Highly-Selective Butyrylcholinesterase Inhibitors to Treat Alzheimer's Disease

A Novel Drug Platform to Improve Brain Function in Patients with Neurodegenerative Disease

JAL THERAPEUTICS

February 5, 2024

JAL Therapeutics is a preclinical life sciences company, focused on drug development to treat Alzheimer's disease (AD) and other neurodegenerative conditions

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Our novel, small molecule drug platform of highly selective butyrylcholinesterase (BChE)* inhibitors is intended to treat AD more effectively with fewer side effects than current treatments

*(aka BuChase, BuChE, pseudocholinesterase, serum cholinesterase, plasma cholinesterase)

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AD Cause Theories

i. Cholinergic Hypothesis:

Significantly diminished levels of the neurotransmitter acetylcholine (ACh) and concomitant loss of cognitive function.

ii. Amyloid Cascade Hypothesis:

Enhanced synthesis of amyloid precursor protein (APP), β-amyloid peptides (Aβ42 & Aβ40) protein and subsequent aggregation to amyloid plaques form neurotoxic fibrils, and tau tangles.

- Butyrylcholinesterase (BChE) and acetylcholinesterase (AChE) have both been implicated in the progression of AD. Both enzymes are capable of hydrolyzing ACh and may be responsible for the decrease in ACh and loss of cognitive function.[1]
- AD treatments inhibiting AChE result in side effects, since AChE is required for brain function.[2] In contrast, people without BChE have no life risks and are healthy in normal conditions.[3]
- In AD, there is a nearly 55-fold increase in BChE activity versus AChE, and BChE becomes the dominant enzyme between the two.[4]
- Elevated BChE is directly connected with the pathogenesis of AD (diminished ACh, and increased APP, Aβ42 & Aβ40, β-amyloid plaques and Tau).[5, 6, 7]

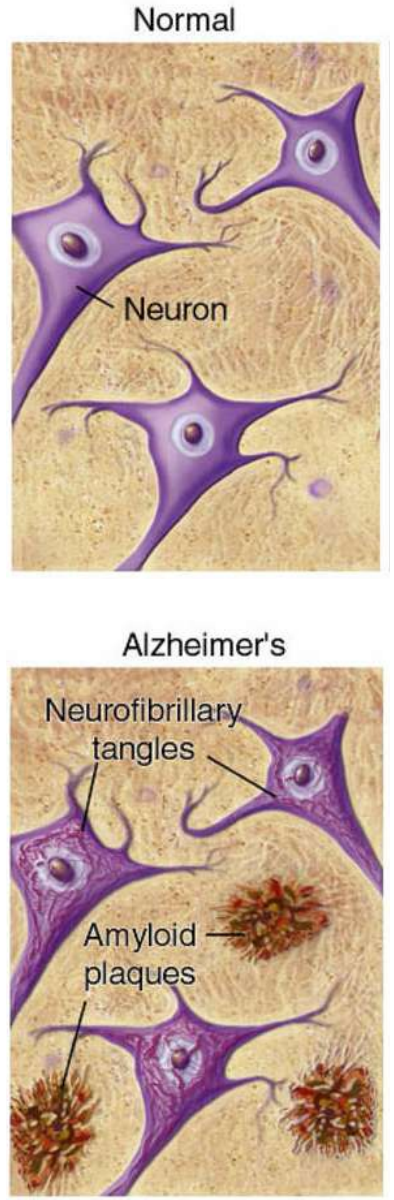
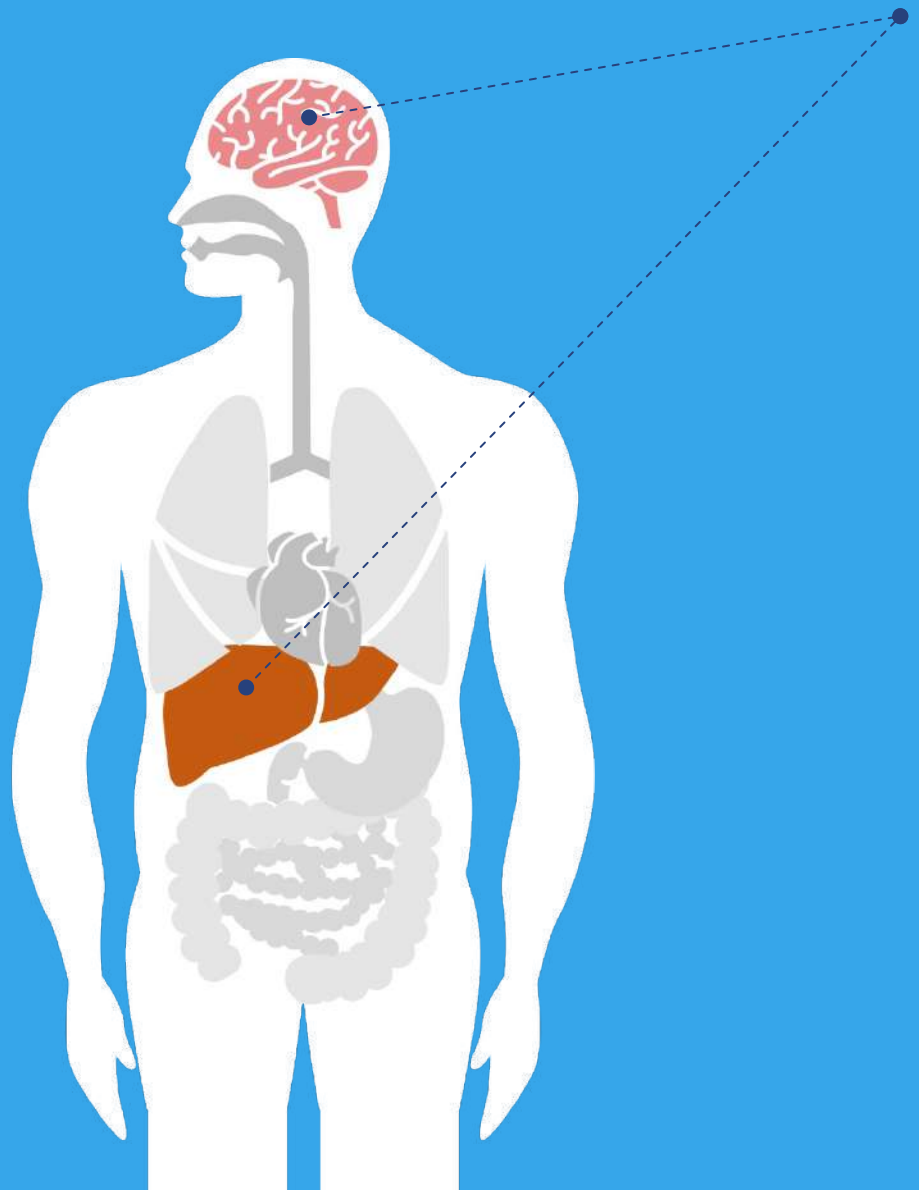


Image source:
BrightFocus Foundation



Elevated BChE is directly connected with Alzheimer's disease progression[5,6,7]

- Our MoA is focused on the highly selective inhibition of BChE, designed to:
- **Restore** acetylcholine (ACh) and cognitive function[5]
- **Decrease** APP, Aβ40 & Aβ42, β-amyloid plaques and Tau[5, 6, 7]

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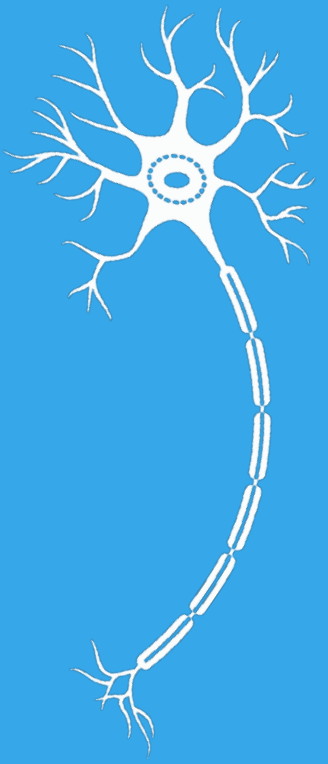
Support for Highly Selective BChE Inhibition as a Viable Therapeutic Approach in AD

- BChE is an attractive target for AD drugs and represents a biomarker for progression of the disease. (2020)[8]
- AChE inhibitors are the mainstay of AD treatments, despite having only short-term symptomatic benefits and severe side effects. Selective BChE inhibitors may be more effective with fewer side effects. (2020)[9]
- Selective BChE inhibition has been regarded as a viable therapeutic approach in AD. However, few highly selective and potent BChE inhibitors have been reported. (2021)[10]
- BChE inhibitors may positively affect not only the cholinergic anti-inflammatory pathway but also other unknown pathways involved in regulating immunity. (2014)[11]



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Lead Drug Candidate Profile: JAL-001

FOR THE TREATMENT OF AD AND BEYOND

Key Biomarker: Butyrylcholinesterase (BChE)

Small molecule drug platform of highly selective BChE inhibitors with lead irreversible (covalent) compound JAL-001.

Highly Selective BChE Inhibitor

68,000-fold selectivity over AChE, essentially nonactive with AChE. Lowered BChE does not cause side effects while lowered AChE does. JAL-001 does not lower AChE.

Promotes Anti-Inflammatory Pathways

MoA designed to activate the cholinergic anti-inflammatory pathway treating both the symptoms and slowing the pathogenesis of AD. Lowered BChE increases acetylcholine, restoring it normal levels, thereby improving brain function.

