1. INTRODUCTION

A healthy human brain has around 100 billion neurons with expanding augmentations that are associated with one another by synapse.[1] The information between two synapses is transferred through tiny bursts of chemicals known as a neurotransmitter. The synapse allows rapid travel of neuronal signs forming a cellular basis of recollections, thoughts, feelings, sensations, and development of physical abilities.[2] The diseases, injuries, and illnesses during the normal aging of the brain obliterate the synapses. These causes slacken nerve connections that slow down the speed of communication between neurons. It is common that as people grow older, they lose some neurons, where normal age-related changes in memory and thinking are observed, but confusion, perplexity and cognition impairments that interfere with routine life are not part of the ordinary maturing process.[3]

Dementia damages the neurons in the brain so the nerve impulse can't be sent adequately, which prevents the brain from functioning normally. Dementia is a Latin word, meaning "madness" from *de-* "without" + *mens-* "mind".[4] It is a syndrome characterized by disruption of multiple cognitive functions such as thinking, reasoning, and behavioral abilities that intervenes the individual's routine life and activities. The impairments of cognitive functions are commonly followed by disturbed social behavior and emotional control. The definite signs of dementia that patients encounter depend on the affected area of the brain inducing dementia.[5]

Alzheimer's disease (AD), semantic dementia, vascular dementia, frontotemporal dementia, and dementia with Lewy bodies are the most widely recognized types of dementia. Among them, AD is the most common type of dementia, causing up to 60-80 % of the dementia cases.[6]

1.1. Alzheimer's Disease

AD was first reported by a German scientist, Dr. Alois Alzheimer in 1907. He termed it as a 'neurodegenerative disease' meaning continuous degeneration of the neuronal system. It was characterized by a continual decline in cognitive ability and serious behavioral abnormalities like restlessness, irritability, disorientation, depression, and anxiety. It was later defined as "A progressive neurodegenerative disorder resulting in irrevocable

loss of neurons within the cerebral cortex and hippocampus area characterized by loss of memory, confusion, impairment of intellect, social withdrawal, poor judgment, etc."[7]

AD is the most prominent form of dementia. More than 50 million people are suffering from it worldwide, and the number will significantly rise up to 152 million by 2050 if no cure or preventive measures are found.[8] The prevalence of AD quickly rises from around 2-3 % among the people with age of 70-75 years to 20–25 % for the people of 85 years or more of age. Over this, there's not adequate information to affirm whether AD pervasiveness continues expanding or balances out. Especially in old age, ladies are more likely to be afflicted with AD than men, fundamentally because of age-adjusted increased risk of AD. A few investigations have demonstrated that the general pervasiveness of AD differs broadly among nations, being impacted by social and financial components.[9]

The decline in acetylcholine (ACh) levels in the synapse,[10] accumulation of the extracellular beta-amyloid (Aβ) plaques[11] and intraneuronal tangles of the tau protein[12] are some hallmarks associated with AD. Low levels of ACh have been accounted to disrupt typical reasoning and memory. Aβ plaques may interrupt the signal transfer from neuron to neuron which leads to cell death, whereas tau tangles block the transport of supplements and other vital nutrients inside neurons.[13] When the early changes of AD occur, the brain can initially compensate for these changes, and the affected individual is able to function normally. As the nerve cells start getting damaged, the cerebrum cannot make up for the changes, and the individual shows an inconspicuous decrease in intellectual capacity. Over the long haul, plaques and tangles show up not just in specific areas of the brain engaged with subjective capacity, but also in other parts of the brain. Afterward, the damage to nerve cells is critical to such an extent that people show a clear intellectual drop, e.g., memory loss or confusion in place or time, social side effects such as despondency, character changes and loss of enthusiasm for exercises they used to appreciate. Even later, fundamental real capacities, e.g., swallowing, are impeded.[14]

The most common symptoms associated with AD are as follow:[15,16]

-Reduced ability to take in and remember new information

The most extensively seen signs of AD are cognitive dysfunctions, particularly overlooking recently learned information, missing significant dates or occasions, asking a piece of similar information over and over, and progressively relying on memory aids.

