PLASMA PROTEOMIC ANALYSIS FROM ALZHEIMER'S PATIENTS IN SPARC CLINICAL TRIAL TO IDENTIFY PHARMACODYNAMIC BIOMARKERS OF THE S2R MODULATOR CT1812

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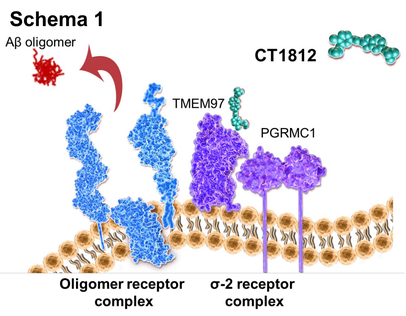
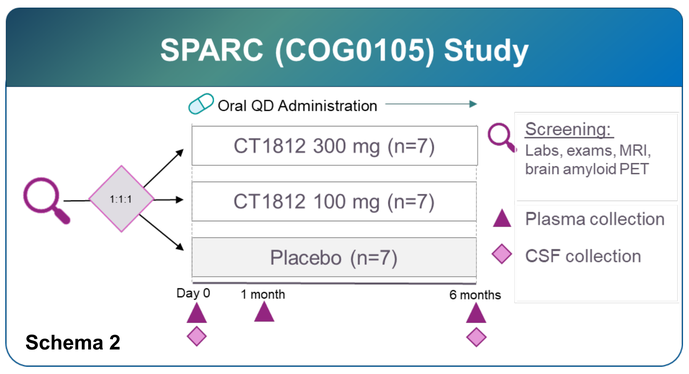
INTRODUCTION

CT1812 is a small molecule modulator of the sigma-2 receptor (S2R, *TMEM97*) that displaces Aβ oligomers bound to neuronal synapses[1] (**Schema 1**). In preclinical studies, CT1812 protects synapses, facilitates their restoration and improves cognitive performance in transgenic Alzheimer's disease (AD) mice[1].

Schema 1



SPARC was a randomized, double-blind, placebo-controlled Phase 1 clinical trial assessing effects of two CT1812 doses (100mg, 300mg; oral, once daily) in patients with mild to moderate AD (**Schema 2**). Treatment was generally well-tolerated and was consistent with the overall safety and tolerability profile from previous studies.

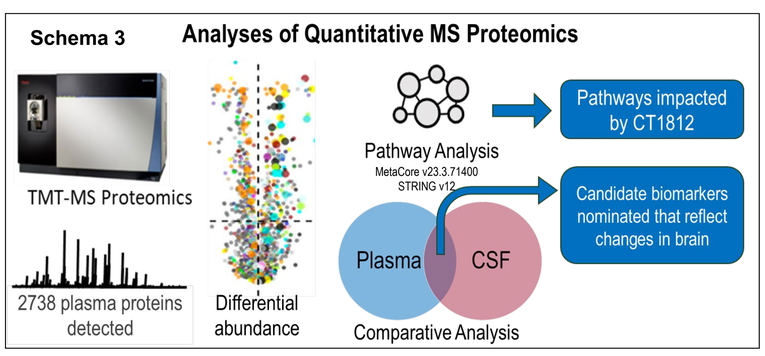


Schema 2

GOALS: 1) Identify plasma pharmacodynamic biomarkers of target/pathway engagement by CT1812 using unbiased proteomic analysis; 2) compare with previous analyses identifying CSF pharmacodynamic biomarkers of CT1812 from SPARC trial participants.

METHODS

Tandem-mass tag mass spectrometry (TMT-MS) followed by unbiased quantification of plasma proteomes was conducted on baseline, 1-month (N=21) and 6-month (N=18) plasma. All biomarker analyses reported herein were exploratory, and for the purpose of identifying pharmacodynamic changes of CT1812, only patients who were actively taking their treatment, as indicated by bioanalysis of drug exposure levels (herein referred to as treatment-compliant patients), were included in the analysis. Change from baseline was calculated, and treatment effects were assessed through differential abundance analysis (pooled drug vs placebo; p≤0.1), followed by pathway analyses. Plasma proteomes were compared across timepoints and to CSF proteome at 6 mo, to identify plasma and CSF biomarkers commonly altered by CT1812.(**Schema 3**).



