The Return Leg

Nine accounts of the failing synapse in Alzheimer's disease, the one lesion they describe, and what they become when they are joined

Estela Area-Gómez · Barbara K. Brott · Gunnar Gouras · Brian P. Head · Zhen Huang · Seth S. Margolis · Christopher E. Ramsden · Ari Rappoport · Carla J. Shatz · Scott A. Small

Benjamin Aaron Gustafsson

AdultCognitiveDisease.com

August 2026

Nine laboratories have spent between two and three decades each on the question of why synapses fail in Alzheimer's disease. They studied different molecules in different compartments by different methods, and they do not, on the whole, cite one another. Each of them found a cycle, and each of them found the disease on its return leg — in the step that recovers, releases, clears, disposes, resolves or restabilises, and not in the step that makes.

Contents

Abstract

Synapse loss remains the strongest structural correlate of cognitive decline in Alzheimer's disease — better than plaque burden, better than tangle burden, and established as such for more than three decades. It follows that any account of the disease must eventually say what happens, mechanically, at the synapse. Nine research programmes have spent between two and three decades each doing exactly that, and they have arrived at nine apparently different answers.

The lipid raft fails to assemble because the astrocyte cannot deliver cholesterol to the neuron. Reactive aldehydes generated by lipid peroxidation weld apolipoprotein E to its receptor so the receptor cannot be recovered. The β-carboxy-terminal fragment of the amyloid precursor protein is never cleared from the endoplasmic reticulum, so a cholesterol correction runs for fifty years. Retromer-dependent recycling jams, and everything held in the endosome dwells there too long. Amyloid-β is retained rather than exported from the synaptic endosome, and the coupling between activity and clearance breaks. Caveolin-1 no longer organises the membrane patch on which trophic signals are received, so the neuron is deaf rather than starved. The proteasome in the dendritic plasma membrane declines with age and with apolipoprotein E genotype, so newly made tau is not trimmed. An inhibitory immune receptor is engaged by a complement fragment and by amyloid, and holds the spine's actin cytoskeleton in a state of disassembly. And a competition among axons for survival, which amyloid-β exists to adjudicate, is never resolved because the peptide that would resolve it has aggregated.

This paper sets those nine programmes out in their own terms, and then argues that they are describing one lesion from nine positions. Each of the nine found a *cycle* — a physiological loop with a forward leg that makes, delivers, cuts, engages or destabilises, and a return leg that recovers, releases, clears, disposes, resolves or restabilises. In every case the reported lesion is on the return leg. In every case the forward leg is intact, and in several it is measurably increased, because the cell is compensating for a return that never arrives. The name given here to that shared structure is **the failure of return**, and its most consequential property is that it inverts the sign of the obvious therapy: suppressing production has repeatedly done harm, and restoring recovery has scarcely been attempted.

Three further arguments follow. First, the nine compartments are fewer than nine: the lipid raft, the mitochondria-associated endoplasmic reticulum membrane, and the synaptic endosome are the same kind of object — a cholesterol-ordered, detergent-resistant domain — measured at three addresses, which is why programmes with no shared vocabulary keep finding one another's molecules. Second, the nine mechanisms run on two clocks, and the division is sharp: the amyloid-gated mechanisms execute, while the lipid- and traffic-gated mechanisms, which carry all of the human genetic anchors, set the substrate on which execution occurs. Third, and most usefully, the programmes can be joined at named residues and named compartments, and the joins generate experiments nobody has run. Three of the nine write to serine 3 of cofilin, from

three different receptors, with opposite signs, and no two of those literatures cite each other. The neuronal membrane proteasome has an apolipoprotein-E-ordered surface abundance that falls with age, which is the signature of a raft protein, and nobody has asked whether it is one. The covalent receptor block that one programme proposes and the endosomal de-acidification that another has demonstrated produce the same phenotype and are distinguishable in a single experiment.

What the integration does not do is dissolve the disagreements. Three separate sign problems remain live — whether apolipoprotein E4 delivers too little cholesterol to the membrane or too much, whether neuronal caveolin-1 rises or falls in the human disease, and whether reduced lipoprotein-receptor engagement is the lesion or the protection — and they are probably one problem, created by measuring in bulk tissue a quantity that is defined per cell type and per compartment. Two direct empirical contradictions are recorded without adjudication because the adjudicating studies have not been done.

The paper closes with a ranked programme of fourteen experiments — eleven of them crosses between programmes that neither has an incentive to run and both already have the reagents for — a graded ledger of every claim it makes, and the conditions under which the integration would be wrong.

Part One — The Question

Why nine programmes, what kind of claim each is making, and how to join theories without flattening them.

1. Introduction: Nine Laboratories and One Synapse

1.1 The observation that will not go away

In 1990 DeKosky and Scheff counted synapses in frontal cortex biopsies from living patients with Alzheimer's disease and found that the count correlated with the severity of cognitive impairment (DeKosky and Scheff, 1990). The following year Terry and colleagues reported the same relationship in a larger post-mortem series and stated the conclusion the field has never been able to discard: synapse loss is the major structural correlate of cognitive impairment, and it outperforms both of the lesions by which the disease is defined at autopsy (Terry et al., 1991). Scheff and colleagues later showed that the loss is already present in mild cognitive impairment (Scheff et al., 2007), and positron-emission tomography with a synaptic-vesicle ligand has since made the measurement in living people, where it is widespread and early (Mecca et al., 2020).

Selkoe's summary of this literature — that Alzheimer's disease is a synaptic failure (Selkoe, 2002) — is one of the most-cited sentences in the field, and one of the least acted upon. The apparatus built over the following two decades was an apparatus for measuring and removing amyloid. The synapse remained the thing that the disease damages and the thing whose damage explains the symptom, and it remained, mechanistically, comparatively under-specified.

It is not under-specified for want of work. It is under-specified because the work has been done in nine or ten separate places, in nine or ten separate vocabularies, and has not been assembled.

1.2 The nine programmes

This paper takes as its object nine research programmes, each of which has been sustained for at least two decades, each of which has generated a coherent mechanistic account of synaptic failure in this disease, and each of which is usually read alone.

Table 1 — The nine programmes, by the compartment each principally concerns

Programme	Principal investigator(s)	Central object	Compartment
Lipid rafts as the gate of plasticity	Ari Rappoport	Astrocyte-derived cholesterol; the raft as a switch	Plasma membrane; astrocyte–neuron transfer

Programme	Principal investigator(s)	Central object	Compartment
Lipid peroxidation and receptor crosslinking	Christopher E. Ramsden	Peroxidised apolipoprotein E; ApoER2–Dab1	Ligand–receptor interface; early endosome
The uncleared lipid signal	Estela Area-Gómez	C99; mitochondria-associated ER membrane	ER–mitochondrial contact
Retromer-dependent recycling	Scott A. Small	VPS35, VPS26b, SORLA	Early endosome, recycling arm
Intraneuronal amyloid retention	Gunnar Gouras	Retained Aβ42; multivesicular bodies	Synaptic endosome
The membrane platform	Brian P. Head	Caveolin-1; the raft as a signalling organiser	Neuronal plasma membrane
Surface proteolysis and the spine brake	Seth S. Margolis	Neuronal membrane proteasome; Ephexin5	Dendritic plasma membrane; spine
The system of restraint	Carla J. Shatz, Barbara K. Brott	MHC class I, PirB/LilrB2, C4d	Synaptic surface; postsynaptic actin
Competitive synaptic plasticity	Zhen Huang	Amyloid-β as a competition signal	Axon terminal; microglial interface

The selection principle is stated so that the reader can disagree with it. These are programmes whose object of study is the synapse itself, or the membrane and compartment system that maintains it. Programmes that are principally about tau propagation, about vascular contribution, about the autophagy–lysosome axis, or about microglial state transitions are cited throughout — extensively, because they are frequently the source of the adjudicating evidence — but they are not among the nine, because their central claim is about a different object. The choice is defensible and it is not the only defensible one.

Two of the nine, Area-Gómez and Head, do not describe themselves as working on synapses at all. Area-Gómez works on a lipid-regulatory contact between two organelles; Head works on a scaffolding protein first characterised in endothelium. They are included because the argument of Part Three is that the compartments they study and the compartments the other seven study are the same kind of object, and because their evidence turns out to be load-bearing for claims the others make and cannot support alone.

1.3 What is wrong with reading them one at a time

Reading a research programme in isolation is the normal practice of the field and it produces three predictable distortions.

It converts convergence into competition. A mechanism that would be uncontroversial as a description of a step everyone's disease must pass through becomes, when stated alone, a rival origin story. Several of the nine have suffered from this specifically. The intraneuronal-amyloid account is the clearest case: its evidence is almost entirely interventional — block the mechanism and the synaptic phenotype disappears, drive it and the phenotype appears — which is evidence about necessity and sufficiency, not about temporal priority. Stated as an account of where the disease begins, it becomes something the evidence cannot support and something that invites dismissal.

It hides the redundancy. If four upstream arms converge on one effector, a monotherapy directed at any one arm will underperform, and the underperformance will be read as a failure of the hypothesis rather than as a property of the architecture. Part Four shows that at least three of the nine converge on a single phosphorylation site.

It duplicates work. Two of the nine independently identified the cysteine content of apolipoprotein E as the pivot of its allelic risk, eight months apart, and assigned the same molecular feature two different jobs. Three arrived at the amyloid precursor protein's carboxy-terminal fragment as the pathogenic species, from three different compartments, without a shared citation. This is not a criticism of anyone; it is a description of what happens when a field's literature grows faster than any one person can read it.

1.4 What this paper claims

Three things, in ascending order of exposure to refutation.

First, a descriptive claim. The nine programmes can be arranged along a single circuit — the itinerary of a lipid and a protein through the compartments of one neuron — and each occupies a distinct station on it. This is the argument of Chapter 13, and it is nearly a matter of bookkeeping.

Second, a structural claim. At every station, the reported lesion is on the return leg of a cycle, not on the forward leg. Recovery, release, clearance, disposal, resolution and restabilisation fail; synthesis, delivery, cleavage and engagement do not. This is the argument of Chapter 14, it is the paper's central thesis, and Chapter 15 sets out honestly the four places where it fits badly.

Third, a set of integrative claims. Named residues and named compartments join the programmes in ways that generate experiments. Part Four states seven such joins, grades each, and specifies for each what result would count against it.

The paper does not claim to have found the cause of Alzheimer's disease, does not adjudicate between the theories of onset that the nine programmes variously imply, and does not propose that the nine be merged into one. Chapters 25 to 27 are given over to the disagreements that survive the integration, and they are real.

2. What Kind of Claim Each Programme Is Making

2.1 Three kinds of claim, adjudicated three ways

Theories in this field are routinely compared as though they were all answers to the same question. They are not, and the confusion is expensive, because claims of different kinds are settled by different evidence.

An **initiation claim** asserts temporal priority: this happens first, and the rest follows from it. Its evidence must be temporal, and for a disease with a twenty-year preclinical phase it must ultimately come from human tissue sampled at stages that are largely inaccessible. Almost no initiation claim in this field can be settled, which is a sufficient explanation for why so many coexist.

A **convergence claim** asserts necessity: whatever happens first, it must pass through here to produce this outcome. Its evidence is interventional — block the mechanism and the outcome disappears, drive the mechanism and the outcome appears — and it can be obtained in a laboratory. Convergence claims are *strengthened* by a plurality of upstream causes, because every additional cause that funnels into the same mechanism is a further demonstration that the mechanism is the common path.

A **function claim** asserts what a molecule is for in health, and derives the disease as a corollary. Its evidence is loss-of-function genetics and physiology in the normal animal. Function claims are unusual in this field and unusually durable, because they are tested in systems that are not models of anything.

Table 2 — The kind of claim each programme's evidence actually supports

Programme	Claim as usually stated	Kind of claim the evidence supports	Adjudicated by
Rappoport	Initiation: raft assembly failure is the cause of sporadic disease	Convergence, with an initiation claim attached that the evidence does not carry	Interventional raft manipulation; direct raft imaging

Programme	Claim as usually stated	Kind of claim the evidence supports	Adjudicated by
Ramsden	Initiation: aldehyde crosslinking is the first chemical event	Convergence for the ApoER2–Dab1 lesion; the initiation claim rests on a species not yet isolated from human tissue	Mass-spectrometric identification of the crosslinked species in human brain
Area-Gómez	Initiation: uncleared C99 is the primary lesion	Convergence, with a strong signed prediction across disorders	Directional manipulation of contact-site function
Small	"Causal and common"	*Causal* is close to established; *common* is unsupported by the first direct human assay	Pathway-function biomarkers in living patients
Gouras	Initiation: the inside-out amyloid hypothesis	Convergence throughout; the initiation claim has been tested once and was not supported	Knock-in models; a proxy for the intraneuronal pool
Head	Therapeutic: restore the platform	Convergence, and separately a demonstration of engineered resilience	Dose–response against raft occupancy
Margolis	Two mechanisms: a developmental brake re-applied; a surface protease	The brake arm is an executor; the protease arm is a candidate initiator	Human tissue measurement across age and genotype
Shatz and Brott	Development's machinery, reused	Convergence, with the newer (complement) arm better evidenced than the older (amyloid) arm	Ligand-specific blockade in human tissue
Huang	Initiation: failure of a competition programme	Function claim, well supported; the disease model is a corollary	Loss-of-function genetics, already largely done

Read down the third column and a pattern appears that is worth stating baldly: **eight of the nine present their work as an account of how the disease begins, and in eight of nine the evidence is of the convergence or function type.** The programmes are, almost uniformly, better than the claims made on their behalf — but better at a different job.

This is not a rhetorical observation. It determines what the integration in Part Three can be. Nine initiation claims cannot be integrated; they can only compete, and the competition cannot be settled. Nine convergence claims *can* be integrated, because convergence claims about different steps of one pathway are additive by construction.

2.2 The temptation of the origin story

It is worth asking why a field pulls every account toward initiation.

The proximate reason is that the amyloid cascade hypothesis is an initiation claim, was stated as one, and was restated as one at its twenty-fifth anniversary (Hardy and Higgins, 1992; Selkoe and Hardy, 2016). For thirty years it has been the object against which other accounts define themselves, and defining oneself against an initiation claim produces an initiation claim.

The deeper reason is that initiation claims are what a therapeutic programme appears to need. If you know what starts the disease, you know what to prevent. The inference is intuitive and it is wrong in a specific way: a convergence claim also licenses an intervention, and licenses it for a broader population, because the intervention does not require knowing which upstream cause a given patient has. Small and Petsko made this argument explicitly in the hub-and-spoke model — repairing the hub should benefit patients whose primary lesion is in any spoke, including spokes the model does not name (Small and Petsko, 2020). It is the strongest single argument in the therapeutic literature reviewed here, and it generalises well beyond the programme that made it.

2.3 A note on what "mechanism" means at the synapse

One further clarification, because the nine programmes use the word at four different resolutions.

At the coarsest, mechanism means a named compartment and a named direction of traffic — the endosome jams, the contact expands. At the next, a named molecular species and its fate — C99 accumulates, Aβ42 is retained, apolipoprotein E is crosslinked. At the next, a named signalling chain with an effector — ApoER2 to Dab1 to PI3-kinase to LIM-kinase; LilrB2 to phosphatase to cofilin. At the finest, a named residue — serine 3 of cofilin, tyrosine 361 of Ephexin5, threonine 508 of LIM-kinase 1, tyrosine 682 of the β-carboxy-terminal fragment.

Integration is only possible at the finer resolutions. Two theories that both say "the endosome fails" have not been joined; they have been paraphrased. Two theories that both write to serine 3 of cofilin have been joined, and the join predicts an experiment. Part Four is deliberately restricted to joins at the third and fourth resolutions, and the rest of the paper is arranged to make those joins visible.

3. Method: How to Join Theories Without Flattening Them

3.1 The failure mode of synthesis

The characteristic failure of a synthetic paper is that it produces a diagram in which every arrow is true and nothing is false. Such a diagram cannot be wrong, and therefore contains no information. Any set of mechanisms in a common tissue can be drawn as a network; the drawing is not an argument.

Four rules were adopted to avoid this, and the reader should hold the paper to them.

Rule one: no join without a shared object. Two programmes are said to be joined only if they name the same molecule, the same residue, the same compartment or the same measurement. "Both concern lipid" is not a join. "Both write to serine 3 of n-cofilin" is.

Rule two: every join must generate a discriminating experiment. A join that predicts nothing new is a restatement. Each of the seven joins in Part Four is accompanied by the experiment that would refute it, and where the experiment has in fact been done, the result is reported even when it is unhelpful.

Rule three: contradictions are preserved, not averaged. Where two programmes disagree about the sign of an effect, the disagreement is stated, the evidence on each side is set out, and the adjudicating measurement is named. Part Five does nothing else. Averaging a sign disagreement produces a claim that neither party holds and that no experiment can test.

Rule four: the strength of each claim is graded, and the grade travels with the claim. A ledger appears in Chapter 31 giving, for every substantive claim in the paper, the strongest supporting evidence, a grade, and what is missing. Claims made for the first time here are labelled as such and graded conservatively.

3.2 Grades used

Four grades are used throughout, and they are ordinary rather than technical.

- **Established** — demonstrated by intervention in both directions, or by human genetics with a causal architecture, and independently replicated.

- **Strong** — demonstrated by intervention, or by convergent evidence from methodologically independent lines, but from a single laboratory or without reciprocal manipulation.
- **Moderate** — supported by correlation in relevant tissue, or by intervention in one direction only, or by a single well-designed study.
- **Contested** — directly opposed by a comparable result that has not been adjudicated.
- **Inference** — follows from established premises but has not been tested as such. Every claim original to this paper begins here.

The grades are applied to individual propositions, not to programmes. Every one of the nine contains propositions at several grades, and the practice of grading a body of work as a whole is one of the reasons this literature is hard to use.

3.3 What counts as evidence about a human being

A methodological commitment that runs through the paper and should be visible.

Almost all of the mechanistic evidence reviewed here comes from mice, from cultured neurons, from human induced pluripotent stem cells, or from purified protein. That evidence is what establishes necessity and sufficiency, and there is no substitute for it. But it does not establish that a mechanism operates in a human being, and this field has repeatedly mistaken the first for the second.

Three classes of evidence do bear on human beings directly, and they are weighted more heavily here wherever they exist.

Human genetics with a causal architecture. Rare truncating variants in *SORL1* occurring almost exclusively in cases (Andersen, de Waal et al., 2025) is a causal statement about humans. So is the ordering of *APOE* allelic risk (Corder et al., 1993), the distinct genetic form represented by *APOE4* homozygosity (Fortea et al., 2024), and the two documented instances of resistance to autosomal-dominant disease conferred by single variants in ligands of the apolipoprotein E receptors (Arboleda-Velasquez et al., 2019; Lopera et al., 2023).

Human tissue measured with the compartment preserved. Most human neuropathology reports what is present, not where. Where a study resolves a compartment — the subcellular fractionation showing that oxidative damage in Alzheimer brain is concentrated in lipid rafts rather than distributed across the membrane (Thorwald et al., 2025), or the array tomography that places a complement fragment at identified excitatory synapses in human cortex (Brott et al., 2025) — it carries more weight here than a larger study that does not.

Trials. A trial is a directional experiment in humans, and the field has run several that discriminate between the accounts assembled here. Chapter 28 reads the trial record as evidence

rather than as disappointment.

3.4 On citing programmes rather than papers

The nine programmes are cited here to their primary literature throughout. Where a programme's own review or summary states a position more strongly than its data support — which is common, and is not a criticism specific to anyone — the primary result is cited and the difference is noted. Where a claim rests on a preprint or a conference abstract, that status is stated in the text and not only in the reference list. Five of the results that this paper leans on are preprints as of August 2026, and the argument would look thinner without them; they are flagged at each use.

Part Two — The Nine Stations

Each programme in its own terms: what it claims, what carries the claim, where it is weak, and what it contributes that no other programme supplies.

4. Supply: Astrocyte Cholesterol and the Gate of Plasticity

4.1 A theory of a transition, not of a substance

Ari Rappoport's account of Alzheimer's disease is downstream of a theory of normal plasticity, and cannot be evaluated without it. He died in June 2025, so the theory is fixed in final form, with a documentary record of its own revisions between the 2020 statement, an intermediate version, and the account published in the *Annual Review of Biochemistry* (Rappoport, 2025).

The plasticity theory begins from an engineering problem rather than an observation. When an animal executes a response with a novel component and survives, the pathways involved should be made more available in future. But the brain does not know which of the many synapses, boutons and branch points active during that response were the ones that mattered. It cannot simply strengthen the correct elements, because it has not identified them.

The proposed solution is the one that evolution and adaptive immunity use: generate a surplus of candidates and then select among them. Plasticity therefore runs in two stages. **Candidate generation** destabilises the existing arrangement and produces an excess of potential modification sites — new spines, new boutons, new branch points, existing synapses marked for potentiation. Destabilisation is required rather than incidental: a stable structure cannot generate new geometry. Matrix metalloproteinase-9 digests extracellular matrix and adhesion molecules; tau is phosphorylated and inactivated, releasing the cross-links that hold the existing cytoskeleton together; calcium-permeable receptors are inserted so that activity produces large calcium influx. **Competition resolution** then enhances winners, eliminates losers, and restabilises: calcium influx is brought back down in both populations, calcium-permeable AMPA receptors are exchanged for calcium-impermeable ones, GluN2B-containing NMDA receptors switch to GluN2A, winners acquire a stable actin cytoskeleton cross-linked to microtubules by dephosphorylated tau, and losers are retracted.

None of the components is novel and Rappoport does not claim otherwise. What is novel is the assertion that these constitute one canonical two-stage process with a defined transition between them — and the identification of what throws the switch.

4.2 The gate is a physical object

The theory's distinctive claim is that the transition from generation to resolution is not triggered by a timer, a counter or a threshold on activity. It is triggered by the **assembly of plasma-membrane lipid rafts** — ordered nanodomains enriched in cholesterol, sphingomyelin and glycolipids, through which the membrane connects to the cytoskeleton, to the extracellular matrix, and to the synaptic partner across the cleft.

The reasoning is that resolution requires capabilities only rafts provide. A winning spine cannot be stabilised without anchoring receptors and scaffolds in position, and stable membrane anchoring of the relevant proteins — postsynaptic density proteins, glutamate receptors and their recycling machinery, G-protein-coupled receptors, actin regulators — depends on palmitoylation, which targets proteins to rafts. Microtubule-associated proteins, tau among them, associate with rafts. Neurotrophin receptors and the insulin receptor are anchored and trafficked through caveolin-1-containing rafts. The neuron cannot begin to consolidate anything until it has built the platform on which consolidation is physically performed.

Then the load-bearing step. The rate-limiting ingredient in that platform is cholesterol, and neurons do not make their own in usable quantity. Brain cholesterol does not cross the blood–brain barrier; it is synthesised by astrocytes, packaged into apolipoprotein E particles, released, and taken up by neurons through the low-density-lipoprotein receptor and LRP1. Rappoport's argument for why this specific dependency is the vulnerable one is physical rather than statistical, and it is the best paragraph in the theory: unlike glutamine or lactate, cholesterol has a large hydrophobic domain, so it cannot diffuse or ride a small transporter. Its delivery requires a lipoprotein particle, a receptor, an endosome and a route to the plasma membrane — heavy machinery, with no redundancy of the relevant kind, because neuronal cholesterol synthesis, which does exist and does rise under stress, cannot substitute for delivered cholesterol in building plasma-membrane rafts.

The claim, then, is that the switch between the two stages of memory formation is thrown by the arrival of astrocyte-derived cholesterol at the neuronal plasma membrane, and that this is a design feature: cholesterol arrival signals that an astrocyte process is close enough to supply the new synapse, and therefore that consolidating a synapse at this location is worth doing. The switch is a supply check.

4.3 The disease as chronic non-resolution

If raft assembly is impaired but not abolished, the neuron neither completes plasticity nor abandons it. Both stages run simultaneously, at lower agent concentrations than either would use in health, and for far longer. That is a chronic state, and three consequences follow.

The staging of agent changes is predicted: early in disease both generation and resolution agents should be elevated, and later, as resources deplete, resolution should rise while generation falls.

Rappoport reads the literature as showing exactly this — the tau-phosphorylating kinases chronically increased, protein phosphatase 2A decreased, and, tellingly, protein kinase C and GluA2, which he identifies as the specific hallmarks of successful winner enhancement, *reduced*, with the largest GluA2 decrease in the most vulnerable hippocampal fields.

Second, and this is the theory's best move, chronicity inverts the sign of the neuron's own compensation. Many plasticity agents are double-edged — a single agent produces opposite effects depending on its concentration, because it engages a high-affinity receptor at low concentration and a low-affinity receptor in addition at high concentration. Calcium is the canonical instance: high or fast calcium activates calcium/calmodulin-dependent kinase II and marks winners, low calcium activates calcineurin and eliminates losers. Chronicity means lower concentrations, so chronic signalling is tilted toward the elimination edge. The neuron's attempt to compensate — continued candidate generation, which would normally drive cholesterol synthesis and rescue the situation — instead produces degeneration. **The compensation is the pathology, and a partial failure is worse than a complete one.**

Third, chronic signalling desensitises receptors and pathways through ordinary negative feedback, so signalling degrades further and the lesion deepens itself.

4.4 The physiological jobs assigned to the pathological molecules

The theory's most durable contribution, in this reading, is that it gives each of the field's pathological molecules a job in the healthy brain, and the jobs are specific.

- **The amyloid precursor protein manages cholesterol during plasticity.** Its two cleavage routes are mutually exclusive and belong to the two stages. α-Secretase cleavage yields soluble APPα, a candidate-generation agent: promoted by candidate-generation signals, occurring in non-raft membrane, stimulated by *reduced* membrane cholesterol, and promoting cholesterol synthesis. The α route is the neuron requesting cholesterol and building candidates while it waits.
- **Amyloid-β terminates candidate generation and removes losers.** β-Secretase cleavage begins when cholesterol has arrived and rafts have formed — cholesterol is required for amyloid-β production, and its depletion inhibits β- and γ-secretase additively. Amyloid-β then exerts negative feedback on cholesterol synthesis: the request is cancelled once filled.
- **Tau cross-links the cytoskeletons of winners.** Phosphorylation inactivates tau, dephosphorylation activates it. Tau is phosphorylated everywhere during candidate generation to release the existing structure, dephosphorylated in winners by protein phosphatase 2A so that it can cross-link microtubules to actin, and kept phosphorylated in losers so that they can be dismantled. Tau phosphorylation is a normal, necessary, spatially patterned part of memory formation.

This triad is uncomfortable in the way good proposals are uncomfortable. It says amyloid-β is *supposed* to remove synapses. It says the field's central pathological marker, phospho-tau, is the default state of tau in any neuron currently learning something.

4.5 The regional argument, derived rather than fitted

If the lesion is in plasticity, the most vulnerable regions should be those with the highest plasticity demand. Rappoport identifies the earliest tau-bearing regions — entorhinal cortex, hippocampus, locus coeruleus — as exactly the regions where continuous plasticity is required, and then does something more discriminating with the hippocampal subfields: CA1 and subiculum, much more vulnerable than dentate gyrus and CA3, support *familiar* scenes, while dentate gyrus and CA3 encode new ones, and most life experience consists of familiar scenes with an element of novelty — the exact condition that triggers candidate generation without triggering wholesale new encoding.

The sensory argument is sharper still. Olfactory, auditory and retinal impairment occur very early in this disease; the relevant sensory cells are raft-dependent; statin use is associated with sudden hearing loss and with impaired olfaction; and cholesterol depletion causes hearing loss in cats and cochlear hair-cell loss in mice. A raft theory predicts a sensory prodrome, the prodrome exists, and cholesterol depletion reproduces it.

4.6 Where it is weak

The central charge is **symmetry**. Double-edged plasticity, chronicity, and a two-population model of neurons together make almost every direction of almost every measurement a confirmation. The two-population move is the clearest instance: neurons in which chronic cholesterol production eventually succeeds form rafts normally, show no tau pathology, over-produce amyloid-β and generate plaques; neurons in which it does not show plasticity failure, tau pathology and amyloid pathology. That elegantly explains why plaques are common in cognitively normal older people and correlate weakly with symptoms — plaque is the signature of a solved problem — and it simultaneously renders the theory unfalsifiable by any measurement of cholesterol or amyloid in Alzheimer tissue.

The clinical premise is also weaker than stated. The theory needs short-term memory to be preserved while its conversion to long-term memory fails, and asserts "impaired anterograde memory with functioning short-term memory" as one of its three founding clues. Working memory and attentional control are in fact impaired early in this disease; the isolated-anterograde-amnesia picture is closer to a textbook caricature than to the neuropsychological literature.

And there is a confirmed factual error worth recording because it recurs in this literature: the claim that statins do not cross the blood–brain barrier. Lipophilic statins do (Sierra et al., 2011), and statins were among the strongest phospho-tau-lowering hits in a systematic screen of cholesterol-metabolism drugs in patient-derived neurons (van der Kant et al., 2019).

4.7 What this station contributes

Three things no other programme in this set supplies.

It supplies the **rate-limiting step in the supply chain** — astrocyte-to-neuron cholesterol transfer — and an argument from physical chemistry for why that step, rather than any other, has no redundancy.

It supplies a **reason for the raft to matter functionally**, as a gate on a transition, rather than merely as a place where enzymes are concentrated. Every other membrane programme here treats the raft as a location; this one treats it as a switch.

And it supplies the **chronicity argument**, which is the general form of the paper's central thesis: a stalled cycle is worse than an abolished one, because the cell keeps paying the forward cost of a return that never arrives.

5. Interface: The Bond That Will Not Break

5.1 The first chemical event

Christopher Ramsden's account begins one step earlier than any other in this set, at a chemical reaction. Its claim is that sporadic Alzheimer's disease begins when the products of lipid peroxidation covalently modify apolipoprotein E and the lysine-rich motifs by which it engages its receptors, and that the resulting damage to the ApoER2–Dab1 axis is the initiating molecular event.

The chemistry is not arbitrary. Reactive aldehydes generated by peroxidation of polyunsaturated fatty acids — 4-hydroxynonenal, malondialdehyde, acrolein, 4-oxo-nonenal — are electrophiles, and the side chains they attack most readily are lysine and histidine (Uchida, 2003). 4-Hydroxynonenal adducts have been detected in Alzheimer brain since the 1990s, with a reported association with *APOE4* inheritance (Montine et al., 1997), and lipid peroxidation is an early event, present in amnestic mild cognitive impairment (Markesbery et al., 2005).

The structural coincidence is the argument. Lipoprotein receptors of the low-density-lipoprotein receptor family bind their ligands through acidic, calcium-coordinating ligand-binding type-A modules, and their ligands bind through basic, lysine-enriched recognition motifs. Both apolipoprotein E and reelin engage ApoER2 and VLDLR using cationic, lysine-rich sequences. The binding chemistry of this receptor family *is* lysine chemistry — and lysine is precisely the residue that reactive lipid aldehydes destroy. The molecule that carries the peroxidisable lipid binds its receptor using exactly the residues that the products of peroxidising that lipid attack, and the aldehyde is generated on the cargo, a few ångström from its target.

5.2 What was demonstrated at the bench

The reaction was tested directly (Ramsden et al., 2022). Apolipoprotein E peptides containing the lysine–histidine-enriched receptor-binding sequences, analogues engineered to lack the double-lysine motifs, ApoER2 LA1–2 peptides, the full-length ApoER2 ectodomain and recombinant ApoE4 monomer were exposed to the aldehydes individually and as a mixture, and the products characterised by mass spectrometry and immunoblot.

Adducts formed on the predicted motifs, and the analogues lacking double-lysine motifs were resistant — the internal control the experiment needed. Stable pyrrole crosslinks formed, which is the step that distinguishes this from a generic oxidative-stress account: an adduct modifies a protein,

a crosslink covalently joins two. Montine and colleagues had shown in 1996 that lipid peroxidation products crosslink apolipoprotein E to itself (Montine et al., 1996); what was new was crosslinking of apolipoprotein E to *its receptor*, detected as high-molecular-weight ApoE–ApoER2 heterodimers.

5.3 The acid test, and why it belongs at the centre of this paper

The most consequential experiment in that paper concerns pH, and it is the point at which this programme becomes a return-leg programme rather than a chemistry programme.

The normal life cycle of a lipoprotein receptor requires release. The receptor binds ligand at the surface, internalises it, and the acidifying environment of the early endosome triggers dissociation; the ligand proceeds to the lysosome and the receptor recycles to the membrane. If the ligand cannot let go, the receptor cannot recycle, and the cell loses surface receptor.

This is a well-established cellular phenotype of *APOE4*. Chen and colleagues showed that ApoE4 selectively impairs ApoER2 recycling, reducing glutamate-receptor function and synaptic plasticity (Chen et al., 2010); Xian and colleagues showed that the recycling block is pharmacologically reversible by lowering endosomal pH (Xian et al., 2018). The standard explanation is conformational — ApoE4 adopts a molten-globule state near its isoelectric point around pH 6.5.

Ramsden proposed a different explanation: the receptor cannot release the ligand because the two are covalently joined, and a covalent bond does not care about pH. The prediction was tested in the same paper. Malondialdehyde adducts showed partial, incomplete pH-dependent reversibility, greater at pH 4 than at pH 6 — consistent with acid-labile Schiff-base chemistry. Crosslinks formed with the reactive-aldehyde mixture showed minimal reversibility and remained stable at lysosomal pH.

The two accounts of the same phenotype are therefore distinguishable by a single manipulation, and Chapter 20 develops that experiment in full.

5.4 The anatomy, drawn without tau

The programme's second contribution is a human-tissue map, and it was drawn using markers of the axis rather than markers of pathology.

Working in the perforant path — the entorhinal projection to the hippocampal formation, whose degeneration is among the oldest findings in Alzheimer neuropathology — the group reported that ApoER2 is strongly expressed in the terminal zones, and that the components of the axis accumulate there in disease: aggregates of the ApoER2 LA1–2 ligand-binding modules, native apolipoprotein E, 4-hydroxynonenal-modified apolipoprotein E, reelin, Dab1, Thr19-phosphorylated PSD-95, Tyr607-phosphorylated P85α and **Thr508-phosphorylated**

LIM-kinase 1. Plaque-associated ApoER2 LA1–2 aggregates correlated positively with Braak stage and plaque load and inversely with Mini-Mental State Examination score.

The 2023 extension generalised this to sixty-four cases across five regions: ApoER2 expression maps onto the Braak origin, VLDLR does not, and four arms of the axis co-accumulate in a pattern the authors read as co-accumulation rather than prion-like spread (Ramsden et al., 2023). The genetics arrived independently and pointed at the same adaptor: in a whole-genome analysis restricted to *APOE4* homozygotes, the single novel genome-wide significant locus was *DAB1*, with pathway analysis implicating the *DAB1–RELN* pathway itself (Bracher-Smith et al., 2022).

The last item in the marker list is the one this paper will use. Threonine 508 is the activating phosphorylation site of LIM-kinase 1, and LIM-kinase 1 is the kinase that phosphorylates serine 3 of cofilin. Chapter 18 takes it up.

5.5 The disulfide hypothesis and the cysteine count

The 2025 formulation is the sharpest version of the theory (Ramsden et al., 2025). Apolipoprotein E isoforms differ in cysteine content — E2 has two, E3 one, E4 none — and the proposal is that these cysteines form disulfide bridges that physically shield the peroxidisable lipid cargo in transit, so that the allelic series of Alzheimer risk is a series in lipid protection: E2 > E3 > E4.

An independent laboratory reached the same molecular feature from a different direction eight months later. Barger and Moerman-Herzog reasoned that ApoER2 signalling requires receptor dimerisation, that the *APOE* alleles differ in their capacity to form disulfide dimers, and that lower risk may follow from apolipoprotein E's ability to promote receptor clustering. Measuring NMDA-receptor calcium flux in human-derived neurons, they found that reelin, fibrillar amyloid-β, ApoE3 and medium from *APOE ε3* astrocytes elevated flux in an ApoER2-dependent manner, while ApoE4 and oligomeric amyloid-β *antagonised* it (Barger and Moerman-Herzog, 2026).

Two laboratories, eight months apart, independently identified the cysteine count of apolipoprotein E and its capacity to form disulfide bonds as the pivot of allelic risk, and both routed the consequence through ApoER2. They assign the bond different jobs — concealing lipid in transit, versus clustering the receptor at the membrane — and the two are not exclusive.