-Impairments to reasoning and complex tasking

A few people feel changes in their capability to plan or to operate with numerals. They may encounter challenges in following a natural formula, monitoring day to day expenses. They may feel focusing issues and take longer time to complete the task than they did previously.

-Difficulty in executing everyday tasks

Individuals with AD usually imagine that it is challenging to complete everyday tasks. Once in a while, individuals may encounter difficulty in traveling to a known place or dealing with a financial limit at work.

-Misplacing things and forgetting the capability to retrace steps

People with AD may put things in random places, and forget them. Then they are not able to go back to their steps for finding them again. Often, they start accusing others for stealing them. It might happen now and again after some time.

-Impaired speaking, reading, and writing

People with AD may face difficulties in following or participating in a discussion. They may stop talking in a discussion and have no idea how to continue or they may start retelling the things. They may battle with jargons, have difficulties in finding proper words or call things by an inappropriate name.

-Changes in mood and personality

The character and mind-set of persons with AD may change as they become more confused, anxious, frustrated, frightful or suspicious. They might be agitated at home and work, with friends or in spots where they are out of their comfort zone.

-Impaired visuospatial abilities

Some people with AD might feel vision problems. They may feel difficulties in deciding distances and judging the shades or complexes, which may cause issues while driving.

-Confusion with time or place

People with AD may find difficulties in tracking the dates, seasons and time.

The symptoms of AD worsen with the time, although the speed at which the disease advances varies from person to person. Based on how the ability of a person changes once the symptoms appear due to cognitive functional impairments in AD, it is divided into seven stages.[16]

Stage 1: No Cognitive Impairment

Symptoms: -No obvious signs

Stage 2: Very Mild Cognitive Impairment

Symptoms: -Slight forgetfulness associated with aging.

Stage 3: Mild Cognitive Impairment

Symptoms: -Forgetfulness, e.g., misplaced car keys, person's name
-Memory loss, losing items
-Impaired work performance
-Loss of concentration, impaired organization
-Trouble with problem-solving and complex tasks.

Stage 4: Moderate Cognitive Impairment

Symptoms: -Trouble with routine tasks and increased social withdrawal
-Emotional moodiness and lack of responsiveness
-Reduced intellectual ability and trouble holding urine
-Increased memory loss and forgetfulness
-Inability to use or find the correct words
-Difficulty doing challenging mental maths.

Stage 5: Moderately Severe Cognitive Impairment

Symptoms: -Worsening of symptoms in previous stages
-Pronounced memory loss, including memory of personal details.

-Confusion about the location or previous events.

-Further reduced mental activity and problem-solving ability.

Stage 6: Severe Cognitive Impairment

Symptoms: -Needing assistance when getting dressed

-Wondering and getting lost

-Unable to recognize or recall names

-Sleep disturbances

-Incontinence

-Dramatic personality changes including paranoia or delusions

-Changes in personality (paranoia or hallucinations).

Stage 7: Very Severe Cognitive Impairment

Symptoms: -Loss of language skills

-Loss of awareness of surroundings

-Assistance when eating

-Unable to control urination

-Loss of muscle control to smile, swallow, walk, or sit without support.

1.1.1. Causes and Pathophysiology

The etiology of AD is still enigmatic. Different factors, like low levels of neurotransmitter ACh, aggregation of the Aβ peptide, accumulation of hyperphosphorylated tau protein, dyshomeostasis of biometals, oxidative stress, mitochondrial dysfunction, and neuroinflammation, are proposed to play pivotal roles in the pathogenesis of AD. Based on these important factors some hypothesis have been put forward to address the pathophysiology of AD.

