The Effect of APOE Gene SNPs on Brain Tissue Specific Gene Expression

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Introduction and Objective

The gene *Apolipoprotein E* (*APOE*) has been found to be significantly associated with longevity and age-related diseases. The 3 major *APOE* alleles $\varepsilon 2$, $\varepsilon 3$, and $\varepsilon 4$ are derived from genotypic combinations of single nucleotide polymorphisms (SNPs) rs7412 and rs429358. Populations with high frequencies of the $\varepsilon 4$ allele are more susceptible to Alzheimer's disease (AD) and cognitive decline, while the $\varepsilon 2$ allele serves as a neuroprotective factor. The *APOE* gene, however, may have other significant undiscovered effects on age-associated diseases through its interaction with other genes.

In this project, we investigated the effect of *APOE* alleles on brain tissue-specific gene expression in hopes of finding highly associated genes that may modify AD risk by interaction. We chose to investigate brain tissues because of the previously found link between *APOE* and AD.

Methods

1. We used normalized RNA-seq expression data from 13 different brain tissues, including: the Amygdala, Anterior Cingulate Cortex, Caudate, Cerebellar Hemisphere, Cerebellum, Cortex, Frontal Cortex, Hippocampus, Substantia Nigra, Hypothalamus, Nucleus Accumbens, Putamen, and Spinal Cord.
2. The data were obtained from the v7 release of the Genotype-Tissue Expression (GTEx) portal database, representing 80 to 154 subjects—depending on the brain tissue site. APOE genotype data were also available for all subjects.
3. *APOE* genotypes were coded using the following schema.

APOE SNPS		
rs7412	**rs429358**	***APOE* allele**
T	C	$\varepsilon 1$
T	T	$\varepsilon 2$
C	T	$\varepsilon 3$
C	C	$\varepsilon 4$

***APOE* SNP combinations**			
	rs429358		
rs7412	TT	TC	CC
TT	$\varepsilon 2\varepsilon 2 \rightarrow \varepsilon 2$	.	.
TC	$\varepsilon 2\varepsilon 3 \rightarrow \varepsilon 2$	$\varepsilon 2\varepsilon 4 \rightarrow \varepsilon 4$	.
CC	$\varepsilon 3\varepsilon 3 \rightarrow \varepsilon 3$	$\varepsilon 3\varepsilon 4 \rightarrow \varepsilon 4$	$\varepsilon 4\varepsilon 4 \rightarrow \varepsilon 4$

3. For each genotype, we computed mean gene expression and log fold change values setting our reference level as $\varepsilon 3$.
4. Genes with log(fold change) $< \pm 1.5$ were removed from the data in order to remove outliers
5. With the homozygous $\varepsilon 3$ allele as a reference factor, we fit the following linear regression model using *APOE* alleles and 20 GTEx-provided covariates (such as gender) as predictors for gene expression in brain tissue.

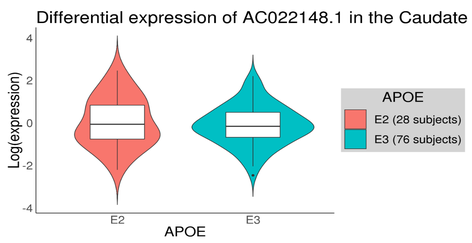
$$Log(expression) = \beta_0 + \beta_1 * (\varepsilon 2 - \varepsilon 3) + \beta_2 * (\varepsilon 4 - \varepsilon 3) + \beta_3 * (Covariate_1) + \cdots + \beta_{22} * (Covariate_{20})$$

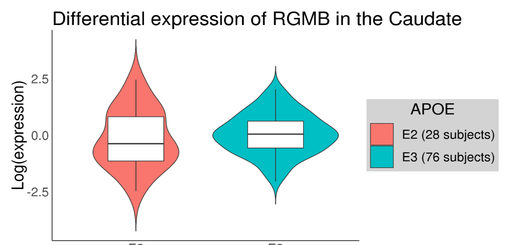
Results

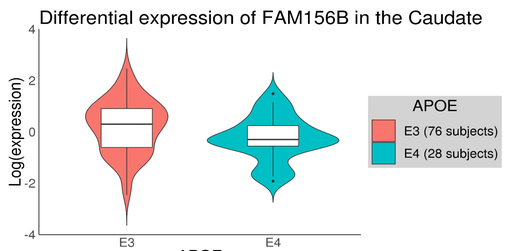
Tissue-specific associations between APOE genotype and genes (FDR 10% adjusted) are shown below. Significant results were only found in 3 tissues

