Medical Hypotheses (2004) **63**, 8–20

ELSEVIER

medical hypotheses

http://intl.elsevierhealth.com/journals/mehy

A "mitochondrial cascade hypothesis" for sporadic Alzheimer's disease

Russell H. Swerdlow[a,b,*], Shaharyar M. Khan[b,c,d]

[a] *Department of Neurology, McKim Hall, University of Virginia Health System, P.O. Box 800394, 1 Hospital Drive, Charlottesville, VA 22908, USA*
[b] *Center for the Study of the Neurodegenerative Diseases, University of Virginia Health System, Charlottesville, VA 22908, USA*
[c] *Department of Neuroscience, University of Virginia Health System, Charlottesville, VA 22908, USA*
[d] *Gencia Corporation, 706 D Forrest Street, Charlottesville, VA 22903, USA*

Received 14 October 2003; accepted 30 December 2003

Summary Alzheimer's disease (AD) includes etiologically heterogenous disorders characterized by senile or presenile dementia, extracellular amyloid protein aggregations containing an insoluble amyloid precursor protein derivative, and intracytoplasmic tau protein aggregations. Recent studies also show excess neuronal aneuploidy, programmed cell death (PCD), and mitochondrial dysfunction. The leading AD molecular paradigm, the "amyloid cascade hypothesis", is based on studies of rare autosomal dominant variants and does not specify what initiates the common late-onset, sporadic form. We propose for late-onset, sporadic AD a "mitochondrial cascade hypothesis" that comprehensively reconciles seemingly disparate histopathologic and pathophysiologic features. In our model, the inherited, gene-determined make-up of an individual's electron transport chain sets basal rates of reactive oxygen species (ROS) production, which determines the pace at which acquired mitochondrial damage accumulates. Oxidative mitochondrial DNA, RNA, lipid, and protein damage amplifies ROS production and triggers three events: (1) a *reset* response in which cells respond to elevated ROS by generating the β-sheet protein, beta amyloid, which further perturbs mitochondrial function, (2) a *removal* response in which compromised cells are purged via PCD mechanisms, and (3) a *replace* response in which neuronal progenitors unsuccessfully attempt to re-enter the cell cycle, with resultant aneuploidy, tau phosphorylation, and neurofibrillary tangle formation. In addition to defining a role for aging in AD pathogenesis, the mitochondrial cascade hypothesis also allows and accounts for histopathologic overlap between the sporadic, late-onset and autosomal dominant, early onset forms of the disease.

Introduction

As described by Alois Alzheimer in 1906 and named by Emil Kraepelin in 1910, Alzheimer's disease (AD) applied to a state of presenile dementia, extraneuronal protein aggregations (plaques), and intraneuronal protein aggregations (tangles) [1,2]. Although it was recognized at the time that brains of persons with senile dementia could also manifest plaques and tangles, in the elderly this was not felt to represent an actual disease state [3–6].

* Corresponding author. Tel.: +1-434-924-5785; fax: +1-434-982-1726.
E-mail address: rhs7e@virginia.edu (R.H. Swerdlow).

0306-9877/$ - see front matter
doi:10.1016/j.mehy.2003.12.045

In the latter half of the 20th century, the AD spectrum expanded to include all plaque and tangle dementias regardless of age [7—10]. It was further proposed that this now common neurodegenerative condition was not a consequence of either normal or accelerated aging, but rather distinctly abnormal pathophysiologic events. To clarify the nature of this abnormal pathophysiology, investigators elucidated genetic defects underlying multiple (albeit rare) families with autosomal dominant, early onset forms. It was found mutation of the amyloid precursor protein (APP) gene and two other genes likely involved with APP processing, presenilin 1 and presenilin 2, cause presenile dementia with plaque formation [11—13]. In particular, the ability of APP mutation to cause an AD-consistent clinical and histopathologic phenotype justified the "amyloid cascade hypothesis" [14,15]. According to this hypothesis, the primary event in AD neurodegeneration is production of the beta amyloid (Aβ) derivative of APP [16—18].

Accumulating evidence suggests that although the amyloid cascade hypothesis is potentially (if not likely) viable in cases of APP, presenilin 1, or presenilin 2 derived AD, it may not apply in its current form to the late-onset, sporadic type of the disease (the vast majority) [19]. First, persons with the common form of AD generally lack mutations of these genes, and so it is unclear what initiates plaque formation in such cases. Second, plaques are a relatively common finding in the non-demented elderly [20—22]. Third, pathways through which plaques generate tangles and other recently described AD pathophysiology are unknown. This includes neuronal apoptosis, neuronal aneuploidy, and cerebral/extracerebral mitochondrial dysfunction [19,23—25].

AD is now identified as a "disease of aging", which implies aging itself is not a disease (otherwise the term is an oxymoron). This semantic trap requires one to overlook the fact that boundaries between late-onset AD and "normal" aging are not absolute. Neuropsychologic test performance decline, brain atrophy, neuronal loss, and plaque/tangle deposition all occur with aging in the absence of frank dementia [26]. For late-onset AD, therefore, it is reasonable to place the causal molecular events within an aging spectrum, rather than consider them distinct disease phenomena. By this logic, some individuals are "set" to develop sporadic AD at a relatively young age, others at an intermediate age, and yet others only at a very advanced age.

We now propose a hypothesis that places AD within the context of developmental and aging theory. The hypothesis takes into account current molecular knowledge of cell division, differentiation, de-differentiation, and demise. We first review relevant scientific principles.

The cell cycle, redox status, and reactive oxygen species

All nucleus-endowed cells contain genetic programs that allow for their division and execution. Recent data suggest a single mediator, the cell redox state (which is reflected by ratios of particular oxidized and reduced substrate variants, such as NAD^+ and NADH), and by extension reactive oxygen species (ROS), regulates the balance between these diametric processes [27—30]. The main determiner of intracellular ROS and overall cell redox states is the mitochondrial electron transport chain (ETC) [31—33]. In experimental systems, limited ROS (H_2O_2 and O_2^-) exposures induce multiple cell types to enter the proliferation cycle, while increasing ROS amounts above such limited thresholds activates apoptotic cell death pathways [34]. Redox status and ROS levels outside ranges specifically associated with either cell proliferation or cell demise are found in cells that are neither dividing nor dying, but rather existing in a stable state of physiologic growth arrest ("G_0"). Stem or progenitor cells comprise a unique category of cells that can undergo growth arrest, yet do not lose their ability to pass through the cell cycle [35].

The avascular status of a developing organism during embryogenesis limits aerobic metabolism. Thus, the expanding, unperfused cell mass must flourish under relatively anaerobic conditions [36]. It is by necessity over-reliant on glycolytic (anaerobic) metabolism, which generates NADH. Mitochondrial ROS production is limited [37]. Accordingly, when embryo cells are delivered from mitosis ("M") into the initial "gap" period (G_1) of interphase, ROS and NAD^+/NADH regulation signals are not set to prompt the cell's exit from reproductive cycling [38,39]. G_0 status is not achieved, G_1 proceeds, and proteins ultimately necessary for cell division are produced. Subsequent DNA replication (in the "S" phase) results in tetraploidy.

Cells reaching the post-S phase "second gap" (G_2) are not obligated to proceed from interphase to mitosis ("G_2-M arrest"). The bioenergetic status of the cell, in particular, regulates whether passage from G_2 to M occurs. Low ATP levels are associated with G_2-M arrest [40,41].

When mitosis does occur, microtubules form spindles that appropriately segregate chromosomes

into daughter nuclei. Tau protein is likely relevant to cell cycling physiology at this point, because as a microtubule-associated protein it is designed to bind microtubules [42]. This transpires whether microtubules act as cytoskeletal elements in differentiated cells or mitotic spindles in undifferentiated cells [43]. In the rapidly dividing cells of developing organisms, tau is present in a phosphorylated state (fetal tau). Tau phosphorylation is therefore seen not only in the neurofibrillary tangles of AD and normal aging, but also during early development and, in general, mitotic cells [26,42,44—46].

Mitochondria: relation to aging, cell death, and APP

A "mitochondrial" or "free radical" theory of aging derives from data suggesting (1) ETC activity declines with age [47—50], and (2) mitochondrial-based oxidative stress increases with age [51—61]. The underlying basis for this age-dependent mitochondrial decline is controversial. Some emphasize mitochondrial DNA (mtDNA) deletions and point mutations accumulate with age, perhaps due to oxidative stress [57,58,62—69]. Detractors counter demonstrable mutational burdens are low, and question their phenotypic significance [70]. Some argue within post-mitotic cells malfunctioning mitochondria have a replicative advantage, and thereby assume an ever-increasing proportion of the total cell mitochondria [71,72]. Others hypothesize damaged mitochondria are favored because of reduced degradation rates [73—75].

