REVIEW ARTICLE

RECORDS OF PHARMACEUTICAL AND BIOMEDICAL SCIENCES

Molecular mechanisms of Alzheimer's disease

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Abstract

Alzheimer's disease is characterized by cognitive decline, progressive neurodegeneration, the development of amyloid βeta-containing plaques and tau hyperphosphorylated neurofibrillary tangles. Initial signs of the neurodegenerative process in Alzheimer's disease include loss of neurons and synaptic impairment. An imbalance between the amounts of amyloid βeta production, aggregation, and clearance leads to amyloid βeta accumulation due to abnormal cleavage of amyloid precursor protein. Furthermore, abnormal tau phosphorylation, which affects tau structure, distribution, and function in neurons, is recognized as a crucial pathogenic mechanism. The primary question driving current research is "What is the primary mechanism leading to the molecular development of Alzheimer's disease pathology?" This review provides an overview of the current knowledge of the molecular processes that result in the production of amyloid βeta, hyperphosphorylation of tau, and amyloid βeta-induced neurotoxicity, including calcium dyshomeostasis, mitochondrial dysfunction, increased oxidative stress, cholinergic dysfunction, and neuroinflammation, which ultimately lead to neuronal dysfunction and neuronal loss.

Keywords: Alzheimer's disease, amyloid βeta, hyperphosphorylated tau, oxidative stress.

1. Introduction

Alzheimer's disease (AD) is the most prevalent type of dementia in elderly people and is defined by an irreversible progression that affects memory, thinking, and behavior. Extracellular amyloid plaques (AP) and intracellular neurofibrillary tangles (NFTs) in the cortex and hippocampus are the two primary histopathological indicators of AD **(Selkoe, 2004)**. The majority of the amyloid βeta (Aβ) peptides, known as senile plaques or AP, are 38 to 43 amino acids long **(Martins et al., 2019)**. The intracellular NFTs, which are the second distinctive pathological hallmark of AD, are made up of an abnormally phosphorylated form of the microtubule-associated protein tau **(Buée et al., 2000)**. NFTs are located inside neurons. In contrast, AP build up extracellularly in the brains of AD patients and induce neuronal damage and cell death **(Mohamed et al., 2011)**.

Multiple hypotheses have been reported explaining AD, including the cholinergic theory, the amyloid hypothesis, and the tau hypothesis. Significant alterations in the brain's cholinergic system, which is linked to cognitive function, have been demonstrated to take place. Acetylcholinesterase (AChE) aggregates in senile plaques with Aβ deposits. Additionally, they promote the creation of harmful Aβ-AChE complexes that are more toxic

than amyloid fibrils. Its consequences include attacks on emotional reactions, difficulties recalling spatial sequences, and effects on learning and memory **(O'Hara et al., 2001)**.

According to earlier studies, there is evidence that oxidative stress (OS), which is particularly sensitive to reactive oxygen species (ROS) in the brain, contributes to AD. The brain also has a weak antioxidant system and fatty acids, which promote peroxidation. Additionally, the brain is more vulnerable to free radicals due to high oxygen consumption **(Uttara et al., 2009)**.

2. Neuropathology of AD

Direct pathologic analysis of brain tissue obtained from an autopsy or biopsy remains the only proven way to diagnose AD in the absence of biologic indicators. Gross cortical atrophy is the typical macroscopic image. Microscopically, the temporal and frontal cortical regions that support cognition are initially more affected than the parietal and occipital cortices due to widespread cellular degeneration and neuronal loss that primarily affects the outer three layers of the cerebral cortex. Reactive gliosis, extensive synaptic and neuronal loss, as well as the appearance of intracellular NFTs and extracellular AP, which are the disease's clinical hallmarks, accompany these alterations **(Masters et al., 2006)**.

3. Molecular mechanism of AD

3.1. Amyloidogenic APP processing

Amyloid precursor protein (APP) is a large Type-I transmembrane protein that is produced sequentially to produce Aβ peptides. APP is a member of an evolutionary conserved protein family that also includes the mammalian APP-like protein 1 (APLP1) and 2 (APLP2), with the Aβ domain being specific to APP **(Jacobsen and Iverfeldt, 2009)**.

A basic cellular function involving β-secretase (BACE) and γ-secretase activity is the proteolytic digestion of APP to produce Aβ peptides. BACE 1 (also known as Asp2 or memapsin2), a membrane-bound protease that is a member of the pepsin family of aspartyl proteases, has been identified as the primary BACE. The cleavage of APP by BACE1, which produces the N-terminus of Aβ, is the initial step in the synthesis of Aβ. In contrast to BACE2, which is expressed at extremely low levels in the majority of brain areas, BACE1 is substantially expressed in neurons **(Bennett et al., 2000)**.

