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Summary of Alzheon Peer-Reviewed Scientific Publications

1. *"Hippocampal Atrophy on Magnetic Resonance Imaging as a Surrogate Marker for Clinical Benefit and Neurodegeneration in Early Symptomatic Alzheimer's Disease: Synthesis of Evidence from Observational and Interventional Trials"*

Published in December 2025 in *Drugs*

Susan Abushakra, MD – Chief Medical Officer of Alzheon, Murali Doraiswamy, **John A. Hey, PhD** – Chief Scientific Officer of Alzheon, Duygu Tosun, Frederik Barkhof, Jerome Barakas, Jeffrey Petrella, Patrick Kesslak, **Aidan Power, MB, MRCPsych** – Chief Development Officer of Alzheon**,** Marwan Sabbagh, Anton Porsteinsson, Sharon Cohen, Serge Gauthier, Craig Ritchie, David Watson, Emer McSweeney, Merce Boada, Earvin Liang, Luc Bracoud, Rosalind McLaine, Susan Flint, Jean Schaefer, Jeremy Yu, Margaret Bray, Suzanne Hendrix, Sam Dickson, Abe Durrant, **Adem Albayrak** – Chief Data Officer of Alzheon**, Martin Tolar, MD, PhD** – Founder, President & CEO of Alzheon

Actionable Insights: This comprehensive analysis evaluated hippocampal volume (HV) as a potential surrogate outcome to predict clinical benefit in early Alzheimer's disease (AD), addressing the limitations of amyloid-plaque reduction as the current surrogate accepted for accelerated approval. Using data from 30 observational studies (>13,000 participants) and nine placebo-controlled clinical trials of five anti-amyloid agents (aducanumab, lecanemab, donanemab, gantenerumab, and valiltramiprosate), the study examined correlations between HV, cognitive performance, drug effects, and neurodegeneration. Across trials totaling ~10,000 treated participants, drug-related slowing of hippocampal atrophy showed a linear correlation with slowing of cognitive decline, supporting HV as a meaningful predictor of clinical outcomes. Two valiltramiprosate studies demonstrated significant subject-level correlations between HV preservation and cognitive benefit ($r = -0.40$ to -0.44), with complementary DTI findings revealing decreased mean diffusivity, consistent with reduced neurodegeneration rather than edema. The analysis estimates that ≥40 mm^3 HV preservation or ≥10% slowing of hippocampal atrophy over 18 months corresponds to clinically meaningful benefit at the MCI stage. Drawing from more than 23,000 subjects across three decades, the findings reinforce HV as an early, sensitive, and biologically grounded measure that predicts cognitive trajectories and treatment effects in early symptomatic AD.

Key Points: (A) Amyloid-plaque clearance is currently the only recognized surrogate outcome for predicting clinical benefit but is not useful for evaluating non-plaque-clearing therapies. Hippocampal volume (HV), affected early in AD, may serve as a more broadly applicable surrogate outcome. (B) Data from 30 observational studies in early AD demonstrated significant and consistent cross-sectional correlations between baseline HV and cognition, and

longitudinal correlations between HV atrophy and cognitive decline. (C) Across nine anti-amyloid clinical trials (~10,000 subjects), slowing of hippocampal atrophy showed a linear relationship with slowing of cognitive decline. Subject-level correlations were confirmed in two valiltramiprosate studies, supported by DTI microstructural measures indicative of neuroprotection. (D) A minimum threshold of ≥40 mm^3 or ≥10% hippocampal atrophy slowing over 18 months corresponds to meaningful clinical benefit at the MCI stage. Data from over 23,000 individuals support HV as a robust, predictive surrogate marker for early symptomatic AD, sensitive to pharmacologic treatment effects and reflective of underlying neurodegenerative processes.

2. *Clinical Efficacy, Safety and Imaging Effects of Oral Valiltramiprosate in APOE ε4/ε4 Homozygotes with Early Alzheimer's Disease: Results of the Phase III, Randomized, Double-Blind, Placebo-Controlled, 78-Week APOLLOE4 Trial*

Published in September 2025 in *Drugs*

Susan Abushakra, MD – Chief Medical Officer of Alzheon, **Aidan Power, MB, MRCPsych** – Chief Development Officer of Alzheon**,** David Watson, Anton Porsteinsson, Marwan Sabbagh, Emer MacSweeney, Sharon Cohen, Mercè Boada Rovira, Murali Doraiswamy, Earvin Liang, Susan Flint, J. Patrick Kesslak, Rosalind McLaine, **Adem Albayrak** – Chief Data Officer of Alzheon**,** Jean Schaefer, Jeremy Yu, Luke Tolar, Sam Dickson**, John A. Hey, PhD** – Chief Scientific Officer of Alzheon**, Martin Tolar, MD, PhD** – Founder, President & CEO of Alzheon

Actionable Insights: This pivotal Phase 3 APOLLOE4 trial evaluated oral ALZ-801/valiltramiprosate (265 mg BID) in 325 APOE4/4 homozygotes with early Alzheimer's disease (AD), including mild cognitive impairment (MCI) and mild dementia, over 78 weeks. The primary endpoint was ADAS-Cog13, with secondary measures including CDR-Sum of Boxes, Amsterdam-IADL, Disability Assessment for Dementia, hippocampal MRI, and diffusion tensor imaging. In the overall population, ALZ-801 did not significantly improve clinical outcomes versus placebo but significantly slowed hippocampal atrophy by 18%. Prespecified analyses showed marked benefits in the MCI subgroup, with 52% slowing of cognitive decline (ADAS-Cog13), 96% slowing of functional decline (DAD), and significant hippocampal and cortical atrophy slowing. Safety was favorable, with no increased risk of amyloid-related imaging abnormalities (ARIA) compared with placebo, and gastrointestinal events (nausea, vomiting, appetite loss) as the most common side effects.

