

Human cortical organoids recapitulate early APOE ϵ 4-associated immune dysregulation

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INTRODUCTION

APOE ϵ 4 is associated with immune dysregulation across human tissues, independent of clinical Alzheimer's disease (AD). However, whether this immune phenotype represents an intrinsic, early biological state or arises secondary to AD pathology remains unresolved. Establishing the temporal emergence of APOE ϵ 4-associated immune dysregulation and determining whether it can be accurately modelled in human experimental models are critical for mechanistic interpretation and translational relevance.

METHOD

Human induced pluripotent stem cell (iPSC)-derived cortical organoids incorporating microglia were generated from an APOE ϵ 4 carrier and a non-carrier donor. Organoid amyloid- β and tau pathology was assessed using immunofluorescence and ELISA. Quantitative proteomics was used to profile organoids pre- and post-AD pathology. Differential protein abundance and pathway enrichment analyses were performed to characterize immune processes over time. To assess translational relevance, immune pathways identified in cortical organoids were systematically compared with APOE ϵ 4-associated immune pathways derived from large-scale human proteomic profiling of plasma (n = 9,028), cerebrospinal fluid (n = 1,099), dorsolateral prefrontal cortex (dlPFC; n = 720) and superior temporal gyrus (STG; n = 105).

RESULTS

At four weeks maturation, neither APOE ϵ 4 nor non-carrier organoids showed evidence of p-tau217 or amyloid- β accumulation (Fig. 1a,b). By eight weeks, however, APOE ϵ 4 organoids exhibited marked accumulation of p-tau217 (t(2.5) = 4.48, p = 0.0203, unpaired two-tailed t-test; Fig. 4a,c) and amyloid- β (t(2.5) = 10.02, p = 0.0010, unpaired two-tailed t-test; Fig. 1a,c).

