Oxidative Stress in Alzheimer Disease

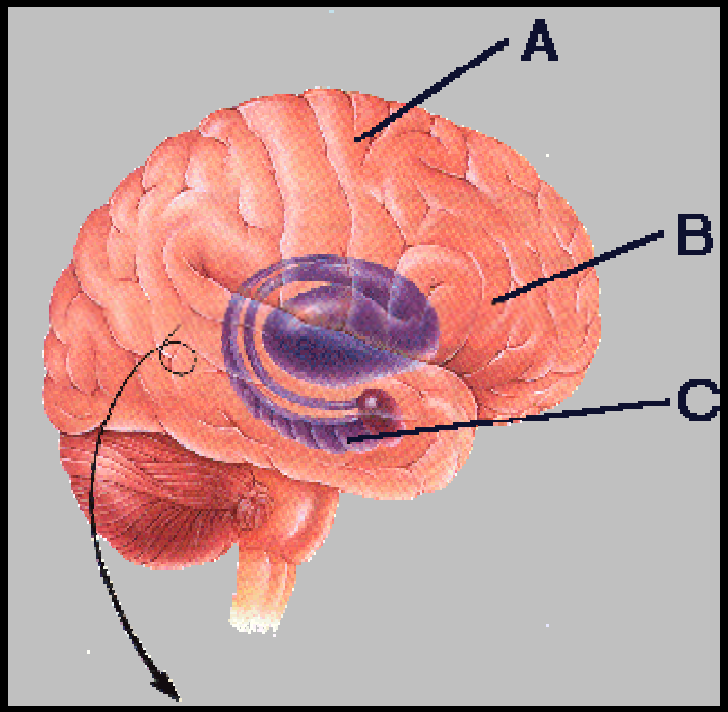
George Perry, Ph.D.

The University of Texas at San Antonio

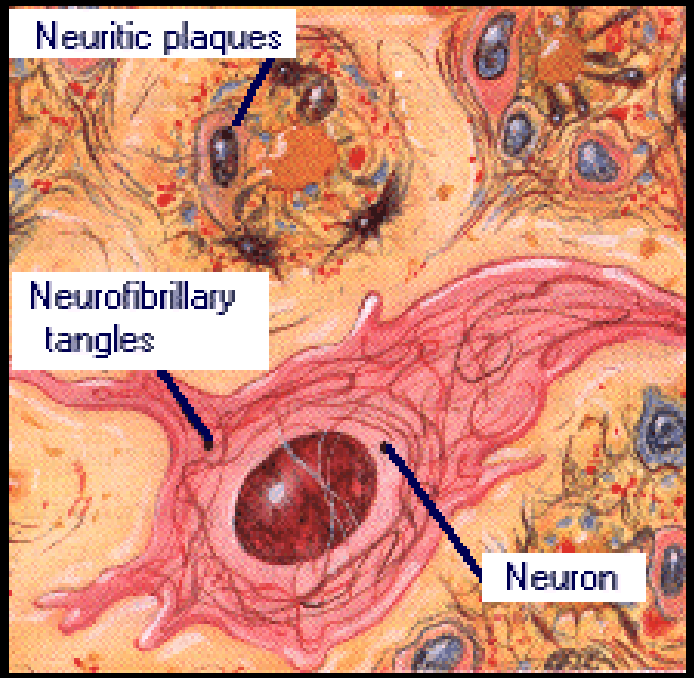
Texas State Plan on Alzheimer's Disease

Public Input Meeting and Scientific Symposium

Austin, Texas 23 April 2010



Alzheimer disease attacks nerve cells in several regions of the brain.



A. Cerebral Cortex: Involved in conscious thought and language.

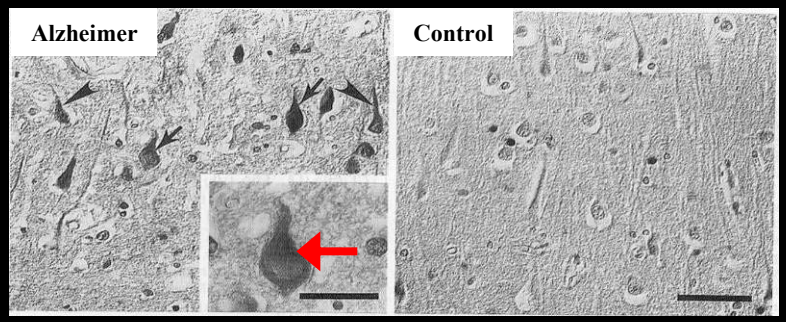
B. Basal forebrain: Has large numbers of neurons containing acetylcholine, a chemical important in memory and learning.

C. Hippocampus: Essential to memory storage. The earliest signs of Alzheimer's are found in the nearby entorhinal cortex.

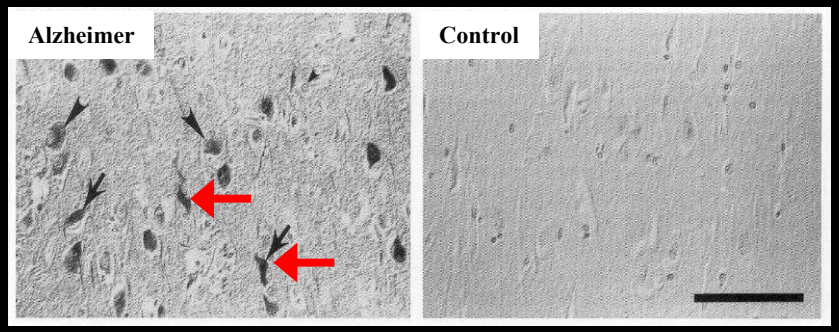
Definition of Oxidative Stress

1. Classic definition: The production of reactive oxygen in excess of antioxidant mechanisms
2. Evolving definition: Altered homeostatic balance resulting from oxidant insult.

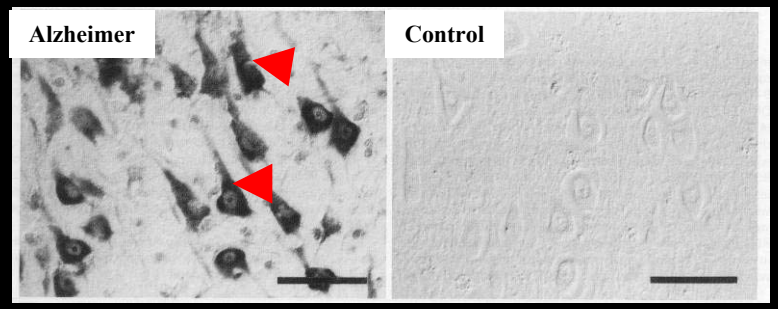
Oxidative damage increases in Alzheimer disease



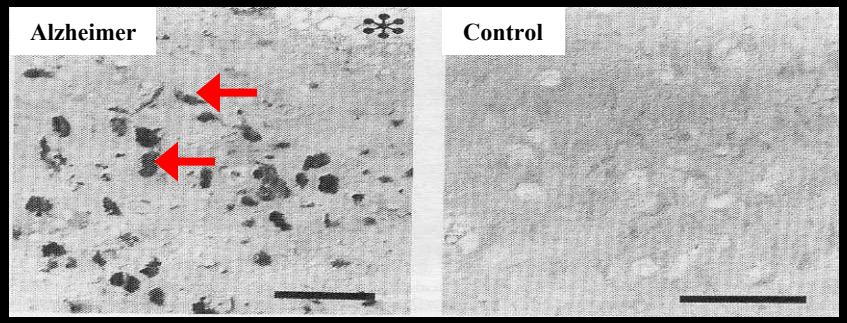
Lipid Peroxidation/Protein Adduction (4-HNE)



Protein Oxidation (Free Carbonyl Groups)



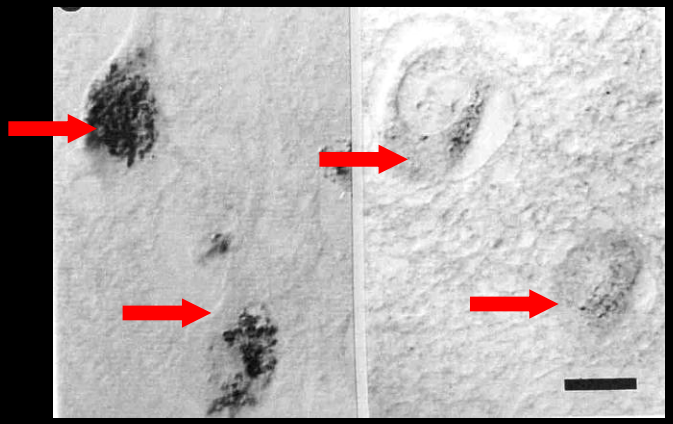
Nucleic Acids (8-OH-Guanosine)



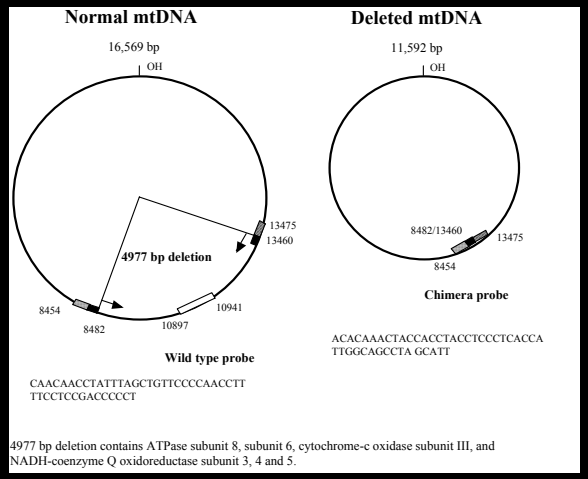
Glycoxidation (Carboxymethyllysine)