Key Points: (A) In the overall Early AD APOE4/4 population, ALZ-801 showed significant slowing of hippocampal atrophy but no statistically significant clinical efficacy. (B) In the prespecified MCI subgroup, ALZ-801 achieved nominally significant benefits in cognition, function, and brain atrophy, with statistically significant positive imaging–clinical correlations.

(C) The safety profile was favorable, with no ARIA risk in this genetically high-risk group. (D) Conclusions: ALZ-801 may provide a favorable benefit–risk profile and practical oral treatment option for APOE4/4 homozygotes at the MCI stage of AD, informing future intervention and prevention trials.

3. *Clinical Pharmacokinetics of Oral ALZ-801/Valiltramiprosate in a Two-Year Phase 2 Trial of APOE4 Carriers with Early Alzheimer's Disease*

Published in February 2025 in *Clinical Pharmacokinetics*

John A. Hey, PhD – Chief Scientific Officer of Alzheon, Jeremy Y. Yu, **Susan Abushakra, MD** – Chief Medical Officer of Alzheon, Jean F. Schaefer, **Aidan Power, MB, MRCPsych** – Chief Development Officer of Alzheon, Pat Kesslak, Jijo Paul and **Martin Tolar MD, PhD** – Founder, President & CEO of Alzheon

Actionable Insights: ALZ-801/valiltramiprosate is an oral, small-molecule inhibitor of β-amyloid (Aβ) oligomer formation in late-stage development as a potential disease-modifying therapy for Alzheimer's disease (AD). The ALZ-801 Phase 2 study was designed to evaluate longitudinal effects of ALZ-801 (265 mg BID) on plasma, CSF and volumetric MRI AD biomarkers, and clinical outcomes over 104 weeks in APOE4 carriers with Early AD. Eighty-four subjects (31 APOE4/4 homozygotes and 53 APOE3/4 heterozygotes) with positive CSF biomarkers of amyloid and tau pathology were enrolled. Following oral dosing, ALZ-801 was rapidly and fully converted to the active moieties, tramiprosate and 3-SPA. The inter-subject variability in plasma drug levels was low, confirming the superior performance of ALZ-801 vs. oral tramiprosate tablet (150 mg BID) from the earlier tramiprosate Phase 3 trials. ALZ-801 was well-tolerated without drug-related severe or serious adverse events or laboratory findings. Over the 2 years of treatment, there was no event of amyloid-related imaging abnormalities with edema (ARIA-E).

Key Points: (A) ALZ-801 was rapidly absorbed and converted to its active moieties, tramiprosate and 3-SPA. The steady-state plasma exposures of tramiprosate and 3-SPA were sustained, consistent with an earlier 2-week Phase 1b pharmacokinetic (PK) study in APOE4 carrier AD subjects, as well as a 7-day Phase 1 PK study in heathy elderly volunteers. The inter-individual variability was low. (B) The plasma exposures for ALZ-801, tramiprosate and 3-SPA were not affected by sex, APOE genotype, age, body mass index, concomitant acetylcholinesterase inhibitor use, or tablet lot. The plasma exposures of both tramiprosate and 3-SPA, but not ALZ-801, inversely correlated with estimated glomerular filtration rate (eGFR), consistent with renal excretion as the primary route of elimination. (C) These results demonstrate favorable PK properties of ALZ-801 and support its development as a potential oral disease-modifying agent for AD therapy.

4. *Point of View: Challenges in Implementation of New Therapies for Alzheimer's Disease*

Published in January 2025 in *The Journal of Prevention of Alzheimer's Disease*

Sandar Aye, Gunilla Johansson, Christoph Hock, Lars Lannfelt, John R Sims, Kaj Blennow, Kristian S Frederiksen, Caroline Graff, José Luis Molinuevo, Philip Scheltens, Sebastian Palmqvist, Michael Schöll, Anders Wimo, Miia Kivipelto, Ron Handels, Lutz Frölich, Norbert Zilka, **Martin Tolar MD, PhD** – Founder, President & CEO of Alzheon, Peter Johannsen, Linus Jönsson, Bengt Winblad

