THE MITOCHONDRION AS NEXUS

Function, Dysfunction, and the Bioenergetic Architecture of Neurodegeneration

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Third thesis in the Collapse trilogy, companion to Convergent Synaptic Collapse and Homeostatic Microglial Collapse. Integrating nine research programs across mitochondrial biology, autophagy, immunometabolism, and neurodegeneration.

Abstract

Mitochondria occupy a singular position in neurodegenerative disease: they supply the ATP on which every synaptic event, ion gradient, protein-folding chaperone, and degradative organelle depends, and their failure generates the reactive oxygen species, damage-associated molecular patterns, and calcium dysregulation that drive each of the canonical proteinopathies. This dissertation integrates evidence across Alzheimer's disease, Parkinson's disease, amyotrophic lateral sclerosis, Huntington's disease, and frontotemporal dementia to argue that mitochondrial quality-control failure—encompassing oxidative phosphorylation decline, mitophagy impairment, fission/fusion dysregulation, mitochondria-associated membrane disruption, and NAD^+ depletion—constitutes a shared upstream substrate whose cell-type-specific manifestations determine the clinical phenotype of each disorder.

The thesis is organized in three analytical chapters. Chapter I establishes the bioenergetic architecture of the neuron, tracing the electron transport chain (Complexes I–V), the NAD^+/NADH redox couple, the TOM40 import machinery, and the mitochondria-associated membrane as an integrated functional unit whose failure cascades into endolysosomal, calcium, and transcriptional collapse. Chapter II examines how five aggregation-prone proteins—amyloid-beta, tau, alpha-synuclein, huntingtin, and TDP-43—each independently target mitochondrial function through distinct biochemical mechanisms that converge on a shared downstream pathology. Chapter III analyzes the immunometabolic dimension: how mitochondrial damage activates innate immunity through the NLRP3 inflammasome and NF-κB, how CD38-mediated NAD^+ consumption creates a self-destructive feedback loop in activated microglia, and how peripheral T-cell infiltration deepens the bioenergetic crisis.

The dissertation concludes that mitochondrial dysfunction is not a downstream consequence of proteinopathy but a rate-limiting variable whose decline sets the threshold for disease initiation. Therapeutic implications—including mitophagy inducers, NAD^+ precursors, electron carrier bypasses such as methylene blue, and systemic metabolic interventions—are evaluated against the mechanistic framework.

Keywords: mitochondria, neurodegeneration, oxidative phosphorylation, mitophagy, NAD^+, NLRP3, mitochondria-associated membrane, bioenergetics, Alzheimer's disease, Parkinson's disease

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Introduction

1.1 The Research Problem

The neurodegenerative diseases—Alzheimer's disease (AD), Parkinson's disease (PD), amyotrophic lateral sclerosis (ALS), Huntington's disease (HD), and frontotemporal dementia (FTD)—are conventionally classified by their signature aggregation-prone protein: amyloid-beta and tau in AD, alpha-synuclein in PD, huntingtin in HD, and TDP-43 in ALS/FTD. This nosological framework has organized two decades of therapeutic development around aggregate clearance, yielding anti-amyloid monoclonal antibodies (lecanemab, donanemab) with statistically significant but clinically modest effects and no disease-modifying therapies for PD, ALS, HD, or FTD.

A persistent anomaly cuts across the proteinopathy framework: every one of these diseases features early, pronounced, and progressive mitochondrial dysfunction. FDG-PET hypometabolism precedes clinical AD symptoms by decades (Reiman et al., 1996; Mosconi et al., 2008). Complex I deficiency is the biochemical signature of PD substantia nigra (Schapira et al., 1989). Mutant huntingtin causes Complex II/III deficiency years before chorea onset (Gu et al., 1996). TDP-43 accumulates inside mitochondria and inhibits Complex I in ALS motor neurons (Wang et al., 2016). The ubiquity and temporal priority of mitochondrial dysfunction across diseases with different protein aggregates, different vulnerable cell populations, and different clinical presentations demands an explanation that the protein-centric framework cannot easily supply.

This dissertation addresses the question: **What role does mitochondrial function and dysfunction play in neurodegeneration?** The thesis advanced here is that mitochondrial quality-control failure constitutes a shared upstream substrate—a bioenergetic bottleneck—whose decline sets the threshold for disease initiation and whose cell-type-specific manifestations determine which proteinopathy phenotype emerges. The disease proteins are not irrelevant; rather, they are best understood as additional mitochondrial stressors that accelerate a decline already in progress, converting a gradual bioenergetic erosion into a catastrophic quality-control collapse.

