

A Combination of Caffeine Supplementation and Enriched Environment in an Alzheimer's Disease Mouse Model

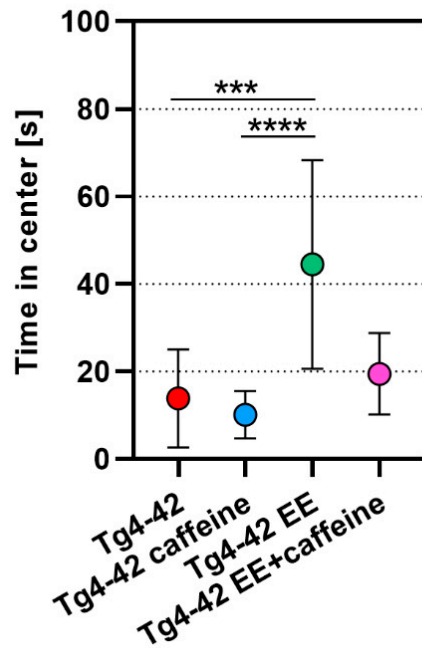


Figure S1. Tg4-42 mice housed under EE conditions spent significantly more time in the central area of the open field compared to Tg4-42 and Tg4-42 caffeine mice. One-way ANOVA followed by Bonferroni's multiple comparison test. *** $p < 0.001$, **** $p < 0.0001$. All data are given as mean \pm SD. Data from vehicle- and caffeine-treated SH and vehicle-treated EE Tg4-42 groups have been partially included in previous studies [1, 2] and were part of a larger set of experiments in order to minimize experimental animal numbers.