Prevents AB-Plaque Formation

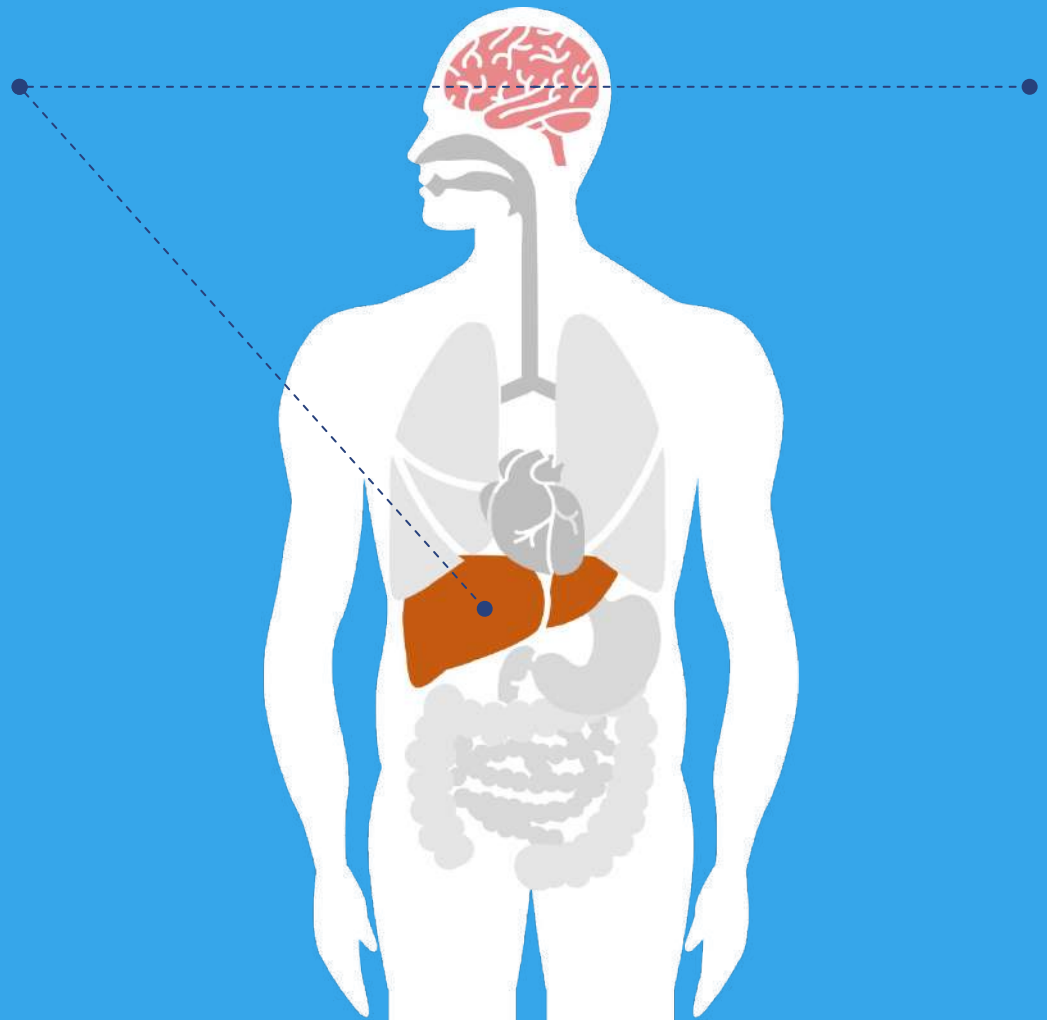
Two successful rounds of animal testing have validated JAL-001 as having high potential for treating AD and other diseases associated with chronic inflammation.

Butyrylcholinesterase & Acetylcholinesterase enzymes are often confused

BChE

Butyrylcholinesterase (BChE) is a known toxin fighter in the body, is not considered essential for basic CNS functions, and can be inhibited without off-target effects

But the industry has overlooked BChE as a target for a new class of cholinesterase inhibiting drugs

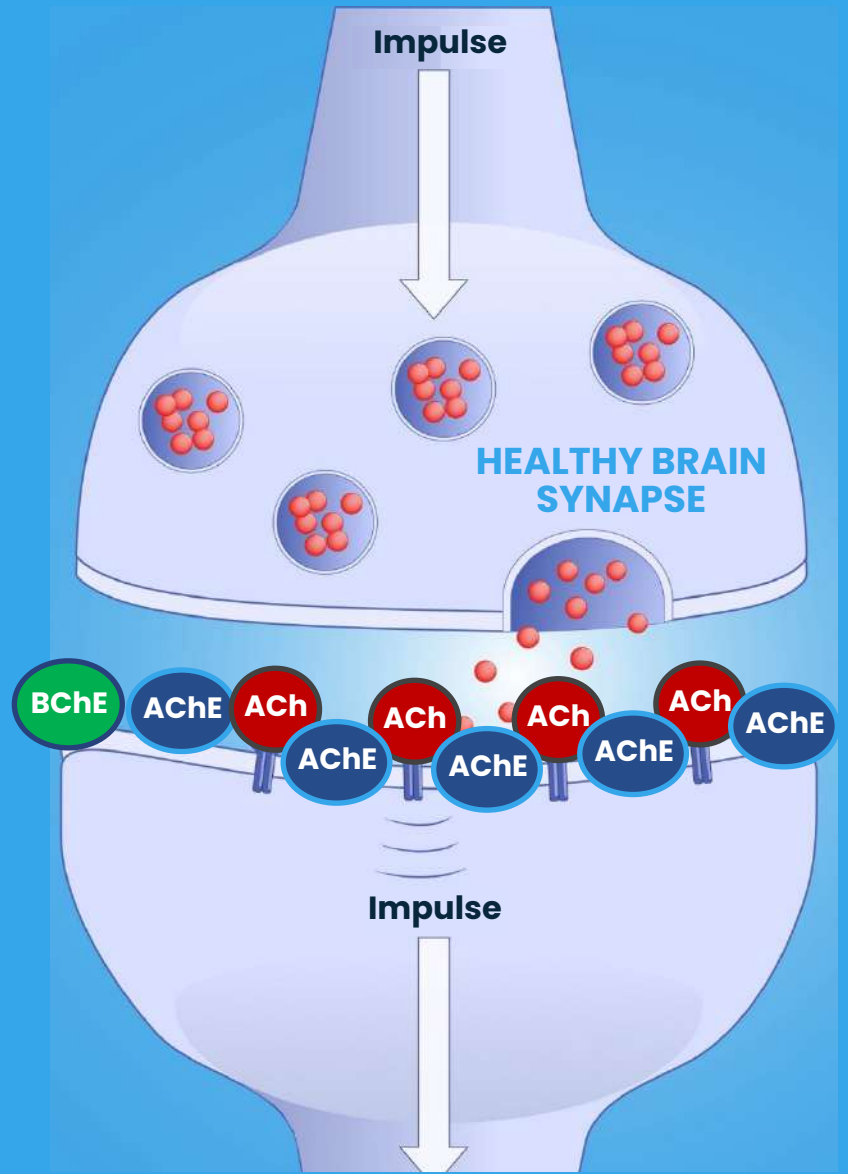


AChE

Acetylcholinesterase (AChE) is crucial to basic CNS and PNS functions, which can lead to cholinergic crisis and even death when inhibited in excess

But other drug companies have focused on the inhibition of AChE in the treatment of AD

The Advantage of Highly Selective BChE Inhibition



Healthy Brain

ACh

Acetylcholine:
The brain's chief neurotransmitter, is hydrolyzed (broken down) at a normal rate

AChE

AChE:
Hydrolyzes acetylcholine resulting in normal neurotransmission

BChE

BChE:
Plays a minor role in acetylcholine hydrolysis, while AChE outnumbers BChE 5:1

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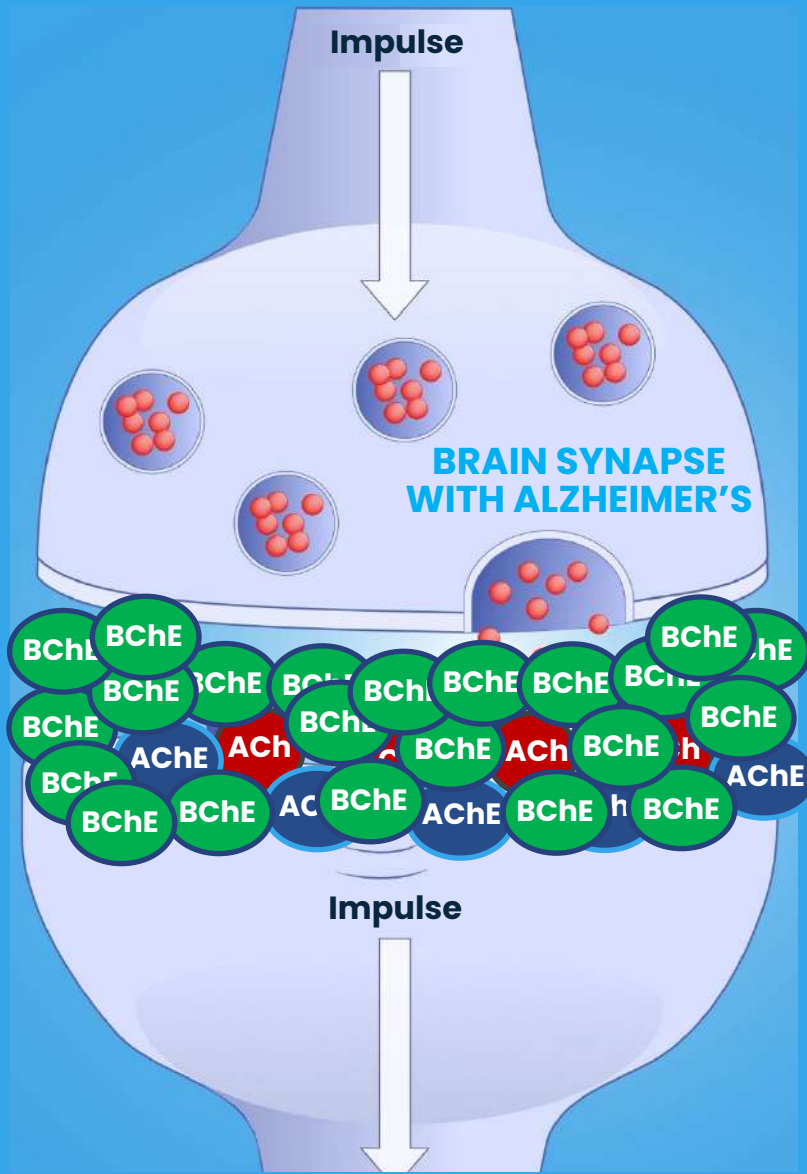
The Advantage of Highly Selective BChE Inhibition



Alzheimer's Brain

ACh — **Acetylcholine:**
Is hydrolyzed at an elevated rate

AChE — **AChE:**
Increases in brains further accelerating acetylcholine hydrolysis

BChE — **BChE:**
Greatly increases 55-fold, outnumbering AChE by 11:1, intensifying acetylcholine hydrolysis