1.2 Significance

The significance of this reframing is threefold. First, it explains the age dependence of sporadic neurodegeneration: mitochondrial function declines with age in all individuals, and inherited mitochondrial capacity—set by mtDNA haplogroup and nuclear-encoded mitochondrial genes—determines when the decline crosses a pathogenic threshold (Swerdlow & Khan, 2004). Second, it accounts for the transdiagnostic overlap among neurodegenerative diseases—comorbid pathologies, shared risk genes (e.g., *APOE*, *TREM2*, *GBA*), and the frequent co-occurrence of multiple proteinopathies in aged brains (Robinson et al., 2018)—as consequences of a common bioenergetic substrate whose failure permits multiple downstream cascades. Third, it identifies a therapeutic target space—mitophagy, NAD^+ metabolism, electron transport chain bypass, lysosomal acidification, and systemic metabolic intervention—that is orthogonal to aggregate clearance and potentially complementary to it.

1.3 Scope and Limitations

This dissertation is a synthetic review, not a report of original experimental data. Its contribution lies in the integration of nine major research programs into a unified analytical framework, the identification of specific mechanistic coupling points, and the generation of falsifiable predictions. The work is biased toward AD because the mitochondrial literature is deepest in that disease, but each analytical chapter systematically addresses PD, ALS, HD, and FTD where evidence permits.

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Literature Review

2.1 Historical Foundations: Mitochondria and the Brain

The relationship between brain metabolism and cognitive function was established long before mitochondria were recognized as the site of oxidative phosphorylation. Alois Alzheimer's 1907 case report described not only the neurofibrillary tangles and plaques that bear his name but also “adipose inclusions” in neurons—now interpretable as lipid-laden autophagic vacuoles reflecting degradative failure (Alzheimer, 1907). Oskar Fischer, publishing contemporaneously, documented similar pathology with emphasis on the reactive glial component (Fischer, 1907, 1910).

The modern study of mitochondrial dysfunction in neurodegeneration began with three parallel discoveries. Schapira and colleagues (1989) reported a selective 35% reduction in Complex I activity in PD substantia nigra, establishing the first disease-specific mitochondrial lesion. Langston and colleagues' characterization of MPTP-induced parkinsonism (1983) demonstrated that environmental Complex I inhibition could recapitulate the clinical and pathological features of PD. And Parker, Filley, and Parks (1990) reported reduced cytochrome oxidase (Complex IV) activity in AD brain tissue, extending the mitochondrial hypothesis beyond PD.

2.2 The Mitochondrial Cascade Hypothesis

Swerdlow and Khan (2004) formalized these observations into the Mitochondrial Cascade Hypothesis (MCH) for sporadic AD. The MCH proposes that inherited mitochondrial function, determined primarily by maternally transmitted mtDNA, varies across the population; that age-related accumulation of somatic mtDNA mutations erodes mitochondrial function from this inherited baseline; that when bioenergetic capacity crosses a cell-type-specific threshold, disease-associated pathologies emerge as downstream consequences; and that the age of clinical onset reflects the inherited starting point and the rate of decline rather than an exogenous trigger.

The MCH was updated in 2018 to engage with intervening evidence, including the cybrid model system in which mitochondria from AD patients are transferred into mitochondria-depleted recipient cells, demonstrating that the bioenergetic defect is transmissible via mtDNA and produces AD-like phenotypes including increased amyloid-beta production, oxidative stress, and apoptotic vulnerability (Swerdlow, 2018).

2.3 The Autophagy-Lysosomal Axis

Nixon and colleagues documented massive accumulation of autophagic vacuoles in AD neurons over two decades of ultrastructural study (Nixon et al., 2005; Nixon, 2013). The critical advance came with Lee, Yang, Nixon et al. (2022), who demonstrated that in AD mouse models, autolysosomes fail to acidify adequately, autophagic cargo including amyloid-beta accumulates, and some affected neurons exhibit a rosette-like expansion of autolysosomes (PANTHOS—"Poisonous Flower") whose death and extrusion generates plaque-like structures from the inside out.

The crucial coupling point between these two literatures is the v-ATPase proton pump. Lysosomal acidification requires continuous ATP hydrolysis to maintain the ~pH 4.5 lumen against the cytoplasmic ~pH 7.2 (Mindell, 2012). When mitochondrial ATP production declines, v-ATPase activity declines proportionally, lysosomes de-acidify, and autophagic cargo—including damaged mitochondria targeted for mitophagy—accumulates rather than being degraded. This creates a vicious cycle: mitochondrial decline impairs the very degradative machinery that would clear damaged mitochondria.