1.1.1.1. Cholinergic hypothesis

The cholinergic framework is involved in the physiological processes such as consideration, learning, memory, stress reaction, alertness, rest and tactile information. Deficits in the cholinergic transmission and cholinergic neurons located in the basal forebrain can affect all parts of cognition and conduct, including cortical and hippocampal functions. ACh is synthesized from acetyl-CoA and choline by choline acetyltransferase (ChAT). It is moved into the synapse where it binds to postsynaptic muscarinic and nicotinic

receptors. ACh available at the synapse is readily hydrolyzed by the protein acetylcholinesterase (AChE), forming acetic acid and choline, which is reused by the presynaptic neurons (**Figure 1.1**).[17]

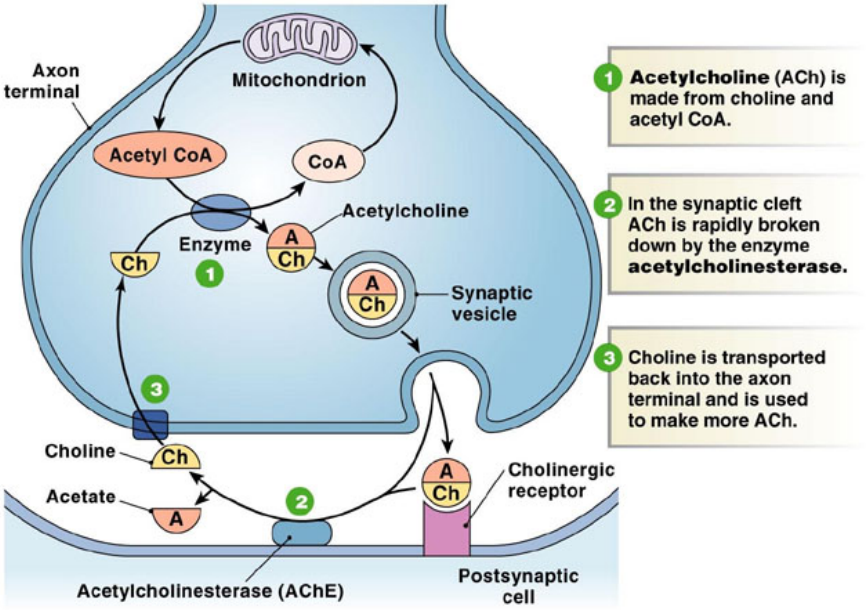

Figure 1.1. Cholinergic hypothesis explaining AD.[18]

Two types of cholinesterase enzymes (ChEs), namely, acetylcholinesterase (AChE) (EC 3.1.1.7) and butyrylcholinesterase (BuChE) (EC 3.1.1.8), are found in the central nervous system. Both of these enzymes belong to the carboxylesterase family of enzymes and play an important role in cholinergic transmission through the hydrolysis of the neurotransmitter ACh. Although AChE and BuChE are produced by different genes, they are highly homologous with more than 65% similarity in their active sites.[19,20] AChE has two major binding subsites, a peripheral anionic site (PAS) and a catalytic active site (CAS).[21] The CAS of the enzyme is actively involved in the maintenance of cholinergic neurotransmission. PAS is involved in the formation of β-amyloid fibrils that are associated with plaque deposition.[22,23] AChE inhibitors blocking both CAS and PAS simultaneously could alleviate the cognitive defects in AD patients by elevating ACh levels, and they have also been endowed with disease-modifying ability by inhibiting amyloid plaque formation.[24] In healthy brains, AChE is more active than BuChE and can hydrolyze about 80% of ACh. Current studies have demonstrated that, as the disease progresses, the ability of BuChE increases by 40–90%, and that of AChE declines in the hippocampus and temporal cortex areas of the brain.[25]