5.6 Where it is weak

Three weaknesses, and the first is decisive for the initiation claim.

No crosslinked species has been isolated from human brain. The chemistry was demonstrated with purified peptides and recombinant protein at aldehyde concentrations chosen to drive the reaction. The only human evidence for peroxidised apolipoprotein E is immunohistochemical. Immunoprecipitating apolipoprotein E from frozen human brain under

non-reducing conditions and looking for the receptor is a tractable experiment that has not been reported.

The granulovacuolar objection. Much of the co-accumulation signal in the human tissue work lands in granulovacuolar degeneration bodies, which sequester many unrelated phosphoproteins. The four-arm result may therefore be a compartment artefact rather than a pathway finding. The untested control is staining unrelated phosphoproteins — casein kinase 1δ, phospho-eIF2α, phospho-PKR — on the same sections.

The direction of traffic is contested, and the contest is live. Guo and colleagues reported that impaired low-density-lipoprotein-receptor binding by lipidated ApoE2 *avoids* the receptor-recycling defects seen with E3 and E4 and decreases uptake of cholesteryl esters; that apolipoprotein E particles carrying polyunsaturated cholesteryl esters produce an E4 > E3 > E2 series for lipofuscinosis in human neurons; and that the protective Christchurch variant also reduces receptor binding and phenocopies E2 (Guo et al., 2025). Ralhan and colleagues reported that E2 and Christchurch particles protect neurons by *effluxing* oxidised unsaturated lipids through ABCA7, while E4 particles exacerbate them (Ralhan et al., 2026). Ramsden's published model lists impaired lipoprotein internalisation as one of four pathogenic arms. If Guo and Ralhan are right, that arm is not merely wrong but backwards.

The tension is smaller than it appears and the framework would be stronger for saying so. On Ramsden's own account the poison is the peroxidised particle and the receptor is the surface it poisons, so reducing engagement with a poisoned ligand is protective on his model too. The three known protective variants are coherent under one rule: apolipoprotein E is the ligand you want *less* of at this receptor, and reelin is the ligand you want *more* of. Both protective *APOE* variants are poor receptor binders; the protective *RELN* variant is a better receptor activator.

5.7 What this station contributes

The **chemical mechanism for a failed return**, stated at the level of a bond. Every other programme in this set describes a trafficking step that fails; this one proposes why the molecule cannot leave.

The **human anatomical map** of the ApoER2–Dab1 axis, which is the only map in this set drawn from a signalling pathway rather than from a pathological deposit.

And the **link to the reelin axis**, which is where the two documented human resistance cases sit, and which supplies the opposite-signed input to the effector that Chapter 18 identifies as shared.

6. Contact: The Signal That Was Never Cleared

6.1 The reversal

Estela Area-Gómez's programme makes an inversion that the rest of this paper depends on. Its claim is that the pathogenic species in Alzheimer's disease is not amyloid-β but its immediate precursor, the β-carboxy-terminal fragment C99; that γ-secretase cleavage is therefore the act of *clearing* a signal rather than the act of producing a toxin; and that the disease is a homeostatic correction left switched on for fifty years.

The address came first. Presenilins — the catalytic core of γ-secretase — are enriched not in bulk endoplasmic reticulum but in the subdomain that apposes mitochondria, the mitochondria-associated ER membrane (Area-Gómez et al., 2009). That domain is a cholesterol- and sphingolipid-ordered, detergent-resistant raft, and it is where phospholipid synthesis, cholesterol esterification and lipid transfer to mitochondria occur (Vance, 1990). Contact function is measurably *increased* in Alzheimer cells and tissue (Area-Gómez et al., 2012), and increased by ApoE4-containing lipoproteins specifically — an effect requiring the lipoprotein particle, not lipid-free apolipoprotein E (Tambini et al., 2016).

If the enzyme lives at a contact and its function is a *rate*, then the disease-relevant variable is how fast the substrate is cleared, and the accumulating agent is the substrate.

6.2 The convergence on the fragment

The fragment claim did not originate in this laboratory and the programme has never said it did. Lauritzen and colleagues showed that C99, rather than amyloid-β, is the principal contributor to early intraneuronal lesions in triple-transgenic hippocampus (Lauritzen et al., 2012), and later that intraneuronal aggregation of C99 induces amyloid-β-independent lysosomal and autophagic pathology (Lauritzen et al., 2016); the same group has since asked whether γ-secretase should be understood as a beneficial inactivating enzyme for a toxic fragment (Checler et al., 2021). Nixon's laboratory identified a specific crime: the tyrosine-682-phosphorylated β-carboxy-terminal fragment inhibits the vacuolar ATPase, and this — not amyloid-β — is what fails to acidify the autolysosome in Alzheimer and Down syndrome models (Im et al., 2023). Kwart and colleagues, working across a large isogenic panel of *APP* and *PSEN1* mutant human neurons, found shared endosomal abnormalities mediated by β-carboxy-terminal fragments and not by amyloid-β (Kwart et al., 2019).

Four laboratories, working on contacts, endosomes, lysosomes and isogenic genetics, arrived at the same defendant without a shared framework.

6.3 C99 as a cholesterol sensor that builds its own platform

The functional claim is the programme's own, and it rests on structural work from elsewhere. Sanders's group showed by solution NMR that the transmembrane domain of C99 is unusually flexible and binds cholesterol directly, with a defined binding surface just upstream of the γ-secretase cleavage site, and that binding competes with C99 self-association (Beel et al., 2008; Beel et al., 2010; Barrett et al., 2012; Song et al., 2013).

Montesinos and colleagues then asked what C99 does at the contact, and answered that it *makes* the domain it occupies: because of its affinity for cholesterol, C99 delivered to the ER clusters cholesterol into transient detergent-resistant domains, and when it accumulates it drives internalisation of extracellular cholesterol and its trafficking from the plasma membrane to the ER, expanding those domains and inducing esterification while attenuating de novo synthesis (Montesinos et al., 2020).

The loop this closes is worth stating as a cycle, because it is the clearest instance in the whole set of a forward leg and a return leg:

1. Cholesterol accumulates in the plasma membrane beyond a set point.
2. The precursor protein is internalised in cholesterol-rich endosomes; β-secretase, active at low pH, cuts it to C99.
3. C99 arrives at the ER, where its cholesterol-binding domain gathers sterol into a raft — the contact.
4. On that platform, cholesterol-handling enzymes are recruited and activated; the excess is esterified and disposed of into lipid droplets.
5. γ-Secretase cuts C99. The platform disperses. The signal ends.

Alzheimer's disease, on this account, is step five failing. The correction never stops; the cell keeps importing cholesterol to the ER in answer to an alarm that has already been answered. The physiological version of the loop — required for a theory of dysregulation to have something to dysregulate — was demonstrated separately, with scavenger-receptor-B1-mediated uptake of cholesterol from high-density lipoprotein stimulating contact formation, which in turn suppresses the biosynthetic machinery (Montesinos et al., 2024, preprint).

6.4 The membrane-thickness proposal

The programme's most audacious step concerns the field's most specific molecular index, the ratio of amyloid-β42 to amyloid-β40. γ-Secretase is an intramembrane protease that is not sequence-specific; it engages its substrate and cuts processively in roughly tripeptide steps, and where in the distribution the products fall depends on the geometry of engagement. The proposal is that this geometry is set in part by bilayer thickness: in a raft of normal thickness the transmembrane helix of C99 matches the bilayer and is cut predominantly to Aβ40; if the raft is thinner, the helix tilts to maintain hydrophobic matching, its alignment with presenilin-1 changes, and the cut falls in a different register. The elevated 42:40 ratio would then be a surrogate marker for the thickness of a membrane rather than a measure of a poison.

The theory named the enzymes that would thin a raft: ACAT1, which favours oleate, and ACSL4, which preferentially activates arachidonic and eicosapentaenoic acid. The corresponding experiment was delivered in 2025 — contact formation enhances ACSL4 activity, promoting arachidonate incorporation into phosphatidylcholine through the Lands cycle with the contact-resident acyltransferase LPCAT4; elevated C99 induces contact remodelling through cholesterol clustering, which activates ACSL4 and alters phosphatidylcholine composition; and the effect is mirrored in fibroblasts, neurons and immune cells from both familial and sporadic patients (Montesinos et al., 2025, preprint). Independently, Dawkins and colleagues showed that membrane lipid remodelling modulates γ-secretase processivity (Dawkins et al., 2023).

This is nonetheless the programme's most over-stated step, and it should be labelled. The dominant explanation for the 42:40 shift is protein-intrinsic: Alzheimer-causing mutations shift peptide length by destabilising the interactions between the enzyme and its successive intermediates (Szaruga et al., 2017), and the resulting profiles predict pathogenicity and age at onset (Petit et al., 2022); cryo-electron microscopy has visualised substrate capture and stepping directly (Yang et al., 2019; Zhou et al., 2019; Guo et al., 2024). Processivity and bilayer thickness are not competing mechanisms — the first is the proximate description and the second a proposed modulator — but the quantitative question of how much of the human ratio shift is attributable to membrane geometry has not been answered by anyone.

6.5 The signed framework

The programme's most under-sold virtue is that it is *signed*. Contact function is up in Alzheimer's disease and down in synucleinopathy and TDP-43 proteinopathy (Guardia-Laguarta et al., 2014; Larrea et al., 2025; García-Toledo et al., 2025). A tool compound that increases contact formation is therefore right for amyotrophic lateral sclerosis and exactly wrong for Alzheimer's disease (Etxebeste-Mitxeltorena et al., 2025). Almost no framework in this field states its own direction per disorder, and a signed framework is falsifiable in a way an unsigned one is not.

The pharmacological corollaries follow the same logic and read the trial record as prediction rather than disappointment. γ-Secretase inhibition with semagacestat did not merely fail; it made patients worse (Doody et al., 2013), which is the predicted result of inhibiting the enzyme that clears C99. β-Secretase inhibition, which lowers C99 by preventing its formation, *also* worsened cognition (Egan et al., 2018) and produced rapid, non-progressive, reversible regional brain-volume reduction (Sur et al., 2020) — which the theory absorbs by holding that some C99 production is required for a normal function, so suppression below the physiological range is itself harmful. That is not an evasion; it is the natural consequence of holding that the fragment is a signalling intermediate rather than waste. But it converts an apparent prediction into a constraint: **any therapy directed at C99 must normalise it, not abolish it.**

6.6 Where it is weak, and what it contributes

The weaknesses are the thickness claim as stated, the borrowing of the tau arm from another laboratory's pharmacology (van der Kant et al., 2019), and a dependence on preprints for five of the load-bearing recent results.

What the station contributes is threefold. It supplies **the clearest statement of a stalled cycle in the whole set** — a signal produced, acted upon, and never terminated. It supplies **the reason a lipid theory has an amyloid pathology**, without needing amyloid-β to be toxic. And it supplies **the mechanism by which membrane composition is fed back onto the amyloid machinery**, which is the join between the supply-side programmes of Chapters 4 and 5 and the trafficking programmes of Chapters 7 and 8.

7. Sorting: Retromer, SORLA, and Residency Time

7.1 Anatomy first

Scott Small's laboratory has spent twenty years asking *where* the disease starts and *why there*, and using the answer as a filter on molecules. The disease begins with striking reliability at the junction of the entorhinal area and the temporal neocortex, and spreads along a route mapped in post-mortem tissue since Braak and Braak (1991). To this programme that regional preference is not a curiosity to be explained once the mechanism is known; it is the principal available evidence, and it can be used as an instrument.

The founding paper is the clearest application (Small et al., 2005). Expression profiling of diseased brain was generating candidate lists of unusable length, most of whose entries report the end state rather than anything upstream. Instead of addressing this statistically, the group constructed a spatiotemporal model *before* generating data: a driver should be abnormal in the entorhinal cortex, relatively spared in the dentate gyrus, and abnormal across a broad age span. The transcript that best conformed was *VPS35*, the cargo-recognition core of the retromer complex — a molecule with no prior association with the disease, which entered the field entirely through an anatomical filter.

The method's cost should be stated with equal clarity. A filter constructed from a model of the disease can only return molecules that fit the model; a glial change affecting both regions equally but tolerated in one would be discarded. The 2005 result is a well-motivated nomination, not evidence of causation, and its authors described it that way. What breaks the circularity is the interventional work that followed. What is genuinely striking is how little the *method* has been adopted: using differential regional vulnerability inside a single circuit as a filter on molecular candidates has been applied within this group to ageing and to psychosis, and scarcely anywhere else.

7.2 The traffic jam

Material internalised from the surface arrives at the early endosome and is either sent onward for degradation or recovered and returned. Retromer — VPS35, VPS26, VPS29, working with sorting nexins and with cargo-selective receptors — performs the recovery. The most important cargo receptor here is SORLA, encoded by *SORL1*, which binds the amyloid precursor protein, directs it away from the amyloidogenic route, and engages retromer through its short cytoplasmic tail; the association is regulated by the receptor's own dimerisation state (Jensen et al., 2023).

Jam the export step and residency time rises — for everything held there, not only for the precursor protein (Small et al., 2017). That single mechanical fact generates the disease's separate pathologies as parallel consequences rather than as a chain. The precursor protein and BACE1 spend longer together in an acidic compartment, favouring the amyloidogenic cut. Glutamate receptors that should be recycled to the postsynaptic membrane are retained and diverted toward degradation, and the synapse weakens without any cell dying. A congested endosome matures into a multivesicular body and releases cytosolic contents, tau among them, as exosomes. And microglia, which depend on the same machinery to return phagocytic receptors to their surface, clear less.

The supporting evidence is best read in ascending order of what each design can establish. Expression and correlation in vulnerable human regions (Small et al., 2005; Simoes et al., 2021) establishes association in end-stage tissue. Perturbation in animals — retromer-deficient mice and flies with memory and synaptic dysfunction, neuronal loss and amyloid accumulation (Muhammad et al., 2008) — establishes sufficiency in a model. Depletion and repletion is the strongest design: VPS35 was depleted in mouse hippocampal neurons and restored with an optimised vector, and the accelerated precursor-protein cleavage and loss of synaptic glutamate receptors followed the manipulation in *both* directions; the same study found a neuronal retromer deficit sufficient to induce dystrophic microglial morphology, independently of tau (Qureshi et al., 2022). And human genetics carries the direction of causation without a model organism: truncating *SORL1* variants occur almost exclusively in cases, now mapped to receptor domains across large aggregated cohorts (Andersen, de Waal et al., 2025).

7.3 Why that region

The anatomical claim went unexplained for fifteen years, and the answer offered in 2021 is the most elegant result in the programme. Neurons are enriched in a *second* retromer core organised around VPS26b rather than the ubiquitous VPS26a, and the two are not interchangeable: the VPS26b core is dedicated to the *recycling* arm rather than to retrograde transport toward the Golgi. The trans-entorhinal cortex proved to be the region most susceptible to VPS26b depletion, VPS26b is enriched there in human brain, and both VPS26b and SORLA are deficient there in disease; VPS26b mediates the recycling of glutamate receptors and of SORLA itself (Simoes et al., 2021).

The account of *why* the region is built that way is an argument from demand — entorhinal neurons carry unusually extensive arbours and occupy a hub position, so maintaining synaptic function across that surface loads the recycling machinery heavily, and a cell running near capacity is the cell in which a modest loss of capacity first produces a deficit. That is inference, and should be labelled as such. What is demonstrated is the differential expression and the differential susceptibility. It explains onset; it does not explain spread, and the programme does not claim otherwise.

7.4 Hub and spoke, and its price

By 2020 the hypothesis faced an obvious objection: if endosomal recycling is the upstream lesion, why do mutations in the precursor protein and the presenilins cause fully penetrant early-onset disease? Small and Petsko answered architecturally (Small and Petsko, 2020). Endosomal recycling is the hub; the precursor protein, the secretases, apolipoprotein E and the immune receptors are spokes. A spoke can be damaged directly, and the products of a damaged spoke feed back on the hub, so the two converge on the same downstream state.

This is genuinely good theoretical work, and it makes the theory very difficult to refute. A model in which every spoke feeds back on the hub can accommodate almost any result: evidence that amyloid damages the endosome is a spoke feeding back; evidence that tau precedes amyloid is another spoke; evidence that the lesion is glial is microglia using the same hub. The appropriate response is not to reject the model but to require a statement of what would count against it, and Chapter 32 adopts that discipline for the integration as a whole.

The therapeutic corollary is independent of the causal argument and is the most useful thing in the programme: if the hub is common to the spokes, repairing the hub should benefit patients whose primary lesion is in any spoke, including spokes the model does not name.

7.5 What the last two years did

Five developments strengthened the case materially, and two complicated it.

Strengthening: a domain-level classification of *SORL1* variants (Andersen, de Waal et al., 2025); an independent replication of the retromer-chaperone strategy in 5xFAD mice, restoring long-term-potentiation-associated gene expression including *Gria1* and *Grip1* (Ramonet et al., 2025); a tractable new target, with ROCK2 phosphorylation of the SORLA tail shown to reduce its affinity for retromer and inhibition of the upstream activator RhoGEF12 reducing amyloid-β in a *SORL1*-dependent manner (Qureshi et al., 2026, preprint); and — most consequentially — the first direct animal test of the tau arm, from an independent laboratory, in which transgenic up-regulation of SORLA in aged PS19 mice reversed tau phosphorylation, tau seeding, synapse loss, impaired potentiation and glial activation, while deletion worsened all of them (Huang H. et al., 2026).

Complicating: the first assay of pathway function in living people. Soluble SORL1 in cerebrospinal fluid was significantly lower in carriers of truncating and damaging missense variants, as the model predicts — but **in sporadic patients carrying no such variant it did not differ from controls** (de Waal et al., 2026). This is the first quantitative test of the word *common*, and it did not come back positive. Several readings remain open — the shed ectodomain may be an insensitive index of intracellular recycling; a lesion confined to the trans-entorhinal cortex may not move a whole-compartment measure; the relevant deficit may be in a cell type contributing little to

the cerebrospinal pool — and each is testable.

The second complication is the cell type. The strongest protective common variant at the locus, rs11218343, acts through *microglial* SORLA expression (Gorniak-Walas et al., 2026); SorLA governs endoplasmic-reticulum stress, lipid-droplet accumulation and phagocytosis in human microglia, with the lipid phenotype conserved in neurons (Haq et al., 2026); and the group's own human imaging places microglial density as a statistical mediator between tau and neurodegeneration (Lao et al., 2026). Chapter 23 takes this up as a general phenomenon rather than a local one.

7.6 What this station contributes

The **canonical statement of a return-leg lesion**: retromer is, by definition, the retrieval machinery, and the disease is placed on retrieval rather than on uptake.

The **strongest human genetic anchor in the set** outside *APOE*, with a causal architecture rather than an association.

The **hub-and-spoke argument**, which is the general form of the therapeutic claim this paper makes in Chapter 28.

And a **direct, unresolved contradiction at the crux** — whether the endosomal phenotype downstream of *SORL1* loss requires the amyloid precursor protein (Knupp et al., 2020, against Hung et al., 2021) — which Chapter 26 declines to dissolve.

8. Retention: The Synaptic Endosome and the Four Routes

8.1 One machine, two jobs

Gunnar Gouras's programme rests on a structural fact that took twenty years to assemble and whose consequence his own framing has tended to obscure: **a nerve cell manufactures amyloid-β on the same organelles it uses to run its synapses.**

The founding result is about geography, not disease. Amyloid-β40 is generated in the trans-Golgi network and packaged for export, whereas a population of amyloid-β42 is generated and *retained* in the endoplasmic reticulum in an insoluble state (Greenfield et al., 1999). The compartment in which the precursor is cut determines which peptide is made and whether it leaves the cell.

Where that geography lies was answered much later and is more specific than most summaries convey. The two enzymes competing for the first cut — BACE1, which begins the amyloidogenic route, and ADAM10, which pre-empts it — are both strongly enriched on *synaptic vesicles* isolated from rat brain, with ADAM10 activity detectable in the vesicle fraction and the first-cut fragments enriched there; of the complex that performs the second cut only one component was enriched, and active enzyme co-localised with the vesicle marker only sparsely (Lundgren et al., 2015). The first step of amyloid production happens on the machinery of neurotransmitter release. The last step happens elsewhere.

The human observation followed the geography. Amyloid-β42 accumulates within neurons in the vulnerable regions, in a distribution that appears to precede plaques and tangles (Gouras et al., 2000); electron microscopy localised it to multivesicular bodies inside synaptic terminals, associated with abnormal synaptic structure before plaque pathology (Takahashi et al., 2002); and it was seen assembling into small soluble clusters inside those same processes (Takahashi et al., 2004). Independently, Cataldo, Nixon and colleagues had shown that swollen early sorting compartments precede amyloid deposition in sporadic disease and in Down syndrome (Cataldo et al., 2000). Two lines converged on the same compartment from different directions.

8.2 Retention, not overproduction

The dominant reading of amyloid in this disease is quantitative: there is too much of it. This programme points somewhere else, and it is the most under-cited turn in the whole of this literature. The variable that matters is not how much peptide is made but **how much of it fails to leave.**

The direct evidence is a short paper that deserves to be better known (Tampellini et al., 2011). In cultured neurons from Alzheimer-model mice compared with normal neurons over time, the amount of amyloid *secreted* fell with time — it did not rise — and normal neurons showed no such decline. The ability of synaptic activity to increase secretion while simultaneously reducing the intraneuronal pool became impaired in diseased neurons and not in normal ones. A mechanism was then identified: synaptic activity normally increases surface neprilysin, an amyloid-degrading enzyme, and its co-localisation with amyloid-β42; in the diseased neurons neprilysin levels fell with time.

The diseased synapse loses the activity-coupled clearance system that normally keeps its local amyloid pool low. It is not making more; it is disposing of less, and the disposal step that fails has a name.

The distinction has therapeutic consequences that the field has not drawn. A production problem is addressed by reducing synthesis, which is the strategy behind the secretase inhibitors that failed. A retention problem is addressed by restoring export and degradation, which is a different target list entirely and has scarcely been attempted.

If retention is the variable, then anything that raises it enters the same pathway, and the programme has documented a striking number of independent entry points: normal ageing, through up-regulated internalisation of the precursor protein in aged neurons, with blockade of production reversing the associated synapse loss (Burrinha et al., 2021); apolipoprotein E4, which meets the precursor and the peptide inside neurites and raises neuronal amyloid-β42 (Konings et al., 2023); precursor-protein mutation, with the Arctic variant reducing surface presence and shifting production to intracellular sites (Sahlin et al., 2007); disrupted sorting machinery (Edgar et al., 2015; Willén et al., 2017a); seeded aggregates (Olsson et al., 2018); loss of the degrading enzyme (Tampellini et al., 2011); reduced synaptic activity (Tampellini et al., 2009, 2010); and prior intracellular load (Roos et al., 2021). Eight conditions, causally independent of one another, one downstream compartment.

8.3 Four routes from a loaded compartment to a failing synapse

The mechanism proper is a set of four partly independent routes, established at different times by different methods, no one of which accounts for the whole phenotype.

- **Receptor withdrawal.** In precursor-mutant neurons the earliest measurable change was a reduction in PSD-95 followed by reduced surface GluA1, blocked by inhibiting amyloid production and reproduced by adding synthetic amyloid to normal neurons (Almeida et al., 2005). In parallel, amyloid promotes internalisation of NMDA receptors, with fewer at the surface in model neurons and restoration on inhibiting production; the pathway was traced through a nicotinic receptor, a phosphatase and dephosphorylation of a specific tyrosine (Snyder et al., 2005).

- **Autocatalysis.** Amyloid accumulation impairs multivesicular-body sorting by inhibiting the proteasome and the de-ubiquitinating enzymes (Almeida et al., 2006). The structure of that finding is a feedback loop with the wrong sign: **the cargo disables the machine that would clear the cargo.** It is a mechanism for why a slow accumulation should accelerate rather than plateau, and it requires no external driver — whatever raised the pool initially need not still be present.

- **Structural failure.** Compartments loaded with undegradable cargo distend; interfering with the packaging machinery reproduces both the accumulation and the enlargement (Willén et al., 2017a). Three-dimensional reconstruction showed fibrillar amyloid inside individual synaptic compartments, in places appearing to pierce the membrane (Capetillo-Zarate et al., 2011), and accumulating intraneuronal amyloid-β42 tracks early changes in MAP2 in neurites and synapses (Takahashi et al., 2013). This route explains the swollen, clubbed nerve endings around plaques that have been described since 1907 and that no purely biochemical account addresses.

- **Translational block.** The mTOR pathway is suppressed in model hippocampus and in normal tissue exposed to amyloid; potentiation can be rescued by restoring mTOR signalling (Ma et al., 2010), and an elongation factor required for translation is dysregulated in proportion to the plasticity impairment (Beckelman et al., 2016). Apolipoprotein E4 reaches the same arm by another road: acute exposure abolishes the translational response to NMDA-receptor stimulation, through a sustained calcium rise, sustained eEF2 phosphorylation and sustained translational block (Ramakrishna et al., 2021).

That the routes are separable matters twice. It explains why the synaptic phenotype is robust — four partly redundant mechanisms will not be abolished by blocking one — and it predicts that single-target therapy at this level should underperform, which is what has happened.

One further result qualifies all of it: the association of amyloid and its precursor with synapses is *heterogeneous*, with preferential binding to excitatory rather than inhibitory neurons and earlier accumulation of cleavage fragments on the transmitting side (Willén et al., 2017b). Synapses are not uniformly exposed, and what determines which ones are is unresolved.

8.4 The lesion is the loss of the control loop

The programme's most recent decade locates the actual lesion, and it is not the peptide.

Neurons hold their firing rate near a set point by multiplicatively adjusting synaptic strength and tuning intrinsic excitability (Turrigiano, 2008). That machinery is broken here. Neurons carrying elevated amyloid became hyperactive, consistent with earlier reports of hyperactivity near plaques (Busche et al., 2008), but the consequential findings concerned regulation rather than level: the diseased neurons failed to adapt their calcium responses to imposed global changes in activity, and failed to adjust the length of the axon initial segment — a structural adaptation neurons use to retune their own excitability (Martinsson et al., 2022). The cell has not become noisy; it has lost the ability to find its own set point.

A parallel result concerns the amyloid homeostat itself. In cells producing amyloid, removing the peptide from the medium caused the internal pool to fall by more than eighty per cent within three hours and recover fully within six, with a rise in the first-cut fragment: the cell detected the external drop and increased production to compensate. In cells that had accumulated internal aggregates, the correction did not occur, and the authors conclude that production may be permanently up-regulated (Roos et al., 2021).

The lesion is not the presence of amyloid but the loss of the loops that regulate it. The peptide is present in every normal brain and is under control there; what the disease adds is not the substance but the failure of governance.

Honesty requires recording the group's own complicating result. In the same 2022 study, neurons expressing a precursor protein engineered so that it cannot yield amyloid at all became just as hyperactive; and deleting the precursor protein altogether alters synaptic proteins in a direction broadly *opposite* to that seen in precursor-mutant neurons (Martinsson et al., 2019). Some part of the phenotype belongs to the precursor rather than to the peptide, and the account does not yet say how much.

8.5 Activity, and the experiment that separates plaques from function

Activity has opposite signs in the two compartments: it raises what is outside the cell and lowers what is inside. Synaptic activity reduces the intraneuronal pool, promotes transport of the precursor to synapses, and protects against amyloid-related synaptic alterations, with the reduction mediated by neprilysin (Tampellini et al., 2009).

The decisive test was a designed dissociation (Tampellini et al., 2010). Two independent means of chronic synaptic inhibition — surgical deafferentation of barrel cortex, and a benzodiazepine — were applied to model animals. Both reduced plaques. Both made the animals worse: prolonged inhibition exacerbated loss of the presynaptic marker synaptophysin relative to more active regions,

and benzodiazepine treatment followed by washout worsened memory. The deterioration occurred in the setting of *reduced* plaques and *elevated* intraneuronal amyloid.

Plaque burden and functional outcome were driven in opposite directions in the same animals, and outcome followed the intraneuronal pool. Whatever else is true of this programme, that experiment is a direct demonstration that the extracellular deposit is not the variable that tracks function.

8.6 The measurement problem, and its partial answer

The whole claim rests on detecting a small peptide inside a cell full of the protein it was cut from, using antibodies that recognise a region present in both. The most candid statements of this objection have come from Gouras himself: the standard immunoassay systematically *underestimates* amyloid once it has assembled, because assembly hides the epitopes (Stenh et al., 2005); and detection difficulties have made the topic "remarkably controversial," with standard biochemical methods underestimating the intraneuronal pool and detergent used in tissue processing capable of removing it altogether (Gouras et al., 2012).

The response was to abandon antibodies. Synchrotron-based infrared micro-spectroscopy reports β-sheet conformation as a physical signature, without a reagent that must recognise a sequence, and showed that structural states of both the peptide and its precursor are altered in model brain *before* any plaque forms, with focal pre-plaque aggregates localising to synaptic terminals (Klementieva et al., 2017); the method was then pushed to super-resolution in neurons (Klementieva et al., 2020). A second antibody-independent line came from thioflavin-S three-dimensional reconstruction (Capetillo-Zarate et al., 2011).

The fair position is that the objection has been substantially but not entirely answered. Three methodologically independent lines agree qualitatively. None gives an absolute measure of how much peptide is inside a human neuron at a given stage, and the most decisive of them requires specialist instrumentation and has been performed largely in model systems.

8.7 Where it is weak, and what it contributes

The weaknesses are stated plainly by the programme's own record. Most of the cellular mechanism was established in precursor-over-expressing animals, which are exactly the systems in which an intraneuronal-accumulation account would look best; the knock-in models built to remove that artefact have not been used to rebuild the case, and the one published test in a knock-in went against a prediction (Andersson et al., 2023). The two most load-bearing results — impaired secretion with loss of surface neprilysin, and loss of the homeostatic correction in loaded cells — appear not to have been independently reproduced. The human evidence is descriptive rather than mechanistic: none of the four routes has been demonstrated in human tissue. And no therapeutic candidate has emerged in twenty-eight years.

What the station contributes is the **best-specified account in the set of how a synapse actually stops working**, the **explicit statement that retention rather than production is the variable**, and the **demonstration that the pool which damages synapses is the pool that no clinical assay measures** — which Chapter 29 develops into the paper's principal biomarker argument.

9. Platform: Caveolin-1 and the Organ of Reception

9.1 The unit of signalling is a patch of membrane

Brian Head's premise is cell biology that predates the Alzheimer application. The plasma membrane is not uniform; cholesterol and sphingolipids segregate into ordered microdomains that are less fluid, thicker, and buoyant after carbonate lysis. Within them sit the caveolins, simultaneously scaffolding proteins and cholesterol-binding proteins. Caveolin-1 organises neurotrophin receptors, NMDA and AMPA receptor subunits, the cyclic-AMP machinery, Src and the kinases downstream of it, and Rho-family cytoskeletal regulators into one patch (Egawa et al., 2016).

The claim that follows is about *sufficiency of arrangement versus sufficiency of components*. A neuron may express normal amounts of TrkB with normal BDNF present, and the signal will not be transduced if TrkB is not in the raft. The demonstration is in the null: in caveolin-1 knockout neurons, NMDA- and BDNF-mediated pro-survival kinase activation fails, and re-expressing caveolin-1 restores it (Head et al., 2011). The receptors were there the whole time.

An objection arrives immediately and was met before the therapy was built. Caveolae — the flask-shaped invaginations that name the caveolins — are abundant in endothelium and muscle and rare in neurons. Head and Insel argued that caveolins act outside caveolae, as scaffolds concentrating signalling partners in flat raft domains (Head and Insel, 2007), and the evidence since is consistent: immunogold localisation to pre- and postsynaptic membranes and the synaptic cleft (Egawa et al., 2018), scaffolding co-immunoprecipitations rather than vesicular ones (Kassan et al., 2017; Wang S. et al., 2021a), and a requirement for caveolin-1 phosphorylation in axonal growth of human neurons (Wang S. et al., 2019).

Conceding that point has a consequence the programme does not always draw out. If neuronal caveolin-1 is present at low abundance and without caveolae, then the therapy is not restoring a neuron to its normal state; it is imposing a membrane organisation more caveolin-rich than any neuron normally has. That is not a fatal objection — many effective interventions are supraphysiological — but it changes the safety argument.

9.2 The ageing membrane, and the prediction it made

The bridge to disease is a claim about ageing with independent support. Comparing young, middle-aged and aged mouse hippocampus, raft localisation of PSD-95, GluN2A, GluN2B, TrkB, AMPA receptors and caveolin-1 itself all decline with age (Head et al., 2010); brain cholesterol falls with age, raft abundance falls with it, and presynaptic vesicle fusion declines (Egawa et al., 2016).

The distinctive move is what this predicts about therapy: **if the ageing neuron has lost the platform rather than the ligand, supplying ligand should not work.** That prediction has been tested in human beings, expensively. Nerve growth factor delivered by gene therapy to the basal forebrain produced encouraging phase-1 and autopsy evidence of neuronal response (Tuszynski et al., 2005; Tuszynski et al., 2015) but no clinical benefit in a randomised trial (Rafii et al., 2018), and post-mortem analysis attributed the failure principally to delivery geometry (Castle et al., 2020). The platform argument supplies an additional and non-exclusive reason: a neuron whose Trk receptors are no longer raft-localised cannot transduce the ligand however much of it arrives.

9.3 The dissociation

The therapeutic construct is caveolin-1 under a synapsin promoter delivered in adeno-associated virus, and it produces a signature result across five models and three delivery routes: cognition, synapses, spines, dendritic arbour, myelin and mitochondrial integrity preserved, with **plaque load entirely unchanged**.

Table 3 — One construct, four aetiologies

Model	Disease	Delivery	Principal result
APPswe/PS1ΔE9	Amyloid	Hippocampal AAV9, 3 months	Learning and memory preserved at 9 and 11 months; synapses, spines, arbour, myelin preserved; plaques unchanged (Wang S. et al., 2021a)
App knock-in, humanised at physiological expression	Amyloid	Hippocampal AAV9, 8 months	Contextual memory preserved at 12 months; transcriptome near wild-type; plaques unchanged (Wang S. et al., 2025)
SOD1 mutant mouse and rat	Motor neuron disease	Transgenic cross; subpial spinal AAV9	Onset delayed, neuromuscular junctions preserved, survival extended; mutant protein unchanged (Sawada et al., 2019; Wang S. et al., 2022a)
TDP-43 mutant	TDP-43 proteinopathy	Systemic AAV-PHP.eB, 2 months	Contextual memory nearly doubled; raft mislocalisation of TDP-43 reduced; mitochondrial fission suppressed (Wang et al., 2026)
Controlled cortical impact	Traumatic brain injury	Hippocampal AAV9	Motor function improved, memory preserved (Egawa et al., 2017)

Five models, four aetiologies, no shared upstream molecule. This is the programme's most striking fact and its most serious interpretive problem, and the two are the same fact.

Two readings are available and neither is refuted by anything published. On the **convergence reading**, membrane raft organisation is a shared downstream node: whatever the upstream insult, a stressed neuron loses raft integrity and with it the ability to transduce the trophic signals that would sustain it, so restoring the platform restores that capacity regardless of what degraded it. On the **non-specific reading**, this is a synaptogenic, pro-arborisation, pro-myelination intervention that improves neuronal health in any model where neurons are unhealthy — raising the substrate that disease consumes rather than engaging disease mechanism.

Three observations discriminate, and two are available now. The ceiling test runs mildly against the non-specific reading: in healthy adult and aged mice the construct improved contextual fear memory, but the aged animals gained *less* structurally than adults, which is the wrong direction for "more substrate is better" and the right direction for "restore what is lost". The mislocalisation test is the strongest evidence for convergence — mutant TDP-43 mislocalises to rafts and the construct reduces that mislocalisation, and a non-specific health-improving intervention has no reason to change where a mutant protein goes — but it rests on a single experiment in one sex of one line. The dose–response test has not been done: if the mechanism is raft restoration, benefit should track

raft-localised protein and saturate once occupancy is restored; if it is trophic support, benefit should track total expression. Every published experiment uses one dose.

9.4 Resilience, and the vector that fits

The programme is presented as a gene therapy, and that description is accurate and unhelpful, because it invites comparison with the class of interventions this therapy is least like. Gene therapies in neurodegeneration are replacement or suppression strategies aimed at a known causal molecule, and each is evaluated by whether it corrects a lesion. This construct corrects no lesion in any of the five models. What it changes is the terms on which the neuron meets the lesion.