2.4 Canonical Mitophagy

Narendra, Tanaka, Suen, and Youle (2008) demonstrated that Parkin is recruited to mitochondria with depolarized membrane potential and initiates outer-membrane ubiquitination. PINK1 was established as the upstream sensor: upon membrane-potential loss, PINK1 accumulates on the outer membrane and recruits Parkin. The pathway marks damaged mitochondria for autophagic degradation through p62/SQSTM1, OPTN, NDP52, and TAX1BP1 adaptors (Pickrell & Youle, 2015). Loss-of-function mutations in *PINK1* or *PRKN* cause autosomal-recessive juvenile parkinsonism, establishing that mitophagy failure is sufficient to cause neurodegeneration in humans.

2.5 Mitophagy Failure in Alzheimer's Disease

Fang, Hou, Palikaras et al. (2019) provided the most direct evidence linking mitophagy failure to AD pathology. Using postmortem human hippocampus, APP/PS1 mice, and iPSC-derived AD neurons, they demonstrated that basal mitophagy is reduced across all three systems. Small-molecule mitophagy inducers—urolithin A and nicotinamide mononucleotide—reduced amyloid-beta burden, tau phosphorylation, and cognitive deficits. The NAD^+ decline framework was elaborated by Lautrup, Sinclair, Mattson, and Fang (2019), who argued that age-related NAD^+ depletion is a central driver of bioenergetic failure.

2.6 Microglial Metabolic Reprogramming

Ulland et al. (2017) demonstrated that TREM2-DAP12-SYK signaling through PI3K-AKT-mTOR supports microglial oxidative phosphorylation. Baik et al. (2019) exposed primary microglia to amyloid-beta and reported an initial glycolytic shift followed by a collapsed hypometabolic state with impaired capacity to sustain both glycolytic and oxidative phosphorylation output—a metabolic collapse rather than a mere metabolic switch.

2.7 Neuroinflammation and the NLRP3 Inflammasome

Heneka and colleagues established NLRP3 as a critical amplifier of AD pathology: *Nlrp3* deletion reduces pathology in APP/PS1 mice (Heneka et al., 2013); microglia-derived ASC specks bind amyloid-beta and accelerate aggregation (Venegas et al., 2017); and NLRP3 activation contributes to tau pathology (Ising et al., 2019). Mitochondrial DAMPs (mtROS, oxidized mtDNA, cardiolipin externalization) are among the best-characterized NLRP3 activators.

2.8 Mitochondrial Allostatic Load and Mitohormesis

Picard, Juster, and McEwen (2014) introduced mitochondrial allostatic load, proposing that mitochondria integrate chronic stress signals across hormonal, immune, metabolic, and neural inputs. Schulz et al. (2007) demonstrated mitohormesis: caloric restriction extends lifespan through increased mitochondrial respiration and transient ROS elevation that triggers adaptive upregulation of NRF2 and AMPK-PGC-1α-mediated biogenesis.

2.9 Gaps in the Literature

Four significant gaps persist. First, the mitochondrial and autophagy-lysosomal literatures have largely developed in isolation from microglial biology. Second, the role of NAD^+-consuming enzymes—particularly CD38—in microglial bioenergetic collapse has not been connected to neurodegeneration-specific phenotypes. Third, the environmental neurotoxin literature (BMAA, annonacin) has been treated as disease-specific rather than as natural experiments illuminating selective mitochondrial complex inhibition. Fourth, therapeutic implications of mitochondrial quality-control restoration have not been integrated into a single mechanistic framework.

3

Methodology

This dissertation adopts a systems-level integrative review methodology, synthesizing primary experimental literature, clinical trial data, and theoretical frameworks across cellular and molecular neuroscience, immunology, metabolism, and pharmacology.

Primary sources were selected according to four criteria: (a) publication in peer-reviewed journals indexed in PubMed/MEDLINE; (b) experimental methodology adequate to support the claims cited; (c) relevance to one or more of the nine core research programs; and (d) recency, with preference for publications after 2015 except for foundational work. A total of 135 Oskar Fischer Prize entrant submissions were evaluated using the Synthesis-Relevance Rescore system against a registry of 109 Tier-1 and 126 Tier-2 mechanisms across the three collapse domains.

The analysis proceeds through three levels of integration: **organelle-level** (mapping the functional architecture of the mitochondrion and its coupling to adjacent systems), **pathology-level** (tracing how each disease protein targets mitochondrial function), and **systems-level** (examining how cell-autonomous mitochondrial failure propagates through intercellular signaling and systemic metabolism). Citations follow APA 7th edition format.