CSF Proteomic Analysis Identified Pharmacodynamic (PD) Biomarkers Impacted by CT1812

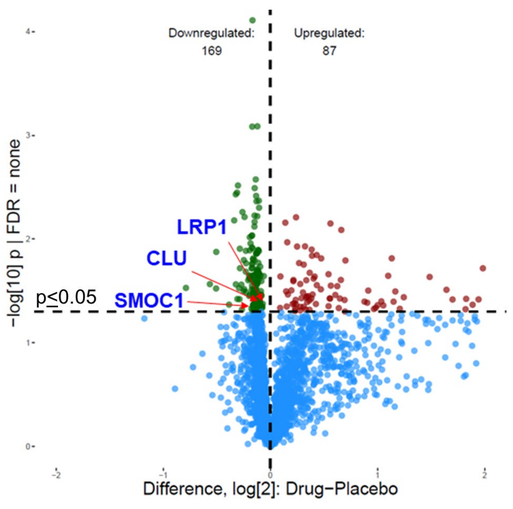


Fig 1. Volcano plot illustrates differentially abundant proteins (256 total; CT1812 vs Placebo) at p≤0.05, with proteins of interest labeled with red arrows.

PD Biomarkers of CT1812 (155) Identified in Plasma After 1 Month of Treatment

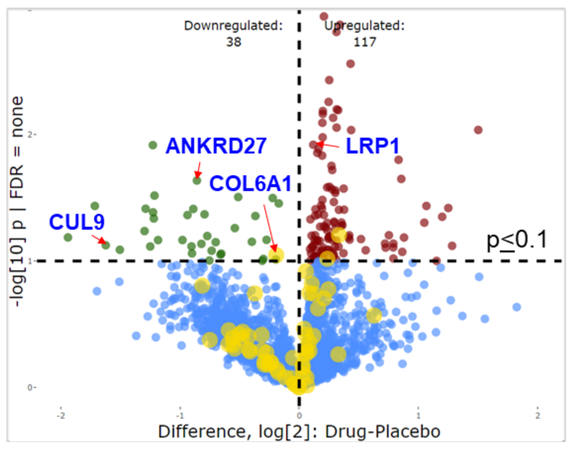


Fig 2. Volcano plot illustrates differentially abundant proteins (155 total; CT1812 vs Placebo) at p≤0.1, with proteins of interest labeled with red arrows.

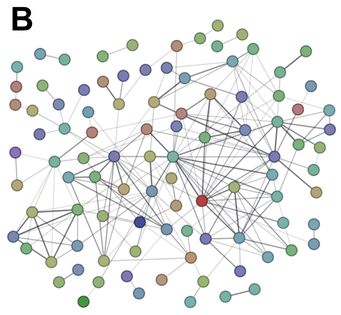
Early Effects (1 mo) of CT1812 in Regulating Amyloid Biology, Immune Response, and β-Catenin Signaling

A

Top Metacore Pathway Maps (SPARC 1mo Plasma; CT1812 vs placebo; p≤0.1)	p-value
Gamma-secretase proteolytic targets	1.32E-06
Cell adhesion - Cell-matrix glycoconjugates	1.36E-04
Gamma-secretase regulation of osteogenesis	1.66E-04
Immune response - Lectin induced complement pathway	3.97E-04
Gamma-Secretase regulation of neuronal cell development and function	6.56E-04
Development - Negative regulation of STK3/4 (Hippo) pathway and positive regulation of YAP/TAZ function	9.03E-04
Development - WNT/Beta-catenin signaling in organogenesis	1.28E-03
Signal transduction - PDGF signaling via PI3K/AKT and NFkB pathways	1.42E-03
Transport - Low density lipoproteins assembly and remodeling	1.67E-03
Immune response - IL-1 signaling	2.66E-03

N.B. non-relevant disease pathologies/organs excluded.

B



STRING v12.0
PPI enrichment p-value: 1.0e-16

Fig 3. A) Differentially abundant proteins (p≤0.1) in 1 mo plasma were analyzed for pathway enrichment using Metacore. **B)** STRING analysis illustrates the interconnectivity between proteins, with Protein-Protein enrichment p value of 1.0e-16.