BuChE plays several roles both in neural and non-neural functioning. Clinical data suggested that the high cortical levels of BuChE were associated with some important AD hallmarks, such as extracellular deposition of the Aβ and aggregation of hyperphosphorylated tau protein.[26] This reflects the important role played by both cholinesterases and the necessity to develop novel therapeutics which could act on both of these ChEs. Three of the four FDA-approved anti-AD drugs are acetylcholinesterase inhibitors (AChEIs), which act through inhibition of the AChE.[27]

1.1.1.2. Amyloid hypothesis

Two important pathological hallmarks present in AD patients' brains are extracellular Aβ plaques (senile plaques) and intraneuronal tangles of hyperphosphorylated tau protein (neurofibrillary tangles).[11,12] Both the deposits were described way back a century ago, but Aβ was isolated first time in 1984 by George Glenner, a pathologist from the University of California. Glenner also reported that Aβ is produced from a protein named amyloid precursor protein (APP). APP is an integral membrane glycoprotein that can be cleaved by the proteases in two different pathways i.e., α-pathway (non-amyloidogenic) and β-pathway (amyloidogenic) (**Figure 1.2**).[28]

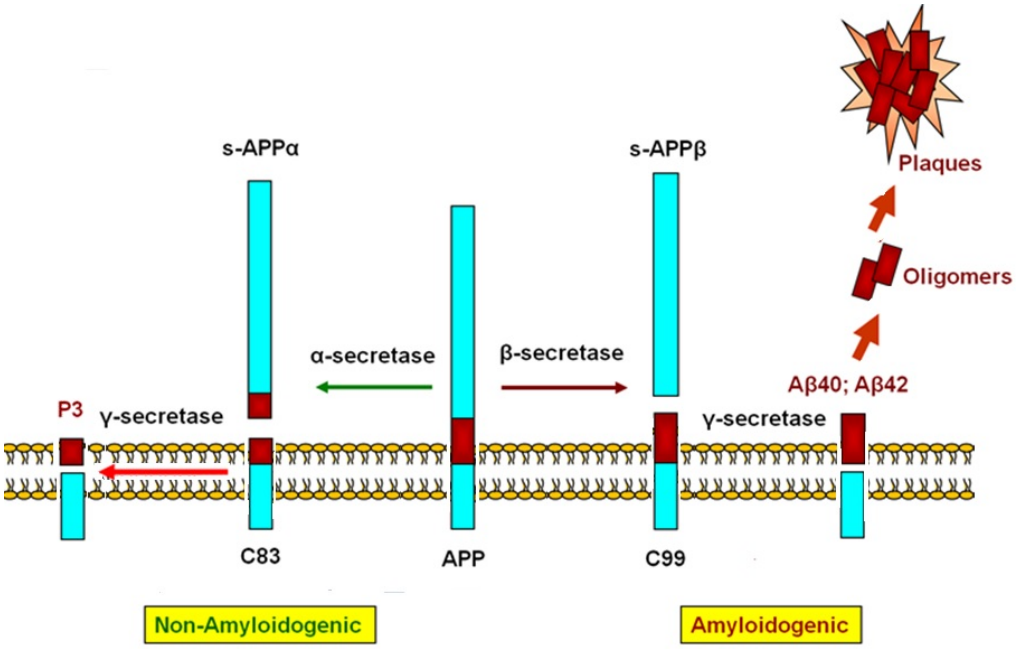

Figure 1.2. Amyloid cascade hypothesis of AD.[29]

In most instances, APP is cleaved sequentially by α- and γ-secretases in the α-path. The APP is first cleaved by α-secretase to produce a soluble