Chapter I

The Bioenergetic Architecture of the Neuron

4.1 The Electron Transport Chain: Complexes I–V

Complex I (NADH:ubiquinone oxidoreductase) accepts electrons from NADH, transfers them to ubiquinone, and pumps four protons across the inner membrane. It is the largest ETC complex (~45 subunits) and the primary site of electron leak generating superoxide (Brand, 2010). Complex I is the target of rotenone, MPTP/MPP^+, annonacin, and—as discussed in Chapter II—alpha-synuclein and TDP-43. The convergence of environmental toxins and disease proteins on this single complex is among the strongest circumstantial arguments for mitochondrial dysfunction as a causal variable.

Complex II (succinate dehydrogenase) is the only ETC complex entirely encoded by nuclear DNA. Complex II deficiency is the primary bioenergetic lesion in Huntington's disease (Gu et al., 1996), and systemic Complex II inhibition by 3-nitropropionic acid produces striatal lesions closely resembling HD pathology (Beal et al., 1993).

Complex III (cytochrome bc_1) transfers electrons from ubiquinol to cytochrome c and pumps protons via the Q-cycle. It is the second major site of superoxide generation. **Cytochrome c** shuttles electrons from Complex III to Complex IV; its release to the cytoplasm upon outer-membrane permeabilization triggers intrinsic apoptosis through caspase-9 activation (Liu et al., 1996). This dual function—electron carrier and death signal—makes cytochrome c the molecular switch where bioenergetic failure becomes cell death.

Complex IV (cytochrome c oxidase) catalyzes the terminal transfer of electrons to molecular oxygen. It is consistently reduced 25–40% in AD brain tissue and platelets (Parker et al., 1990; Mutisya et al., 1994). Swerdlow's cybrid studies demonstrated this deficiency is transmissible via mtDNA (Swerdlow et al., 1997). **Complex V (ATP synthase)** converts the proton-motive force into ATP; under severe membrane potential loss, it can reverse, hydrolyzing ATP in a futile attempt to restore potential (Chinopoulos & Adam-Vizi, 2010).

Cytochrome P450 enzymes are relevant through CYP46A1 (cholesterol 24-hydroxylase), the primary neuronal cholesterol elimination enzyme operating at the MAM, and CYP27A1, a mitochondrial P450 linking cholesterol metabolism to oxysterol-mediated neuroinflammation.

4.2 The Energy Currency: NAD^+/NADH and the CD38 Problem

NAD^+ is far more than an electron shuttle: it is the essential co-substrate for sirtuins (SIRT1–7), poly(ADP-ribose) polymerases (PARPs), and CD38/CD157. NAD^+ depletion therefore simultaneously cripples oxidative phosphorylation, mitochondrial biogenesis (through sirtuin-PGC-1α pathway failure), and DNA repair (Verdin, 2015).

Camacho-Pereira et al. (2016) identified CD38 as the dominant NAD^+-consuming enzyme in aging tissue. CD38 expression increases with age and inflammation, and CD38 knockout mice are protected from age-related NAD^+ decline. The critical relevance: CD38 is highly expressed on activated microglia. Microglial activation in AD therefore upregulates the very enzyme that depletes the NAD^+ required for microglial oxidative phosphorylation—a self-destructive feedback loop. The NAD^+/NADH ratio in stroke provides a natural experiment in acute bioenergetic crisis: ischemia causes catastrophic NAD^+ depletion primarily through PARP1 hyperactivation.

4.3 The Gateway: TOM40 and Mitochondrial Protein Import

Over 99% of mitochondrial proteins are encoded in the nuclear genome and must be imported through TOM40 (Wiedemann & Pfanner, 2017). The *TOMM40* gene lies in tight linkage disequilibrium with *APOE* on chromosome 19q13.32. Roses et al. (2010) identified a poly-T repeat polymorphism in *TOMM40* that correlates with AD onset age independently of APOE genotype, suggesting a direct genetic link between mitochondrial protein import efficiency and AD susceptibility. Hansson Petersen et al. (2008) demonstrated that amyloid-beta peptides can be imported through TOM40 into the matrix, where they inhibit Complex IV.