Mitochondrial components are increased in Alzheimer disease

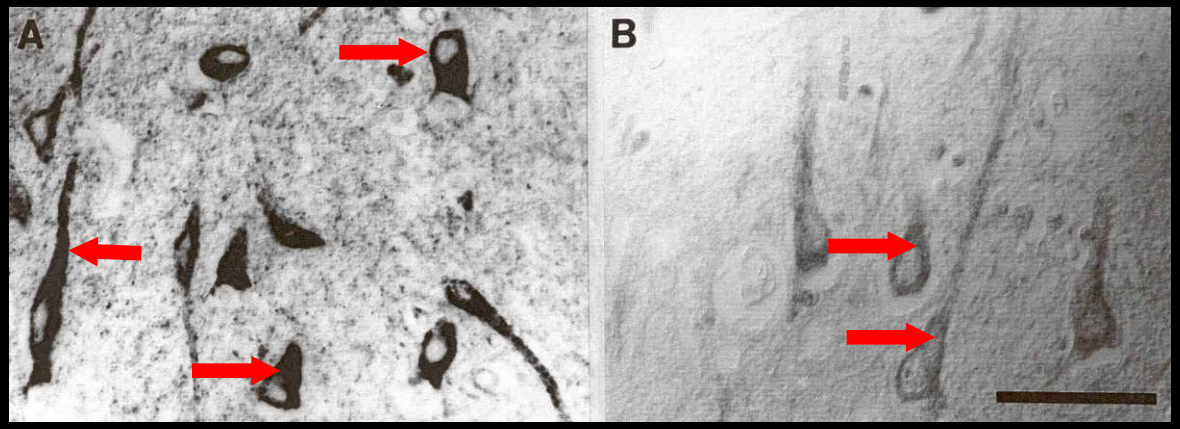
In situ hybridization of mtDNA



Alzheimer Control

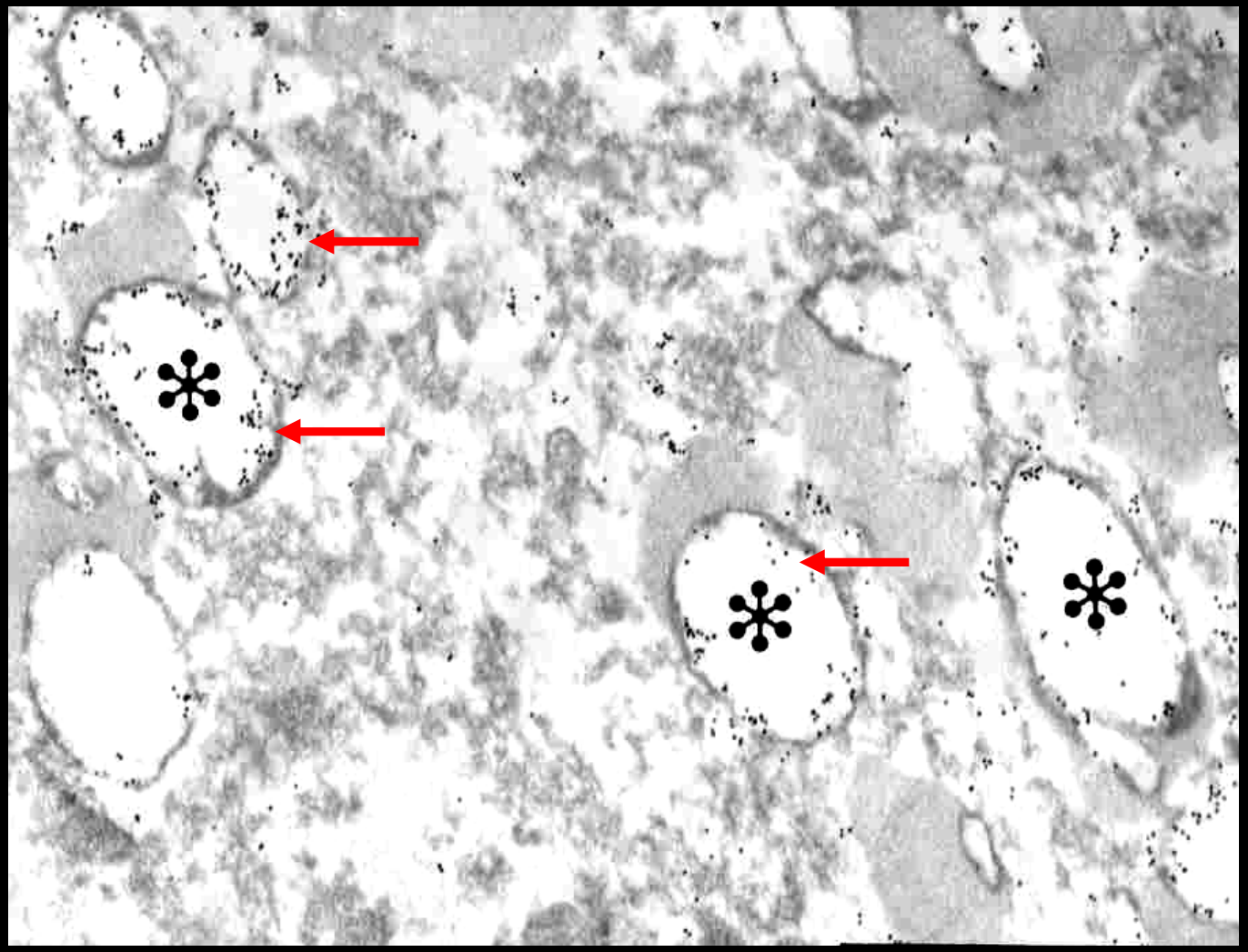


Immunolocalization of Cytochrome oxidase 1 is increased in AD

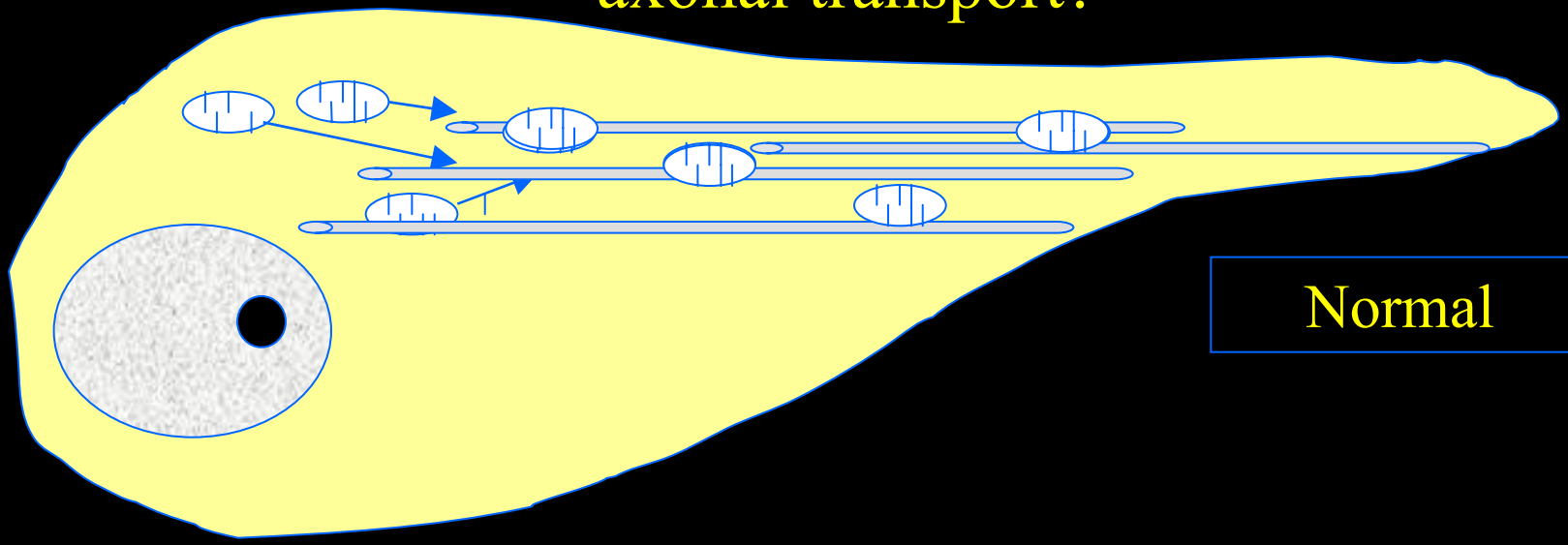


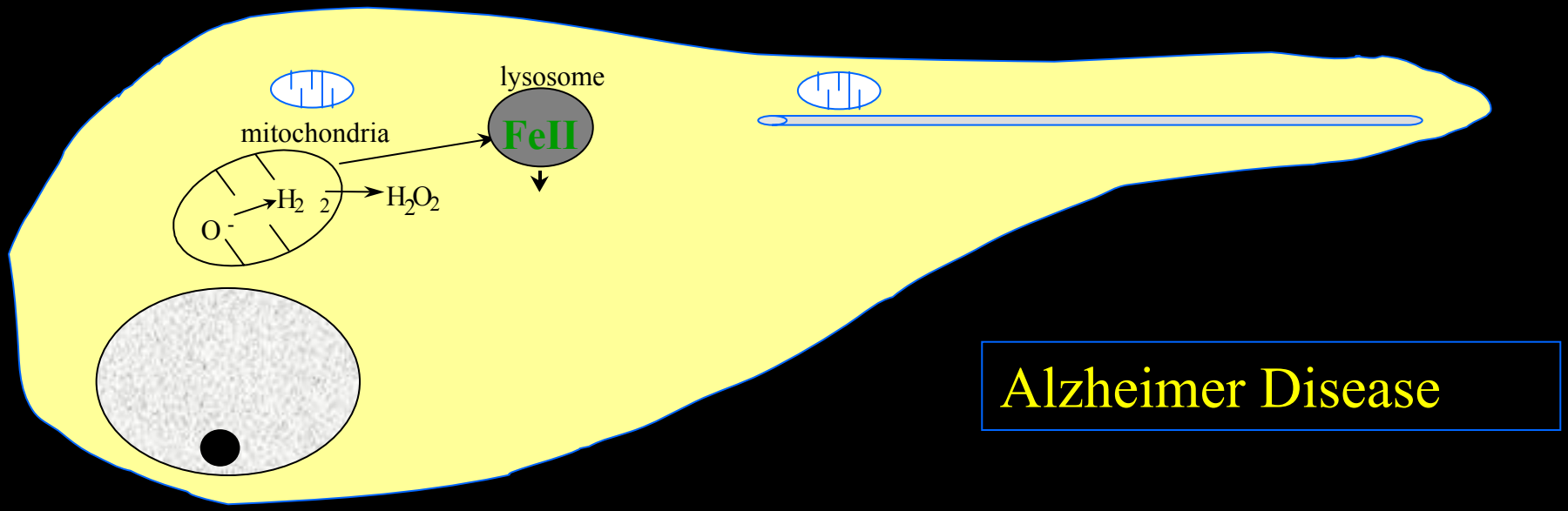
Alzheimer disease Control

Mitochondria components are in autophagosomes



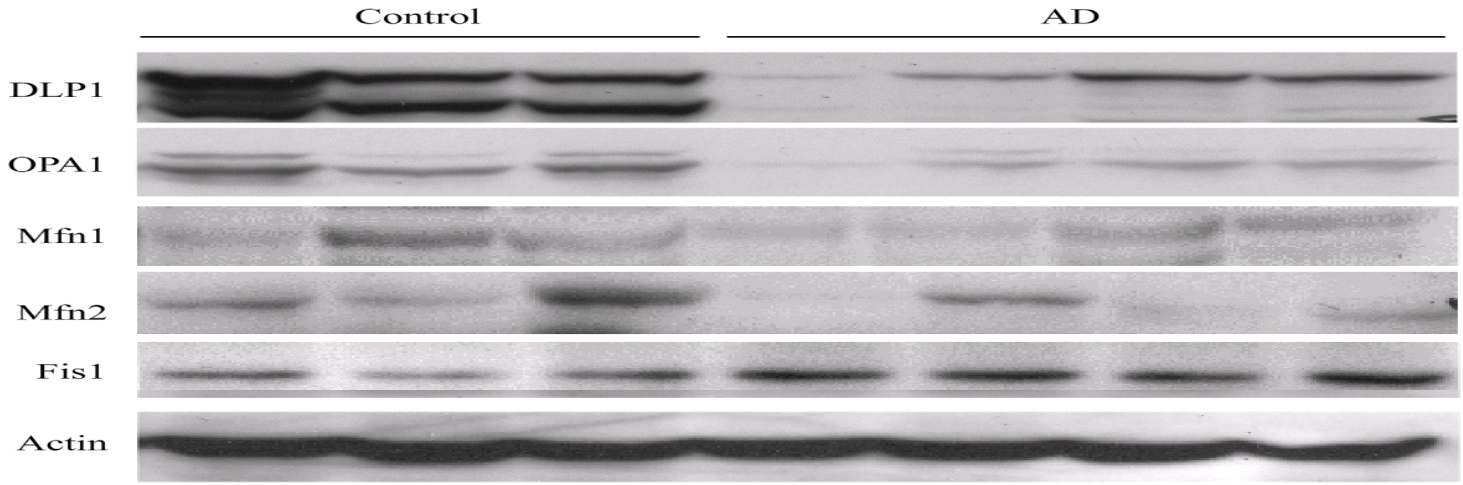
Could the mitochondrial problem be related to reduced axonal transport?



