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Pleiotropic Effects of CSF Levels of Alzheimer's Disease Proteins

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Background & Motivation

Cerebrospinal fluid (CSF) analytes harbor potential as diagnostic biomarkers for Alzheimer's Disease (AD). Quantitative measures of CSF proteins comprise a set of often highly correlated endophenotypes that have previously shown promise in genetic analyses (Cruchaga et al., 2013; Kauwe et al., 2014). Pleiotropic impact of genetic variations on this set may provide additional insights into AD pathology at its earliest stages. To determine which specific endophenotypes are pleiotropic, one can employ methods based on the reverse regression of genotype on phenotypes. Recently, we proposed a method based on functional linear models (Vsevolozhskaya et al, 2016) that utilizes reverse regression and simultaneously evaluates all variants within a genetic region for an association with multiple correlated phenotypes. Here we apply our novel methodology to explore pleiotropic eff ects of CSF analtyes using Alzheimer's Disease Neuroimaging Initiative (ADNI) data.

Materials

- CSF levels of β-amyloid (Aβ) and τ proteins are potentially early diagnostic markers for probable AD. We investigated the influence of genetic variation on these markers for two candidate genes: *APOE* and *CLU*.
- Data were obtained from the ADNI database. A total of 309 (AD = 79, MCI = 148, NL = 82) individuals were included.
- 526 *APOE* single nucleotide polymorphism (SNP) and 569 *CLU* SNPs were available for the analysis.

Normal (n = 82)	Mean MCI (n = 148)	Mean AD (n = 79) Mean
Age (year)	75.0	74.4	74.3
CSF Aβ levels (pg/ml)	205.5	161.9	144.9
CSF tau levels (pg/ml)	70.5	102.1	120.7
	N	**N**	**N**
Male	45	101	44
Female	37	47	35

Table 1: Demogphic, clinical and biomarker data for ADNI subjects.

Methods

- Our method is gene- or region-based. What makes it stand apart is the fact that a genetic region serves as an outcome in the model, not a predictor (i.e., the so-called reverse regression).

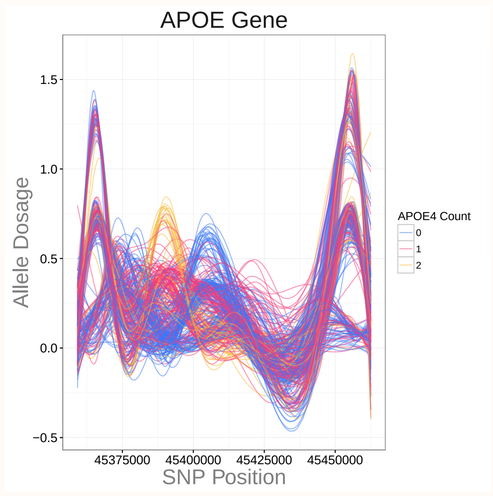

Figure 1: Smoothed allele counts within the *APOE* region for 309 subjects.

- The genetic information does not enter the model as discretized allele counts (e.g., 0, 1, 2) but rather as a smooth function $G(t)$, with t indexing a genetic variants' position over a genetic region.
- Fig. 1 illustrates genotypic functions fitted for each subject, $G_i(t), i = 1, \ldots 309$, over the *APOE* region.

Statistical Analysis

- To examine the effect of SNPs on the 2 CSF biomarkers, we fit the following functional linear model:

$$E[G_i(t)] = \beta_0(t) + \beta_1(t)X_{Tau_i} + \beta_2(t)X_{Abeta_i} + \beta_3(t)X_{\epsilon 4}$$
$$\beta_4(t)X_{Sex_i} + \beta_5(t)X_{Age_i} + \beta_6(t)X_{Diag_i} + \sum_{j=1}^{3}\beta_{6+j}(t)X_{PC_{ij}}.$$

- The model is looking for an association with a CSF biomarker, while adjusting for the level of the other protein, gender, age and diagnosis status. Also, to ensure the significance is not due to population stratification, we incorporated the top three principal components to further control for the population structure.
- Two analyses were performed: with *APOE* $\epsilon 4$ genotype (number of APOE $\epsilon 4$ alleles; 0, 1, 2) as a covariate and without.
- The analysis is Bayesian; the estimated $\hat{\beta}_j(t)$'s are obtained as posterior expectations; the confidence bands are obtained as 95% posterior crediable intervals.