Actionable Insights: To highlight and address the challenges of implementing the new upcoming Alzheimer's disease (AD) disease modifying treatment (DMT), a symposium was convened at the Nobel Forum at the Karolinska Institutet, Sweden, where international researchers and experts were brought together. The initial step in managing DMT in clinical practice involves identifying patients eligible for treatment. These recommendations suggest that eligibility may be assessed across four domains such as diagnosis and staging, biomarker assessment, structural imaging, and comorbidities assessment. One important factor to consider in people diagnosed with AD who may be eligible for treatment with DMT is the presence or absence of the apolipoprotein E (*APOE*) gene variant *APOEε4*. Individuals carrying the *APOEε4* variant have an increased risk of developing AD compared to non-carriers. *APOEε4* carriers are at a higher risk for amyloid- related imaging abnormalities (ARIA), which are adverse events associated with donanemab and lecanemab, and may lead to serious symptoms. Moreover, the clinical efficacy of lecanemab differs between individuals who carry the *APOEε4* gene and those who do not, with reduced efficacy observed in people who are *APOEε4* homozygote carriers. The worldwide societal economic burden of AD and related dementias is estimated at $1.3 trillion. Direct medical costs account for a small fraction of this total, with the majority attributed to social services and informal care outside the healthcare system. Given the high drug price of DMTs (for lecanemab, $26,500 per year in the U.S. and $20,500 in Japan), there is concern about the financial implications. From the reimbursement agency perspective, drug reimbursement decisions are guided by three principles: non-discrimination to all human life, equitable distribution of healthcare resources, and maximizing health gains, i.e., cost-effectiveness. Due to the complexity of AD pathophysiology, meaningful improvements are more likely to come gradually through ongoing refinement of molecular strategies, better patient selection, and the use of combination therapies. Examples of therapies under development include AADvac1, ALZ-801, and glucagon-like peptide-1 receptor agonist (GLP1-RA), each offering potential benefits for treating AD.

Key Points: (A) The initial step for appropriate disease modifying treatment (DMT) involves the identifying the right patient. For most patients, the diagnostic pathway starts with primary care physicians (PCP). Today, standard practice with PCPs does not involve disease-related

biomarkers or genetic testing. Blood based biomarkers may help with this in the future. (B) Appropriate Use Recommendations for DMT recommend biomarker and genetic testing as well as clinicians discussing the risks and benefits of treatment. However, implementing these recommendations in clinical practice presents challenges at the individual, health care system, and society levels. (C) Many recent clinical trials, including TRAILBLAZER-ALZ2 study, suggest a greater benefit from DMTs if initiated at an earlier disease stage. (D) Conclusions: The advancement of DMTs for AD presents a significant and positive development in the treatment landscape, offering new hope in addressing the substantial unmet need for effective AD treatment. However, several challenges and considerations must be addressed before implementing such treatment in clinical settings.

Therefore, the expert group who convened in symposium recommends the following: 1. *Establish treatment guidelines for the right patient*: Develop clear inclusion and exclusion criteria to ensure that only the most suitable patients receive these treatments, i.e., personalized medicine, thereby maximizing benefits and minimizing risks; stopping rules should also be discussed. 2. *Physician training to identify patients at the right time:* Equip both primary care providers and memory clinic specialists with the necessary skills to interpret biomarker results, effectively communicate them with patients, and recognize treatment side effects. 3. *Follow-up studies and invest in patient registry*: Conduct non-company-sponsored unbiased follow-up studies to confirm the clinical effectiveness and economic value of ATTs in routine care. Invest in registries to characterize patient subgroups who benefit more from treatment and guide the optimal use of these therapies. 4. *Consideration of new and combination therapies to address the right biologic targets*: Explore the potential of novel combination therapies to fully address the complex pathology of AD.

ALZ-801, being developed by Alzheon, is a promising potential future therapy due to the precision approach focusing on patients with the *APOEε4* gene who are at the earliest symptomatic stages of AD, its novel mechanism of action, oral formulation which makes it more easily available and potential stabilization efficacy without the risk of vasogenic brain edema.

5. *APOLLOE4 Phase 3 Study of Oral ALZ-801/Valiltramiprosate in APOE ε4/ε4 Homozygotes with Early Alzheimer's Disease: Trial Design and Baseline Characteristics*

Published in August 2024 in *Alzheimer's and Dementia: Translational Research & Clinical Interventions*

Susan Abushakra, MD – Chief Medical Officer of Alzheon, Anton Porsteinsson, Marwan Sabbagh, David Watson, **Aidan Power, MB, MRCPsych** – Chief Development Officer of Alzheon, **Earvin Liang,** Emer MacSweeney, Merce Boada, **Susan Flint, Rosalind McLaine, Patrick Kesslak**, **John A. Hey, PhD** – Chief Scientific Officer of Alzheon, Jeremy Y. Yu, **Martin Tolar MD, PhD** – Founder, President & CEO of Alzheon

Actionable Insights: The APOLLOE4 Phase 3, placebo-controlled, 78-week study is designed to evaluate the efficacy and safety of oral ALZ-801 265 mg twice per day in early Alzheimer's disease (AD) subjects with the apolipoprotein E (APOE) 4/4 genotype. This trial which enrolled 325 subjects with early symptomatic AD is unique in its focus on APOE4/4 homozygotes who typically have earlier onset of AD pathology and symptoms than other APOE genotypes. The primary outcome is the cognitive Alzheimer's Disease Assessment Scale 13-item Cognitive Subscale (ADAS-Cog13), with two functional measures as key secondary outcomes (Clinical Dementia Rating-Sum of Boxes, Amsterdam-Instrumental Activities of Daily Living), and with hippocampal volume and fluid biomarkers as additional outcomes. Safety is paramount for new therapies in Alzheimer's in the APOE4/4 population, since the currently approved amyloid-antibody therapies for early Alzheimer's have boxed warnings for the high risk of Amyloid Related Imaging Abnormalities (ARIA, with both edema and hemorrhages) in APOE4/4 patients. This study is unique in allowing APOE4/4 patients with a large number of microhemorrhages or siderosis at baseline magnetic resonance imaging, lesions that indicate concomitant cerebral amyloid angiopathy (CAA) that is an important risk factor for ARIA. Study results will be available in the second half of 2024 and, if positive, ALZ-801 may become the first oral amyloid-targeted drug to demonstrate a favorable benefit/risk profile in APOE4/4 subjects.