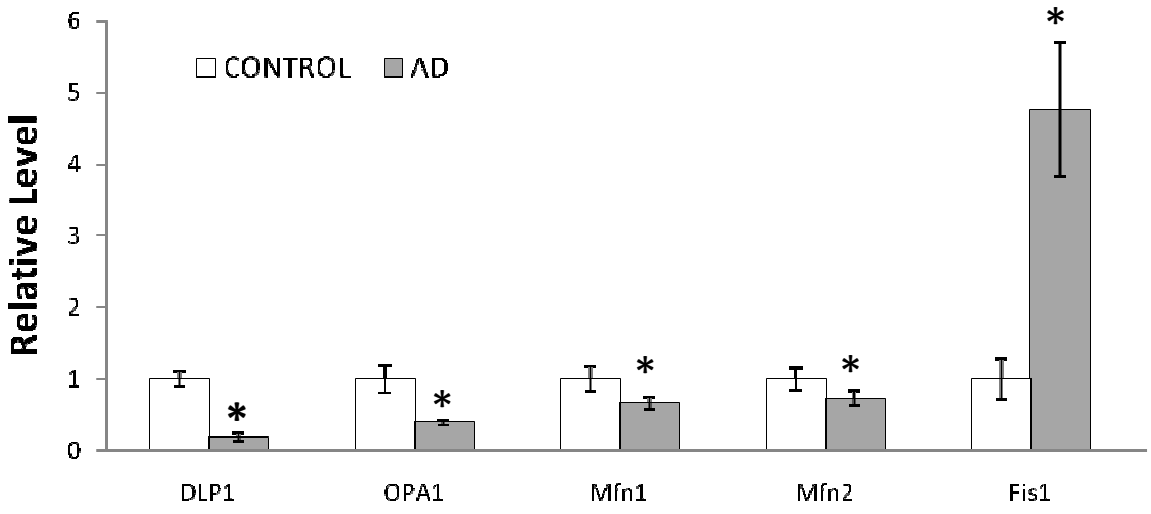


Mitochondrial Fission/Fusion Proteins are Changed in AD Brains

A



B

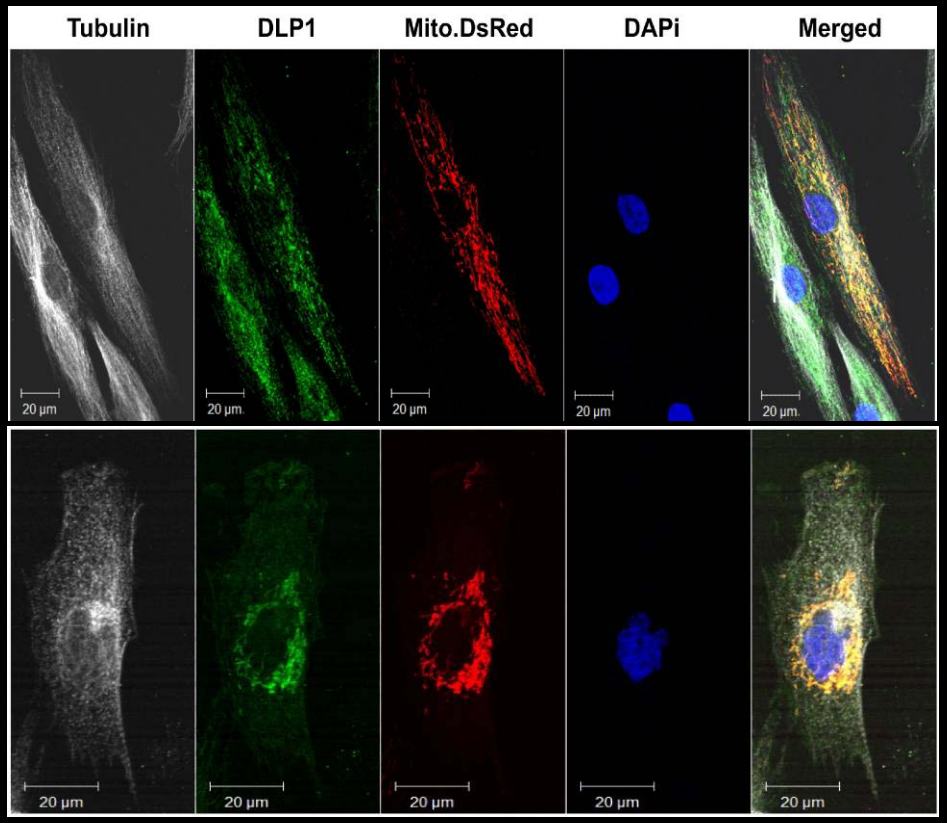


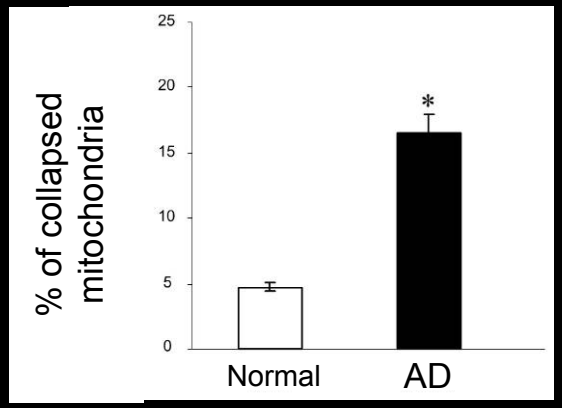
Control: n=12; AD: n=13

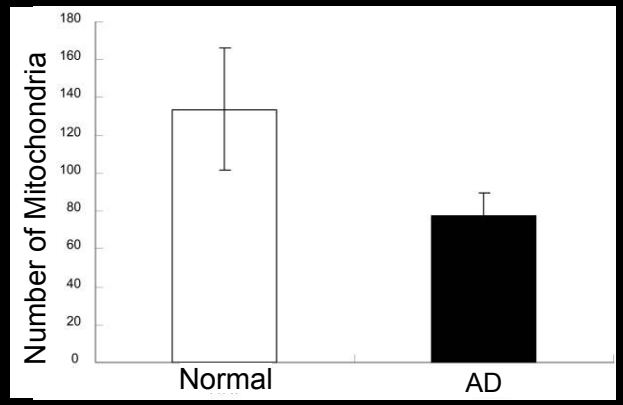
…and perinuclear clumping

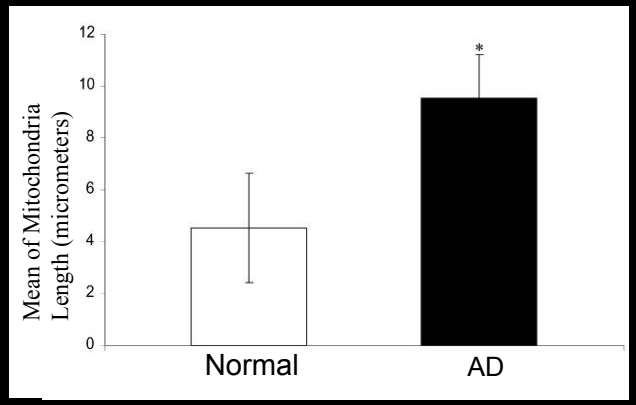
Disbursed mt

Collapsed mt

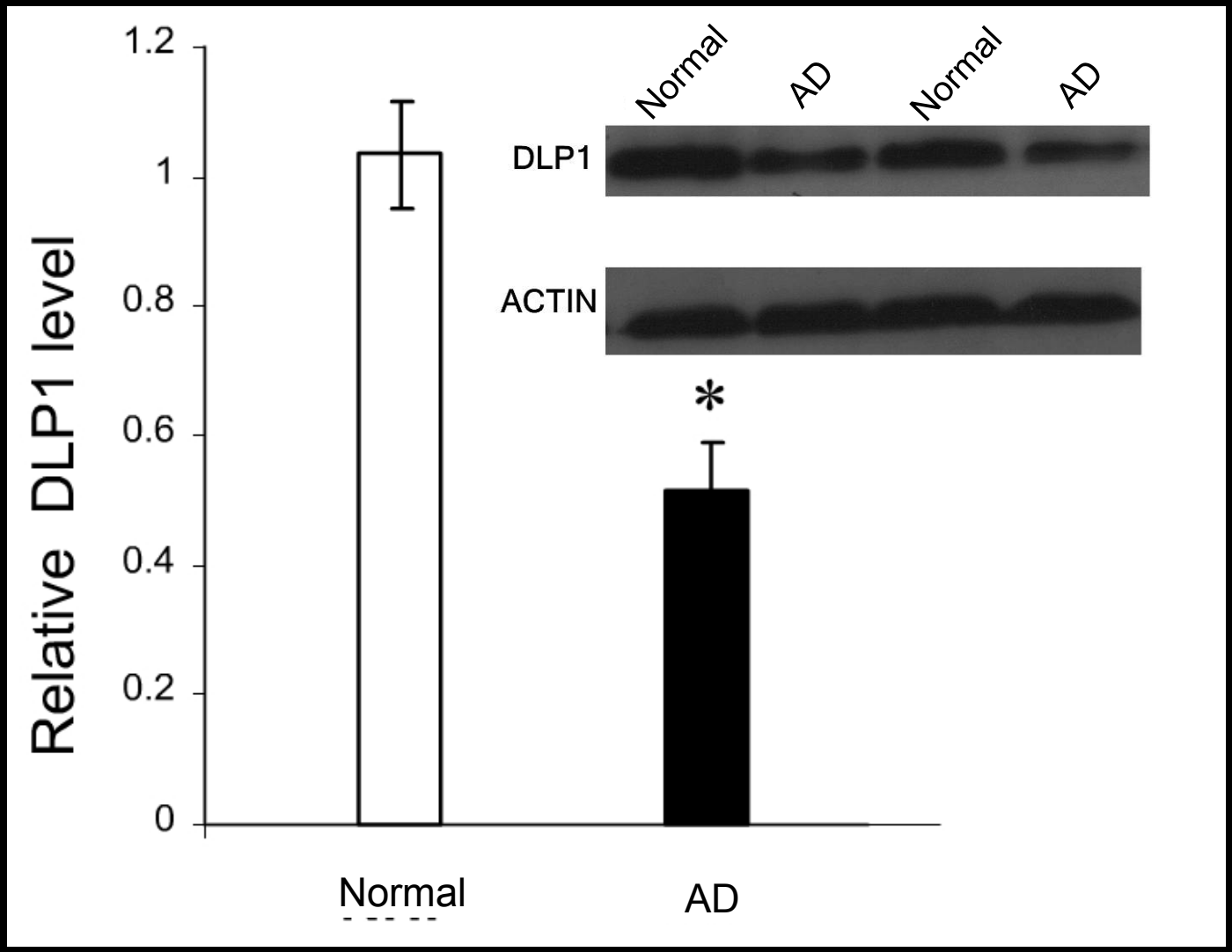




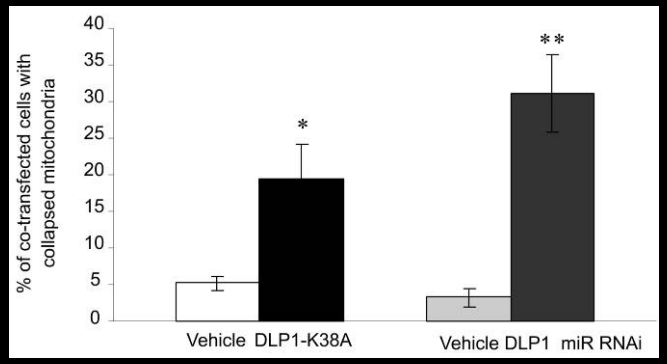


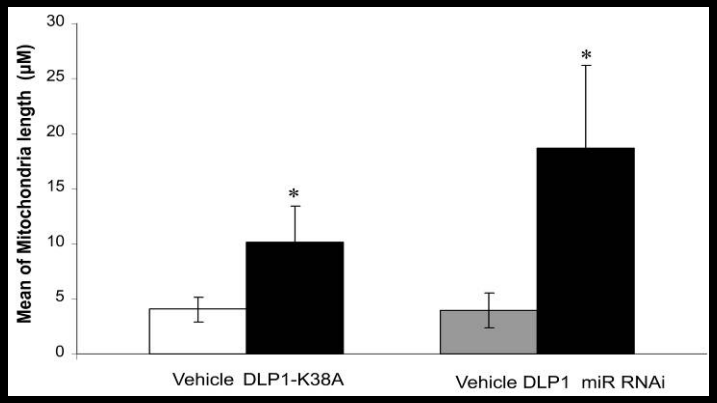


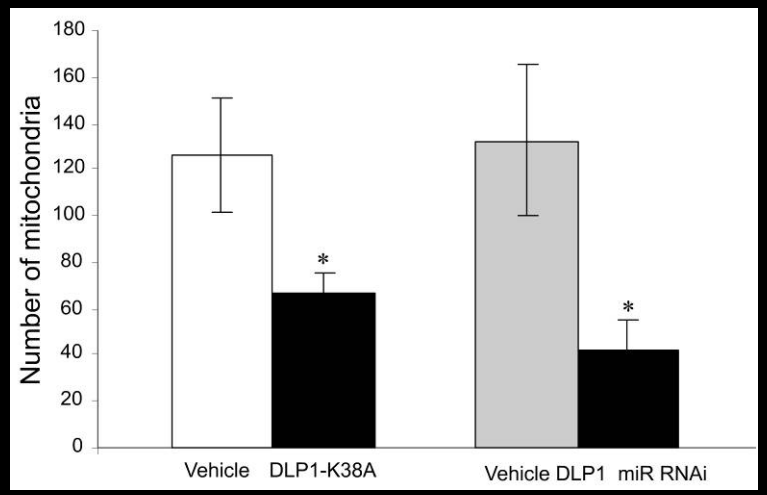
Decreased DLP1 levels are found in AD fibroblasts

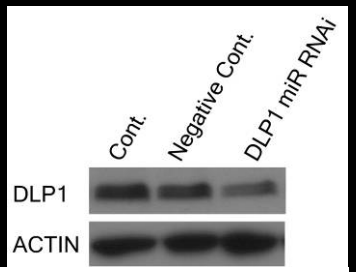


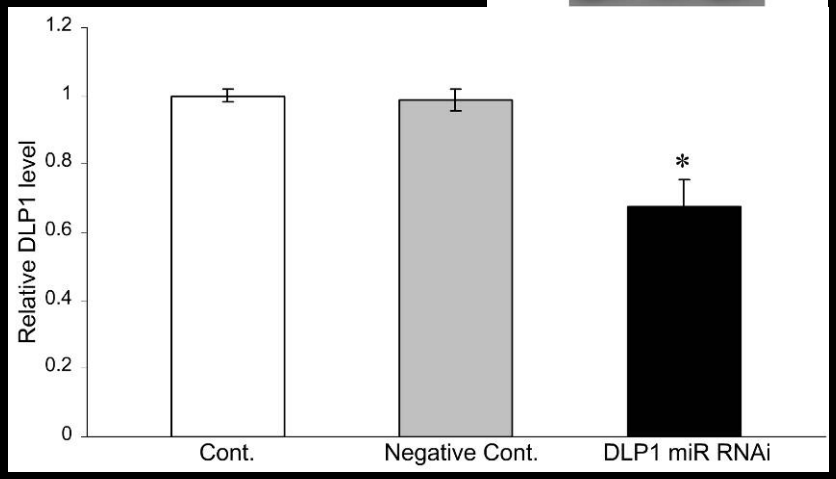
Reduction of DLP1 levels in normal cell fibroblasts by iRNA affects mitochondria