extracellular APP fragment (sAPP-α) and C83 fragment, that is further cleaved by γ-secretase to p3 fragment. The specific physiological role of the p3 fragment has not been completely elucidated yet. As the α-pathway is not involved in the release of Aβ, this path is recognized as the non-amyloidogenic pathway. In the β-pathway, APP is initially cleaved by the β-secretase (BACE-1) to the C-terminal fragment (C99) and a soluble extracellular fragment (sAPP-β). The C99 fragment is further cleaved by the γ-secretase to 38–43 amino acids containing peptides known as Aβ and the intracellular C-terminal domain (AICD). $A\beta_{1-40}$ and $A\beta_{1-42}$ are the two main isoforms of Aβ peptides produced by this amyloidogenic path. $A\beta_{1-40}$ is the predominant product in the proteolytic cleavage, whereas $A\beta_{1-42}$ is more fibrillogenic in nature. These Aβ monomers undergo misfolding and aggregation to form amyloid fibrils and extracellular plaques. These aggregates trigger the pathogenic cascade and eventually result into neuronal loss and dementia.

The Aβ plaques generated from $A\beta_{1-42}$ are neurotoxic and continuously activate inflammatory mediators, such as TNF-α and IL-6. Furthermore, $A\beta_{1-42}$ itself can serve as an oxygen-free radical donor that generates reactive oxygen species (ROS) and directly influences the typical physiological functions of neurocytes.[30] Thus the hypothesis states that there is a fault in the production of Aβ or in the mechanism by which it clears from the brain, or possibly both which ultimately leads to the development of AD.[31]

1.1.1.3. Tau protein hypothesis

Tau protein, a highly soluble microtubule-associated protein (MAP) performs a salient role in regulating the stability and dynamics of microtubules (MTs), axonal transport, and neurite outgrowth in the normal phosphorylation state.[32] The tau protein is regulated primarily by post-translational modifications (PTMs) involving truncation, phosphorylation, acetylation, glycation and methylation. The most common PTM of tau is phosphorylation. Overexpression of GSK-3β modifies the functions and isoform expressions of tau that directs to hyperphosphorylation.[33]

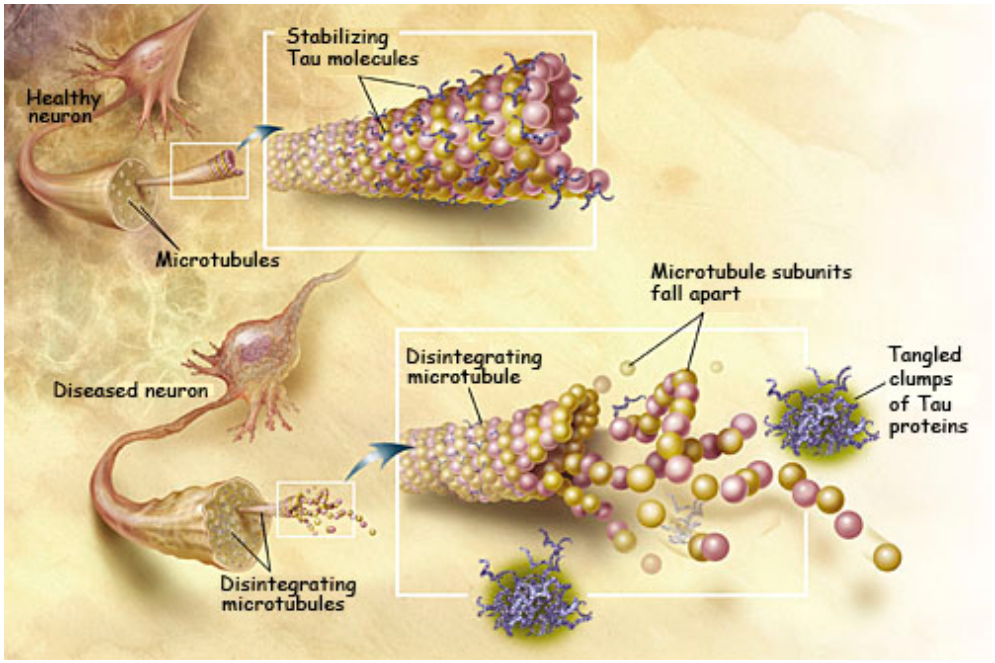

Figure 1.3. Formation of neurofibrillary tangles illustrating tau hypothesis of AD.[34]