Mechanistic issues notwithstanding, oxidative stress does appear to influence longevity. Life extension occurs in fruit flies engineered to better detoxify the free radical byproducts of oxidative metabolism [76]. Experimental caloric restriction in animals also extends life span, perhaps by indirectly reducing oxidative metabolism-related oxidative stress [77,78].

Recent data now implicate mitochondrial dysfunction as an initiating event in apoptotic programmed cell death (PCD) pathways [79,80]. In the "intrinsic" apoptosis pathway, when mitochondrial depolarization, oxidative stress, or bioenergetic failure surpasses a threshold, permeability transition is triggered. This allows efflux of molecules typically sequestered within the mitochondrial compartment, and subsequent activation of cell death cascades [81—86].

Proteins that affect ETC function may influence mitochondrial ROS production [87—89]. In this respect APP is relevant, since it is partly targeted to mitochondria and under pathologic conditions may induce ETC dysfunction and alter oxidative stress levels [90]. Oxidative stress, in turn, can induce soluble proteins to adopt insoluble β-pleated sheet conformations, or else yield β-sheet derivatives. Interestingly, precedent exists for the insertion of β-sheet proteins in mitochondrial membranes, where they are predicted to form pores [91]. It is tempting to consider existence of a feedback loop, in which mitochondria overproducing ROS initiate conformational changes in local proteins that then "shut down" the mitochondria that drive their formation.

The ability of the APP derivative Aβ to complex elemental and organic cations may also serve to alter mitochondrial function. Aβ is a β-sheet "biobioflocculant" that chelates organic and elemental iron and copper, redox-active metal ions abundant in mitochondria [92]. Attomolar concentrations of iron and copper induce monomeric Aβ to oligomerize, forming insoluble precipitates that in turn sequester the ions that enable their aggregation [93]. As copper and iron are required for electron transport, chelation of these ions may indirectly inhibit oxidative phosphorylation. In indirect support of this are two findings: (1) micromolar concentrations of Aβ(25—35) peptide have no effect on cells that do not possess a functional ETC [94], and (2) glycolytic upregulation ameliorates Aβ toxicity by decreasing cell reliance on oxidatively derived ATP production [95]. Subsequent extracellular secretion of metal-chelated Aβ from the cytoplasm via the Golgi apparatus would predictably give rise to insoluble amyloid plaques, which presumably would activate local gliosis and microglial invasion.

Pre-translational mRNA oxidation may also contribute to a protein aggregation diathesis in both aging and AD [96,97]. Peptides produced from oxidized mRNA species are more likely to aggregate than peptides produced from non-oxidized mRNA species [98]. Excessive mRNA oxidation is observed in AD brain, but appears to represent a highly selective process that affects only particular transcripts [98]. This selectivity may arise from the fact that translation is a cytoanatomically specific event. Indeed, as is the case with yeast, in human cells certain mRNA species are translated by ribosomes that reside tethered to the mitochondrial outer membrane [99—101]. In the case of increased mitochondrial ROS production, peri-mitochondrial translation would promote cytoanatomically selective mRNA free radical exposure, with subsequent aggregation of the translational products. To date, however, it remains to be shown that APP and tau mRNA from AD brain exhibit excessive oxidation [98].

One final point about ROS production is in order. ROS are an unavoidable byproduct of cell metabolism. Cell metabolism, in turn, is defined by the interplay between multiple interdependent enzyme systems that are designed to facilitate substrate cycling. Compromise of one biochemical system tends to induce compensatory (although not necessarily advantageous) changes in other systems. In addition to the mitochondrial ETC, other sites and enzyme systems participate in the redox cycling reactions that maintain appropriate cell NAD^+/NADH ratios. These include cytoplasmic glycolysis and lactate production, fatty acid ∃-oxidation and conversion of pyruvate to acetyl CoA in the mitochondrial matrix, peroxisomal oxidation of fatty acids, and activity of the plasma membrane oxidoreductase system. Further, it appears that diminished redox cycling by the mitochondrial ETC is associated with increased redox cycling at other cell sites, specifically the plasma membrane oxidoreductase complex [102—104]. "Shifting" of certain redox chemical reactions from one cell locale to another facilitates conservation.

Cytoanatomic redox shifts are seen in various cell types and under various conditions. Cells that lack a functional ETC because of mtDNA depletion (ρ0 cells) show elevated plasma membrane ROS production [102,103]. Tumor cells are also characterized by low levels of cytoplasmic ROS and elevated levels of plasma membrane ROS [105,106]. Specific ETC enzyme activities and overall oxidative phosphorylation are reduced in tumor cells [107]. Similar mechanisms may also apply to non-tumor hepatocytes, in which reduced oxidation phosphorylation capacity is part of a physiologic "de-differentiation" process that occurs when local tissue repair responses are activated [108]. Taken together, these findings are consistent with the view that relatively anaerobic/glycolytic cells are capable of cell division, and shift redox maintenance from the mitochondrial ETC to the plasma membrane oxidoreductase system.

Mitochondrial function in development and aging

The most distinctive feature of mitochondria is their ability to perform electron transport. Evolution has facilitated the development of several ETC enzyme complexes for this purpose. Four particular ETC complexes (I, II, III, and IV) harness energy from mobilized free electrons, and use this energy to drive proton translocation. An additional complex (V) allows protons to re-access the matrix, and couples energy from this proton flux to ADP phosphorylation.

Multimeric ETC complexes contain protein subunits that derive from two cell genomes, the nuclear and mitochondrial. For example, 7 of the over 40 proteins that comprise complex I are mitochondrial DNA (mtDNA) encoded. One of 11 complex III, three of 13 complex IV, and two of 14 complex V subunits also arise from mtDNA.

There is substantial polymorphic variability in both the mtDNA and nuclear DNA (nDNA) ETC subunit genes [109,110]. These polymorphisms frequently alter amino acids. With so many polymorphic genes giving rise to participant peptides, considerable ETC structural variation exists between individuals. Emerging data indicate this variability may influence a spectrum of ontologic events, including development, aging, and neurodegeneration [111—114]. Current paradigms emphasizing mitochondrial contributions to embryogenesis, aging, and PCD implicate mitochondrial ROS as a crucial intermediate in each case. Indeed, a small percentage (1—4%) of electron transfer normally goes towards production of the superoxide radical [53,60]. Although classically considered detrimental in any form, there is an emerging consensus that ROS in *physiologic* amounts are required to regulate intracellular signaling mechanisms [115,116].

ETC efficiency therefore determines an individual's basal ROS production rate. ETC efficiency, in turn, is likely influenced by the large number of polymorphism combinations generated by the over 80 ETC peptide-encoding genes of mtDNA and nDNA. Basal ROS production is potentially relevant to the rate at which mitochondrial oxidative damage accumulates in an individual over time. Specifically, over the course of physiologic aging mtDNA progressively acquires deletion and point mutations [57,58,62,64,65,67]. Precedents exist that show somatic mtDNA mutation influences ETC function [117—119].

Unifying hypothesis for AD histopathology and pathophysiology

We believe low rates of mitochondrial oxidative phosphorylation, increased reliance on anaerobic glycolysis, and high rates of mitochondrial ROS production ultimately account for, either directly or indirectly, the histopathologic and pathophysiologic

features of sporadic, late-onset AD. We therefore propose a unifying "mitochondrial cascade hypothesis" for this form of the disorder. In formulating the hypothesis, we considered recent data on cell cycle regulation, programmed cell death dynamics, ROS-mediated protein modification, and ROS-mediated DNA modification. Much of this data post-dates introduction of the mitochondrial theory of aging and amyloid cascade hypothesis. Accordingly, we attempted to update these two constructs within the context of an advancing body of knowledge. Whenever possible, we modify rather than discard aspects of both constructs, and synthesize the most relevant parts into a comprehensive whole. The mitochondrial cascade hypothesis for sporadic, late-onset AD maintains:

(1) Inherited polymorphic variations in the mtDNA and nDNA genes that encode ETC subunits determines ETC efficiency and basal mitochondrial ROS production;
(2) A correlation exists between basal mitochondrial ROS production rates and accumulating mtDNA damage, with higher basal ROS production rates causing more rapid accumulation of mtDNA damage;
(3) Somatic mtDNA mutation decreases mitochondrial ETC efficiency from its inherited set point, which manifests as reduced oxidative phosphorylation and/or increased mitochondrial ROS production. This triggers a three part compensatory response-
   (a) Reset the system: mitochondrial ROS overproduction in terminally differentiated neurons triggers Aβ production from APP, which further reduces ETC activity. Cell redox activities may eventually shift to the plasma membrane oxidoreductase system, where excess plasma membrane ROS would increase extracellular Aβ production and contribute to amyloid plaque formation.
   (b) Remove the most dysfunctional cells: apoptosis is activated in terminally differentiated neurons that continue to manifest or go on to manifest suprathreshold ROS and/or subthreshold oxidative phosphorylation.
   (c) Replace lost cells: impaired mitochondrial oxidative phosphorylation increases cell reliance on anaerobic glycolysis, initiating hypoxic signaling and also altering ROS homeostasis. In neurons with residual proliferative ability, this provides a signal for reentry into mitotic cycling. Cell cycle re-entry ultimately fails (perhaps due to bioenergetic considerations), but before or during G_2-M arrest there is cyclin protein upregulation, DNA synthesis/aneuploidy, tau phosphorylation, and tangle formation. These cells eventually lose viability not from their failure to complete the cell cycle per se, but rather from the underlying mitochondrial dysfunction that prompted cell cycle re-entry in the first place.