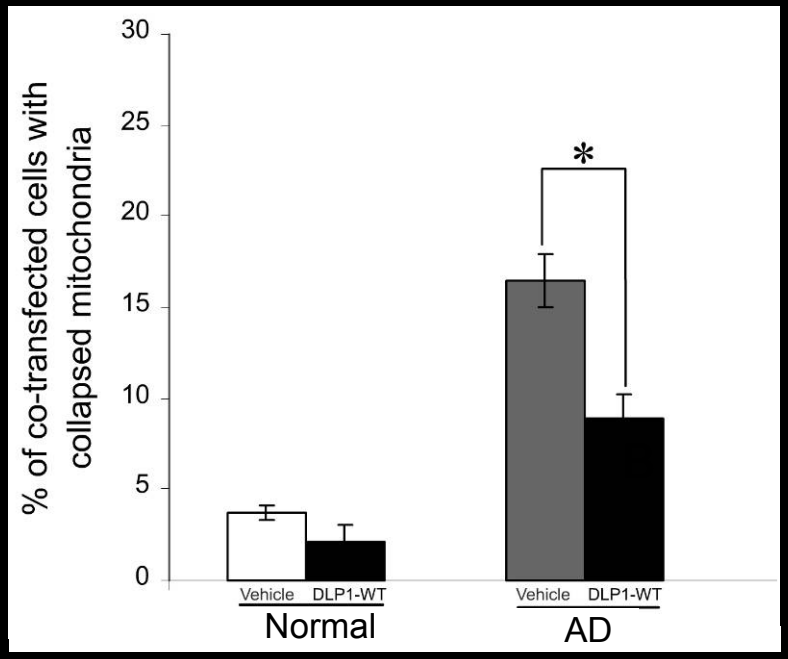




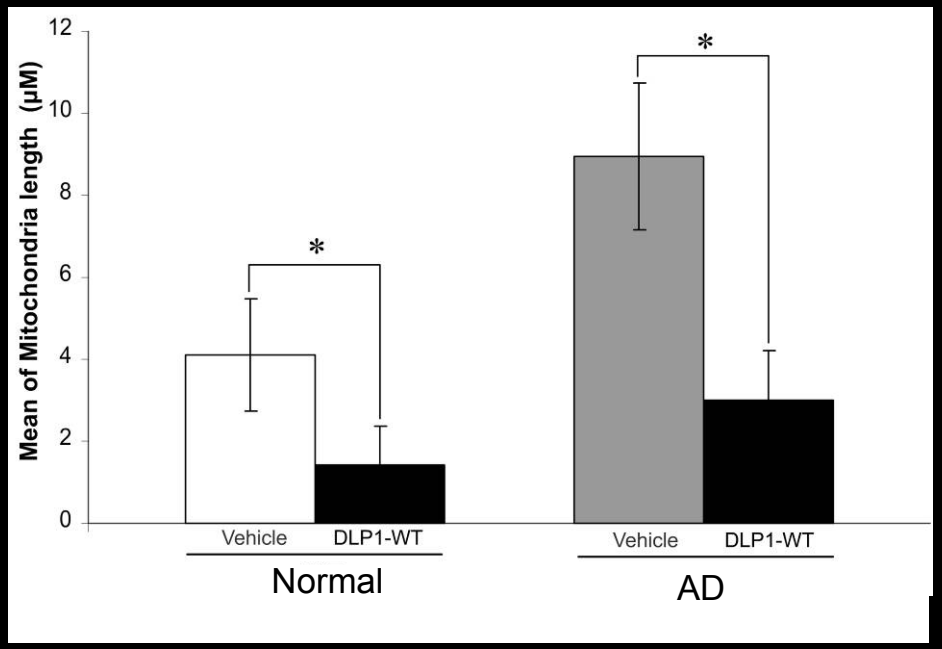


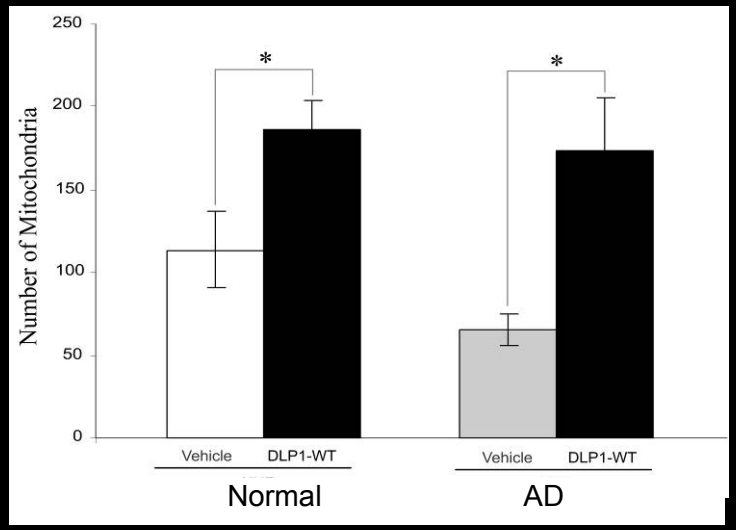




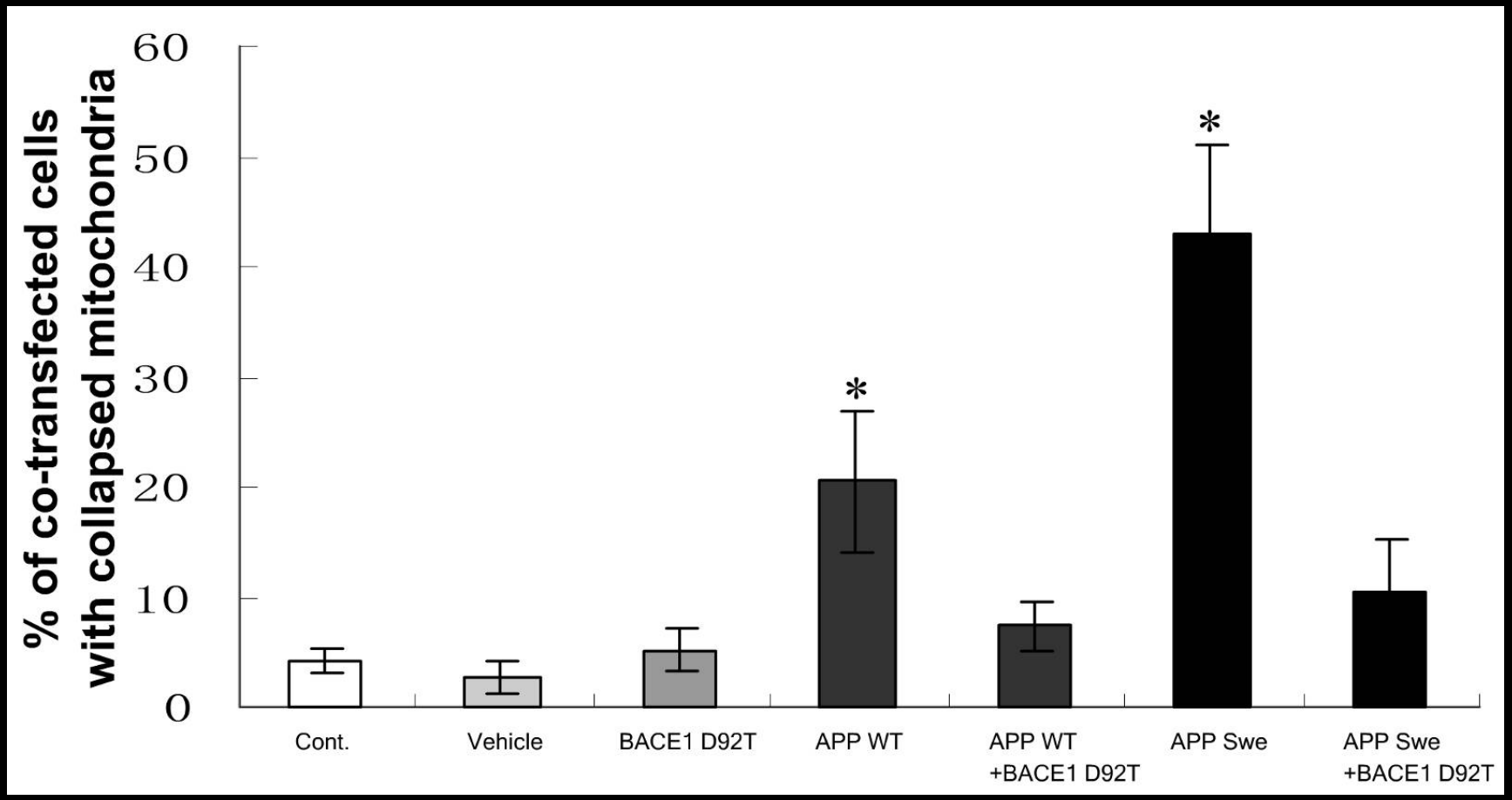


Increase of DLP1 prevents mitochondrial changes found in AD

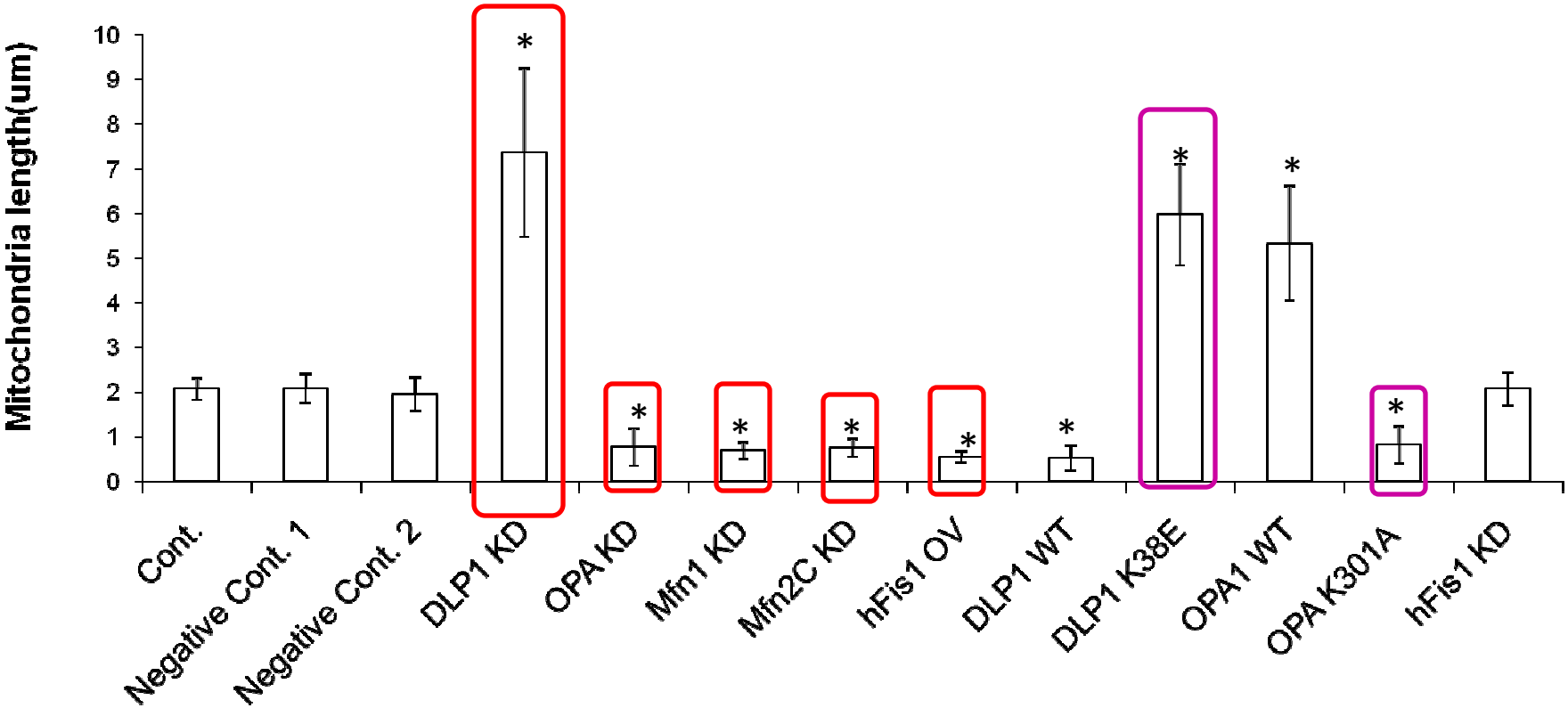




APP plays a role in mitochondrial network collapse

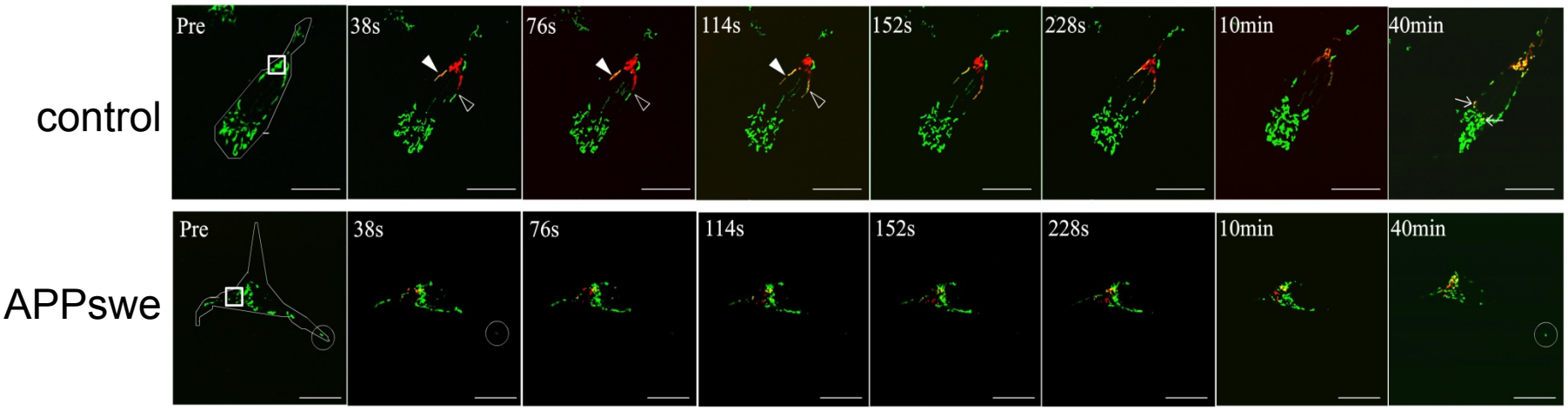


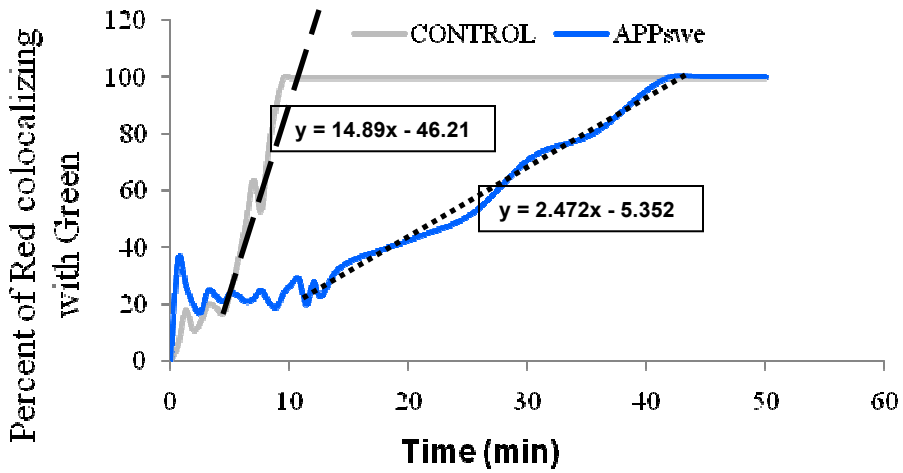
Mitochondrial Length in Neuronal Processes