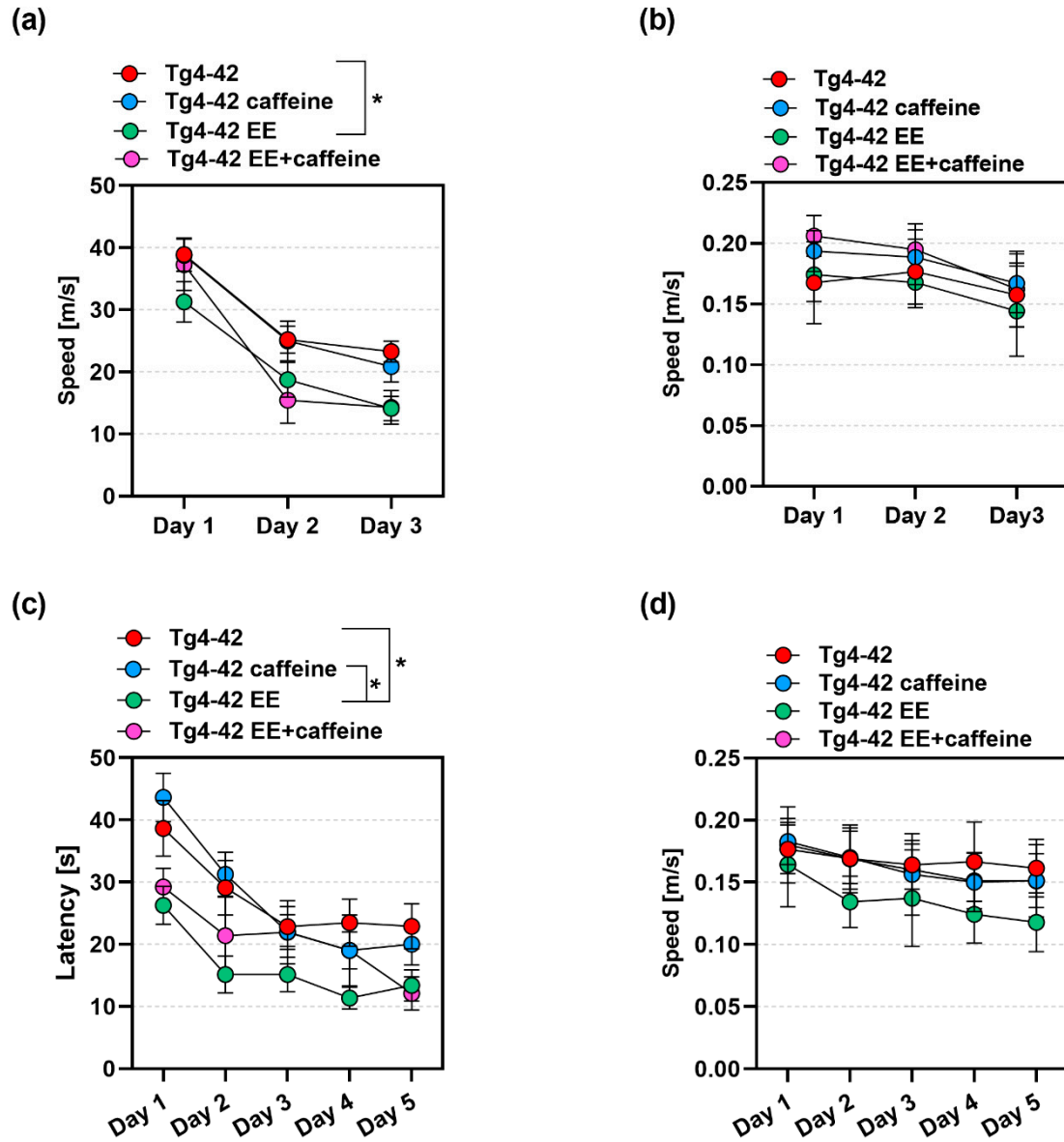


Figure S2. During the cued (a, b) and acquisition training phase (c, d), Tg4-42 mice housed under EE conditions mice displayed a reduced escape latency compared to Tg4-42 SH and Tg4-42 caffeine groups (a, c), while swimming speeds were unaltered (b, d). Two-way repeated measures ANOVA followed by Bonferroni's multiple comparison test. $*p < 0.05$. All data are given as mean \pm SD. Data from vehicle- and caffeine-treated SH and vehicle-treated EE Tg4-42 groups have been partially included in previous studies [1, 2] and were part of a larger set of experiments in order to minimize experimental animal numbers.

