

APOE*4 Risk-Modifying Genes and Drug Targets in Alzheimer's Disease Through Cell-type Specific Genomic Analyses

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INTRODUCTION

- APOE**4 has broad effects on Alzheimer's disease (AD) pathogenesis, which can differ across various brain cell types [1-2].
- Identifying cell-type-specific AD risk-modifying genes according to an individual's *APOE**4 status may provide new therapeutic targets.
- This study aimed to uncover such genes and potential drug targets by integrating AD genetic association analyses with single-cell multi-omics data.

METHODS

- APOE**4-stratified cell-type-specific transcriptome-wide association analyses (cTWAS) were performed using FUSION by integrating newly generated European ancestry *APOE**4-stratified AD GWAS with gene expression data derived from seven brain cell types: astrocytes, endothelial cells, excitatory neurons, inhibitory neurons, microglia, oligodendrocytes, and oligodendrocyte progenitor cells [3-4].
- Identified cTWAS genes were further prioritized using five approaches: (a) Replicate cTWAS using newly generated *APOE**4 stratified GWAS in African ancestry, (b) summary-data-based Mendelian Randomization (SMR) [5], (c) Genetic colocalization analysis (COLOC) [6], (d) single-cell ATAC-seq datasets [7], and (e) single cell differential expression gene analysis (scRNA-seq DEG) [4,8].
- Prioritized genes were further interrogated through gene-drug/compound interaction networks and drug/compound enrichment analyses using DSigDB and curated druggable gene resources to identify potential therapeutic drugs/compounds.

DISCUSSION

- APOE**4 stratified cTWAS identified 84 cell-type-gene pairs. 11/17 *APOE**4+ cell-type-gene pairs and 33/67 *APOE**4- cell-type-gene pairs were further prioritized (supported by ≥ 4 multi-omics approaches).
- The distinct distribution of prioritized genes across *APOE**4 strata suggests that AD risk is mediated through genotype-specific and cell-type-specific biological mechanisms; for example, four prioritized genes in *APOE**4 carriers include *CISD1*, *SLC23A2*, *UBXN4*, and *TNS3*, whereas in *APOE**4 non-carriers, genes such as *MAPT*, *KANSL1*, *TRAF3*, and *RPS6KA2* are prioritized.
- These findings support the potential for *APOE**4-stratified therapeutic strategies, where targeting pathways such as oxidative stress and neuroinflammation (e.g., Hydrocortisone, Trolox) may preferentially benefit *APOE**4+ individuals.