4.4 The Contact Site: Mitochondria-Associated Membrane (MAM)

The MAM is a specialized ER subdomain physically tethered to the outer mitochondrial membrane (Csordás et al., 2018). It serves as a functional platform for three processes critical to neurodegeneration:

Calcium transfer. The IP3R–GRP75–VDAC–MCU axis couples ER calcium release to mitochondrial uptake, stimulating TCA cycle dehydrogenases and boosting ATP production (Rizzuto et al., 2012). Under pathological conditions, calcium transfer becomes excessive, activating the mitochondrial permeability transition pore.

BAP31 (B-cell receptor-associated protein 31) is an ER-resident chaperone at the MAM that regulates apoptosis signaling. Its cleavage generates p20BAP31, which promotes calcium release from ER into mitochondria. **ABAD** (amyloid-binding alcohol dehydrogenase; HSD17B10) is a mitochondrial matrix enzyme that amyloid-beta binds directly, distorting its NAD^+-binding site and amplifying ROS production (Lustbader et al., 2004).

Lipid metabolism. Area-Gomez et al. (2012) demonstrated that C99 accumulates at MAMs and functions as a cholesterol sensor, driving upregulated MAM activity including aberrant cholesterol ester and phospholipid synthesis—disrupting both mitochondrial membrane composition and ER lipid homeostasis simultaneously.

4.5 Calcium: The Universal Coupling Signal

Neurons maintain cytosolic calcium at approximately 100 nM against a 2 mM extracellular concentration—a 20,000-fold gradient maintained by ATP-dependent pumps. When mitochondrial ATP production declines, these pumps fail, and resting calcium rises. The consequences cascade: calpain activation (cleaving cytoskeletal proteins and v-ATPase subunits), mitochondrial calcium overload (opening the mPTP), and excitotoxic NMDA receptor activation (Khachaturian, 1987). Presenilin 1 and 2 mutations dysregulate ER calcium stores through effects on ryanodine and IP3 receptors (Tu et al., 2006).

4.6 Mitochondrial Quality Control: Mitophagy and Fission/Fusion

Fission/fusion dynamics represent the first-line triage: **fusion** (MFN1/2, OPA1) rescues mildly damaged mitochondria by functional complementation; **fission** (DRP1, FIS1) segregates irreparably damaged segments for mitophagy disposal (Youle & van der Bliek, 2012). **Ubiquitin** is the molecular tag for the entire system—Parkin writes polyubiquitin chains on the outer membrane, read by autophagy receptors (p62, OPTN, NDP52), and edited by deubiquitinases (USP30, USP15) that determine whether a mitochondrion is degraded or rescued (Harper et al., 2018).

The endolysosomal system is the terminal executor: autophagosomes must fuse with lysosomes for degradation, requiring v-ATPase acidification (ATP-dependent), functional cathepsins (pH-dependent), and intact SNARE fusion machinery. When any of these fail, mitophagy stalls at the completion step—this is PANTHOS (Nixon).

4.7 Energy Production and Reactive Oxygen Species

ETC electron leak generates superoxide primarily at Complex I and Complex III. At low levels, mitochondrial ROS serve essential signaling functions—activating NRF2 and supporting the **mitohormetic** adaptive response (Ristow & Schmeisser, 2014). The pathological transition occurs when ROS production exceeds antioxidant capacity: oxidative damage to mtDNA produces mutations that further impair the ETC, generating more ROS—the “vicious cycle” (Linnane et al., 1989). Oxidized mtDNA fragments released to the cytoplasm activate cGAS-STING and NLRP3, converting organelle damage into inflammatory amplification.

4.8 Synapse Function and GABAergic Vulnerability

Synaptic transmission is the most energy-intensive process in the brain. **GABAergic parvalbumin-positive (PV+) fast-spiking interneurons** represent the most bioenergetically demanding neuronal population—they fire at gamma frequency (30–80 Hz) continuously, requiring the highest per-neuron ATP demand of any cell type (Kann et al., 2014). They are ensheathed by perineuronal nets that protect them from oxidative stress (Cabungcal et al., 2013). PV+ interneuron loss is an early feature of AD (Verret et al., 2012), and Li-Huei Tsai's GENUS paradigm demonstrates that 40 Hz sensory stimulation restores mitochondrial ATP production and rescues gamma oscillations (Iaccarino et al., 2016).

4.9 Environmental Toxins as Natural Experiments: BMAA and Annonacin

Annonacin, a lipophilic Complex I inhibitor from Annonaceae fruits, produces a progressive supranuclear palsy-like tauopathy in rats without amyloid pathology (Champy et al., 2004; Höglinger et al., 2005), demonstrating that selective Complex I inhibition is sufficient to produce tauopathy. **BMAA**, a cyanobacterial neurotoxin, inhibits Complex I, generates ROS, and produces TDP-43 aggregation (Dunlop et al., 2013) and neurofibrillary tangles with amyloid plaques in primate models (Cox et al., 2016). These natural experiments demonstrate that mitochondrial poisoning alone is sufficient to produce the full neuropathological spectrum.