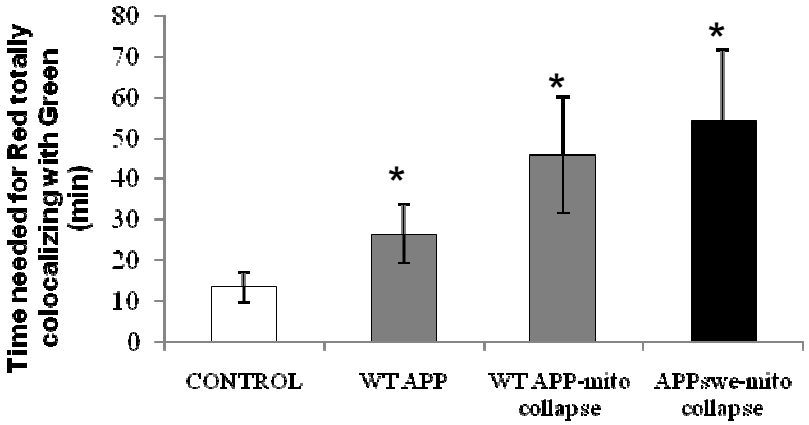


Quantification of mitochondria length in a segment of neuronal process 200 µm in length beginning from the cell body of neurons either overexpressing or knocking down mitochondria fission/fusion proteins. At least 500 cells were analyzed in each experiment for each cell line, experiments were repeated three times. (*p<0.05, student-t-test)