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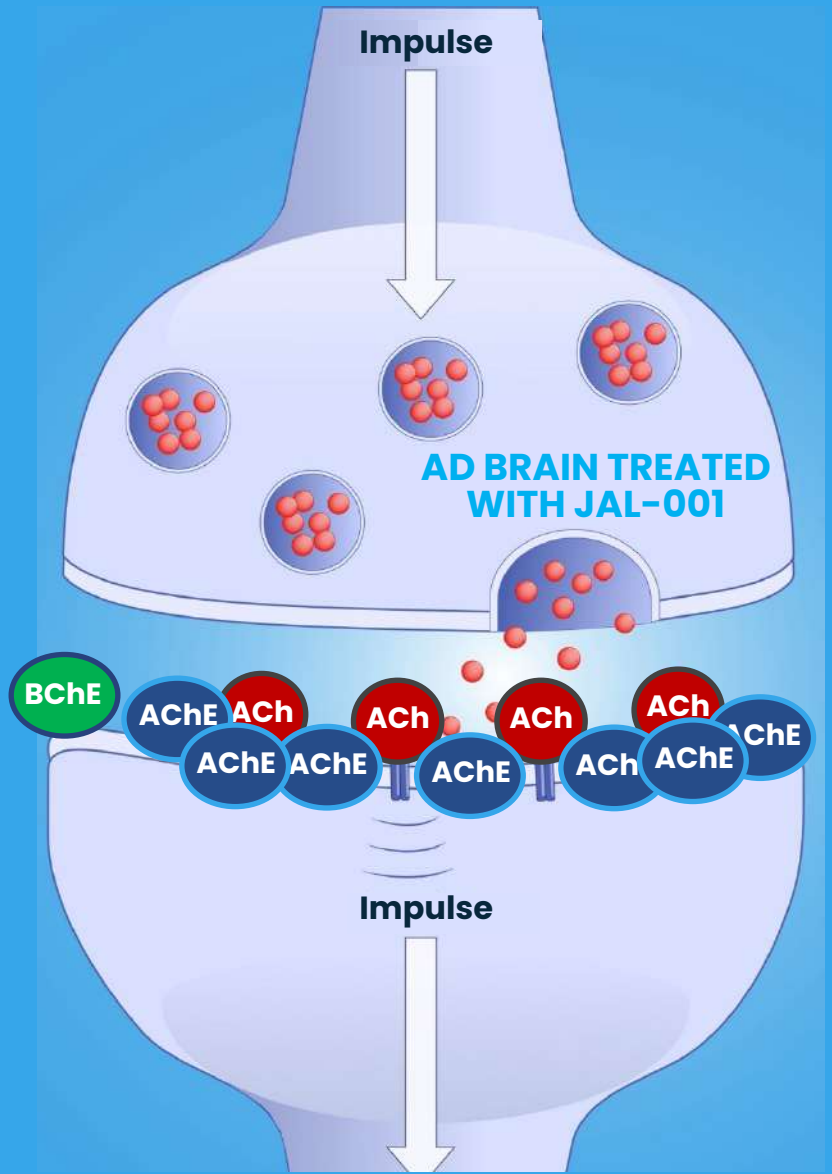
The Advantage of Highly Selective BChE Inhibition



Alzheimer's Brain Treated with JAL-001*

ACh

Acetylcholine:
Hydrolysis is reduced and restored to a normal rate

AChE

AChE:
Is effectively untouched

BChE

BChE:
Is reduced to normal levels, thereby restoring and normalizing acetylcholine levels to improve brain function with no off-target effects

*From modeled data.
Not evaluated by the FDA.

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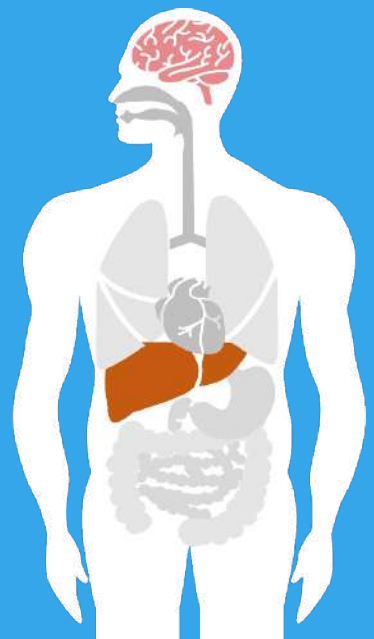
The old-school class of cholinesterase inhibitors currently used to treat AD, inhibit both AChE and BChE as a MoA.

But all have serious side effects and poor efficacy due to low selectivity of BChE over AChE.



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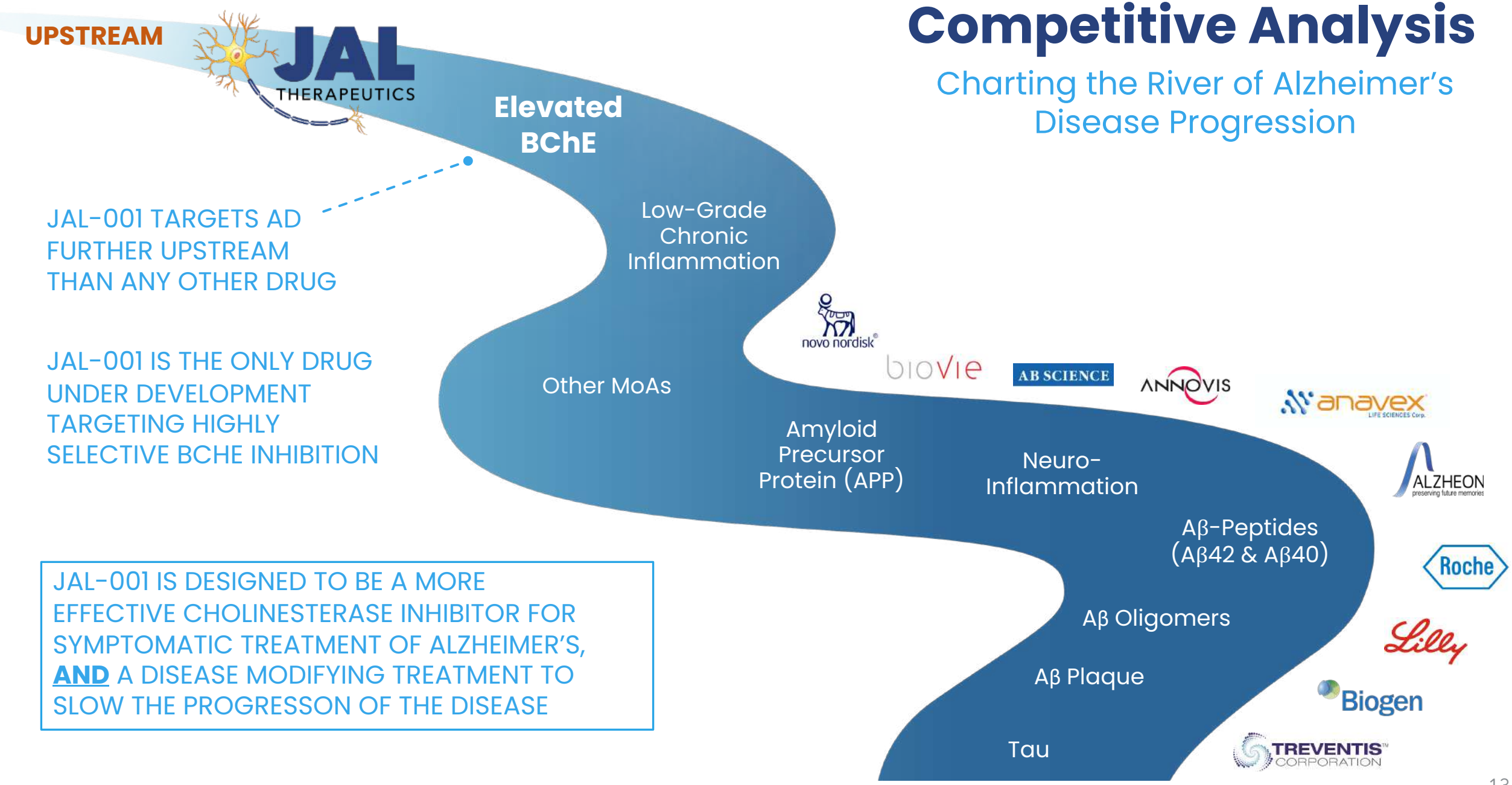
Competitive Analysis

Charting the River of Alzheimer's Disease Progression



JAL-001 TARGETS AD FURTHER UPSTREAM THAN ANY OTHER DRUG

JAL-001 IS THE ONLY DRUG UNDER DEVELOPMENT TARGETING HIGHLY SELECTIVE BCHE INHIBITION

JAL-001 IS DESIGNED TO BE A MORE EFFECTIVE CHOLINESTERASE INHIBITOR FOR SYMPTOMATIC TREATMENT OF ALZHEIMER'S, **AND** A DISEASE MODIFYING TREATMENT TO SLOW THE PROGRESSON OF THE DISEASE

JAL Executive & Advisory Team

Ken Nakayama, PhD
Chief Executive Officer

Company founder, leading international expert on BuChase inhibition, professor of organic chemistry at CSULB

Kevin Sinchak, PhD
Chief Scientific Officer

Preclinical animal testing lead for JAL, professor of neurobiology at Cal State University, Long Beach

M H Khalil, PhD
Pharmaceutical Advisor

Former Principal Scientist, Cardinal Health, Head of CMC, Avanir. Led Nuedexta development (drug for agitation in Alzheimer's), and OTC Abreva

Mark Maricich
Chief Strategy Officer

Med device and life sciences strategic consultant and communications advisor for 20+ years, B+L Surgical, Beckman Coulter and many others.