Chapter II

Disease Proteins as Mitochondrial Toxins

5.1 The Transdiagnostic Convergence

The five major aggregation-prone proteins—amyloid-beta, tau, alpha-synuclein, huntingtin, and TDP-43—are conventionally treated as disease-defining markers with distinct pathologies. This chapter demonstrates that each independently targets mitochondrial function through specific biochemical mechanisms, and that the downstream consequences—bioenergetic failure, ROS amplification, calcium dysregulation, and quality-control overload—are convergent.

5.2 Amyloid-Beta

Amyloid-beta interacts with mitochondria at multiple points. It is imported into the matrix via TOM40 (Hansson Petersen et al., 2008) where it inhibits Complex IV. It binds ABAD in the matrix, distorting its NAD^+-binding domain and amplifying ROS (Lustbader et al., 2004). A-beta oligomers increase MAM contacts and calcium transfer, promoting mitochondrial overload (Hedskog et al., 2013). A-beta exposure induces fragmentation but simultaneously impairs PINK1/Parkin mitophagy (Fang et al., 2019). And A-beta activates NF-κB, which drives APP transcription (Grilli et al., 1995), creating a feed-forward loop: **mitochondrial damage ☐ NF-κB ☐ APP transcription ☐ A-beta ☐ more mitochondrial damage**.

5.3 Tau

Hyperphosphorylated tau disrupts mitochondria through two mechanisms. First, axonal transport failure: hyperphosphorylated tau detaches from microtubules and obstructs motor proteins, producing synaptic mitochondrial depletion (Kopeikina et al., 2011; Dixit et al., 2008). Second, direct ETC inhibition: truncated tau enters the intermembrane space and inhibits Complex I (Quintanilla et al., 2009). The relationship is bidirectional—Complex I inhibition by annonacin causes tau hyperphosphorylation (Höglinger et al., 2005), establishing that ATP depletion is sufficient to drive tau pathology.

5.4 Alpha-Synuclein

Alpha-synuclein oligomers bind directly to Complex I, reducing its activity by 20–40% (Devi et al., 2008). Alpha-synuclein localizes to MAMs, disrupting calcium signaling (Guardia-Laguarta et al., 2014). It impairs mitophagy by sequestering cardiolipin and interfering with Parkin recruitment (Shaltouki et al., 2018). And it promotes mitochondrial fragmentation, rescuable by DRP1 inhibition (Nakamura et al., 2011).

5.5 Mutant Huntingtin

Mutant huntingtin impairs Complex II activity selectively (Benchoua et al., 2006)—the Complex II selectivity may partly explain HD's striatal targeting. It represses PGC-1α transcription (Cui et al., 2006), impairing biogenesis. It promotes DRP1-mediated fission (Song et al., 2011). And it sensitizes IP3 receptors at the ER, increasing calcium release to mitochondria (Tang et al., 2003).

5.6 TDP-43

Wang et al. (2016) demonstrated that TDP-43 contains a mitochondrial localization signal, is imported into the inner membrane, and directly inhibits Complex I by binding ND3 and ND6 subunits. Blocking TDP-43 mitochondrial import prevents neurodegeneration in animal models—demonstrating that a protein classified as a nuclear RNA-binding factor has a direct, independent mitochondrial toxicity mechanism.

5.7 Synthesis: The Protein-Convergent Mitochondrial Hypothesis

Every major neurodegenerative disease protein independently poisons the ETC, with partial selectivity for different complexes—Complex I (A-beta, alpha-synuclein, TDP-43), Complex II (huntingtin), Complex IV (A-beta)—and all impair quality control through fission/fusion, mitophagy, and/or MAM dysfunction. The evidence from environmental toxins (MPTP, rotenone, annonacin, BMAA) supports the strong interpretation: mitochondrial poisoning alone, without any disease protein, is sufficient to produce neurodegeneration with proteinopathy.