PD Biomarkers of CT1812 (68) Identified in Plasma After 6 Months of Treatment

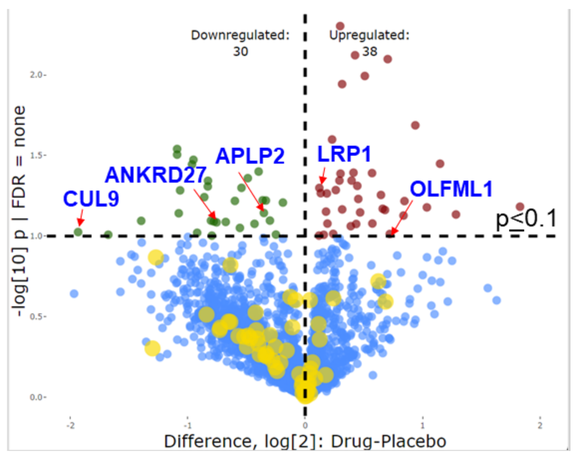


Fig 4. Volcano plot illustrates differentially abundant proteins (68 total; CT1812 vs Placebo) at p≤0.1, with proteins of interest labeled with red arrows.

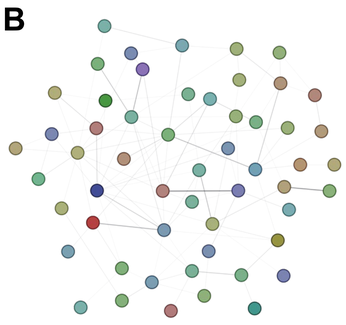
Pathways Enriched in 6 Month Plasma: Early Pathway Effects of CT1812 are Sustained

A

Top Metacore Pathway Maps (SPARC 6mo Plasma; CT1812 vs placebo; p≤0.1)	p-value
Cell adhesion – Desmosomes	4.33E-05
Regulation of **immune cell** differentiation by Notch signaling	7.17E-04
Signal transduction - Angiotensin II/AGTR1 signaling via **Notch, Beta-catenin and NF-kB pathways**	2.92E-03
Gamma-secretase proteolytic targets	3.14E-03
Development - NOTCH signaling activation	3.36E-03
Development - NOTCH signaling in the nervous system	4.24E-03
Development - Transcription regulation of granulocyte development	6.11E-03
Cytoskeleton remodeling - Keratin filaments	7.69E-03
Development - Negative regulation of **WNT/Beta-catenin signaling** at the receptor level	1.18E-02
Development - NOTCH signaling in myogenesis	1.29E-02

N.B. non-relevant disease pathologies/organs excluded.

B

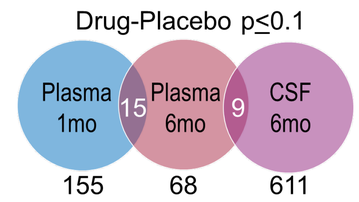


STRING v12.0
PPI enrichment p-value: 0.0138

Fig 5. A) Differentially abundant proteins (p≤0.1) in 6 mo plasma were analyzed for pathway enrichment using Metacore. **B)** STRING analysis illustrates the interconnectivity between proteins, with Protein-Protein enrichment p value of 0.0138.

Candidate Plasma Biomarkers Identified, Altered in a Similar Direction at Both 1 and 6 Months

A



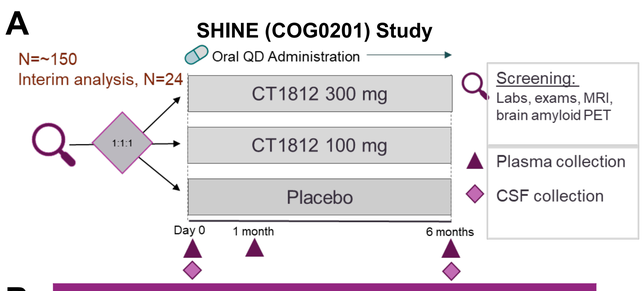
B **Overlapping proteins across biofluids**

Protein	1mo Plasma log2FC	6mo Plasma log2FC	6mo CSF log2FC	1mo Plasma p-value	6mo Plasma p-value	6mo CSF p-value
LRP1	0.12	0.12	-0.10	<0.05	<0.1	<0.05
IGHA1	0.20	0.18	0.32	<0.05	<0.1	<0.1

Fig 6. A) Venn diagram illustrates overlapping proteins between 1 mo and 6 mo plasma or between 6 mo plasma and CSF (CT1812 vs placebo (p≤0.1)). **B)** Listed are overlapping significant (p≤0.1) proteins changed across biofluids with CT1812 treatment vs placebo (red: increased; blue: decreased).