Charlie Moffett
Business Development

Biz dev specialist with 20+ years of life sciences sales experience. MS in cell and molecular biology, CSULB

David Maricich
Chief Operating Officer

Med device and life sciences strategic consultant and communications advisor for 20+ years, Dutch Ophthalmic (DORC), Vocera and many others

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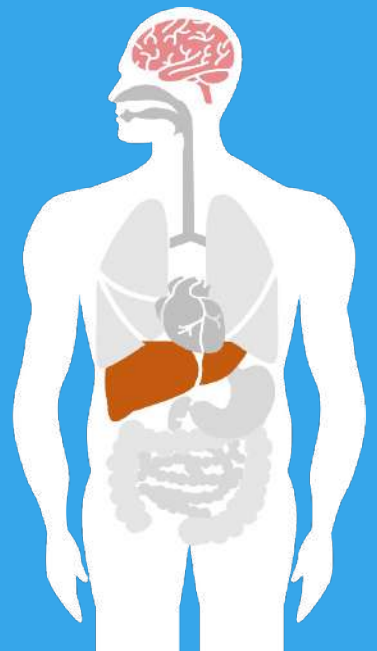
Testing Results & Validation



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JAL-001: Highly Selective BChE Inhibition

High selectivity of JAL-001 is much greater than any other drug

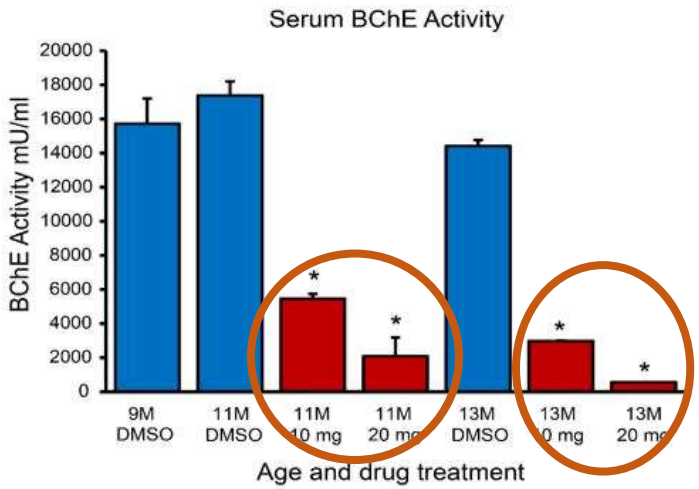
Drug	BChE Inhibition IC_{50} Value (nM)	AChE Inhibition IC_{50} Value (nM)	Selectivity of BChE over AChE
JAL-001 • JAL Therapeutics	**0.69**	**47,000**	**68,000-fold**
FDA Approved Drugs			
Donepezil • (Aricept™, Donepezil hydrochloride, Eranz®, E 2020) • Eisai Co., Ltd., Pfizer	4150	22	---
Galantamine • (Razadyne™, Reminyl™, Nivalin®) • Sanochemia Pharmazeutika, Shire, Takeda Pharmaceutical Company	10,000	1070	---
Rivastigmine • (Exelon™, Rivastigmine tartrate, Rivastach® Patch, Prometax®, SDZ ENA 713) • Novartis Pharmaceuticals Corp	260	3030	12-fold

Source: JAL Data on File + Internet Sourced IC50 Values

Testing Results — Inhibition of BChE: Brain / Serum

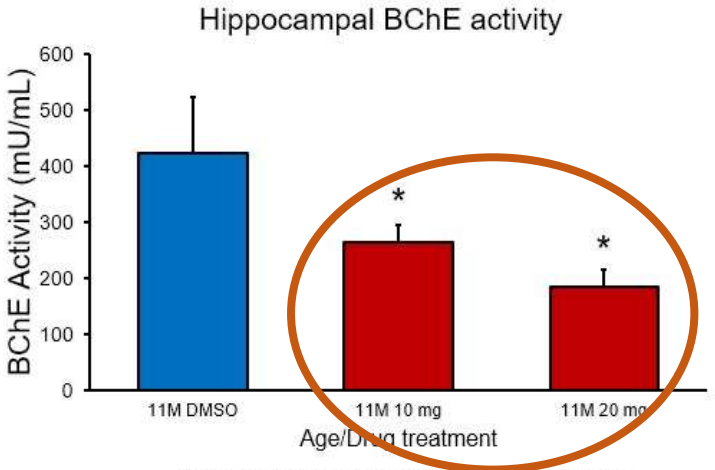
JAL-001 inhibits BChE in brain and serum without inhibiting AChE in serum

Serum BChE activity in mouse model for AD is reduced by treatment with JAL-001



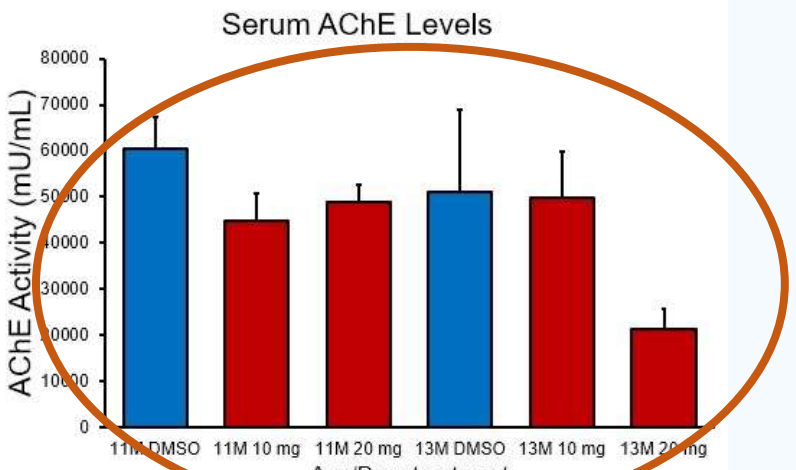
- Significantly lower than all DMSO groups ANOVA, df (6,45) F = 26.1; P < 0.001; Holm-Sidak post hoc P < 0.05

Hippocampal BChE activity in mouse model for AD is reduced by treatment with JAL-001 in 11M treated animals



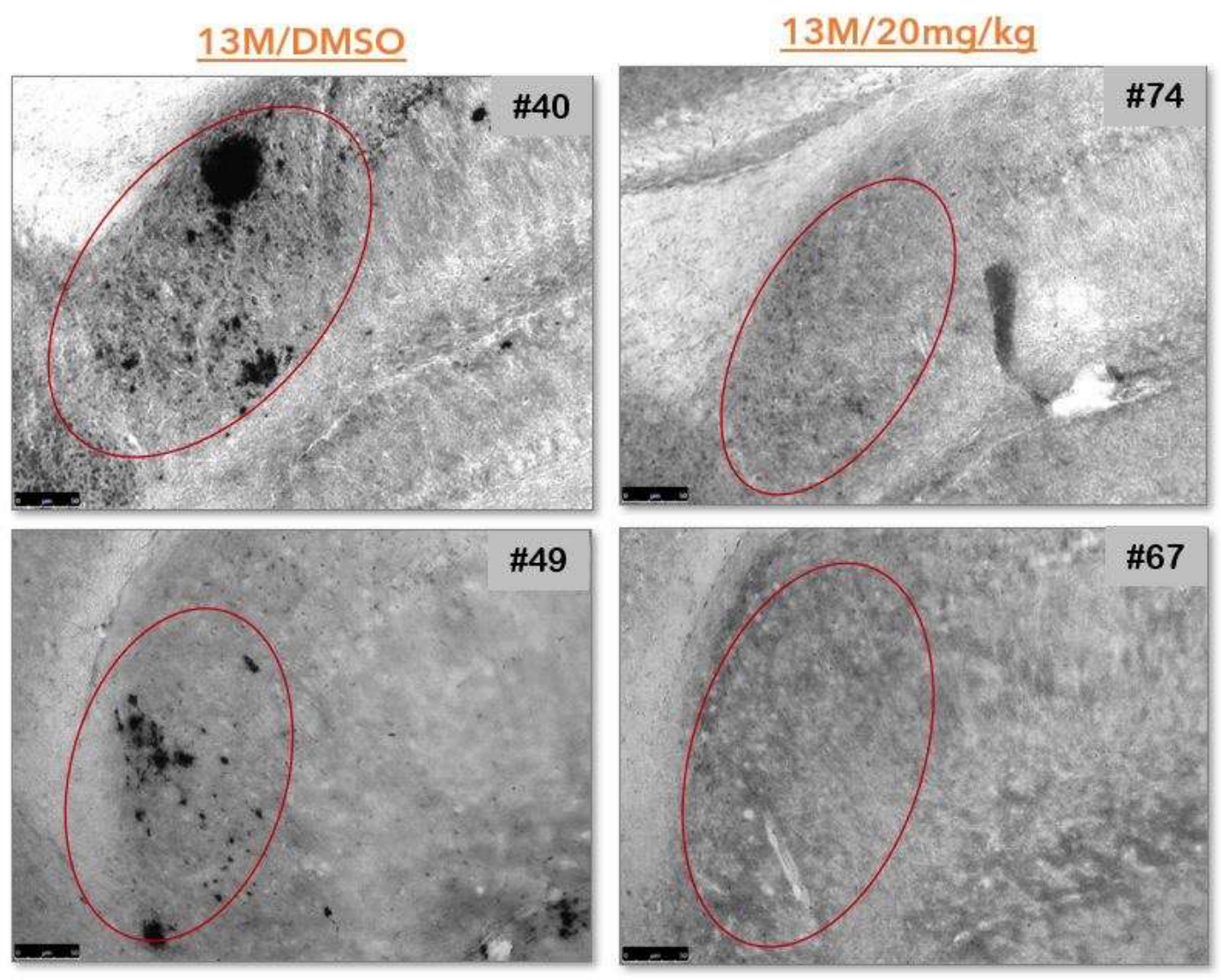
- Significantly lower than 11M DMSO group ANOVA, df (2,24) F = 5.037; P = 0.016; Holm-Sidak post hoc P < 0.05

Serum acetylcholinesterase (AChE) activity in mouse model for AD is not altered by treatment with JAL-001



