Supplementary Data for *Drugs* Article

Plasma Biomarker Effects of Oral Valiltramiprosate/ALZ-801 in Early Alzheimer's Disease from Phase 3 and Phase 2 Trials: Analysis of Core Biomarkers and Correlations with Clinical and Neuroimaging Outcomes

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Table S1. Summary of plasma Aβ42 concentrations (pg/ml) in valiltramiprosate Phase 3 study in APOE4/4 homozygotes.

Disease severity	Subject group	Baseline	Week 26	Week 52	Week 78
Overall	ALZ-801 (n =145)	19.4 ± 0.3	19.8 ± 0.3	19.8 ± 0.3	20.0 ± 0.3
	Placebo (n =150)	19.9 ± 0.3	20.2 ± 0.5	20.2 ± 0.4	22.6 ± 1.6
MCI	ALZ-801 (n = 60)	18.9 ± 0.5	19.2 ± 0.5	19.9 ± 0.5	19.6 ± 0.5
	Placebo (n = 56)	19.1 ± 0.6	20.8 ± 1.2	20.4 ± 0.6	20.6 ± 0.7
Mild AD	ALZ-801 (n = 85)	19.7 ± 0.4	20.3 ± 0.4	19.8 ± 0.4	20.4 ± 0.4
	Placebo (n = 94)	20.3 ± 0.4	19.9 ± 0.4	20.0 ± 0.4	23.8 ± 2.5

Plasma concentration values are expressed as mean ± SEM. No statistical significance across treatment arms or time points were observed (NS, p>0.05).

Table S2. Correlations between the plasma p-tau217 drug effect (change from baseline at Week 78) and the clinical and neuroimaging outcomes (change from baseline at Week 78) in Phase 3 APOE4/4 subjects with Mild AD: ALZ-801 treatment vs. placebo.

	ALZ-801 treatment			Placebo		
Outcome	**n**	**r**	**p-value**	**n**	**r**	**p-value**
Positive correlation coefficient r values consistent with association						
ADAS-Cog 13	72	0.058	0.629	86	0.141	0.194
CDR-SB	70	0.086	0.479	85	**0.292**	**0.007**
Ventricular volume	60	0.063	0.632	75	**0.246**	**0.033**
Negative correlation coefficient r values consistent with association						
DAD	71	-0.093	0.441	86	-0.118	0.279
Hippocampal volume	60	-0.051	0.700	75	-0.180	0.123
Cortical thickness	60	-0.112	0.395	75	-0.138	0.237
Whole brain volume	60	-0.003	0.982	75	-0.214	0.065

r, Spearman's correlation coefficient. Significant correlations with p<0.05 are shown in bold. ADAS-Cog13, 13-item Alzheimer's Disease Assessment Scale-Cognitive Subscale; CDR-SB, Clinical Dementia Rating-Sum of Boxes; DAD, Disability Assessment for Dementia. For plasma p-tau217, ADAS-Cog13, CDR-SB and ventricular volume, larger values indicate more severe disease. For DAD, hippocampal volume, cortical thickness and whole brain volume, smaller values indicate more severe disease. These results show no significant correlations of plasma p-tau217 changes and clinical/neuroimaging outcomes at Week 78 in valiltramiprosate-treated Mild AD subjects. In Mild AD placebo subjects, p-tau217 increase correlated with CDR-SB worsening and increase in ventricular volume (an index of whole brain atrophy).

Table S3. Correlations between baseline plasma p-tau217 and baseline clinical and neuroimaging measures in the overall Phase 3 APOE4/4 homozygous MCI population.