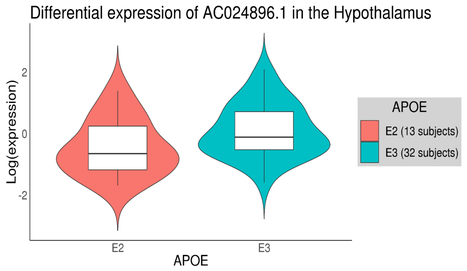
Tissue	Gene	Genotype	Estimate	Std Error	t-statistic	adj. p-value
Caudate Nucleus	*AC022148.1*	$\varepsilon 2$ v. $\varepsilon 3$	0.51	0.11	4.7	0.09
	RGMB	$\varepsilon 2$ v. $\varepsilon 3$	-0.49	0.10	-4.7	0.09
	FAM156B	$\varepsilon 4$ v. $\varepsilon 3$	-0.71	0.14	-4.9	0.07
Hypothalamus	*AC024896.1*	$\varepsilon 2$ v. $\varepsilon 3$	-1.0	0.20	-5.1	0.06
Nucleus Accumbens	*TSPAN7*	$\varepsilon 2$ v. $\varepsilon 3$	-0.47	0.09	-5.1	0.05
	LNX1	$\varepsilon 2$ v. $\varepsilon 3$	-0.35	0.07	-4.8	0.06

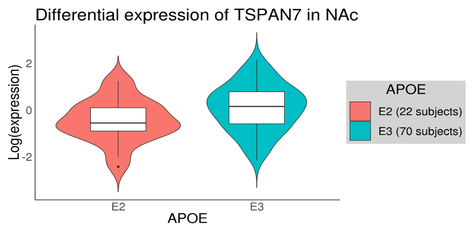
Gene	Functionality
AC022148	• Long non-coding RNA
RGMB	• Plays negative roles in breast cancer
FAM156B	• Associated with X-linked Intellectual disability
AC024896.1	• Long non-coding RNA
TSPAN7	• Associated with X-linked intellectual disability • In lung cancer, associated tumor size, and poor prognosis • Plays role in synapse development and cognition
LNX1	• Contributes to tumor growth by down-regulating p53 (tumor suppressor) stability. • Associated with synaptic transmission at electrical synapses

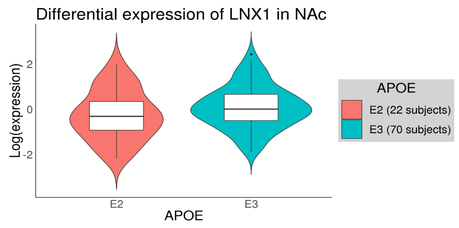












Conclusion

Based on our modelling process, we can conclude that there are indeed significant tissue-specific interactions between APOE genotypes and gene expression.

Future directions for this study may include:

- Incorporating more data
- Further in-depth analysis of the significant genes that we identified in order to assess the nature of their relationship with APOE and disease
- Study whether tissue-specific expression informs disease risk or disease risk informs expression levels
- The influence of subjects' racial profiles is also worth investigating as previous studies have revealed APOE expression to be differentially affected by race

References and Acknowledgements

1. Paola Sebastiani, Anastasia Gurinovich, et al. APOE Alleles and Extreme Human Longevity, The Journals of Gerontology: Series A, Volume 74, Issue 1, January 2019, Pages 44–51, https://doi.org/10.1093/gerona/gly174
2. Carithers, Latarsha J, Ardlie, et al., on behalf of the GTEx consortium, A Novel Approach to High-Quality Postmortem Tissue Procurement: The GTEx Project, Biopreservation and Biobanking 13(5), 2015, p 311–319.
3. Zhang A, Zhao Q, Xu D, Jiang S. Brain APOE expression quantitative trait loci-based association study identified one susceptibility locus for Alzheimer's disease by interacting with APOE ε4. *Sci Rep*. 2018;8(1):8068. Published 2018 May 23. doi:10.1038/s41598-018-26398-1
4. Wang X, Lin M, Zhao J, Zhu S, Xu M, Zhou X. TSPAN7 promotes the migration and proliferation of lung cancer cells via epithelial-to-mesenchymal transition. *Onco Targets Ther*. 2018;11:8815–8822. Published 2018 Dec 13. doi:10.2147/OTT.S167902
5. Li, Jin & Ye, Lin & Sanders, Andrew & Jiang, Wen. (2012). Repulsive guidance molecule B (RGMB) plays negative roles in breast cancer by coordinating BMP signaling. Journal of cellular biochemistry. 113. 2523-31. 10.1002/jcb.24128.
6. Park, Rackhyun & Kim, Hyunju & Jang, Minsu & Jo, Daum & Park, Yea-In & Namkoong, Sim & Lee, Jin & Jang, Ik-Soon & Park, Junsoo. (2019). LNX1 contributes to tumor growth by down-regulating p53 stability. The FASEB Journal. fj.201900366R. 10.1096/fj.201900366R.
7. Lombard, Zané & Park, Chungoo & Makova, Kateryna & Ramsay, M.. (2011). A computational approach to candidate gene prioritization for X-linked mental retardation using annotation-based binary filtering and motif-based linear discriminatory analysis. Biology direct. 6. 30. 10.1186/1745-6150-6-30.

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