In the AD brain, tau is highly phosphorylated, around 3-fold in comparison to the normal brain which leads to the disruption of the MTs and formation of filaments (**Figure 1.3**). Hyperphosphorylated tau disintegrates microtubules and sequesters normal tau, ubiquitin, MAP-1 and MAP-2 into tangles of paired helical filament (PHFs). These insoluble structures alter cytoplasmic functions and impede the axonal transports, and lead to cell death.[35]

1.1.1.4. Oxidative stress

Oxidative stress is identified to be an event that appears prior to all other hallmarks of the AD.[36] Reactive oxygen species (ROS) are generated under normal conditions and their levels are maintained by the delicate balance between the rate of their generation and the rate of their clearance by antioxidants and related enzymes such as superoxide dismutase, catalase and some antioxidant agents like ascorbic acid, glutathione and vitamin E.[37]

In this way, either enhanced ROS generation or weakened antioxidant system framework shifts the cell redox balance to oxidative unevenness and causes ROS overproduction.[37] Additionally, redox-active metal ions i.e., Fe(II/III) and Cu(I/II), in association with Aβ, are reported to generate reactive oxygen species (ROS) by Fenton-like reactions. ROS can denature

biomolecules like proteins, lipids, and nucleic acids. This can induce tissue damage through necrosis and apoptosis (**Figure 1.4**).[38]

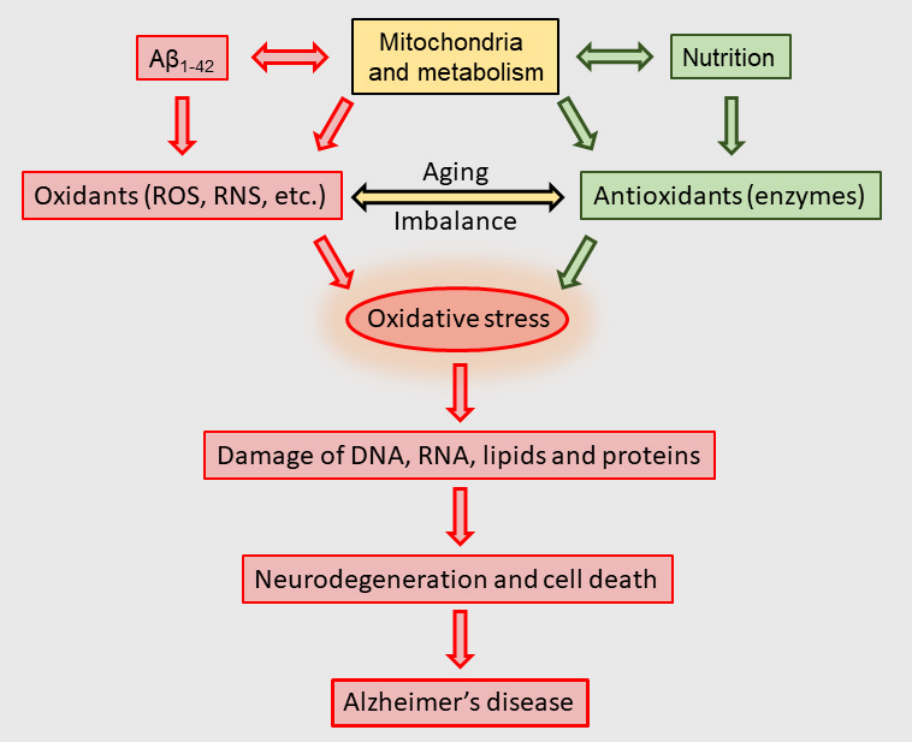

Figure 1.4. Oxidative stress hypothesis.

