Hexachlorophene reduces Tau aggregation and potential therapeutic agent for treatment of Alzheimer's disease

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Abstract

Alzheimer's disease (AD) is a rapidly progressive neurodegenerative disease that affects over 5 million people in the United States alone. The molecular mechanism of AD pathogenesis is not completely understood yet and there only exist very few pharmacological means of intervention for the disease. Tau, a microtubule-associated protein, plays a vital role in stabilizing microtubule networks in neurons. An increase in phosphorylation of tau (hyper phosphorylation) leads to aggregation of the protein resulting in neurofibrillary tangles, a pathological hallmark associated with Alzheimer's disease (AD). The identification of pharmacological agents that can help decrease levels of phosphorylated tau would be advantageous. Recent work in our laboratory has found that the molecule hexachlorophene can regulate levels of tau in cellular models. Our validation experiments using hexachlorophene showed significant reduction of both total and phosphorylated forms of endogenous tau in M17 neuroblastoma cells, inducible tau in HEK280 cells and over-expressed tau in HeLa C3 cells. We also analyzed effect of hexachlorophene on various pathological forms of tau such as the phosphoserine 396 tau form that localizes to the soluble fraction. Treatment with hexachlorophene decreased levels of phospho serine 396 tau in soluble fraction. Additionally, staining with Thioflavin-S clearly indicates a potential reduction in tau aggregation upon hexachlorophene treatment. Overall, our data suggest that hexachlorophene could be a potential drug molecule for the treatment of tau proteinopathies and AD.

Methods

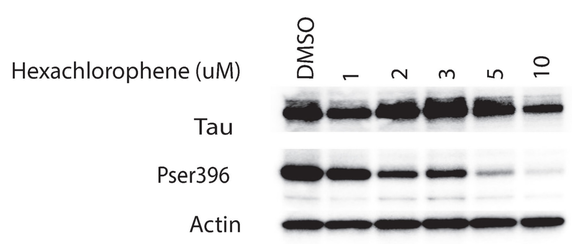
-Western Blotting

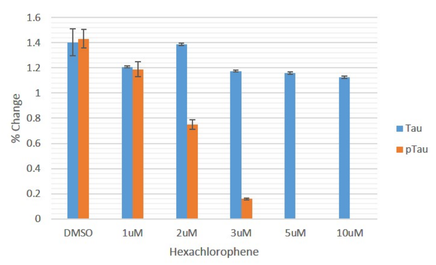
M17 neuroblastoma, HeLa C3 over-expressed tau, Inducible tau model iHEK 280 cells treated with hexachlorophene with different doses and lysates were analyzed by Western blotting for total tau, phosphorylated tau, and Actin.

-Thioflavin-S staining

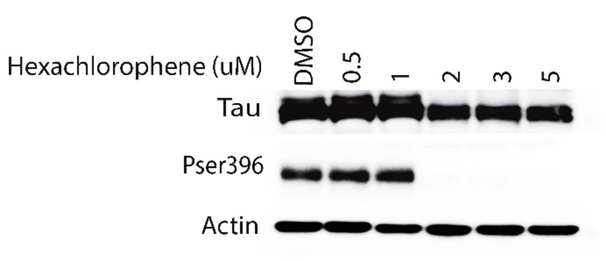
Thioflavin-S staining was performed using 0.1% Thioflavin-S incubated with 5 min, and primary rabbit polyclonal Tau Antibody diluted with 5% goat serum and secondary anti-rabbit labeled with red. Cells containing distinct ThioS signals indicating the presence of aggregated protein were scored in many independent fields containing a total of 500 cells.

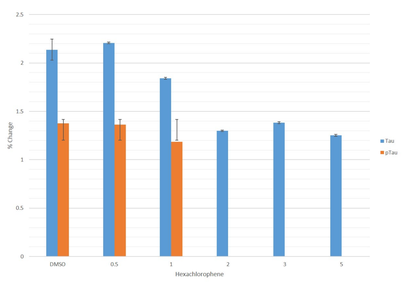
Hexachlorophene decreases levels of endogenous tau in M17 neuroblastoma cells



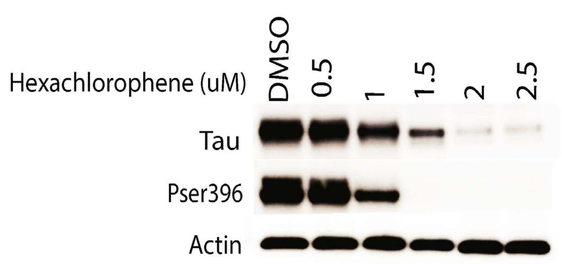


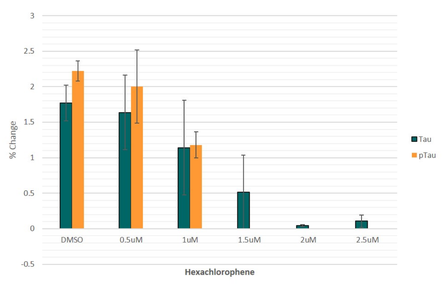
Hexachlorophene decreases total and phospho tau levels HeLa C3 cells over-expressed tau



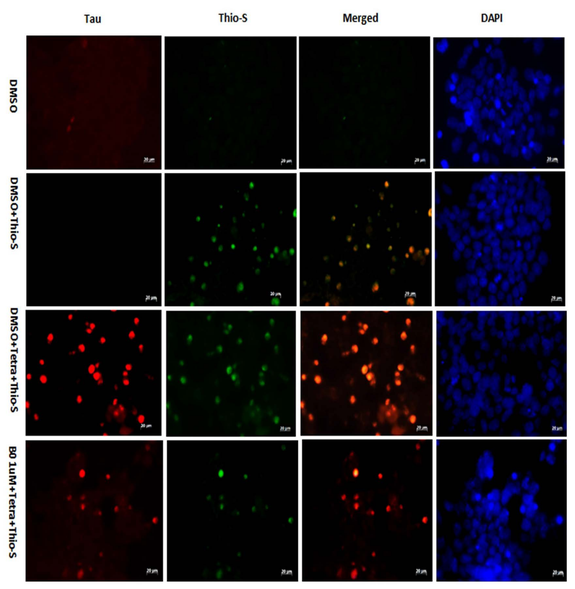


Hexachlorophene decreases levels total and phospho tau in iHEK280 cell Inducible tau model





Hexachlorophene reduces tau aggregates in a HEK280 cell inducible tau model stained with Thioflavin-S



Conclusions

-For the first time, these data demonstrate that hexachlorophene reduces tau aggregation.

-M17 neuroblastoma cells treated with hexachlorophene 10µM completely clears endogenous total and phosphorylated pSer396 tau proteins.

-In the over-expressed tau cell model in HeLa C3 2uM hexachlorophene partially clears total tau and completely clears the phosphorylated tau pSer396.

-Inducible tau in iHEK 280 cells 1.5 µM hexachlorophene partially clears the total tau and completely clears the phosphorylated tau Pser396. For 2 uM concentration of drug clears the both tau protein levels.

-Additionally, staining with Thioflavin-S clearly indicates significant reduction of tau aggregation at a concentration of 1 µM.

-Overall, our data suggest that hexachlorophene could be a potential drug molecule for the treatment of tau proteinopathies and AD.

References

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