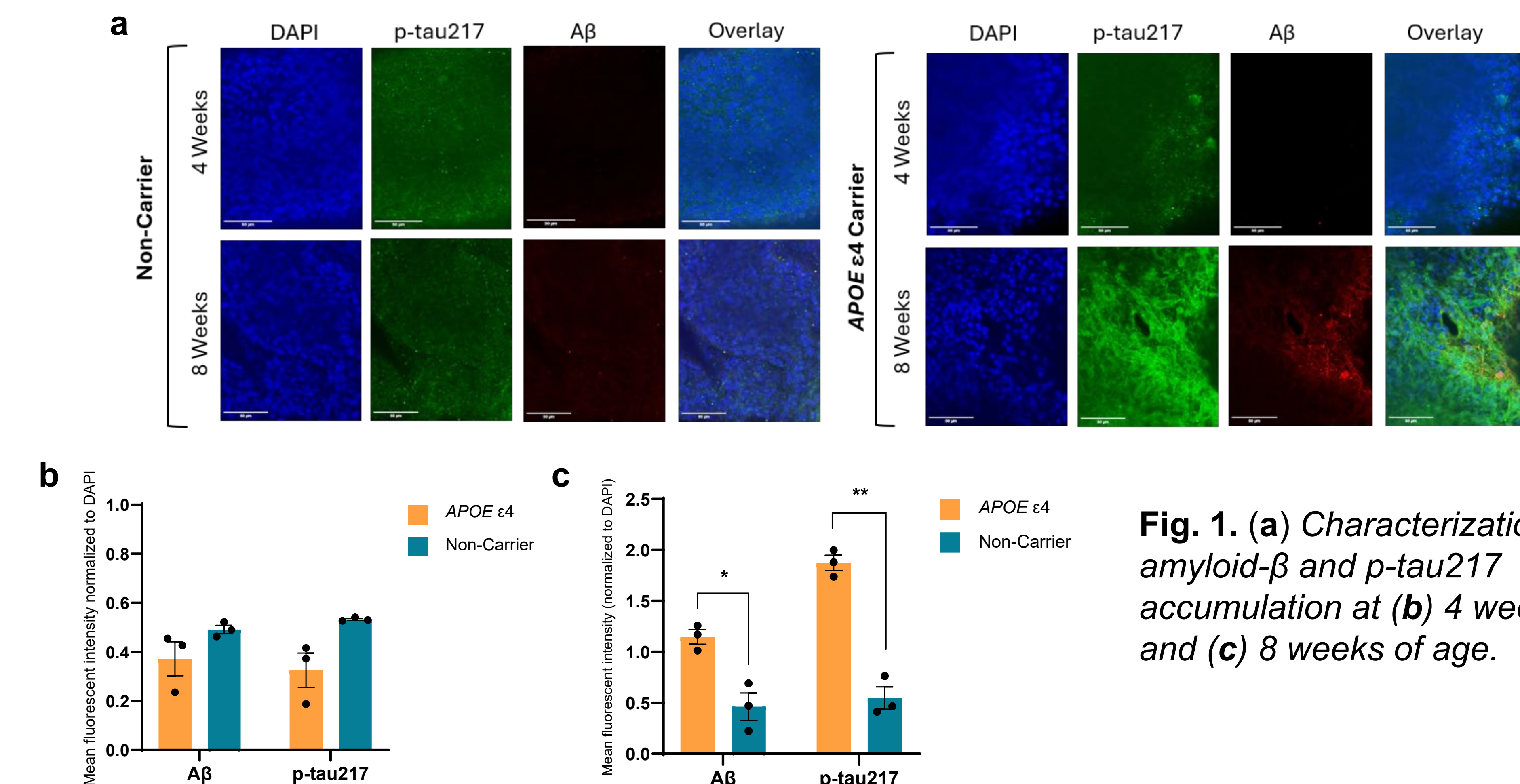


Fig. 1. (a) Characterization of amyloid- β and p-tau217 accumulation at **(b)** 4 weeks and **(c)** 8 weeks of age.

There were no differences in secreted amyloid- β 42/40 or p-tau217 at four weeks (Fig. 2a,b). At eight weeks, there were significantly reduced levels of secreted amyloid- β 42/40 compared to non-carriers (t(8) = 2.40, p = 0.043; unpaired two-tailed t-test; Fig. 2c), consistent with amyloid- β retention in APOE ϵ 4 carrier organoids. Despite intracellular p-tau217 accumulation in APOE ϵ 4 organoids at eight weeks, there was no change in secreted p-tau217 (Fig. 4d).

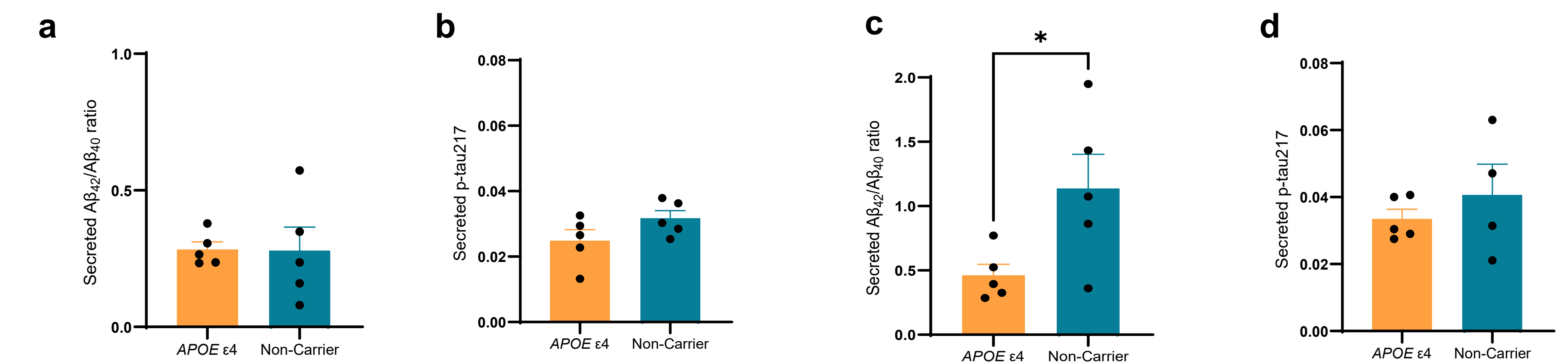


Fig. 2. ELISA of secreted amyloid- β 42/40 and p-tau217 at (a-b) 4 weeks and (c-d) eight-week-old organoids.