Key Points: (A) The APOE4 allele is the strongest genetic risk factor for sporadic AD, with double alleles (APOE4/4 homozygotes) increasing risk by 12-fold. Further, the approved amyloid antibodies for early Alzheimer's disease (AD) carry a boxed warning about the risk of ARIA which is highest in APOE4 homozygotes. The APOE4/4 genotype is a major risk factor for both developing AD and CAA. (B) Utilizing a precision approach for APOLLOE4, approximately 6500 individuals were screened to identify ~650 APOE4 homozygotes, of whom 325 fulfilled all inclusion/exclusion criteria and were randomized. The randomized patients included early AD patients with MMSE ≥ 22 and were individuals pre-specified with either mild cognitive impairment (MCI) or mild AD (National Institute on Aging-Alzheimer's Association clinical criteria). Volumetric MRI measures and fluid biomarkers were additional outcomes, including the assessment of hippocampal atrophy and cortical thickness. (C) Significant drug effects on hippocampal atrophy may indicate neuroprotection and further support clinical outcomes and provide valuable insights into its correlation to clinical benefit. This would aid the design of future trials in presymptomatic individuals, as the development of preventative treatments for this population is a high priority. (D) Conclusions: The APOE4 homozygote population constitutes 10-15% of the 6.7 million individuals living with AD in the United States alone. APOE4/4 patients have a pressing need for an effective and safe treatment that avoids the risk of serious ARIAs and its challenging and burdensome management in clinical practice. The APOLLOE4 Phase 3 trial has the potential to demonstrate the meaningful clinical benefits of ALZ-801 with favorable safety and long-term tolerability. ALZ-801 has the additional advantages of a convenient oral formulation, a simple dosing regimen, limited MRI monitoring, and potentially wider accessibility to patients from diverse communities and geographies.

6. *Analysis of Cerebrospinal Fluid, Plasma β-Amyloid Biomarkers, and Cognition from a 2-Year Phase 2 Trial Evaluating Oral ALZ-801: Valiltramiprosate in APOE4 Carriers with Early AD Using Quantitative Systems Pharmacology Model*

Published in June 2024 in *Drugs*

John A. Hey, PhD – Chief Scientific Officer of Alzheon, Jeremy Y. Yu, **Susan Abushakra, MD** – Chief Medical Officer of Alzheon, **Jean F. Schaefer**, **Aidan Power, MB, MRCPsych** – Chief Development Officer of Alzheon, **Patrick Kesslak**, **Martin Tolar MD, PhD** – Founder, President & CEO of Alzheon**.**

Actionable Insights: This analysis leveraged a comprehensive, validated well established Quantitative Systems Pharmacology (QSP) model of the amyloid cascade to evaluate the effects of treatment with ALZ-801/valiltramiprosate on amyloid fluid biomarkers, an oral inhibitor of beta-amyloid (Aβ) aggregation, in an open-label Phase 2 study of APOE4 carriers with early Alzheimer's disease. Utilizing longitudinal Alzheimer's fluid biomarker and RAVLT-total score data, the analysis showed ALZ-801 significantly arrested or reversed the decline in cerebrospinal fluid (CSF) Aβ42 and plasma Aβ42/Aβ40 levels, respectively, an action consistent with an effective inhibition of amyloid aggregation in AD subjects, as well as reversing the decline in RAVLT-total scores over 104 weeks. The QSP model provides a robust framework for understanding and assessing the ALZ-801's pharmacodynamic effects using fluid biomarkers, and its potential disease-modifying capabilities in an investigational clinical trial setting.

Key Points: (A) The positive Phase 2 study achieved a significant reduction from baseline in plasma p-tau$_{181}$ at all timepoints, reaching 31-43% over 52-104 weeks following treatment with ALZ-801/valiltramiprosate. In addition, a significant reduction in hippocampal volume atrophy was also observed, with a clear correlation between decreased atrophy and stable memory scores at the end of the study. (B) Utilizing published longitudinal trajectory CSF and plasma data, the QSP model indicated that ALZ-801 significantly increased the free non- aggregated fraction of amyloid in both CSF and plasma vs. the natural trajectory of AD progression: 92% slowing of the decrease in Aβ42 in CSF and a 2.4% increase in plasma Aβ42/Aβ40 (vs. a 5.2% decrease in natural progression), supporting its mechanism of action in inhibiting amyloid aggregation. (C) 104-week treatment with ALZ-801 resulted in a 4.1% improvement in the RAVLT-total score vs. natural history trajectories (ADNI-1 and Mayo Normative Studies). (D) Conclusions: The significant reduction in plasma p-tau$_{181}$ and increase in non-aggregated, monomeric amyloid levels, coupled with the observed cognitive stabilization and decreased hippocampal atrophy, highlight ALZ-801's potential as a disease- modifying treatment. The QSP analysis added critical insights into the mechanism of action and its effects to arrest the progression of amyloid pathology and delineates how the upstream change in fluid amyloid biomarkers predict downstream benefit on cognitive outcomes. These results support further evaluation of treatment with ALZ- 801/valiltramiprosate in a broader population of APOE4

carriers and in prevention trials in amyloid positive AD subjects.