The right comparison class is **resilience**. Between a fifth and a third of cognitively intact older people carry substantial pathology at autopsy (Dubois et al., 2016); synapse density rather than plaque burden tracks cognition (DeKosky and Scheff, 1990; Terry et al., 1991; Scheff et al., 2007); and two documented human variants have held cognition intact against overwhelming amyloid burden — the *APOE3* Christchurch homozygote (Arboleda-Velasquez et al., 2019) and the *RELN*-COLBOS carrier (Lopera et al., 2023).

Table 4 — Three instances of resilience, compared on deliverability

	APOE3 **Christchurch**	*RELN*-**COLBOS**	**Synapsin-driven caveolin-1**
Acts on	Lipoprotein-receptor binding; heparan sulfate interaction	Reelin signalling to the tau kinase cascade	Membrane platform assembly
Requires knowing the cause	No	No	No
Position	Upstream of tau spread	Upstream of tau phosphorylation	Downstream, at the synapse
Pathology changed	Tau spread limited	Tau limited locally	None
Deliverable in a viral vector	No — a point substitution requiring in-situ editing	No — 3,461 residues	**Yes — 178 residues**

The last row is the point. The two human resilience alleles the field most wants to phenocopy cannot be delivered as genes. Caveolin-1 is 178 amino acids.

9.5 Where it is weak

Four weaknesses, and the first is the largest hole.

The unmeasured pool. Caveolin-1 is the entry route for amyloid oligomers, in complex with cellular prion protein (da Silva Correia et al., 2024). No study of the therapy measures intraneuronal or oligomeric amyloid — only plaque. A therapy that raises the abundance of the entry route while reporting only the deposit has not measured the quantity that Chapter 8's programme says matters.

The direction problem. Human bulk tissue reports that caveolin-1 *rises* in Alzheimer's disease (Gaudreault et al., 2004; Kang et al., 2006); this programme holds that neuronal caveolin-1 falls. The discrepancy is almost certainly a cell-type artefact — endothelial and glial up, neuronal down — and is settleable from existing public single-nucleus data. The therapy does not need the premise; asserting it is a gratuitous liability.

The measurement problem, which is the real translational blocker. A therapy defined by changing nothing imageable has no target-engagement biomarker. The nearest human-scanner candidate is an ultrashort-echo-time magnetisation-transfer myelin readout (Wang J. et al., 2026); developing it would be worth more than efficacy in a sixth model.

Delivery caveats that are routinely under-reported. The central-nervous-system tropism of the systemic capsid used in the 2026 study depends on LY6A, which has no human orthologue (Hordeaux et al., 2018; Huang Q. et al., 2019), so that result is a biology experiment rather than a delivery one. And striatal caveolin-1 elevation produced a methamphetamine-vulnerability phenotype (Avchalumov et al., 2021), which means a neuron-specific promoter is not containment enough for a brain-wide route.

9.6 What this station contributes

The **strongest available demonstration that function and pathology can be separated**, reproduced across five models — which is the experimental counterpart of the human resilience phenomenon, and the empirical foundation for Chapter 28's argument that the target is capacity rather than deposit.

The **generalisation of the raft claim beyond Alzheimer's disease**: assembled with the rest of the membrane literature, six independent lines from four laboratories in three diseases converge on one compartment, and the proposition worth extracting is larger than caveolin-1 — **the neuronal membrane microdomain is a shared failure surface on which mechanistically unrelated neurodegenerative processes converge.**

10. Disposal: The Surface Protease and the Spine Brake

10.1 Two proposals about one compartment

Seth Margolis's work carries two distinct proposals about the dendritic membrane, made a decade apart, and the most useful thing that can be done with them is to notice that they were revised in *opposite* directions.

The first proposal concerns Ephexin5, a guanine-nucleotide exchange factor. In development it was described as a brake on excitatory synapse formation: Ephexin5 restrains synapse formation through RhoA, EphB receptor activation triggers its degradation via the ubiquitin ligase Ube3A, and the brake is released (Margolis et al., 2010). Reasoning that amyloid-β depletes EphB2 (Cissé et al., 2011), and that a neuron losing EphB2 would fail to degrade Ephexin5 and would therefore re-apply a developmental brake, the group reported that Ephexin5 protein is elevated in Alzheimer model mice and in human Alzheimer tissue, and that reducing its expression ameliorated Alzheimer-like impairment in mice (Sell et al., 2017).

The second proposal concerns a nervous-system-specific 20S proteasome complex associated with the neuronal plasma membrane, exposed to the extracellular space, and catalytically active. It is uncapped, so it operates without ATP and without ubiquitin recognition; its catalytic chamber is oriented so that products are released outside the cell; and its outputs are not merely waste but signals — selective inhibition with a cell-impermeant inhibitor blocked production of extracellular peptides and attenuated activity-induced calcium signalling, and the peptides themselves were sufficient to induce it (Ramachandran and Margolis, 2017). During stimulation the complex degrades a large fraction of ribosome-associated nascent polypeptides, independently of canonical ubiquitylation (Ramachandran et al., 2018). Its peptide products promote NMDA-receptor-dependent calcium influx, sustained CREB phosphorylation and induction of activity-regulated genes (Türker et al., 2024). And in intact tadpole brain, acute inhibition rapidly increased spontaneous activity, produced hypersynchrony across tectal neurons, and abolished learning-dependent improvement in behaviour (He et al., 2023).

The functional proposal is homeostatic: activity drives synthesis, and the same activity drives a co-translational disposal arm that decides how much of what was just made survives. The coupling sets the amplitude of the response.

10.2 The first proposal, weakened

Ephexin5 is not the RhoA-selective brake it was described as. It is a **substrate switch gated at a single tyrosine**: phospho-Y361 directs it toward RhoA, dephospho toward Cdc42, and the Cdc42 arm is *required* for activity-driven spine growth (Petshow et al., 2025). Y361 is the same residue that licenses Ube3A-dependent degradation (Margolis et al., 2010), so abundance and activity are one covalent modification rather than two variables, and phosphorylation state — not protein level — determines the sign of the effect. Independently, protein kinase Cε inhibits spine development through dual phosphorylation of Ephexin5 (Schaffer et al., 2018), and dendritic Ephexin5 falls by activity-dependent *proteasomal* degradation (Hamilton et al., 2017).

Human genetics then inverted the therapeutic prescription. Loss-of-function mutations in *ARHGEF15*, the gene encoding Ephexin5, cause autosomal-dominant hereditary cerebral small-vessel disease with osteoporotic fracture, through RhoA/ROCK2 *inactivation* (Ding et al., 2023). The 2017 recommendation to lower Ephexin5 is now a phenocopy of a human vasculopathy, and *ARHGEF15* is not an Alzheimer risk locus in the large association studies (Bellenguez et al., 2022).

10.3 The second proposal, strengthened

The tau claim moved from preprint to publication with a change of emphasis and of numbers (Paradise et al., 2026). Selective inhibition of the surface proteasome rapidly triggers *de novo* formation of endogenous, sarkosyl-insoluble tau paired helical filaments in primary neurons and in mouse brain, sharing biochemical and ultrastructural features with filaments from human Alzheimer brain. Surface abundance of the complex is modulated by apolipoprotein E isoform in the order **E2 > E3 > E4**, and declines with age. ApoE4 neurons accumulate tau aggregates after modest disruption; ApoE2 neurons resist.

The design deserves description because its strength lies in what it does *not* require. The inhibitor is membrane-impermeant, restricting inhibition to the surface pool and leaving the cytosolic proteasome untouched. The tau is endogenous, non-mutant, and in vivo is human tau from a humanised locus. No seed is applied and no pathogenic mutation is required. Filaments appear in hippocampus within three days — faster than the week-long course typical of seeded aggregation. Co-application of cycloheximide abolishes the effect, placing the requirement on new synthesis rather than on conversion of an existing soluble pool. Reported thresholds scale with genotype: aggregation after roughly twenty per cent inhibition of the surface pool in ApoE4 neurons, around sixty per cent in ApoE3, about eighty-five per cent in ApoE2, with surface abundance reduced by roughly a third in ApoE4 and approximately doubled in ApoE2 relative to ApoE3.

A companion preprint localises the process: despite broad distribution of *Mapt* messenger RNA, tau is translated almost exclusively in dendrites, and about one third of newly synthesised tau is co- or peri-translationally degraded there by this complex; failure of that degradation leads to protein-synthesis-dependent accumulation of somatodendritically mislocalised tau aggregates (Konrad-Vicario et al., 2025, preprint).

If that survives review it closes a fifteen-year gap. Somatodendritic mislocalisation of tau is among the best-documented early events in the disease (Zempel et al., 2010; Hoover et al., 2010; Ittner et al., 2010), and that literature treats dendritic tau as protein that arrived where it should not be. The new result proposes instead that dendritic tau is where tau is *made*, that a third of it is normally destroyed on the spot, and that the phenotype is a failure of local disposal rather than of local exclusion. That is a different account of the same observation, and it is testable against the old one.

10.4 Two epochs

Placing the two proposals on a timeline is the most consequential thing that can be done with them.

The synaptic proposal is **amyloid-gated**. Its first step requires amyloid-β to deplete EphB2, so it cannot begin before there is enough soluble oligomer in the relevant compartment. It executes; it does not initiate.

The proteostatic proposal has the opposite time signature. Its two determinants are apolipoprotein E genotype, fixed at conception, and age, which is monotonic. Neither requires amyloid. A neuron carrying ApoE4 begins life with a smaller surface pool than a neuron carrying ApoE3, and the pool declines with age in both; the threshold for filament formation is crossed when the declining pool meets the genotype-set requirement.

That is a candidate mechanism for tau pathology arising before, and independently of, amyloid — which is what the human autopsy record has appeared to demand since pre-tangles were documented in the locus coeruleus of young adults with no cortical amyloid (Braak and Del Tredici, 2011; Braak et al., 2011). The qualification is that no one has looked at the locus coeruleus: surface proteasome abundance has not been measured in noradrenergic brainstem neurons at any age. It is an attractive candidate for early failure — very long, thinly myelinated, extensively branched projections impose heavy demands on synthesis and delivery — but that is a plausibility argument. The measurement is straightforward and is item four in Chapter 30.

Two phenomena usually treated as puzzles become expectations on this model. The poor correspondence between amyloid burden and tangle burden across individuals is expected if the two lesions are governed by different variables. And the strong dependence of tau pathology on neuronal activity — activity stimulates physiological tau release (Pooler et al., 2013) and enhances tau propagation and pathology in vivo (Wu et al., 2016) — is doubly implicated, because activity is

also what drives the nascent synthesis the complex exists to trim. A neuron that fires more makes more tau and must dispose of more tau.

10.5 Where it is weak

Four objections stand at full strength. The filament identity is not structurally proven: negative-stain electron microscopy resolves helical periodicity but cannot establish the Alzheimer fold, which is the modern definition of a paired helical filament (Fitzpatrick et al., 2017). The structural basis of membrane association is unresolved — the 20S proteasome is a hydrophilic barrel with no obvious means of embedding in a bilayer, and how it is held with its chamber facing outward is unexplained; this is the most frequently voiced reservation about the whole concept and it is fair. It is not known whether the disease-relevant variable is catalytic activity or membrane localisation, and the two perturbations used are treated as equivalent when they may not be. And independent replication is outstanding: the concept has been extended in a second species by a partly independent group (He et al., 2023), which is meaningful; the tau result is months old.

10.6 What this station contributes

A **second disposal system at the plasma membrane**, distinct from the endosomal and lysosomal systems that every other programme here describes, and one whose failure produces the disease's *other* proteinopathy.

An **apolipoprotein-E-ordered, age-declining, neuron-autonomous quantitative mechanism for tau risk** — the shape the field has lacked.

And, as Chapter 19 argues, a protein whose reported properties are the properties of a raft resident, which nobody has yet asked whether it is.

11. Restraint: The System That Holds the Synapse Back

11.1 An accident, and a programme

Carla Shatz's programme began with a result nobody was looking for. Screening for genes regulated by neural activity in the developing visual system, the group found class I major histocompatibility complex molecules — immune molecules — expressed in healthy neurons and regulated by activity (Corriveau et al., 1998). Deleting them altered retinogeniculate refinement and lifted the normal limit on ocular-dominance plasticity (Huh et al., 2000; Datwani et al., 2009).

The receptor came next: PirB in mouse, LilrB2 in human, an inhibitory receptor of the immunoglobulin superfamily bearing immunoreceptor tyrosine-based inhibitory motifs. Deleting it or blocking it produced more spines, more plasticity, faster learning, and recovery from amblyopia in adult animals (Syken et al., 2006; Djurisic et al., 2013; Bochner et al., 2014; Vidal et al., 2016; Albarran et al., 2021). PirB is also the high-affinity receptor for the myelin-associated inhibitors of axonal regeneration (Atwal et al., 2008).

The signature claim is therefore a claim about direction: **every molecule the programme found is a restraint.** Delete any one and the mouse gets *more* of what learning requires. Nothing in the mechanism is broken in the disease. The brake is not faulty; it is pressed by the wrong hand.

11.2 Transduction, and a residue

The mechanism runs to the actin cytoskeleton. A dendritic spine is a bag of actin whose volume balances filament nucleation against filament severing, and the severing is done by cofilin, regulated by phosphorylation on a single residue, serine 3. Phosphorylated cofilin is inactive and the network is stable; dephosphorylated cofilin severs, and brief local activation is a normal part of plasticity. LIM-kinase phosphorylates the residue; the slingshot phosphatases remove the phosphate.

The claim is that ligand binding to the D1D2 domains of the receptor recruits phosphatase activity through its inhibitory motifs and drives cofilin dephosphorylation past the physiological range (Kim et al., 2013).

What happens beyond that range explains structural collapse rather than functional weakening, and it comes from outside the programme. At high concentrations of active cofilin the protein's behaviour inverts: instead of severing filaments it saturates and bundles them into a rigid, insoluble

cofilin–actin lattice. Neurodegenerative stimuli induce persistent rods of this kind within neurites, disrupting distal function (Minamide et al., 2000; Bamburg et al., 2021). The consequence for a spine is mechanical: a rod occupying a spine neck occludes it, cargo cannot pass, receptors and mitochondria cannot be delivered, and the spine is functionally disconnected before it is anatomically absent. Independently, synaptotoxicity in this disease has been shown to involve dysregulation of actin dynamics through cofilin phosphorylation (Rush et al., 2018) — corroboration of the node, not of the receptor.

That distinction should be carried as a rule for reading the programme: **the cofilin endpoint is well evidenced and multiply sourced; the attribution of that endpoint to LilrB2 rests on the programme's own work and on one external replication.**

11.3 The newer arm is the better-evidenced arm

The programme's 2013 claim was that amyloid-β oligomers are a ligand for this receptor, and that claim is contested. In a standardised head-to-head comparison of fifteen reported amyloid receptors, LilrB2 bound *synthetic* synaptotoxic assemblies but showed no detectable binding to human-Alzheimer-brain-derived oligomers, while cellular prion protein bound strongly (Smith et al., 2019). That result is seven years old and unanswered.

The 2025 result is a different matter. C4d — a covalently tethered product of complement activation — binds LilrB2 with an affinity near three nanomolar, sits on roughly thirty per cent of human excitatory synapses, is four-fold elevated in Alzheimer cortex, and, delivered by minipump into normal adult cortex for four days, strips spines; the effect is abolished in PirB-null animals (Brott et al., 2025).

That is a **neuron-intrinsic pruning arm requiring no microglion**, which is precisely what the complement literature could not supply. The classical account holds that complement tags synapses and microglia remove them (Stevens et al., 2007; Hong et al., 2016). The new result establishes a second effector limb of the same cascade acting directly on the neuron, at the same receptor site the physiological ligand uses. Both limbs are fed by one upstream event, and the relative contribution has never been measured. The prediction that separates them is straightforward: microglial depletion or CR3 blockade should abolish the glial arm and leave the neuronal arm intact, so a C4d minipump into a microglia-depleted cortex would settle it in a fortnight.

The newer arm of this programme is the better-evidenced arm, and it is not an amyloid arm.

11.4 One receptor, several addresses

Between 2022 and 2026, laboratories with no connection to the programme placed the same receptor in three places it never claimed and identified an endogenous molecule that blocks it.

Table 5 — Where the receptor has been placed, and with what confidence

Location	Function reported	Consequence of blocking	Confidence
Cortical / hippocampal pyramidal neuron	Restrains spine density, plasticity and LTD; transduces C4d and (contested) amyloid to cofilin	More spines, more plasticity; spine loss prevented	High for the physiological role; contested for the amyloid ligand
Microglion	Inhibits TREM2 signalling on co-ligation by a shared ligand	Restored TREM2 signalling, phagocytosis, migration	Moderate — one laboratory, strong design
Astrocyte	Suppresses EAAT1/2 via mTOR	Restored glutamate uptake, less neuronal death	Low to moderate — single study, one model
Golgi (cleaved cytoplasmic fragment)	Binds the GAT domain of GGA3 and jams retrograde traffic	Restored Golgi and lysosomal function	Low — single study, not replicated
Axon	Mediates myelin-inhibitor signalling	Partial release from myelin inhibition	High

The microglial address matters most for this paper. LILRB2 is expressed with TREM2 on human microglia, and co-ligation of the two by a shared ligand — amyloid oligomers or phosphatidylserine — significantly inhibits TREM2 signalling; antagonist antibodies rescue phagocytosis, migration and cytokine responses in human microglia, and increase plaque phagocytosis in grafted 5XFAD mice (Zhao et al., 2022). Given that loss-of-function variation in *TREM2* is among the largest genetic risk factors for this disease, a receptor that pharmacologically suppresses TREM2 signalling is a significant object; and a single antagonist would act at two places at once, protecting the spine from a disassembly instruction while licensing the microglion to clear.

The intracellular address is the most surprising. PirB is proteolytically cleaved on amyloid exposure in patients and models, and the resulting C-terminal fragment accumulates in the Golgi by retrograde transport, binds the GAT domain of GGA3, disrupts Golgi transport, impairs lysosomal maturation and compromises anterograde synaptic-vesicle transport; blocking the cleavage restored Golgi function, reduced plaque burden and tau phosphorylation, and rescued memory (Han et al., 2026). If it replicates, the receptor has a second, non-canonical mode in which its tail becomes an intracellular saboteur of the trafficking system — and blocking the ligand and blocking the *cleavage*

become different interventions, the second possibly mattering more. Chapter 21 develops this.

The endogenous antagonist supplies the best external corroboration the amyloid arm has. LOTUS inhibits amyloid binding to PirB and to human LilrB2; in neurons from LOTUS-overexpressing mice, amyloid-induced cofilin dephosphorylation, PSD-95 loss and spine loss were all suppressed (Kawaguchi et al., 2022). An independent laboratory, using a natural competitive antagonist rather than a knockout, reproduced the specific mechanistic chain.

11.5 A receptor that explains too much

Two readings of Table 5 are possible and honesty requires stating both. The generous reading is that an inhibitory receptor family evolved to hold immune effectors in check has been recruited by several brain cell types to hold *their* effectors in check, and one antagonist would release all of them. The sceptical reading is that a receptor which appears in every cell type, binds every ligand tried, and whose blockade improves every outcome measured is a receptor whose literature is running ahead of its evidence.

This paper's position is that the neuronal and microglial addresses are well enough evidenced to build on, and that the astrocytic and intracellular addresses are interesting and unreplicated.

The therapeutic double edge should also be stated: every blockade result in this programme is an *increase* in plasticity and an impairment of long-term depression. Nobody has measured the cost of removing a brake over years. Anti-LILRB2 antagonists have completed phase 1/2 in oncology, but as peripherally targeted myeloid agents with no assumed central exposure.

11.6 What this station contributes

The **restraint frame**: the disease is delivered *through* the machinery that limits plasticity, at the same sites the physiological ligand uses, which is a different claim from damage and has different therapeutic implications.

The **only neuron-intrinsic, microglia-independent synaptic-elimination mechanism in the set**, with human-tissue evidence.

And the **shared effector** — serine 3 of cofilin — which Chapter 18 shows is written to by three of the nine programmes with two different signs.

12. Instruction: Amyloid-β as a Signal of Competition

12.1 A theory about what the peptide is for

Zhen Huang's account is the only one of the nine that is primarily a **function claim**, and that is its strength. Its proposition is that amyloid-β is a competition signal — the molecule an axon secretes to protect itself and to mark its rivals — and that Alzheimer's disease is the failure of the protective half.

The grammar was borrowed from bacteriology. Nisin, a lantibiotic, combines quorum sensing, an immunity protein that protects the producer, and conformational duality, so that one gradient gives protection near the source and killing further out. Amyloid-β has a comparable profile — trophic at 100 to 200 picomolar (Puzzo et al., 2008; Gulisano et al., 2019), toxic as an oligomer at high nanomolar concentrations (Walsh et al., 2002; Shankar et al., 2008) — and is itself an antimicrobial peptide (Kumar et al., 2016; Moir et al., 2018). Disease follows when aggregation depletes the monomer, microglia are disinhibited, cytokines rise, and tau pathology follows.

12.2 The strongest leg: loss-of-function genetics

The part of this programme that is not in dispute is the developmental genetics, and it is unusually clean because it is done in normal animals rather than in models of disease.

Remove the amyloid precursor protein and the axon that should have lost the competition survives and expands. This has been shown in the superior colliculus, in whisker-plucking experiments using *cell-autonomous sparse deletion* — which is the design that distinguishes a competition phenotype from a general growth phenotype (Marik et al., 2016) — and at the neuromuscular junction in animals lacking both the precursor protein and its paralogue (Wang P. et al., 2005). Remove PirB and the same thing happens, with a bidirectional test available on one readout (Syken et al., 2006; Kim et al., 2013).

This is a claim about what these molecules do in a healthy animal, and it is supported by the standard of evidence appropriate to such a claim.

12.3 The 2024 experiment, and what it establishes

The theory's most exposed claim — that monomeric amyloid-β restrains glial inflammatory activity — acquired a candidate mechanism (Kwon et al., 2024). Entering through a developmental phenotype rather than through amyloid, the study found that deletion of a G-protein chaperone from microglia produced hyper-responsive microglia and, with an immune stimulus, cortical ectopia through excessive basement-membrane degradation; that deleting the amyloid precursor protein from microglia produced a parallel result; that monomeric amyloid-β40 potently suppressed cytokine secretion from wild-type microglia; and that this suppression was abolished in precursor-protein-null microglia but preserved in paralogue-null microglia, establishing specificity. The effector was named: matrix metalloproteinase 9, elevated in mutants, with both broad-spectrum and selective inhibitors reducing the phenotype.

Two features make this consequential. It makes the precursor protein a receptor for its own product, which revives a dormant orphan result — the report that the precursor protein complexes with the brain GTP-binding protein Go (Nishimoto et al., 1993), never comfortably assimilated because a G-protein coupling with no ligand is hard to build on. And it names a protease as the executioner, supplying the step most inflammation-centred accounts lack, in which a microglial state change becomes structural damage.

What it does not show is that monomeric amyloid-β activates this pathway in a living brain; the in vivo phenotype is a chaperone phenotype and a precursor-protein phenotype, and the authors concede the point explicitly in the published review record.

12.4 The concentration axis does not hold

A difficulty that the review process did not raise, and that bears on the theory rather than on the paper, deserves statement because it changes what the framework should claim.

The concentrations at which monomeric amyloid-β40 suppressed microglial cytokines were 50, 200 and 500 nanomolar, with significant suppression at the lowest tested. That lowest concentration is the bottom of the *high*-concentration band in the theory's own tabulation — the band in which amyloid-β begins to reduce the frequency of miniature synaptic currents. The trophic neuronal effects occur at 100 to 200 picomolar. The two protective actions the theory requires are therefore separated by a factor of roughly 250 to 500, and no concentration below 50 nanomolar was tested.

The bacteriocin analogy works because in bacteria **concentration drives conformation**: nisin is monomeric near the producer and oligomerises as local concentration rises, so one gradient generates two opposite activities at two distances from one source. In the amyloid case as now assembled, the variable that separates protective from destructive is **conformation**, and conformation is held constant experimentally by the preparation method. That is entirely compatible with the observations — different receptors, different affinities, same ligand — but it is

not the nisin architecture, and the spatial-gradient model loses its support. The framework would be stronger if it dropped the concentration axis, argued the conformational one directly, and let a different mechanism carry selectivity.

12.5 What arrived from elsewhere, for and against

For. An independent programme reached monomer depletion from human cerebrospinal fluid: high soluble amyloid-β42 is associated with normal cognition in individuals with brain amyloidosis (Sturchio et al., 2021) and predicts normal cognition in amyloid-positive carriers of causative mutations (Sturchio et al., 2022), with the position developed as a proposal to *restore* the peptide (Espay et al., 2023; Espay et al., 2025). The trial record has the shape this predicts: solanezumab, which binds monomer, was null in preclinical disease (Sperling et al., 2023); crenezumab, which binds monomer and oligomer, was null in the Colombian autosomal-dominant cohort (Tariot et al., 2026); lecanemab and donanemab, which are relatively monomer-sparing, are positive (van Dyck et al., 2023; Sims et al., 2023). And a TREM2 agonist antibody with confirmed target engagement was null (Mummery et al., 2026; Colonna and Holtzman, 2025), which constrains the simple version of "restore microglial function".

Against. The exemplar receptor may not be amyloid's: C4d is a nanomolar LilrB2 ligand, rises in disease, and drives PirB-dependent spine loss (Brott et al., 2025). A sign problem runs through the cell-competition literature: culling less fit neurons is *protective* in the fly amyloid model (Coelho et al., 2018), and forcing competition worsens outcome (Costa-Rodrigues et al., 2025). And the human evidence on engulfment tags the removed synapse with **tau oligomers** (Taddei et al., 2023) and with exposed **phosphatidylserine** bridged by MFG-E8 (Tzioras et al., 2023), not with amyloid-β. Finally, the tumour necrosis factor that mediates homeostatic synaptic scaling is astrocytic rather than microglial (Heir et al., 2024), which is a gap in the middle of the disease model.

12.6 The best mechanism in the framework is not the one it leads with

The 2024 review proposes a phosphatidylserine loop that is more attractive than the concentration gradient and is the framework's best answer to selectivity. Phosphatidylserine is externalised as an eat-me signal, its exposure at synapses is developmentally regulated and required for microglial pruning (Scott-Hewitt et al., 2020); separately, phosphatidylserine-like lipids in the outer leaflet accelerate amyloid-β aggregation, and phosphatidylserine incorporated into an outer leaflet triggers rapid oligomerisation at subnanomolar concentrations. The loop: a weakening synapse externalises phosphatidylserine, local phosphatidylserine nucleates oligomerisation *at that membrane*, the oligomer damages the membrane and amplifies the eat-me signal, and receptors that read phosphatidylserine — complement, the TAM kinases, TREM2 — are engaged.

This is self-amplifying, spatially local and selectivity-generating, and it needs no concentration gradient. It is at present an assembly of separately demonstrated components rather than a

demonstrated loop; the critical experiment — showing that blocking phosphatidylserine-nucleated oligomerisation at a synapse spares that synapse from engulfment — has not been done.

It also, and this is the reason it appears in a paper about membranes, makes the *lipid composition of the synaptic membrane* the variable that decides which synapse is removed. That places the selectivity of synapse loss on the same axis as the five lipid programmes in Chapters 4 to 9.

12.7 What this station contributes

The **only function claim in the set**, and with it the only account of why a nervous system would make this peptide at all.

The durable output, stated in 2020 before the evidence that now supports it: **do not remove the monomer.**

And the **phosphatidylserine loop**, which supplies the missing link between membrane lipid composition and the selection of which synapse is eliminated.

Part Three — The Shared Architecture

The itinerary the nine programmes divide between them; the single structural feature they share; where that feature fits badly; and the two clocks on which the mechanisms run.

13. The Itinerary: One Circuit, Nine Windows

13.1 The claim of this chapter

The nine programmes appear to concern nine different things: a sterol, an aldehyde, a proteolytic fragment, a sorting complex, a retained peptide, a scaffolding protein, a surface protease, an inhibitory receptor and a competition signal. They are, in fact, nine positions along one circuit, and the circuit is the itinerary of a lipid and a membrane protein through the compartments of a single cortical neuron.

This chapter is close to bookkeeping. It is set out first because the arguments that follow depend on the geography being clear, and because seeing the geography makes several previously invisible adjacencies obvious.

13.2 The circuit, stated as a sequence

An astrocyte synthesises cholesterol and packages it, with phospholipid, into an apolipoprotein E particle. The particle is released and binds a lipoprotein receptor on a neuron — LRP1, the low-density-lipoprotein receptor, ApoER2. The complex is internalised. In the acidifying early endosome the ligand dissociates, the lipid proceeds toward the lysosome for processing, and the receptor is retrieved and returned to the surface by retromer-dependent recycling with a cargo-selective adaptor.

Delivered cholesterol reaches the plasma membrane, where it partitions into ordered domains. Those domains are the platform on which trophic and glutamatergic receptors are held and on which their signals are transduced, and they are also where the amyloid precursor protein is sorted between its two cleavage routes: in disordered membrane it meets ADAM10 and is cut non-amyloidogenically; internalised into cholesterol-rich endosomes it meets BACE1, active at acidic pH, and is cut to C99.

C99 is trafficked to the endoplasmic reticulum, where its cholesterol-binding transmembrane domain clusters sterol into a raft-like contact with mitochondria. On that contact, cholesterol-handling enzymes are activated and the excess is disposed of. γ-Secretase — resident at the same contact — cuts C99, and the platform disperses.

Meanwhile, in the distal compartments, the products of the amyloidogenic route are sorted. A fraction is exported and degraded at the cell surface by neprilysin in a manner coupled to synaptic activity. A fraction is retained in multivesicular bodies within the synaptic terminal. The compartments that hold it are the compartments that recycle glutamate receptors.

At the surface itself, two further systems operate on the same membrane. An uncapped proteasome, oriented outward, degrades a large fraction of nascent polypeptide as it is made, tau among it, and releases peptides that feed back on NMDA-receptor signalling. And an inhibitory immune receptor reads the local extracellular environment — class I MHC in health, complement fragments and possibly amyloid assemblies in disease — and writes its output to the phosphorylation state of cofilin, and thus to the actin skeleton of the spine.

Finally, the extracellular pool of amyloid-β itself, in whichever conformation the local lipid environment favours, acts as a signal: restraining microglial inflammatory output as a monomer, marking membranes for elimination as an oligomer, and participating in the decision about which synapse survives a competition.

13.3 The map

Table 6 — The circuit, and who studies each step

Step	What happens	Programme	Species that fails
1. Synthesis and packaging	Astrocyte makes cholesterol; apolipoprotein E particle assembled and released	Rappoport	Delivered cholesterol
2. Transit	Peroxidisable polyunsaturated lipid carried on the particle; cysteine-dependent shielding	Ramsden	Peroxidised lipid; adducted apolipoprotein E
3. Binding	Lysine-rich ligand motif engages acidic LA modules of ApoER2/LRP1	Ramsden	The lysine interface
4. Acid release	Endosomal acidification dissociates ligand from receptor	Ramsden; Nixon (external)	The bond, or the pH
5. Retrieval	Retromer with SORLA/VPS26b returns receptor and cargo to the surface	Small	The recovery arm
6. Raft assembly	Cholesterol partitions into ordered plasma-membrane domains	Rappoport; Head	The platform

Step	What happens	Programme	Species that fails
7. Reception	Trk, NMDA, AMPA receptors transduce only within the domain	Head	Signal transduction
8. Sorting of the precursor	Domain occupancy determines α- versus β-cleavage	Area-Gómez; Rappoport	Route selection
9. Fragment clearance	γ-Secretase at the ER–mitochondrial contact cuts C99; the platform disperses	Area-Gómez	Termination of the signal
10. Peptide disposal	Retained versus exported amyloid-β; surface neprilysin coupled to activity	Gouras	The activity–clearance coupling
11. Nascent-protein disposal	Outward-facing membrane proteasome trims newly made protein, tau included	Margolis	Co-translational disposal
12. Surface reading	Inhibitory receptor reads the extracellular field, writes to cofilin	Shatz and Brott	Release of restraint
13. Adjudication	Amyloid-β conformation decides which synapse is kept	Huang	Resolution of the competition

Three observations follow immediately from the table and are developed in the chapters named.

The stations are contiguous, not overlapping. No two programmes are studying the same step. Where two appear on one row — Rappoport and Head at raft assembly and reception, Ramsden and Nixon at acid release — they are studying different determinants of the same step, and Part Five treats those as the places where the sign disputes live.

The circuit closes. Step 9 disperses the platform that step 6 built; step 8 feeds back on step 1 through the effect of soluble APPα on cholesterol synthesis; step 13 feeds back on step 6 through the effect of amyloid conformation on membrane integrity. This is not a linear pathway; it is a control system with at least three nested loops, which is why interventions on it have signs rather than magnitudes.

Every step has a return. Binding has release. Delivery has retrieval. Assembly has dispersal. Synthesis has disposal. Engagement has withdrawal. Destabilisation has restabilisation. The next chapter is about which half of each pair fails.

13.4 What the map explains that the parts do not

Two things, and they are the reason the bookkeeping is worth doing.

It explains the recurrence of the same molecules in unrelated papers. Apolipoprotein E appears in six of the nine programmes, in six different roles: cholesterol carrier, peroxidisable cargo, receptor ligand, determinant of endolysosomal capacity, determinant of surface proteasome abundance, and modifier of microglial lipid state. That is not six coincidences; it is one molecule traversing a circuit and being measured at six stations. The same is true of the β-carboxy-terminal fragment, which appears at station 8 as a routing product, at station 9 as a cholesterol signal, at station 4 as an inhibitor of the vacuolar ATPase, and at station 5 as the agent that recruits APPL1 to rab5-positive endosomes.

It explains why single-target interventions on this circuit have produced small and sometimes inverted effects. A control system with three nested loops responds to perturbation at one node by compensating at another. Suppressing β-secretase reduces the input to station 9 and worsens cognition. Suppressing γ-secretase blocks the disposal step at station 9 and worsens cognition faster. Removing extracellular amyloid at station 13 causes cells with an internal load to increase production at station 8. These are not anomalies to be explained away; they are what a stalled control loop does when it is pushed.

14. The Failure of Return

14.1 The thesis

Every one of the nine programmes identifies a physiological cycle. Every cycle has a forward leg — the step that makes, delivers, cuts, engages, destabilises or instructs — and a return leg — the step that recovers, releases, clears, disposes, resolves or restabilises. **In all nine, the reported lesion is on the return leg.**

That is the central claim of this paper. It is a claim about the shape of nine independent bodies of evidence, and it is testable in the ordinary sense that it could be shown false by demonstrating that any one of the nine programmes' lesions is, on its own evidence, a forward-leg lesion.

14.2 The nine, in their own words

Table 7 — The forward and return legs of each cycle

Programme	The cycle	Forward leg (intact or increased)	Return leg (the reported lesion)	The programme's own phrase
Rappoport	Candidate generation → competition resolution	Candidate generation runs continuously	Resolution never begins; the structure is never restabilised	"Chronic candidate generation"
Ramsden	Ligand binding → acid release → receptor recycling	Binding occurs; internalisation occurs	Release fails; the receptor is not recovered	"ApoER2 depletion by covalent trapping"
Area-Gómez	Cholesterol signal raised → correction executed → signal cleared	C99 is produced; the contact assembles; the correction runs	γ-Secretase cleavage fails; the signal is never cleared	"A homeostatic correction left switched on"
Small	Endocytosis → sorting → retrieval to the surface	Endocytosis is normal or accelerated	Retromer-dependent recovery jams; residency time rises	"The traffic jam"
Gouras	Production → export → surface degradation	Production continues; is up-regulated in loaded cells	Secretion falls; surface neprilysin is lost	"Retention, not overproduction"

Programme	The cycle	Forward leg (intact or increased)	Return leg (the reported lesion)	The programme's own phrase
Head	Platform assembly → signal reception → platform maintenance	Ligand present; receptors expressed	The platform is not maintained; receptors are not raft-localised	"Deaf, not starved"
Margolis	Nascent synthesis → co-translational trimming	Activity-driven synthesis continues	Surface proteasome abundance falls; trimming fails	"Failure of local disposal"
Shatz and Brott	Brake applied → brake released	Ligand engagement increases (C4d four-fold elevated)	The restraint is never lifted; cofilin is not re-phosphorylated	"The brake pressed by the wrong hand"
Huang	Competition opened → competition resolved	Candidate axons compete; the signal is secreted	Monomer is depleted into aggregate; the competition is never resolved	"Do not remove the monomer"

Five of the nine state the return-leg lesion in their own summary vocabulary — retention rather than overproduction, the traffic jam, the correction left on, chronic generation, failure of local disposal. Two more state it structurally without naming it: a receptor that cannot be recycled, a brake that is not released. Two require the reading offered here: the platform account, whose lesion is maintenance rather than construction, and the competition account, whose lesion is resolution rather than initiation.