Results: Exploring the estimated $\hat{\beta}_{Tau}(t)$ and $\hat{\beta}_{Abeta}(t)$

APOE: no $\epsilon 4$ adjustment

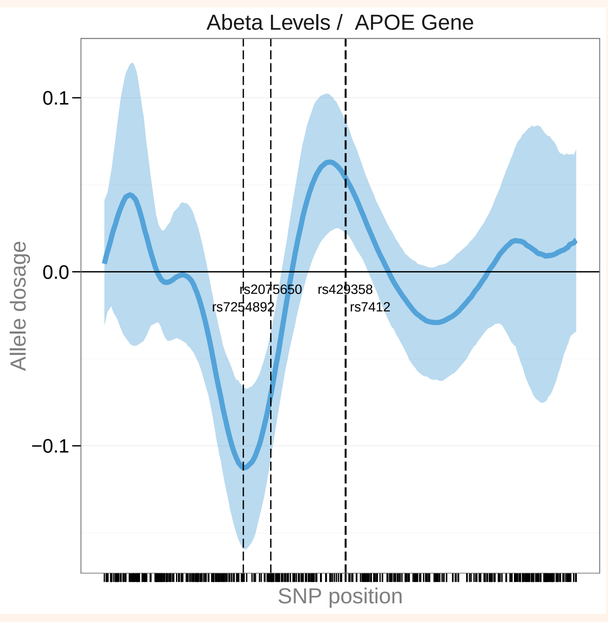

Figure 2: Estimated posterior expectation of *APOE* association with Aβ biomarker and the corresponding 95% credible intervals.

APOE genotype is the established risk factor for LOAD. The two previously identified *APOE* SNPs (rs429358, rs7412) that define the $\epsilon 2/\epsilon 3/\epsilon 4$ alleles for AD susceptibility are among the top SNPs that drive the association with the elevated Aβ levels in our model. Additionally, *TOMM40/APOE* rs7254892 that was recently identified to be associated with longer human longevity (Lin et al., 2016) is clearly associated with lower Aβ levels in our analysis. *TOMM40/APOE* rs2075650 is also associated with lower Aβ levels under the functional inference, confirming prior report by Kim et al., 2011.

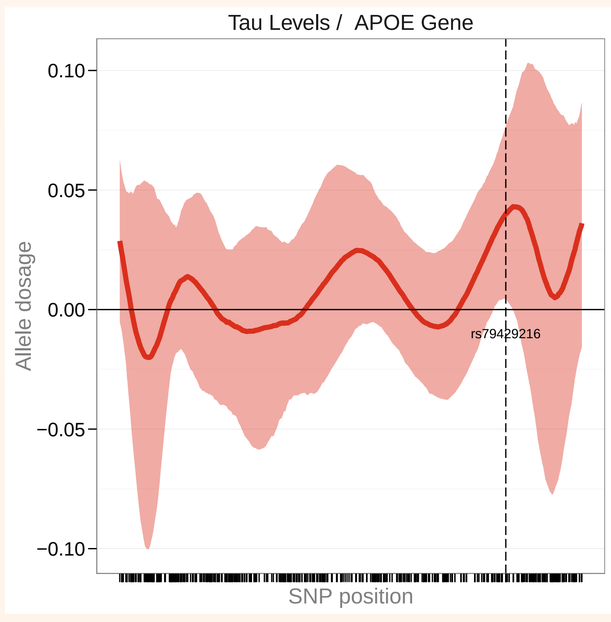

Figure 3: Estimated posterior expectation of *APOE* association with τ biomarker and the corresponding 95% credible intervals.

Cervantes et al., 2011, tested *APOE* rs79429216 and rs76214972 for an association with AD, but after multiple test correction their allelic P-value were found to be non-significant (0.747 and 0.053 respectively). Nonetheless, in our analysis, the credible interval for the genetic effect of both of these SNPs does not capture zero and they were found to be associated with the elevated τ levels.