7. *Effects of Oral ALZ-801 on Plasma Biomarkers, Brain Hippocampal Volume, and Cognition: Results of 2 Year Single Arm, Open Label, Phase 2 Trial in APOE4 Carriers with Early AD*

Published in June 2024 in *Drugs*

John A. Hey, PhD – Chief Scientific Officer of Alzheon, **Susan Abushakra, MD** – Chief Medical Officer of Alzheon**,** Kaj Blennow, Eric M. Reiman, Jakub Hort, Niels D. Prins, Katerina Sheardova, **Patrick Kesslak**, Larry Shen, Xinyi Zhu, **Adem Albayrak**, Jijo Paul, **Jean F. Schaefer, Aidan Power, MB, MRCPsych** – Chief Development Officer of Alzheon, **Martin Tolar MD, PhD** – Founder, President & CEO of Alzheon.

Actionable Insights: In APOE4 carriers with early AD, open-label treatment with ALZ-801/valiltramiprosate at a dose 265 mg twice daily by mouth significantly reduced plasma p-tau$_{181}$ levels, starting at 13 weeks and sustained at every visit through 104 weeks. Compared with an external control (ADNI-1 matched cohort), ALZ-801 treatment resulted in stabilization of memory test scores over 104 weeks which significantly correlated with the observed reduction in hippocampal atrophy. ALZ-801 also displayed a favorable safety profile, with no instances of vasogenic brain edema.

Key Points: (A) This Phase 2 study included 84 participants, 52% female, mean age of 69.0 years, 37% homozygotes, and mean MMSE score of 25.7 with 43% considered to have mild cognitive impairment (MCI). (B) The primary outcome was achieved with a significant 31% reduction from baseline in plasma p-tau$_{181}$ at 104 weeks. (C) The key secondary outcome of hippocampal volume atrophy, compared to an external control, also demonstrated a statistically significant reduction at 104 weeks. (D) Memory scores remained stable over 104 weeks and correlated significantly with the observed reduction in hippocampal volume atrophy. (E) Safety assessments revealed common adverse events such as COVID-19 infection and mild nausea, with no drug-related serious adverse events despite enrolling APOE4 carriers, who are at higher risk for ARIA-E events due to higher CAA burden. (F) Conclusions: Treatment with ALZ-801 resulted in a significant reduction in plasma p-tau$_{181}$ and hippocampal volume atrophy, clear demonstrations of target engagement. Together with the observed stabilization of cognitive function and safety profile, these data strongly support the potential of ALZ-801/valiltramiprosate as a disease-modifying treatment for early AD in the high-risk APOE4 carrier population.

8. *The Single Toxin Origin of Alzheimer's Disease and Other Neurodegenerative Disorders Enables Targeted Approach to Treatment and Prevention*

Published in February 2024 in *International Journal of Molecular Sciences*

Martin Tolar MD, PhD – Founder, President & CEO of Alzheon, **John A. Hey, PhD** – Chief Scientific Officer of Alzheon, **Aidan Power, MB, MRCPsych** – Chief Development Officer of Alzheon, **Susan Abushakra, MD** – Chief Medical Officer of Alzheon.

Actionable Insights: Novel biomarkers and disease-modifying treatments have advanced our understanding of Alzheimer's pathogenesis and provide support for our thesis that the aggregation of misfold ed native proteins initiates and drives the pathogenic cascade that leads to Alzheimer's disease and other age-related neurodegenerative disorders. The destruction of brain structures by oligomer assemblies results in characteristic clinical syndromes described in Alzheimer's disease and Parkinson's disease, amyotrophic lateral sclerosis, progressive supranuclear palsy, and many other neurological diseases. These well-conserved pathways of brain neurodegeneration offer multiple opportunities for potential disease-modifying interventions to slow the progression or even prevent the onset of clinical symptoms if administered early in the pathogenic process. ALZ-801 may provide advantages as an oral agent that efficiently crosses the blood brain barrier, selectively interacts with amyloid monomers to inhibit their misfolding, and blocks the formation of neurotoxic soluble amyloid oligomers in a concentration-dependent manner, without affecting insoluble amyloid plaques or fibrils.

Key Points: (A) Soluble beta amyloid aggregates called oligomers or protofibrils initiate and drive pathogenesis of Alzheimer's disease. (B) All other biochemical effects and neurodegenerative changes observed in Alzheimer's brain are a direct response to or downstream effect of initial toxic insult by amyloid oligomers. (C) Other neurodegenerative disorders follow a similar path in which normal brain proteins become trapped in aging brain due to impaired clearance and then misfold and aggregate into neurotoxic species that exhibit prion-like behavior. (D) Inhibition of amyloid misfolding and aggregation by ALZ-801/valiltramiprosate could slow disease progression in Alzheimer's patients.

