Assessment of Oral LISPRO Treatment in Ameliorating Amyloid and Tau Pathology in Transgenic Alzheimer's Mice Model

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Abstract

Lithium is used primarily as a mood stabilizer for bipolar disorder and has been used to treat mania, depression and suicidal thoughts for a long time. In addition, it has also been shown to prevent cognitive decline which indicated that lithium has a potential therapeutic effect in Alzheimer's disease (AD). However, one of the main problems that exist in the currently FDA-approved lithium pharmaceutics (carbonate and citrate) is that it has narrow therapeutic index and lithium plasma level change drastically which can cause adverse side effects. Here we investigated the safety, pharmacokinetics and therapeutic efficacies of LISPRO (ionic co-crystals of lithium salicylate with organic l-proline), lithium salicylate, Li_2CO_3 (currently used) and placebo. We found that LISPRO attenuate β-amyloid plaques and phosphorylation of tau through modulation of inflammation and GSK3β inactivation. Cytokine profiles in the brain, plasma and splenocytes suggest that LISPRO (8-weeks) down-regulates pro-inflammatory, up-regulates anti-inflammatory and suppresses renal COX2 expression in Tg2576 mice. Plasma and brain pharmacokinetics of lithium indicated that LISPRO showed significantly higher brain and steady plasma lithium levels on C57BL/6J (2-weeks) and Tg2576 (8-weeks) mice. Interestingly, chronic (20-weeks) administration of LISPRO produces a slightly higher, but non-significant brain to plasma lithium levels and reduces β-amyloid plaques, and tau-phosphorylation through modulation of presynaptic (synaptophysin) and post-synaptic protein (PSD95) expression in 3xTg-AD mice.

Methods

Tg2576 APP KM670/671NL (Swedish) and triple transgenic (3xTg-AD) mice harboring [APP_{SWE}, PSEN1 (PS1/M146V) and tau (P301L) mutations] were divided into different experimental groups and were fed for 8- and 20- weeks with Lithium containing diets. These treatments consisted of a normal mice chow diet and the same chow diets supplemented with lithium carbonate (LC) [0.05%, 2.26 mmol/kg/day equivalent to 83 mg/kg/day, with chow food], lithium salicylate (LS) [0.195%, equivalent to 2.24 mmol/kg/day equivalent to 325 mg/kg/day, with chow food], and lithium salicylate proline (LISPRO) [0.35%, 2.25 mmol/kg/day equivalent to 583 mg/kg /day, with chow food]. These dosages are chosen based on the literature and preliminary study conducted at our lab using low- and high- concentration of lithium salts. All mice were sacrificed 20 weeks after the treatments. The plasma and brain lithium levels were measured using Atomic Absorption Spectroscopy (AA-6200, Shimadzu, Japan). Mice brain tissue sections were analyzed for immunohistochemical and immunoblot analyses to see whether LISPRO could ameliorate amyloid tau pathology compared to other lithium salts (lithium salicylate and lithium carbonate) and placebo.

Conclusions

Ionic co-crystals of lithium salicylate with organic proline (LISPRO) showed better safety and pharmacokinetic profile compared to two other lithium salts. Oral treatment with LISPRO attenuate β-amyloid and tau pathology in cell culture and transgenic AD mice model.

Acknowledgements

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Results

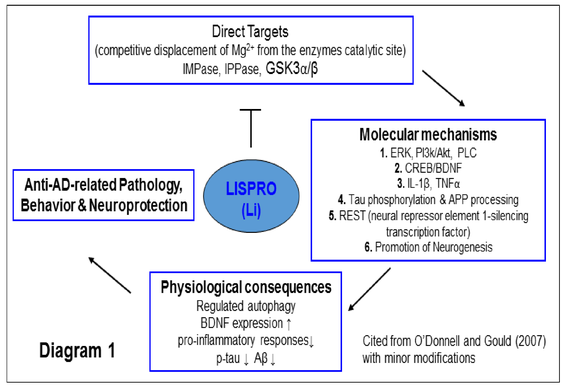

Diagram. 1 Mechanisms of Lithium Efficacy for AD

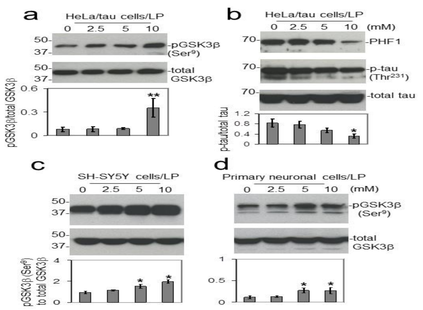

Fig. 3 Treatment with LISPRO decreases tau phosphorylation in HeLa/tau cells while increasing inhibitory GSK3β (Ser^9) phosphorylation .