RESULTS

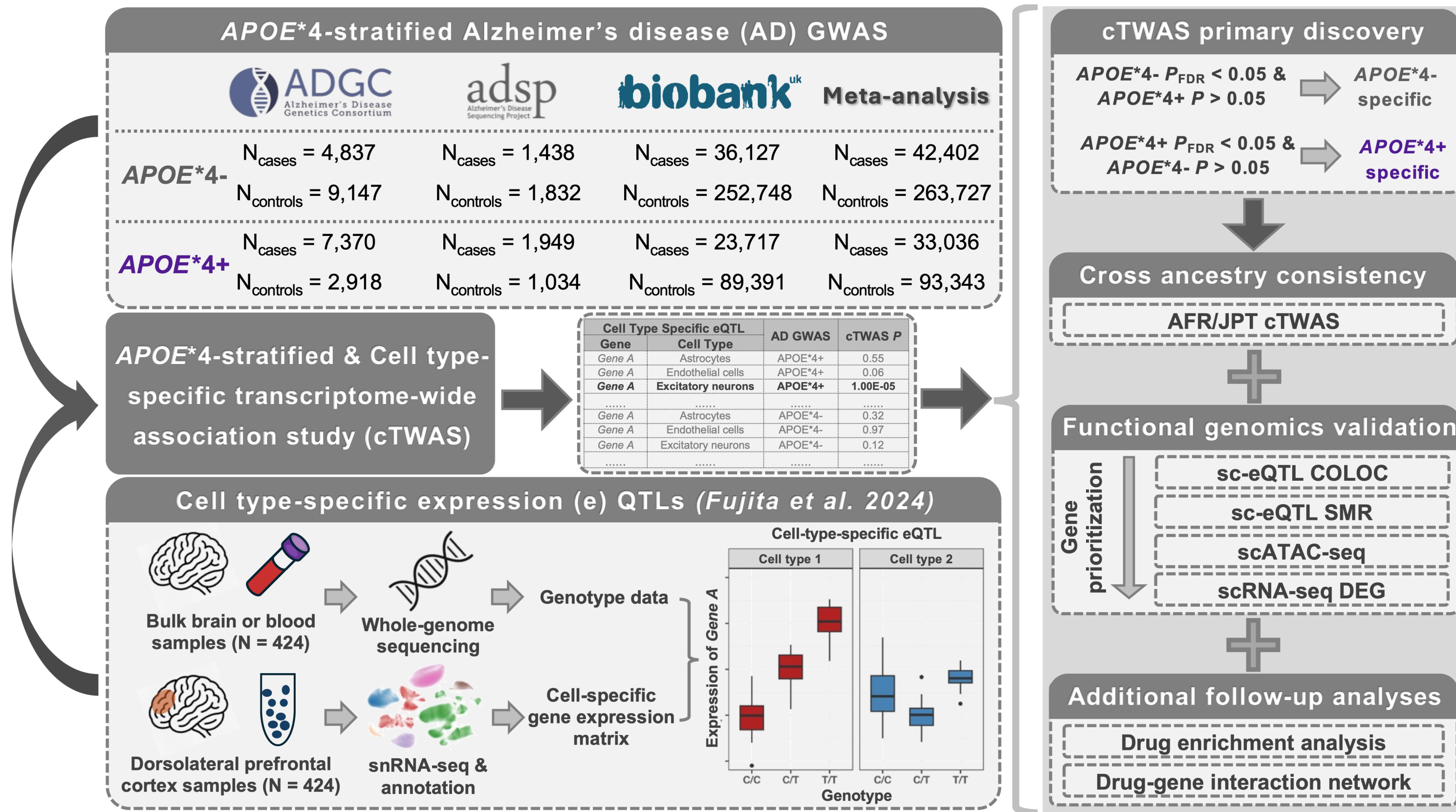


Figure 1. Schematic overview of the study design. Left) *APOE**4 stratified GWAS of AD in European ancestry subjects were integrated with snRNA-seq and paired whole genome sequencing data from ROSMAP brain tissue to perform an *APOE**4 stratified cTWAS. This approach implicates cell-type-gene pairs that are likely causal to AD in an *APOE**4 dependent manner. Right top) cTWAS findings were filtered for *APOE**4 specificity by retaining only significant ($P_{FDR} < 0.05$) genes in a given *APOE**4 stratum that were not significant ($P > 0.05$) or showed discordant effect directions in the opposite stratum. Right middle) Following cTWAS gene discoveries, multiple types of gene prioritization analyses were conducted, including cross-ancestry cTWAS, leveraging smaller AD genetic datasets in AFR and Japanese cohorts to assess consistent *APOE**4 heterogeneity effects, and a series of cell-type-specific multi-omics analyses. Right bottom) Top prioritized genes were used to perform drug repurposing analyses.