	Overall MCI at baseline (ALZ-801 + Placebo subjects combined)		
Outcome	**n**	**r**	**p-value**
Positive correlation coefficient r values consistent with association			
ADAS-Cog 13	116	**0.478**	**<0.0001**
CDR-SB	116	**0.239**	**0.0098**
Ventricular volume	112	**0.236**	**0.012**
Negative correlation coefficient r values consistent with association			
DAD	116	**−0.205**	**0.027**
Hippocampal volume	112	**−0.350**	**0.0002**
Cortical thickness	112	**−0.286**	**0.002**
Whole brain volume	112	−0.124	0.191

r, Spearman's correlation coefficient. Significant correlations with p<0.05 are shown in bold. ADAS-Cog13, 13-item Alzheimer's Disease Assessment Scale-Cognitive Subscale; CDR-SB, Clinical Dementia Rating-Sum of Boxes; DAD, Disability Assessment for Dementia. For plasma p-tau217, ADAS-Cog13, CDR-SB and ventricular volume, larger values indicate more severe disease. For DAD, hippocampal volume, cortical thickness and whole brain volume, smaller values indicate more severe disease. These results show significant correlations between baseline plasma p-tau217 and baseline clinical/neuroimaging measures in MCI APOE4/4 homozygotes.

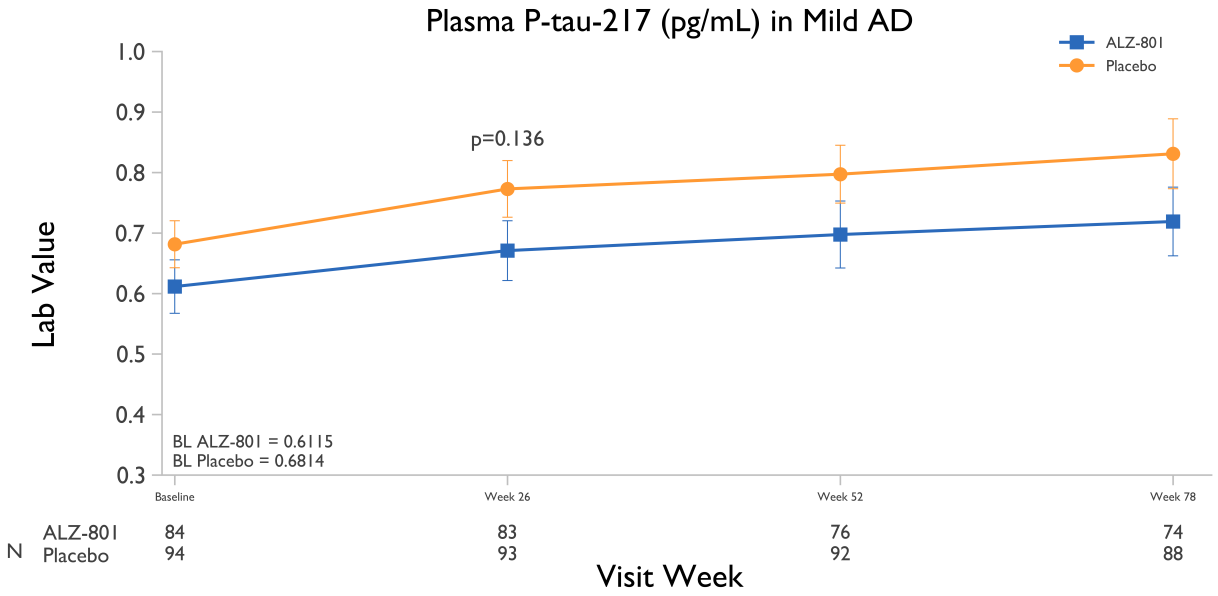

Figure S1. Longitudinal absolute value time course of plasma p-tau217 concentrations (pg/mL) following valiltramiprosate (ALZ-801) 265 mg BID vs. placebo treatment in the Mild AD APOE4/4 population of the Phase 3 trial. Data are shown as observed case analysis; means ± SEM, n=84 for valiltramiprosate and n=94 for placebo. Unpaired, two-tailed t-test. Valiltramiprosate-treated participants showed numerically lower plasma p-tau217 levels relative to placebo across all timepoints; the between-group differences are not statistically significant (p=0.136, 0.172 and 0.173 at Weeks 26, 52 and 78, respectively).

