p=2x10^{-8}) in CN subjects, and for ERBB4 rs839506 (p=1.8x10^{-7}) in AD subjects. GWS associations were also found for LMT with NRG1 SNP rs10095844 in MCI subjects (p=2x10^{-9}) and for HPV with AKAP9 SNP rs149454736 in AD subjects (p=7x10^{-9}). The GWS SNPs in GRIN2B, NACA2, and NRG1 are significant cis-acting eSNPs (p<0.05) in hippocampus and frontal cortex. Pathway analysis demonstrated that the top ranked genes are involved in function of neuronal cells and significantly enriched in nNOS signaling, reelin signaling, and glutamate receptor signaling pathways (p<6.9x10^{-3}).

The top-ranked ERBB4 SNPs are located in exon 3 which encodes residues located close to the site that binds with NRG1. Subjects with the AD risk genotypes of ERBB4 SNP rs89506 showed significantly steeper decline of CSF Aβ_{42} level over time. Subjects with AD-related risk genotypes for the other loci identified in our GWAS showed a higher AD conversion rate and significantly lower of CSF Aβ_{42} or higher of CSF tau levels.

**Conclusion:** We identified association of several AD-related endophenotypes with genes not previously associated with AD (NACA2 and ERBB4), as well as with several known AD risk genes GRIN2B, NRG1, and AKAP9 which contains rare missense mutations in African Americans). Our results suggest that some AD-related traits are influenced by unique sets of genetic factors during different stages of cognitive impairment. In addition, these genes are involved in neuron-specific pathways. Our study also suggests that gene influences on AD-related endophenotypes may be mechanistically specific and that each of these effects of are more prominent at certain stages of disease progression. Further studies of these genotype-
endophenotype-stage associations may lead to the development of pre-clinical biomarkers for AD.