We intend the mitochondrial cascade hypothesis to apply only in cases of sporadic, late-onset AD. For the early onset, autosomal dominant cases that arise from mutation of APP, presenilin 1, or presenilin 2, we see no reason to challenge the ascendancy of the amyloid cascade hypothesis. At the same time, our hypothesis predicts mitochondria should occupy a relatively upstream position in the amyloid cascade hierarchy, rather than the peripheral, downstream location most reviews typically ascribe them [17,18]. This view is consistent with tissue culture data indicating ETC function is requisite for Aβ toxicity. The fact that Aβ does not harm cells artificially depleted of mtDNA (and as a consequence lack a functional ETC) potently argues direct Aβ-mitochondria interactions are highly relevant in autosomal dominant AD [94].

We propose bioenergetic dysfunction and mitochondrial ROS overproduction represents a nexus between the mitochondrial cascade hypothesis of sporadic AD and the amyloid cascade hypothesis (which we feel is most likely to apply in early onset, autosomal dominant cases). Further, the mitochondrial cascade hypothesis helps address some of the more poorly defined aspects of the amyloid cascade hypothesis, such as the mechanisms through which Aβ production might drive neurofibrillary tangle formation. Mitochondrial dysfunction also results in synaptic degradation [120], and our hypothesis provides a mechanism through which both sporadic, late-onset and autosomal dominant, early onset AD cases acquire synaptic pathology.

Two essential features distinguish the mitochondrial and amyloid cascade hypotheses. First, unlike what is the case with the early onset, autosomal dominant forms of AD, in sporadic, late-onset AD the defining histopathology and pathophysiology are initiated by mitochondrial dysfunction. Second, in sporadic, late-onset AD, increased Aβ production may represent a compensatory event occurring in response to the primary mitochondrial pathology, while in early onset, autosomal dominant AD Aβ production is strictly a toxic phenomenon.

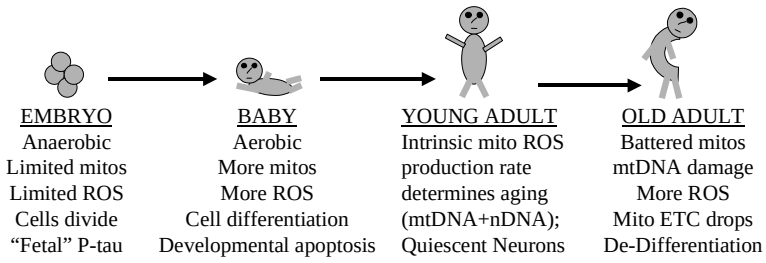

Figure 1 Mitochondrial function helps regulate cell proliferation, differentiation, and senescence.

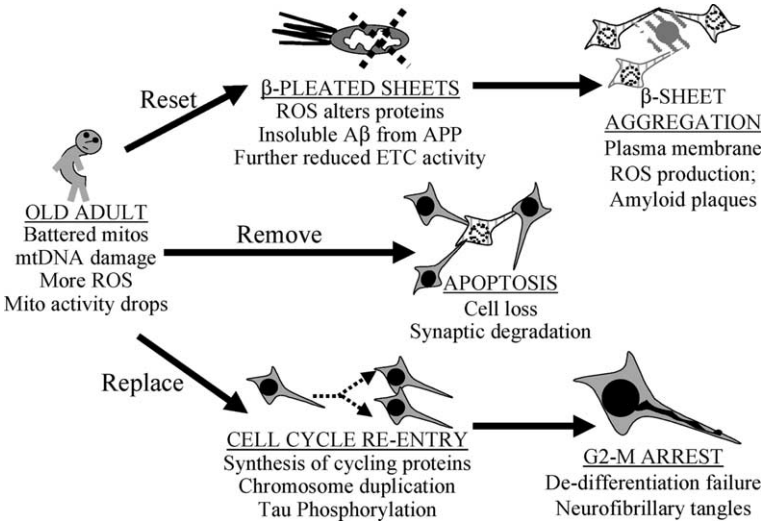

Figure 2 Mitochondrial dysfunction initiates compensatory events that result in the histopathologic sequelae of AD.

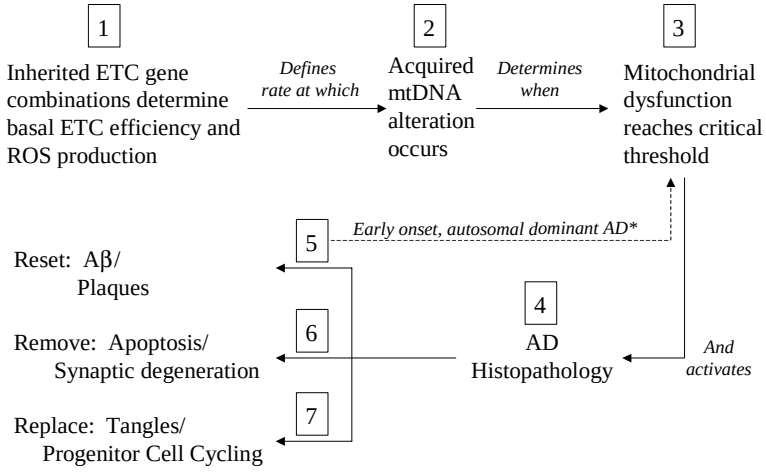

Figure 3 The mitochondrial cascade hypothesis for late-onset, sporadic AD. A key difference between late-onset, sporadic AD and early-onset, autosomal dominant AD is that in the early, autosomal dominant forms of the disorder Aβ formation is the primary pathologic event, and causes secondary mitochondrial dysfunction (indicated by the asterisk). In the mitochondrial cascade hypothesis, mitochondrial dysfunction ultimately causes the pathology indicated in steps 5–7. In the amyloid cascade hypothesis mitochondrial dysfunction leads to the pathology of steps 6 and 7.

Key points of the mitochondrial cascade hypothesis are emphasized in Figs. 1 and 2. Fig. 1 summarizes components that derive from cell cycling and differentiation theory and relates them to the aging process. Fig. 2 addresses how mitochondrial dysfunction gives rise to AD histopathology. Fig. 3 synthesizes both aspects into one construct, and indicates the proposed point of

overlap between the mitochondrial and amyloid cascade hypotheses.

Support for the hypothesis

Data supporting a role for mitochondria and, in particular, mtDNA in aging and age-related diseases are generally consistent with the first part of the AD mitochondrial cascade hypothesis (Fig. 1). Some of these data derive from basic epidemiologic studies of aging. Longevity analysis of the Framingham cohort, for instance, reveals that although the best predictor of an individual's longevity is biparental longevity, maternal longevity carries a greater impact [121]. Epidemiologic studies of both AD and PD further suggest for subjects that have a parent with the disease, among the affected parents there is maternal overrepresentation [122—124]. Taken together, these studies argue that a maternally inherited genetic factor (mtDNA) helps determine how long one lives, as well as contributes to AD risk.

Some studies suggest mtDNA haplogroups (which are defined by mtDNA polymorphism patterns) influence longevity. In northern Italy, haplogroup J is present to statistical excess in centenarians [112]. Certain mtDNA polymorphisms are more common in the extremely old than they are in the general population [113]. Mitochondrial DNA haplogroup variations may also affect an individual's odds of developing a neurodegenerative disease. One recent study of mtDNA polymorphisms in Parkinson's disease (PD) found that mtDNA haplogroups J and K (which share a common SNP 10398G polymorphism) are associated with a robust PD risk reduction [114].

Cytoplasmic hybrid ("cybrid") studies of persons with various sporadic neurodegenerative diseases also argue mtDNA inheritance at least partly determines mitochondrial ETC efficiency, oxidative-phosphorylation capacity, and ROS production. Cybrid cell lines are generated when mtDNA from a designated subject are expressed within cultured cells depleted of endogenous mtDNA [125,126]. This technique allows explorations of mitochondrial genotype—phenotype relationships while controlling for nDNA variability. If cybrid cell lines with a common nuclear background but mtDNA from different donors have distinct mitochondrial phenotypes, the root cause is likely differences between the donor mtDNAs [127].

