INPP5D is associated with tau seeding and tau pathogenesis

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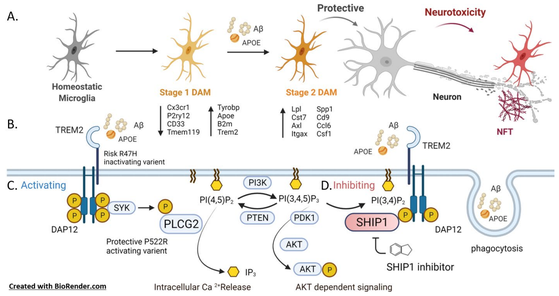
BACKGROUND

Background: Alzheimer's disease (AD) is the most common type of dementia characterized by extracellular amyloid deposits and intracellular neurofibrillary tangles composed of hyperphosphorylated tau aggregates. GWAS studies have identified variants in several inflammatory genes as significant risk factors for AD, highlighting that neuroinflammation is a critical, underlying pathological component in AD. INPP5D, a family member of Src homology 2 domain containing inositol 5' polyphosphatases (SHIPs), is an AD risk gene related to innate immunity in late-onset AD (LOAD). Inpp5d plays a role in regulating signal transduction initiated by immune cell surface receptors. INPP5D binds receptor ITIMs, competes with kinases, and converts PI(3,4,5)P3 to PI(3,4)P¬2, thereby limiting downstream signaling. We previously reported that Inpp5d expression is increased in LOAD and is positively correlated with amyloid burden. However, the relationship of Inpp5d in tau pathology remains unclear. We hypothesize that inhibiting INPP5D will release the break and increase microglial function.

METHODS

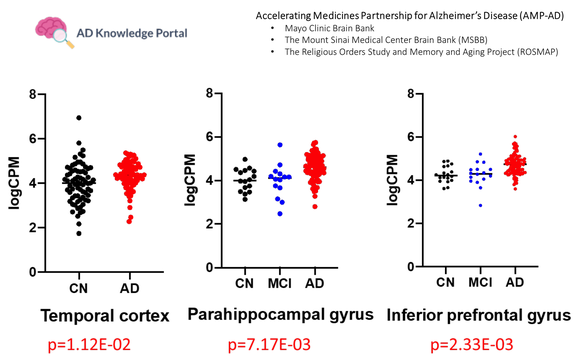
To assess the role of Inpp5d in tau pathology, we performed a fluorescence resonance energy transfer (FRET) assay on human-AD brains obtained from NCRAD. We also crossed a mouse model deficient in Inpp5d (Inpp5d+/-) with a PS19 model of tau pathology and measured tau phosphorylation and transcriptomics. Primary microglia were utilized to measure microglial internalization of Tau.

HYPOTHESIS



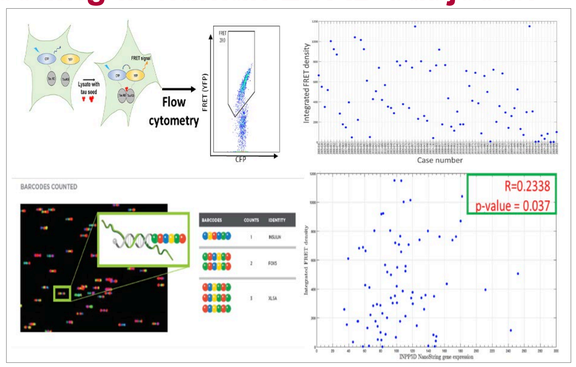
SHIP1 inhibition will increase PLCγ2/AKT-mediated signaling and increase the protective phagocytic activity of microglia to clear extracellular neurotoxins before they accumulate and drive the neuroinflammation that causes neurotoxicity. **A.** Microglia respond to Aβ and APOE with protective or neurotoxic phenotypes depending on their microenvironment. **B.** TREM2 binds Aβ and APOE, which activate microgliosis. Inactivating TREM2 variants increase risk. **C.** DAP12 mediated activation through SYK and PLCγ2. Activating PLCG2 variant is protective. **D.** SHIP1 completes with SYK and limits PIP3-dependent PLCγ2 and AKT signaling downstream from TREM2.