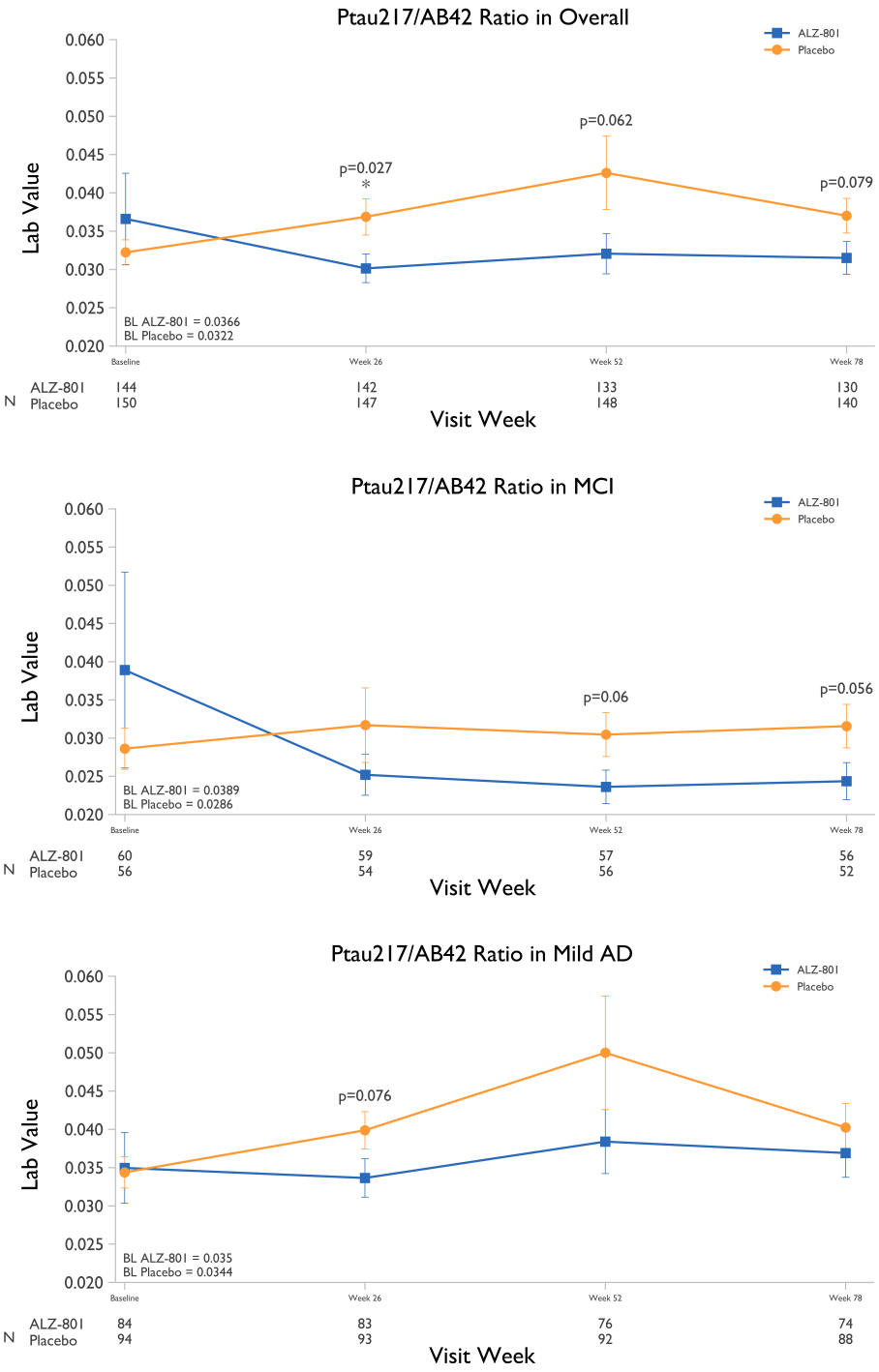

Figure S2. Longitudinal absolute value time course of plasma p-tau217/Aβ42 ratio following valiltramiprosate (ALZ-801) 265 mg BID vs. placebo treatment in the Overall, MCI and Mild AD APOE4/4 subjects of the Phase 3 trial. Data are shown as observed case analysis; means ± SEM, n=84 for valiltramiprosate and n=94 for placebo. Unpaired, two-tailed t-test. These results

show a rapid and sustained reduction of plasma p-tau217/Aβ42 ratio after valiltramiprosate treatment vs. an increase with placebo over 78 weeks in both the overall early AD and MCI populations. In Mild AD subjects, the longitudinal change of p-tau217/Aβ42 ratio was flattened following valiltramiprosate treatment as compared to an increase with placebo. Valiltramiprosate treatment reduced plasma p-tau217/Aβ42 ratio relative to placebo in the overall and MCI subjects ($p<0.10$). A non-significant numerical trend effect was observed in the Mild AD subgroup with valiltramiprosate vs. placebo.