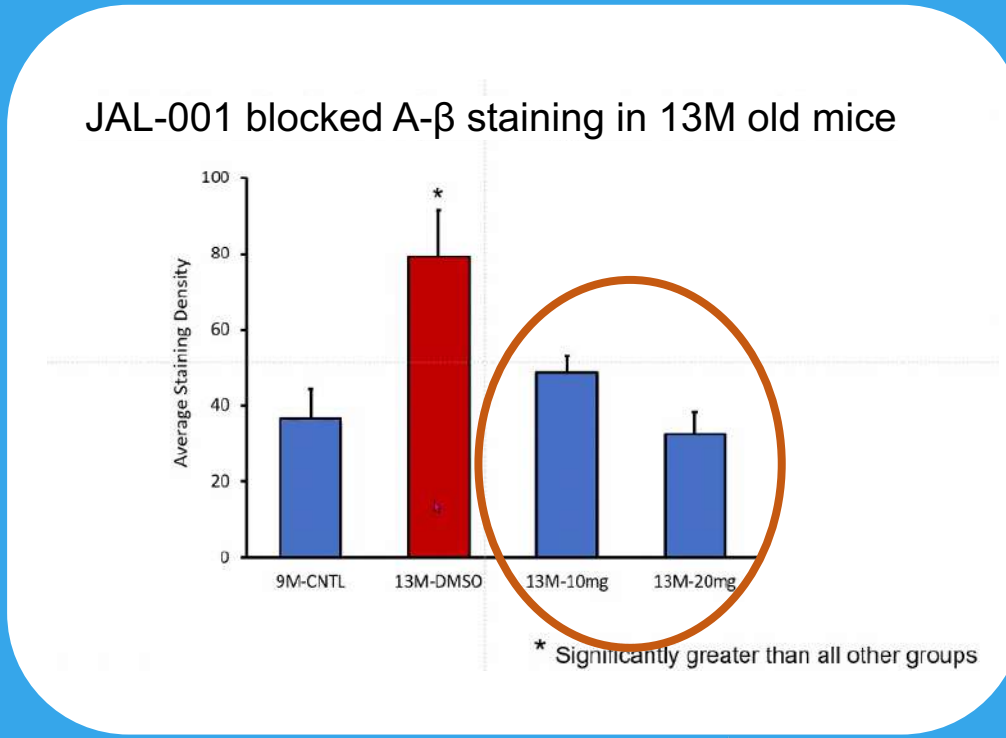
Source: Transgenic AD Mice Studies 2022

Immunohistochemistry Testing



Transgenic mouse models of AD were treated with JAL-001 and the DMSO control from 9 months - 13 months / n=43. Source: Transgenic AD Mice Studies 2022 (currently in progress)

JAL-001 reduced beta amyloid plaques immunopositive staining in the hippocampus compared to untreated mice



* Sig difference from 13M DMSO

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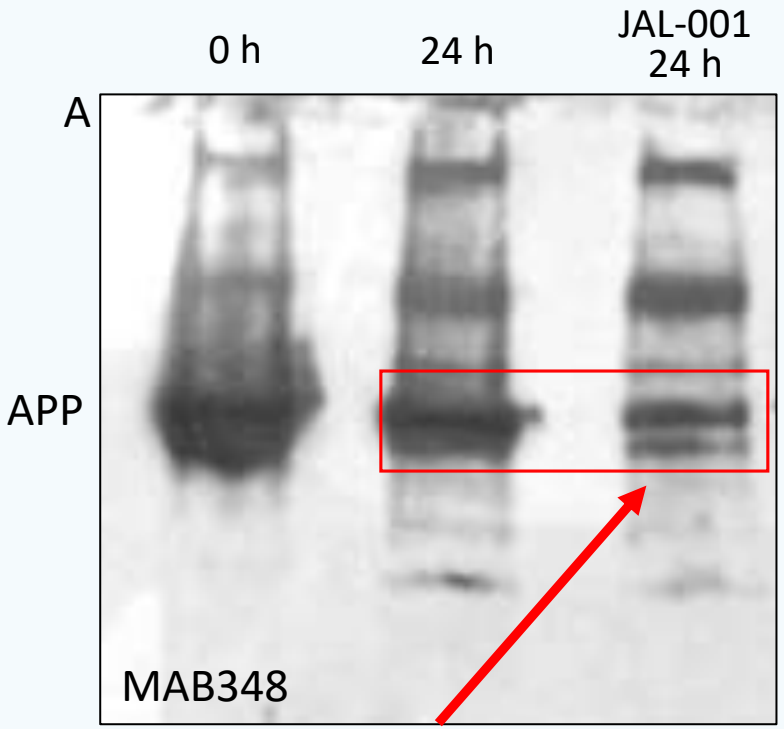
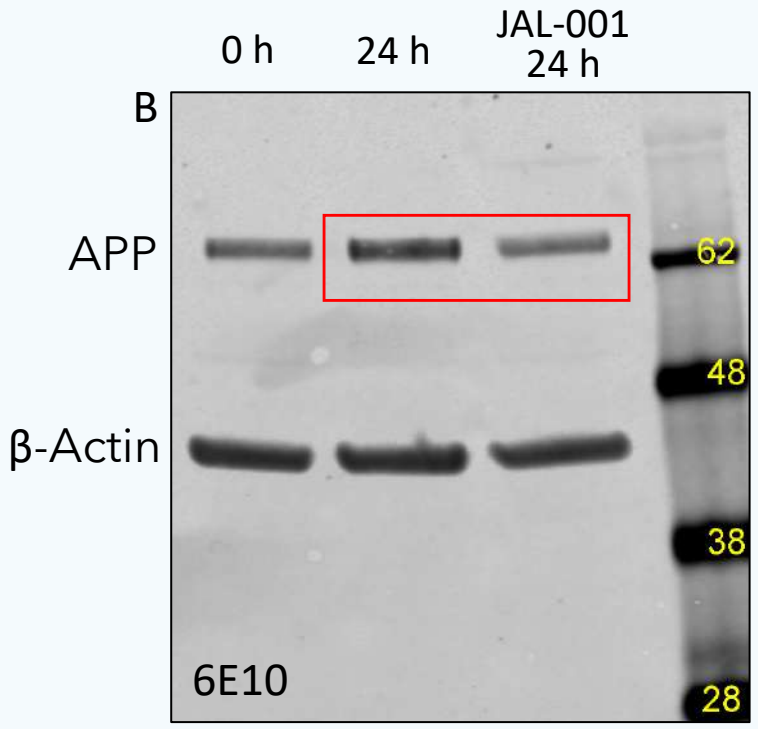
Testing Results — Inhibition of APP Expression

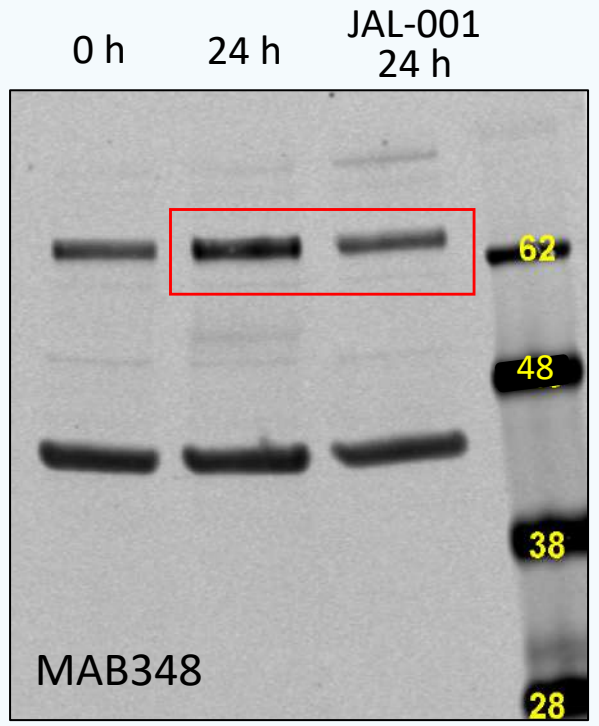
JAL-001 inhibits Amyloid Precursor Protein in human neuroblastoma cells, ex vivo



Reduced APP Expression



37.2% decrease in cells treated with JAL-001



49.4% decrease in cells treated with JAL-001

Tris-HEPES 4-12%, reducing conditions

10% Bis-Tris gel, reducing conditions

Source: JAL Ex Vivo Data

In Vitro Testing Results — β-Amyloid Peptide Inhibition

JAL-001 inhibits the secretion of β-amyloid peptide in vitro

	Aβ-40	Aβ-42
JAL-001	[pg/mg]	[pg/mg]
10^{-8} M	87.3 ± 3.4	56.9 ± 3.1
No Inhibitor	122.5 ± 1.7	88.5 ± 0.2

β-amyloid peptide inhibition appears to be most effective at smaller doses

Source: JAL In Vitro Data

In Vitro Testing Results — Cell Viability

JAL-001 had no effect on cell viability at concentrations as high as 10μM in vitro

Fig. A. Effect of JAL-001 on Cellular Proliferation.
Absorbance values are directly proportional to rates of cell proliferation. Solvent control, 1% Methanol. Results are expressed as the mean of three replicates ±SEM.

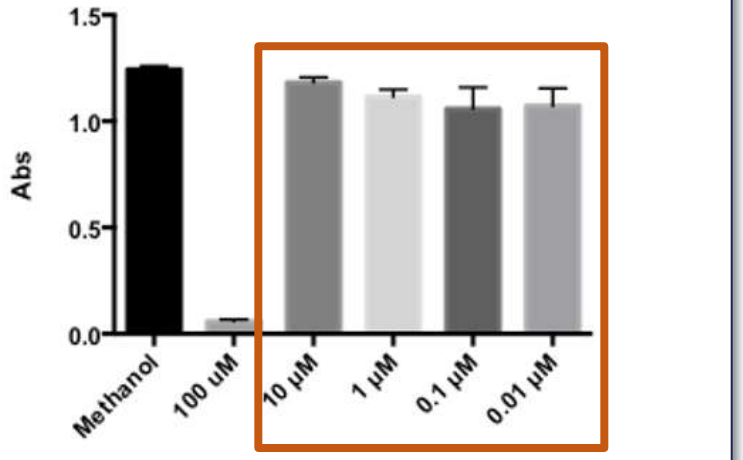


Fig. B. Effect of JAL-001 on Membrane Integrity.
RFU: Relative fluorescent units. Solvent control, 1% Methanol. Results are expressed as the mean of three replicates ±SEM.