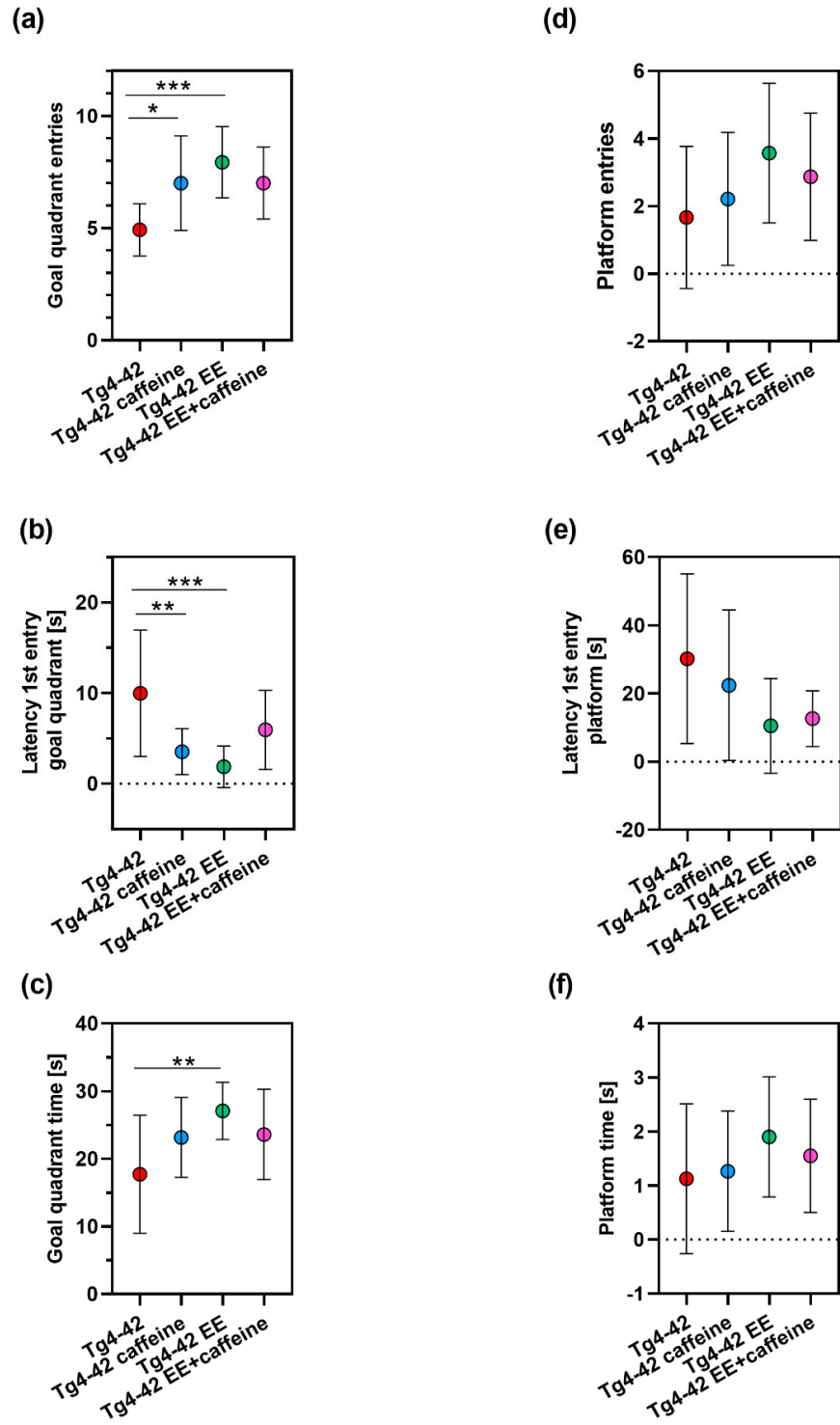


Figure S3. Compared to Tg4-42 mice, Tg4-42 caffeine and Tg4-42 EE groups showed more goal quadrant entries (a), as well as a significantly reduced latency for the initial goal quadrant entry (b). Tg4-42 EE mice spent significantly more time in the goal quadrant than Tg4-42 mice (c), while no statistically significant differences in the number of platform entries (d), the latency to the initial entry in the former platform position (e) or the platform time (f) were detected. One-way ANOVA followed by Bonferroni's multiple comparison test. $*p < 0.05$, $**p < 0.01$, $***p < 0.001$. All data are given as mean \pm SD. Data from vehicle- and caffeine-treated SH and vehicle-treated EE Tg4-42 groups have been partially included in previous studies [1, 2] and were part of a larger set of experiments in order to minimize experimental animal numbers.