Mitochondria fusion is impaired by APP overexpression

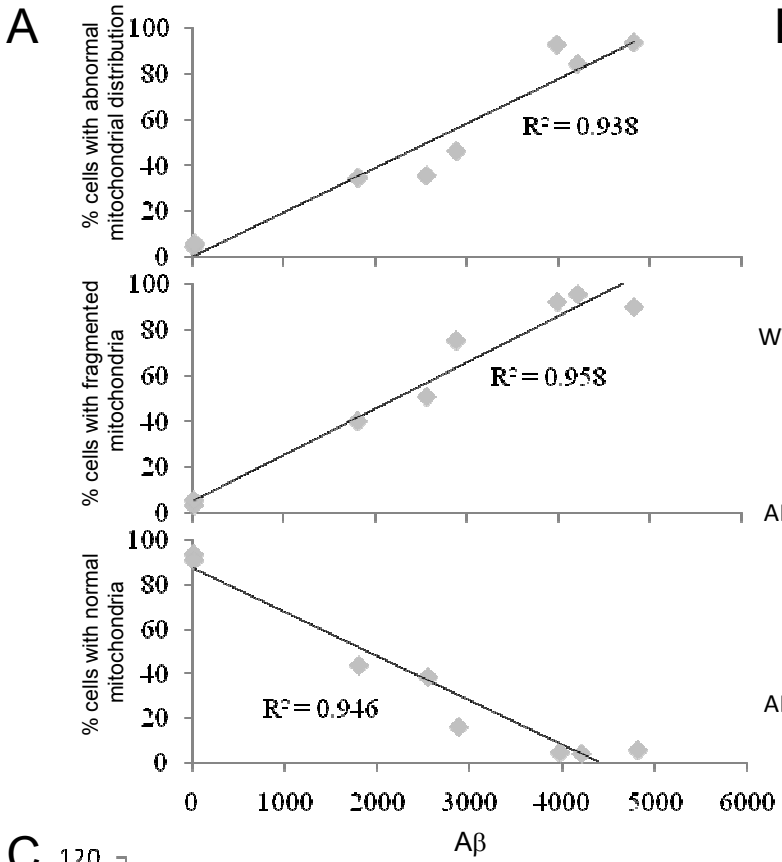


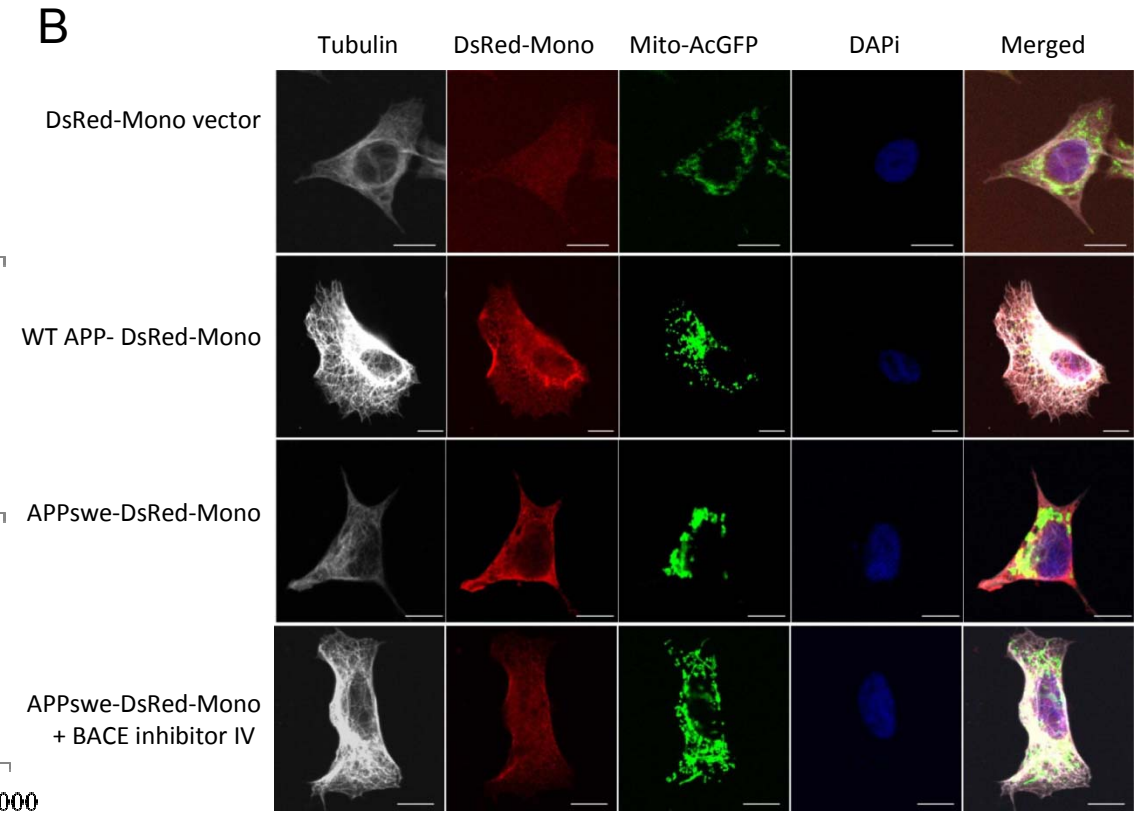


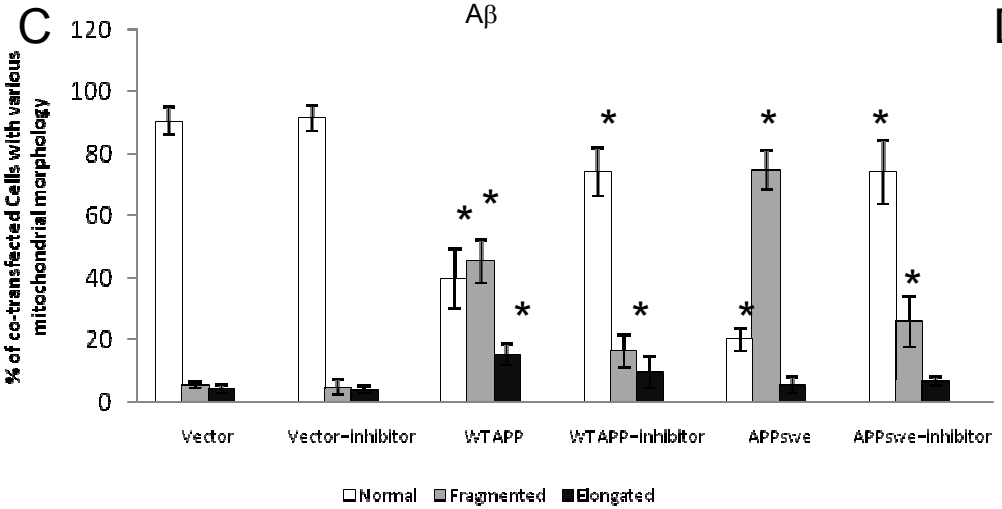


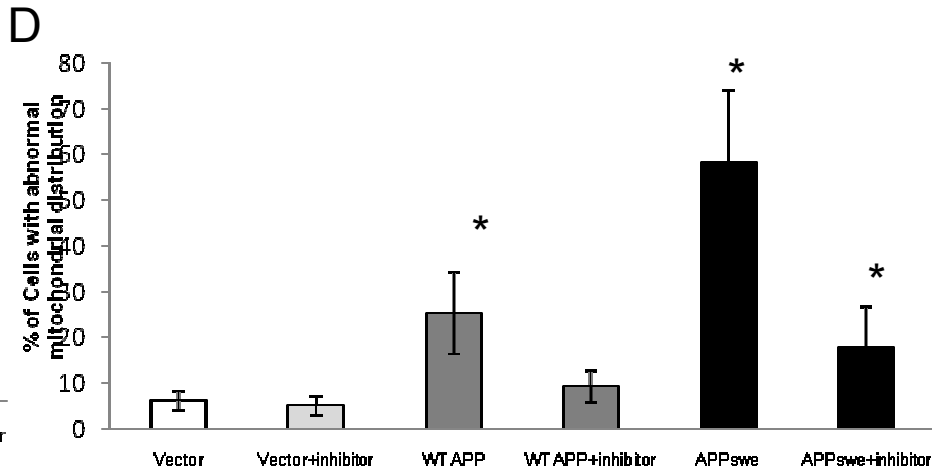
N=5 for each

Abeta Overproduction Underlies APP-Induced Mitochondrial Abnormalities

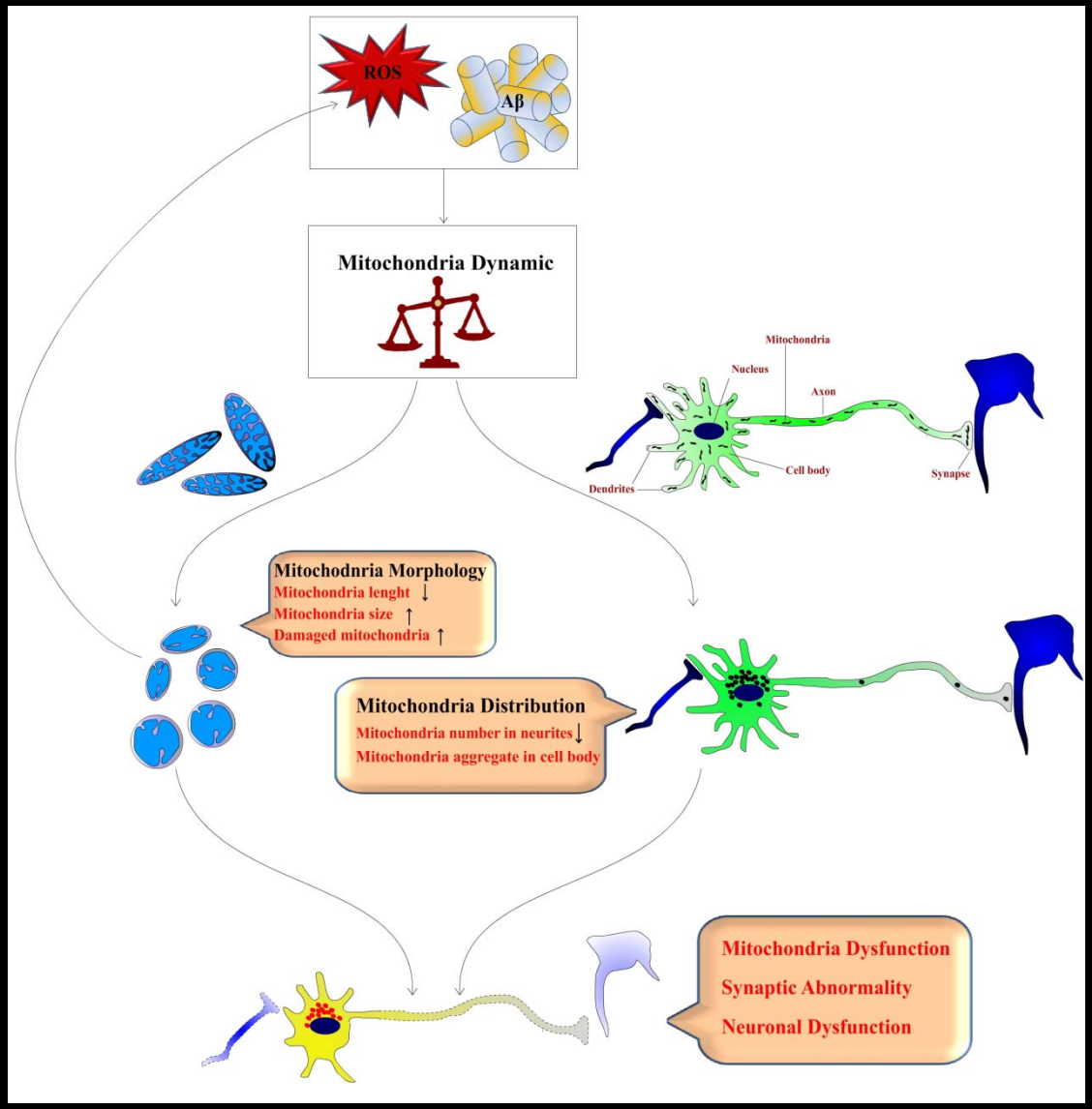








Conclusion


ROS
Aβ
Mitochondria Dynamic
Mitochondria
Nucleus
Axon
Cell body
Dendrites
Synapse
Mitochodnria Morphology
Mitochondria lenght ↓
Mitochondria size ↑
Damaged mitochondria ↑
Mitochondria Distribution
Mitochondria number in neurites ↓
Mitochondria aggregate in cell body
Mitochondria Dysfunction
Synaptic Abnormality
Neuronal Dysfunction

2768 *Biochemistry* **2003**, *42*, 2768−2773

Accelerated Publications

Metal Binding and Oxidation of Amyloid-β within Isolated Senile Plaque Cores: Raman Microscopic Evidence†

Jian Dong,‡ Craig S. Atwood,§ Vernon E. Anderson,‡ Sandra L. Siedlak,§ Mark A. Smith,§ George Perry,§ and Paul R. Carey*,‡

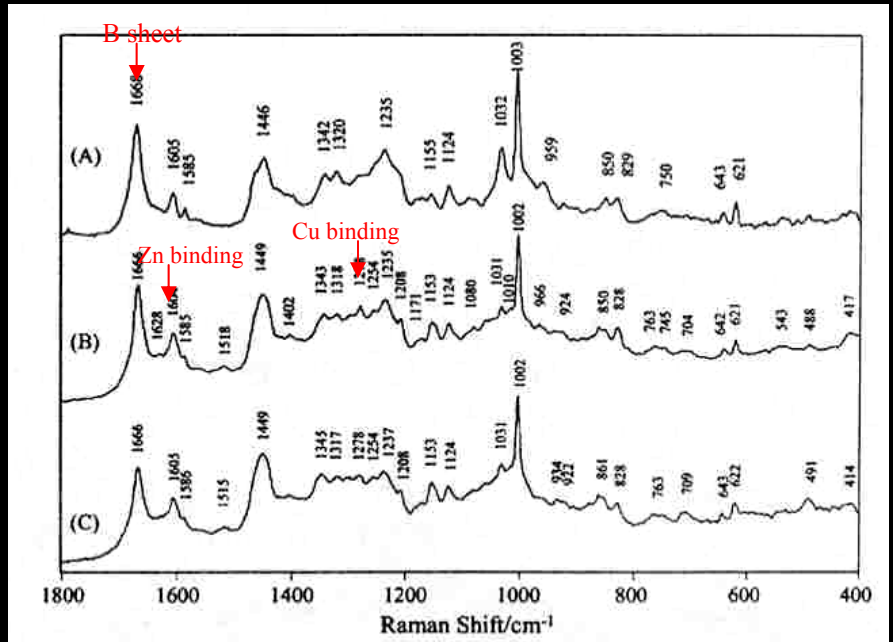
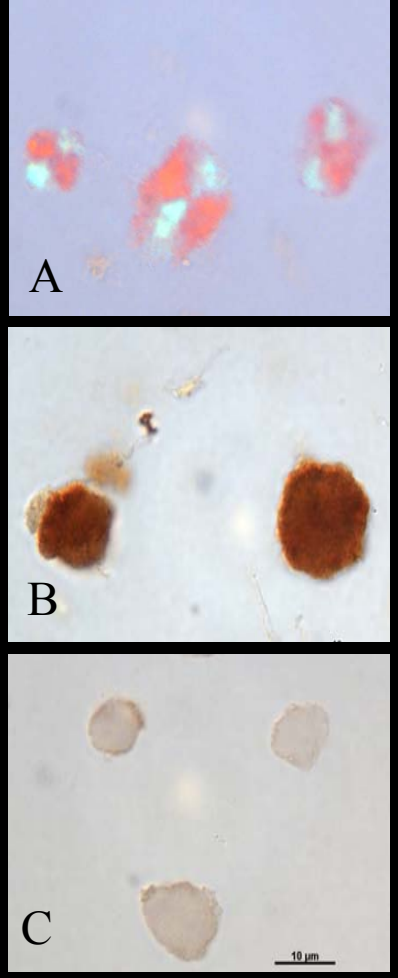


FIGURE 2: Raman spectra of (A) synthetic Aβ(1−42) fibrils, (B) SP cores, and (C) SP core treated with 2.5 mM EDTA (pH 7.0) for 24 h.

Spectra of plaque cores (B) are virtually identical to synthetic peptide (A). Intensity at 1604 is indicative of Zn binding and at 1278 for Cu binding, both increased in purified cores.

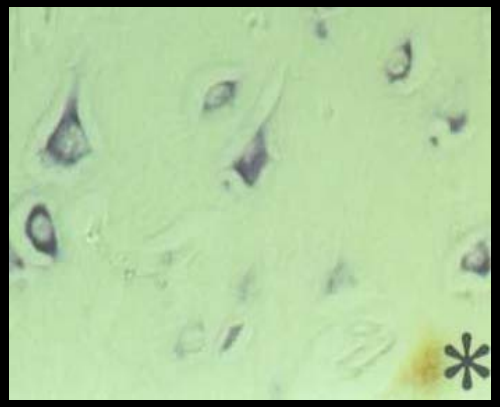


Amyloid plaque cores purified from brain demonstrate characteristic Congo red birefringence (A), are strongly reactive with antisera to Aβ 1-42 (B), but not with irrelevant antisera (C).

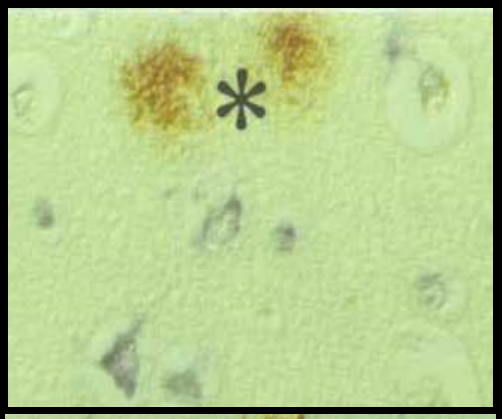
Oxidative damage (8OHG, blue) decreases with increased amyloid - β (brown)

In vivo

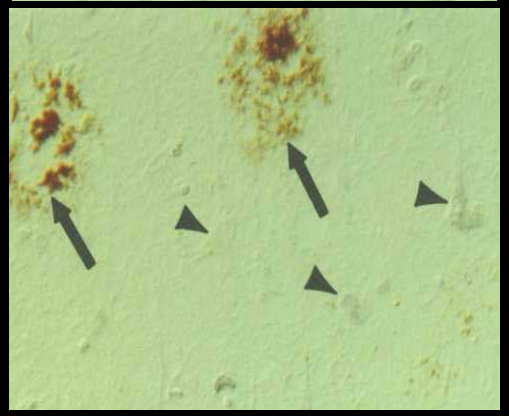
17 yr.

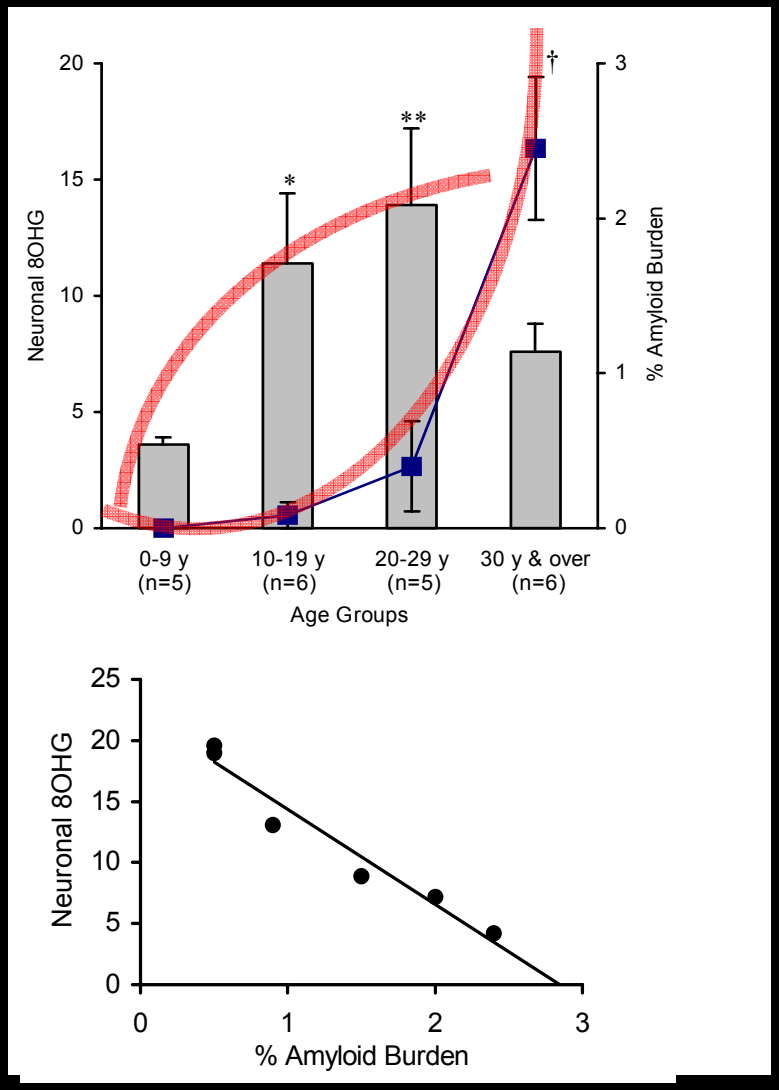


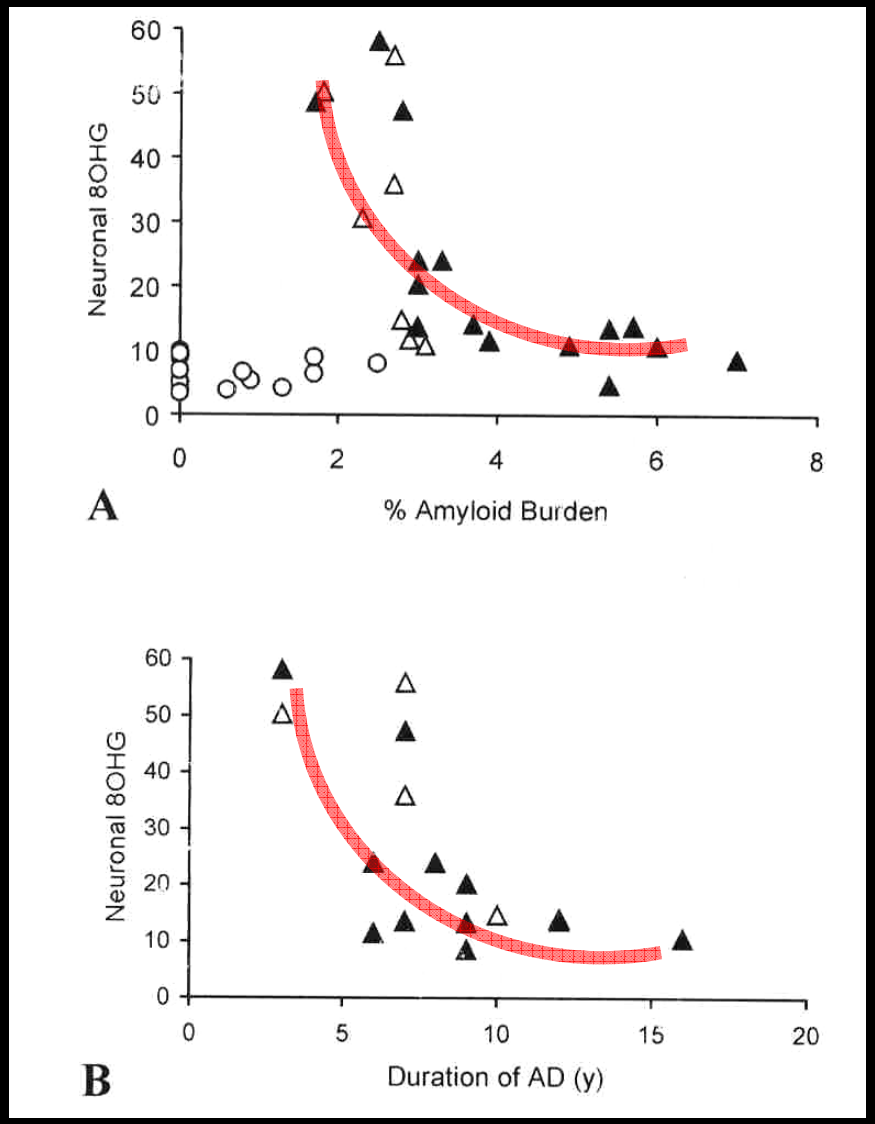
31 yr.