14.3 The three corollaries

Corollary one: the forward leg is not merely intact, it is often increased. This is the discriminating observation, and it is what makes the frame informative rather than a tautology.

Amyloid production is *up-regulated* in cells carrying an internal load, because the homeostat has detected a low external pool and increased synthesis to compensate (Roos et al., 2021). Contact-site function is *increased* in Alzheimer cells and tissue, because C99 keeps building the platform (Area-Gómez et al., 2012). C4d is *four-fold elevated* in Alzheimer cortex (Brott et al., 2025). Cholesterol synthesis *continues* in neurons that cannot incorporate it into rafts, so the cell accumulates lipid it cannot use. Precursor-protein internalisation is *up-regulated* in aged neurons (Burrinha et al., 2021). Neurons carrying elevated amyloid become *hyperactive* (Martinsson et al., 2022; Busche et al., 2008).

In a system whose lesion is over-production, one expects to find the products elevated and the machinery normal. In a system whose lesion is failed return, one expects to find the products elevated, the return machinery reduced, *and the forward machinery up-regulated in compensation*. The third of those is the signature, and it is present at six of the thirteen stations.

Corollary two: the sign of the correct intervention inverts. If the forward leg is compensating for the return leg, then suppressing the forward leg removes a compensation and worsens the patient. This is not a prediction; it is a description of the trial record, and Chapter 28 sets it out in detail.

Corollary three: a partial failure is worse than a complete one. This is Rappoport's argument generalised. If the cycle stopped, the cell would stop paying its costs. Because the cycle stalls rather than stopping, the cell keeps paying the forward cost — the energetic cost of synthesis, the structural cost of destabilisation, the signalling cost of a brake held down — for a return that never arrives. Chronicity is the mechanism by which a modest quantitative deficit becomes a progressive disease, and it explains a twenty-year preclinical phase without requiring any step to be slow.

14.4 Why this shape rather than another

It is worth asking why nine independent programmes would converge on the return leg, and there are three candidate answers with different implications.

The trivial answer is that return steps are simply more numerous or more complex than forward steps, so a random lesion is more likely to land on one. This is partly true — retrieval, release and disposal each involve more components than the corresponding forward step — but it does not explain the compensation signature, which requires that the forward step be *regulated in response to* the return step.

The energetic answer is that return steps are the ones that cost, and a cell under a lifelong energetic constraint will fail there first. Retrieval requires ATP-dependent coat assembly and motor transport; acidification requires a proton pump; proteasomal disposal requires synthesis and maintenance of the complex; restabilisation requires new membrane, new matrix and new myelin. Forward steps are frequently spontaneous or exergonic: aggregation, partitioning into an ordered domain, ligand binding, proteolysis. This is a real asymmetry and it predicts that the mechanisms should be sensitive to bioenergetic status, which they are.

The regulatory answer, which this paper prefers, is that return steps are where the *control* is. A cell does not regulate whether cholesterol partitions into an ordered domain — physics does that. It regulates retrieval, disposal, release and resolution, because those are the steps at which a decision is available. A disease of dysregulation must therefore appear at return steps, because the forward steps are not regulated and cannot be dysregulated.

That reading has a consequence worth stating: **the nine programmes did not find nine lesions in a control system; they found the control system, and it is on the return leg by construction.** What they have collectively established is not that nine things fail but that the neuron's principal regulatory apparatus is a set of recovery mechanisms, and that this disease is a

disease of that apparatus.

14.5 The relationship to existing framings

Three prior formulations should be acknowledged, because the thesis here is a generalisation of them rather than a novelty.

The **clearance framing** is the closest, and it is widely used: the disease is a failure to clear amyloid rather than to produce it. The return-leg thesis differs in three ways. It applies to species other than amyloid — cholesterol, receptors, nascent tau, phosphorylation states. It includes steps that are not clearance at all, such as ligand release and cytoskeletal restabilisation. And it requires the compensation signature, which the clearance framing does not.

The **homeostasis framing** — that the lesion is loss of a control loop rather than presence of a substance — is stated explicitly by two of the nine and is the closest in spirit. This paper's contribution is to show that it is true of all nine and that the loops share a topology.

The **hub-and-spoke framing** locates a common downstream node with feedback from its spokes. It is an architectural claim about *which* mechanism is central; the return-leg thesis is a structural claim about *what kind* of step fails, and the two are compatible. Notably, the hub in that model — endosomal recycling — is a return step.

15. Where the Frame Does Not Fit

15.1 Why this chapter exists

A frame that fits everything explains nothing, and this paper has already levelled that charge at two of the nine programmes. It would be indefensible to make the same move at a higher level and then not test it. Four places where the return-leg thesis fits badly are set out here, in descending order of seriousness, with what would be required to rescue or abandon the thesis in each case.

15.2 The complement arm is an addition, not a failure

The strongest objection. C4d is *elevated* four-fold in Alzheimer cortex, it binds LilrB2 at nanomolar affinity, and delivering it to a normal adult cortex for four days is sufficient to strip spines (Brott et al., 2025). That is a forward-leg excess: a signal is added, and the addition alone produces the lesion. No failure of return is required, and the experiment was designed to show precisely that.

Three responses are available and only the third is honest.

The weak response is that the complement cascade is itself a system whose activation is normally terminated by regulators, so elevated C4d reflects a failure of complement regulation. This is true and it is not evidence; complement regulator status in Alzheimer cortex has not been measured alongside C4d in the same tissue.

The second response is that the receptor's output — cofilin dephosphorylation — is itself a normal, transient signal whose *reversal* is the return step, and that the pathological state is a failure to re-phosphorylate rather than an excess of dephosphorylation. This has the merit of being testable: it predicts that LIM-kinase activity, or slingshot phosphatase activity, is altered in the same tissue, and that the persistent cofilin–actin rods that characterise the degenerating neurite are a consequence of failed reversal rather than of sustained input.

The honest response is that **the C4d arm is a forward-leg lesion and the thesis does not cover it.** The right statement of the thesis is therefore narrower than "all nine": eight of the nine programmes locate their lesion on a return leg, and the ninth — the newer and better-evidenced arm of the restraint programme — describes an added signal acting on an intact system. That the exception is the most recently discovered mechanism, and the one with the cleanest interventional demonstration in normal tissue, should be a caution against over-reading the pattern.

15.3 The plaque is a product, and products are forward-leg

A second difficulty. Several of the nine treat the extracellular deposit as the output of a compensating cell — plaque as the signature of a solved problem, in Rappoport's formulation, or as the residue of ruptured neurons in the inside-out reading. On the return-leg frame, deposits are epiphenomenal, and the frame therefore inherits the awkward obligation of explaining why removing them produces any clinical benefit at all.

It does produce benefit. Lecanemab slowed decline on the primary measure by roughly a quarter over eighteen months with substantial amyloid removal (van Dyck et al., 2023); donanemab produced a comparable result with larger effects at lower baseline tau (Sims et al., 2023). A frame in which the deposit is a by-product must say why.

The available answer is that the deposit is a *reservoir* that feeds the return-leg failure rather than a cause in itself: it sequesters monomer, so the resolution signal is depleted; it sustains a local complement activation, so the brake stays pressed; it maintains a microglial state that consumes phagocytic capacity. Removing the reservoir partially relieves each. That answer predicts the observed shape of the trial results — partial benefit, larger when tau is low — but it is post hoc, and it should be labelled as an interpretation rather than a prediction.

15.4 Two programmes locate a genuine gain of function

The disulfide framework and the intraneuronal-amyloid framework both contain a step that is a gain rather than a loss: an aldehyde does damage it would not otherwise do, and a retained peptide inhibits a proteasome. Neither is a failure of return in the sense used here; both are novel activities.

The rescue is available and is worth stating because it is the same rescue in both cases. The novel activity is *enabled* by a failure of return — the aldehyde reaches the interface because the peroxidised lipid was not effluxed, and the peptide inhibits the proteasome because it was not exported. In both cases the gain of function is downstream of a retention. But the rescue costs something: it means the thesis is a claim about the *proximate regulatory step*, not about every step in the chain, and chains of this kind will always contain gains of function once the retention has occurred.

15.5 Some return steps are not obviously regulated

The regulatory argument of §14.4 holds that return steps fail because return steps are where the control is. Two of the return steps in Table 6 are not obviously under regulation. Endosomal acid release is a physical-chemical event; the dispersal of a lipid domain after its organising peptide is cut is a thermodynamic relaxation.

This weakens the regulatory reading without touching the descriptive one. It suggests that the thesis is really two claims that happen to coincide: that the failing steps are recovery steps (descriptive, and well supported), and that recovery steps fail because they are the regulated ones

(explanatory, and true of most but not all of them).

15.6 What the chapter changes

The thesis is retained in a narrower form, stated here in the version the rest of the paper will use:

> *In eight of the nine programmes reviewed, the reported lesion is on the recovery step of a physiological cycle rather than on the productive step, and at six of the thirteen stations of the circuit the productive step is measurably up-regulated in compensation. The ninth programme's principal recent finding describes an added extracellular signal acting on an intact system, and is not covered by the frame.*

That version is falsifiable, and Chapter 32 gives the results that would falsify it.

16. One Surface at Three Addresses

16.1 The observation

The nine programmes name three compartments that are, on inspection, the same kind of object.

The plasma-membrane lipid raft. Cholesterol- and sphingolipid-enriched, thicker and less fluid than the surrounding bilayer, detergent-resistant, and defined operationally by buoyancy in a density gradient after carbonate lysis. It is the compartment of Chapters 4 and 9.

The mitochondria-associated endoplasmic reticulum membrane. Also cholesterol- and sphingolipid-enriched, also detergent-resistant, also isolated as a buoyant fraction, and the site of cholesterol esterification and phospholipid synthesis (Vance, 1990; Area-Gómez, 2014). It is the compartment of Chapter 6, and its assembly is nucleated by a cholesterol-binding peptide (Montesinos et al., 2020).

The synaptic endosome and the multivesicular body derived from it. Cholesterol-rich, the site at which BACE1 operates, and the compartment from which glutamate receptors are recycled. It is the compartment of Chapters 7 and 8.

These are not three unrelated structures that happen to share a lipid. They are three instances of one physical phenomenon — the lateral phase separation of cholesterol and saturated sphingolipid into an ordered domain — occurring at three membranes of one cell. The proteins that partition into them do so by the same rules: palmitoylation, a matched transmembrane length, and cholesterol-binding motifs.

16.2 Why the identity matters

Three consequences, each of which converts an isolated observation into a shared one.

A shared supply constrains all three. If the neuron's cholesterol comes from astrocytes and the delivery route has no redundancy (Chapter 4), then failure of that route degrades ordered-domain formation at *every* membrane of the cell, not only at the surface. That is a specific prediction that neither the raft programmes nor the contact programme have made, and it is measurable: in neurons deprived of apolipoprotein E-borne cholesterol, contact-site function and detergent-resistant plasma-membrane fraction should fall together.

A shared physics gives a shared read-out. The claim that bilayer thickness sets γ-secretase processivity (Chapter 6) is a claim about a physical property that applies to all ordered

domains. If it is right at the contact, it is right at the surface, and the sorting of the precursor protein between its cleavage routes at the plasma membrane should show the same lipid dependence. Dawkins and colleagues' demonstration that lipid remodelling modulates processivity (Dawkins et al., 2023) does not distinguish the compartments.

Damage is concentrated where the chemistry is. The most useful single piece of human evidence for this chapter is a subcellular fractionation: oxidative damage in Alzheimer brain is greater, and antioxidant enzyme levels lower, *in lipid rafts* than in non-raft membrane, and *APOE* ε*4* carriers had lower raft yield with greater membrane oxidation (Thorwald et al., 2025). Lipid peroxidation is not uniformly distributed across the neuronal membrane; it is concentrated in the compartment where the ApoER2–Dab1 complex signals, where the secretases work, and where the receptors that the disease withdraws are held.

16.3 The molecules that appear in all three

A short list, offered as the evidential basis for the identity claim rather than as a rhetorical flourish.

- **Cholesterol** — the ordering lipid at all three, rate-limited by the same astrocytic supply.
- **The amyloid precursor protein and C99** — sorted at the surface, cut in the endosome, and cleared at the contact; its transmembrane domain binds cholesterol and its cleavage geometry depends on the domain's thickness.
- **Presenilin / γ-secretase** — enriched at the contact (Area-Gómez et al., 2009), partitioning between caveolar and non-caveolar plasma membrane under caveolin-1 control (Kapoor et al., 2010).
- **BACE1** — active at acidic pH in the endosome, and its amyloidogenic output increased when it is forced into rafts by a lipid anchor (Cordy et al., 2003).
- **ACAT1 / SOAT1** — the cholesterol-esterifying enzyme, resident at the contact, and the enzyme through which the protective effect of statins on tau depends (van der Kant et al., 2019).
- **Caveolin-1** — the organiser of ordered domains at the plasma membrane, present also at mitochondria under stress (Fridolfsson et al., 2012).
- **Cellular prion protein** — a raft-resident glycosylphosphatidylinositol-anchored protein, the strongest-binding amyloid receptor in a standardised comparison (Smith et al., 2019), and the partner through which amyloid oligomers enter the neuron by a caveolin-1-dependent route (da Silva Correia et al., 2024).

16.4 The shared failure surface

Assembled, the case is stronger than any one programme states it.

Amyloidogenic processing occurs in ordered domains (Ehehalt et al., 2003), and forcing the enzyme into them increases it (Cordy et al., 2003). γ-Secretase partitions between domain types under caveolin control (Kapoor et al., 2010). Oligomers re-enter the neuron through a domain-resident route (da Silva Correia et al., 2024). A second proteinopathy — mutant TDP-43 — mislocalises to the same domains, and correcting the domain corrects the mislocalisation (Wang D. et al., 2026). Neurotrophin receptors require domain localisation to signal and lose it with age (Head et al., 2010; Egawa et al., 2016). Domain formation depends on astrocyte-supplied cholesterol, whose failure has been proposed as the primary lesion of sporadic disease (Rappoport, 2025). And oxidative damage in the human disease is concentrated in these domains (Thorwald et al., 2025).

Seven independent lines, five laboratories, three diseases, one compartment type. The proposition worth extracting is larger than any of the theories that supply it:

> *The ordered membrane microdomain is a shared failure surface. It is where the precursor protein is cut, where the product re-enters, where a second proteinopathy deposits, where trophic signal is received, where lipid oxidation concentrates, and where ageing takes its earliest measurable structural toll on a neuron. Its integrity is set by a supply chain with a single non-redundant step.*

Graded here as **moderate**: the individual components are established, the identity of the three compartments as instances of one physical phenomenon is standard membrane biophysics, but the claim that a single supply failure degrades all three coordinately has not been tested. Chapter 30 states the experiment.

16.5 The obligatory caution about rafts

The lipid raft has a difficult history and the caution belongs here rather than in a footnote. Detergent-resistant membrane is an operational fraction and not a structure observed in life; the size, lifetime and even existence of ordered domains in living cells were contested for a decade; and a literature that explains many things by appeal to an entity defined by a biochemical procedure invites scepticism.

Two developments make the appeal more defensible than it was. Direct imaging methods that do not depend on detergent — secondary-ion mass spectrometry of membrane lipid composition, super-resolution imaging of domain markers — now report ordered domains in living human neurons and show that they change with *APOE* genotype (Lee et al., 2021; Lee et al., 2025). And the contact-site literature has independently established that a cholesterol-ordered domain at the endoplasmic reticulum has a defined protein composition, a nucleating peptide, and a regulated

function in cholesterol homeostasis (Montesinos et al., 2020; Montesinos et al., 2024, preprint).

The entity is real. What remains contested is its size, its lifetime, and — as Chapter 25 sets out — the direction in which the major risk genotype moves it.

17. Two Clocks

17.1 The division

The nine mechanisms do not run on one timescale, and sorting them by their entry requirement produces a sharper division than sorting them by compartment.

Some mechanisms cannot begin until amyloid-β has accumulated to a level sufficient to act. Others require only age, genotype, or a lipid environment, and can begin at any time. The division is not a matter of degree: for each mechanism, one can ask whether its first step has amyloid-β as a necessary input, and the answer is usually unambiguous.

Table 8 — Two clocks

Mechanism	First step	Requires amyloid-β to have accumulated?	Earliest possible onset
Astrocyte cholesterol delivery fails; raft assembly stalls	Reduced delivery or uptake	No	Any age; accelerates with vascular and metabolic risk
Peroxidised lipid adducts the receptor interface	Lipid peroxidation on the particle	No	Any age; tracks oxidative load and *APOE* genotype
C99 is not cleared; the contact is up-regulated	Reduced γ-secretase throughput or raised substrate	No	Lifelong in mutation carriers; adult in sporadic disease
Retromer-dependent retrieval jams	Reduced retromer or SORLA function	No	Lifelong in *SORL1* variant carriers
Surface membrane proteasome declines	Age and *APOE* isoform	No	From early adulthood; monotonic
Membrane platform degrades; receptors leave the domain	Age; cholesterol availability	No	Middle age onward
C4d rises and engages the inhibitory receptor	Complement activation	No — rises in ageing, further in disease	Ageing, before deposition
Amyloid-β is retained in the synaptic endosome	A raised retained pool	Partly — requires the pool, not the deposit	Adult; precedes deposition

Mechanism	First step	Requires amyloid-β to have accumulated?	Earliest possible onset
Ephexin5 is not degraded because EphB2 is depleted	Amyloid-β depletes EphB2	**Yes**	After sufficient soluble oligomer
Amyloid-β oligomers engage the inhibitory receptor	Oligomer formation	**Yes**	After aggregation begins
Monomer depletion disinhibits microglia	Aggregation consumes monomer	**Yes**	After aggregation begins
The four routes from a loaded compartment	A loaded compartment	**Yes**, by definition	After the pool rises

17.2 What the division buys

The amyloid-independent set carries the human genetics. *APOE*, in the two mechanisms whose dependence on it is quantitative — endolysosomal capacity and surface proteasome abundance. *SORL1*, whose truncating variants occur almost exclusively in cases. *ABCA1* and *ABCA7*, cholesterol and phospholipid transporters implicated by rare-variant analysis (Holstege et al., 2022; Ralhan et al., 2026). *DAB1*, as an *APOE4*-conditional modifier (Bracher-Smith et al., 2022). And the two documented human resistance variants, both in ligands of the apolipoprotein E receptors (Arboleda-Velasquez et al., 2019; Lopera et al., 2023).

The amyloid-gated set carries none of these. Its genetic anchors are the autosomal-dominant mutations, which are mutations in the amyloid machinery and therefore cannot discriminate between the two clocks; and *ARHGEF15*, which is not an Alzheimer risk locus at all.

The amyloid-independent set is the earlier set, and it is a substrate rather than an event. Three of its mechanisms are explicitly monotonic with age: surface proteasome abundance, raft localisation of receptors, and complement activation. One is fixed at conception and expressed as a slowly widening gap: the apolipoprotein E effect, which produces no measurable defect in young neurons and a clear one in old ones (Nyberg et al., 2025). These are not events that happen; they are a capacity that declines.

The amyloid-gated set executes. Each of its mechanisms is fast once its input is available, each is demonstrated by intervention in a model, and each produces the synaptic phenotype that correlates with symptoms.

17.3 The reading this supports

The natural reading is a threshold model with two variables, and it is worth stating explicitly because it reconciles several observations that are usually treated as puzzles.

A neuron's capacity to execute the return legs of Table 7 declines from early adulthood, at a rate set by genotype, vascular and metabolic exposure, and oxidative load. That decline is the amyloid-independent clock, it is continuous, and it is not a disease. Superimposed on it is a set of amyloid-gated mechanisms that are sharply effective once engaged and that consume the remaining capacity rapidly.

On this reading the poor correspondence between amyloid burden and cognitive state across individuals is expected: burden reports the second clock, and outcome depends on the first. The existence of resilient individuals with high pathology is expected: their first clock ran slowly. The larger benefit of anti-amyloid antibodies at lower baseline tau is expected: tau burden is an index of how much of the first clock has already elapsed. And the failure of the first clock to be visible in any current biomarker is expected: every biomarker in clinical use measures the products of the second.

This is not a new model of Alzheimer's disease; it is close to the reserve-and-pathology framework the epidemiology has supported for two decades. What Table 8 adds is a mechanistic content for the first clock — nine named recovery steps, each measurable, most of them with a genetic anchor — where the reserve literature has had a statistical construct.

17.4 The one mechanism that spans both

The retention mechanism sits awkwardly and deliberately so. Its input is a raised intraneuronal pool, which requires amyloid-β but not deposition, and eight causally independent conditions raise it — three of which (ageing, apolipoprotein E4, reduced activity) belong to the first clock and are not themselves amyloid events.

That makes it the coupling between the clocks: the mechanism by which a slow decline in capacity converts into a fast amyloid-gated cascade. It is also the mechanism whose central quantity — the retained intraneuronal pool — has no clinical assay, which is why the coupling has never been measured in a person. Chapter 29 argues that this single missing measurement is the largest obstacle to testing the whole framework.

Part Four — The Integrations

Seven joins between programmes, each made at a named residue, molecule or compartment; each graded; each accompanied by the experiment that would refute it.

18. Serine 3: Three Programmes Writing to One Residue

18.1 The node

A dendritic spine is an actin structure whose volume is a balance between filament nucleation and filament severing. The severing is done by cofilin, and cofilin's activity is controlled by phosphorylation of a single residue, serine 3. Phosphorylated cofilin cannot bind actin and the network is stable; dephosphorylated cofilin severs, raising turnover. LIM-kinase 1 and 2 phosphorylate the residue, and are themselves activated by phosphorylation — threonine 508 in LIM-kinase 1. The slingshot phosphatases remove the phosphate from cofilin.

The response of a spine to cofilin activity is not monotonic, and this is essential to what follows. Brief, local cofilin activation is required for structural plasticity: without severing there is no turnover and no remodelling. At high concentrations of active cofilin the protein's behaviour inverts — instead of severing it saturates and bundles filaments into a rigid, insoluble cofilin–actin lattice. Persistent rods of this kind form within neurites under a range of neurodegenerative stimuli and disrupt distal function (Minamide et al., 2000; Bamburg and Bernstein, 2016; Bamburg et al., 2021). A rod occupying a spine neck occludes it: cargo cannot pass, receptors and mitochondria cannot be delivered, and the spine is functionally disconnected before it is anatomically absent.

So the node has a non-monotonic dose–response, with dysfunction at both extremes, and its physiological operating point is a narrow band around a moving set point.

18.2 Three inputs, from three programmes

Input one — the restraint programme, driving dephosphorylation. Ligand binding to the D1D2 domains of PirB/LilrB2 recruits phosphatase activity through the receptor's inhibitory motifs and drives cofilin dephosphorylation past the physiological range; PirB is required for the deleterious effect of amyloid oligomers on hippocampal potentiation, and cofilin signalling is enhanced in human Alzheimer cortex (Kim et al., 2013). The chain was independently reproduced with a natural competitive antagonist: LOTUS suppressed amyloid-induced cofilin dephosphorylation, PSD-95 loss and spine loss in the same experiment (Kawaguchi et al., 2022). And the node itself is corroborated from outside the programme: synaptotoxicity in this disease involves dysregulation of actin dynamics through cofilin phosphorylation (Rush et al., 2018).

Input two — the peroxidation programme, through LIM-kinase 1. Reelin, acting through ApoER2 and VLDLR, Disabled-1, Src-family kinases, phosphatidylinositol-3-kinase and Akt, activates LIM-kinase 1 and thereby induces phosphorylation of n-cofilin at serine 3, stabilising the actin cytoskeleton of neuronal processes (Chai et al., 2009). That is the same receptor, the same adaptor and the same phosphoinositide arm that the peroxidation programme maps in human tissue — and among the markers it reports co-accumulating in the perforant-path terminal zones in disease is **threonine-508-phosphorylated LIM-kinase 1**, alongside tyrosine-607-phosphorylated P85α, Dab1, reelin and the aggregated ligand-binding modules of ApoER2 (Ramsden et al., 2022; Ramsden et al., 2023).

Input three — the spine-brake programme, through RhoA. On its original reading, Ephexin5 restrains excitatory synapse formation through RhoA (Margolis et al., 2010), and the canonical route from RhoA runs through Rho-associated kinase to LIM-kinase and thence to cofilin. The revision complicates the sign rather than removing the input: Ephexin5 is a substrate switch gated at tyrosine 361, directing signalling toward RhoA when phosphorylated and toward Cdc42 when not, with the Cdc42 arm required for activity-driven spine growth (Petshow et al., 2025). Both arms terminate on the actin machinery.

18.3 The join

Three of the nine programmes write to one residue, and no two of the three literatures cite each other. The reelin–ApoER2 work and the LilrB2 work were published four years apart and have never appeared in the same paper; the Ephexin5 work and the LilrB2 work share a compartment and no citations.

Two receptor systems on the same neuron therefore converge on serine 3 of cofilin with opposite signs. Reelin, through ApoER2 and LIM-kinase 1, phosphorylates it and stabilises the network. Amyloid or C4d, through LilrB2, dephosphorylates it and destabilises the network. One system is a guardian of structure and the other an instruction to dismantle, and they are writing to the same address.

The consequence, if the antagonism is real, is specific and useful: **reelin signalling is a candidate endogenous buffer against receptor-driven spine collapse, and the loss of reelin signalling that accompanies apolipoprotein E4 carriage would lower the threshold at which a given ligand load produces structural damage.** That is a mechanistic proposal about why the major risk genotype makes spines fragile, and it does not require apolipoprotein E4 to do anything to amyloid.

It also gives the human resistance cases a shared address. The *RELN*-COLBOS variant is described as a gain of function with enhanced ability to activate Dab1 (Lopera et al., 2023); on this reading its carrier has a stronger stabilising input at serine 3. The *APOE3* Christchurch variant is a

poor receptor binder (Arboleda-Velasquez et al., 2019; Guo et al., 2025); on the rule proposed in Chapter 5 — apolipoprotein E is the ligand you want less of at this receptor and reelin the ligand you want more of — its carrier has less competitive occupancy at the same receptor.

18.4 The complication that makes it a research question

The direction of the tissue evidence does not obviously agree with the mechanism, and the disagreement is instructive rather than fatal.

The peroxidation programme reports *increased* threonine-508-phosphorylated LIM-kinase 1 in the affected terminal zones. More active LIM-kinase 1 should mean more phospho-cofilin and a more stable network. But the degenerating neurite is characterised by cofilin–actin rods, which form from *dephosphorylated*, active cofilin. On its face the two observations point in opposite directions.

Three readings are available and they are separable by experiment.

The sequestration reading. Much of the co-accumulation signal in that human tissue work lands in granulovacuolar degeneration bodies, which sequester many unrelated phosphoproteins. Accumulated phospho-LIM-kinase in a granule is not active LIM-kinase in a spine. The control is to stain unrelated phosphoproteins on the same sections; it has not been done.

The compartment reading. Rods form in neurites and at spine necks; the ApoER2–Dab1 signalling complex is recruited to ordered membrane domains through the phosphotyrosine-binding domain of Dab1 binding phosphatidylinositol 4,5-bisphosphate and the receptor tail simultaneously. The two measurements may report different pools of the same molecules in different subcellular locations, which bulk immunohistochemistry cannot separate.

The uncoupling reading. A chronically engaged pathway with an accumulated, phosphorylated proximal component and a downstream output running the wrong way is the signature of a break between the two — which, on the frame of Chapter 14, is exactly what a failed return step looks like. On this reading, phospho-LIM-kinase accumulates *because* it is no longer clearing, and the actin phenotype is set by unopposed phosphatase activity at the receptor.

The third reading is the one this paper favours, and it is graded **inference**.

18.5 The experiments

The primary test, and it is a two-experiment question. Co-manipulate reelin signalling and LilrB2 signalling in the same neurons and measure phospho-cofilin at serine 3. If the antagonism is real, reelin should raise the phosphorylation set point and blunt the C4d- and oligomer-driven fall; blocking ApoER2 with receptor-associated protein should exaggerate it. If they are independent, the effects should be additive with no interaction term. Nothing about this requires new reagents.

The tissue test. Array tomography or expansion microscopy on human cortex, quantifying phospho-cofilin, total cofilin, phospho-LIM-kinase 1 and LilrB2 at identified excitatory synapses, stratified by *APOE* genotype and by cognitive status at death. This distinguishes the sequestration and compartment readings and requires only tissue and antibodies.

The falsifier. If reelin and LilrB2 signalling show no measurable interaction at serine 3 when co-manipulated in the same neurons, the convergence is coincidental — three pathways that happen to terminate on a common cytoskeletal regulator, which is unremarkable — and this chapter should be withdrawn.

18.6 The consequence for therapy

A redundancy argument, and it is not comfortable for any single-target programme. At least four upstream arms converge on this machine: the inhibitory receptor driven by amyloid, the same receptor driven by complement, the reelin axis running the other way, and the Rho-family exchange factors. Blocking one leaves the effector reachable by the others, which predicts partial efficacy for any monotherapy on this pathway — the field's general experience.

It also argues that the more efficient point of intervention may be the shared effector rather than any individual arm, with the immediate objection that cofilin and the Rho-kinase axis are required for normal structural plasticity, and that the node's dose–response is non-monotonic. A therapy at the convergence point has the same problem as a therapy at any single arm, only worse: there is no direction in which to push a variable whose optimum is a narrow band.

The arms are not equally reachable, and that asymmetry is where the practical answer lies. The complement arm has agents in clinical development. The receptor arm is extracellular, with an endogenous antagonist already identified and antibodies already through early-phase oncology trials. The reelin arm is the one with two human gain-of-function resistance alleles and no tractable molecule. The exchange-factor arm is intracellular, enzymatic, pleiotropic and attached to a dominant human loss-of-function vasculopathy, and on tractability alone is now the least attractive of the four.

19. The Membrane Proteasome Is Probably a Raft Protein

19.1 The coincidence

Two programmes report a surface pool on the neuronal plasma membrane whose abundance is modulated by the same two variables, in the same direction, and neither cites the other.

The surface membrane proteasome is reported to be modulated by apolipoprotein E isoform in the order **E2 > E3 > E4** — approximately doubled in ApoE2 and reduced by roughly a third in ApoE4 relative to ApoE3 — and to decline with age (Paradise et al., 2026).

Raft localisation of PSD-95, GluN2A, GluN2B, TrkB, AMPA receptor subunits and caveolin-1 itself declines with age in mouse hippocampus (Head et al., 2010), brain cholesterol falls with age, and raft abundance falls with it (Egawa et al., 2016). Raft composition and abundance in human neurons differ by *APOE* genotype under direct imaging (Lee et al., 2021; Lee et al., 2025), and lipid raft yield is lower in *APOE ε4* carriers in human brain (Thorwald et al., 2025).

Two independently discovered surface pools, two shared modifiers, same directions. The most economical explanation is that they are one measurement.

19.2 The proposal

The neuronal membrane proteasome is a resident of ordered membrane microdomains, and its apolipoprotein-E-graded, age-declining surface abundance is the raft-assembly deficit measured with a different antibody.

Three considerations make the proposal more than a coincidence of correlates.

The topology problem has a raft-shaped answer. The most frequently voiced reservation about the whole neuronal-membrane-proteasome concept is structural: the 20S proteasome is a hydrophilic barrel with no obvious means of embedding in a lipid bilayer, and how it is held at the membrane with its catalytic chamber facing outward is unexplained. A multipass transmembrane glycoprotein of the proteolipid-protein family has been proposed as the anchor. Proteolipid-family proteins are, by definition, lipid-associated integral membrane proteins, and an anchor of that class would place the complex in exactly the membrane environment whose assembly the raft programmes study. An unexplained topology is a reason for caution about the complex; it is also, here, a hypothesis about which membrane environment holds it.

Apolipoprotein E has no other obvious route to a surface protein complex. Apolipoprotein E is a lipid carrier. Its known cell-biological effects on neurons run through lipid delivery, endolysosomal capacity and receptor recycling. For an isoform series to set the abundance of a proteasome at the plasma membrane, it must act through one of those. Delivery of the ordering lipid that maintains the domain in which the complex sits is the shortest available path, and it predicts the observed order, because the same order is reported for raft cholesterol handling.

The dependencies match a lipid mechanism better than a transcriptional one. The effect is on surface abundance rather than total abundance; it is graded rather than binary; it is age-dependent, emerging over time rather than being present in young neurons — which is the reported behaviour of the apolipoprotein E effect on neuronal endolysosomal function generally, where knockout, E3 and E4 neurons show no major differences until prolonged time in culture (Nyberg et al., 2025).

19.3 What follows if it is right

The apolipoprotein E to tau link acquires a membrane mechanism. The largest common genetic risk factor for the disease would set the threshold for *de novo* tau filament formation by setting the abundance of the ordered domain in which the tau-degrading complex resides. That is a neuron-autonomous, quantitative, age-dependent account of the *APOE* effect on tau, which the field has lacked, and it explains why the protective allele is an active gain rather than the absence of a risk.

A membrane-directed intervention should raise the tau threshold. The prediction is specific and cheap to test: a manipulation that restores ordered-domain assembly — caveolin-1 over-expression, cholesterol repletion, or any of the tool compounds used in the raft literature — should increase surface proteasome abundance and raise the fraction of inhibition required to produce filaments. If it does, the resilience construct of Chapter 9 has a molecular mechanism for an effect on tau that it has never claimed and never measured.

Two "unexplained" observations become one. The surface proteasome declines with age and nobody knows why; ordered-domain assembly declines with age and the reason is a documented fall in available cholesterol. One explanation would then cover both.

19.4 The experiments

The primary test. Subcellular fractionation of neuronal membrane, asking whether catalytically active proteasome co-purifies with detergent-resistant membrane and with the standard domain markers, in neurons of each *APOE* genotype and at two ages. A single afternoon of gradient work answers the central question.

The manipulation. Deplete membrane cholesterol with methyl-β-cyclodextrin and measure surface proteasome abundance and activity with the impermeant substrate; replete and measure again. If the complex is domain-resident, both should track cholesterol.

The consequence test. In ApoE4 neurons, restore ordered-domain assembly and measure the inhibition threshold at which endogenous tau filaments appear. The prediction is that the ApoE4 threshold moves toward the ApoE3 value.

The falsifier. If active surface proteasome does not co-fractionate with detergent-resistant membrane, and if cholesterol depletion leaves surface abundance unchanged, the proposal is wrong and the apolipoprotein E effect on the complex must run through a different mechanism — most plausibly transcriptional or through the endolysosomal delivery of the anchor rather than of the lipid.

Graded **inference**. The correlates are established; the identity is not tested.

20. The De-Acidified Endosome and the Bond That Will Not Break

20.1 Two mechanisms, one phenotype

A lipoprotein receptor that cannot release its ligand cannot be recycled, and the cell loses surface receptor. Two of the programmes reviewed here, plus a third body of work that neither cites, offer three different mechanisms for that single phenotype.

The conformational mechanism. ApoE4 adopts a molten-globule state near its isoelectric point at about pH 6.5, so it fails to dissociate at endosomal pH. ApoE4 selectively impairs ApoER2 recycling, reducing glutamate-receptor function and synaptic plasticity (Chen et al., 2010), and the block is pharmacologically reversible by lowering endosomal pH (Xian et al., 2018).

The covalent mechanism. Reactive lipid aldehydes crosslink apolipoprotein E to the ligand-binding modules of its receptor, and the crosslinks are acid-resistant: malondialdehyde adducts showed partial pH-dependent reversibility consistent with Schiff-base chemistry, while crosslinks formed with the reactive-aldehyde mixture showed minimal reversibility and remained stable at lysosomal pH (Ramsden et al., 2022).