INPP5D expression is increased in LOAD subjects.



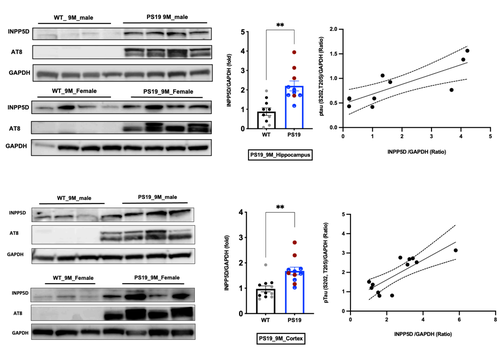
Association of INPP5D expression with amyloid plaque mean density. The scatter plots show the positive association between INPP5D expression and plaque mean density in parahippocampal gyrus, inferior frontal gyrus, and superior temporal gyrus from the MSBB cohort.

Inpp5d gene is correlated with tau seeding in human LOAD subjects.



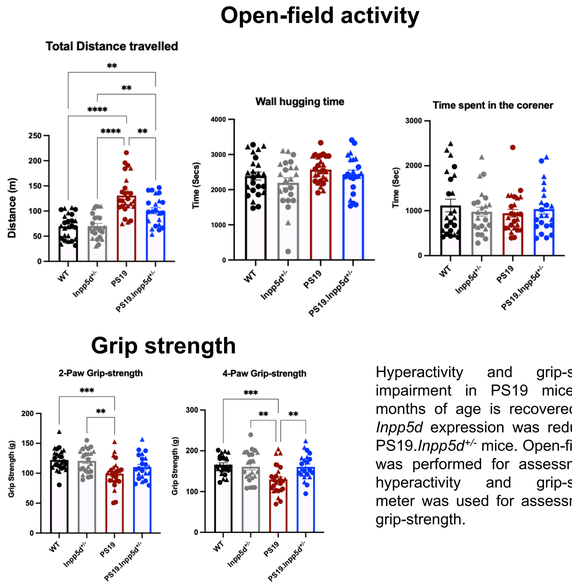
Fluorescence-resonance energy transfer (FRET)-based tau seeding assay exhibits tau seeding in human-LOAD brain-samples. Nano-string technology was used on same samples to examine 770 neuroinflammation genes and among them INPP5D was positively correlated with tau seeding.

Inpp5d gene and protein expression are increased in 5xFAD mice.



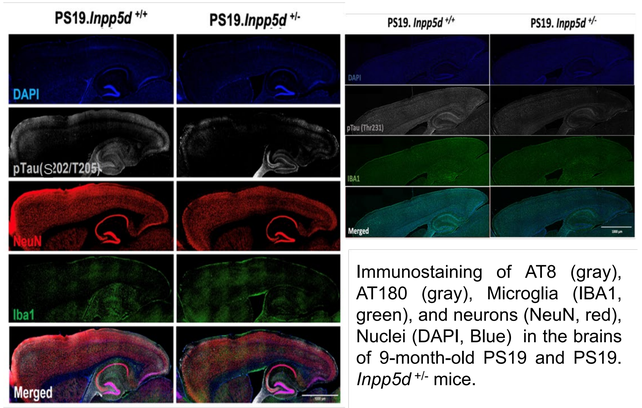
INPP5D protein expression increases in hippocampus and cortex of PS19 mice brains at 9 months of age. Increased INPP5D expression correlates positively with AT8 (pTau,S202/T205) in both hippocampus (r2=0.66) and cortex (r2=0.67)

Inpp5d deficiency mitigates the motor deficits in PS19 mice.