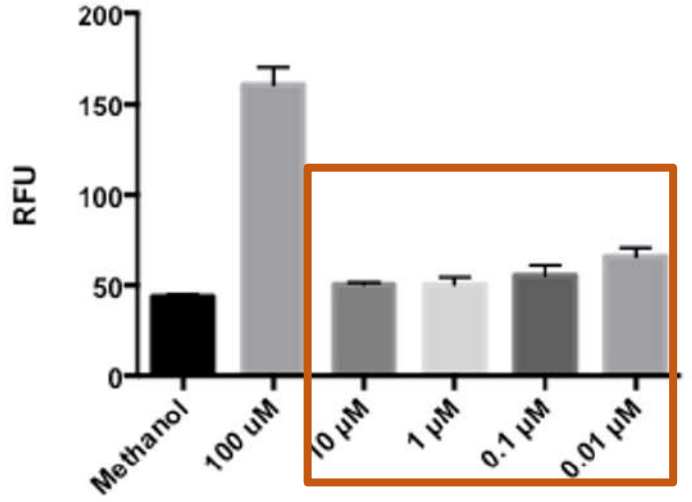
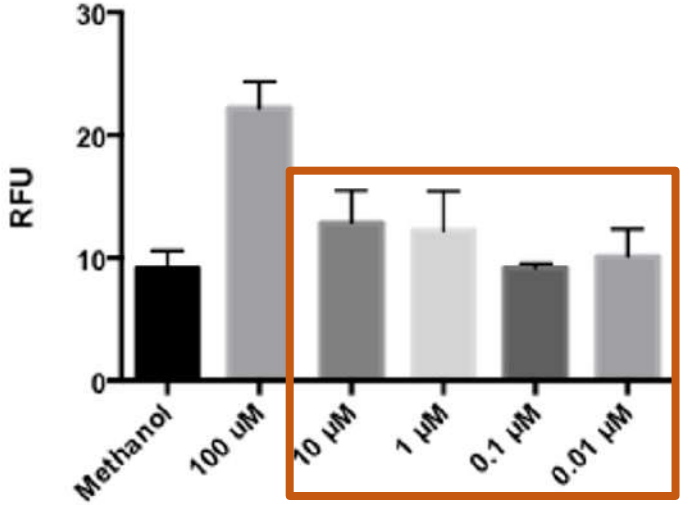


Fig. C. Effect of JAL-001 on Apoptotic Induction.
RFU: Relative fluorescent units. Solvent control, 1% Methanol. Results are expressed as the mean of three replicates ±SEM.



Source: JAL In Vitro Data

In Vitro Testing Results — No Off-Target Effects

JAL-001 has no detectible off-target effects in vitro

	Solvent	I*	JAL-001
Acetylcholinesterase	1.0	0.32	**1.02**
Chymotrypsin	1.0	0.09	**0.98**
Trypsin	1.0	0.49	**0.98**
Hexokinase/G6PDH	1.0	0.71	**0.98**
Protein Kinase A	1.0	0.08	**1.12**

Off Target Inhibitory Activity of JAL-001

Results are expressed as the ratio of enzyme activity of the control (solvent=1.000) to inhibitor treated enzyme. I* Enzyme Specific Inhibitor: Acetylcholinesterase: BW254c; Chymotrypsin: PMSF; Trypsin: TLCK; Hexokinase/G6PDH: Iodoacetamide; Protein Kinase A: PKA-I.

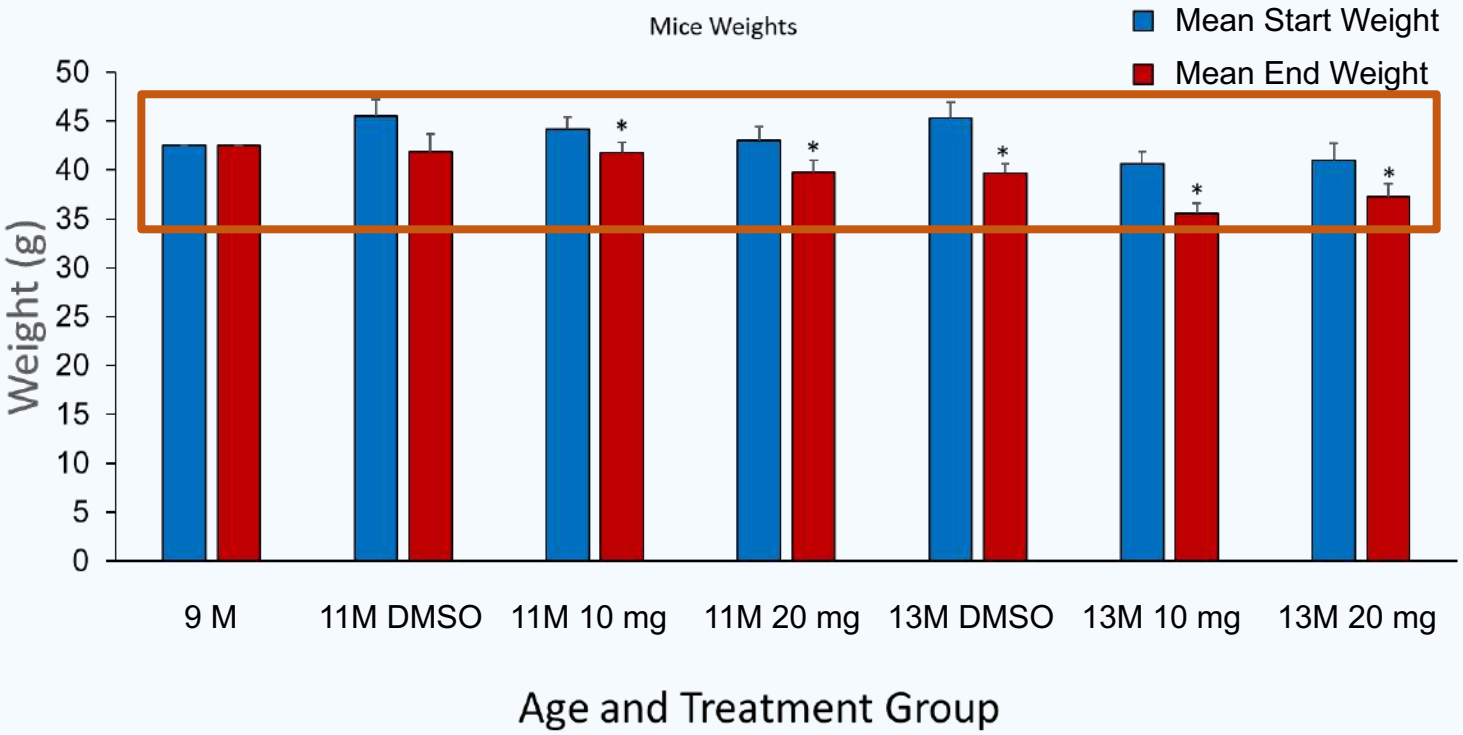
A score of 1.0 = no off-target effects

Source: JAL In Vitro Data

Animal Testing Results — No Off-Target Effects

Standard C57 Lab Mice + Transgenic Mice Model for Alzheimer's

JAL-001 has no detectible off-target effects in animals



- All experimental mice appeared healthy during ongoing visual inspections.
- Mean weights remained consistent among DMSO and JAL-001 mice.
- The first experimental group of standard lab mice (n=53) had zero (0) deaths. These mice received JAL-001 doses of 1, 5, 7.5, 10, 20 and 50mg.
- The second experimental group of transgenic AD mice (n=88) had two (2) deaths at a later stage of 13 months, possibly from stress of handling.*

*Note: These mice had both received 20mg JAL-001 doses, but this was seen as coincidental and not the cause of death. This is since deaths occurred at such a late stage in the experiment (51 & 75 days after initial injections), and because higher 50mg doses had previously been administered with zero (0) deaths.

Source: JAL C57 Mice Studies + Transgenic AD Mice Studies 2022

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JAL-001 Data: In Summary

- ✓ JAL's organophosphorus compounds are highly selective irreversible inhibitors of BChE
- ✓ JAL-001 inhibits the secretion of β-amyloid peptide in vitro
- ✓ JAL-001 crosses the blood brain barrier in vivo
- ✓ JAL-001 significantly and selectively inhibits both brain and serum BChE activity in vivo
- ✓ JAL-001 completely metabolizes within 24-48 hours in vivo
- ✓ JAL-001 has no detectible off-target effects in vivo
- ✓ JAL-001 inhibits amyloid precursor protein (APP) in human neuroblastoma cells, ex vivo
- ✓ JAL-001 inhibits β-amyloid plaque in vivo

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JAL Product Pipeline

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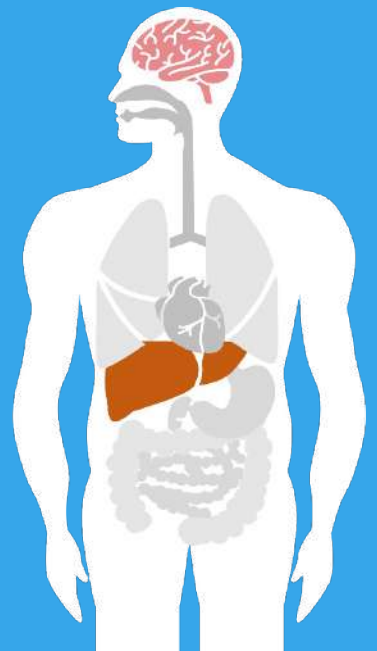
Market Analysis