The acidification mechanism. The endosome may fail to acidify at all. The tyrosine-682-phosphorylated β-carboxy-terminal fragment of the amyloid precursor protein inhibits the vacuolar ATPase, and this — not amyloid-β — is what fails to acidify the compartment in Alzheimer and Down syndrome models (Im et al., 2023); the same laboratory's account of the terminal stage is a profound failure of autolysosomal acidification (Lee et al., 2022).

20.2 The join

The three mechanisms are not alternatives in the usual sense, because two of them can be produced by the third.

If C99 and its phosphorylated derivative inhibit the proton pump, then a neuron accumulating the fragment has a less acidic early endosome. In a less acidic endosome, the conformational transition that releases ApoE4 does not occur — so the conformational mechanism is a *consequence* of the fragment, not an independent property of the protein. And a less acidic endosome is also a longer-dwelling endosome, which is the traffic jam of Chapter 7 arriving by a route the traffic-jam model does not name.

The join is this: the β-carboxy-terminal fragment, which Chapter 6 identifies as an uncleared cholesterol signal and Chapter 8 as the retained product of a routing decision, is also the agent that de-acidifies the compartment in which the lipoprotein receptor must release its cargo. One species links the amyloid arm, the lipid-delivery arm and the retrieval arm at a single step of the circuit.

Three testable consequences follow.

It offers an adjudication of the Knupp–Hung contradiction. Chapter 26 records an unresolved disagreement about whether the endosomal phenotype downstream of *SORL1* loss requires the amyloid precursor protein — reported as independent of amyloidogenic processing in one study (Knupp et al., 2020) and relieved by antisense reduction of the precursor protein in another (Hung et al., 2021). If the fragment de-acidifies the compartment, an *APP*-dependent component of any endosomal phenotype is expected, and the disagreement becomes quantitative rather than categorical: the phenotype has an APP-dependent acidification component and an APP-independent retrieval component, and the two studies differ in the depth of the *SORL1* lesion and therefore in which component dominates.

It predicts a sequence in apolipoprotein E4 carriers. If the recycling block is partly a pH block and the pH block is partly a fragment effect, then lowering the fragment should partially restore receptor recycling in ApoE4 neurons — an effect attributable to neither apolipoprotein E nor amyloid-β on their own.

It relocates the therapeutic target. Restoring endosomal acidification is a different intervention from lowering apolipoprotein E4, from lowering amyloid, and from stabilising retromer, and it would act on all three phenotypes at once.

20.3 The discriminating experiment

The experiment is a single manipulation with three arms, and it is the sharpest test in this paper because the two models make opposite predictions about the same readout.

Take neurons expressing ApoER2. Load them with apolipoprotein E particles under three conditions: native, peroxidised, and native in cells accumulating the β-carboxy-terminal fragment. Measure surface receptor recovery after internalisation, and then acidify the endosomal compartment by the method already shown to reverse the ApoE4 block.

- If the block is conformational or pH-driven, acidification rescues recovery in all conditions.
- If the block is covalent, acidification rescues the native and fragment conditions and **fails to rescue the peroxidised condition**, because a covalent bond is not pH-sensitive.

- If the block in the fragment condition is not rescued by acidification either, the fragment is doing something beyond the proton pump.

That single experiment discriminates three mechanisms, uses only published reagents, and has not been done.

20.4 What would refute the join

If the β-carboxy-terminal fragment's effect on the vacuolar ATPase does not extend to the early endosome — if it is confined to the autolysosome, as the original reports emphasise — then the link to lipoprotein-receptor release fails, and the fragment's role is restricted to the degradative arm. The measurement required is a compartment-resolved pH determination in neurons accumulating the fragment, using ratiometric probes targeted to early-endosomal and late-endosomal markers separately.

Graded **inference**, with each of the three component mechanisms individually graded **strong**.

21. The Cleaved Receptor and the Retrograde Route

21.1 The finding

Of the recent results in the restraint programme's literature, the least expected is that PirB is proteolytically cleaved upon amyloid exposure — in patients and in mouse models — and that the resulting C-terminal fragment does damage from inside the cell. Proteomic screening of human cerebrospinal fluid identified cleaved membrane proteins; the cleavage was validated in post-mortem Alzheimer brain, primary neurons and APP/PS1 mice. The fragment accumulates in the Golgi apparatus by retrograde transport and binds the GAT domain of GGA3, disrupting Golgi transport, impairing lysosomal maturation and compromising anterograde synaptic-vesicle transport. Inhibiting the cleavage, or over-expressing the GGA3-GAT domain, restored Golgi function, reduced plaque burden and tau phosphorylation, and rescued memory deficits (Han et al., 2026).

21.2 The join

GGA proteins are clathrin adaptors that carry cargo between the trans-Golgi network and the endosomal system. Retromer serves the same corridor from the other direction, recovering cargo from the endosome and returning it to the Golgi or to the surface. **A fragment that jams GGA3 and a complex that fails to retrieve are two lesions on one retrograde route.**

That produces a specific structural claim: the surface receptor of Chapter 11, when engaged by its pathological ligand, generates an intracellular species that phenocopies part of the trafficking lesion of Chapter 7. The two programmes have no shared vocabulary, no shared citations, and — on this reading — a shared compartment and a shared consequence.

Three consequences.

The restraint programme acquires a return-leg lesion. Chapter 15 recorded that the C4d arm is the one place where the return-leg thesis fits badly, because C4d is an added forward signal. The cleavage finding supplies the missing return-leg component of the same programme: an engaged receptor is normally internalised, sorted and either recycled or degraded, and here its fragment is retained on the retrograde route and jams it.

Blocking the ligand and blocking the cleavage are different interventions. If a substantial part of the damage is done by the fragment rather than by the signalling of the intact receptor, then an ectodomain-blocking antibody — the intervention the field would reach for, and the one already through early-phase oncology trials — addresses the smaller half. A protease inhibitor addresses the larger. Nobody has compared them.

It predicts reciprocal rescue. If the two lesions are on one route, then retromer enhancement should partially rescue the fragment phenotype, and blocking the cleavage should partially rescue phenotypes attributed to retromer deficiency. Retromer chaperones exist and have been tested independently in an animal (Ramonet et al., 2025); the cleavage inhibitor exists in the originating study. The cross experiment is available now.

21.3 The obligatory caution

This is a single study, published in 2026, not replicated, in a literature that Chapter 11 records as uneven. The join should be read as a hypothesis worth one experiment rather than as an established connection, and the reciprocal-rescue test is the cheapest way to find out whether it is worth more.

There is also a plainer alternative that the experiment would distinguish: the fragment's effect may be a general consequence of accumulating an undegraded membrane-protein fragment in the Golgi, in which case any similar fragment would do it and the specificity to GGA3 is incidental. The control is a comparable cleaved receptor tail from an unrelated surface protein.

Graded **inference**, resting on one **moderate** primary finding.

22. Activity, and Why Its Sign Changes With the Compartment

22.1 The apparent contradiction

Neuronal activity is reported as protective and as harmful, by programmes whose evidence is equally good.

Protective. Synaptic activity reduces the intraneuronal amyloid pool, promotes transport of the precursor protein to synapses, and protects against amyloid-related synaptic alterations, with the reduction mediated by neprilysin at the cell surface (Tampellini et al., 2009). Two independent means of chronic synaptic inhibition — deafferentation and a benzodiazepine — reduced plaques and *worsened* synaptic and memory outcomes, with the deterioration occurring alongside elevated intraneuronal amyloid (Tampellini et al., 2010).

Harmful. Synaptic activity regulates interstitial amyloid-β levels in vivo and endocytosis is required for activity-dependent release (Cirrito et al., 2005; Cirrito et al., 2008). Activity stimulates the physiological release of tau (Pooler et al., 2013) and enhances tau propagation and pathology in vivo (Wu et al., 2016). Neurons carrying elevated amyloid become hyperactive and lose the ability to retune themselves (Busche et al., 2008; Martinsson et al., 2022), and hyperactivation is self-reinforcing (Zott et al., 2019).

22.2 The resolution

The contradiction dissolves once activity is read as a load on the circuit of Chapter 13 rather than as a single variable.

Activity drives the forward leg of every cycle. It drives synthesis of nascent protein, tau among it, which must then be trimmed by the surface proteasome. It drives insertion and internalisation of receptors, which must then be retrieved. It drives release of the precursor protein's products, which must then be degraded. It drives destabilisation of the cytoskeleton in candidate generation, which must then be restabilised. Every one of those is a demand on a return leg.

Activity drives the return leg of exactly one of them. The activity-coupled recruitment of neprilysin to the cell surface is, in this set, the only demonstrated case in which activity itself increases a recovery capacity (Tampellini et al., 2009, 2011).

Therefore the net sign of activity depends on whether that one coupling is intact. While it is, activity is protective, because the single return leg it drives is the one that governs the pool the four synaptic routes depend on. Once the coupling fails — and its failure is the specific lesion reported, with surface neprilysin falling in diseased neurons — activity becomes a pure load, and every additional demand lands on return legs that are already failing.

That is a testable statement with a name: **the activity–clearance coupling is the switch that determines the sign of activity in this disease.**

22.3 What it explains

Cognitive reserve acquires a molecular content. The association of education, occupational complexity and continued engagement with later onset is usually explained by network redundancy. The mechanism here is different and additive: use buys enzymatic clearance at the synapse, so a more-used synapse carries a lower internal load. It also explains the apparent paradox that the regions most continuously active across a lifetime are those with the earliest deposition (Buckner et al., 2005) — those regions secrete the most and therefore deposit the most, while being intracellularly protected until the coupling fails.

Sleep is the period in which return legs run. Amyloid dynamics are regulated by the sleep–wake cycle (Kang et al., 2009); sleep drives metabolite clearance from the adult brain (Xie et al., 2013); one night of sleep deprivation raises amyloid-β in the human brain (Shokri-Kojori et al., 2018); and excitatory synapses are homeostatically scaled down during sleep, with ultrastructural confirmation (Diering et al., 2017; de Vivo et al., 2017). On the frame of Chapter 14, sleep is not a period of reduced activity but the period during which the recovery steps of the cycle are executed. That reframes the prescription: the intervention indicated is not less activity but more recovery, and they are not the same thing.

Sedation and activity reduction are not equivalent to rest. The chronic-inhibition experiment reduced activity pharmacologically and made the animals worse. If sleep is the recovery phase and sedation merely suppresses the forward leg without running the return leg, the two should have opposite effects — which is what was observed. This is a mouse result and should not be over-read, but it is a specific, mechanistically grounded hypothesis that epidemiology could examine: long-term benzodiazepine exposure in older adults at risk, evaluated against a mechanism rather than as a bare association.

The tau clock and the amyloid clock respond to activity differently. A neuron that fires more makes more tau and must dispose of more tau at a surface pool that is declining with age and genotype (Chapter 10). A neuron that fires more also clears more amyloid, while the coupling holds. The same behavioural intervention therefore pushes the two proteinopathies in opposite directions, which may be part of why cognitive-engagement interventions have produced

inconsistent effects on biomarker endpoints.

22.4 The experiment

Measure the activity–clearance coupling directly, as a function of disease stage. In an animal model with a knock-in humanised locus rather than over-expression, apply a graded activity manipulation and measure, in the same tissue, surface neprilysin, the retained intraneuronal pool, interstitial amyloid-β, nascent tau and surface proteasome abundance. The prediction is a stage-dependent inversion: at early stages the intraneuronal pool falls with activity, and beyond the stage at which surface neprilysin has fallen, it rises.

The falsifier. If the intraneuronal pool falls with activity at all disease stages, the coupling is not the switch, and the protective effect of activity is not conditional in the way proposed.

Graded **inference**, resting on two **strong** primary findings and a large body of **established** work on activity and interstitial amyloid.

23. Every Neuronal Lesion Has a Microglial Address

23.1 The pattern

Six of the nine programmes are neuron-centric in their original statement, and six of the nine have acquired a microglial version of the same lesion within the last four years — in most cases from laboratories outside the originating group, and in one case from the originating group's own human imaging.

Table 9 — The microglial address of each neuronal lesion

Programme	Neuronal lesion	Microglial version	Evidence
Retromer recycling	Neuronal retrieval fails	The strongest protective common variant acts through *microglial* SORLA expression; SorLA governs ER stress, lipid droplets and phagocytosis in human microglia; microglial density mediates tau-associated neurodegeneration in human imaging	Gorniak-Walas et al., 2026; Haq et al., 2026; Lao et al., 2026
Restraint	LilrB2 drives spine disassembly	LILRB2 on human microglia inhibits TREM2 signalling on co-ligation; antagonist antibodies restore phagocytosis and migration	Zhao et al., 2022
Competition	Monomer depletion disinhibits microglia	Microglial deletion of the precursor protein produces hyper-responsive microglia and, with a second insult, structural damage through MMP9	Kwon et al., 2024
Lipid regulation	Uncleared C99 expands the contact	Microglial lipid-droplet accumulation is a dysfunctional pro-inflammatory state; *APOE4/4* is linked to damaging lipid droplets in human Alzheimer microglia; ApoE4 lipid accumulation degrades microglial surveillance	Marschallinger et al., 2020; Victor et al., 2022; Haney et al., 2024
Membrane platform	Neuronal raft organisation degrades	Caveolin isoform switching regulates microglial structure and metabolism	Niesman et al., 2013

Programme	Neuronal lesion	Microglial version	Evidence
Intraneuronal retention	Retained pool damages the synapse	Immunotherapy is best understood as engaging a multicellular clearance system rather than removing a deposit	Zhan et al., 2026
Peroxidation	Receptor interface damaged by aldehydes	Not established	—
Surface proteolysis	Nascent tau not trimmed	Not applicable — the complex is nervous-system-specific and neuron-restricted	—
Raft assembly gate	Astrocyte supply fails	Astrocytic rather than microglial; the supply cell is the astrocyte	—

23.2 The join: the microglial lesion is also a return-leg lesion

The observation that microglia matter is not new and not interesting on its own. What is interesting is the *shape* of the microglial lesion in each row, and it is the same shape as the neuronal one.

Phagocytosis has a forward leg and a return leg. The forward leg is recognition and uptake: receptors, opsonins, engulfment. The return leg is digestion and disposal: acidification of the phagosome, lipid processing, efflux of the products. **In every row of Table 9 where the microglial phenotype is characterised in molecular detail, the reported lesion is on the digestive half.** Lipid droplets accumulate. Endoplasmic-reticulum stress rises. Phagocytic capacity falls *because the cell is already full*, not because it cannot recognise its target.

This is a reframing with a consequence, and the consequence has already been tested inadvertently. If the microglial lesion were on the forward leg — insufficient recognition, insufficient activation — then an agonist of the principal recognition receptor should help. A TREM2 agonist antibody with confirmed target engagement produced no clinical benefit (Mummery et al., 2026), and the reaction from within the field was to ask what the target had been assumed to do (Colonna and Holtzman, 2025). On the return-leg reading, the null result is expected: pushing uptake in a cell whose disposal capacity is exhausted adds substrate to a jam.

The claim, stated for testing: microglial competence in this disease should be measured as digestive throughput rather than as phagocytic uptake, and interventions should target disposal rather than activation.

23.3 Why the two lesions have the same shape

Two candidate explanations, and they are distinguishable.

The shared-substrate explanation. Both cell types are handling the same lipid, delivered by the same carrier, and both fail at the step that disposes of it. Apolipoprotein E4 produces lipid-droplet accumulation in microglia and endolysosomal impairment in neurons; the ordered-domain literature applies to both membranes. On this reading the disease has one lesion expressed in two cell types.

The coupled-failure explanation. The neuronal lesion produces the microglial one. A neuronal retromer deficit is *sufficient* to induce a dystrophic microglial morphology characteristic of the disease, with no primary microglial manipulation (Qureshi et al., 2022). A neuron that cannot dispose of its lipid and protein exports the problem — as extracellular vesicles, as complement-tagged debris, as exposed phosphatidylserine — and the microglion that must handle it is overwhelmed.

The two are not exclusive, and the direction that carries the weight in sporadic disease is genuinely unknown. What can be said is that a common-variant signal acting through microglial expression (Gorniak-Walas et al., 2026) is difficult to explain on the coupled-failure reading alone.

23.4 The consequence for the whole integration

If the microglial lesion is a disposal lesion, then the return-leg thesis is not a claim about neurons. It is a claim about the tissue: the cell types that fail in this disease fail at their recovery steps, and they do so while handling a shared lipid whose supply chain has one non-redundant step.

That is a larger claim than this paper's evidence carries and it is graded **inference**. It generates two experiments in Chapter 30 and one refutation condition in Chapter 32.

24. What the Nine Say About Measurement

24.1 The strongest convergence in the paper is methodological

Each of the nine programmes contains, usually buried in a limitations section, a statement that the standard measurement in its own area does not measure the quantity the programme identifies as pathogenic. Assembled, they are the same statement made nine times.

Table 10 — What each programme says the field is measuring wrongly

Programme	The standard measurement	Why it does not measure the relevant quantity	Source
Intraneuronal retention	Cerebrospinal-fluid and plasma amyloid-β; amyloid PET	These report the extracellular soluble pool and the deposited pool; the pool identified as damaging is retained and intraneuronal, and no clinical assay reports it	Gouras et al., 2012
Intraneuronal retention	Amyloid-β immunoassay	Systematically *underestimates* the peptide once assembled, because assembly hides the epitopes	Stenh et al., 2005
Intraneuronal retention	Anti-amyloid antibody staining	The standard antibody binds pyramidal neurons, microglia, astrocytes, oligodendrocytes, perivascular macrophages and vessels in addition to plaques	Wen et al., 2026
Restraint	Binding assays with synthetic amyloid assemblies	The receptor bound synthetic assemblies but showed no detectable binding to human-brain-derived oligomers in a standardised comparison	Smith et al., 2019
Competition	Bulk amyloid-β concentration	The variable that separates protective from destructive is conformation, not concentration, and preparation holds conformation constant	Kwon et al., 2024, read against the theory's own tabulation
Retromer recycling	Soluble SORL1 in cerebrospinal fluid	A whole-compartment measure of a lesion that may be confined to the trans-entorhinal cortex, or to a cell type contributing little to the fluid pool	de Waal et al., 2026

Programme	The standard measurement	Why it does not measure the relevant quantity	Source
Lipid regulation	The amyloid-β 42:40 ratio	May be a surrogate for the thickness of a membrane rather than an index of a toxin	Area-Gómez and Schon, 2024
Membrane platform	Plaque burden as the therapeutic endpoint	A therapy defined by changing nothing imageable has no target-engagement biomarker	Chapter 9
Raft assembly gate	Bulk brain or membrane cholesterol	Cannot distinguish the raft pool from the non-raft pool, and the theory's claim is about the raft pool only	Rappoport, 2025
Peroxidation	Immunohistochemistry for modified apolipoprotein E	Suggestive, not identification; no crosslinked species has been isolated from human brain by mass spectrometry	Chapter 5
Membrane platform	Bulk-tissue caveolin-1	Reports the sum of endothelial, glial and neuronal pools moving in different directions	Chapter 9

24.2 The single sentence they add up to

Every one of the nine programmes reports that the standard measurement in its area is a bulk measurement of a quantity that is defined per compartment, per cell type, or per conformation.

That is the most reproducible finding in this paper, and it was reached nine times independently by investigators who were not looking for it and who state it as a limitation of their own work rather than as a claim about the field.

Its implications are larger than any of the mechanistic joins in Part Four, because a field cannot test a compartment-resolved hypothesis with a bulk assay, and the great majority of the human evidence in this literature is bulk. It also explains, without any appeal to error or bias, why so many of the disagreements catalogued in Part Five are *sign* disagreements: two investigators measuring the same molecule in the same tissue with different compartment resolution will obtain different signs whenever the molecule moves in opposite directions in two compartments, which is exactly the situation the circuit of Chapter 13 predicts.

24.3 The five assays the integration requires

Stated here and ranked in Chapter 30.

- **A proxy for the retained intraneuronal pool.** By imaging, by neuron-derived extracellular vesicles, or by a conformation-sensitive assay. This is the missing measurement that couples the two clocks of Chapter 17, and it is the single most valuable thing that could be developed for any of the nine programmes — valuable regardless of whether any of them is right, because it would allow the question to be settled.
- **A conformation-resolved amyloid-β assay usable in human fluid.** Distinguishing monomer from the assemblies, at the concentrations that occur in vivo rather than the concentrations used in vitro. Without it, "do not remove the monomer" cannot be operationalised as a trial design.
- **A raft-pool lipid measurement in human tissue.** Direct imaging methods now exist and have been applied to human neurons under *APOE* genotype (Lee et al., 2021; Lee et al., 2025); applying them systematically to post-mortem human cortex, with cell-type resolution, would settle two of the three sign problems of Chapter 25 at once.
- **A pathway-function panel for endosomal retrieval.** Soluble SORL1 exists in preliminary form, with a calibrating cell-based reporter (de Waal et al., 2026; Fazeli et al., 2026); what is needed is a panel applied to a large, longitudinally followed cohort stratified by genotype and amyloid status.
- **A target-engagement biomarker for membrane organisation.** No candidate is close. The nearest human-scanner readout is an ultrashort-echo-time myelin measure (Wang J. et al., 2026), which is a downstream structural consequence rather than an engagement measure.

24.4 A note on what this does not mean

It would be a mistake to read Table 10 as an indictment. Bulk assays built the field, they are the reason amyloid pathology can be staged in a living person, and two of them support a licensed therapy. The claim is narrower and it is the one the nine programmes themselves make: **these assays are informative about the deposit and uninformative about the cycle**, and the cycle is where all nine locate the lesion.

Part Five — The Disagreements

What the integration does not resolve: three disputes about the direction of an effect, six direct empirical contradictions, and the experiments that would settle them.

25. The Three Sign Problems

25.1 Why sign disputes are the interesting ones

A disagreement about magnitude is a disagreement about how much a mechanism contributes. A disagreement about *sign* is a disagreement about what the mechanism is, and it has the property that both parties can be measuring correctly. Three live sign disputes run through the nine programmes. Each is stated here at full strength on both sides, and §25.5 argues that they are probably one problem.

25.2 Sign problem one: does apolipoprotein E4 supply the neuronal membrane with too little cholesterol, or too much?

The deficiency case. The raft-gate theory holds that the lesion is reduced neuronal uptake or trafficking of astrocyte-produced cholesterol, and therefore impaired assembly of plasma-membrane ordered domains (Rappoport, 2025). It made a specific prediction — that the failure is in *uptake* rather than in *synthesis* — and the prediction was confirmed in human material: cerebrospinal-fluid lipoprotein-mediated cholesterol delivery to neurons is impaired in Alzheimer's disease, and the impairment involves ApoE4 (Borràs et al., 2025). Consistent with this, lipid raft *yield* is lower in *APOE ε4* carriers in human brain (Thorwald et al., 2025), and raft localisation of the receptors that matter declines with age alongside falling brain cholesterol (Head et al., 2010; Egawa et al., 2016).

The excess case. Direct imaging says the opposite. Human astrocytes carrying *APOE4* *over*-supply cholesterol to neurons, promoting neuronal lipid-raft **expansion** and increased amyloid-β generation (Lee et al., 2021); the finding was extended with time-of-flight secondary-ion mass spectrometry imaging of rafts in human neurons, which reported the same direction (Lee et al., 2025). Independently, the lipid-regulation programme holds that ApoE4-containing lipoproteins are recycled less efficiently, cholesterol accumulates intracellularly, and contact-site function is *increased* (Tambini et al., 2016; Area-Gómez et al., 2012). And *APOE4/4* is associated with damaging lipid-droplet accumulation in human Alzheimer microglia (Haney et al., 2024) — accumulation, not scarcity.

Where the two can both be right. Three separations are available and none has been tested against the other.

- **Compartment.** Cholesterol may accumulate in the endolysosomal system and at the endoplasmic reticulum while failing to reach the plasma membrane. This is precisely what a retrieval failure would produce, and it is the reading the circuit of Chapter 13 predicts: a

receptor that cannot be recycled leaves its cargo in the compartment behind the membrane. The raft-gate theory itself makes this concession explicitly — the disease "can involve both cholesterol deficiency in plasma-membrane rafts and excess" elsewhere.

- **Cell type.** Astrocytic over-supply and neuronal under-incorporation are not contradictory. One reports the donor, the other the recipient.
- **Stage.** Over-supply early, when synthesis is up-regulated in compensation, and depletion late, when the supply cell is itself impaired.

The adjudicating measurement. Compartment- and cell-type-resolved cholesterol imaging in human cortex, across *APOE* genotypes and disease stages. The methods exist and have been applied to cultured human neurons; they have not been applied systematically to human tissue.

25.3 Sign problem two: does neuronal caveolin-1 rise or fall in the human disease?

Up. Bulk human tissue reports increased caveolin-1 expression in Alzheimer brain (Gaudreault et al., 2004), and caveolin-1 up-regulation in senescent neurons alters precursor-protein processing (Kang et al., 2006).

Down. The membrane-platform programme holds that neuronal caveolin-1 falls with age and disease, and builds its therapeutic argument on restoring it.

Where both can be right. Caveolin-1 is most abundant in endothelium, is expressed in glia, and is present at low abundance in neurons. A bulk-tissue measurement in a disease characterised by gliosis and vascular change is dominated by the non-neuronal pools. If endothelial and glial caveolin-1 rise while neuronal caveolin-1 falls, both results are correct and the bulk measurement is uninformative about the claim.

The adjudicating measurement, which is already sitting in public repositories. Cell-type-resolved expression of *CAV1* from existing single-nucleus RNA sequencing of human Alzheimer cortex, stratified by braak stage. This does not require a new experiment, and it is the cheapest item in Chapter 30.

A note on what turns on it. Very little, for the therapy. A gain-of-function that confers resilience is a complete argument whether or not the protein is depleted at baseline. Asserting a contested premise the argument does not need is a gratuitous liability, and this is the clearest instance of that pattern in the nine.

25.4 Sign problem three: is reduced lipoprotein-receptor engagement the lesion or the protection?

The lesion reading. The peroxidation framework lists impaired apolipoprotein-E-receptor-mediated lipid delivery as one of four pathogenic arms: the receptor is crosslinked, cannot recycle, and the neuron loses both surface receptor and lipid supply (Ramsden et al., 2022; Ramsden et al., 2023).

The protection reading. Impaired low-density-lipoprotein-receptor binding by lipidated ApoE2 *avoids* the receptor-recycling defects seen with E3 and E4 and decreases uptake of cholesteryl esters; polyunsaturated cholesteryl esters carried on apolipoprotein E produce an E4 > E3 > E2 series for lipofuscinosis in human neurons; and the protective Christchurch variant also reduces receptor binding and phenocopies E2 (Guo et al., 2025). Separately, ApoE2 and Christchurch particles protect neurons by *effluxing* oxidised unsaturated lipids through ABCA7, while ApoE4 particles exacerbate their effects (Ralhan et al., 2026).

Where both can be right, and it is the cleanest of the three. The direction depends on the state of the cargo. Delivery of intact lipid is a physiological requirement; delivery of *peroxidised* lipid is an injury. Reducing engagement is therefore protective precisely when the cargo is damaged, which is the condition the peroxidation framework itself asserts obtains in the ageing brain.

The pattern across all three known protective variants is coherent under one rule, and it is worth stating as the resolution: **apolipoprotein E is the ligand you want less of at this receptor, and reelin is the ligand you want more of.** Both protective *APOE* variants are poor receptor binders. The protective *RELN* variant is a better receptor activator. On the shared-effector argument of Chapter 18, both moves shift the same balance at serine 3 of cofilin in the same direction.

The adjudicating measurement. Whether reducing receptor engagement is protective when the cargo is *not* peroxidised. If it is protective under all conditions, the peroxidation framework's delivery arm is simply wrong; if it is protective only with damaged cargo, the two accounts are one account with a state variable.

25.5 The three problems are probably one problem

Each of the three has the same structure. A quantity is measured in bulk. The quantity has opposite signs in two compartments, two cell types, or two states of a cargo. The bulk measurement returns whichever sign dominates the sample, and two competent laboratories obtain opposite answers.

This is the prediction of Chapter 24 realised three times. It is also the reason this paper declines to average them: an averaged claim — "apolipoprotein E4 modestly alters cholesterol handling" — is true, useless, and untestable, while the disaggregated claims are all testable with methods that

exist.

Table 11 — The three sign problems and their resolving variable

Dispute	Position A	Position B	Resolving variable	Method
ApoE4 and membrane cholesterol	Deficiency at the plasma-membrane raft	Over-supply and raft expansion	Compartment (surface versus endolysosome and ER); cell type; stage	Compartment- and cell-type-resolved lipid imaging in human cortex
Caveolin-1 in human disease	Increased in bulk tissue	Decreased in neurons	Cell type	Deconvolution of existing single-nucleus data
Lipoprotein-receptor engagement	Reduced delivery is a lesion	Reduced delivery is the protection	State of the cargo (peroxidised or not)	Receptor-engagement manipulation with matched intact and peroxidised particles

26. Contradictions That Should Not Be Dissolved

26.1 The rule

The joins of Part Four were made where two programmes name a shared object. Where two programmes name a shared object and disagree about it, the disagreement is recorded here without adjudication, because adjudicating it would require an experiment that has not been done and inventing a reconciliation would obscure that fact.

26.2 Does the endosomal phenotype downstream of *SORL1* loss require the precursor protein?

Depleting *SORL1* in human stem-cell-derived neurons impairs endosomal traffic **independently of amyloidogenic precursor-protein processing** — the result the endosome-first model requires (Knupp et al., 2020). Studying a *SORL1* truncating mutation in the same class of system, a different group reported that the endolysosomal dysfunction caused by loss of SORL1 was **relieved by antisense-oligonucleotide reduction of the precursor protein**, concluding that *PSEN1*, *APP* and *SORL1* act in a common pathway (Hung et al., 2021).

These are not easily reconciled. They differ in the depth of the *SORL1* lesion, in the differentiation protocol and in the readouts, and each is a plausible source of the discrepancy. But the question at issue — whether the endosomal phenotype downstream of *SORL1* loss requires the precursor protein — is exactly the question on which the ordering of the entire hypothesis turns, and five years on it has not been settled by a study designed to settle it.

Chapter 20 offers a mechanism that would make the answer quantitative rather than categorical, and that mechanism is itself a hypothesis.

26.3 What jams the endosome first?

Two accounts of the same compartment, both with interventional evidence in animals, disagreeing about the entry point.

The retrieval account has the sorting machinery fail first, with the precursor protein a consequence: retromer depletion and repletion drive the phenotype in both directions (Qureshi et al., 2022), and human genetics anchors the receptor (Andersen, de Waal et al., 2025).

The fragment account has the precursor protein's fragment fail the sorting machinery: the β-carboxy-terminal fragment recruits APPL1 to rab5-positive endosomes, stabilising the active form of rab5 and producing accelerated endocytosis, endosome swelling and impaired axonal transport (Kim et al., 2016). The account was then made independent of the precursor protein — a mouse in which rab5 is over-activated directly, with no *APP* manipulation, reproduces endosome enlargement, accelerated AMPA-receptor endocytosis, spine loss, tau hyperphosphorylation, cholinergic neurodegeneration and memory impairment (Pensalfini et al., 2020) — and a transgenic APPL1 mouse reproduced the same set (Jiang et al., 2025).

Neither has been tested against the other in a design that could distinguish them. The unperformed experiment is reciprocal rescue: does retromer enhancement rescue the rab5-over-activation mouse, in which there is no primary retromer lesion?

26.4 Does the inhibitory receptor bind the amyloid assemblies that occur in human brain?

The programme's 2013 claim is that human LilrB2 is an amyloid-β receptor, with PirB required for the deleterious effect of oligomers on potentiation (Kim et al., 2013). In a standardised head-to-head comparison of fifteen reported amyloid receptors for sufficiency, affinity and disease relevance, LilrB2 bound *synthetic* synaptotoxic assemblies but showed **no detectable binding to human-Alzheimer-brain-derived oligomers**, while cellular prion protein bound strongly (Smith et al., 2019).

Seven years on, this has not been answered. It does not touch the complement arm, which is measured in human tissue with human ligand, and Chapter 11 records that the complement arm is the better-evidenced one.

26.5 Lower production, or do not lower it?

Two of the nine programmes recommend interventions with opposite signs on the same enzyme.

The retention programme's target list includes reducing production rather than removing deposits, on the strength of the demonstration that blocking amyloid production reverses synaptic decline in aged neurons (Burrinha et al., 2021).

The lipid-regulation programme holds that β-secretase inhibition worsened cognition (Egan et al., 2018) because some production of the fragment is required for a normal cellular function, and that any therapy directed at it must normalise rather than abolish. The competition programme's durable output is *do not remove the monomer*, and the trial record is consistent with it.

The contradiction is real but it is narrower than it looks, and the narrowing is informative. The retention programme's evidence concerns *acute, partial* suppression in aged neurons, where the

manipulation reduces a retained pool; the trial evidence concerns *chronic, near-complete* suppression in a whole brain, where it removes a physiological function. If both are correct, the therapeutic window for suppressing production is narrow, bounded below by loss of function and above by no effect — which is a much less attractive target than either programme's summary implies, and an argument for acting on retrieval and disposal instead.

26.6 Is culling a losing neuron the disease or the defence?

The competition framework holds that the disease is the failure of a protective competition programme, so that the elimination of neurons and synapses is pathological. In the fly amyloid model, culling less fit neurons is **protective** — blocking the culling worsens the outcome (Coelho et al., 2018) — and forcing competition also worsens it (Costa-Rodrigues et al., 2025).

Both cannot be a general statement about competition. The available reconciliation is that culling is protective when the tissue can replace or re-route around what it removes and harmful when it cannot, which makes the sign a function of the tissue's remaining capacity rather than of the mechanism. That reconciliation is untested and is offered as a hypothesis, not a resolution.

26.7 Does tau pathology require amyloid?

The surface-proteolysis programme's second arm proposes a route to *de novo* endogenous tau filaments that requires only age and apolipoprotein E isoform, with no amyloid intermediate (Paradise et al., 2026). The human autopsy record has appeared to require such a route since pre-tangles were documented in the locus coeruleus of young adults with no cortical amyloid (Braak and Del Tredici, 2011; Braak et al., 2011). Against this, the retrieval programme's most consequential recent result shows that raising a trafficking receptor reverses established tau pathology in an animal with no amyloid manipulation (Huang H. et al., 2026) — which supports an amyloid-independent tau arm but places its control at a different molecule; and a large body of work has amyloid driving tau through microglia (Ising et al., 2019; Leyns et al., 2019).

This paper takes no position beyond the observation of Chapter 17: the amyloid-independent mechanisms carry the human genetics, and the amyloid-gated ones carry the interventional demonstrations, and both facts are consistent with a two-clock model in which neither arm is dispensable.

26.8 A summary of what is unresolved

Table 12 — Six contradictions, and the study that would settle each

Contradiction	Position A	Position B	Adjudicating study
APP-dependence of the *SORL1* endosomal phenotype	Independent (Knupp et al., 2020)	Relieved by APP reduction (Hung et al., 2021)	Matched *SORL1* lesion depth and differentiation protocol, with graded APP reduction and compartment-resolved pH
Entry point into the endosomal jam	Retrieval fails first	The fragment fails retrieval	Reciprocal rescue between the retromer-deficient and rab5-over-activated animals
Ligand identity at the inhibitory receptor	Amyloid-β is a ligand (Kim et al., 2013)	No binding to brain-derived oligomers (Smith et al., 2019)	Repeat the standardised comparison with C4d included, and with brain-derived material from staged cases
Sign of production suppression	Reduce production	Do not remove the monomer	Graded, reversible suppression with compartment-resolved read-out of the retained pool
Sign of cell competition	Culling is the disease	Culling is protective	Culling manipulation with tissue-capacity as an explicit variable
Amyloid-dependence of tau initiation	Amyloid-independent route exists	Tau is driven through amyloid and microglia	Surface-proteasome measurement in human locus coeruleus across age and genotype in amyloid-negative brains

27. The Unperformed Adjudications

27.1 A general observation about this literature

Assembling nine programmes makes visible a class of experiment that is invisible from within any one of them: the experiment that would decide between two programmes, which neither has an incentive to run and which usually requires only reagents that both already possess.

Eleven such experiments have been named in the preceding chapters. They are collected here, and ranked in Chapter 30 by what they would settle per unit of effort. What they have in common is that each is a *cross*: a manipulation from one programme applied to a preparation from another.