Relative to cybrids expressing mtDNA from age-matched control individuals, cybrids expressing mtDNA from AD, PD, amyotrophic lateral sclerosis (ALS), and progressive supranuclear palsy subjects show bioenergetic impairment and increased oxidative stress [128—139]. These cybrid lines also exhibit altered calcium homeostasis, mitochondrial membrane potential depolarization, abnormal mitochondrial morphology, molecular stress response pathway activation, increased activation and expression of apoptotic proteins, and excessive protein aggregation (including Aβ in AD cybrids and α-synuclein in PD cybrids) [128,132,134, 136, 40—149]. Interestingly, mtDNA used to generate cybrid cell lines in these experiments is derived not from brain but rather from platelets, a non-degenerating tissue. This suggests the relative functional impairment observed in cell lines with disease subject mtDNA represents a systemic defect. Systemic mitochondrial dysfunction is more consistent with inherited rather than somatic mtDNA aberration.

To date, only limited published data indicate inherited mtDNA variation influences somatic mtDNA mutation acquisition [61]. There is, however, considerable evidence showing mtDNA does acquire mutations during the course of an individual's lifespan, including the brain, the tissue with the greatest rate of oxidative metabolism and therefore ROS production [65,67,150,151]. Over a decade ago it was shown that mtDNA deletions (specifically, a particular 5 kb deletion called the "common deletion") accumulate with age [152—156]. Tissues with lower rates of oxidative mutation than brain acquire less deletion burden. More recently, investigators have uncovered an entire layer of age-dependent mtDNA mutation in the brains of deceased individuals [67,157]. These presumed somatic mutations are not detected using routine sequencing strategies, but rather require laborious clonal mtDNA analysis. It is currently unclear whether these mutations represent low abundance "microheteroplasmy" that is widely distributed between many or most cells of a brain parenchyma region, or if it arises from limited numbers of individual cells that carry unique homoplasmic mutations. These scenarios, however, may not be mutually exclusive, since somatic mutations (that by definition are in low abundance when they arise) tend to clonally expand towards high abundance intracellular heteroplasmy or even homoplasmy [158—160]. Ultimately, through either clonal expansion or "compound microheteroplasmy", mtDNA mutational burdens within individual cells may reach thresholds at which the resultant mitochondrial dysfunction becomes critical.

There is some debate as to how aging effects mitochondrial ETC function. Data supporting an age-related decline are strongest for liver, and also indicate a similar phenomenon in muscle,

fibroblasts, and brain [161–170]. The status of brain mitochondria ETC function has also been extensively evaluated in subjects with neurodegenerative disorders. Relevant to this discussion is the well-replicated finding that complex IV (cytochrome oxidase) activity is reduced in AD brain [19]. Some argue this is an epiphenomenal consequence of neuronal de-afferentation. Indeed, it has been shown that surgically de-afferented neurons downregulate cytochrome oxidase [171,172]. The mechanistic explanation for this is that de-afferented neurons have reduced synaptic connections, less synaptic activity, and require less ATP to maintain ion gradients across their membranes. This cannot entirely explain the AD cytochrome oxidase deficit, which is also present in sporadic AD subject platelets and fibroblasts [173–178].

One AD chicken-and-egg controversy revolves around whether mitochondrial dysfunction causes Aβ over-production, or whether Aβ over-production causes mitochondrial dysfunction. When viewed outside the charged confines of the AD debate, it certainly seems clear that cytochrome oxidase inhibition promotes amyloidgenic fragmentation of APP, and that Aβ inhibits cytochrome oxidase [179–183]. For the sporadic late-onset forms, available data argue amyloidgenesis follows mitochondrial dysfunction. Specifically, in sporadic AD mitochondrial dysfunction is more anatomically widespread than is Aβ deposition, and therefore mitochondrial dysfunction cannot entirely be accounted for by Aβ [19]. Further, cybrid cell lines that express mtDNA from AD subjects, in addition to showing reduced cytochrome oxidase activity and increased ROS production relative to control cybrid lines, also produce substantially increased amounts of both intracellular and extracellular Aβ [19,146].

AD brain shows evidence of increased apoptotic cell death, cyclin protein expression, and nuclear DNA replication with excess aneuploidy [23–25]. The concept that tau phosphorylation promotes tangle formation in neurons attempting to re-enter the cell replication cycle is not novel [24], nor is the idea that mitochondrial function can determine tau phosphorylation. Indeed, tau phosphorylation is promoted in AD fibroblasts exposed to an ETC uncoupler [184].

Conclusion

The mitochondrial cascade hypothesis provides a unifying framework for AD pathology. In developing this hypothesis, we approached sporadic AD from an aging theory perspective, since sporadic AD incidence and prevalence progressively rise at least into the ninth decade [185]. Indeed, if half those over age 85 meet criteria for AD [186], can it truly be considered a disease?

Our hypothesis posits mitochondrial dysfunction represents primary pathology in sporadic, late-onset AD, and drives both Aβ plaque and neurofibrillary tangle formation. We further provide a rationale for how mitochondrial dysfunction surpassing certain thresholds triggers compensatory events that cause the various histopathologic and pathophysiologic features of AD. Other sporadic neurodegenerative diseases also manifest mitochondrial dysfunction, oxidative stress, and protein aggregation [127,187,188]. It is tempting to consider whether similar principles may underlie these disorders at the molecular level.

References

[1] Alzheimer A. Uber eine eigenartige Erkrankung der Hirnrinde. Allg Z Psychiat Psych- Gerichtl Med 1907;64:146–8.
[2] Kraepelin E. Psychiatrie. Ein Lehrbuch fur Studierende und Artze. II. Band, Klinische Psychiatrie. Leipzig: Verlag Johann Ambrosius Barth; 1910.
[3] Fischer O. Miliare Nekrosen mit drusigen Wucherungen der Neurofibrillen, eine regelmabige Veranderung der Hirnrinde bei seniler Demenz. Monatsschr Psychiatr Neurol 1907;22:361–72.
[4] Perusini G. Sul valore nosografico di alcuni reperti istopatlogici caratteristici per la senilita. Rivista italiana di Neuropatologia. Psichiatria e Elettroterapia 1911;4:145–51.
[5] Boller F, Forbes MM. History of dementia and dementia in history: an overview. J Neurol Sci 1998;158:125–33.
[6] Amaducci LA, Rocca WA, Schoenberg BS. Origin of the distinction between Alzheimer's disease and senile dementia: how history can clarify nosology. Neurology 1986;36:1497–9.
[7] Katzman R. The prevalence and malignancy of Alzheimer's disease: a major killer. Arch Neurol 1976;33:217–8.
[8] Katzman R, Terry R, Bick K. Recommendations of the nosology, epidemiology, and etiology and pathophysiology commissions of the workshop-conference on Alzheimer's disease: senile dementia and related disorders. In: Katzman R, Terry RD, Bick KL, editors. Alzheimer's disease: senile dementia and related disorders. New York: Raven Press; 1978. p. 579–85.
[9] Katzman R. Alzheimer's disease. New England J Med 1986;314:964–73.
[10] Amaducci L, Rocca W, Schoenberg B. Origin of the distinction between Alzheimer's disease and senile dementia. How history can clarify nosology. Neurology 1986;36:1497–9.
[11] Goate A, Chartier-Harlin MC, Mullan M, Brown J, Crawford F, Fidani L, et al. Segregation of a missense mutation in the amyloid precursor protein gene with familial Alzheimer's disease. Nature 1991;349:704–6.
[12] Sherrington R, Rogaev EI, Liang Y, Rogaeva EA, Levesque G, Ikeda M, et al. Cloning of a gene bearing missense mutations in early-onset familial Alzheimer's disease. Nature 1995;375:754–60.