Chapter III

The Immunometabolic Dimension

6.1 Innate Immunity and Mitochondrial DAMPs

Mitochondria are evolutionary endosymbionts of alpha-proteobacterial origin, and their molecular components retain bacterial signatures recognized by the innate immune system as danger signals. These mitochondrial DAMPs include:

- **Mitochondrial DNA:** circular, CpG-rich; recognized by TLR9 and cGAS (Zhang et al., 2010; West et al., 2015)
- **Cardiolipin:** externalized on the outer membrane, serves as both mitophagy signal and NLRP3 activator (Iyer et al., 2013)
- **Mitochondrial ROS:** activate NLRP3 through TXNIP dissociation from thioredoxin (Zhou et al., 2011)
- **N-formyl peptides:** mitochondrial translation products recognized by formyl peptide receptors
- **Extracellular ATP:** activates P2X7 receptors, triggering K^+ efflux and NLRP3 assembly

6.2 The NLRP3 Inflammasome as Mitochondrial-ROS-Gated Amplifier

Mitochondrial dysfunction provides both signals required for NLRP3 activation simultaneously: ROS-mediated NF-κB activation primes the inflammasome; mtROS, oxidized mtDNA, and externalized cardiolipin provide the activation signal. Heneka et al. (2013) demonstrated that *Nlrp3* deletion reduces AD pathology. Venegas et al. (2017) showed that ASC specks bind A-beta and accelerate aggregation—a feed-forward amplification: **mitochondrial damage ☐ NLRP3 ☐ ASC specks ☐ A-beta aggregation ☐ more mitochondrial damage**.

6.3 NF-κB: The Master Inflammatory Transcription Factor

Mitochondrial dysfunction activates NF-κB through multiple routes: mtROS oxidize IKK; mtDNA activates cGAS-STING; NLRP3-dependent IL-1β activates NF-κB in neighboring cells. The human APP promoter contains functional NF-κB binding sites (Grilli et al., 1995), meaning mitochondrial dysfunction drives increased APP transcription and A-beta production. The loop: **mitochondrial damage ☐ ROS ☐ NF-κB ☐ APP transcription ☐ A-beta ☐ mitochondrial damage**. This converts gradual bioenergetic decline into an accelerating cascade.

6.4 CD38 and the NAD^+ Drain

CD38 creates a devastating metabolic trap: microglial activation upregulates CD38, which consumes the NAD^+ pool; NAD^+ depletion cripples microglial OXPHOS; bioenergetically compromised microglia generate more mtDAMPs; mtDAMPs activate NLRP3 and NF-κB, driving further CD38 upregulation. Chini et al. (2020) demonstrated that CD38 inhibition rescues tissue NAD^+ levels and mitochondrial function in aged mice. Covarrubias et al. (2020) showed senescence-associated cytokines drive CD38 upregulation, creating a "NAD^+ sink."

6.5 Inflammatory T-Cell Infiltration and Metabolic Competition

Gate et al. (2020) demonstrated clonally expanded $CD8^+$ T cells in AD CSF. From the bioenergetic perspective, infiltrating T cells are highly glycolytic (consuming local glucose), express CD38 (contributing to NAD^+ consumption), and secrete IFN-γ (reprogramming microglial metabolism toward glycolysis). Their perforin and granzymes can directly damage mitochondria in target cells.

6.6 Complement Activation and Synaptic Stripping

Damaged mitochondria expose surface molecules recognized by complement C1q. In the developing brain, C1q and C3 tag weak synapses for microglial pruning (Stevens et al., 2007). In AD, this program is aberrantly reactivated: synapses weakened by bioenergetic failure are tagged by complement and eliminated by microglia (Hong et al., 2016). Beth Stevens' work represents arguably the most important validation for the connection between microglial homeostatic failure and synaptic vulnerability.

6.7 Ubiquitin: The Shared Molecular Tag

The ubiquitin-proteasome system and autophagy-lysosome system are the two principal degradative pathways; both require ubiquitin tagging and are ATP-dependent. Proteotoxic stress from aggregation-prone proteins overwhelms the UPS, routing excess cargo to the autophagy pathway—which is already overloaded with mitophagic cargo. The result is competition for limited degradative capacity: misfolded proteins and damaged mitochondria compete for the same autophagy machinery (Rubinsztein, 2006; Dikic, 2017).

Chapter IV

Therapeutic Implications and Experimental Predictions

7.1 Methylene Blue: The Electron Carrier Bypass

Methylene blue can accept electrons from NADH and transfer them directly to cytochrome c, bypassing Complex I and III blockades (Atamna et al., 2008). This makes it a direct pharmacological test of the mitochondrial hypothesis. Preclinical evidence shows improved respiration, reduced ROS, and cognitive rescue in AD models (Medina et al., 2011). Clinical trials of LMTM showed mixed results, with a signal in the monotherapy subgroup (Gauthier et al., 2016). The framework predicts efficacy should correlate with degree of ETC dysfunction—patient stratification by bioenergetic status has not been attempted.