In the AP of AD brains, both peptides—full-length Aβ and N-terminally condensed Aβ peptides—can be detected. Other Aβ isoforms with various lengths at the C-terminus of Aβ are released by γ-cleavage in addition Aβ40, which accounts for 80–90% of Aβ produced and is the most abundant species, and Aβ42 peptides, which account for only approximately 10% of Aβ peptides **(Schieb et al., 2011)**.

The action of γ-secretase is found in a hetero-tetrameric protein complex, according to molecular analysis of the enzyme. γ-secretase complex members include presenilin (PSEN) 1 and 2. Notably, just like the APP substrate itself, every part of the γ-secretase complex is a transmembrane protein. Although all γ-secretase components are required for enzymatic activity, presenilins have the greatest influence on AD development. The well-known early-onset type of AD is caused by more than 150 autosomal dominant point mutations in the presenilins and the production of Aβ-42 peptides is enhanced by these mutations **(Patterson et al., 2008) (Figure 1)**.

3.2. Tau hyperphosphorylation

Neurofibrillary tangles are regarded as another important pathological marker of AD. Insoluble paired helical fragments made primarily of hyperphosphorylated tau protein make up NFTs in neurons. Tau proteins can be detected in axons and the somato-dendritic compartment **(McKibben and Rhoades, 2019)**, and are mostly expressed in neurons. They belong to a group of proteins that are essential for the stabilization of the neuronal microtubule network and the assembly of tubulin monomers into microtubules, both of which are necessary for controlling cell shape and axonal transport **(Nizynski et al., 2017)**.

As a result of hyperphosphorylation and polymerization into paired helical pieces, tau proteins in AD create intraneuronal NFTs **(Schöll et al., 2019)**. Protein kinases and protein phosphatases control tau function and the degree of tau phosporylation. There are already 79 possible serine and threonine phosphorylation sites in tau that have been identified **(Mishan et al., 2019)** and it is thought that the aberrant hyperphosphorylation seen in AD is brought on either by activated protein

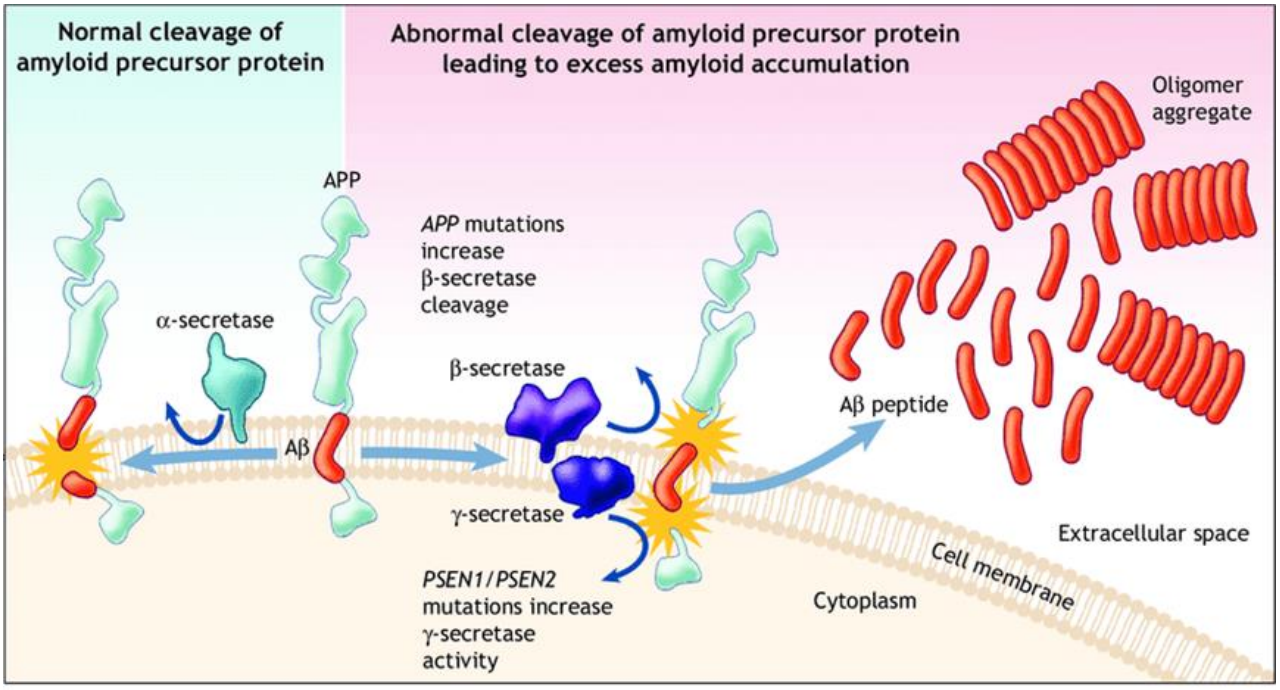

Figure 1: A series of proteolytic cleavage of APP by secretase enzymes **(McNaull et al., 2010)**.

kinases or by decreased protein phosphatase activity.