1.1.1.5. Metal ion hypothesis

Metals are important for the function of enzymes and numerous intracellular signaling proteins.[39] In a healthy individual, the levels of these metals are highly regulated. In the case of normal aging or a neurodegenerative disease state, such homeostatic mechanisms become disturbed, leading to deviation of metal-dependent enzyme functions, mitochondrial dysfunction and the production of ROS all of which are well-known etiologies associated with AD.[40] The metal-mediated Aβ aggregation is caused by the coordination of Cu^{2+} ion to Aβ precursor protein (AβPP) at its amino-terminus leading to the export of neuronal Cu^{2+}. The diminished cellular Cu^{2+} level triggers the expression of AβPP mRNA leading to the overproduction of AβPP. It is further sequentially cleaved by β- and γ-secretase to produce $Aβ_{40-42}$.[41] The Aβ directly binds to copper and zinc, but not to iron or other metal ions. β-Secretase has a Cu^{2+} binding site in its C-terminal cytoplasmic domain by which it interacts with superoxidase dismutase-1.[42] Similarly, γ-secretase activity has been inhibited by low

concentrations of Zn^{2+}.[43] Aβ peptides form complexes with the Cu/Zn metals, which induce toxicity to the neuronal cell in different ways. The metal-Aβ complex increases the Aβ fibrilization which leads the senile plaque formation, increases Aβ oligomerization which impairs the synaptic functions and generates the ROS which induces oxidative stress (**Figure 1.5**).[44]

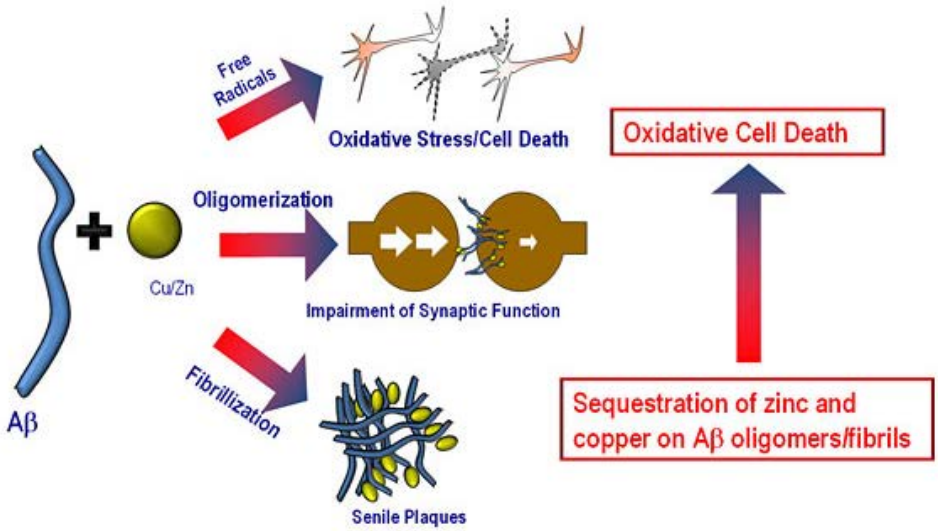

Figure 1.5. Metal mediated toxicity to neuronal cell.[45]

The tau protein is also involved in metal-related variations observed in AD. Tau protein binds to Cu^{2+} in a *p*H and stoichiometric-dependent manner resulting in a conformational change in tau. Hyperphosphorylation of tau protein is also modulated by Cu by activation of GSK-3β and cyclin-dependent kinase (CDK)5/p25 complex and that are crucial in the formation of PHFs.[42,46] This results in aggregation of tau protein thereby causing the formation of neurofibrillary tangles which ultimately leads to cell death.

Apart from the cholinergic, Aβ, tau, oxidative stress, and metal hypotheses, other factors i.e., excitotoxicity,[47] apoptosis,[48] neuroinflammation,[49] etc. are reported to perform critical roles in a complex pathology of AD.