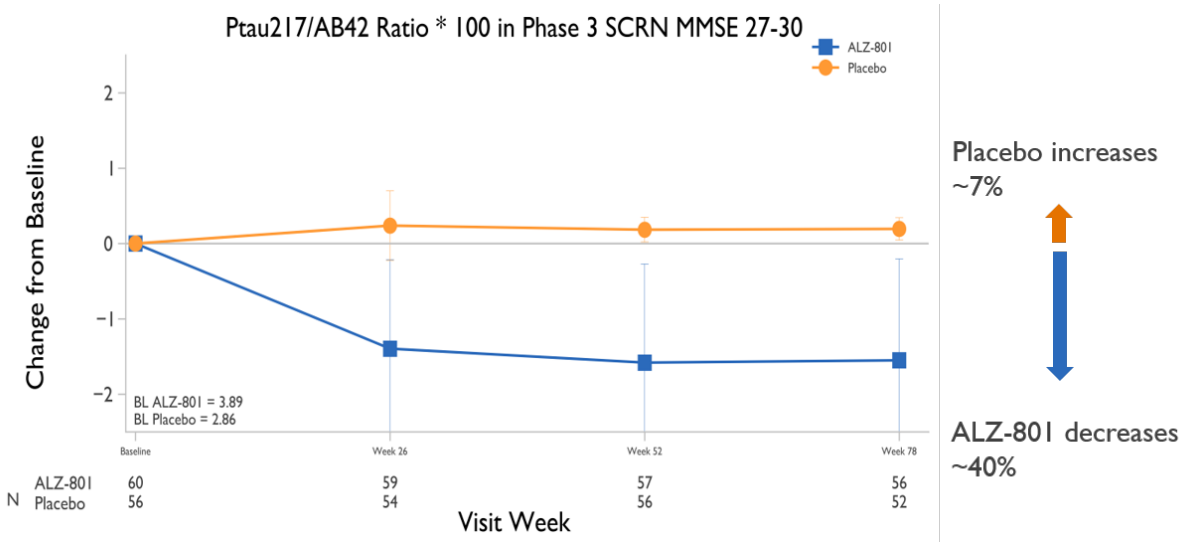

Figure S3. Longitudinal changes from baseline (CBL) of plasma p-tau217/Aβ42 ratio following valiltramiprosate (ALZ-801) 265 mg BID vs. placebo treatment in the MCI APOE4/4 population of the Phase 3 trial. Data are shown as observed case analysis; means ± SEM, n=60 for valiltramiprosate and n=56 for placebo. These data show a rapid and sustained reduction of plasma p-tau217/Aβ42 ratio after valiltramiprosate treatment vs. placebo over 78 weeks, similar to the plasma p-tau217 levels in the same subjects.