61 yr.







Levels of neuronal 8OHG in AD decrease with:

A. increasing levels of amyloid ($p=0.002$ in cases with ApoE 4 and $p=0.05$ in cases lacking ApoE4) and

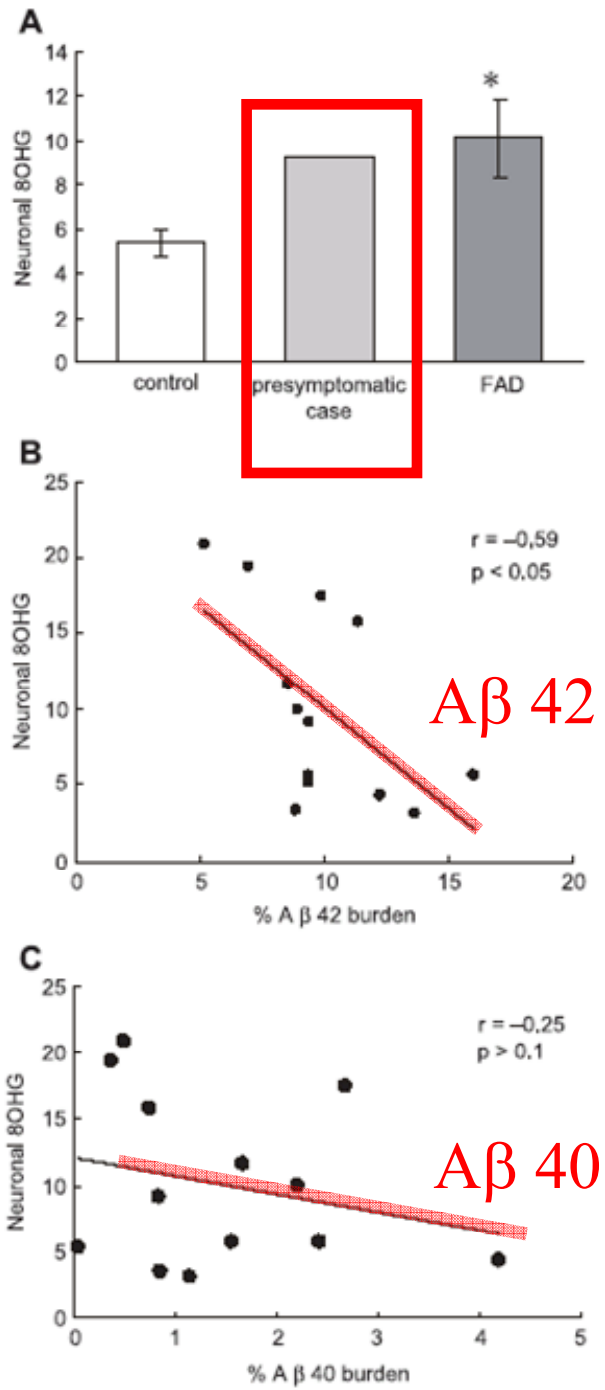
B. the duration of the disease ($p<0.03$).

▲ AD with ApoE4

△ AD lacking ApoE 4

○ Control

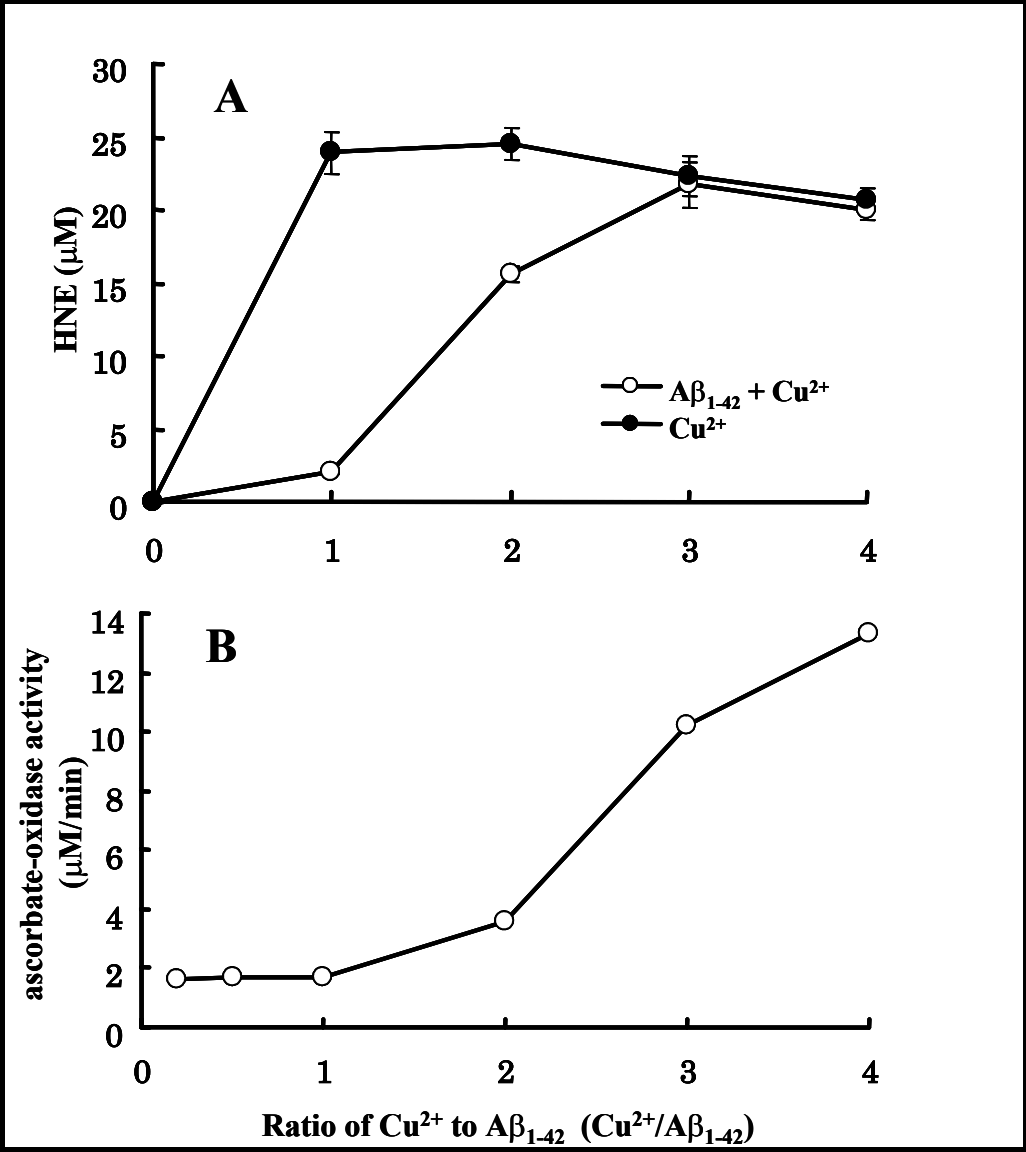
Familial AD Cases



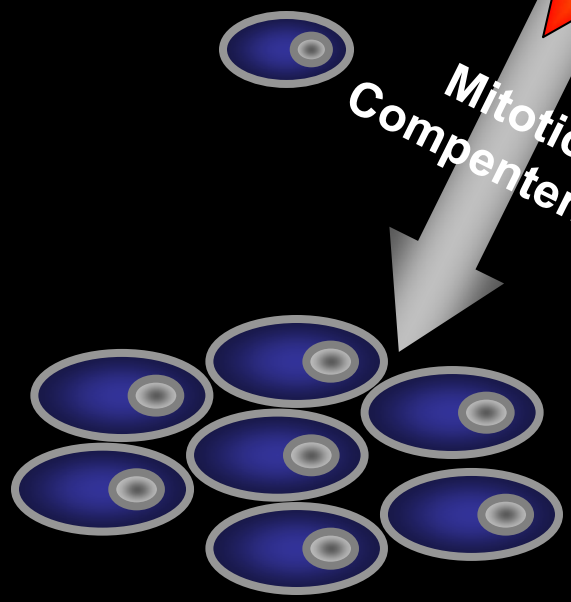
Levels of 8OHG immunoreactivity are significantly increased in familial AD cases (FAD) cases (n=13, avg age 59 yr) compared with controls (n=15,avg age 66 yr). One presymptomatic case with PS-1 mutation shows a similar level to average FAD cases (A).

In (FAD) there is a significant inverse correlation of Aβ42 burden (B) but not with Aβ 40 (C).

Aβ reduces copper-induced oxidation



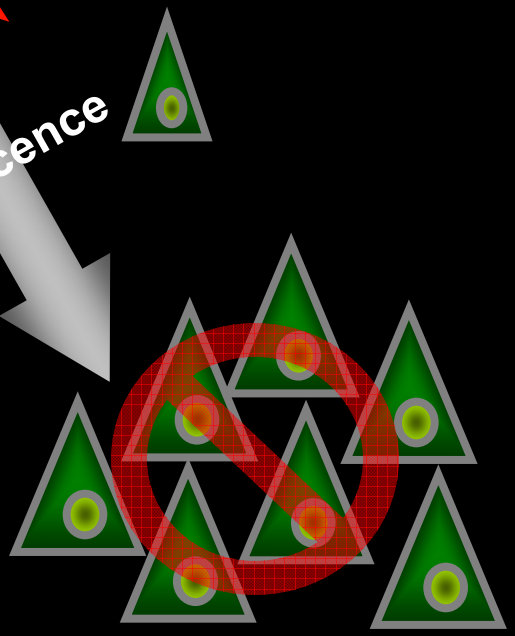
Cell Cycle Abnormalities



Mitotic Compentence

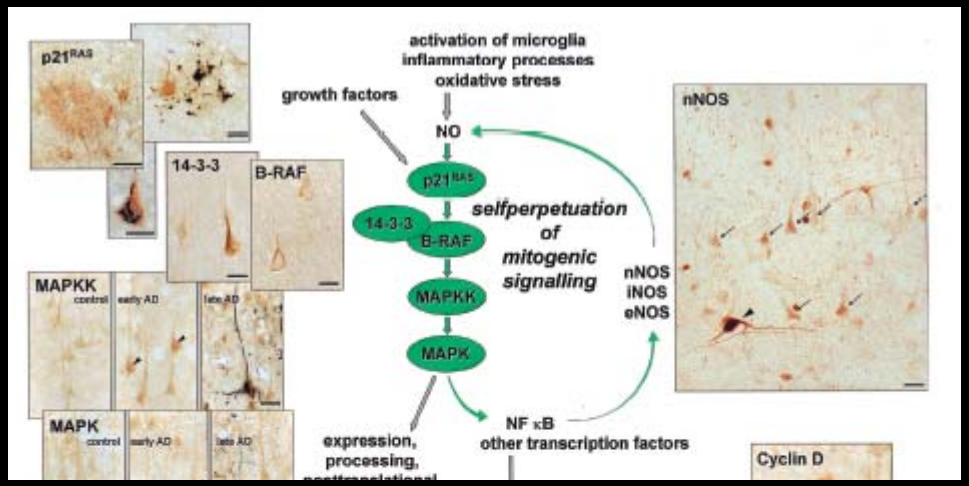
Proliferation:
Cancer

Quiescence

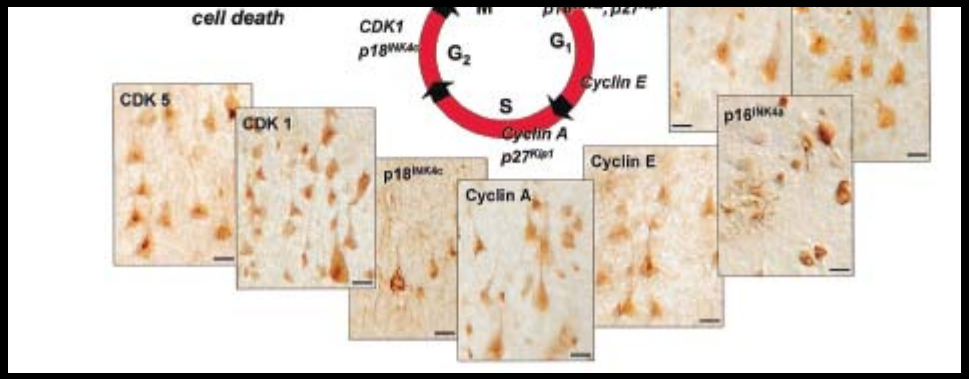


Degeneration:
Alzheimer Disease

Cell Cycle related Proteins in AD



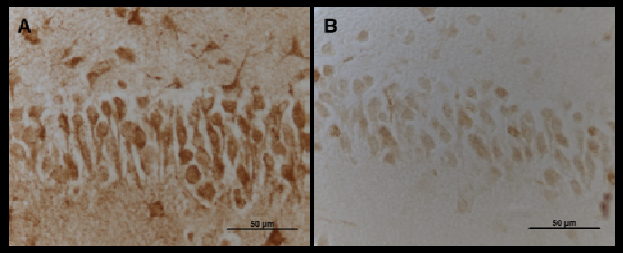
Causes or Consequences?