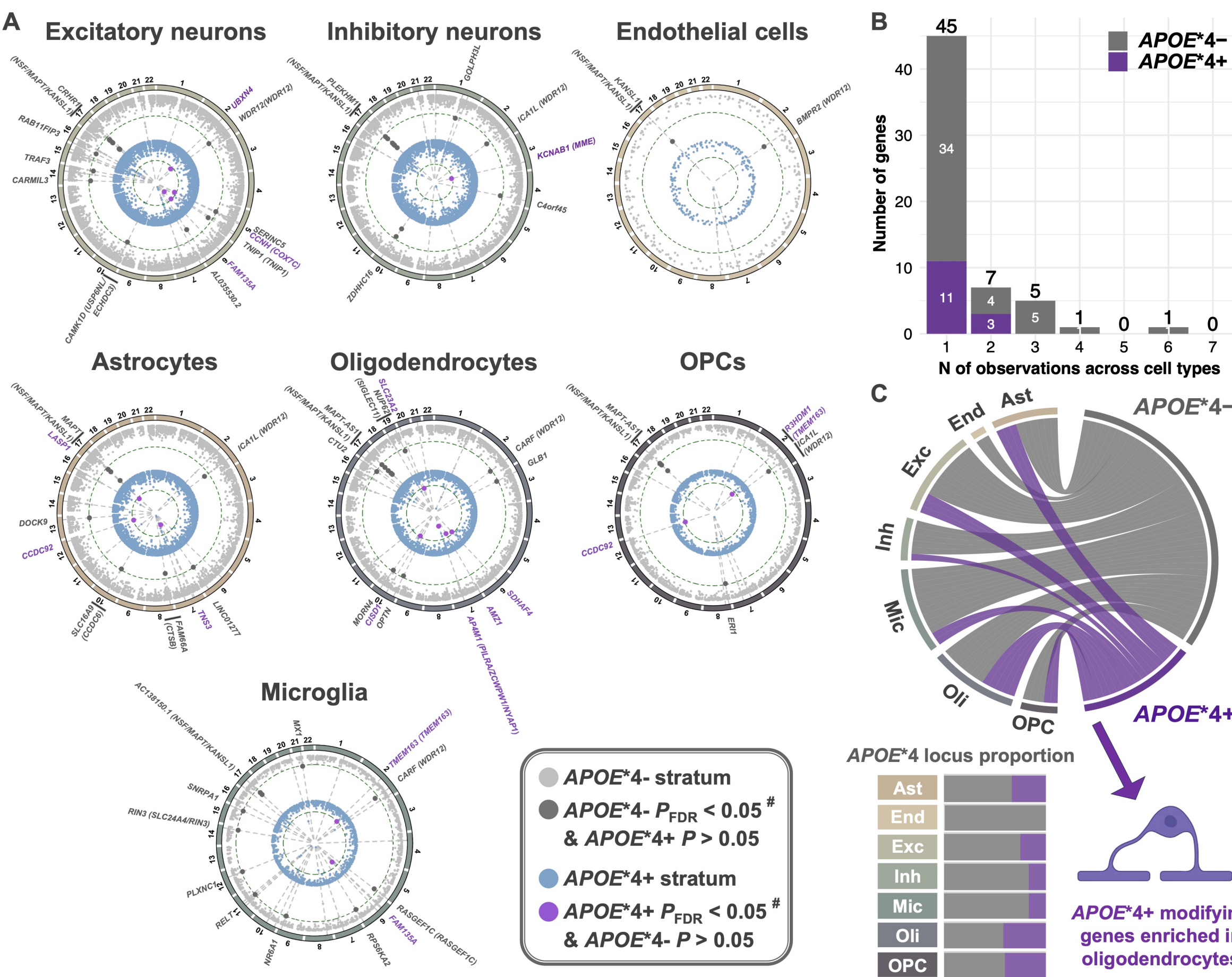


Figure 2. *APOE4 stratified cell-type-specific transcriptome-wide association study (cTWAS) findings.** A) For each cell-type, a circos plot is shown, depicting *APOE**4- findings on the outer layer and *APOE**4+ findings on the inner layer. The radial axes indicate the $-\log_{10}(P\text{-value})$, i.e. significance of gene association, with increasing significance toward the center of the plots. Dashed circular lines indicate $P_{FDR} < 0.05$ thresholds. Significant associations that displayed evidence of *APOE**4 specificity (dotted lines show alignment across *APOE**4 strata) are labeled with unique colors (cf. legend) and their respective gene names, including further gene annotation if they fell within known AD risk loci (genes between parentheses). If multiple genes were located within the same locus (within a 1 Mb window), only the top significant gene is labeled in the outer track. B) The distribution of respective gene observations across cell-types is shown, indicating that most genes were observed in only one cell-type. C) Chord diagram (left) illustrates AD risk loci across *APOE**4 and cell-type groups, while the matching distributions are visualized in bar graphs (right). Notably, Oligodendrocytes showed the highest proportion of *APOE**4+ risk modifying loci.