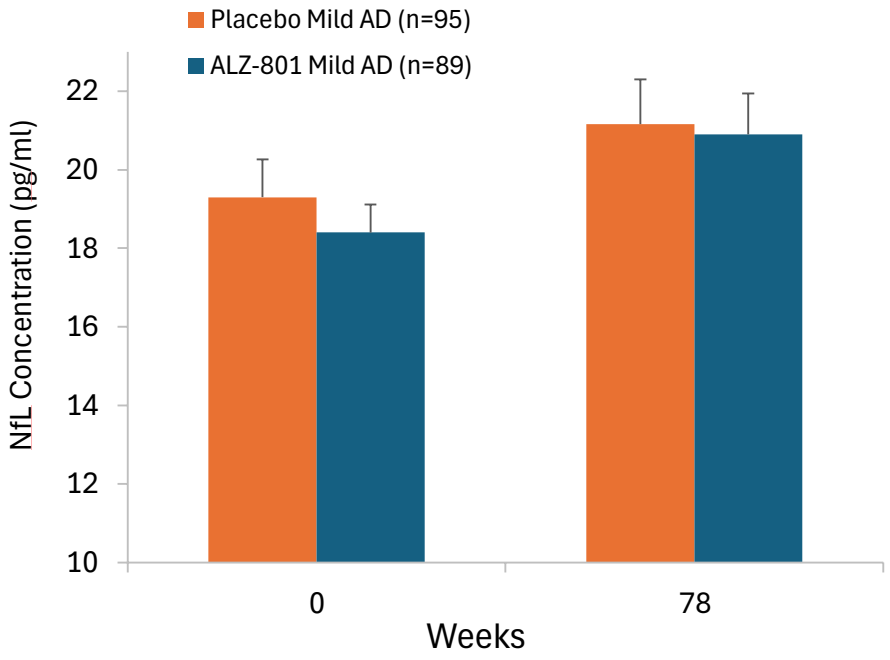

Figure S4. Plasma NfL concentrations (pg/mL) at baseline and at Week 78 following valiltramiprosate (ALZ-801) 265 mg BID vs. placebo treatment in the Mild AD APOE4/4 population of the Phase 3 trial. Data are shown as observed case analysis; n=89 for valiltramiprosate and n=95 for placebo, unpaired, two-tailed t-test. No significant difference ($p>0.05$) was observed between valiltramiprosate-treated and placebo subjects at 78 weeks.