9. *Neurotoxic Soluble Amyloid Oligomers Drive Alzheimer's Pathogenesis and Represent a Clinically Validated Target for Slowing Disease Progression*

Published in June 2021 in *International Journal of Molecular Sciences*

Martin Tolar MD, PhD – Founder, President & CEO of Alzheon, **John A. Hey, PhD** – Chief Scientific Officer of Alzheon, **Aidan Power, MB, MRCPsych** – Chief Development Officer of Alzheon, **Susan Abushakra, MD** – Chief Medical Officer of Alzheon.

Actionable Insights: Therapeutic agents that can selectively target amyloid oligomers can provide clinical efficacy without risk of vasogenic edema and microbleeds. Developing an assay that could detect and measure amyloid oligomers in the brain would greatly advance drug development efforts of such agents and serve as a tool for clinicians to help diagnose and better treat patients.

Key Points: (A) Analyses of clinical efficacy versus the binding profiles of drugs in recent late-stage AD trials suggests that efficacy outcomes correspond to level of amyloid oligomer binding, while risk of vasogenic edema and microbleeds corresponds with amyloid plaque binding. (B)The finding that soluble neurotoxic amyloid oligomers are important therapeutic targets is consistent with genetic findings in several familial AD mutations: The Osaka and Arctic mutations overproduce soluble amyloid oligomers (protofibrils) and develop early AD symptoms with minimal amyloid plaque. (C) Therapeutic agents that can selectively target amyloid oligomers can provide clinical efficacy without risk of vasogenic edema and microbleeds. Several such agents are in development and the most advanced is ALZ-801, which inhibits oligomer formation and is currently in a Phase 3 trial in AD patients with the APOE4/4 genotype.

10. *APOE4/4 Homozygotes with Early Alzheimer's Disease Show Accelerated Hippocampal Atrophy and Cortical Thinning that Correlates with Cognitive Decline*

Published in December 2020 in *Alzheimer's & Dementia: Translational Research & Clinical Interventions*

Susan Abushakra, MD – Chief Medical Officer of Alzheon, **John A. Hey, PhD** – Chief Scientific Officer of Alzheon, A. Porsteinsson, M. Sabbagh, L. Bracoud, J. Schaerer, **Aidan Power**, D. Scott, J. Suhy, **Martin Tolar MD, PhD** – Founder, President & CEO of Alzheon for the Alzheimer's Disease Neuroimaging Initiative.

Actionable Insights: Rates of hippocampal atrophy and cortical thinning in Alzheimer's

Disease Neuroimaging Initiative (ADNI-1) cohort and tramiprosate Phase 3 trial were analyzed. APOE4/4 subjects with Mild Cognitive Impairment (MCI) or Mild Alzheimer's disease (AD) exhibited significantly smaller baseline hippocampal volumes and accelerated atrophy and cortical thinning rates over 12 months, compared to APOE3/3 subjects. Atrophy rates were significantly correlated with cognitive decline in MCI, but not in Mild AD. Hippocampal atrophy and cortical thinning seem to contribute to cognitive impairment early in the disease process and may be a harbinger of future cognitive decline.

Future Directions: Correlation of drug effects on hippocampal atrophy or cortical thinning to cognitive benefits in Early AD would support their use as potential surrogate outcomes in prevention trials of presymptomatic subjects, allowing trials of shorter duration. These imaging-cognitive correlations in MCI support the utility of these imaging biomarkers in Early AD trials. Rates of hippocampal atrophy or cortical thinning can be used as main outcomes in Phase 2 drug trials of ≥12 months, to predict clinical effects in Early AD.

Key Points: (A) APOE4/4 subjects with MCI were approximately 5 years younger and had significantly smaller hippocampal volumes and worse cognitive scores than APOE3/3 subjects. (B) APOE4/4 subjects with MCI and Mild AD showed significantly faster hippocampus atrophy and cortical thinning over 2 years compared to APOE3/3 subjects. (C) Atrophy rates of hippocampal volume and cortical thinning were significantly correlated with cognitive decline in MCI subjects, but not in Mild AD.

11. Aducanumab, Gantenerumab, BAN2401, and ALZ-801 — the First Wave of Amyloid-Targeting Drugs for Alzheimer's Disease with Potential for Near Term Approval

Published in August 2020 in *Alzheimer's Research & Therapy*

Authors: **Martin Tolar MD, PhD** – Founder, President & CEO of Alzheon, **Susan Abushakra, MD** – Chief Medical Officer of Alzheon, **John A. Hey, PhD** – Chief Scientific Officer of Alzheon, A. Porsteinsson, M. Sabbagh

Actionable Insights: Four anti-amyloid agents with potential for near term approval share a common characteristic of targeting soluble Aβ oligomers, albeit to varying degree. The degree of selectivity for Aβ oligomers and brain exposure drive the magnitude and onset of clinical efficacy, while the clearance of plaques is associated with vasogenic brain edema. Only the highest doses of aducanumab and BAN2401 show modest efficacy, as higher doses are limited by increased risk of vasogenic edema, especially in APOE4 carriers. These limitations can be avoided, and efficacy improved by small molecule agents that selectively inhibit the formation or block the toxicity of Aβ oligomers without clearing amyloid plaques. The most advanced highly selective anti-oligomer agent is ALZ-801, an optimized tablet oral prodrug of tramiprosate, which demonstrated efficacy in homozygous APOE4/4 AD subjects. ALZ-801

selectively and fully inhibits the formation of Aβ42 oligomers at the clinical dose, without evidence of vasogenic edema, and will be evaluated in a Phase 3 trial in homozygous APOE4/4 patients with Early AD.

