

Cysteine Protease Inhibitor, E64d, of Cathepsin B Reduces pGlu-Abeta and Abeta, and Improves Memory Deficits in the APPLon Mouse Model of AD

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Background: Pyroglutamate amyloid-beta peptides (pGlu-Abeta) are particularly pernicious forms of Abeta. pGlu-Abeta and full-length Abeta peptides accumulate in Alzheimer's disease (AD) brains, leading to severe memory deficits. pGlu-Abeta peptides are N-terminally truncated forms of full-length Abeta peptides with modification of the N-terminal glutamate to form pGlu-Abeta(3-40/42). The prominent presence of pGlu-Abeta in AD brains and involvement of pGlu-Abeta to initiate formation of oligomeric neurotoxic Abeta forms may be key in AD. Our recent research indicates the key role of the alternative beta-secretase cathepsin B (CatB) in the production of pGlu-Abeta and Abeta (Hook et al., 2014, in press; Kindy et al., 2012), suggesting that inhibitors of CatB can reduce pGlu-Abeta and Abeta. Therefore, this study investigated the cysteine protease inhibitor E64d, that inhibits CatB, for its effectiveness in reducing Abeta peptide forms and improving memory deficits in APPLon AD mice, which express APP-695 and have the wild-type (wt) beta-secretase activity present in most AD patients.

Methods: APPLon mice were administered E64d (oral), using E64d prepared in the food chow. Memory deficits were then assessed by the Morris water maze test. Brain tissue samples were measured for levels of pGlu-Abeta and flAbeta peptides by ELISAs, and amyloid plaque load was assessed by quantitative immunohistochemistry.

Results: E64d treatment reduced brain levels of pGlu-Abeta(3-40/42), flAbeta(1-40/42), and pGlu-Abeta/Abeta plaque load in APPLon mice. E64d treatment of APPLon mice with CatB gene knockout resulted in similar level of Abeta peptide reduction. Notably, E64d resulted in substantial and significant improvement in memory deficits.

Conclusion: Administration (oral) of the cysteine protease inhibitor E64d to APPLon mice resulted in decreased brain levels of pGlu-Abeta and flAbeta, decreased amyloid plaque load, and substantial improvement in memory deficits. E64d is known to be safe in patients, based on extensive clinical trials in Japan for muscular dystrophy. These data strongly suggest that the E64d type compound(s) will be useful as therapeutic drug candidates for treating AD patients.