[13] Levy-Lahad E, Wasco W, Poorkaj P, Romano DM, Oshima J, Pettingell WH, et al. Candidate gene for the chromosome 1 familial Alzheimer's disease locus. Science 1995;269: 973—7.
[14] Hardy J, Allsop D. Amyloid deposition as the central event in the aetiology of Alzheimer's disease. Trends Pharmacol Sci 1991;12:383—8.
[15] Hardy JA, Higgins GA. Alzheimer's disease: the amyloid cascade hypothesis. Science 1992;256:184—5.
[16] Selkoe DJ. Cell biology of the amyloid β-protein precursor and the mechanism of Alzheimer's disease. Annu Rev Cell Biol 1994;10:373—403.
[17] Selkoe DJ. Toward a comprehensive theory for Alzheimer's disease. Ann NY Acad Sci 2000;924:17—25.
[18] Cummings JL, Cole G. Alzheimer disease. JAMA 2002;287: 2335—8.
[19] Swerdlow RH, Kish SJ. Mitochondria in Alzheimer's disease. Int Rev Neurobiol 2002;53:341—85.
[20] Davis DG, Schmitt FA, Wekstein DR, Markesbery WR. Alzheimer neuropathologic alterations in aged cognitively normal subjects. J Neuropath Exp Neurol 1999;58: 376—88.
[21] Neuropathology Group of the Medical Research Council Cognitive Function and Ageing Study. Pathological correlates of late-onset dementia in a multicentre, community-based population in England and Wales. Lancet 2001;357: 169—75.
[22] Snowdon DA. Healthy aging and dementia: findings from the nun study. Ann Int Med 2003;139:450—4.
[23] Barinaga M. Is apoptosis key in Alzheimer's disease. Science 1998;281:1303—4.
[24] Arendt T. Alzheimer's disease as a loss of differentiation control in a subset of neurons that retain immature features in the adult brain. Neurobiol Aging 2000;21: 783—96.
[25] Yang Y, Geldmacher DS, Herrup K. DNA replication precedes neuronal cell death in Alzheimer's disease. J Neurosci 2001;21:2662—8.
[26] Anderton BH. Ageing of the brain. Mech Ageing and Dev 2002;123:811—7.
[27] Kwon YW, Masutani H, Nakamura H, Ishii Y, Yodoi J. Redox regulation of cell growth and cell death. Biol Chem 2003;384:991—6.
[28] Fulco M, Schiltz RL, Iezzi S, King MT, Zhao P, Kashiwaya Y, et al. Sir2 regulates skeletal muscle differentiation as a potential sensor of the redox state. Mol Cell 2003;12: 51—62.
[29] Chueh PJ. Cell membrane redox systems and transformation. Antioxid Redox Signal 2000;2:177—87.
[30] Klein JA, Ackerman SL. Oxidative stress, cell cycle, and neurodegeneration. J Clin Invest 2002;111:785—93.
[31] Boveris A, Oshino N, Chance B. The cellular production of hydrogen peroxide. Biochem J 1972;128:617—30.
[32] Chance B, Sies H, Boveris A. Hydroperoxide metabolism in mammalian organs. Physiol Rev 1979;59:527—605.
[33] Shigenaga MK, Hagen TM, Ames BN. Oxidative damage and mitochondrial decay in aging. Proc Natl Acad Sci USA 1994;91:10771—8.
[34] Burdon RH. Superoxide and hydrogen peroxide in relation to mammalian cell proliferation. Free Rad Biol Med 1995; 18:775—94.
[35] Johansson CB. Mechanism of stem cells in the central nervous system. J Cell Physiol 2003;196:409—18.
[36] Jauniaux E, Gulbis B, Burton GJ. The human first trimester gestational sac limits rather than facilitates oxygen transfer to the foetus — A review. Placenta 2003;24(suppl A): S86—93.
[37] Nasr-Esfahani MH, Aitken JR, Johnson MH. Hydrogen peroxide levels in mouse oocytes and early cleavage stage embryos developed in vitro or in vivo. Development 1990; 109:501—7.
[38] Johnson MH, Nasr-Esfahani MH. Radical solutions and cultural problems: could free oxygen radicals be responsible for the impaired development of preimplantation mammalian embryos in vitro. Bioessays 1994;16: 31—8.
[39] Trimarchi JR, Liu L, Porterfield DM, Smith PJS, Keefe DL. Oxidative phosphorylation-dependent and — independent oxygen consumption by individual preimplantation mouse embryos. Biol Reprod 2000;62:1866—74.
[40] Sweet S, Singh G. Accumulation of human promyelocytic leukemic (HL-60) cells at two energetic cell cycle checkpoints. Cancer Res 1995;55:5164—7.
[41] Dorward A, Sweet S, Moorehead R, Singh G. Mitochondrial contributions to cancer cell physiology: redox balance, cell cycle, and drug resistance. J Bioenerg Biomem 1997; 29:385—92.
[42] Lovestone S, Reynolds CH. The phosphorylation of tau: a critical stage in neurodevelopment and neurodegenerative processes. Neuroscience 1997;78:309—24.
[43] Connolly JA, Kalnins VI, Cleveland DW, Kirschner MW. Immunoflourescent staining of cytoplasmic and spindle microtubules in mouse fibroblasts with antibody to tau protien. Proc Natl Acad Sci USA 1977;74:2437—40.
[44] Delobel P, Plament S, Hamdane M, Mailliot C, Sambo AV, Begard S, et al. Abnormal tau phosphorylation of the Alzheimer-type also occurs during mitosis. J Neurochem 2002;83:412—20.
[45] Billingsley ML, Kincaid RL. Regulated phosphorylation and dephosphorylation of tau protein: effects on microtubule interaction, intracellular trafficking and neurodegeneration. Biochem J 1997;323:577—91.
[46] Hartigan JA, Johnson GVW. Tau protein in normal and Alzheimer's disease brain: an update. Alzheimer's Disease Rev 1998;3:125—41.
[47] Trounce I, Byrne E, Marzuki S. Decline in skeletal muscle mitochondrial respiratory chain function: possible factor in ageing. Lancet 1989;1:637—9.
[48] Yen TC, Chen YS, King KL, Yeh SH, Wie YH. Liver mitochondrial respiratory function declines with age. Biochem Biophys Res Commun 1989;165:994—1003.
[49] Cooper JM, Mann VM, Schapira AHV. Analysis of mitochondrial respiratory cahin function and mitochondrial DNA deletion in human skeletal muscle: effect of aging. J Neurol Sci 1992;113:91—8.
[50] Ojaimi J, Masters CL, Opwskin K, McKelvie P, Byrne E. Mitochondrial respiratory chain activity in the human brain as a function of age. Mech Ageing Dev 1999;111: 39—47.
[51] Harmon D. The biologic clock: the mitochondria. J Am Geriatr Soc 1972;20:145—7.
[52] Miquel J, Economos AD, Fleming J, Johnson JE. Mitochondrial role in cell aging. Exp Gerontol 1980;15:575—91.
[53] Beckman KB, Ames BN. Mitochondrial aging: open questions. Ann NY Acad Sci 1998;854:118—27.
[54] Melov S. Mitochondrial oxidative stress. Physiologic consequences and potential for a role in aging. Ann NY Acad Sci 2000;908:219—25.
[55] Lenaz G, Bovina C, D'Aurelio M, Fato R, Formiggini G, Genova ML, et al. Role of mitochondria in oxidative stress and aging. Ann NY Acad Sci 2002;959:199—213.
[56] Cadenas E, Davies KJA. Mitochondrial free radical generation, oxidative stress, and aging. Free Rad Biol Med 2000; 29:222—30.