Key Points: (A) Targeting of amyloid oligomers drives clinical and biomarker efficacy. (B) Time to peak brain levels is an important factor for achieving optimal target engagement and, together with peak brain exposure, drives clinical benefit. (C) Selectivity for Aβ oligomers and the individual pharmacokinetic properties underlie the different safety profiles of the anti-amyloid agents, namely risk of brain edema, ARIA-E, and microhemorrhage, ARIA-H. (D) The consistency of effects of anti-amyloid antibodies on phosphorylated tau, total tau, and neurofilament light chain protein measured in cerebrospinal fluid, supports the important role of these biomarkers in future AD trials. (E) The most advanced selective anti-oligomer agent is ALZ-801, entering Phase 3 for the treatment of Early AD, in a genetically defined high-risk population.

12. *The Path Forward in Alzheimer's Disease Therapeutics: Reevaluating the Amyloid Cascade Hypothesis*

Published in January 2020 in *Alzheimer's & Dementia*

Authors: **Martin Tolar MD, PhD** – Founder, President & CEO of Alzheon, **Susan Abushakra, MD** – Chief Medical Officer of Alzheon, M. Sabbagh

Actionable Insights: Substantial evidence from clinical trials, as well as basic science, now supports primary initiating role of soluble Aβ oligomers in Alzheimer's. Soluble Aβ oligomers, rather than insoluble fibrils or plaques, cause synaptic toxicity and neurodegeneration. Inhibiting the formation of soluble toxic oligomers, or their removal, is the key to clinical efficacy of anti-amyloid treatments.

Key Points: (A) Prioritize development of therapeutic agents, which are highly selective for Aβ oligomers, efficiently cross the blood-brain barrier and achieve sustained brain levels that prevent oligomer toxicity. (B) Test treatments in AD populations enriched for Aβ oligomers, such as carriers of APOE4 genotype. (C) Poor brain penetration of anti-amyloid antibodies as a class result in suboptimal drug levels in the brain, below the sustained levels needed to continuously prevent the formation or effective removal of Aβ oligomers. (D) ALZ-801, an oral drug with favorable long-term safety and high brain penetration that fully inhibits Aβ oligomer formation, will be evaluated in a Phase 3 trial in patients with Early AD who are homozygous for APOE4 genotype.

13. *Discovery and Identification of an Endogenous Metabolite of Tramiprosate and Its Prodrug ALZ-801 that Inhibits Beta Amyloid Oligomer Formation in the Human Brain*

Published August 2018 in *CNS Drugs*

Authors: **John A. Hey, PhD** – Chief Scientific Officer of Alzheon, P. Kocis, J. Hort, **Susan Abushakra, MD** – Chief Medical Officer of Alzheon, **Aidan Power**, M. Vyhnálek, J. Y. Yu, **Martin Tolar MD, PhD** – Founder, President & CEO of Alzheon

Actionable Insights: Discovery that 3-SPA, the primary metabolite of tramiprosate and Alzheon clinical lead ALZ-801, occurs naturally in the human brain, and has Aβ anti-oligomer activity comparable to that of tramiprosate. Study shows that 3-SPA may contribute to the clinical activity of tramiprosate and its prodrug ALZ-801 in AD. The potential protective role of endogenous 3-SPA on normal brain function, and in the pathogenesis of AD, warrants further investigation.

Key Points: (A) We describe the discovery and identification of an endogenous Aβ anti-oligomer substance, 3-sulfopropanoic acid (3-SPA), in the human brain. (B) 3-SPA is the primary metabolite of ALZ-801, which is in late-stage clinical development for the treatment of AD. (C) 3-SPA penetrates the brain and in a tramiprosate Phase 3 trial achieved brain concentrations associated with prevention of Aβ oligomer formation and clinical benefit in AD patients carrying the APOE4 genotype. (D) The levels of 3-SPA were up to 12.6-fold greater in patients with AD receiving tramiprosate than in drug-naïve patients.

14. *Clinical Pharmacokinetics and Safety of ALZ-801, a Novel Prodrug of Tramiprosate in Development for the Treatment of Alzheimer's Disease*

Published in October 2017 in *Clinical Pharmacokinetics*

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Actionable Insights: ALZ-801 showed excellent oral safety and tolerability in healthy adults and elderly volunteers, with significantly improved PK characteristics over oral tramiprosate. The clinical dose of ALZ-801 administered 265 mg twice daily, achieves AUC exposure of 150 mg of tramiprosate twice daily, which had previously shown positive cognitive and functional improvements in APOE4 homozygous AD patients. These bridging data support the advancement of ALZ-801 to Phase 3 in early AD patients homozygous for APOE4.