APOE: $\epsilon 4$ adjustment

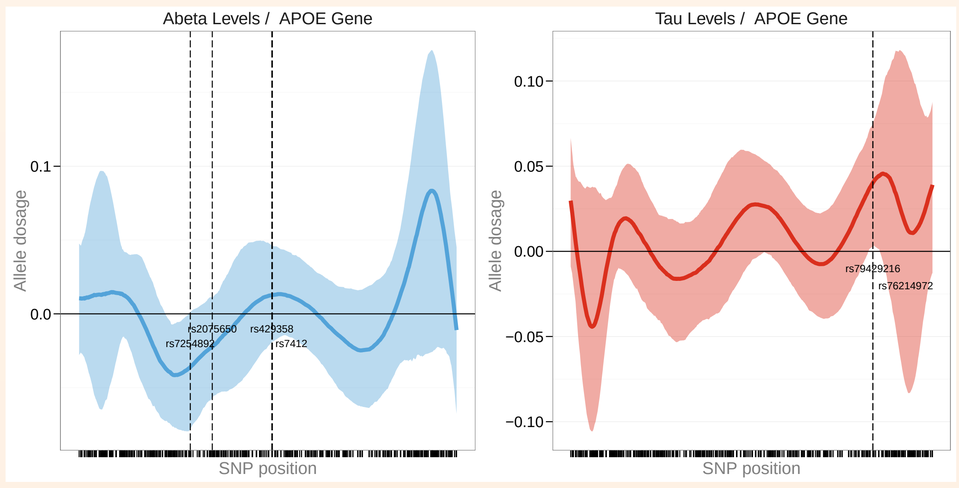

Figure 4: Estimated posterior expectation over *APOE* after $\epsilon 4$ covariate adjustment.

- The $\epsilon 4$ adjustment did not have a significant impact on the association between τ levels and AD; for Aβ the region over *APOE* SNPs rs429358, rs7412 is not longer significant. Nonetheless, the second interval still contains a statistically significant signal

CLU gene

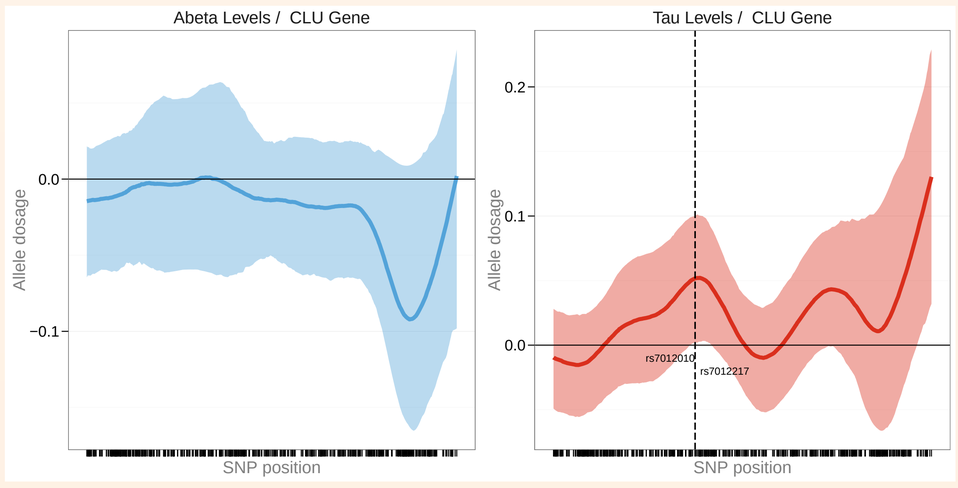

Figure 5: Estimated posterior expectation of *CLU* association with Aβ biomarker, τ biomarker and the corresponding 95% credible intervals.

- No *CLU* SNPs were found to be associated with Aβ levels
- Two markers (rs7012010, rs7012217) are in the constellation of SNPs that are associated with the elevated τ levels. Both of these SNPs were previously reported