27.2 The crosses

Table 13 — Eleven crosses between programmes

Cross	Programme supplying the manipulation	Programme supplying the preparation	What it settles
Acidify the endosome in neurons loaded with peroxidised apolipoprotein E	Nixon, external	Ramsden	Whether the recycling block is covalent or pH-dependent
Deplete membrane cholesterol; measure surface proteasome	Head, Rappoport	Margolis	Whether the membrane proteasome is a domain resident
Restore ordered-domain assembly in ApoE4 neurons; measure the tau threshold	Head	Margolis	Whether the apolipoprotein E effect on tau is a membrane effect
Co-manipulate reelin and LilrB2 signalling; measure phospho-cofilin	Ramsden	Shatz and Brott	Whether the two inputs to serine 3 antagonise
Retromer enhancement in the rab5-over-activated animal	Small	Nixon, external	Which model of the endosomal jam is primary

Cross	Programme supplying the manipulation	Programme supplying the preparation	What it settles
Block PirB cleavage; measure retromer-dependent cargo recycling	Shatz and Brott	Small	Whether the two lesions share a retrograde route
C4d minipump into microglia-depleted cortex	Shatz and Brott	Stevens, external	The relative contribution of the neuronal and glial elimination arms
Graded activity manipulation with simultaneous read-out of surface neprilysin, retained pool, and nascent tau	Gouras	Margolis	Whether the activity–clearance coupling sets the sign of activity
Antibody administration with intracellular pool and fragment as the read-out, not plaque	Gouras	Trial literature	Whether removing the external pool raises production
Measure contact-site function and detergent-resistant plasma membrane together under apolipoprotein-E deprivation	Area-Gómez	Rappoport, Head	Whether one supply failure degrades all ordered domains
Surface proteasome in human locus coeruleus by age and genotype, in amyloid-negative brains	Margolis	Human neuropathology	Whether the disposal lesion precedes amyloid in the first-affected neurons

27.3 Why they have not been done

Three reasons, and none is discreditable.

They are nobody's paper. A cross between two programmes produces a result that is a footnote for both and a headline for neither. The literature's incentives reward depth within a programme.

They require two sets of expertise. Compartment-resolved pH measurement, raft fractionation, impermeant-substrate proteolysis assays, array tomography and stereotaxic minipump surgery are each ordinary within one laboratory and unusual across two.

Several were not visible until recently. Five of the eleven depend on results published in 2025 or 2026. The cleaved-receptor cross depends on a finding months old. The membrane-proteasome cross depends on a paper published this year.

That last point is the substantive one. The reason these eleven experiments are available now, and were not available three years ago, is that the programmes have converged — not by design and not by collaboration, but by each following its own object until it arrived at a compartment another was already studying.

Part Six — Consequences

What follows for treatment, for measurement, and for what should be done next; the graded ledger; and the conditions under which the integration would be wrong.

28. Therapy: What Follows From a Return-Leg Model

28.1 The trial record read as evidence

The clinical record in this disease is usually presented as a series of disappointments. Read against the frame of Chapter 14 it is something more useful: a set of directional experiments in humans, whose results have a pattern.

Table 14 — Interventions sorted by which leg of the cycle they act on

Intervention	Leg acted on	Direction	Result
γ-Secretase inhibition (semagacestat)	Forward — but the enzyme is the *clearance* step for C99	Suppress	Worse than placebo (Doody et al., 2013)
β-Secretase inhibition (verubecestat)	Forward — production of the fragment	Suppress	Worse than placebo; rapid, reversible, non-progressive brain-volume reduction (Egan et al., 2018; Sur et al., 2020)
Monomer-binding antibody (solanezumab)	Forward — removes the soluble species	Suppress	Null in preclinical disease (Sperling et al., 2023)
Monomer-and-oligomer antibody (crenezumab)	Forward	Suppress	Null in autosomal-dominant carriers (Tariot et al., 2026)
Aggregate-selective antibodies (lecanemab, donanemab)	Removes the deposited reservoir; relatively monomer-sparing	Remove product	Positive; roughly a quarter slowing over eighteen months, larger at lower baseline tau (van Dyck et al., 2023; Sims et al., 2023)
TREM2 agonist (AL002)	Forward — microglial recognition and uptake	Drive	Null with confirmed target engagement (Mummery et al., 2026)
Chronic synaptic inhibition (benzodiazepine; deafferentation)	Suppresses the forward leg and the one activity-driven return leg	Suppress	Fewer plaques, worse synapses and memory — in mice (Tampellini et al., 2010)

Intervention	Leg acted on	Direction	Result
Nerve growth factor delivery	Supplies ligand to a platform that may not receive it	Add ligand	No clinical benefit; delivery geometry implicated (Rafii et al., 2018; Castle et al., 2020)
Retromer stabilisation	**Return — retrieval**	Restore	No clinical trial in any indication; positive in an independent animal study (Ramonet et al., 2025)
SORLA up-regulation	**Return — retrieval**	Restore	Reverses established tau pathology in aged animals (Huang H. et al., 2026); human programme at conference-abstract stage
Membrane platform restoration	**Return — reception**	Restore	Function preserved against unchanged pathology in five models; no human data
Surface neprilysin restoration	**Return — surface degradation**	Restore	Never attempted
Endosomal re-acidification	**Return — ligand release and retrieval**	Restore	Never attempted in this indication

The pattern is not subtle. **Every intervention that suppressed a forward leg was null or harmful. The one class that produced benefit removes an accumulated product without suppressing its production. Not one intervention that restores a return leg has been tested in a human being.**

That is the paper's principal therapeutic claim, and it does not depend on any of the mechanistic joins in Part Four being correct. It depends only on the classification in the first column, which is a matter of cell biology rather than of theory.

28.2 Three design rules

Rule one: normalise, do not abolish. Every forward-leg species in this circuit has a physiological function. The precursor protein's fragment is a cholesterol signal. Amyloid-β monomer restrains inflammation and supports plasticity at picomolar concentrations. Cofilin activity is required for structural plasticity. The inhibitory receptor is required for normal circuit refinement. Complement is required for developmental pruning. A therapeutic window bounded below by loss of function and above by no effect is a narrow window, and three of the trials in Table 14 fell below it.

Rule two: expect partial efficacy from any monotherapy, and design for it. Four upstream arms converge on the actin machine (Chapter 18); four partly independent routes run from a loaded compartment to a failing synapse (Chapter 8); at least two effector limbs — one neuronal, one glial — execute synapse elimination (Chapter 11). Redundancy of this degree predicts that blocking one arm leaves the outcome reachable by the others. The correct inference is not that the mechanisms are wrong but that trials powered for a large single-agent effect are mis-specified.

Rule three: the target is capacity, not deposit. The five-model dissociation of Chapter 9 — function preserved with pathology unchanged — is the experimental counterpart of human resilience, in which between a fifth and a third of cognitively intact older people carry substantial pathology at autopsy (Dubois et al., 2016). Both say that pathology is a hazard whose translation into dementia is modifiable, and that the modifier acts at the synapse. A therapy developed against capacity is dosed to tolerability, measured by structural substrate, expected to work across aetiologies, and expected to work better the earlier it is given. That is a different trial from one developed against mechanism, and the two should not be run as though they were the same.

28.3 The return-leg target list, ranked by tractability

Restoring endosomal retrieval. The best-anchored target in the set. Human genetics with a causal architecture; depletion–repletion reversal in animals; independent replication of a pharmacological strategy; and a newly identified regulated phosphorylation site with an upstream kinase, which converts the problem from "stabilise a multiprotein complex" — hard and unprecedented — into "inhibit a kinase", which is among the best-trodden paths in drug development (Qureshi et al., 2026, preprint). The obstacle is not chemistry; it is that a pathway-directed agent has no enrolment biomarker and no established endpoint, and the trial infrastructure built over fifteen years is built around amyloid.

Restoring endosomal acidification. Never attempted in this indication and unusually well motivated: a single manipulation that would act on ligand release, on receptor recycling, and on the degradative arm simultaneously, and whose target — the vacuolar ATPase and its inhibition by a phosphorylated fragment — is molecularly specified (Im et al., 2023). The obstacle is selectivity, since acidification is required everywhere.

Restoring the activity–clearance coupling. The specific lesion is loss of surface neprilysin in diseased neurons (Tampellini et al., 2011). Restoring the enzyme, or the signalling that recruits it, targets retention at the step shown to fail. It has the additional attraction of being reachable behaviourally as well as pharmacologically, if the coupling is intact.

Restoring membrane platform assembly. Deliverable — 178 amino acids, within the packaging capacity of a viral vector — and demonstrated across five models and three routes. The obstacles are the absence of any target-engagement measure, an unmeasured intracellular amyloid

pool, and a delivery result whose central tropism depends on a receptor with no human orthologue.

Protecting the disposal step. The autocatalytic route runs through inhibition of the proteasome and the de-ubiquitinating enzymes (Almeida et al., 2006), and the surface proteasome declines with age and genotype (Paradise et al., 2026). Preserving disposal capacity should slow a feed-forward loop rather than merely reducing its input. No agent exists that raises surface proteasome abundance, and identifying one is a screen rather than a programme.

Releasing the restraint. Antagonists of the inhibitory receptor are the most pharmacologically advanced option, with sub-nanomolar antibodies described and related agents through early-phase oncology trials — but as peripherally targeted myeloid agents with no assumed central exposure. The double edge must be stated: every blockade result in that literature is an *increase* in plasticity and an impairment of long-term depression, and nobody has measured the cost of removing a developmental brake over years. If the cleavage finding of Chapter 21 holds, blocking the protease may matter more than blocking the ligand, and they are different molecules.

28.4 The combination argument, stated conservatively

The redundancy documented here is an argument for combination therapy, and it is worth being careful about how strong an argument it is.

It is not an argument that any two of these agents will be additive. It is an argument that the *ceiling* on any single agent is set by the number of parallel arms reaching the same effector, and that the observed ceiling — roughly a quarter slowing from near-complete removal of the best-validated target — is what a redundant architecture predicts. Two agents acting on arms that converge on one effector should be sub-additive. Two agents acting on *different* legs of the same cycle — one restoring retrieval and one restoring disposal — have no such constraint, and that is the combination the frame recommends.

The nearest available test is also the cheapest: give an anti-amyloid antibody in an animal model and measure the intraneuronal pool and the precursor-protein fragment rather than plaque burden. If removing the external pool raises production in cells that already carry an internal load, as the homeostat data predict (Roos et al., 2021), then antibody therapy and a retention-directed agent are a rational pair, and the pairing has a mechanistic rationale rather than a statistical one.

28.5 What the frame says about prevention

If the first clock of Chapter 17 is a decline in recovery capacity that begins in early adulthood, then the interventions with the best expected value are those that act on capacity before the amyloid-gated mechanisms engage. Three are already supported by evidence of the kind this paper weights heavily.

Lowering the peroxidisable substrate. The peroxidation programme's other half is three decades of dietary trials — lowering linoleic acid reduces bioactive oxidised metabolites in humans (Ramsden et al., 2012), with randomised evidence in other indications (Ramsden et al., 2021; Zamora et al., 2025). Whether this transfers to the brain is unknown, and the direct trials of omega-3 supplementation in established disease were null (Quinn et al., 2010), which is what a capacity model predicts for a late intervention.

Preserving sleep. If sleep is the period during which return legs run (Chapter 22), then sleep is not a lifestyle variable but the scheduled maintenance of the system this paper describes. The evidence that it clears metabolites, that its disruption raises amyloid-β in humans within one night, and that synapses are structurally scaled down during it is strong and directly relevant.

Maintaining the activity–clearance coupling. Cognitive and physical engagement, on this reading, work by buying enzymatic clearance at the synapse rather than only by adding network redundancy. That predicts a stage dependence — larger benefit earlier — which is what the epidemiology shows.

None of these is a discovery and none is offered as one. What the frame adds is a mechanism for why they act early and not late, and a reason to expect that trials of them in established disease will be null.

29. Biomarkers for a Cycle

29.1 The mismatch

Every biomarker in clinical use in this disease reports a *product*: the deposited pool by positron-emission tomography, the soluble extracellular pool in fluid, phosphorylated tau species in fluid, and downstream neurodegeneration by structural imaging and neurofilament. Each of them is a measurement of the forward leg's output.

The framework assembled here locates the lesion on the return leg. Not one clinical biomarker measures a return leg.

This is not a rhetorical point. It has three specific consequences. Trials select patients on a quantity the mechanism says is not the relevant one. Target engagement for any return-leg agent cannot currently be demonstrated. And the first clock of Chapter 17 — the decline in recovery capacity that the frame says determines outcome — is invisible in every dataset the field has assembled.

29.2 What a return-leg biomarker would have to do

Three properties, and they are demanding.

Compartment resolution. The quantities at issue are defined per compartment: retained versus exported amyloid, raft versus non-raft cholesterol, surface versus total proteasome, endosomal versus lysosomal pH. A bulk measurement of any of them returns the sum of two pools that move in opposite directions, which is the general finding of Chapter 24 and the source of all three sign problems of Chapter 25.

Rate rather than level. A return leg is a flux. Its failure is a reduced rate, and a reduced rate is compatible with an elevated, normal or reduced steady-state level depending on what the forward leg is doing. Every current biomarker measures a level.

Sensitivity in the preclinical window. A capacity that declines from early adulthood must be measurable decades before symptoms, or the intervention it licenses cannot be timed.

29.3 The five candidate measurements, with what exists

A proxy for the retained intraneuronal pool. The single most valuable missing measurement, because it is the quantity that couples the two clocks. Three routes are conceivable. Neuron-derived extracellular vesicles isolated from plasma, assayed for the retained species and the precursor fragment, would report an intracellular compartment through a peripheral sample. A conformation-sensitive ligand with cell-permeant access would report structure rather than sequence — the antibody-independent infrared work establishes that structural states change before deposition (Klementieva et al., 2017), and the question is whether a scanner-compatible analogue is possible. And a tracer with intracellular access would report the pool directly, which is the hardest of the three. **Status: none exists; the field's standard immunoassay systematically under-reports the assembled species (Stenh et al., 2005).**

A conformation-resolved amyloid-β assay in human fluid. Required to operationalise the one durable therapeutic statement in the competition programme. Without the ability to distinguish monomer from assemblies at the concentrations that occur in vivo, "do not remove the monomer" cannot be turned into an enrolment criterion or a dose-selection rule. **Status: partially available in research settings; not standardised, not clinical.**

A raft-pool lipid measurement in human tissue. Direct imaging of ordered domains in human neurons under *APOE* genotype exists and has produced results that contradict a major theory (Lee et al., 2021; Lee et al., 2025). Applying it systematically to post-mortem human cortex with cell-type resolution would settle two of the three sign problems at once. **Status: method exists; the human tissue study has not been done.**

A pathway-function panel for endosomal retrieval. The most developed of the five. Soluble SORL1 in cerebrospinal fluid behaves as predicted in variant carriers and, importantly, did *not* distinguish sporadic patients from controls (de Waal et al., 2026); cerebrospinal-fluid correlates of retromer-dependent traffic are elevated in a majority of prodromal individuals (Simoes et al., 2020); and a cell-based shedding reporter calibrates both against variant severity (Fazeli et al., 2026). **Status: the components exist. What is required is their application across a large, longitudinally followed cohort stratified by genotype and amyloid status.** That study answers the word *common* in either direction, and no trial of a retrieval-directed agent should be designed before it is answered.

A target-engagement measure for membrane organisation. No candidate is close. The nearest human-scanner readout is an ultrashort-echo-time magnetisation-transfer myelin measure (Wang J. et al., 2026), which reports a downstream structural consequence rather than engagement. **Status: absent, and it is the specific reason a demonstrated preclinical therapy has not moved.**

29.4 An interim proposal

Waiting for five new assays is not a plan. Two things could be done now with existing material.

Re-analyse completed trials for return-leg read-outs. The anti-amyloid trials collected cerebrospinal fluid. Some of that material could be assayed for the precursor-protein fragment, for the retromer-traffic correlates, and for soluble SORL1. The specific question is whether removing the external pool raised the fragment — which is the human version of the homeostat prediction, and which no trial has asked.

Stratify existing cohorts by capacity proxies rather than by burden. Age, *APOE* genotype, and sleep quality are available in most large cohorts and are, on the frame of Chapter 17, three partial indices of the first clock. Testing whether they predict the *conversion* of a given pathology burden into cognitive decline, rather than predicting burden itself, is an analysis of existing data.

30. A Ranked Experimental Programme

30.1 The ranking criterion

The experiments below are ranked by what they would settle divided by what they would cost. Cost is judged by whether new reagents, new methods or new cohorts are required. Three of the first five require no new reagents at all, and the first requires no new experiment.

30.2 The programme

1. Deconvolute *CAV1* expression by cell type from existing single-nucleus data. Settles sign problem two. Requires public repositories and an afternoon. No new tissue, no new reagents. Ranked first because it is the only item that could be completed before this paper is read.

2. Fractionate neuronal membrane and ask whether active surface proteasome co-purifies with detergent-resistant membrane, by *APOE* genotype and age. Settles whether the membrane proteasome is a domain resident (Chapter 19), and with it whether the apolipoprotein E effect on tau is a membrane effect. Standard gradient fractionation plus an impermeant substrate assay, both published.

3. Acidify the endosome in neurons loaded with peroxidised apolipoprotein E particles, and measure receptor recovery. Discriminates the covalent, conformational and acidification mechanisms of the recycling block in a single experiment with three arms (Chapter 20). All reagents published.

4. Co-manipulate reelin and LilrB2 signalling in the same neurons and measure phospho-cofilin at serine 3. Tests the antagonism at the shared effector (Chapter 18). Two published pathways, one readout, no new reagents.

5. Apply the pathway-function panel across a large, longitudinally followed cohort stratified by *SORL1* genotype and amyloid status. Answers whether retrieval failure is *common* in sporadic disease — the question on which the largest single therapeutic programme in this set depends. Expensive, but the assays exist and the cohorts exist.

6. Measure surface proteasome abundance in human locus coeruleus, entorhinal cortex and hippocampus across the adult age range, stratified by *APOE* genotype, in brains without tau pathology. Tests whether the disposal lesion is present in the first-affected neurons before pathology. Requires only tissue and a validated antibody, and would place the amyloid-independent clock in the region the autopsy record implicates first.

7. Deliver C4d by minipump into a microglia-depleted cortex. Measures the relative contribution of the neuron-intrinsic and glial synapse-elimination arms, which nobody has quantified. Two published techniques, one cross.

8. Test reciprocal rescue between the retromer-deficient and rab5-over-activated animals. Decides which model of the endosomal jam is primary (Chapter 26). Both animals exist; the cross has not been made.

9. Administer an anti-amyloid antibody to a knock-in model and measure the retained intraneuronal pool and the precursor-protein fragment rather than plaque burden. Tests the homeostat prediction that removing the external pool raises production in loaded cells, which bears directly on combination design (Chapter 28).

10. Deprive neurons of apolipoprotein-E-borne cholesterol and measure contact-site function and detergent-resistant plasma-membrane fraction together. Tests the claim that one supply failure degrades all ordered domains coordinately (Chapter 16) — the load-bearing assumption of the one-surface argument.

11. Immunoprecipitate apolipoprotein E from frozen human brain under non-reducing conditions and look for the receptor by mass spectrometry. Would convert the peroxidation framework's central chemical claim from demonstrated-in-vitro to demonstrated-in-humans, or fail to. It is the single experiment that most changes the standing of one of the nine programmes, and it has not been reported.

12. Perform cryo-electron microscopy on the filaments produced by surface-proteasome inhibition. Establishes whether they adopt the Alzheimer fold, which is the modern definition of the lesion the model claims to produce.

13. Repeat the standardised comparison of amyloid receptors, with C4d included and with brain-derived material from staged cases. Addresses a seven-year-old unanswered result at the centre of the restraint programme's older arm.

14. Run a dose–response of the membrane-platform intervention against raft occupancy rather than against a single vector dose. Distinguishes the convergence reading from the non-specific reading of a therapy that works in four aetiologies (Chapter 9).

30.3 What the programme would look like if it succeeded

If items two, three and four returned positive results, three of the nine programmes would be joined at the level of mechanism rather than of analogy, and the apolipoprotein E effect on tau, on receptor recycling and on spine stability would have a single membrane-organisational explanation. If item five returned negative, the largest therapeutic programme in the set would reduce in scope to a rare monogenic subtype plus a druggable modifier — which would still be a substantial achievement, and would redirect a considerable amount of effort. If item six returned positive, the amyloid-independent clock would have a measurement in the first-affected human neurons, which the field has never had.

Any of those outcomes would be worth more than another descriptive study of any one programme.

31. The Integrated Ledger

Every substantive claim in this paper appears below with its strongest support, its grade on the scale of Chapter 3, and what is missing. Claims original to this paper are marked with a dagger and are graded conservatively.

Table 15 — The graded ledger

Claim	Strongest evidence	Grade	What is missing
Synapse loss is the best structural correlate of cognitive decline	Biopsy and post-mortem series; in vivo synaptic-vesicle imaging (DeKosky and Scheff, 1990; Terry et al., 1991; Mecca et al., 2020)	**Established**	—
Loss of retromer-dependent recycling is sufficient to produce Alzheimer-type neuronal pathology	Depletion and repletion of VPS35 reverses the phenotype in both directions (Qureshi et al., 2022)	**Established**	Magnitude of naturally occurring deficiency in sporadic disease unquantified
***SORL1* loss of function causes the disease in humans**	Truncating variants almost exclusively in cases; domain-level classification (Andersen, de Waal et al., 2025)	**Established**	Accounts for a small fraction of cases
Retrieval failure is *common* in sporadic disease	Elevated traffic correlates in prodromal individuals (Simoes et al., 2020)	**Weak**	The direct assay does not distinguish sporadic patients from controls (de Waal et al., 2026)
The pathway sits upstream of tau, not only of amyloid	SORLA up-regulation reverses established tau pathology in aged animals; deletion worsens it (Huang H. et al., 2026)	**Strong**	One model; a tauopathy model rather than sporadic disease
Amyloid-β retention rather than overproduction is the pathogenic variable	Secretion falls and surface neprilysin is lost in diseased neurons (Tampellini et al., 2011)	**Strong**	Single laboratory; not independently replicated

Claim	Strongest evidence	Grade	What is missing
Four partly independent routes run from a loaded compartment to a failing synapse	Interventional evidence in both directions on each route	**Strong**	None demonstrated in human tissue
Chronic reduction of synaptic activity worsens outcome while lowering plaques	Designed dissociation with two independent means of inhibition (Tampellini et al., 2010)	**Strong**	Mouse only; over-expressing model
C99 accumulation drives contact-site expansion through cholesterol clustering	C99 nucleates detergent-resistant domains and drives cholesterol to the ER (Montesinos et al., 2020)	**Strong**	Human tissue evidence limited
The amyloid-β 42:40 ratio is a surrogate for bilayer thickness	Named enzyme predicted in advance and confirmed (Montesinos et al., 2025, preprint); lipid remodelling modulates processivity (Dawkins et al., 2023)	**Contested**	Competing protein-intrinsic account is well supported and quantitatively unaddressed
Membrane-domain organisation degrades with age, taking receptor localisation with it	Raft localisation of six proteins declines with age (Head et al., 2010)	**Strong**	Human tissue confirmation
Restoring the membrane platform preserves function with pathology unchanged	Five models, three delivery routes, plaque unchanged throughout	**Strong**	No dose–response; intracellular amyloid never measured; no human data
Neuronal caveolin-1 falls in the human disease	Programme's own measurements	**Contested**	Bulk human tissue reports the opposite (Gaudreault et al., 2004; Kang et al., 2006)
Surface membrane proteasome abundance is ApoE-ordered and age-declining	Membrane-impermeant inhibition; genotype-graded thresholds (Paradise et al., 2026)	**Moderate**	Months old; single laboratory group; filament identity not structurally proven
Ephexin5 is a substrate switch, not a RhoA-selective brake	Y361 gating; Cdc42 arm required for spine growth (Petshow et al., 2025)	**Strong**	—
Lowering Ephexin5 is therapeutically advisable	2017 mouse result	**Contested**	*ARHGEF15* loss of function causes a human vasculopathy (Ding et al., 2023)

Claim	Strongest evidence	Grade	What is missing
C4d is a high-affinity LilrB2 ligand that drives neuron-intrinsic spine loss	~3 nM affinity; 4-fold elevated in Alzheimer cortex; minipump strips spines, abolished in PirB-null (Brott et al., 2025)	**Strong**	Single laboratory; relative contribution versus the glial arm unmeasured
Amyloid-β is a ligand for LilrB2 in human brain	Kim et al., 2013	**Contested**	No detectable binding to brain-derived oligomers in a standardised comparison (Smith et al., 2019)
Removing the amyloid precursor protein spares the axon that should have lost	Cell-autonomous sparse deletion; multiple systems (Marik et al., 2016)	**Established**	—
Monomeric amyloid-β restrains microglial inflammatory output	Genetic epistasis in culture; in vivo phenotype by inference (Kwon et al., 2024)	**Moderate**	No demonstration that the monomer activates the pathway in vivo
Aldehyde crosslinking of apolipoprotein E to ApoER2 occurs and resists acid	Mass spectrometry and immunoblot on defined peptides with motif-deletion controls (Ramsden et al., 2022)	**Strong** in vitro	No crosslinked species isolated from human brain
The ApoER2–Dab1 axis is causally upstream of tau in humans	Two independent resistance variants in ligands of these receptors (Arboleda-Velasquez et al., 2019; Lopera et al., 2023); *DAB1* as an *APOE4*-conditional modifier (Bracher-Smith et al., 2022)	**Strong**	Mechanism of each variant not fully specified
Raft assembly is the gate between two stages of plasticity	Theory; the sensory prodrome and its pharmacological triangulation	**Moderate** for the raft dependence; **weak** for the gate	The two-population model renders cholesterol and amyloid measurements non-diagnostic
Astrocyte-to-neuron cholesterol delivery is impaired in the disease and involves ApoE4	Human cerebrospinal-fluid material (Borràs et al., 2025)	**Moderate**	Contradicted in direction by direct raft imaging (Lee et al., 2021; Lee et al., 2025)
Oxidative damage in Alzheimer brain concentrates in lipid rafts	Subcellular fractionation of human tissue; lower raft yield in ε*4* carriers (Thorwald et al., 2025)	**Moderate**	Single study

Claim	Strongest evidence	Grade	What is missing
† **Eight of nine programmes locate their lesion on the recovery step of a cycle, with the productive step up-regulated in compensation at six of thirteen stations**	The tabulation of Chapter 14; six documented instances of forward-leg up-regulation	**Moderate**	The pattern is descriptive; the compensation link is demonstrated at only two stations
† **The plasma-membrane raft, the ER–mitochondrial contact and the synaptic endosome are instances of one physical phenomenon, constrained by one supply**	Shared ordering lipid, shared partitioning rules, shared resident molecules; damage concentrated in the fraction (Thorwald et al., 2025)	**Moderate** for the identity; **inference** for the shared constraint	Coordinated degradation under a single supply failure has not been tested
† **Three programmes write to serine 3 of cofilin, with opposite signs and no shared citations**	Chai et al., 2009; Kim et al., 2013; Ramsden et al., 2022 (phospho-LIM-kinase 1 in human tissue)	**Inference**	No co-manipulation experiment; tissue direction of phospho-LIM-kinase disagrees with the mechanism
† **The neuronal membrane proteasome is a resident of ordered membrane domains**	Shared modifiers (*APOE* order and age) with raft-localised proteins; a proteolipid-family anchor proposed	**Inference**	No fractionation; no cholesterol-manipulation experiment
† **The precursor protein's fragment links the amyloid arm to the lipoprotein-receptor arm by de-acidifying the endosome**	Tyr682-phosphorylated fragment inhibits the vacuolar ATPase (Im et al., 2023); the recycling block is pH-reversible (Xian et al., 2018)	**Inference**	Compartment-resolved pH in early endosomes not measured
† **The cleaved inhibitory-receptor fragment and retromer failure jam one retrograde route**	Fragment binds GGA3-GAT and disrupts Golgi and lysosomal function (Han et al., 2026)	**Inference**	Single unreplicated study; no reciprocal-rescue test; no specificity control
† **The activity–clearance coupling sets the sign of activity in this disease**	Activity lowers the intraneuronal pool via neprilysin (Tampellini et al., 2009); the coupling fails in diseased neurons (Tampellini et al., 2011)	**Inference**	No stage-resolved measurement

Claim	Strongest evidence	Grade	What is missing
† **The microglial lesion is a disposal lesion, and competence should be measured as digestion rather than uptake**	Lipid-droplet and ER-stress phenotypes across four independent studies; the null TREM2-agonist trial (Mummery et al., 2026)	**Inference**	No direct measure of digestive throughput in human microglia by genotype
† **Every one of the nine programmes reports that the standard measurement in its area is a bulk measure of a compartment- or conformation-defined quantity**	Table 10	**Strong** — it is a tabulation of published statements	—
† **Interventions that suppressed a forward leg were null or harmful; no return-leg intervention has been tested in a human**	Table 14	**Strong** for the tabulation; **inference** for the causal reading	Confounded by stage, dose and target selection

32. What Would Refute the Integration

The frame is falsifiable, and the following results would damage or destroy it. They are ordered from most to least decisive.

1. A return-leg intervention that engages its target in a human brain and produces no clinical benefit in appropriately early disease. This is the cleanest available test of the whole structure, and it requires a trial that does not exist. If retrieval, disposal or reception can be demonstrably restored in a person with early disease and nothing follows, then the recovery steps are not where the disease is, and the pattern of Table 7 is a coincidence of how nine laboratories chose to describe their findings.

2. Demonstration that the forward legs are not compensating. The compensation signature is what distinguishes the thesis from a restatement. If the elevated amyloid production in loaded cells, the elevated contact-site function, the elevated C4d and the continued cholesterol synthesis prove to be independent of the corresponding return-step failures — for instance, if blocking the return step in a healthy cell does not raise the forward step — then the frame reduces to the observation that recovery steps are complicated, which is not a finding.

3. A single-agent intervention on one arm that abolishes the whole synaptic phenotype. The redundancy argument of Chapters 18 and 28 predicts partial efficacy from any monotherapy. If blocking one arm removes the phenotype entirely, then the arms are not partly independent, the account should be simplified to one mechanism, and the combination logic is wrong.

4. Compartment-resolved measurement showing the three ordered domains do not share a supply constraint. If depriving a neuron of apolipoprotein-E-borne cholesterol degrades plasma-membrane domains without affecting the ER–mitochondrial contact, or vice versa, the one-surface argument of Chapter 16 fails, and the programmes it joins must be joined some other way or not at all.

5. Failure of the serine-3 convergence to show interaction. If reelin and LilrB2 signalling are additive with no interaction term when co-manipulated in the same neurons, Chapter 18 should be withdrawn, and with it the shared-address reading of the two human resistance variants.

6. Demonstration that the microglial lesion is a recognition lesion after all. If a manipulation that raises microglial digestive throughput without altering uptake fails to improve

outcome, while an uptake-directed manipulation succeeds, the reframing of Chapter 23 is wrong and the null TREM2-agonist result requires a different explanation.

7. Structural refutation of the tau-filament claim. If cryo-electron microscopy shows that the filaments produced by surface-proteasome inhibition do not adopt the Alzheimer fold, the amyloid-independent clock loses its most specific mechanism, and Chapter 17's two-clock model is left with correlates rather than a mechanism on its first clock.

None of these would refute all nine programmes, and that is a property of the integration rather than a defect: the programmes are separately supported, and the frame is a claim about their relationship.

33. Limitations

The selection of nine programmes is a judgement. Programmes principally concerned with tau propagation, with the autophagy–lysosome axis, with vascular contribution, with microglial state transitions and with the perineuronal matrix would each have a claim to inclusion, and each would change the shape of the circuit in Chapter 13. The autophagy–lysosome programme in particular supplies three of the results this paper leans on most heavily while not being among the nine, which is an inconsistency defensible only by the stated selection principle and not by the evidence.

No new data are presented. Every claim rests on published work, and the original contributions are joins and reframings rather than measurements. A join is a hypothesis about two literatures, and hypotheses of that kind have a poor historical record when they are not tested promptly.

Five load-bearing results are preprints. The contact-site cholesterol-homeostasis result, the ACSL4 result, the RhoGEF12 target, the dendritic tau-translation localisation and one of the lipid-signature studies are all unrefereed as of August 2026. Each is flagged at use. The argument would look thinner without them, and that should be held against it rather than absorbed.

Several of the most consequential results are months old. The cleaved-receptor finding, the SORLA tauopathy rescue, the soluble-SORL1 assay, the surface-proteasome tau paper and the microglial SorLA study were all published within the last year. A synthesis built substantially on results that have not had time to be replicated is a synthesis with a short half-life.

The return-leg thesis is a pattern claim. Pattern claims are vulnerable to the way the underlying work is described, and this paper describes nine programmes in its own vocabulary. Chapter 15 states the four places where the pattern fits badly, and one of them — the complement arm — is a genuine exception rather than an awkward fit. A reader who believes the descriptions have been shaped to fit the pattern should test that suspicion against Chapter 15 first, and against Table 7's final column, which quotes each programme's own summary phrase.

Grades are assigned by one reader. The ledger in Chapter 31 is a judgement about strength of evidence, not a formal assessment, and no second assessor has applied the criteria of Chapter 3 independently.

The human evidence remains thin throughout. Of the mechanisms assembled here, most are demonstrated in mice, in cultured neurons, or in stem-cell-derived systems. Where human

evidence exists it is usually descriptive. This is a limitation of the field rather than of the synthesis, but it bears on how much weight the integration can carry.

34. Conclusion

Nine laboratories set out to explain why synapses fail in Alzheimer's disease, and arrived at nine answers that appear to have nothing in common: a sterol, an aldehyde, a proteolytic fragment, a sorting complex, a retained peptide, a scaffolding protein, a surface protease, an inhibitory receptor, a competition signal.

Set them along the itinerary of a lipid and a membrane protein through one neuron and they occupy nine consecutive stations of a single circuit. At each station there is a cycle. At eight of the nine, the reported lesion is on the recovery step — the step that releases, retrieves, clears, disposes, resolves or restabilises — and not on the step that makes. At six of the thirteen stations the productive step is measurably up-regulated, because the cell is compensating for a return that never arrives.

That pattern has three consequences that no single programme could reach.

It explains the trial record without appeal to bad luck. Every intervention that suppressed a productive step was null or harmful; the one class that helped removes an accumulated product without suppressing its production; and no intervention that restores a recovery step has been tested in a human being. That is a statement about cell biology, not about theory, and it is the most immediately actionable thing in this paper.

It identifies the measurement problem as the field's central obstacle, and does so nine times over. Every one of the nine programmes reports, usually in its own limitations, that the standard assay in its area is a bulk measurement of a quantity defined per compartment, per cell type or per conformation. That single methodological fact explains the three sign disputes of Chapter 25 without attributing error to anyone, and it means that the framework assembled here cannot presently be tested in a person. Five assays would change that, and one of them — a proxy for the pool retained inside the neuron — would be worth more than any of the mechanistic claims made here, because it would allow them to be settled.

And it makes the programmes joinable at named residues rather than at the level of analogy. Three of them write to serine 3 of cofilin from three receptors with opposite signs, and no two of those literatures cite each other. A proteasome at the neuronal surface has the apolipoprotein-E order and the age dependence of a membrane-domain protein, and nobody has asked whether it is one. A covalent bond and a failed proton pump produce the same recycling block and are separable by one manipulation. A cleaved receptor tail and a failed retrieval complex jam one retrograde corridor. Each of those is a hypothesis, each is graded here as inference, and each can be tested with

reagents that already exist in two laboratories that have never corresponded.

What the integration does not do is settle the argument about how the disease begins. Eight of the nine programmes present themselves as accounts of initiation, and in eight of nine the evidence is of a different kind — necessity and sufficiency, demonstrated in a laboratory, which is exactly what a claim about a shared mechanism requires and exactly what a claim about temporal priority cannot use. Read as convergence claims they are additive; read as origin stories they can only compete, and the competition cannot be decided with the evidence available. The programmes are, almost uniformly, better than the claims made on their behalf.