[57] Linnane AW, Marzuki S, Ozawa T, Tanaka M. Mitochondrial DNA mutations as an important contributor to ageing and degenerative diseases. Lancet 1989;1:642—5.
[58] Wallace DC. Mitochondrial genetics: a paradigm for aging and degenerative diseases? Science 1992;256:628—32.
[59] Beckman KB, Ames BN. The free radical theory of aging matures. Physiol Rev 1998;78:548—81.
[60] Gadaleta MN, Cormio A, Pesce V, Lezza AM, Cantatore. Aging and mitochondria. Biochimie 1998;80:863—70.
[61] Ozawa T. Mechanism of somatic mitochondrial DNA mutations associated with age and diseases. Biochim Biophys Acta 1995;1271:177—89.
[62] Melov S, Shoffner JM, Kaufman A, Wallace DC. Marked increase in the number and variety of mitochondrial DNA rearrangements in aging human skeletal muscle. Nucl Acids Res 1995;23:4122—6.
[63] Kovalenko SA, Kopsidas G, Kelso JM, Linnane AW. Deltoid human muscle mtDNA is extensively rearranged in old age subjects. Biochem Biophys Res Commun 1997;232:147—52.
[64] Melov S, Schneider JA, Coskun PE, Bennett DA, Wallace DC. Mitochondrial DNA rearrangements in aging human brain in situ PCR of mtDNA. Neurobiol Aging 1999;20: 565—71.
[65] Michikawa Y, Mazzucchelli F, Bresolin N, Scarlato G, Attardi G. Aging-dependent large accumulation of point mutations in the human mtDNA control region for replication. Science 1999;286:774—9.
[66] Wang Y, Michikawa Y, Mallidis C, Bai Y, Woodhouse KE, Yarasheski KE, et al. Muscle-specific mutations accumulate with aging in critical human mtDNA control sites for replication. Proc Natl Acad Sci USA 2001;98:4022—7.
[67] Lin MT, Simon DK, Ahn CH, Kim LM, Beal MF. High aggregate burden of somatic mtDNA point mutations in aging and Alzheimer's disease brain. Hum Mol Genet 2002;11:133—45.
[68] Richter C, Park JW, Ames BN. Normal oxidative damage to mitochondrial and nuclear DNA is extensive. Proc Natl Acad Sci USA 1988;85:6465—7.
[69] Cheng KC, Cahill DS, Kasai H, Nishimura S, Loeb LA. 8-Hydroxyguanine, an abundant form of oxidative DNA damage, causes G—T and A—C substitutions. J Biol Chem 1992;267:166—72.
[70] Brierley EJ, Johnson MA, James OF, Turnbull DM. Mitochondrial involvement in the ageing process. Facts and controversies. Mol Cell Biochem 1997;174:325—8.
[71] Yoneda M, Chomyn A, Martinuzzi A, Hurko O, Attardi G. Marked replicative advantage of human mtDNA carrying a point mutation that causes the MELAS encephalomyopathy. Proc Natl Acad Sci USA 1992;89:11164—8.
[72] Taylor DR, Zeyl C, Cooke E. Conflicting levels of selection in the accumulation of mitochondrial defects in *Saccharomyces cerevisiae*. Proc Natl Acad Sci USA 2002;99: 3690—4.
[73] De Grey ADNJ. A mechanism proposed to explain the rise in oxidative stress during aging. J Anti Aging Med 1998;1:53—66.
[74] Kowald A. The mitochondrial theory of aging: do damaged mitochondria accumulated by delayed degradation? Exp Gerontol 1999;34:605—12.
[75] Gershon D. The mitochondrial theory of aging: is the culprit a faulty disposal system rather than indigenous mitochondrial alterations. Exp Gerontol 1999;34:613—9.
[76] Soohal RS, Agarwal A, Agarwal S, Orr WC. Simultaneous overexpression of copper- and zinc-containing superoxide dismutase and catalase retards age-related oxidative damage and increases metabolic potential in *Drosophila melanogaster*. J Biol Chem 1995;270:15671—4.
[77] Roth GS, Ingram DK, Lane MA. Slowing ageing by caloric restriction. Nat Med 1995;1:414—5.
[78] Aspnes LE, Lee CM, Weindruch R, Chung SS, Roecker EB, Aiken JM. Caloric restriction reduces fiber loss and mitochondrial abnormalities in aged rat muscle. FASEB J 1997;11:573—81.
[79] Zamzami N, Hirsch T, Dallaporta B, Petit PX, Kroemer G. Mitochondrial implication in accidental and programmed cell death: apoptosis and necrosis. J Bioenerg Biomem 1997;29:185—93.
[80] Ichas F, Mazat JP. From calcium signaling to cell death: two conformations for the mitochondrial permeability transition pore. Switching from low- to high-conductance state. Biochim Biophys Acta 1998;1366:33—50.
[81] Zou H, Li Y, Liu X, Wang X. An APAF-1 cytochrome c multimeric complex is a functional apoptosome that activates procaspase-9. J Biol Chem 1999;27:11549—56.
[82] Liu X, Naekyung Kim C, Yang J, Jemmerson R, Wang X. Induction of apoptotic program in cell-free extracts: requirement for dATP and cytochrome c. Cell 1996;86: 147—57.
[83] Kluck RM, Bossy-Wetzel E, Green DR, Newmeyer DD. The release of cytochrome c from mitochondria: a primary site for Bcl-2 regulation of apoptosis. Science 1997;275: 1132—6.
[84] Petit PX, Zamzami N, Vayssiere JL, Mignotte B, Kroemer G, Castedo M. Implication of mitochondria in apoptosis. Mol Cell Biochem 1997;174:185—8.
[85] Yang J, Liu X, Bhalla K, Kim CN, Ibrado AM, Cai J, et al. Prevention of apoptosis by Bcl-2: release of cytochrome c from mitochondria blocked. Science 1997;275:1129—32.
[86] Li P, Nijhawan D, Budihardjo I, Srinivasula SM, Ahmad M, Alnemri ES, et al. Cytochrome c and dATP-dependent formation of Apaf-1/caspase-9 complex initiates an apoptotic protease cascade. Cell 1997;91:479—89.
[87] Cardoso SM, Swerdlow RH, Oliveira CR. Induction of cytochrome c-mediated apoptosis by amyloid β25—35 requires functional mitochondria. Brain Res 2002;931: 117—25.
[88] Negre-Salvayre A, Hirtz C, Carrera G, Cazenave R, Troly M, Salvayre R, et al. A role for uncoupling proteins-2 as a regulator of mitochondrial hydrogen peroxide generation. FASEB J 1997;11:809—15.
[89] Ledesma A, de Lacoba MG, Rial E. The mitochondrial uncoupling proteins. Genome Biol 2003;3:3015, Reviews.
[90] Anandtheerthavarada HK, Biswas G, Robin MA, Avadhani MG. Mitochondrial targeting and a novel transmembrane arrest of Alzheimer's precursor protein impairs mitochondrial function in neuronal cells. J Cell Biol 2003;16:41—54.
[91] Dyall SD, Lester DC, Schneider RE, Delgadillo_correa MG, Plumper E, Martinez A, Koehler CM, Johnson PJ. *Trichomonas vaginalis* Hmp35, a putative pore-forming hydrogenosomal membrane protein, can form a complex in yeast mitochondria. J Biol Chem 2003;278:30548—61.
[92] Robinson SR, Bishop GM. Abeta as a bioflocculant: implications for the amyloid hypothesis of Alzheimer's disease. Neurobiol Aging 2002;23:1051—72.
[93] Atwood CS, Moir RD, Huang X, Scarpa RC, Bacarra NM, Romano DM, et al. Dramatic aggregation of Alzheimer abeta by Cu (II) is induced by conditions representing physiological acidosis. J Biol Chem 1998;273:12817—26.
[94] Cardoso SM, Santos S, Swerdlow RH, Oliveira CR. Functional mitochondria are required for amyloid beta-mediated neurotoxicity. FASEB J 2001;15:1439—41.
[95] Arias C, Montiel T, Quiroz-Baez R, Massieu L. β-Amyloid neurotoxicity is exacerbated during glycolysis inhibition and mitochondrial impairment in the rat hippocampus