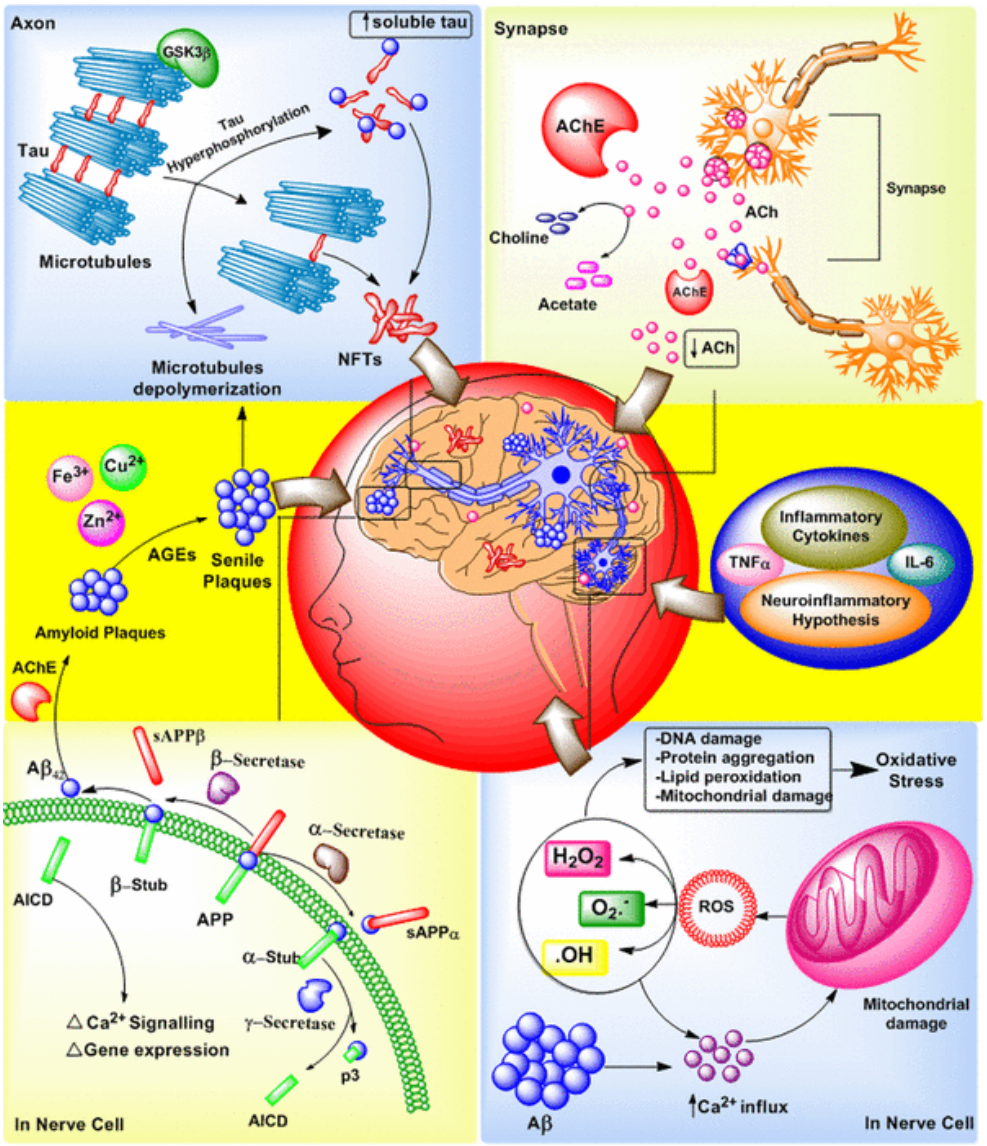

Figure 1.6. An overview of interconnection between various hypotheses of AD.[50]

All these factors discussed above are interconnected to form a complex cellular network and the co-relation between these hypotheses for AD can be represented in **Figure 1.6.** Due to such a complex etiology, AD is categorized as multifactorial or multifaceted disease.[51]