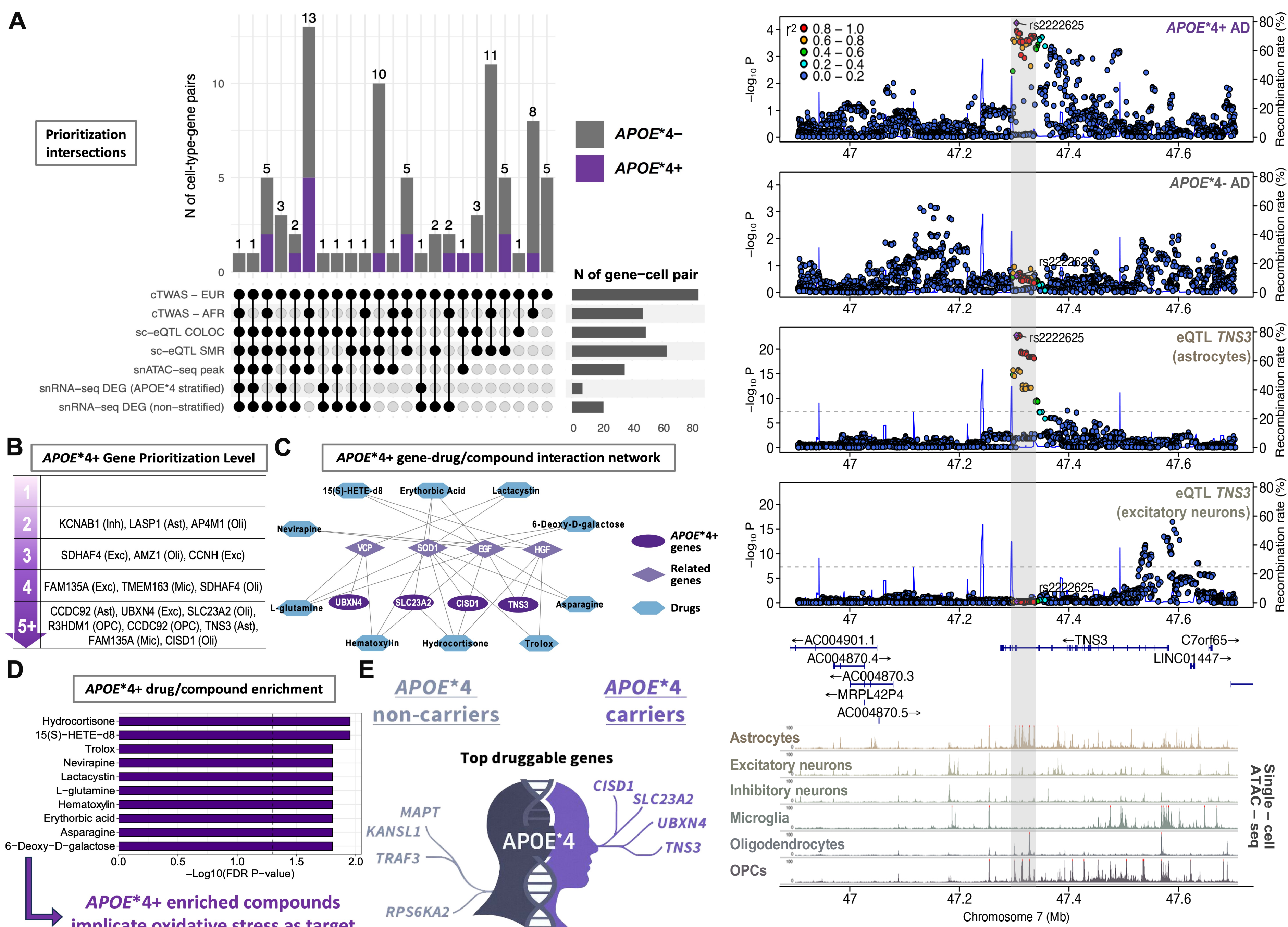


Figure 3. *APOE4 stratified gene and drug target prioritization.** A) The UpSet plot summarizes the European cTWAS significant cell-type-gene pairs with their varying supporting gene prioritization findings, arranged highest to lowest supporting observations from left to right. B) The schematic indicates cell-type-gene pairs with increasing levels of gene prioritization support in the *APOE**4+ stratum. C) Gene-drug interaction network including top *APOE**4+ cTWAS genes and their interacting AD-related genes that were connected to the top 10 enriched compounds or drugs. D) Top 10 drug/compound enrichment results for *APOE**4+ specific genes (with prioritization level ≥ 4) and their interacting AD-related, druggable genes. E) The schematic shows the key candidate druggable targets prioritized in *APOE**4+ and *APOE**4- stratum.