in vivo and in isolated nerve terminals: implications for Alzheimer's disease. Exp Neurol 2002;176:163–74.
[96] Nunomura A, Perry G, Pappolla MA, Wade R, Hirai K, Chiba S, et al. RNA oxidation is a prominent feature of vulnerable neurons in Alzheimer's disease. J Neurosci 1999;19: 1959–64.
[97] Smith MA, Nunomura A, Zhu X, Takeda A, Perry G. Metabolic, metallic, and mitotic sources of oxidative stress in Alzheimer disease. Antioxide Redox Signal 2000; 2:413–20.
[98] Shan X, Tashiro H, Lin CL. The identification and characterization of oxidized RNAs in Alzheimer's disease. J Neurosci 2003;23:4913–21.
[99] Sylvestre J, Vialette S, Corral Debrinski M, Jacq C. Long mRNAs coding for yeast mitochondrial proteins of prokaryotic origin preferentially localize to the vicinity of mitochondria. Genome Biol 2003;4:R44, Epub 2003 Jun 06.
[100] Marc P, Margeot A, Devaux F, Blugeon C, Corral-Debrinski M, Jacq C. Genome-wide analysis of mRNAs targeted to yeast mitochondria. EMBO Rep 2002;3:159–64, Epub 2002 Jan 29.
[101] Ginsberg MD, Feliciello A, Jones JK, Avvedimento EV, Gottesman ME. PKA-dependent binding of mRNA to the mitochondrial AKAP121 protein. J Mol Biol 2003;327: 885–97.
[102] Lawen A, Martinus RD, McMullen GL, Nagley P, Vaillant F, Wolvetang EJ, et al. The universality of bioenergetic disease: the role of mitochondrial mutation and the putative inter-relationship between mitochondria and plasma membrane NADH oxidoreductase. Molec Aspects Med 1994;15(suppl):s13–27.
[103] Larm JA, Vaillant F, Linnane AW, Lawen A. Upregulation of the plasma membrane oxidoreductase as a prerequisite for the viability of human Namalwa rho^0 cells. J Biol Chem 1994;269:30097–100.
[104] Morre DM, Lenaz G, Morre J. Surface oxidase and oxidative stress propagation in aging. J Exp Biol 2000;203:1513–21.
[105] Capuano F, Guerrieri F, Papa S. Oxidative phosphorylation enzymes in normal and neoplastic cell growth. J Bioenerg Biomembr 1997;29:379–84.
[106] Chueh PJ. Cell membrane redox systems and transformation. Antioxid Redox Signal 2000;2:177–87.
[107] Capuano F, Varone D, D'Eri N, Russo E, Tommasi S, Montemurro S, et al. Oxidative phosphorylation and F (O)F (1) ATP synthase activity of human hepatocellular carcinoma. Biochem Mol Biol Int 1996;38:1013–22.
[108] Uriel J. Retrodifferentiation and the fetal patterns of gene expression in cancer. Adv Cancer Res 1979;29:127–74.
[109] MITOMAP: A Human Mitochondrial Genome Database. Center for Molecular Medicine, Emory University, Atlanta, GA, USA. Available from: http://www.gen.emory.edu/mitomap.html, 2001.
[110] National Center for Biotechnology Information, National Library of Medicine, National Institutes of Health. Available from: http://www.ncbi.nlm.nih.gov/entrez/, 2002.
[111] Roubertoux PL, Sluyter F, Carlier M, Marcet B, Maarouf-Veray F, Cherif C, et al. Mitochondrial DNA modifies cognition in interaction with the nuclear genome and age in mice. Nat Genet 2003;35:65–9.
[112] De Benedictis G, Rose G, Carrieri G, De Luca M, Falcone E, Passarino G, et al. Mitochondrial DNA inherited variants are associated with successful aging and longevity in humans. FASEB J 1999;13:1532–6.
[113] Tanaka M, Gong JS, Zhang J, Yoneda M, Yagi K. Mitochondrial gentoype associated with longevity. Lancet 1998;351: 185–6.
[114] van der Walt JM, Nicodemus KK, Martin ER, Scott WK, Nance MA, Watts RL, et al. Mitochondrial polymorphisms significantly reduce the risk of Parkinson disease. Am J Hum Genet 2003;72:804–11.
[115] Wolin MS. Reactive oxygen species and vascular signal transduction mechanisms. Microcirculation 1996;3:1–17.
[116] Droge W. Free radicals in the physiologic control of cell function. Physiol Rev 2001;82:47–95.
[117] Wanagat J, Cao Z, Pathare P, Aiken JM. Mitochondrial DNA deletion mutations colocalize with segmental electron transport system abnormalities, muscle fiber atrophy, fiber splitting, and oxidative damage in sarcopenia. FASEB J 2001;15:322–32.
[118] Fayet G, Jansson M, Sternberg D, Moslemi AR, Blondy P, Lombes A, et al. Aging muscle: clonal expansions of mitochondrial DNA point mutations and deletions cause focal impairment of mitochondrial function. Neuromuscul Disord 2002;12:483–93.
[119] Chomyn A, Attardi G. MtDNA mutations in aging and apoptosis. Biochem Biophys Res Commun 2003;304:519–29.
[120] Mattson MP, Liu D. Energetics and oxidative stress in synaptic plasticity and neurodegenerative disorders. Neuromol Med 2002;2:215–31.
[121] Brand FN, Kiely DK, Kannel WB, Myers RH. Family patterns of coronary heart disease mortality: the Framingham longevity study. J Clin Epidemiol 1992;45:169–74.
[122] Duara R, Lopez-Alberola RF, Barker WW, Loewenstein DA, Zatinsky M, Eisdorfer CE, et al. A comparison of familial and sporadic Alzheimer's disease. Neurology 1993;43: 1377–84.
[123] Edland SD, Silverman JM, Peskind ER, Tsuang D, Wijsman E, Morris JC. Increased risk of dementia in mothers of Alzheimer's disease cases: evidence for maternal inheritance. Neurology 1996;47:254–6.
[124] Swerdlow RH, Parker WD, Currie LJ, Bennett Jr JP, Harrison MB, Trugman JM, et al. Gender ratio differences between Parkinson's disease patients and their affected parents. Parkinsonism Related Disorders 2001;7:47–51.
[125] King MP, Attardi G. Human cells lacking mtDNA: repopulation with exogenous mitochondria complementation. Science 1989;246:500–3.
[126] Chomyn A, Lai ST, Shakeley R, Bresolin N, Scarlato G, Attardi G. Platelet-mediated transformation of mtDNA-less human cells: analysis of phenotypic variability among clones from normal individuals — and complementation behavior of the tRNALys mutation causing myoclonic epilepsy and ragged red fibers. Am J Hum Genet 1994; 54:966–74.
[127] Swerdlow RH. Mitochondrial DNA and dysfunction in neurodegenerative diseases. Arch Path Lab Med 2002;126:271–80.
[128] Swerdlow RH, Parks JK, Miller SW, Tuttle JB, Trimmer PA, Sheehan JP, et al. Origin and functional consequences of the complex I defect in Parkinson's disease. Ann Neurol 1996;40:663–71.
[129] Gu M, Cooper JM, Taanman JW, Schapira AHV. Mitochondrial DNA transmission of the mitochondrial defect in Parkinson's disease. Ann Neurol 1998;44:177–86.
[130] Shults CW, Miller SW. Reduced complex I activity in Parkinsonian cybrids. Mov Dis 1998;13:217.
[131] Davis RE, Miller S, Herrnstadt C, Ghosh SS, Fahy E, Shinobu LA, et al. Mutations in mitochondrial cytochrome c oxidase genes segregate with late-onset Alzheimer disease. Proc Natl Acad Sci USA 1997;94:4526–31.
[132] Swerdlow RH, Parks JK, Cassarino DS, Maguire DJ, Maguire RS, Bennett JP, et al. Cybrids in Alzheimer's disease: a cellular model of the disease. Neurology 1997;49:918–25.
[133] Cassarino DS, Fall CP, Swerdlow RH, Smith TS, Halvorsen EM, Miller SW, et al. Elevated reactive oxygen species and

antioxidant enzyme activities in animal and cellular models of Parkinson's disease. Biochim Biophys Acta 1997;1362:77–86.
[134] Swerdlow RH, Parks JK, Davis JN, Cassarino DS, Trimmer PA, Currie LJ, et al. Matrilineal inheritance of complex I dysfunction in a multigenerational Parkinson's disease family. Ann Neurol 1998;44:873–81.
[135] Ghosh SS, Swerdlow RH, Miller SW, Sheeman B, Parker Jr WD, Davis RE. Use of cytoplasmic hybrid lines for elucidating the role of mitochondrial dysfunction in Alzheimer's disease and Parkinson's disease. Ann NY Acad Sci 2000;893:176–91.
[136] Swerdlow RH, Miller SW, Parks JK, Sheehan JP, Cassarino DS, Maguire DJ, et al. Mitochondria in sporadic amyotrophic lateral sclerosis. Exp Neurol 1998;153:135–42.
[137] Swerdlow RH, Golbe LI, Parks JK, Cassarino DS, Binder DR, Grawey AE, et al. Mitochondrial dysfunction in cybrid lines expressing mitochondrial genes from patients with PSP. J Neurochem 2000;75:1681–4.
[138] Trimmer PA, Keeney PM, Borland MK, Simon FA, Almeida J, Swerdlow RH, et al. Mitochondrial abnormalites in cybrid cell models of sporadic Alzheimer's disease worsen with passage in culture. Neurobiol Dis 2004;15:29–39.
[139] Cardoso SM, Santana I, Swerdlow RH, Oliveira CR. Mitochondria dysfunction of Alzheimer's disease cybrids enchances Aβ toxicity. J Neurochem, in press.
[140] Sheehan JP, Swerdlow RH, Parker WD, Miller SW, Davis RE, Tuttle JB. Altered calcium homeostasis in cells transformed by mitochondria from individuals with Parkinson's disease. J Neurochem 1997;68:1221–33.
[141] Sheehan JP, Swerdlow RH, Miller SW, Davis RE, Tuttle JB. Altered calcium homeostasis and reactive oxygen species production in cells transformed by Alzheimer's disease mitochondrial DNA. J Neurosci 1997;17:4612–22.
[142] Cassarino DS, Halvorsen EM, Swerdlow RH, Abramova NN, Parker WD, Sturgill TW, et al. Interaction among mitochondria, mitogen-activated protein kinases, and nuclear factor-kappa B in cellular models of Parkinson's disease. J Neurochem 2000;74:1384–92.
[143] Trimmer PA, Swerdlow RH, Parks JK, Miller SW, Davis RE, Parker Jr WD. Abnormal mitochondrial morphology in sporadic Parkinson's and Alzheimer's disease cybrid lines. Exp Neurol 2000;162:37–50.
[144] Veech GA, Dennis J, Keeney PM, Fall CP, Swerdlow RH, Parker Jr WD. Disrupted mitochondrial electron transport function increases expression of anti-apoptotic Bcl-2 and Bcl-X_L proteins in SH-SY5Y neuroblastoma and in Parkinson's disease cybrid cell lines. J Neurosci Res 2000;61:693–700.
[145] Cassarino DS, Swerdlow RH, Parks JK, Davis Jr WP, Bennett Jr JP. Cyclosporin A increases resting mitochondrial membrane potential in SY5Y cells and reverses the depressed mitochondrial membrane potential of Alzheimer's disease cybrids. Biochem Biophys Res Commun 1998;248:168–73.
[146] Khan SM, Cassarino DS, Abramova NN, Keeney PM, Borland K, Trimmer PA, et al. Alzheimer's disease cybrids replicate β-amyloid abnormalities through cell death pathways. Ann Neurol 2000;48:148–55.
[147] Bijur GN, Davis RE, Jope RS. Rapid activation of heat shock factor-1 DNA binding by H_2O_2 and modulation by glutathione in human neuroblastoma and Alzheimer's disease cybrid cells. Brain Res Mol Brain Res 1999;71:69–77.
[148] De Sarno P, Bijur GN, Lu R, Davis RE, Jope RS. Alterations in muscarinic receptor-coupled phosphoinositide hydrolysis and AP-1 activation in Alzheimer's disease cybrid cells. Neurobiol Aging 2000;21:31–8.
[149] Trimmer PA, Borland K, Keeney PM, Bennett Jr JP, Parker Jr WD. Parkinson's disease transgenic mitochondrial cybrids generate Lewy inclusion bodies. J Neurochem 2004;88:800–12.
[150] Laderman KA, Penny JR, Mazzucchelli F, Bresolin N, Scarlato G, Attardi G. Aging-dependent functional alterations of mitochondrial DNA (mtDNA) from human fibroblasts transferred into mtDNA-less cells. J Biol Chem 1996;271:15891–7.
[151] Diaz F, Bayona-Bafaluy MP, Rana M, Mora M, Hao H, Moraes CT. Human mitochondrial DNA with large deletions repopulates organelles faster than full-length genomes under relaxed copy number control. Nucl Acids Res 2002;30:4626–33.
[152] Cortopassi GA, Arnheim N. Detection of a specific mitochondrial DNA deletion in tissues of older humans. Nucl Acids Res 1990;18:6927–33.
[153] Cortopassi GA, Shibata D, Soong NW, Arnheim N. A pattern of accumulation of a somatic deletion of mitochondrial DNA in aging human tissues. Proc Natl Acad Sci USA 1992;89:7370–4.
[154] Soong NW, Hinton DR, Cortopassi G, Arnheim N. Mosaicism for a specific somatic mitochondrial DNA mutations in adult human brain. Nat Genet 1992;2:318–23.
[155] Corral-Debrinski M, Horton MT, Lott JM, Shoffner JM, Beal MF, Wallace DC. Mitochondrial DNA deletions in human brain: regional variability and increase with advanced age. Nat Genet 1992;2:324–9.
[156] Corral-Debrinski M, Horton T, Lott MT, Shoffner JM, McKee AC, Beal MF, et al. Marked changes in mitochondrial DNA deletion levels in Alzheimer brains. Genomics 1994;23:471–6.
[157] Smigrodzki R, Parks J, Parker WD. High frequency of mitochondrial complex I mutations in Parkinson's disease and aging. Neurobiol Aging, in press.
[158] Elson JL, Samuels DC, Turnbull DM, Chnnery PF. Random intracellular drift explains the clonal expansion of mitochondrial DNA mutations with age. Am J Hum Genet 2001;68:802–6.
[159] Nekhaeva E, Bodyak ND, Kraytsberg Y, McGrath SB, Van Orsouw NJ, Pluzhnikov A, et al. Clonally expanded mtDNA point mutations are abundant in individual cells in human tissues. Proc Natl Acad Sci USA 2002;99:5521–6.
[160] Coller HA, Bodyak ND, Khrapko K. Frequent intracellular clonal expansions of somatic mtDNA mutations. Ann NY Acad Sci 2002;959:434–47.
[161] Muller-Hocker J. Cytochrome-c-oxidase deficient cardiomyocytes in the human heart – an age-related phenomenon. A histochemical ultracytochemical study. Am J Pathol 1989;134:1167–73.
[162] Muller-Hocker J, Schneiderbanger K, Stefani FH, Kadenbach B. Progressive loss of cytochrome c oxidase in the human extraocular muscles in ageing – a cytochemical–immunohistochemical study. Mutat Res 1992;275:115–24.
[163] Trounce I, Byrne E, Marzuki S. Decline in skeletal muscle mitochondrial respiratory chain function: possible factor in ageing. Lancet 1989;1:637–9.
[164] Yen TC, Chen YS, King KL, Yeh SH, Wei YH. Liver mitochondrial respiratory functions decline with age. Biochem Biophys Res Commun 1989;165:944–1003.
[165] Bowling AC, Mutisya EM, Walker LC, Price DL, Cork LC, Beal MF. Age-dependent impairment of mitochondrial function in primate brain. J Neurochem 1993;60:1964–7.
[166] Greco M, Villani G, Mazzucchelli F, Bresolin N, Papa S, Attardi G. Marked aging-related decline in efficiency of oxidative phosphorylation in human skin fibroblasts. FASEB J 2003;17:1706–8.

