Disease status affects the association between rs4813620 and the expression of Alzheimer's disease susceptibility gene *TRIB3*

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Lorenzi et al. (1) recently applied a functional prioritization method to the Alzheimer's Disease Neuroimaging Initiative dataset and successfully identified a link between tribbles pseudokinase 3 (*TRIB3*) and the stereotypical pattern of gray matter loss in Alzheimer's disease (AD). Lorenzi et al. conducted an expression quantitative trait loci (eQTL) analysis using the dataset from the Genotype Tissue Expression (GTEx) project and found that rs4813620 was the top eQTL variant for *TRIB3*. The rs4813620 variant and its proxy, rs62191440 ($D' = 0.8469$; $r^2 = 0.6559$), could regulate the expression of *TRIB3* in various tissues, including the nervous tissue proxy nerve tibial, as well as two brain tissues, the cortex and the caudate ganglia.

It is reported that disease status may have significant effects on gene expression (2–4) and it is suggested that eQTL analysis using disease samples should account for the effect of disease status, as has been shown in recent studies (2, 5). The causes of death of the donors in GTEx include traumatic injury, cerebrovascular disease, heart disease, and liver, renal, respiratory, and neurological diseases (6). In GTEx, a linear regression analysis was applied to perform the eQTL analysis using Matrix eQTL, assuming an additive model and adjusting for several critical covariates, including genotyping principal components; genotyping array platform; 15, 30, or 35 PEER (probabilistic estimation of expression residuals software) factors; and gender (6, 7). However, GTEx did not account for the effect of disease status.

Hence, disease status may affect the association between the rs4813620 variant and the expression of *TRIB3*. To confirm this view, we comprehensively evaluated the association between rs4813620 and *TRIB3* expression using the eQTL dataset from BRAINEAC (8), the Mayo eQTL dataset (5), and the Brain xQTL Serve database (9). In brief, BRAINEAC includes 10 brain regions of 134 neuropathologically normal individuals with European descent (8). In BRAINEAC, we downloaded the *TRIB3* expression data and the genotype data of genetic variants within 1 million basepairs (Mb) upstream and 1 Mb downstream of the transcription start site (8). We further evaluated the association between rs4813620 and *TRIB3* expression using a linear regression analysis (7, 10). For comparison, we downloaded the summary results from the Mayo eQTL dataset and the Brain xQTL Serve database to directly evaluate the association between rs4813620 and *TRIB3* expression. The significance level was defined as $P < 0.05$.

In BRAINEAC, the results showed no significant association between rs4813620 and *TRIB3* expression in 10 brain regions of 134 neuropathologically normal individuals (Table 1). Compared with the BRAINEAC dataset, there was significant association of rs4813620 with *TRIB3* expression in the Mayo eQTL dataset and the Brain xQTL Serve database. The BRAINEAC dataset consists of 134 neuropathologically normal individuals. Hence, BRAINEAC excluded the effect of disease status on *TRIB3* expression (8).

In summary, these findings from the BRAINEAC dataset do not support the association between rs4813620 and *TRIB3* expression in human brain tissues, as reported by Lorenzi et al. (1). Compared with BRAINEAC, the disease status in GTEx, the Mayo eQTL dataset, and Brain xQTL Serve may have caused the significant association between the rs4813620 variant and *TRIB3* expression. Hence, our findings provide important supplementary information about the role of rs4813620 in AD.

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Table 1. rs4813620 variant and *TRIB3* expression in human brain tissues

Dataset	Ref.	Disease status	Sample size, *n*	Brain tissue	*P*
BRAINEAC	8	Neuropathologically normal	134	Cerebellar cortex	0.28
				Frontal cortex	0.07
				Hippocampus	0.81
				Medulla	0.25
				Occipital cortex	0.45
				Putamen	0.94
				Substantia nigra	0.94
				Temporal cortex	0.09
				Thalamus	0.70
				Intralobular white matter	0.74
Mayo eQTL	5	AD	51	Cerebellum	4.88E-03*
		AD and other brain pathologies†	87	Cerebellum	1.38E-04
		Brain pathologies except AD	36	Cerebellum	5.76E-02
		AD	46	Temporal cortex	3.73E-03
		AD and other brain pathologies	83	Temporal cortex	7.42E-06
		Brain pathologies except AD	37	Temporal cortex	1.32E-02
Brain xQTL Serve	9	Neurodegenerative disease (96%)	496	Prefrontal cortex	1.06E-19

*$P < 0.05$.

†Other brain pathologies included progressive supranuclear palsy, Lewy body disease, corticobasal degeneration, frontotemporal lobar degeneration, multiple system atrophy, and vascular dementia (5).

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