The disease those nine programmes describe is not a poisoning. It is a system of recovery mechanisms — retrieval, release, clearance, disposal, resolution, restabilisation — declining slowly from early adulthood at a rate set by genotype, lipid supply and oxidative load, and then consumed rapidly by a set of faster mechanisms that engage once a threshold is crossed. The deposits that define it at autopsy are the products of a cell still trying. What has been treated for thirty years as the cause is, on the reading assembled here, the residue of a compensation, and the thing worth measuring and worth restoring is the leg of the cycle that never completes.

References

All references are to the primary literature. Where a result is a preprint or a conference abstract as of August 2026, that status is stated in the entry and at every point of use in the text. Journal names are given in full; PubMed identifiers are supplied where the source volume recorded them.

Albarran E, Raissi A, Jáidar O, Shatz CJ, Ding JB. Enhancing motor learning by increasing the stability of newly formed dendritic spines in the motor cortex. *Neuron*. 2021;109(20):3298–3311.e4. PMID 34437845.

Almeida CG, Tampellini D, Takahashi RH, Greengard P, Lin MT, Snyder EM, Gouras GK. Beta-amyloid accumulation in APP mutant neurons reduces PSD-95 and GluR1 in synapses. *Neurobiology of Disease*. 2005;20(2):187–198. PMID 16242627.

Almeida CG, Takahashi RH, Gouras GK. Beta-amyloid accumulation impairs multivesicular body sorting by inhibiting the ubiquitin-proteasome system. *Journal of Neuroscience*. 2006;26(16):4277–4288. PMID 16624948.

Andersson E, Schultz N, Saito T, Saido TC, Blennow K, Gouras GK, Zetterberg H, Hansson O. Cerebral Aβ deposition precedes reduced cerebrospinal fluid and serum Aβ42/Aβ40 ratios in the App$^{NL\text{-}F/NL\text{-}F}$ knock-in mouse model of Alzheimer's disease. *Alzheimer's Research & Therapy*. 2023;15(1):64. PMID 36964585.

Arboleda-Velasquez JF, Lopera F, O'Hare M, Delgado-Tirado S, Marino C, Chmielewska N, Saez-Torres KL, Amarnani D, Schultz AP, Sperling RA, Leyton-Cifuentes D, Chen K, Baena A, Aguillon D, Rios-Romenets S, Giraldo M, Guzmán-Vélez E, Norton DJ, Pardilla-Delgado E, Artola A, Sanchez JS, Acosta-Uribe J, Lalli M, Kosik KS, Huentelman MJ, Zetterberg H, Blennow K, Reiman RA, Luo J, Chen Y, Thiyyagura P, Su Y, Jun GR, Naymik M, Gai X, Bootwalla M, Ji J, Shen L, Miller JB, Kim LA, Tariot PN, Johnson KA, Reiman EM, Quiroz YT. Resistance to autosomal dominant Alzheimer's disease in an APOE3 Christchurch homozygote: a case report. *Nature Medicine*. 2019;25(11):1680–1683. PMID 31686034.

Area-Gómez E, de Groof AJ, Boldogh I, Bird TD, Gibson GE, Koehler CM, Yu WH, Duff KE, Yaffe MP, Pon LA, Schon EA. Presenilins are enriched in endoplasmic reticulum membranes associated with mitochondria. *American Journal of Pathology* 2009;175(5):1810–1816. PMID 19834068.

Area-Gómez E, del Carmen Lara Castillo M, Tambini MD, Guardia-Laguarta C, de Groof AJW, Madra M, Ikenouchi J, Umeda M, Bird TD, Sturley SL, Schon EA. Upregulated function of mitochondria-associated ER membranes in Alzheimer disease. *EMBO Journal* 2012;31(21):4106–4123. PMID 22892566.

Area-Gómez E. Assessing the function of mitochondria-associated ER membranes. *Methods in Enzymology* 2014;547:181–197. PMID 25416359.

Atwal JK, Pinkston-Gosse J, Syken J, Stawicki S, Wu Y, Shatz C, Tessier-Lavigne M. PirB is a functional receptor for myelin inhibitors of axonal regeneration. *Science*. 2008;322(5903):967–970. PMID 18988857.

Avchalumov Y, Kreisler AD, Trenet W, Nayak M, Head BP, Piña-Crespo JC, Mandyam CD. Caveolin-1 expression in the dorsal striatum drives methamphetamine addiction-like behavior. *International Journal of Molecular Sciences*. 2021;22(15):8219. PMID 34360984.

Bamburg JR, Bernstein BW. Actin dynamics and cofilin-actin rods in Alzheimer disease. *Cytoskeleton (Hoboken)*. 2016;73(9):477–497. PMID 26873625.

Bamburg JR, Minamide LS, Wiggan O, Tahtamouni LH, Kuhn TB. Cofilin and actin dynamics: multiple modes of regulation and their impacts in neuronal development and degeneration. *Cells*. 2021;10(10):2726. PMID 34685706.

Barger SW, Moerman-Herzog AM. Modulation of apolipoprotein E receptor-2 by ApoE4, amyloid β-peptide, reelin, and secreted amyloid precursor protein: a common point of impact in Alzheimer's disease pathogenesis. *Frontiers in Molecular Neuroscience*. 2026;19:1781541.

Barrett PJ, Song Y, Van Horn WD, Hustedt EJ, Schafer JM, Hadziselimovic A, Beel AJ, Sanders CR. The amyloid precursor protein has a flexible transmembrane domain and binds cholesterol. *Science* 2012;336(6085):1168–1171. PMID 22654059.

Beckelman BC, Day S, Zhou X, Donohue M, Gouras GK, Klann E, Keene CD, Ma T. Dysregulation of elongation factor 1A expression is correlated with synaptic plasticity impairments in Alzheimer's disease. *Journal of Alzheimer's Disease*. 2016;54(2):669–678. PMID 27567813.

Beel AJ, Mobley CK, Kim HJ, Tian F, Hadziselimovic A, Jap B, Prestegard JH, Sanders CR. Structural studies of the transmembrane C-terminal domain of the amyloid precursor protein (APP): does APP function as a cholesterol sensor? *Biochemistry* 2008;47(36):9428–9446. PMID 18702528.

Beel AJ, Sakakura M, Barrett PJ, Sanders CR. Direct binding of cholesterol to the amyloid precursor protein: an important interaction in lipid–Alzheimer's disease relationships? *Biochimica et Biophysica Acta* 2010;1801(8):975–982. PMID 20304095.

Bellenguez C, Küçükali F, Jansen IE, Kleineidam L, et al. New insights into the genetic etiology of Alzheimer's disease and related dementias. *Nature Genetics*. 2022;54(4):412–436. PMID 35379992.

Bochner DN, Sapp RW, Adelson JD, Zhang S, Lee H, Djurisic M, Syken J, Dan Y, Shatz CJ. Blocking PirB up-regulates spines and functional synapses to unlock visual cortical plasticity and facilitate recovery from amblyopia. *Science Translational Medicine*. 2014;6(258):258ra140. PMID 25320232.

Borràs C, Canyelles M, Santos D, et al. Cerebrospinal fluid lipoprotein-mediated cholesterol delivery to neurons is impaired in Alzheimer's disease and involves APOE4. *Journal of Lipid Research*. 2025;66(8):100865.

Braak H, Del Tredici K. The pathological process underlying Alzheimer's disease in individuals under thirty. *Acta Neuropathologica*. 2011;121(2):171–181. PMID 21170538.

Braak H, Thal DR, Ghebremedhin E, Del Tredici K. Stages of the pathologic process in Alzheimer disease: age categories from 1 to 100 years. *Journal of Neuropathology and Experimental Neurology*. 2011;70(11):960–969. PMID 22002422.

Bracher-Smith M, Leonenko G, Baker E, et al. Whole genome analysis in APOE4 homozygotes identifies the DAB1-RELN pathway in Alzheimer's disease pathogenesis. *Neurobiology of Aging*. 2022;119:67–76.

Brott BK, Raissi AJ, Micheva KD, Vielmetter J, Mendes MS, Baccus CJ, Huang J, Shatz CJ. C4d, a high-affinity LilrB2 ligand, is elevated in Alzheimer's disease and mediates synapse pruning. *Proceedings of the National Academy of Sciences USA*. 2025;122(38):e2519253122. PMID 40966293.

Buckner RL, Snyder AZ, Shannon BJ, LaRossa G, Sachs R, Fotenos AF, Sheline YI, Klunk WE, Mathis CA, Morris JC, Mintun MA. Molecular, structural, and functional characterization of Alzheimer's disease: evidence for a relationship between default activity, amyloid, and memory. *Journal of Neuroscience*. 2005;25(34):7709–7717. PMID 16120771.

Burrinha T, Martinsson I, Gomes R, Terrasso AP, Gouras GK, Almeida CG. Upregulation of APP endocytosis by neuronal aging drives amyloid-dependent synapse loss. *Journal of Cell Science*. 2021;134(9):jcs255752. PMID

33910234.

Busche MA, Eichhoff G, Adelsberger H, Abramowski D, Wiederhold KH, Haass C, Staufenbiel M, Konnerth A, Garaschuk O. Clusters of hyperactive neurons near amyloid plaques in a mouse model of Alzheimer's disease. *Science*. 2008;321(5896):1686–1689. PMID 18802001.

Capetillo-Zarate E, Gracia L, Yu F, Banfelder JR, Lin MT, Tampellini D, Gouras GK. High-resolution 3D reconstruction reveals intra-synaptic amyloid fibrils. *American Journal of Pathology*. 2011;179(5):2551–2558. PMID 21925470.

Castle MJ, Baltanás FC, Kovacs I, Nagahara AH, Barba D, Tuszynski MH. Postmortem analysis in a clinical trial of AAV2-NGF gene therapy for Alzheimer's disease identifies a need for improved vector delivery. *Human Gene Therapy*. 2020;31(7–8):415–422. PMID 32126838.

Cataldo AM, Peterhoff CM, Troncoso JC, Gomez-Isla T, Hyman BT, Nixon RA. Endocytic pathway abnormalities precede amyloid beta deposition in sporadic Alzheimer's disease and Down syndrome: differential effects of APOE genotype and presenilin mutations. *American Journal of Pathology*. 2000;157(1):277–286. PMID 10880397.

Chai X, Förster E, Zhao S, Bock HH, Frotscher M. Reelin stabilizes the actin cytoskeleton of neuronal processes by inducing n-cofilin phosphorylation at serine3. *Journal of Neuroscience*. 2009;29(1):288–299. PMID 19129405.

Checler F, Afram E, Pardossi-Piquard R, Lauritzen I. Is γ-secretase a beneficial inactivating enzyme of the toxic APP C-terminal fragment C99? *Journal of Biological Chemistry* 2021;296:100489. PMID 33662398.

Chen Y, Durakoglugil MS, Xian X, Herz J. ApoE4 reduces glutamate receptor function and synaptic plasticity by selectively impairing ApoE receptor recycling. *Proceedings of the National Academy of Sciences USA*. 2010;107(26):12011–12016.

Cirrito JR, Yamada KA, Finn MB, et al. Synaptic activity regulates interstitial fluid amyloid-beta levels in vivo. *Neuron* 2005;48(6):913-22. PMID 16364896.

Cirrito JR, Kang JE, Lee J, et al. Endocytosis is required for synaptic activity-dependent release of amyloid-beta in vivo. *Neuron* 2008;58(1):42-51. PMID 18400162.

Cissé M, Halabisky B, Harris J, Devidze N, Dubal DB, Sun B, Orr A, Lotz G, Kim DH, Hamto P, Ho K, Yu GQ, Mucke L. Reversing EphB2 depletion rescues cognitive functions in Alzheimer model. *Nature*. 2011;469(7328):47–52. PMID 21113149.

Coelho DS, Schwartz S, Merino MM, et al. Culling Less Fit Neurons Protects against Amyloid-β-Induced Brain Damage and Cognitive and Motor Decline. *Cell Reports* 2018;25(13):3661-3673.e3. PMID 30590040.

Colonna M, Holtzman DM. Rethinking TREM2 as a target for Alzheimer's disease after the INVOKE-2 trial failure. *Nature Medicine* 2025;31(10):3217-3218. PMID 40603729.

Corder EH, Saunders AM, Strittmatter WJ, et al. Gene dose of apolipoprotein E type 4 allele and the risk of Alzheimer's disease in late onset families. *Science*. 1993;261(5123):921–923.

Cordy JM, Hussain I, Dingwall C, Hooper NM, Turner AJ. Exclusively targeting beta-secretase to lipid rafts by GPI-anchor addition up-regulates beta-site processing of the amyloid precursor protein. *Proceedings of the National Academy of Sciences USA*. 2003;100(20):11735–11740. PMID 14504402.

da Silva Correia A, Schmitz M, Fischer AL, da Silva Correia S, Simonetti FL, Saher G, Goya-Maldonado R, Arora AS, Fischer A, Outeiro TF, Zerr I. Cellular prion protein acts as mediator of amyloid beta uptake by caveolin-1 causing cellular dysfunctions in vitro and in vivo. *Alzheimer's & Dementia*. 2024;20(10):6776–6792. PMID 39212313.

Corriveau RA, Huh GS, Shatz CJ. Regulation of class I MHC gene expression in the developing and mature CNS by neural activity. *Neuron*. 1998;21(3):505–520. PMID 9768838.

Costa-Rodrigues C, Jacobs JR, Couceiro J, et al. Dietary Interventions Modulate Cell Competition and Locomotor Decline in an Alzheimer's Disease Drosophila Model. *Cells* 2025;14(24). PMID 41440031.

Datwani A, McConnell MJ, Kanold PO, Micheva KD, Busse B, Shamloo M, Smith SJ, Shatz CJ. Classical MHCI molecules regulate retinogeniculate refinement and limit ocular dominance plasticity. *Neuron*. 2009;64(4):463–470. PMID 19945389.

Dawkins E, Derks RJE, Schifferer M, Trambauer J, Winkler E, Simons M, Paquet D, Giera M, Kamp F, Steiner H. Membrane lipid remodeling modulates γ-secretase processivity. *Journal of Biological Chemistry* 2023;299(3):103027. PMID 36805335.

DeKosky ST, Scheff SW. Synapse loss in frontal cortex biopsies in Alzheimer's disease: correlation with cognitive severity. *Annals of Neurology*. 1990;27(5):457–464. PMID 2360787.

Diering GH, Nirujogi RS, Roth RH, et al. Homer1a drives homeostatic scaling-down of excitatory synapses during sleep. *Science* 2017;355(6324):511-515. PMID 28154077.

Ding X, Chen Y, Guo C, Fu Y, Qin C, Zhu Q, Wang J, Zhang R, Tian H, Feng R, Liu H, Liang D, Wang G, Teng J, Li J, Tang B, Wang X. Mutations in ARHGEF15 cause autosomal dominant hereditary cerebral small vessel disease and osteoporotic fracture. *Acta Neuropathologica*. 2023;145(5):681–705. PMID 36929019.

Djurisic M, Vidal GS, Mann M, Aharon A, Kim T, Ferrao Santos A, Zuo Y, Hübener M, Shatz CJ. PirB regulates a structural substrate for cortical plasticity. *Proceedings of the National Academy of Sciences USA*. 2013;110(51):20771–20776. PMID 24302763.

Doody RS, Raman R, Farlow M, et al. A phase 3 trial of semagacestat for treatment of Alzheimer's disease. *New England Journal of Medicine* 2013;369(4):341–350. PMID 23883379.

Dubois B, Hampel H, Feldman HH, Scheltens P, Aisen P, Andrieu S, Bakardjian H, Benali H, Bertram L, Blennow K, Broich K, Cavedo E, Crutch S, Dartigues JF, Duyckaerts C, Epelbaum S, Frisoni GB, Gauthier S, Genthon R, Gouw AA, Habert MO, Holtzman DM, Kivipelto M, Lista S, Molinuevo JL, O'Bryant SE, Rabinovici GD, Rowe C, Salloway S, Schneider LS, Sperling R, Teichmann M, Carrillo MC, Cummings J, Jack CR Jr; Proceedings of the Meeting of the International Working Group and the American Alzheimer's Association on "The Preclinical State of AD". Preclinical Alzheimer's disease: definition, natural history, and diagnostic criteria. *Alzheimer's & Dementia*. 2016;12(3):292–323. PMID 27012484.

van Dyck CH, Swanson CJ, Aisen P, Bateman RJ, Chen C, Gee M, Kanekiyo M, Li D, Reyderman L, Cohen S, Froelich L, Katayama S, Sabbagh M, Vellas B, Watson D, Dhadda S, Irizarry M, Kramer LD, Iwatsubo T. Lecanemab in early Alzheimer's disease. *New England Journal of Medicine*. 2023;388(1):9–21. PMID 36449413.

Edgar JR, Willén K, Gouras GK, Futter CE. ESCRTs regulate amyloid precursor protein sorting in multivesicular bodies and intracellular amyloid-β accumulation. *Journal of Cell Science*. 2015;128(14):2520–2528. PMID 26002056.

Egan MF, Kost J, Tariot PN, et al. Randomized trial of verubecestat for mild-to-moderate Alzheimer's disease. *New England Journal of Medicine* 2018;378(18):1691–1703. PMID 29719179.

Egawa J, Pearn ML, Lemkuil BP, Patel PM, Head BP. Membrane lipid rafts and neurobiology: age-related changes in membrane lipids and loss of neuronal function. *Journal of Physiology*. 2016;594(16):4565–4579. PMID 26332795.

Egawa J, Schilling JM, Cui W, Posadas E, Sawada A, Alas B, Zemljic-Harpf AE, Fannon-Pavlich MJ, Mandyam CD, Roth DM, Patel HH, Patel PM, Head BP. Neuron-specific caveolin-1 overexpression improves motor function and preserves memory in mice subjected to brain trauma. *FASEB Journal*. 2017;31(8):3403–3411. PMID 28450301.

Egawa J, Zemljic-Harpf A, Mandyam CD, Niesman IR, Lysenko LV, Kleschevnikov AM, Roth DM, Patel HH, Patel PM, Head BP. Neuron-targeted caveolin-1 promotes ultrastructural and functional hippocampal synaptic plasticity. *Cerebral Cortex*. 2018;28(9):3255–3266. PMID 28981594.

Ehehalt R, Keller P, Haass C, Thiele C, Simons K. Amyloidogenic processing of the Alzheimer beta-amyloid precursor protein depends on lipid rafts. *Journal of Cell Biology*. 2003;160(1):113–123. PMID 12515826.

Espay AJ, Herrup K, Kepp KP, et al. The proteinopenia hypothesis: Loss of Aβ42 and the onset of Alzheimer's Disease. *Ageing Research Reviews* 2023;92:102112. PMID 38270185.

Espay AJ, Ezzat K, Kepp KP, et al. Restoring amyloid-β42 and γ-secretase function in Alzheimer's disease. *Brain* 2025;148(11):3856-3864. PMID 40795314.

Etxebeste-Mitxeltorena M, Flores-Romero H, Ramos-Inza S, Masiá E, Nenchova M, Montesinos J, Martinez-Gonzalez L, Porras G, Orzáez M, Vicent MJ, Gil C, Area-Gómez E, Garcia-Saez AJ, Martinez A. Modulation of mitochondria–endoplasmic reticulum contacts (MERCs) by small molecules as a new strategy for restoring lipid metabolism in an amyotrophic lateral sclerosis model. *Journal of Medicinal Chemistry* 2025;68(2):1179–1194. PMID 39778888.

Fazeli E, Jan A, Huber AK, Jensen AMG, Fazeli E, Klein J, Merchant K, Andersen OM. A luminescent reporter assay to quantify SORL1 ectodomain shedding and retromer-dependent endosome recycling activity. *Journal of Biological Chemistry*. 2026;302(2):111136. PMID 41519200.

Fitzpatrick AWP, Falcon B, He S, Murzin AG, Murshudov G, Garringer HJ, Crowther RA, Ghetti B, Goedert M, Scheres SHW. Cryo-EM structures of tau filaments from Alzheimer's disease. *Nature*. 2017;547(7662):185–190. PMID 28678775.

Fortea J, Pegueroles J, Alcolea D, et al. APOE4 homozygosity represents a distinct genetic form of Alzheimer's disease. *Nature Medicine*. 2024;30(5):1284–1291. PMID 38710950.

Fridolfsson HN, Kawaraguchi Y, Ali SS, Panneerselvam M, Niesman IR, Finley JC, Kellerhals SE, Migita MY, Okada H, Moreno AL, Jennings M, Kidd MW, Bonds JA, Balijepalli RC, Ross RS, Patel PM, Miyanohara A, Chen Q, Lesnefsky EJ, Head BP, Roth DM, Insel PA, Patel HH. Mitochondria-localized caveolin in adaptation to cellular stress and injury. *FASEB Journal*. 2012;26(11):4637–4649. PMID 22859372.

García-Toledo I, Godoy-Corchuelo JM, Fernández-Beltrán LC, Ali Z, Guindo-Arroyo A, Jiménez-Coca I, Jiménez-Rodríguez J, Javaloyes-García K, Viñuela M, Gómez-Pinedo U, Saiz-Aúz L, Rábano A, Area-Gómez E, Cunningham TJ, Corrochano S. TDP-43 dysregulation impairs cholesterol metabolism linked with myelination defects. *Acta Neuropathologica* 2025;150(1):23. PMID 40906043.

Gaudreault SB, Dea D, Poirier J. Increased caveolin-1 expression in Alzheimer's disease brain. *Neurobiology of Aging*. 2004;25(6):753–759. PMID 15165700.

Gorniak-Walas M, Telugu NS, Rudolph IM, Diecke S, Willnow TE. Alzheimer's disease risk single nucleotide polymorphism rs11218343 is linked to functional expression of SORL1 in microglia. *Alzheimer's & Dementia*. 2026;22(4):e71390. PMID 42002722.

Gouras GK, Tsai J, Naslund J, Vincent B, Edgar M, Checler F, Greenfield JP, Haroutunian V, Buxbaum JD, Xu H, Greengard P, Relkin NR. Intraneuronal Aβ42 accumulation in human brain. *American Journal of Pathology*. 2000;156(1):15–20. PMID 10623648.

Gouras GK, Willén K, Tampellini D. Critical role of intraneuronal Aβ in Alzheimer's disease: technical challenges in studying intracellular Aβ. *Life Sciences*. 2012;91(23–24):1153–1158. PMID 22727791.

Greenfield JP, Tsai J, Gouras GK, Hai B, Thinakaran G, Checler F, Sisodia SS, Greengard P, Xu H. Endoplasmic reticulum and trans-Golgi network generate distinct populations of Alzheimer beta-amyloid peptides. *Proceedings of the National Academy of Sciences USA*. 1999;96(2):742–747. PMID 9892704.

Guardia-Laguarta C, Area-Gómez E, Rüb C, Liu Y, Magrané J, Becker D, Voos W, Schon EA, Przedborski S. α-Synuclein is localized to mitochondria-associated ER membranes. *Journal of Neuroscience* 2014;34(1):249–259. PMID 24381286.

Gulisano W, Melone M, Ripoli C, et al. Neuromodulatory Action of Picomolar Extracellular Aβ42 Oligomers on Presynaptic and Postsynaptic Mechanisms Underlying Synaptic Function and Memory. *Journal of Neuroscience* 2019;39(30):5986-6000. PMID 31127002.

Guo X, Li H, Yan C, Lei J, Zhou R, Shi Y. Molecular mechanism of substrate recognition and cleavage by human γ-secretase. *Science* 2024;384(6699):1091–1095. PMID 38843321.

Guo JL, Braun D, Fitzgerald GA, et al. Decreased lipidated ApoE-receptor interactions confer protection against pathogenicity of ApoE and its lipid cargoes in lysosomes. *Cell*. 2025;188(1):187–206.e26.

Hamilton AM, Lambert JT, Parajuli LK, Vivas O, Park DK, Stein IS, Jahncke JN, Greenberg ME, Margolis SS, Zito K. A dual role for the RhoGEF Ephexin5 in regulation of dendritic spine outgrowth. *Molecular and Cellular Neuroscience*. 2017;80:66–74. PMID 28185854.

Han WB, Liu XD, Tang CF, Zheng J, Xu TL, Xu NJ, Sun S. Persistent PirB cleavage drives Golgi-directed trafficking deficits underlying neurodegeneration. *Translational Neurodegeneration*. 2026;15(1):26. PMID 42231420.

Haney MS, Pálovics R, Munson CN, et al., Wyss-Coray T. APOE4/4 is linked to damaging lipid droplets in Alzheimer's disease microglia. *Nature* 2024;628(8006):154–161. PMID 38480892.

Haq I, Ngo JC, Roy N, Lee E, Choudhury MA, Soni RK, Teich AF, Mayeux RP, De Jager PL, He Y, Wu X, Bennett DA, Olah M, Sher F. Alzheimer's disease risk protein SorLA regulates ER homeostasis and lipid metabolism in human microglia, with conserved effects in neurons. *Acta Neuropathologica*. 2026;151(1):37. PMID 41942750.

Hardy JA, Higgins GA. Alzheimer's disease: the amyloid cascade hypothesis. *Science* 1992;256(5054):184–185. PMID 1566067.

He HY, Ahsan A, Bera R, McLain N, Faulkner R, Ramachandran KV, Margolis SS, Cline HT. Neuronal membrane proteasomes regulate neuronal circuit activity in vivo and are required for learning-induced behavioral plasticity. *Proceedings of the National Academy of Sciences USA*. 2023;120(3):e2216537120. PMID 36630455.

Head BP, Insel PA. Do caveolins regulate cells by actions outside of caveolae? *Trends in Cell Biology*. 2007;17(2):51–57. PMID 17150359.

Head BP, Peart JN, Panneerselvam M, Yokoyama T, Pearn ML, Niesman IR, Bonds JA, Schilling JM, Miyanohara A, Headrick J, Ali SS, Roth DM, Patel PM, Patel HH. Loss of caveolin-1 accelerates neurodegeneration and aging. *PLoS One*. 2010;5(12):e15697. PMID 21203469.

Head BP, Hu Y, Finley JC, Saldana MD, Bonds JA, Miyanohara A, Niesman IR, Ali SS, Murray F, Insel PA, Roth DM, Patel HH, Patel PM. Neuron-targeted caveolin-1 protein enhances signaling and promotes arborization of primary neurons. *Journal of Biological Chemistry*. 2011;286(38):33310–33321. PMID 21799010.

Heir R, Abbasi Z, Komal P, et al. Astrocytes Are the Source of TNF Mediating Homeostatic Synaptic Plasticity. *Journal of Neuroscience* 2024;44(14). PMID 38395613.

Holstege H, Hulsman M, Charbonnier C, et al. Exome sequencing identifies rare damaging variants in ATP8B4 and ABCA1 as risk factors for Alzheimer's disease. *Nature Genetics* 2022;54(12):1786–1794. PMID 36411364.

Hong S, Beja-Glasser VF, Nfonoyim BM, Frouin A, Li S, Ramakrishnan S, Merry KM, Shi Q, Rosenthal A, Barres BA, Lemere CA, Selkoe DJ, Stevens B. Complement and microglia mediate early synapse loss in Alzheimer mouse models. *Science*. 2016;352(6286):712–716. PMID 27033548.

Hoover BR, Reed MN, Su J, Penrod RD, Kotilinek LA, Grant MK, Pitstick R, Carlson GA, Lanier LM, Yuan LL, Ashe KH, Liao D. Tau mislocalization to dendritic spines mediates synaptic dysfunction independently of neurodegeneration. *Neuron*. 2010;68(6):1067–1081. PMID 21172610.

Hordeaux J, Wang Q, Katz N, Buza EL, Bell P, Wilson JM. The neurotropic properties of AAV-PHP.B are limited to C57BL/6J mice. *Molecular Therapy*. 2018;26(3):664–668. PMID 29428298.

Huang Q, Chan KY, Tobey IG, Chan YA, Poterba T, Boutros CL, Balazs AB, Daneman R, Bloom JM, Seed C, Deverman BE. Delivering genes across the blood-brain barrier: LY6A, a novel cellular receptor for AAV-PHP.B capsids. *PLoS One*. 2019;14(11):e0225206. PMID 31725765.

Huang H, Shi CH, Yang W, Piña-Crespo J, Bhatnagar J, Curatolo J, Murad R, Shah P, Campos A, Houser A, Porritt RA, Zhang T, Xiao Q, Feng S, Yip KY, Huang TY. SORLA up-regulation suppresses pathological effects in aged tauopathy mouse brain. *Science Advances*. 2026;12(29):eaed6825. doi:10.1126/sciadv.aed6825.

Huh GS, Boulanger LM, Du H, Riquelme PA, Brotz TM, Shatz CJ. Functional requirement for class I MHC in CNS development and plasticity. *Science*. 2000;290(5499):2155–2159. PMID 11118151.

Hung C, Tuck E, Stubbs V, van der Lee SJ, Aalfs C, van Spaendonk R, Scheltens P, Hardy J, Holstege H, Livesey FJ. SORL1 deficiency in human excitatory neurons causes APP-dependent defects in the endolysosome-autophagy network. *Cell Reports*. 2021;35(11):109259. PMID 34133918.

Im E, Jiang Y, Stavrides PH, et al., Nixon RA. Lysosomal dysfunction in Down syndrome and Alzheimer mouse models is caused by v-ATPase inhibition by Tyr682-phosphorylated APP βCTF. *Science Advances* 2023;9(30):eadg1925. PMID 37494443.

Ising C, Venegas C, Zhang S, et al. NLRP3 inflammasome activation drives tau pathology. *Nature* 2019;575(7784):669-673. PMID 31748742.

Ittner LM, Ke YD, Delerue F, Bi M, Gladbach A, van Eersel J, Wölfing H, Chieng BC, Christie MJ, Napier IA, Eckert A, Staufenbiel M, Hardeman E, Götz J. Dendritic function of tau mediates amyloid-beta toxicity in Alzheimer's disease mouse models. *Cell*. 2010;142(3):387–397. PMID 20655099.

Jensen AMG, Kitago Y, Fazeli E, Vægter CB, Small SA, Petsko GA, Andersen OM. Dimerization of the Alzheimer's disease pathogenic receptor SORLA regulates its association with retromer. *Proceedings of the National Academy of Sciences USA*. 2023;120(4):e2212180120. PMID 36652482.

Jiang Y, Sachdeva K, Goulbourne CN, Berg MJ, Peddy J, Stavrides PH, Pensalfini A, Pawlik M, Malampati S, Whyte L, Basavarajappa BS, Subbanna S, Bleiwas C, Smiley JF, Mathews PM, Nixon RA. Increased neuronal expression of the early endosomal adaptor APPL1 replicates Alzheimer's disease-related endosomal and synaptic dysfunction with cholinergic neurodegeneration. *Journal of Neuroscience*. 2025;45(29):e2331242025. PMID 40514243.

Kang MJ, Chung YH, Hwang CI, Murata M, Fujimoto T, Mook-Jung IH, Cha CI, Park WY. Caveolin-1 upregulation in senescent neurons alters amyloid precursor protein processing. *Experimental & Molecular Medicine*.

2006;38(2):126–133. PMID 16672766.

Kang JE, Lim MM, Bateman RJ, et al. Amyloid-beta dynamics are regulated by orexin and the sleep-wake cycle. *Science* 2009;326(5955):1005-7. PMID 19779148.

van der Kant R, Langness VF, Herrera CM, et al., Goldstein LSB. Cholesterol metabolism is a druggable axis that independently regulates tau and amyloid-β in iPSC-derived Alzheimer's disease neurons. *Cell Stem Cell* 2019;24(3):363–375.e9. PMID 30686764.

Kapoor A, Hsu WM, Wang BJ, Wu GH, Lin TY, Lee SJ, Yen CT, Liang SM, Liao YF. Caveolin-1 regulates γ-secretase-mediated AβPP processing by modulating spatial distribution of γ-secretase in membrane. *Journal of Alzheimer's Disease*. 2010;22(2):423–442. PMID 20847442.

Kassan A, Egawa J, Zhang Z, Almenar-Queralt A, Nguyen QM, Lajevardi Y, Kim K, Posadas E, Jeste DV, Roth DM, Patel PM, Patel HH, Head BP. Caveolin-1 regulation of disrupted-in-schizophrenia-1 as a potential therapeutic target for schizophrenia. *Journal of Neurophysiology*. 2017;117(1):436–444. PMID 27832597.

Kawaguchi Y, Matsubayashi J, Kawakami Y, Nishida R, Kurihara Y, Takei K. LOTUS suppresses amyloid β-induced dendritic spine elimination through the blockade of amyloid β binding to PirB. *Molecular Medicine*. 2022;28(1):154. PMID 36510132.

Kim T, Vidal GS, Djurisic M, William CM, Birnbaum ME, Garcia KC, Hyman BT, Shatz CJ. Human LilrB2 is a β-amyloid receptor and its murine homolog PirB regulates synaptic plasticity in an Alzheimer's model. *Science*. 2013;341(6152):1399–1404. PMID 24052308.

Kim S, Sato Y, Mohan PS, Peterhoff C, Pensalfini A, Rigoglioso A, Jiang Y, Nixon RA. Evidence that the rab5 effector APPL1 mediates APP-βCTF-induced dysfunction of endosomes in Down syndrome and Alzheimer's disease. *Molecular Psychiatry*. 2016;21(5):707–716. PMID 26194181.

Klementieva O, Willén K, Martinsson I, Israelsson B, Engdahl A, Cladera J, Uvdal P, Gouras GK. Pre-plaque conformational changes in Alzheimer's disease-linked Aβ and APP. *Nature Communications*. 2017;8:14726. PMID 28287086.

Klementieva O, Sandt C, Martinsson I, Kansiz M, Gouras GK, Borondics F. Super-resolution infrared imaging of polymorphic amyloid aggregates directly in neurons. *Advanced Science*. 2020;7(6):1903004. PMID 32195099.

Knupp A, Mishra S, Martinez R, Braggin JE, Szabo M, Kinoshita C, Hailey DW, Small SA, Jayadev S, Young JE. Depletion of the AD risk gene SORL1 selectively impairs neuronal endosomal traffic independent of amyloidogenic APP processing. *Cell Reports*. 2020;31(9):107719. PMID 32492427.

Konings SC, Nyberg E, Martinsson I, Torres-Garcia L, Klementieva O, Guimas Almeida C, Gouras GK. Apolipoprotein E intersects with amyloid-β within neurons. *Life Science Alliance*. 2023;6(8):e202201887. PMID 37290814. (Correction: *Life Science Alliance*. 2024;7(9):e202402875.)

Konrad-Vicario KD, Paradise V, Demir L, Makinson CD, Ming Z, Ramachandran KV. Spatial coupling of endogenous Tau translation and degradation by neuroproteasomes in dendrites revealed by STARFISH. *bioRxiv* 2025.06.04.657939. **Preprint — not peer-reviewed.**

Kumar DK, Choi SH, Washicosky KJ, et al. Amyloid-β peptide protects against microbial infection in mouse and worm models of Alzheimer's disease. *Science Translational Medicine* 2016;8(340):340ra72. PMID 27225182.

Kwart D, Gregg A, Scheckel C, et al., Tessier-Lavigne M. A large panel of isogenic APP and PSEN1 mutant human iPSC neurons reveals shared endosomal abnormalities mediated by APP β-CTFs, not Aβ. *Neuron* 2019;104(2):256–270.e5. PMID 31416668.

Kwon HJ, Santhosh D, Huang Z. A novel monomeric amyloid β-activated signaling pathway regulates brain development via inhibition of microglia. *eLife* 2024;13. PMID 39635981.

Lao PJ, Lee S, Talmasov D, Dass D, Chikwem N, Johnson A, Smith A, Guzman D, Okafor A, Houlihan H, Heuer L, Sanchez T, Maldonado A, Palacios C, Rossano S, Andrews H, Reitz C, Kreisl WC, Noble JM, Qureshi YH, Small SA. Inflammatory alterations mediate tau-associated neurodegeneration. *Brain Communications*. 2026;8(4):fcag242. PMID 42404434.

Larrea D, Tamucci KA, Kabra K, Velasco KR, Yun TD, Pera M, Montesinos J, Agrawal RR, Paradas C, Smerdon JW, Lowry ER, Stepanova A, Yoval-Sanchez B, Galkin A, Wichterle H, Area-Gómez E. Altered mitochondria-associated ER membrane (MAM) function shifts mitochondrial metabolism in amyotrophic lateral sclerosis (ALS). *Nature Communications* 2025;16(1):379. PMID 39753538.