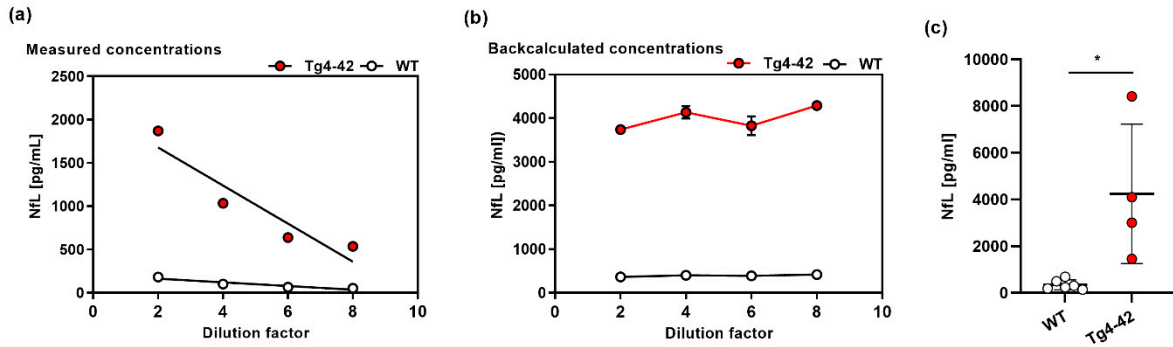


Figure S4. Pooled plasma samples from 6-month-old Tg4-42 and WT mice were measured in different dilutions (a). The plasma NFL concentrations were back-calculated and plotted against the dilution factor (b). Plasma samples of 6-month-old WT ($n = 6$) and Tg4-42 ($n = 4$) mice were measured in 6-fold dilution, revealing significantly increased NFL levels in Tg4-42 mice (c). * $p < 0.05$, two-tailed unpaired t-test.

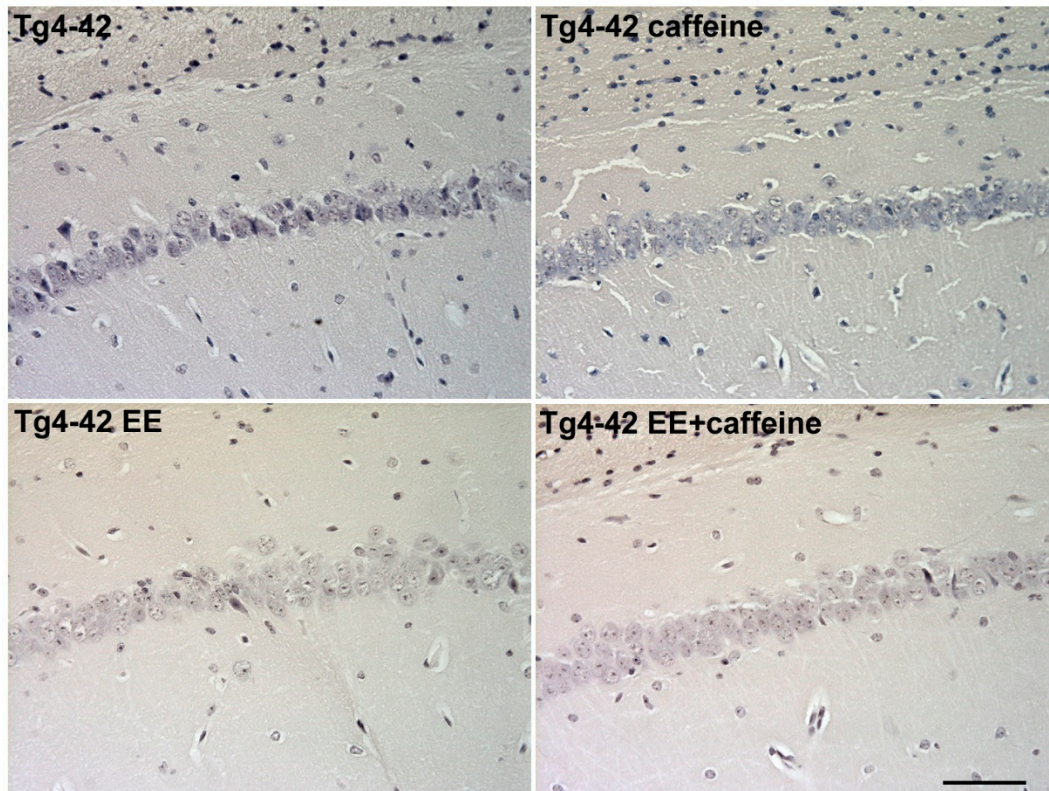


Figure S5. Example images of CA1 pyramidal neurons at 6 months of age. Sagittal brain sections were stained with hematoxylin and images were taken at 400x magnification. Scale bar: 50 μm .