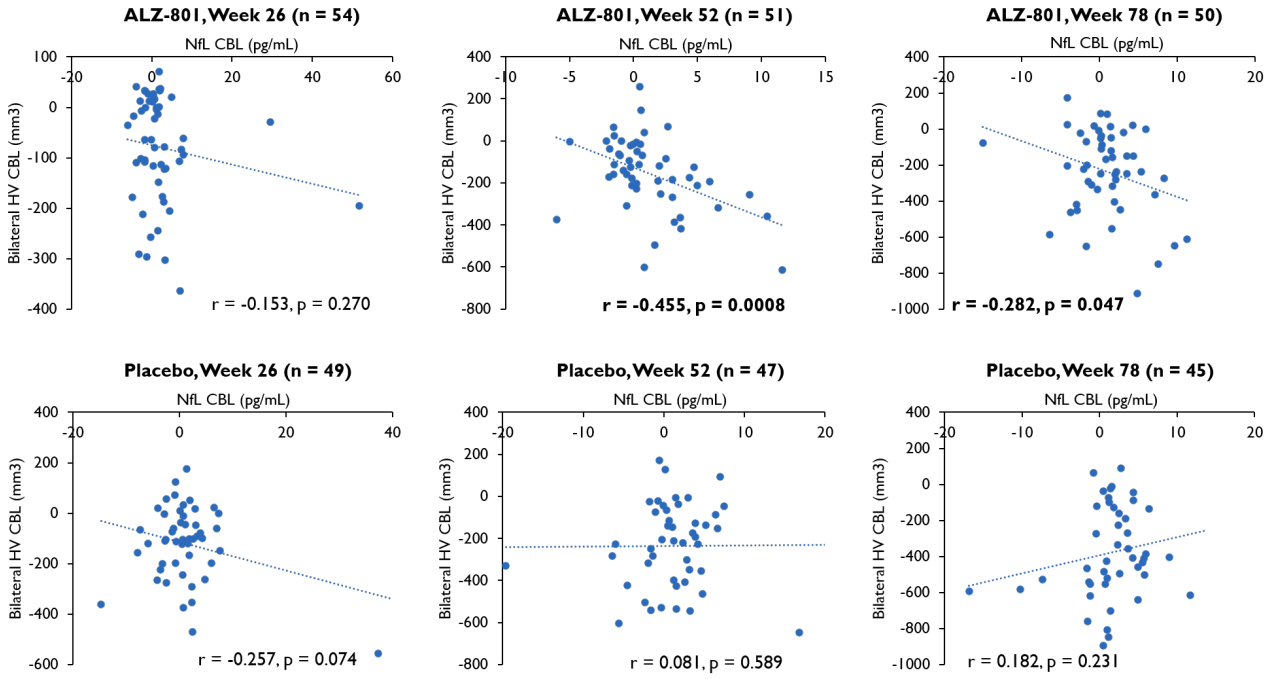

Figure S5. Correlation analysis of plasma NfL changes with bilateral hippocampal volume (HV) changes in valiltramiprosate-treated and placebo subjects at 26, 52 and 78 weeks in the Phase 3 study. Data are shown as observed case analysis. The results show significant correlations between NfL (change from baseline, CBL) and bilateral HV CBL at both Week 52 (p=0.0008) and Week 78 (p=0.047) following valiltramiprosate 265 mg BID treatment, whereas no significant correlations were observed in the placebo subjects.

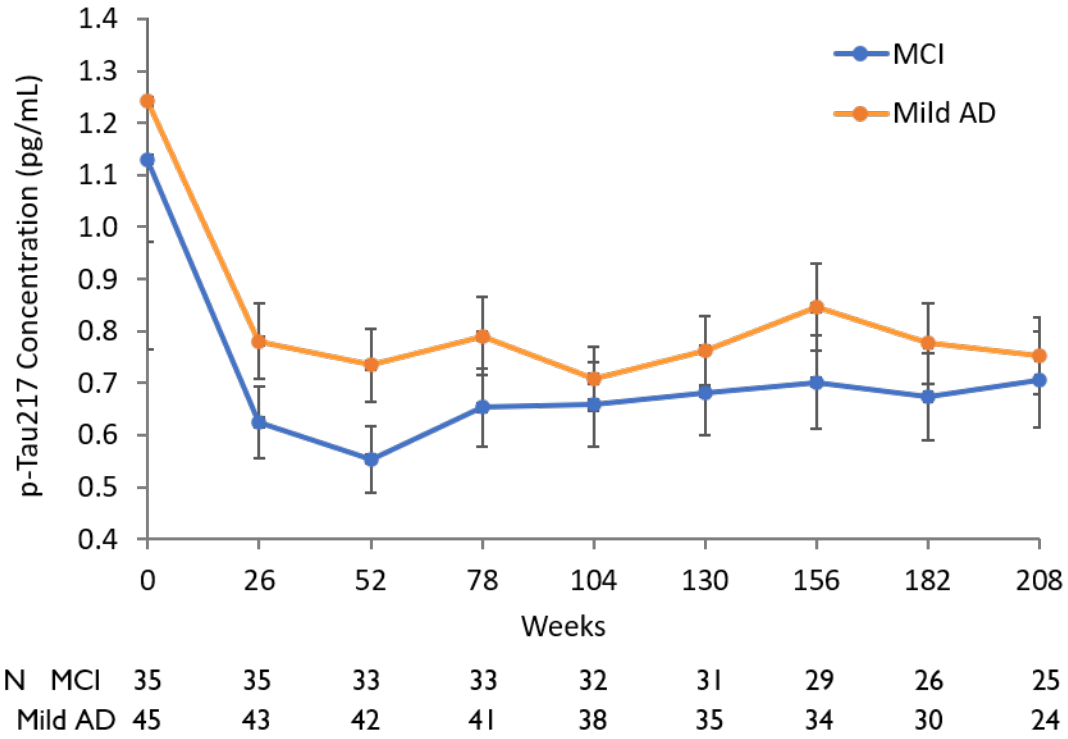

Figure S6. Longitudinal absolute value time course of plasma p-tau217 following valiltramiprosate (ALZ-801) 265 mg BID in MCI and Mild AD APOE4 carriers in the Phase 2 trial over 4 years. Data are shown as observed case analysis; means ± SEM, n=35 for MCI and n=45 for Mild AD subjects. These data show a rapid reduction of plasma p-tau217 after valiltramiprosate treatment. The observed reduction was sustained over 4 years in both subgroups and was numerically greater in MCI vs. Mild AD subjects.