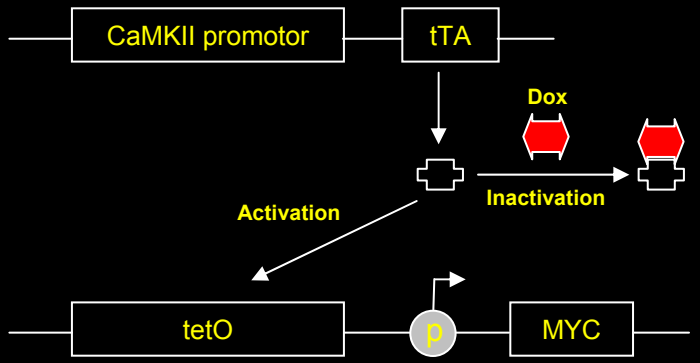


Neurobiology of Aging, 21, 783-796, 2000

Establishment of a new animal model CaMKII-MYC mice

MYC is specifically induced in cerebral cortical and hippocampal neurons by depletion of doxycyline

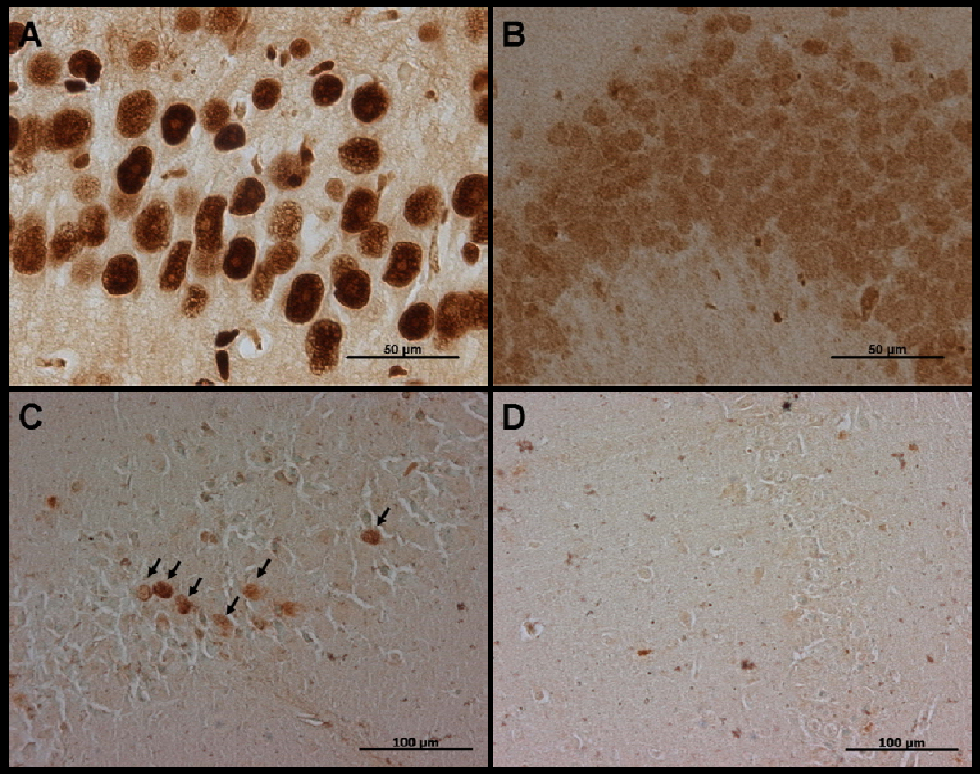




C-MYC (now referred to as MYC) is a member of a family of proto-oncogenes comprising C-MYC, N-MYC, and L-MYC. MYC encodes a transcription factor that, as part of a heterodimeric complex with MAX, regulates the expression of a multitude of genes involved in regulating cellular proliferation and growth. Overexpression of MYC is commonly associated with tumorigenesis. Where MYC exerts its neoplastic function by inducing autonomous cellular proliferation and cellular growth, blocking differentiation, and inducing genomic destabilization.

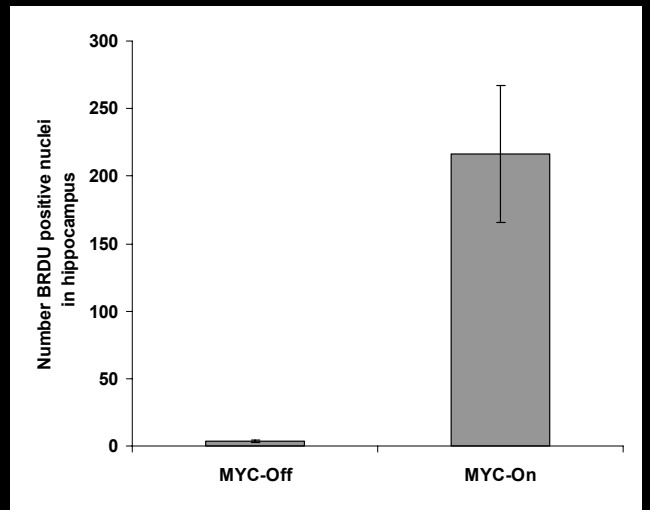
Cell Cycle Re-entry in CaMKII-MYC mice

MYC-ON MYC-OFF

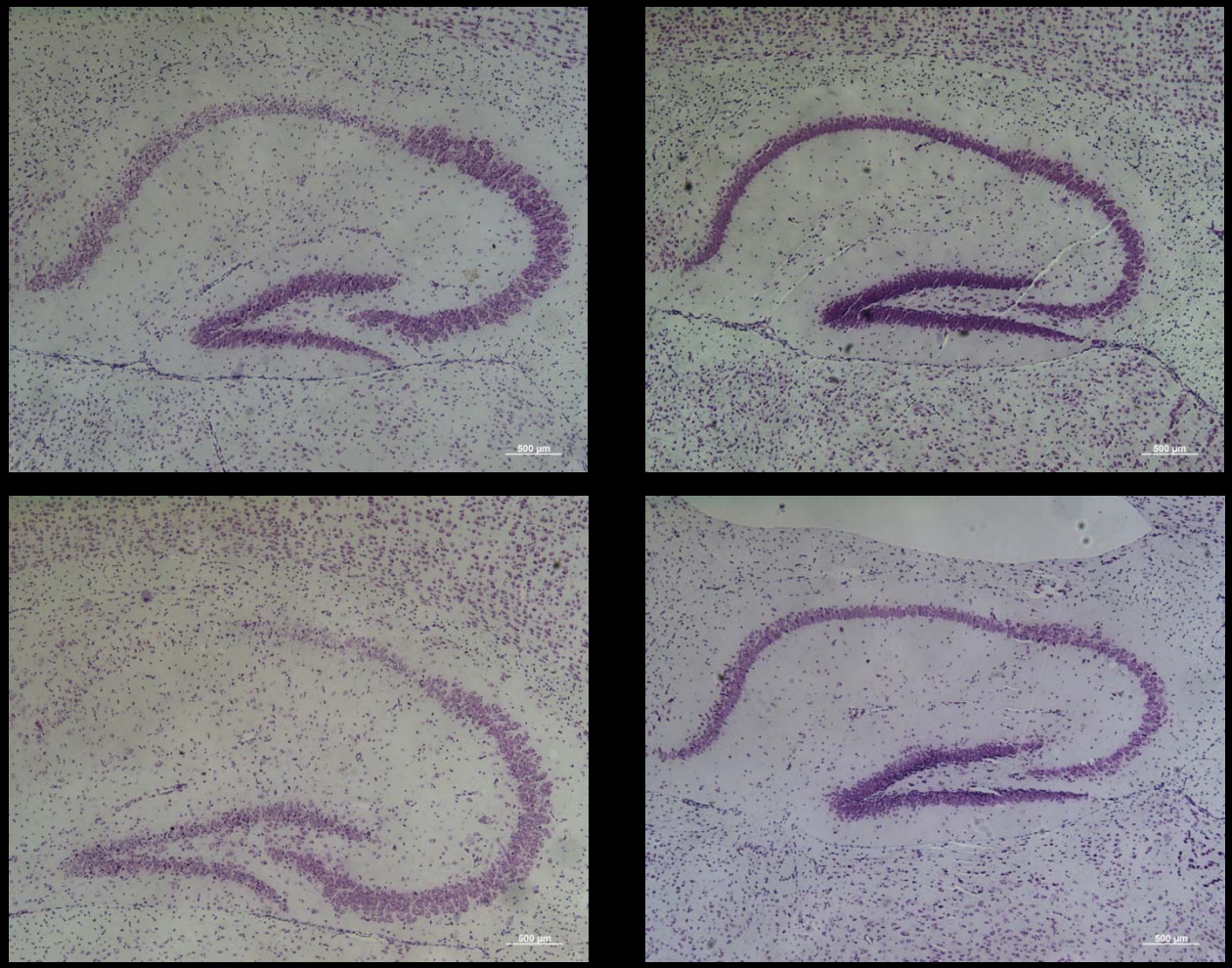


PCNA

BrdU

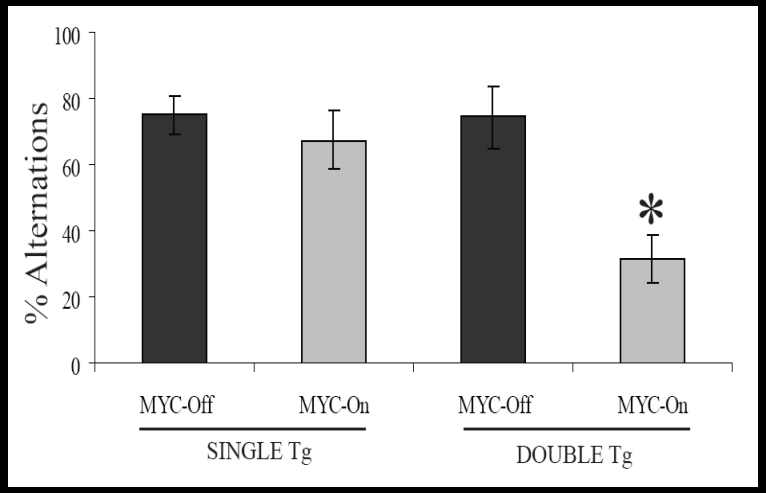


Selective Neurodegeneration in CaMKII-MYC mice



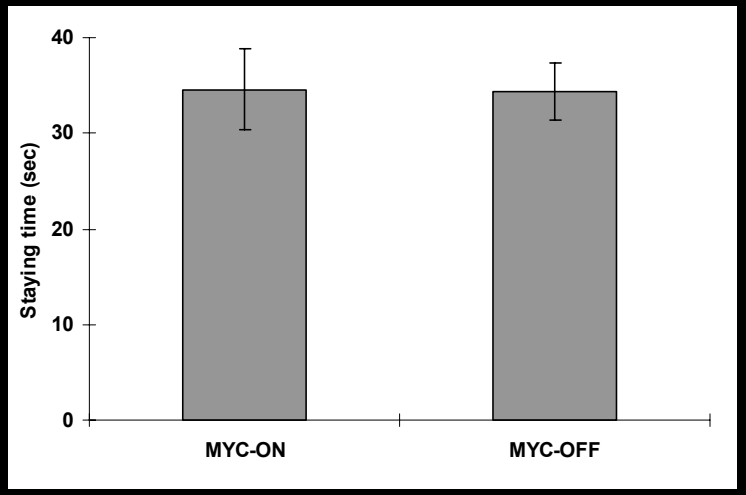
MYC-ON MYC-OFF

Cognitive deficits in CaMKII-MYC mice



Modified T-Maze

(Cognitive function)



Rotarod

(Motor function)

Summary

1. Oxidative damage is a major feature of Alzheimer disease.
2. Mitochondria abnormalities are critical to the oxidative abnormalities.
3. Aβ/AβPP are linked to mitochondrial turnover and oxidative balance.
4. Cell cycle abnormalities may be the second hit in Alzheimer disease.