[167] Cooper JM, Mann VM, Schapira AHV. Analysis of mitochondrial respiratory cahin function and mitochondrial DNA deletion in human skeletal muscle: effect of aging. J Neurol Sci 1992;113:91—8.
[168] Ojaimi J, Masters CL, Opwskin K, McKelvie P, Byrne E. Mitochondrial respiratory chain activity in the human brain as a function of age. Mech Ageing Dev 1999;111:39—47.
[169] Papa S. Mitochondrial oxidative phosphorylation changes in the life span. Molecular aspects and physiopathological implications. Biochim Biophys Acta 1996;1276:87—105.
[170] Chomyn A, Attardi G. MtDNA mutations in aging and apoptosis. Biochem Biophys Res Commun 2003;304:519—29.
[171] Borowsky IW, Collins RC. Histochemical changes in enzymes of energy metabolism in the dentate gyrus accompany deafferentation and synaptic reorganization. Neuroscience 1989;33:253—62.
[172] Wong-Riley MT. Cytochrome oxidase: an endogenous metabolic marker for neuronal activity. Trends Neurosci 1989;12:94—101.
[173] Parker Jr WD, Filley CM, Parks JK. Cytochrome oxidase deficiency in Alzheimer's disease. Neurology 1990;40: 1302—3.
[174] Parker Jr WD, Mahr NJ, Filley CM, Parks JK, Hughes D, Young DA, et al. Reduced platelet cytochrome c oxidase activity in Alzheimer's disease. Neurology 1995;44: 1086—90.
[175] Bosetti F, Brizzi F, Barogi S, Mancuso M, Siciliano G, Tendi EA, et al. Cytochrome c oxidase and mitochondrial F1F0 = ATPase (ATP synthase) activities in platelets and brain from patients with Alzheimer's disease. Neurobiol Aging 2002;23:371—6.
[176] Cardoso SM, Proenca MT, Santos S, Santana I, Oliveira CR. A possible mechanism for the decrease of cytochrome oxidase in Alzheimer's disease. Soc Neurosci Abstr 1999; 25:337.
[177] Mancuso M, Filosto M, Bosetti F, Ceravolo R, Rocchi A, Tognoni G, et al. Decreased platelet cytochrome c oxidase activity is accompanied by increased blood lactate concentration during exercise in patients with Alzheimer disease. Exp Neurol 2003;182:421—6.
[178] Curti D, Rognoni F, Gasparini L, Cattaneo A, Paolllo M, Racchi M, et al. Oxidative metabolism in cultured fibroblasts derived from sporadic Alzheimer's disease (AD) patients. Neurosci Lett 1997;236:13—6.
[179] Gabuzda D, Busciglio J, Chen LB, Matsudaira P, Yankner BA. Inhibition of energy metabolism alters the processing of amyloid precursor protein and induces a potentially amyloidogenic derivative. J Biol Chem 1994;269:13623—8.
[180] Webster MT, Pearce BR, Bowen DM, Francis PT. The effecs of perturbed energy metabolism on the processing of amyloid precursor protein in PC12 cells. J Neural Transm 1998;105:839—53.
[181] Mark RJ, Keller JN, Kruman I, Mattson MP. Basic FGF attenuates amyloid beta peptide-induced oxidative stress, mitochondrial dysfunction, and impairment of Na^+/K^+-ATPase activity in hippocampal neurons. Brain Res 1997; 756:205—14.
[182] Pereira C, Santos MS, Oliveira C. Mitochondrial function impairment induced by amyloid beta-peptide on PC12 cells. Neuroreport 1998;9:1749—55.
[183] Canevari L, Clark JB, Bates TE. β-Amyloid fragment 25—35 selectively decreases complex IV activity in isolated mitochondria. FEBS Lett 1999;457:131—4.
[184] Blass JP, Baker AC, Ko L, Black RS. Induction of Alzheimer antigens by an uncoupler of oxidative phosphorylation. Arch Neurol 1990;47:864—9.
[185] Zabar Y, Kawas CH. Epidemiology and clinical genetics of Alzheimer's disease. In: Clark CM, Trojanowski JQ, editors. Neurodegenerative dementias: clinical features and pathological mechanisms. New York: McGraw-Hill; 2000. p. 79—94.
[186] Evans DA, Funkenstein HH, Albert MS, Scherr PA, Cook NR, Chown MJ, et al. Prevalence of Alzheimer's disease in a community population of older persons. Higher than previously reported. JAMA 1989;262:2551—6.
[187] Swerdlow RH. Role of mitochondria in Parkinson's disease. In: Chesselet MF, editor. Molecular mechanisms of neurodegenerative diseases. New Jersey: Humana Press Inc; 2000. p. 233—70.
[188] Swerdlow RH, Parks JK, Pattee G, Parker Jr WD. Role of mitochondria in amyotrophic lateral sclerosis. Amyotroph Lateral Sclerosis Motor Neuron Disorders 2000; 1:185—90.