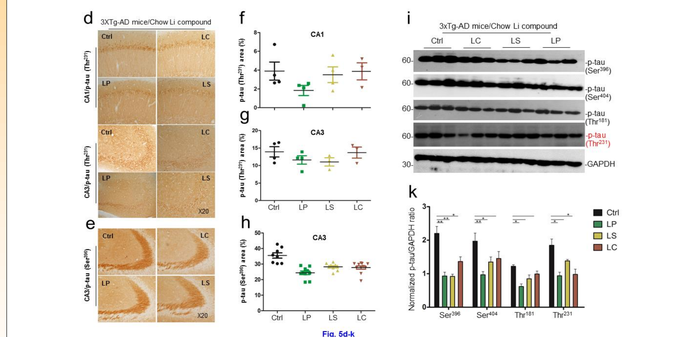

Fig. 5 (d –k) Oral LISPRO promotes autophagy, microglial phagocytosis of Aβ and anti-inflammatory in the mice

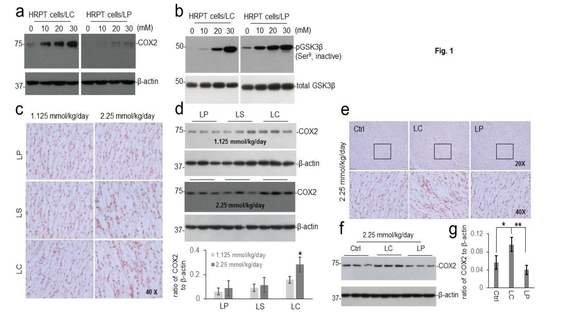

Fig. 1 LISPRO does not increase COX2 expression *in vitro* and *in vivo*

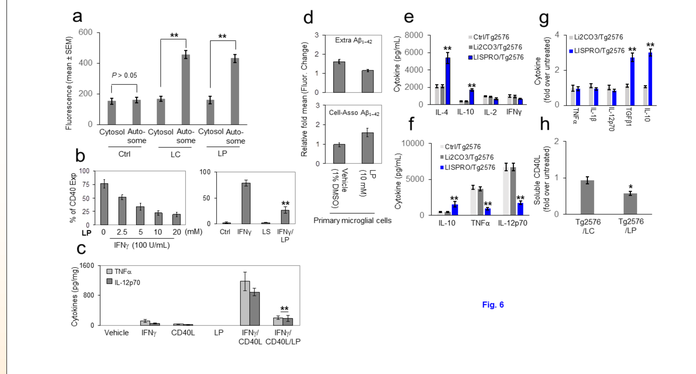

Fig. 6 Oral LISPRO promotes autophagy, microglial phagocytosis of Aβ and anti-inflammatory in the mice

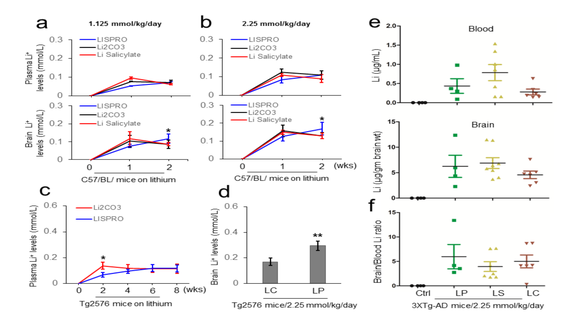

Fig. 2 Plasma and brain pharmacokinetic analysis of LISPRO, lithium salicylate, and Li_2CO_3 in C57BL/6J, Tg2576 and 3XTg-AD mice following oral administration (a & b)

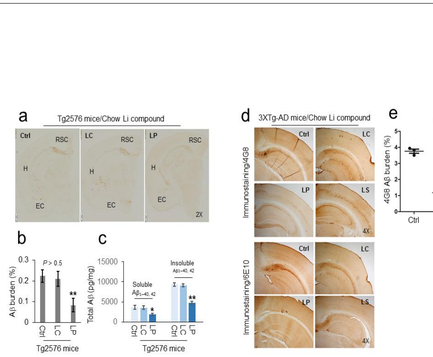

Fig. mice

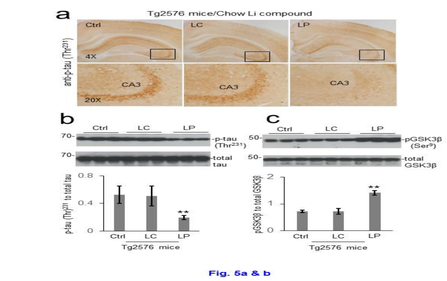

Fig. 5 (a & b) Oral LISPRO reduces tau hyper-phosphorylation in Tg2576 and 3XTg-AD mice

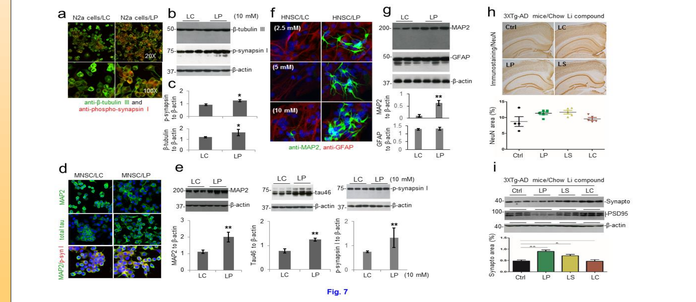

Fig. 7 LISPRO markedly promotes neuronal cell differentiation and prevent pre- and post-synaptic protein loss in 3XTg-AD mice