Pathways Enriched by CT1812 Treatment in SPARC Cohort Are Congruent With Pathways Identified in Independent AD Cohort (SHINE-A) Plasma

A



B

Top Metacore Pathway Maps (SHINE-A 1mo Plasma; CT1812 vs placebo; p≤0.1)	p-value
Cell adhesion – Desmosomes	8.94E-07
Cytoskeleton remodeling – Keratin filaments	1.29E-05
Signal transduction – **Anti-inflammatory** and cardioprotective Adiponectin signaling	2.60E-05
Signal transduction – Angiotensin II/AGTR1 signaling via **Notch, Beta-catenin and NF-kB pathways**	2.76E-04
Signal transduction – NOTCH signaling in tissue homeostasis	3.19E-04

N.B. non-relevant disease pathologies/organs excluded.

C

Top Metacore Pathway Maps (SHINE-A 6mo plasma; CT1812 vs placebo; p<0.1)	p-value
Development – NOTCH signaling activation	5.30E-05
Immune response – Antigen presentation by MHC class I, classical pathway	1.46E-04
Notch signaling in oligodendrocyte precursor cell differentiation in multiple sclerosis	3.92E-04
Gamma-secretase regulation of angiogenesis	4.34E-04
Cell adhesion – Cell-matrix glycoconjugates	8.76E-04

N.B. non-relevant disease pathologies/organs excluded.

Fig 7. A) Schematic representation of SHINE clinical trial. Differentially abundant proteins at p≤0.1 in **B)** 1 mo plasma (N=22) or C**)** 6 mo plasma (N=21) from treatment-compliant patients in the SHINE interim analysis (SHINE-A) trial[2] were analyzed for pathway enrichment using Metacore.

CONCLUSIONS

- **Pathways enriched by proteins significantly affected by CT1812 support a role for CT1812 in modulating Aβ biology, Notch/β-Catenin signaling, and neuroinflammation.**
- **Pathways enriched by CT1812 in SPARC patient biofluids are congruent with pathways altered by CT1812 in SHINE-A trial, indicative of pathway engagement that replicates across independent trial cohorts.**
- **Across plasma and CSF, CT1812 impacted abundance of LRP1, a protein that has functional links to amyloid biology in both brain and periphery.[3,4]**

Data reveal biological effects of S2R modulator CT1812 that replicate across biofluids and across independent trial cohorts

Other Posters on CT1812 by Cognition Therapeutics

Abstract 2964: Identification of New Pharmacodynamic Biomarkers of CT1812 That Correlate With Favorable Functional Connectivity of the Brain
V. Di Caro, K. Pandey, E. Cho, D. Duong, W. de Haan, M Grundman, N. Seyfried, A. Caggiano, E. Vijverberg, M. Hamby

Abstract 2965: Analysis of CSF Samples From a Phase 2 Clinical Trial in Alzheimer's Patients Show That CT1812 Can Modulate α-Synuclein
V. Di Caro, I. Levey, K. Pandey, D. Duong, N. Seyfried, M. Grundman, E. Vijverberg, A.O Caggiano, C. Teunissen, M.E. Hamby

ClinicalTrials.gov: NCT03493282
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Corresponding author: mhamby@cogrx.com

REFERENCES
1. Izzo et al. Preclinical and clinical biomarker studies of CT1812: A novel approach to Alzheimer's disease modification. Alz & Dementia 2021.
2. Lizama et al. Proteomic Analysis of Plasma in a Phase 2 Clinical Trial in Alzheimer's Patients to Identify Pharmacodynamic Biomarkers of the S2R Modulator CT1812. Poster presented at: Clinical Trials on Alzheimer's Disease (CTAD); Boston, Massachusetts.
3. Shinohara et al. Role of LRP1 in the pathogenesis of Alzheimer's disease: evidence from clinical and preclinical studies. J Lipid Res. 2017.
4. Kanekiyo et al. The low-density lipoprotein receptor-related protein 1 and amyloid-6 clearance in Alzheimer's disease. Front aging neurosci. 2014

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