Pazopanib Alters Astrocytes in a Mouse Model of Tauopathy

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Introduction

The hallmark features of Alzheimer’s disease are amyloid beta plaques and neurofibrillary tangles. Plaques are derived from the amyloid precursor protein (APP) cleavage into amyloid beta peptides, which forms pathogenic extracellular plaques in the brain. Tangles are intracellular aggregates containing hyperphosphorylated tau (p-tau), which also contribute to a diverse set of neurodegenerative diseases known as the tauopathies. Previous studies from our lab have shown that tyrosine kinase inhibitors (TKIs) can reduce toxic protein aggregation in mouse models of neurodegenerative disease.

This study focuses on a specific tyrosine kinase inhibitor, pazopanib, that preferentially targets VEGFR. We have found that pazopanib significantly reduces p-tau in the brain as well as preventing neuronal cell death. To determine the effects of pazopanib treatment on glial cells in the brain, we performed immunohistochemistry for astrocytes and microglia in two mouse models of neurodegenerative disease.

Results

1A. Pazopanib reduces astrocyte numbers in the hippocampus of TauP301L mice. However, pazopanib has no effect on microglia in the brain. Furthermore, astrocytes and p-tau co-localize in TauP301L mice, showing that p-tau is present in these cells. This effect is reduced by pazopanib treatment. However, it is unclear whether the presence of p-tau in astrocytes is a mechanism for clearance, or propagation. In progressive supranuclear palsy (PSP), another tauopathy, it has been shown that p-tau accumulation in astrocytes is a degenerative process (1).

1B. Pazopanib does not affect astrocytes or microglia in 3x-APP mice. We found that GFAP and IBA-1 were increased in 3x-APP mice compared to wild type, complementary to our previous studies (2).

Materials & Methods

Treatment

Human mutant TauP301L mice and 3x-APP (Swedish, Dutch, Iowa mutations) mice were treated daily with an intraperitoneal injection of 5mg/kg pazopanib or DMSO for 3-4 weeks. Mice were male and female ranging from 12 to 18 months old.

Immunohistochemistry

20 micron thick brain slices were stained using antibodies for astrocyte marker glial fibrillary acidic protein (GFAP) (Cat ASTRO6, Thermofisher) and microglial marker ionized binding adaptor protein 1 (IBA-1) (Cat 019-19741, WAKO). GFAP co-stain was performed with antibodies to p-tau (AT180) (Cat OPA-03156). Images were taken on a fluorescent microscope (Evos Fl, Thermofisher).

Statistical Analysis

Densitometry was done using ImageJ software. Statistical analysis was performed using Graphpad Prism software, version 7.0. Significance was determined by one-way analysis of variance (ANOVA) with Tukey’s method for multiple comparisons.

Conclusions

1. Pazopanib reduces astrocyte staining in TauP301L mice back to wild type levels
2. The changes in astrocytes in TauP301L mice was independent of microglial changes.
3. Pazopanib has no effect on astrocytes or microglia in 3x-APP mice.
4. In TauP301L mice, astrocytes may take up p-tau, and pazopanib reduces this effect.

References


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