Prior to detectable amyloid- β or tau pathology, APOE ϵ 4 cortical organoids exhibited a distinct immune proteomic signature enriched for innate and adaptive immune pathways, including antiviral, interferon, antigen-presentation, and cytokine signalling. This signature intensified with progressive maturation and the emergence of AD-related pathology. Across both time points, pathway enrichment was driven predominantly by upregulated proteins, indicating an active pro-inflammatory immune state rather than loss of immune competence. Notably, immune pathways identified in cortical organoids showed substantial concordance with APOE ϵ 4-associated immune pathways observed systemically (Fig. 3a) and centrally (Fig. 3b), supporting the fidelity of the organoid model in recapitulating in vivo APOE ϵ 4 immune biology.

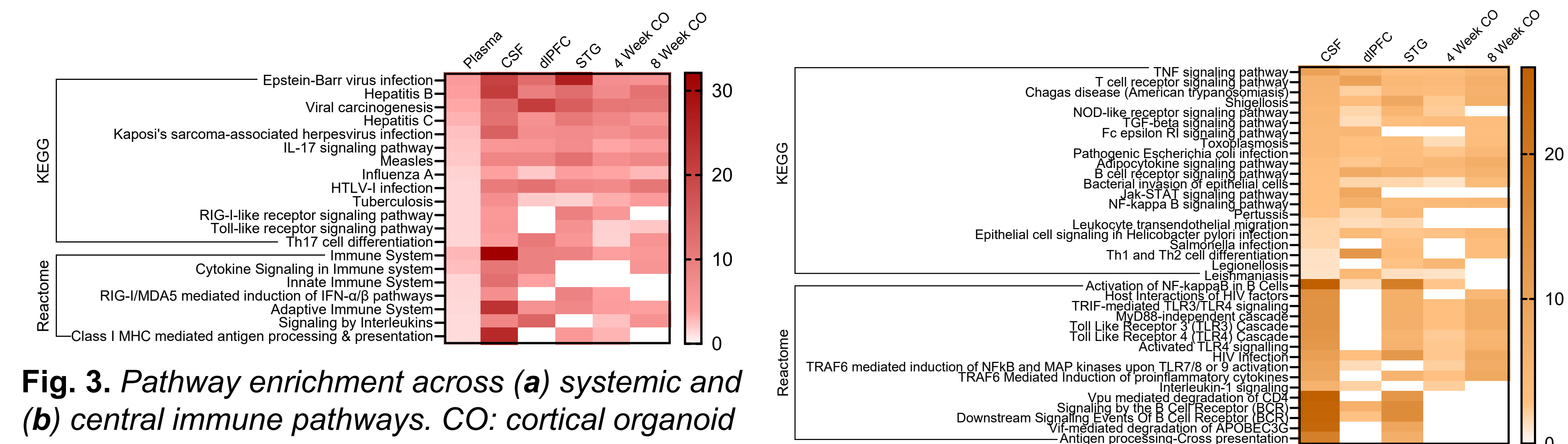


Fig. 3. Pathway enrichment across (a) systemic and (b) central immune pathways. CO: cortical organoid

CONCLUSION

These findings indicate that APOE ϵ 4-associated immune dysregulation emerges prior to hallmark AD pathology and reflects an intrinsic, genotype-linked biological state. The recapitulation of this early immune phenotype in human cortical organoids provides a tractable experimental framework for interrogating APOE ϵ 4-driven immune mechanisms and supports immune dysregulation as a proximal biological feature relevant to AD risk.

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