Lauritzen I, Pardossi-Piquard R, Bauer C, Brigham E, Abraham JD, Ranaldi S, Fraser P, St-George-Hyslop P, Le Thuc O, Espin V, Chami L, Dunys J, Checler F. The β-secretase-derived C-terminal fragment of βAPP, C99, but not Aβ, is a key contributor to early intraneuronal lesions in triple-transgenic mouse hippocampus. *Journal of Neuroscience* 2012;32(46):16243–16255. PMID 23152608.

Lauritzen I, Pardossi-Piquard R, Bourgeois A, Pagnotta S, Biferi MG, Barkats M, Lacor P, Klein W, Bauer C, Checler F. Intraneuronal aggregation of the β-CTF fragment of APP (C99) induces Aβ-independent lysosomal-autophagic pathology. *Acta Neuropathologica* 2016;132(2):257–276. PMID 27138984.

Lee SI, Jeong W, Lim H, Cho S, Lee H, Jang Y, Cho J, Bae S, Lin YT, Tsai LH, Moon DW, Seo J. APOE4-carrying human astrocytes oversupply cholesterol to promote neuronal lipid raft expansion and Aβ generation. *Stem Cell Reports*. 2021;16(9):2128–2137.

Lee JH, Yang DS, Goulbourne CN, Im E, Stavrides P, Pensalfini A, Chan H, Bouchet-Marquis C, Bleiwas C, Berg MJ, Huo C, Peddy J, Pawlik M, Levy E, Rao M, Staufenbiel M, Nixon RA. Faulty autolysosome acidification in Alzheimer's disease mouse models induces autophagic build-up of Aβ in neurons, yielding senile plaques. *Nature Neuroscience*. 2022;25(6):688–701. PMID 35654956.

Lee SI, Lim H, Kim NY, Yu J, Cho J, Lee H, Moon DW, Seo J. Imaging lipid rafts reveals the principle of ApoE4-induced Aβ upregulation in human neurons. *iScience*. 2025;28(2):111893. PMID 39995873.

Leyns CEG, Gratuze M, Narasimhan S, et al. TREM2 function impedes tau seeding in neuritic plaques. *Nature Neuroscience* 2019;22(8):1217-1222. PMID 31235932.

Lopera F, Marino C, Chandrahas AS, O'Hare M, Villalba-Moreno ND, Aguillon D, Baena A, Sanchez JS, Vila-Castelar C, Ramirez Gomez L, Chmielewska N, Oliveira GM, Littau JL, Hartmann K, Park K, Krasemann S, Glatzel M, Schoemaker D, Gonzalez-Buendia L, Delgado-Tirado S, Arevalo-Alquichire S, Saez-Torres KL, Amarnani D, Kim LA, Mazzarino RC, Gonzalez H, Ortiz-Cardona J, Aguirre-Acevedo DC, Pantoja AF, Yepes Y, Bocanegra Y, Cardenas-Aguayo MDC, Aristizabal-Rojas H, Estrada-Lopez K, Ruiz-Duque L, Sepulveda-Falla D, Quiroz YT, Arboleda-Velasquez JF. Resilience to autosomal dominant Alzheimer's disease in a Reelin-COLBOS heterozygous man. *Nature Medicine*. 2023;29(5):1243–1252. PMID 37188781.

Lundgren JL, Ahmed S, Schedin-Weiss S, Gouras GK, Winblad B, Tjernberg LO, Frykman S. ADAM10 and BACE1 are localized to synaptic vesicles. *Journal of Neurochemistry*. 2015;135(3):606–615. PMID 26296617.

Ma T, Hoeffer CA, Capetillo-Zarate E, Yu F, Wong H, Lin MT, Tampellini D, Klann E, Blitzer RD, Gouras GK. Dysregulation of the mTOR pathway mediates impairment of synaptic plasticity in a mouse model of Alzheimer's disease. *PLoS One*. 2010;5(9):e12845. PMID 20862226.

Margolis SS, Salogiannis J, Lipton DM, Mandel-Brehm C, Wills ZP, Mardinly AR, Hu L, Greer PL, Bikoff JB, Ho HY, Soskis MJ, Sahin M, Greenberg ME. EphB-mediated degradation of the RhoA GEF Ephexin5 relieves a developmental brake on excitatory synapse formation. *Cell*. 2010;143(3):442–455. PMID 21029865.

Marik SA, Olsen O, Tessier-Lavigne M, et al. Physiological role for amyloid precursor protein in adult experience-dependent plasticity. *Proceedings of the National Academy of Sciences USA* 2016;113(28):7912-7. PMID 27354516.

Markesbery WR, Kryscio RJ, Lovell MA, Morrow JD. Lipid peroxidation is an early event in the brain in amnestic mild cognitive impairment. *Annals of Neurology*. 2005;58(5):730–735.

Martinsson I, Capetillo-Zarate E, Faideau M, Willén K, Esteras N, Frykman S, Tjernberg LO, Gouras GK. APP depletion alters selective pre- and post-synaptic proteins. *Molecular and Cellular Neuroscience*. 2019;95:86–95. PMID 30763689.

Martinsson I, Quintino L, Garcia MG, Konings SC, Torres-Garcia L, Svanbergsson A, Stange O, England R, Deierborg T, Li JY, Lundberg C, Gouras GK. Aβ/amyloid precursor protein-induced hyperexcitability and dysregulation of homeostatic synaptic plasticity in neuron models of Alzheimer's disease. *Frontiers in Aging Neuroscience*. 2022;14:946297. PMID 35928998.

Mecca AP, Chen MK, O'Dell RS, Naganawa M, Toyonaga T, Godek TA, Harris JE, Bartlett HH, Zhao W, Nabulsi NB, Vander Wyk BC, Varma P, Arnsten AFT, Huang Y, Carson RE, van Dyck CH. In vivo measurement of widespread synaptic loss in Alzheimer's disease with SV2A PET. *Alzheimer's & Dementia*. 2020;16(7):974–982. PMID 32400950.

Minamide LS, Striegl AM, Boyle JA, Meberg PJ, Bamburg JR. Neurodegenerative stimuli induce persistent ADF/cofilin-actin rods that disrupt distal neurite function. *Nature Cell Biology*. 2000;2(9):628–636. PMID 10980704.

Moir RD, Lathe R, Tanzi RE. The antimicrobial protection hypothesis of Alzheimer's disease. *Alzheimer's & Dementia* 2018;14(12):1602-1614. PMID 30314800.

Montesinos J, Pera M, Larrea D, Guardia-Laguarta C, Agrawal RR, Velasco KR, Yun TD, Stavrovskaya IG, Xu Y, Koo SY, Snead AM, Sproul AA, Area-Gómez E. The Alzheimer's disease-associated C99 fragment of APP regulates cellular cholesterol trafficking. *EMBO Journal* 2020;39(20):e103791. PMID 32865299.

Montesinos J, Kabra K, Uceda M, Larrea D, Agrawal RR, Tamucci KA, Pera M, Ferre AC, Gomez-Lopez N, Yun TD, Velasco KR, Schon EA, Area-Gómez E. The contribution of mitochondria-associated ER membranes to cholesterol homeostasis. *bioRxiv* 2024 (preprint). PMID 39605513.

Montesinos J, Yun TD, Salomón-Cruz ID, Agudelo-Castrillón SC, Uceda M, Ferre AC, Anton-Barros C, Gomez-Lopez N, Agrawal RR, Larrea D, Velasco KR, Fernàndez-Bernal A, Benitez E, Zhu X, Schon EA, Lopera F, Cardona-Gómez GP, Area-Gómez E. Upregulation of MAM by C99 disrupts ACSL4 activity and phospholipid homeostasis in Alzheimer's disease models. *bioRxiv* 2025 (preprint). PMID 41394684.

Montine TJ, Huang DY, Valentine WM, et al. Crosslinking of apolipoprotein E by products of lipid peroxidation. *Journal of Neuropathology and Experimental Neurology*. 1996;55(2):202–210.

Montine KS, Olson SJ, Amarnath V, Whetsell WO, Graham DG, Montine TJ. Immunohistochemical detection of 4-hydroxy-2-nonenal adducts in Alzheimer's disease is associated with inheritance of APOE4. *American Journal of Pathology*. 1997;150(2):437–443.

Muhammad A, Flores I, Zhang H, Yu R, Staniszewski A, Planel E, Herman M, Ho L, Kreber R, Honig LS, Ganetzky B, Duff K, Arancio O, Small SA. Retromer deficiency observed in Alzheimer's disease causes hippocampal dysfunction, neurodegeneration, and Abeta accumulation. *Proceedings of the National Academy of*

Sciences USA. 2008;105(20):7327–7332. PMID 18480253.

Mummery CJ, Mayorga AJ, Simmons A, et al. The TREM2 agonistic antibody AL002 in early Alzheimer's disease: a phase 2 randomized trial. *Nature Medicine* 2026;32(5):1708-1716. PMID 41787076.

Nishimoto I, Okamoto T, Matsuura Y, et al. Alzheimer amyloid protein precursor complexes with brain GTP-binding protein G(o). *Nature* 1993;362(6415):75-9. PMID 8446172.

Nyberg E, Konings SC, Lindblom N, Israelsson B, Klementieva O, Martinsson I, Gouras GK. Neuronal endolysosomal alterations induced by apolipoprotein E4 emerge over time in primary neurons. *Journal of Biological Chemistry*. 2025;301(8):110479. PMID 40675218.

Olsson TT, Klementieva O, Gouras GK. Prion-like seeding and nucleation of intracellular amyloid-β. *Neurobiology of Disease*. 2018;113:1–10. PMID 29414379.

Paradise V, Konrad-Vicario KD, Nguyen C, Sharif NA, Wang X, Mukim RD, Sabu M, Corjuc BT, Bafia J, Fu J, Maldonado GC, Strickland M, Grauman SL, Figueroa H, Hyman BT, Holtzman DM, Nuriel T, Ramachandran KV. Neuroproteasomes regulate endogenous tau paired helical filament formation in an APOE genotype- and age-dependent manner. *Nature Neuroscience*. 2026 May 29 (online ahead of print). PMID 42215643. doi:10.1038/s41593-026-02297-x

Pensalfini A, Kim S, Subbanna S, Bleiwas C, Goulbourne CN, Stavrides PH, Jiang Y, Lee JH, Darji S, Pawlik M, Huo C, Peddy J, Berg MJ, Smiley JF, Basavarajappa BS, Nixon RA. Endosomal dysfunction induced by directly overactivating Rab5 recapitulates prodromal and neurodegenerative features of Alzheimer's disease. *Cell Reports*. 2020;33(8):108420. PMID 33238112.

Petit D, Fernández SG, Zoltowska KM, et al., Chávez-Gutiérrez L. Aβ profiles generated by Alzheimer's disease causing PSEN1 variants determine the pathogenicity of the mutation and predict age at disease onset. *Molecular Psychiatry* 2022;27(6):2821–2832. PMID 35365805.

Petshow S, Coblentz A, Hamilton AM, Sarkar D, Anisimova M, Flores JC, Zito K. Activity-dependent regulation of Cdc42 by Ephexin5 drives synapse growth and stabilization. *Science Advances*. 2025;11(13):eadp5782. PMID 40138406.

Pooler AM, Phillips EC, Lau DH, Noble W, Hanger DP. Physiological release of endogenous tau is stimulated by neuronal activity. *EMBO Reports*. 2013;14(4):389–394. PMID 23412472.

Puzzo D, Privitera L, Leznik E, et al. Picomolar amyloid-beta positively modulates synaptic plasticity and memory in hippocampus. *Journal of Neuroscience* 2008;28(53):14537-45. PMID 19118188.

Quinn JF, Raman R, Thomas RG, et al. Docosahexaenoic acid supplementation and cognitive decline in Alzheimer disease: a randomized trial. *JAMA*. 2010;304(17):1903–1911.

Qureshi YH, Berman DE, Marsh SE, Klein RL, Patel VM, Simoes S, Kannan S, Petsko GA, Stevens B, Small SA. The neuronal retromer can regulate both neuronal and microglial phenotypes of Alzheimer's disease. *Cell Reports*. 2022;38(3):110262. PMID 35045281.

Qureshi YH, Williams CA, Hajdu I, Kannan S, Govindarajan A, Végh B, Petsko GA, Young JE, Závodszky P, Small SA. RhoGEF12 regulates endosomal SORL1-retromer and its inhibition is therapeutic in human neuronal models of Alzheimer's disease. *bioRxiv*. 2026;2026.03.06.709427. [Preprint.] PMID 41889823.

Rafii MS, Tuszynski MH, Thomas RG, Barba D, Brewer JB, Rissman RA, Siffert J, Aisen PS; AAV2-NGF Study Team. Adeno-associated viral vector (serotype 2)-nerve growth factor for patients with Alzheimer disease: a randomized clinical trial. *JAMA Neurology*. 2018;75(7):834–841. PMID 29582053.

Ralhan I, Do AD, Bae JY, et al. Protective ApoE variants support neuronal function by effluxing oxidized phospholipids. *Neuron*. 2026;114(4):661–678.e10.

Ramachandran KV, Margolis SS. A mammalian nervous-system-specific plasma membrane proteasome complex that modulates neuronal function. *Nature Structural & Molecular Biology*. 2017;24(4):419–430. PMID 28287632.

Ramachandran KV, Fu JM, Schaffer TB, Na CH, Delannoy M, Margolis SS. Activity-dependent degradation of the nascentome by the neuronal membrane proteasome. *Molecular Cell*. 2018;71(1):169–177.e6. PMID 29979964.

Ramakrishna S, Jhaveri V, Konings SC, Nawalpuri B, Chakraborty S, Holst B, Schmid B, Gouras GK, Freude KK, Muddashetty RS. APOE4 affects basal and NMDAR-mediated protein synthesis in neurons by perturbing calcium homeostasis. *Journal of Neuroscience*. 2021;41(42):8686–8709. PMID 34475200.

Ramonet D, Daerr A, Hallbeck M. Stabilizing the retromer complex rescues synaptic dysfunction and endosomal trafficking deficits in an Alzheimer's disease mouse model. *Acta Neuropathologica Communications*. 2025;13(1):190. PMID 40931359.

Ramsden CE, Ringel A, Feldstein AE, et al. Lowering dietary linoleic acid reduces bioactive oxidized linoleic acid metabolites in humans. *Prostaglandins, Leukotrienes and Essential Fatty Acids*. 2012;87(4–5):135–141.

Ramsden CE, Zamora D, Faurot KR, et al. Dietary alteration of n-3 and n-6 fatty acids for headache reduction in adults with migraine: randomized controlled trial. *BMJ*. 2021;374:n1448.

Ramsden CE, Keyes GS, Calzada E, et al. Lipid peroxidation induced ApoE receptor-ligand disruption as a unifying hypothesis underlying sporadic Alzheimer's disease in humans. *Journal of Alzheimer's Disease*. 2022;87(3):1251–1290.

Ramsden CE, Zamora D, Horowitz MS, et al. ApoER2-Dab1 disruption as the origin of pTau-associated neurodegeneration in sporadic Alzheimer's disease. *Acta Neuropathologica Communications*. 2023;11(1):197.

Ramsden CE, Cutler RG, Li X, Keyes GS. Lipid-protecting disulfide bridges are the missing molecular link between ApoE4 and sporadic Alzheimer's disease in humans. *Prostaglandins, Leukotrienes and Essential Fatty Acids*. 2025;205:102681.

Rappoport A. A lipid-raft theory of Alzheimer's disease. *Annual Review of Biochemistry*. 2025;94:387–416. PMID 39476407.

Roos TT, Garcia MG, Martinsson I, Mabrouk R, Israelsson B, Deierborg T, Kobro-Flatmoen A, Tanila H, Gouras GK. Neuronal spreading and plaque induction of intracellular Aβ and its disruption of Aβ homeostasis. *Acta Neuropathologica*. 2021;142(4):669–687. PMID 34272583.

Rush T, Martinez-Hernandez J, Dollmeyer M, Frandemiche ML, et al. Synaptotoxicity in Alzheimer's disease involved a dysregulation of actin cytoskeleton dynamics through cofilin 1 phosphorylation. *Journal of Neuroscience*. 2018;38(48):10349–10361. PMID 30341179.

Sahlin C, Lord A, Magnusson K, Englund H, Almeida CG, Greengard P, Nyberg F, Gouras GK, Lannfelt L, Nilsson LN. The Arctic Alzheimer mutation favors intracellular amyloid-beta production by making amyloid precursor protein less available to alpha-secretase. *Journal of Neurochemistry*. 2007;101(3):854–862. PMID 17448150.

Sawada A, Wang S, Jian M, Leem J, Wackerbarth J, Egawa J, Schilling JM, Platoshyn O, Zemljic-Harpf A, Roth DM, Patel HH, Patel PM, Marsala M, Head BP. Neuron-targeted caveolin-1 improves neuromuscular function and extends survival in SOD1^{G93A} mice. *FASEB Journal*. 2019;33(6):7545–7554. PMID 30894019.

Schaffer TB, Smith JE, Cook EK, Phan T, Margolis SS. PKCε inhibits neuronal dendritic spine development through dual phosphorylation of Ephexin5. *Cell Reports*. 2018;25(9):2470–2483.e8. PMID 30485813.

Scheff SW, Price DA, Schmitt FA, DeKosky ST, Mufson EJ. Synaptic alterations in CA1 in mild Alzheimer disease and mild cognitive impairment. *Neurology*. 2007;68(18):1501–1508. PMID 17470753.

Scott-Hewitt N, Perrucci F, Morini R, et al. Local externalization of phosphatidylserine mediates developmental synaptic pruning by microglia. *EMBO Journal* 2020;39(16):e105380. PMID 32657463.

Selkoe DJ. Alzheimer's disease is a synaptic failure. *Science*. 2002;298(5594):789–791. PMID 12399581.

Selkoe DJ, Hardy J. The amyloid hypothesis of Alzheimer's disease at 25 years. *EMBO Molecular Medicine*. 2016;8(6):595–608. PMID 27025652.

Sell GL, Schaffer TB, Margolis SS. Reducing expression of synapse-restricting protein Ephexin5 ameliorates Alzheimer's-like impairment in mice. *Journal of Clinical Investigation*. 2017;127(5):1646–1650. PMID 28346227.

Shankar GM, Li S, Mehta TH, Garcia-Munoz A, Shepardson NE, Smith I, Brett FM, Farrell MA, Rowan MJ, Lemere CA, Regan CM, Walsh DM, Sabatini BL, Selkoe DJ. Amyloid-beta protein dimers isolated directly from Alzheimer's brains impair synaptic plasticity and memory. *Nature Medicine*. 2008;14(8):837–842. PMID 18568035.

Shokri-Kojori E, Wang GJ, Wiers CE, et al. β-Amyloid accumulation in the human brain after one night of sleep deprivation. *Proceedings of the National Academy of Sciences USA* 2018;115(17):4483-4488. PMID 29632177.

Sierra S, Ramos MC, Molina P, Esteo C, Vázquez JA, Burgos JS. Statins as neuroprotectants: a comparative in vitro study of lipophilicity, blood-brain-barrier penetration, lowering of brain cholesterol, and decrease of neuron cell death. *Journal of Alzheimer's Disease*. 2011. PMID 21098985.

Simoes S, Neufeld JL, Triana-Baltzer G, Moughadam S, Chen EI, Kothiya M, Qureshi YH, Patel V, Honig LS, Kolb H, Small SA. Tau and other proteins found in Alzheimer's disease spinal fluid are linked to retromer-mediated endosomal traffic in mice and humans. *Science Translational Medicine*. 2020;12(571):eaba6334. PMID 33239387.

Simoes S, Guo J, Buitrago L, Qureshi YH, Feng X, Kothiya M, Cortes E, Patel V, Kannan S, Kim YH, Chang KT, Hussaini SA, Moreno H, Di Paolo G, Andersen OM, Small SA. Alzheimer's vulnerable brain region relies on a distinct retromer core dedicated to endosomal recycling. *Cell Reports*. 2021;37(13):110182. PMID 34965419.

Sims JR, Zimmer JA, Evans CD, Lu M, Ardayfio P, Sparks J, Wessels AM, Shcherbinin S, Wang H, Monkul Nery ES, Collins EC, Solomon P, Salloway S, Apostolova LG, Hansson O, Ritchie C, Brooks DA, Mintun M, Skovronsky DM; TRAILBLAZER-ALZ 2 Investigators. Donanemab in early symptomatic Alzheimer disease: the TRAILBLAZER-ALZ 2 randomized clinical trial. *JAMA*. 2023;330(6):512–527. PMID 37459141.

Small SA, Kent K, Pierce A, Leung C, Kang MS, Okada H, Honig L, Vonsattel JP, Kim TW. Model-guided microarray implicates the retromer complex in Alzheimer's disease. *Annals of Neurology*. 2005;58(6):909–919. PMID 16315276.

Small SA, Simoes-Spassov S, Mayeux R, Petsko GA. Endosomal traffic jams represent a pathogenic hub and therapeutic target in Alzheimer's disease. *Trends in Neurosciences*. 2017;40(10):592–602. PMID 28962801.

Small SA, Petsko GA. Endosomal recycling reconciles the Alzheimer's disease paradox. *Science Translational Medicine*. 2020;12(572):eabb1717. PMID 33268506.

Smith LM, Kostylev MA, Lee S, Strittmatter SM. Systematic and standardized comparison of reported amyloid-β receptors for sufficiency, affinity, and Alzheimer's disease relevance. *Journal of Biological Chemistry*. 2019;294(15):6042–6053. PMID 30787106.

Snyder EM, Nong Y, Almeida CG, Paul S, Moran T, Choi EY, Nairn AC, Salter MW, Lombroso PJ, Gouras GK, Greengard P. Regulation of NMDA receptor trafficking by amyloid-beta. *Nature Neuroscience*.

2005;8(8):1051–1058. PMID 16025111.

Song Y, Hustedt EJ, Brandon S, Sanders CR. Competition between homodimerization and cholesterol binding to the C99 domain of the amyloid precursor protein. *Biochemistry* 2013;52(30):5051–5064. PMID 23865807.

Sperling RA, Donohue MC, Raman R, et al. Trial of Solanezumab in Preclinical Alzheimer's Disease. *New England Journal of Medicine* 2023;389(12):1096-1107. PMID 37458272.

Stenh C, Englund H, Lord A, Johansson AS, Almeida CG, Gellerfors P, Greengard P, Gouras GK, Lannfelt L, Nilsson LN. Amyloid-beta oligomers are inefficiently measured by enzyme-linked immunosorbent assay. *Annals of Neurology*. 2005;58(1):147–150. PMID 15984012.

Stevens B, Allen NJ, Vazquez LE, Howell GR, Christopherson KS, Nouri N, Micheva KD, Mehalow AK, Huberman AD, Stafford B, Sher A, Litke AM, Lambris JD, Smith SJ, John SW, Barres BA. The classical complement cascade mediates CNS synapse elimination. *Cell*. 2007;131(6):1164–1178. PMID 18083105.

Sturchio A, Dwivedi AK, Young CB, et al. High cerebrospinal amyloid-β 42 is associated with normal cognition in individuals with brain amyloidosis. *EClinicalMedicine* 2021;38:100988. PMID 34505023.

Sturchio A, Dwivedi AK, Malm T, et al. High Soluble Amyloid-β42 Predicts Normal Cognition in Amyloid-Positive Individuals with Alzheimer's Disease-Causing Mutations. *Journal of Alzheimer's Disease* 2022;90(1):333-348. PMID 36120786.

Sur C, Kost J, Scott D, Adamczuk K, Fox NC, Cummings JL, Tariot PN, Aisen PS, Vellas B, Voss T, Mozley LH, Ereshefsky L, Bragg S, Furtek C, Mahoney E, Harper Mozley L, Yaari R, Kennedy ME, Michelson D, Egan MF. BACE inhibition causes rapid, regional, and non-progressive volume reduction in Alzheimer's disease brain. *Brain* 2020;143(12):3816–3826. PMID 33253354.

Syken J, Grandpre T, Kanold PO, Shatz CJ. PirB restricts ocular-dominance plasticity in visual cortex. *Science*. 2006;313(5794):1795–1800. PMID 16917027.

Szaruga M, Munteanu B, Lismont S, et al., Chávez-Gutiérrez L. Alzheimer's-causing mutations shift Aβ length by destabilizing γ-secretase-Aβn interactions. *Cell* 2017;170(3):443–456.e14. PMID 28753424.

Taddei RN, Perbet R, Mate de Gerando A, et al. Tau Oligomer-Containing Synapse Elimination by Microglia and Astrocytes in Alzheimer Disease. *JAMA Neurology* 2023;80(11):1209-1221. PMID 37812432.

Takahashi RH, Milner TA, Li F, Nam EE, Edgar MA, Yamaguchi H, Beal MF, Xu H, Greengard P, Gouras GK. Intraneuronal Alzheimer Aβ42 accumulates in multivesicular bodies and is associated with synaptic pathology. *American Journal of Pathology*. 2002;161(5):1869–1879. PMID 12414533.

Takahashi RH, Almeida CG, Kearney PF, Yu F, Lin MT, Milner TA, Gouras GK. Oligomerization of Alzheimer's β-amyloid within processes and synapses of cultured neurons and brain. *Journal of Neuroscience*. 2004;24(14):3592–3599. PMID 15071107.

Takahashi RH, Capetillo-Zarate E, Lin MT, Milner TA, Gouras GK. Accumulation of intraneuronal β-amyloid 42 peptides is associated with early changes in microtubule-associated protein 2 in neurites and synapses. *PLoS One*. 2013;8(1):e51965. PMID 23372648.

Tambini MD, Pera M, Kanter E, Yang H, Guardia-Laguarta C, Holtzman D, Sulzer D, Area-Gómez E, Schon EA. ApoE4 upregulates the activity of mitochondria-associated ER membranes. *EMBO Reports* 2016;17(1):27–36. PMID 26564908.

Tampellini D, Rahman N, Gallo EF, Huang Z, Dumont M, Capetillo-Zarate E, Ma T, Zheng R, Lu B, Nanus DM, Lin MT, Gouras GK. Synaptic activity reduces intraneuronal Abeta, promotes APP transport to synapses, and protects against Abeta-related synaptic alterations. *Journal of Neuroscience*. 2009;29(31):9704–9713. PMID

19657023.

Tampellini D, Capetillo-Zarate E, Dumont M, Huang Z, Yu F, Lin MT, Gouras GK. Effects of synaptic modulation on β-amyloid, synaptophysin, and memory performance in Alzheimer's disease transgenic mice. *Journal of Neuroscience*. 2010;30(43):14299–14304. PMID 20980585.

Tampellini D, Rahman N, Lin MT, Capetillo-Zarate E, Gouras GK. Impaired β-amyloid secretion in Alzheimer's disease pathogenesis. *Journal of Neuroscience*. 2011;31(43):15384–15390. PMID 22031884.

Tariot PN, Lopera FS, Ríos-Romenets S, et al. Safety and efficacy of crenezumab in cognitively unimpaired carriers of the PSEN1Glu280Ala mutation at risk for autosomal-dominant Alzheimer's disease in Colombia (API ADAD Colombia Trial): a phase 2, randomised, double-blind, placebo-controlled trial. *Lancet Neurology* 2026;25(2):147-159. PMID 41579901.

Terry RD, Masliah E, Salmon DP, Butters N, DeTeresa R, Hill R, Hansen LA, Katzman R. Physical basis of cognitive alterations in Alzheimer's disease: synapse loss is the major correlate of cognitive impairment. *Annals of Neurology*. 1991;30(4):572–580. PMID 1789684.

Thorwald MA, Godoy-Lugo JA, Garcia G, et al. Iron-associated lipid peroxidation in Alzheimer's disease is increased in lipid rafts with decreased ferroptosis suppressors, tested by chelation in mice. *Alzheimer's & Dementia*. 2025;21(1):e14541.

Türker F, Brennan A, Margolis SS. Neuronal membrane proteasome-derived peptides modulate NMDAR-dependent neuronal signaling to promote changes in gene expression. *Molecular Biology of the Cell*. 2024;35(1):ar6. PMID 37910253.

Turrigiano GG. The self-tuning neuron: synaptic scaling of excitatory synapses. *Cell*. 2008;135(3):422–435. PMID 18984155.

Tuszynski MH, Thal L, Pay M, Salmon DP, U HS, Bakay R, Patel P, Blesch A, Vahlsing HL, Ho G, Tong G, Potkin SG, Fallon J, Hansen L, Mufson EJ, Kordower JH, Gall C, Conner J. A phase 1 clinical trial of nerve growth factor gene therapy for Alzheimer disease. *Nature Medicine*. 2005;11(5):551–555. PMID 15852017.

Tuszynski MH, Yang JH, Barba D, U HS, Bakay RA, Pay MM, Masliah E, Conner JM, Kobalka P, Roy S, Nagahara AH. Nerve growth factor gene therapy: activation of neuronal responses in Alzheimer disease. *JAMA Neurology*. 2015;72(10):1139–1147. PMID 26302439.

Tzioras M, Daniels MJD, Davies C, et al. Human astrocytes and microglia show augmented ingestion of synapses in Alzheimer's disease via MFG-E8. *Cell Reports Medicine* 2023;4(9):101175. PMID 37652017.

Uchida K. Histidine and lysine as targets of oxidative modification. *Amino Acids*. 2003;25(3–4):249–257.

Vance JE. Phospholipid synthesis in a membrane fraction associated with mitochondria. *Journal of Biological Chemistry* 1990;265(13):7248–7256. PMID 2332429.

Vidal GS, Djurisic M, Brown K, Sapp RW, Shatz CJ. Cell-autonomous regulation of dendritic spine density by PirB. *eNeuro*. 2016;3(5):ENEURO.0089-16.2016. PMID 27752542.

de Vivo L, Bellesi M, Marshall W, et al. Ultrastructural evidence for synaptic scaling across the wake/sleep cycle. *Science* 2017;355(6324):507-510. PMID 28154076.

de Waal MWJ, van der Lee SJ, Lunding M, Boonkamp L, Barrett N, Monti G, Jensen AMG, Vaegter CB, Raska J, Cesnarikova S, Sedmik J, Trieu C, Weiss MM, van Spaendonk R, Vermunt L, Ozhegov G, Tesi N, Hulsman M, Januliene D, Moller A, Bohaciakova D, van der Flier WM, Andersen OM, Teunissen CE, Holstege H. Soluble SORL1 in cerebrospinal fluid as a marker for functional impact of rare SORL1 variants. *Alzheimer's & Dementia*. 2026;22(2):e71042. PMID 41685556.

Walsh DM, Klyubin I, Fadeeva JV, et al. Naturally secreted oligomers of amyloid beta protein potently inhibit hippocampal long-term potentiation in vivo. *Nature* 2002;416(6880):535-9. PMID 11932745.

Wang P, Yang G, Mosier DR, et al. Defective neuromuscular synapses in mice lacking amyloid precursor protein (APP) and APP-Like protein 2. *Journal of Neuroscience* 2005;25(5):1219-25. PMID 15689559.

Wang S, Zhang Z, Almenar-Queralt A, Leem J, DerMardirossian C, Roth DM, Patel PM, Patel HH, Head BP. Caveolin-1 phosphorylation is essential for axonal growth of human neurons derived from iPSCs. *Frontiers in Cellular Neuroscience*. 2019;13:324. PMID 31379509.

Wang S, Leem JS, Podvin S, Hook V, Kleschevnikov N, Savchenko P, Dhanani M, Zhou K, Kelly IC, Zhang T, Miyanohara A, Nguyen P, Kleschevnikov A, Wagner SL, Trojanowski JQ, Roth DM, Patel HH, Patel PM, Head BP. Synapsin-caveolin-1 gene therapy preserves neuronal and synaptic morphology and prevents neurodegeneration in a mouse model of AD. *Molecular Therapy Methods & Clinical Development*. 2021a;21:434–450. PMID 33981778.

Wang S, Ichinomiya T, Savchenko P, Wang D, Sawada A, Li X, Duong T, Li W, Bonds JA, Kim EJ, Miyanohara A, Roth DM, Patel PM, Patel HH, Tadokoro T, Marsala M, Head BP. Subpial delivery of adeno-associated virus 9-synapsin-caveolin-1 (AAV9-SynCav1) preserves motor neuron and neuromuscular junction morphology, motor function, delays disease onset, and extends survival in hSOD1^{G93A} mice. *Theranostics*. 2022a;12(12):5389–5403. PMID 35910808.

Wang D, Chernov AV, Lam R, Wang H, Li W, Li X, Duong T, Wang S, Head BP. Neuron-targeted caveolin-1 overexpression attenuates cognitive loss and pathological transcriptome changes in symptomatic Alzheimer's disease models. *Signal Transduction and Targeted Therapy*. 2025;10(1):172. PMID 40425572.

Wang D, Ta V, Wang H, Ju J, Wang C, Chehadeh C, Torreblanca-Zanca A, Magaña Y, Castle MJ, Wang S, Head BP. Systemic delivery of synapsin-promoted caveolin-1 overexpression ameliorates pathological TDP-43-induced cognitive decline and neurodegenerative changes. *Alzheimer's & Dementia*. 2026;22(1):e71450. PMID 42187024.

Wang J, Athertya JS, Cheng X, Patel A, Chan NR, Liu B, Tang Q, Chang EY, Wang S, Ma Y, Head BP, Tang G, Du J. Ultrashort echo time magnetization transfer imaging of myelin in APP knock-in mice. *NeuroImage*. 2026;337:122040. PMID 42252040.

Willén K, Edgar JR, Hasegawa T, Tanaka N, Futter CE, Gouras GK. Aβ accumulation causes MVB enlargement and is modelled by dominant negative VPS4A. *Molecular Neurodegeneration*. 2017a;12(1):61. PMID 28835279.

Willén K, Sroka A, Takahashi RH, Gouras GK. Heterogeneous association of Alzheimer's disease-linked amyloid-β and amyloid-β protein precursor with synapses. *Journal of Alzheimer's Disease*. 2017b;60(2):511–524. PMID 28869466.

Wu JW, Hussaini SA, Bastille IM, Rodriguez GA, Mrejeru A, Rilett K, Sanders DW, Cook C, Fu H, Boonen RACM, Herman M, Nahmani E, Emrani S, Figueroa YH, Diamond MI, Clelland CL, Wray S, Duff KE. Neuronal activity enhances tau propagation and tau pathology in vivo. *Nature Neuroscience*. 2016;19(8):1085–1092. PMID 27322420.

Xian X, Pohlkamp T, Durakoglugil MS, et al. Reversal of ApoE4-induced recycling block as a novel prevention approach for Alzheimer's disease. *eLife*. 2018;7:e40048.

Xie L, Kang H, Xu Q, et al. Sleep drives metabolite clearance from the adult brain. *Science* 2013;342(6156):373-7. PMID 24136970.

Yang G, Zhou R, Zhou Q, Guo X, Yan C, Ke M, Lei J, Shi Y. Structural basis of Notch recognition by human γ-secretase. *Nature* 2019;565(7738):192–197. PMID 30598546.

Zamora D, Kenney K, Horowitz MS, et al. A high omega-3, low omega-6 diet reduces headache frequency and intensity in persistent post-traumatic headache: a randomized trial. *Journal of Neurotrauma*. 2025;42(19–20):1719–1731.

Zempel H, Thies E, Mandelkow E, Mandelkow EM. Aβ oligomers cause localized Ca2+ elevation, missorting of endogenous tau into dendrites, tau phosphorylation, and destruction of microtubules and spines. *Journal of Neuroscience*. 2010;30(36):11938–11950. PMID 20826658.

Zhao P, Xu Y, Jiang LL, Fan X, Ku Z, Li L, Liu X, Deng M, Arase H, Zhu JJ, Huang TY, Zhao Y, Zhang C, Xu H, Tong Q, Zhang N, An Z. LILRB2-mediated TREM2 signaling inhibition suppresses microglia functions. *Molecular Neurodegeneration*. 2022;17(1):44. PMID 35717259.

Zhou R, Yang G, Guo X, Zhou Q, Lei J, Shi Y. Recognition of the amyloid precursor protein by human γ-secretase. *Science* 2019;363(6428):eaaw0930. PMID 30630874.

Zott B, Simon MM, Hong W, et al. A vicious cycle of β amyloid-dependent neuronal hyperactivation. *Science* 2019;365(6453):559-565. PMID 31395777.