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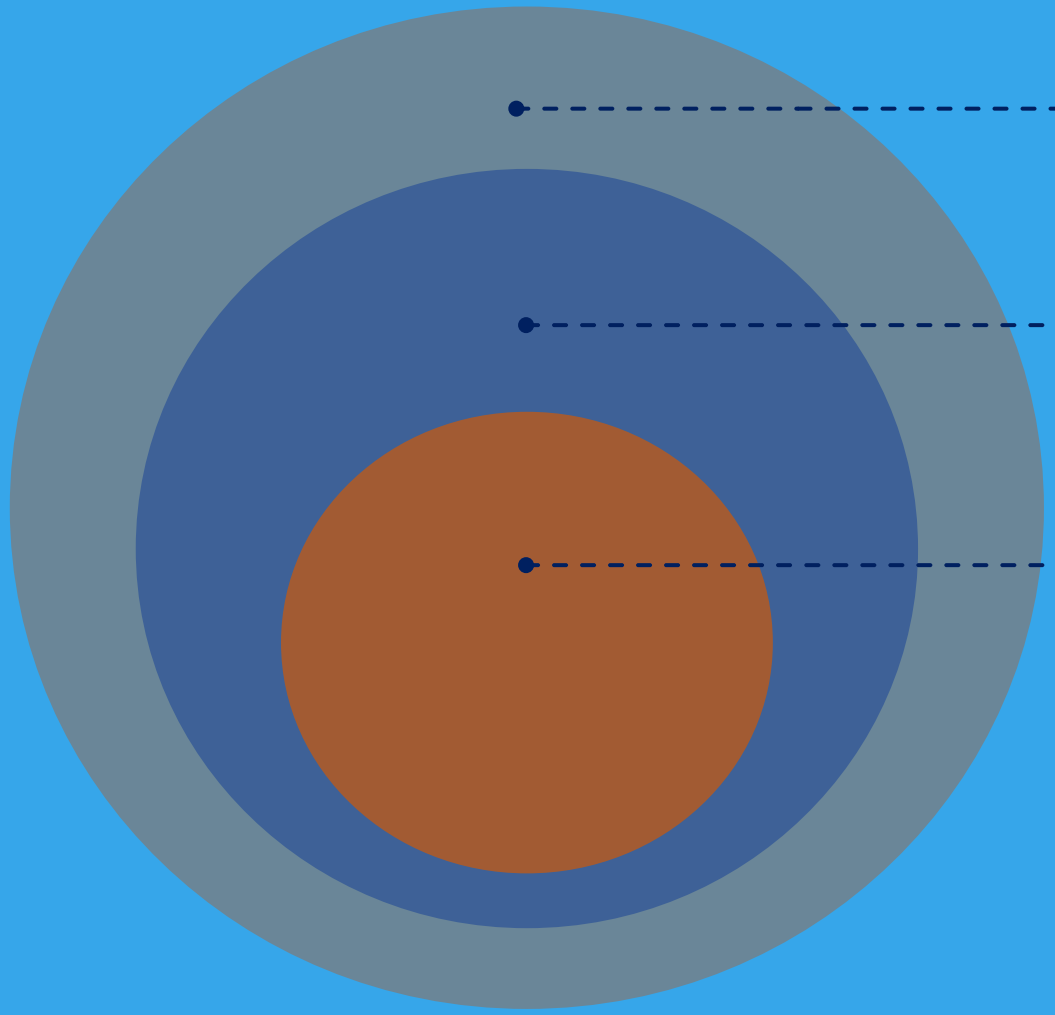
JAL Market Opportunity – Replacing Old AChE Inhibitors with a Novel Platform to Treat AD: Highly Selective BChE Inhibitors



TOTAL AVAILABLE MARKET: $4B

SERVICABLE AVAILABLE MARKET: $3B

SERVICABLE OBTAINABLE MARKET: $2 Billion

The low efficacy of the current acetylcholinesterase inhibiting drugs used for AD justifies large market share potential for JAL-001 if approved

Source: Grand View Market Research, 2023

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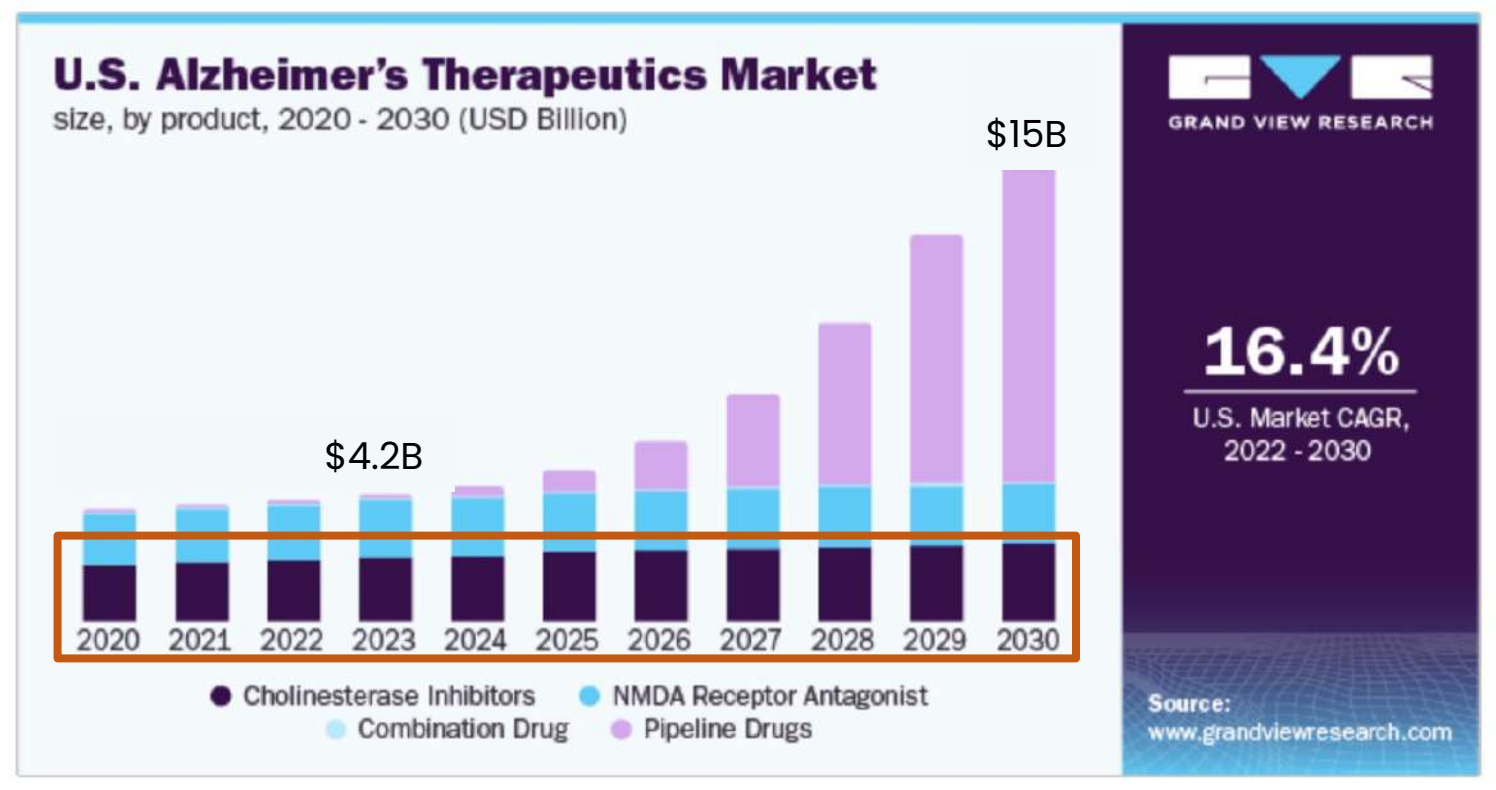
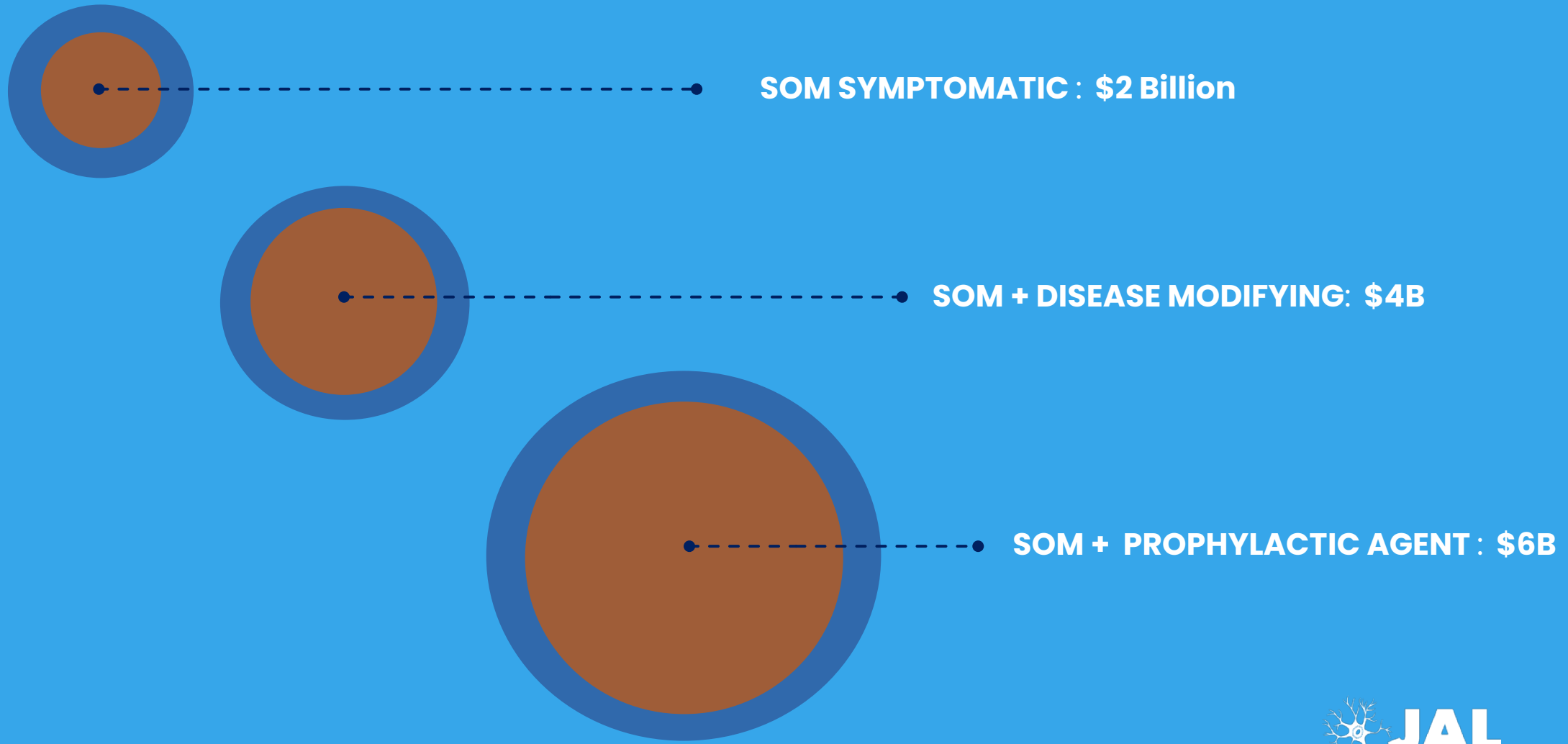
JAL Market Opportunity – A New Class of Cholinesterase Inhibiting Drugs for AD



Source: Grand View Market Research, 2023

Cholinesterase inhibitors are projected to be a steady part of AD treatment protocols for years to come and represent 51% of all AD drugs currently prescribed.