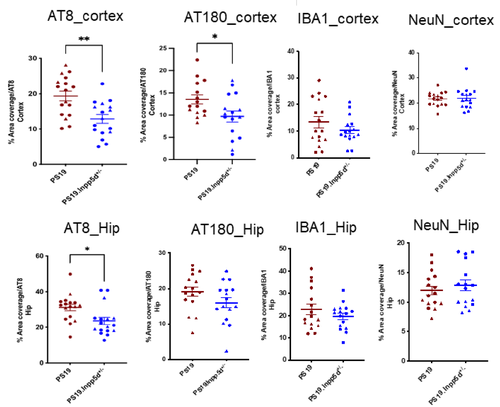


Hyperactivity and grip-strength impairment in PS19 mice at 9 months of age is recovered when *Inpp5d* expression was reduced in PS19.*Inpp5d*$^{+/-}$ mice. Open-field test was performed for assessment of hyperactivity and grip-strength meter was used for assessment of grip-strength.

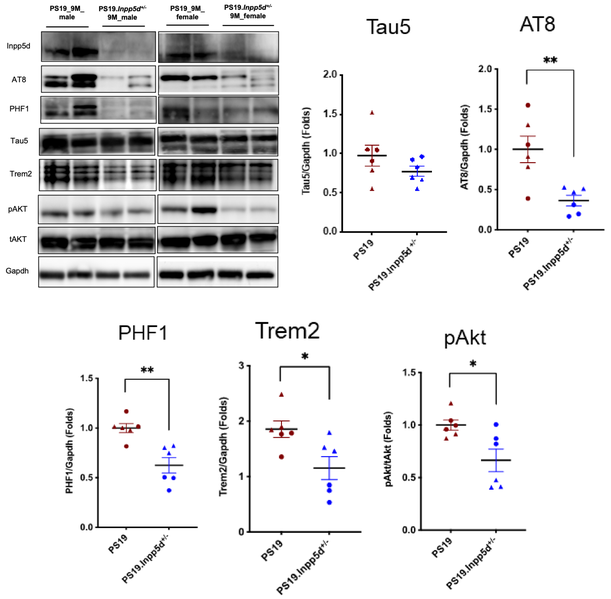
Inpp5d haplodeficiency reduces tau deposition in PS19 mice.



Immunostaining of AT8 (gray), AT180 (gray), Microglia (IBA1, green), and neurons (NeuN, red), Nuclei (DAPI, Blue) in the brains of 9-month-old PS19 and PS19. *Inpp5d* $^{+/-}$ mice.

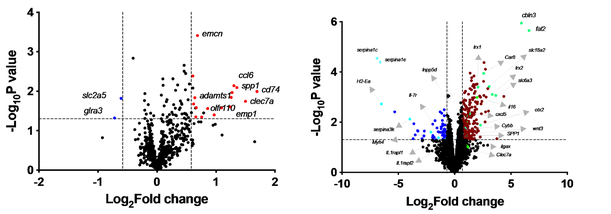


Inpp5d haplodeficiency reduces tau, Trem2 and pAkt.



Immunoblotting showing expression of AT8, PHF1, Trem2 and pAKT in cortices of 9 months old PS19 and PS19.*Inpp5d*$^{+/-}$ mice. Graphs next to blot-image showing quantification of amount of total tau in the brains measured using MSD ELISA Assay.

Differential gene expression analysis revealed distinct neuroinflammatory and cell migration pathways.



Volcano plot showing differential gene expression (DEGs) (P < 0.05, FC > 1.5) in hippocampus and cortex of 9-month-old PS19. *Inpp5d* $^{+/-}$ vs PS19 mice by using Nano-string Glia-profiling panel(Hippocampus) and bulk RNA seq (Cortex).

CONCLUSIONS & FUTURE WORK

- Our findings confirmed that elevated Inpp5d expression in human-AD subjects is recapitulated in PS19 mice and correlated with increased tau pathology.
- In PS19.Inpp5d+/- mice we found alterations in immune related and cell migration related pathways suggesting that Inpp5d modulates the disease progression through these pathways. Furthermore, pathogenesis may be modulated through increased internalization of tau.
- Together these results suggest that therapeutic interventions aimed at reducing Inpp5d may be beneficial in tauopathies.

FUNDING

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