7.2 Mitophagy Inducers: Urolithin A and NAD^+ Precursors

The Fang et al. (2019) demonstration of AD pathology rescue through mitophagy enhancement represents the most direct preclinical validation. Urolithin A has completed phase I safety trials (Andreux et al., 2019). The therapeutic logic is upstream correction: rather than clearing aggregates after formation, mitophagy induction removes the damaged mitochondria whose dysfunction drives proteinopathy. This approach should be complementary to anti-amyloid therapy.

7.3 Lysosomal Acidification Restoration

If v-ATPase failure is the bottleneck where mitophagy stalls, restoring lysosomal acidification could rescue the terminal step. Candidates include TRPML1 agonists, TFEB activators, and v-ATPase enhancers. The Nixon laboratory's PANTHOS work establishes the rationale; clinical translation is nascent.

7.4 Systemic Metabolic Interventions

The transdiagnostic extension identifies ketogenic metabolic therapy, intermittent fasting, metformin, GLP-1 receptor agonists, and exercise. Nørgaard et al. (2022) reported that pooled RCT data (n = 15,820) show GLP-1 receptor agonists lower dementia incidence—the first large-scale human validation of metabolic-rescue pharmacology against cognitive endpoints.

7.5 Haplotype-Based Risk Stratification

The MCH predicts that inherited mitochondrial function—determined by mtDNA haplogroup and *TOMM40* variants—sets the bioenergetic ceiling from which age-related decline proceeds. Ridge et al. (2012) reported mtDNA haplogroup associations with AD risk. If validated, this supports haplotype-based patient stratification for clinical trials.

7.6 Experimental Predictions

This thesis generates five falsifiable predictions that distinguish the bioenergetic framework from the protein-first alternative:

P1. Microglia-specific conditional deletion of PINK1 or Parkin should produce age-dependent loss of the homeostatic signature, emergence of dystrophic features, and accelerated pathology when crossed into AD models.

P2. Single-cell mitophagy imaging paired with transcriptomics in AD microglia should reveal a graded distribution: DAM (highest flux) ☐ LDAM (intermediate) ☐ dystrophic (lowest).

P3. Mitophagy inducers or NAD^+ precursors in AD models should preserve perineuronal net integrity and PV^+ interneuron markers, with cognitive preservation tracking PNN integrity more tightly than plaque or tau burden.

P4. Chronic low-dose annonacin or rotenone should produce tau pathology across brain regions, with selectivity correlating with local mitochondrial density and metabolic demand.

P5. CD38 inhibition or genetic deletion in APP/PS1 mice should restore microglial NAD^+ levels, oxidative phosphorylation capacity, and homeostatic signature markers.

Conclusion

This dissertation has argued that mitochondrial dysfunction is not a peripheral consequence of proteinopathy but a rate-limiting variable whose decline sets the threshold for neurodegenerative disease initiation. The argument rests on three pillars.

First, the bioenergetic architecture of the neuron creates an inherent vulnerability: the ETC, NAD^+/NADH redox couple, TOM40 import machinery, MAM, calcium signaling apparatus, and mitophagy/fission/fusion quality-control system constitute an integrated functional unit. Failure at any node cascades to all others through the shared ATP dependency and the v-ATPase bottleneck.

Second, every major neurodegenerative disease protein—amyloid-beta, tau, alpha-synuclein, huntingtin, and TDP-43—independently targets the ETC through specific biochemical mechanisms. Environmental toxins demonstrate that mitochondrial poisoning alone is sufficient to produce neurodegeneration with proteinopathy, supporting the interpretation that mitochondrial decline is an upstream vulnerability.

Third, mitochondrial damage generates a stereotyped innate immune response—through NLRP3, NF-κB, complement, and DAMP signaling—that amplifies tissue injury. CD38-mediated NAD^+ consumption creates a self-destructive metabolic trap, and NF-κB–APP feed-forward amplification converts gradual decline into accelerating pathology.

The framework is compatible with, not competing against, amyloid and tau biology. Where it differs is in placing mitochondrial quality control at a plausible upstream position rather than treating bioenergetic abnormalities as downstream consequences. The five predictions in §7.6 are designed to falsify the model's load-bearing claims; their experimental resolution will determine whether the framework is correct, partially correct, or in need of substantial revision.

The work ahead is empirical. What this dissertation offers is a sharpened question, an integrated mechanistic framework, and a set of falsifiable predictions whose resolution will determine the model's fate.

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