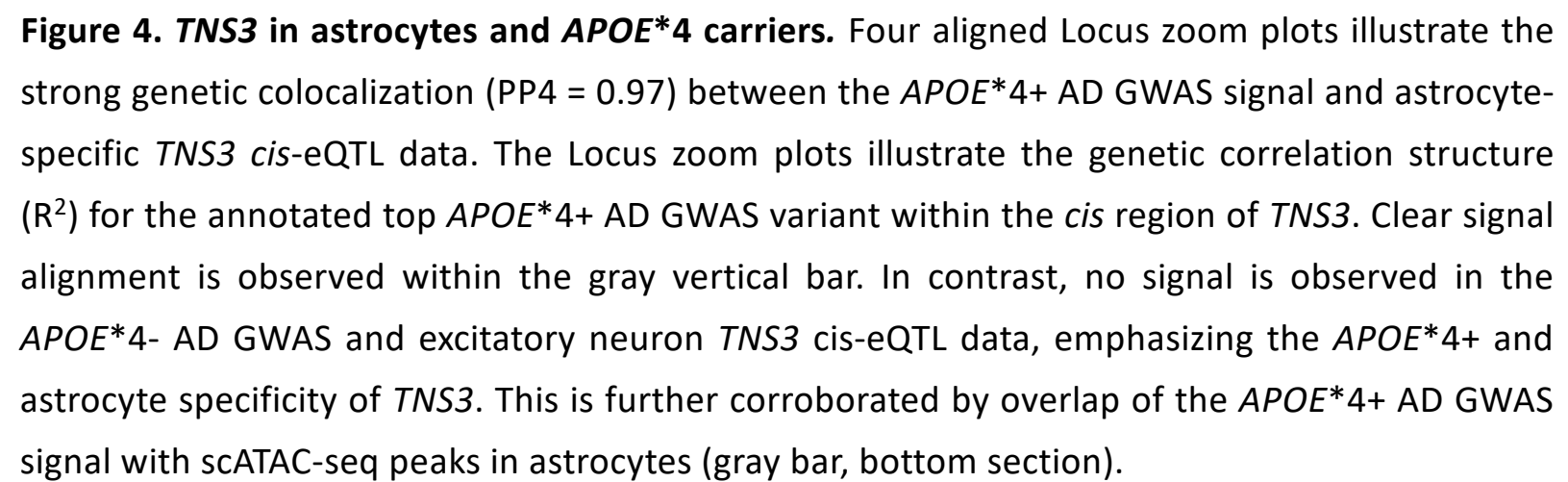


Figure 4. *TNS3* in astrocytes and *APOE4 carriers.** Four aligned Locus zoom plots illustrate the strong genetic colocalization (PP4 = 0.97) between the *APOE**4+ AD GWAS signal and astrocyte-specific *TNS3* cis-eQTL data. The Locus zoom plots illustrate the genetic correlation structure (r^2) for the annotated top *APOE**4+ AD GWAS variant within the cis region of *TNS3*. Clear signal alignment is observed within the gray vertical bar. In contrast, no signal is observed in the *APOE**4- AD GWAS and excitatory neuron *TNS3* cis-eQTL data, emphasizing the *APOE**4+ and astrocyte specificity of *TNS3*. This is further corroborated by overlap of the *APOE**4+ AD GWAS signal with scATAC-seq peaks in astrocytes (gray bar, bottom section).

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