JAL Market Opportunity – Multi Action Strategy Increases Long-Term Market Size Considerably



Source: Grand View Market Research, 2023

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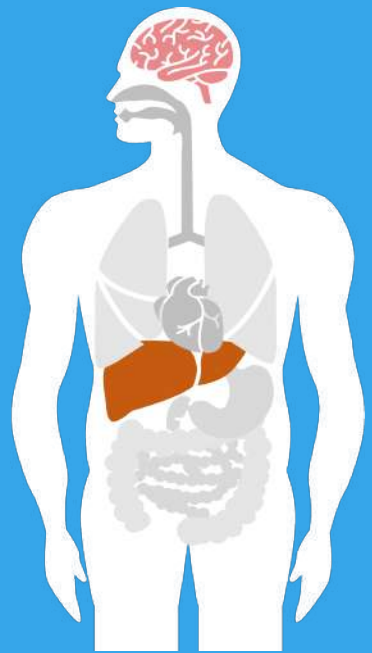
Market Comparison

Financial Reference Based on Recent Comps & Deal Sizes

Company	Market Cap / Deal size	Stage
ANNOVIS	$100M	Phase III
AB SCIENCE	$175M	Phase III
TREVENTIS CORPORATION	$372M	Preclinical
syndesi therapeutics	$870M	Phase I
KARUNA THERAPEUTICS	$14B	Phase III



Sources:

i. Nature; "Trends in Neuroscience Dealmaking" (2018); https://www.nature.com/articles/d43747-020-00598-z

ii. Nature; "High-value dealmaking strengthens big pharma's CNS ambitions" (2022); https://www.nature.com/articles/d43747-022-00178-3

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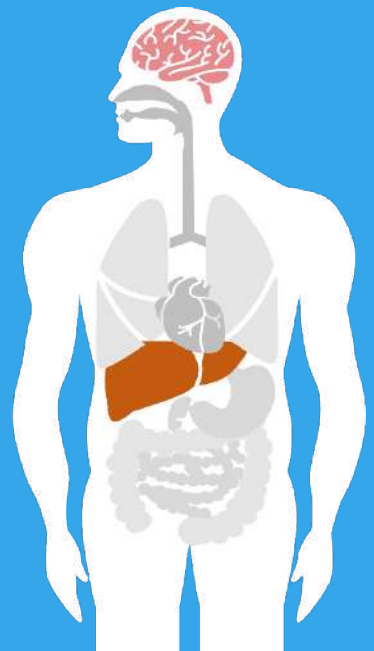
Average discovery stage pharma licensing deals for Alzheimer's and CNS drugs $65M upfront / $1.5B total. [i, ii]

Company History, Timelines & Milestones



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JAL History & Progress

Preclinical Phase II (2024 – 2026)

(De-risking) Safety and tolerability, PK/ADME, PD and MTD studies for JAL-001, Pre-IND studies, patent nationalization, VC and pharma licensing outreach, IND application for human clinical trials.

Strategic Platform (2021 – 2023)

Patent expansion – PCT filing (platform of 98 molecules), indication expansion, application for $2.5M NIH grant, CA Life Sciences FAST member, Academic Venture Exchange listing initial pharma & VC engagement.

Preclinical Phase I (2017 – 2021)

Utility patent issued for JAL-001 for AD. Taconic transgenic mouse model studies (immunohistochemistry, serum & brain).

Discovery (2005 – 2016)

Identification of highly selective BChE inhibiting molecules, *in vitro* assays: β-amyloid peptide secretion. *Ex vivo* assays: inhibition of amyloid precursor protein (APP) in human neuroblastoma cells, provisional patent application (2007). First mouse models testing conducted.

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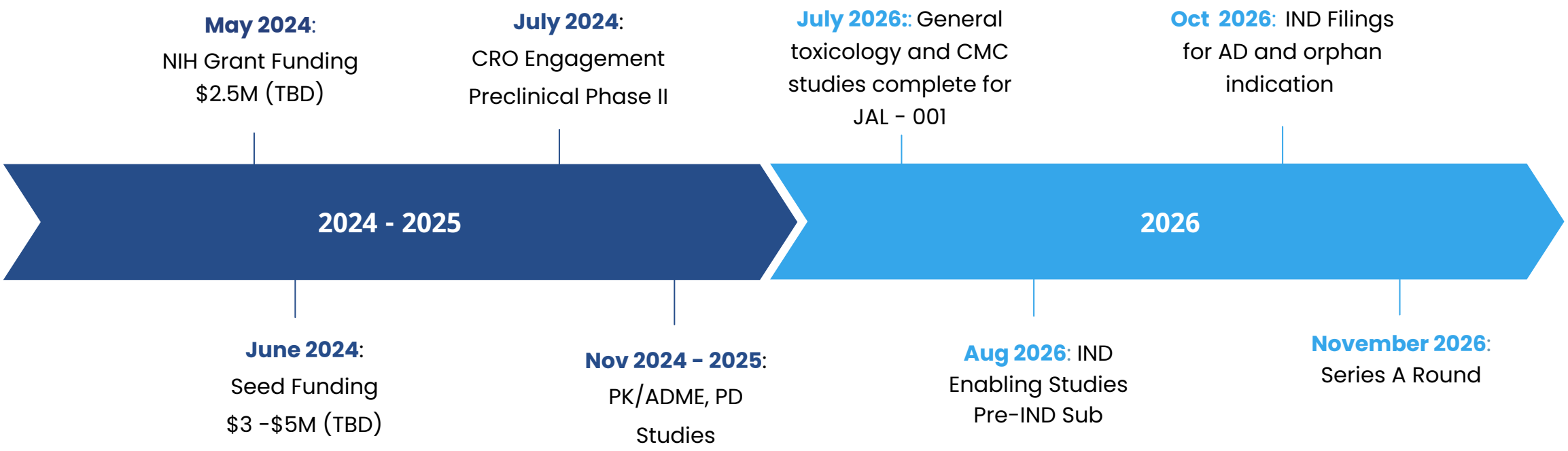
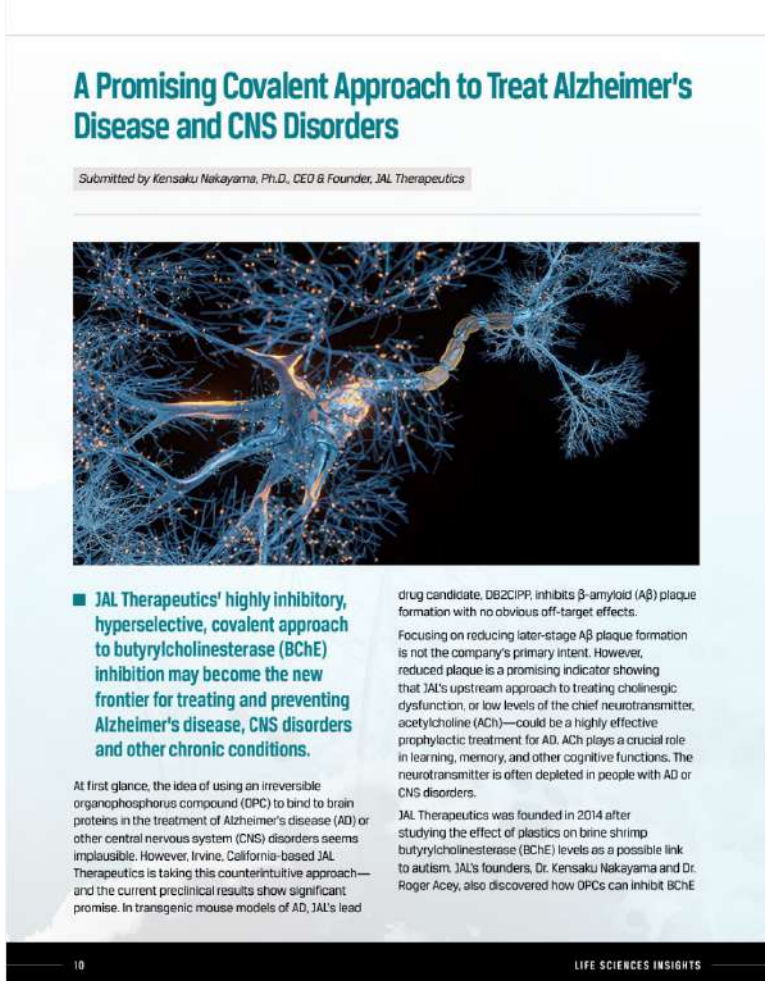
Timeline & Milestones – Path to IND

Seeking Seed Funding of $3 - $5M



Press & Affiliations



A Promising Covalent Approach to Treat Alzheimer's Disease and CNS Disorders

Submitted by Kensaku Nakayama, Ph.D., CEO & Founder, JAL Therapeutics

JAL Therapeutics' highly inhibitory, hyperselective, covalent approach to butyrylcholinesterase (BChE) inhibition may become the new frontier for treating and preventing Alzheimer's disease, CNS disorders and other chronic conditions.

At first glance, the idea of using an irreversible organophosphorus compound (OPC) to bind to brain proteins in the treatment of Alzheimer's disease (AD) or other central nervous system (CNS) disorders seems implausible. However, Irvine, California-based JAL Therapeutics is taking this counterintuitive approach—and the current preclinical results show significant promise. In transgenic mouse models of AD, JAL's lead drug candidate, DB2CIPP, inhibits β-amyloid (Aβ) plaque formation with no obvious off-target effects.

Focusing on reducing later-stage Aβ plaque formation is not the company's primary intent. However, reduced plaque is a promising indicator showing that JAL's upstream approach to treating cholinergic dysfunction, or low levels of the chief neurotransmitter, acetylcholine (ACh)—could be a highly effective prophylactic treatment for AD. ACh plays a crucial role in learning, memory, and other cognitive functions. The neurotransmitter is often depleted in people with AD or CNS disorders.

JAL Therapeutics was founded in 2014 after studying the effect of plastics on brine shrimp butyrylcholinesterase (BChE) levels as a possible link to autism. JAL's founders, Dr. Kensaku Nakayama and Dr. Roger Acey, also discovered how OPCs can inhibit BChE

CALIFORNIA LIFE SCIENCES

Academic Venture eXchange

JAL Therapeutics April 2023 Feature in [California Life Sciences Magazine]

AVX Partner Listing - Feb 2024 [Academic Venture Exchange]

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Conclusion

The New World of BChE as a Therapeutic Target

JAL has reached a crucial inflection point in our journey, with much opportunity for continued acceleration of our progress.
Thank you for your consideration!

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