Key Results: (A) We summarize the Phase 1 bridging program to evaluate the safety,

tolerability, and pharmacokinetics of ALZ-801 in healthy volunteers. (B) ALZ-801 is an orally available, valine-conjugated prodrug of tramiprosate with substantially improved pharmacokinetics properties and gastrointestinal tolerability compared with the parent compound. (C) Oral ALZ-801 represents an advanced and markedly improved clinical candidate for the treatment of Alzheimer's disease.

15. *Clinical Effects of Tramiprosate in APOE4/4 Homozygous Patients with Mild Alzheimer's Disease Suggest Disease Modification Potential*

Published 2017 in *Journal of Prevention of Alzheimer's Disease*

Authors: **Susan Abushakra, MD** – Chief Medical Officer of Alzheon, A. Porsteinsson, P. Scheltens, C. Sadowsky, B. Vellas, J. Cummings, S. Gauthier, **John A. Hey, PhD** – Chief Scientific Officer of Alzheon, **Aidan Power**, P. Wang, L. Shen; **Martin Tolar MD, PhD** – Founder, President & CEO of Alzheon

Actionable Insights: The Mild subgroup of APOE4/4 AD patients (MMSE 22-26) showed larger benefits on high dose tramiprosate than the overall mild to moderate group. Consistent with its preclinical effects on Aβ oligomers, tramiprosate stabilized cognitive performance over the 78-week trial duration, supporting its disease modification potential, and had clinically meaningful benefit on both function and disability. Confirmatory studies using ALZ- 801, an improved pro-drug formulation of tramiprosate, will target APOE4/4 patients with Early/Mild AD.

Key Results: (A) In APOE4/4 homozygotes receiving tramiprosate 150mg twice daily, efficacy in Mild AD patients (MMSE 20-26) was higher than the overall group (MMSE 16-26), and efficacy in very mild patients (MMSE 22-26) was highest of all. (B) Tramiprosate benefits compared to placebo on ADAS-cog, CDR-SB, and DAD were 125%, 81% and 71%, respectively ($p<0.02$). (C) The very mild subgroup (MMSE 22-26) showed cognitive stabilization with no decline over 78 weeks, and both ADAS-cog and DAD effects increased over time. (D) Tramiprosate safety in APOE4/4 patients was favorable over 1.5 years, with no events of vasogenic edema. The most common adverse events were gastrointestinal and mild or moderate in severity.

16. *Elucidating the Aβ42 Anti-Aggregation Mechanism of Action of Tramiprosate in Alzheimer's Disease: Integrating Molecular Analytical Methods, Pharmacokinetic and Clinical Data*

Published in October 2017 in *CNS Drugs*

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Actionable Insights: We have identified the molecular mechanism for the observed clinical efficacy of tramiprosate in patients with APOE4/4 homozygous AD. Additionally, we showed the feasibility of modulating and controlling Aβ42 molecular conformation (shape) by a small molecule, causing favorable, clinically relevant inhibition of oligomer formation. This novel enveloping MOA of tramiprosate has potential utility in the development of disease-modifying therapies for AD and other neurodegenerative diseases caused by misfolded proteins.

Key Results: (A) We have elucidated and characterized the molecular mechanism of action of tramiprosate. (B) Tramiprosate modulates conformational flexibility of amyloid beta Aβ42 and, therefore, prevents amyloid oligomer formation. (C) Translational analysis shows alignment of the molecular effects on Aβ42 with pharmacokinetic and published clinical data.

17. *Clinical Benefits of Tramiprosate in Alzheimer's Disease are Associated with Higher Number of APOE4 Alleles: "The APOE4 Gene Dose Effect"*

Published in October 2016 in *Journal of Prevention of Alzheimer's Disease*

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Actionable Insights: The phenomenon of increased tramiprosate efficacy in patients with one or two copies of the APOE4 gene is likely explained by the high prevalence of amyloid pathology in APOE4 carriers, leading to much higher diagnostic accuracy in these patients compared with APOE3/3. Recent studies have shown that to up 38% of APOE3/3 patients may be amyloid negative, while almost all APOE4/4 patients are amyloid positive. In APOE4/4 patients, high dose tramiprosate showed favorable safety and clinically meaningful efficacy when added to standard of care treatment with cholinesterase inhibitors. Prospective confirmation of these promising efficacy findings in APOE4/4 patients with AD, together with the favorable safety profile and convenience of oral ALZ-801, could provide a major

therapeutic advance for this genetically defined, high unmet medical need population.

Key Results: (A) In APOE4/4 subjects, tramiprosate showed dose-dependent and statistically significant effects on ADAS-cog and positive trends on CDR-SB, 40-66% and 25-45% benefit compared to placebo, respectively. (B) Highest efficacy was observed in APOE4/4 homozygotes receiving tramiprosate 150 mg twice daily. (C) APOE4 heterozygotes showed intermediate efficacy, and non-carriers showed no benefit. (D) In 426 patients with serial MRI scans, no cases of treatment-emergent vasogenic edema were observed. (E) Across the three APOE subgroups treated for up to 1.5 years, the most common adverse events were nausea, vomiting, and decreased weight. (F) Oral tramiprosate showed clinically meaningful efficacy in symptomatic APOE4/4 patients with Mild to Moderate AD at a dose with favorable long-term safety, and no observed brain edema.