Hyperphosphorylated tau has been observed to affect intracellular compartments in the somato-dendritic compartment by fragmenting the Golgi apparatus, decreasing mitochondria, and diminishing rough endoplasmic reticulum (ER). Due to ongoing ER stress, hyperphosphorylated tau buildup in the ER may contribute to neurodegeneration **(Kim et al., 2008)**.

3.3. Disruption of calcium homeostasis

Neuronal vitality and function depend on intracellular calcium (Ca^{2+}) signaling. Neurons at rest have precise mechanisms to maintain low Ca^{2+} concentrations in the cytosol by extruding Ca^{2+} into the extracellular space and pumping Ca^{2+} into the ER **(Verkhratsky et al., 2004)**. This is because Ca^{2+} is involved in second-messenger signaling. When G-protein coupled receptors are stimulated, more inositol-1,4,5-trisphosphate (IP3) is produced, which binds to IP3 receptors on the ER and releases Ca^{2+} from the ER into the cytosol. Ca^{2+} can also flow through voltage-, ligand-, or store-operated channels from the extracellular space into the cytosol upon activation. Neuronal cellular signaling pathways that are involved in membrane excitability, neurotransmitter release, gene expression, cellular growth, differentiation, the generation of FR species, and cell death are activated by elevated cytosolic Ca^{2+} levels **(Fedrizzi and Carafoli, 2011)**.

An essential aspect of AD pathophysiology is Aβ-induced Ca^{2+} dyshomeostasis, which modifies the characteristics of neurotransmitter receptors, compromises membrane integrity, and initiates signaling cascades that lead to cell death, synaptic degeneration, and severe memory loss. It has been demonstrated that Aβ interacts with various Ca^{2+}-permeable channels and boosts Ca^{2+} entrance into the cytosol **(Rovira et al., 2002)**. In neurons, voltage-gated cell membrane Ca^{2+} channels constitute a key mechanism for cytosolic Ca^{2+} entry. Voltage-gated Ca^{2+} channels of the L-, N-, and P-type are activated by Aβ greatly increasing Ca^{2+} inflow and raising postsynaptic responses **(MacManus et al., 2000)**.

The hippocampus and neocortex, which require higher cognitive abilities, express these receptors, and these brain regions are the ones most significantly impacted by the neuronal loss seen in AD. N-methyl-D-aspartate (NMDA) receptor activity has been demonstrated to be modulated by Aβ oligomers **(Ye et al., 2004)** and NMDA receptor-mediated Ca^{2+} influx has been increased, leading to dynamin-1 breakdown and boosting calpain activity, both of which are crucial for the production and operation of synaptic vesicles. However, it has also been noted that Aβ oligomers

decrease the expression of NMDA receptors and consequently Ca^{2+} influx **(Dewachter et al., 2009)**.

3.4. Cholinergic dysfunction

Deregulation of the Ca^{2+}-permeable n-acetylcholine-receptor (nAChR) channels also leads to the breakdown of synaptic integrity. It has been shown that neurons with high levels of nAChR expression, particularly those with the 7 subunits, are the most sensitive to AD. In cortical and hippocampal synaptic membrane preparations, Aβ has been demonstrated to bind to 7- and 4-nAChRs and to influence nAChR functionality. Additionally, the 7-nAChR co-localizes with AP, and the 4- and 7-nACHRs positively correlate with neurons that accumulate Aβ. The development of pharmaceutical drugs that target cholinergic molecular components, primarily the breakdown of acetylcholine by AChE **(Arneric et al., 2007)**, was prompted by the dramatic degeneration of cholinergic neurons in AD and the loss of cholinergic neurotransmission, which contributes to cognitive decline in AD **(Alvarez-Jimenez et al., 2016)**.

3.5. Decreased membrane integrity

Amyloid βeta-induced loss of the integrity of the membrane's lipids results in a similar unidirectional flux of cations in neuronal plasma membranes. Aβ-peptides interact with additional membrane lipids. This alteration in membrane fluidity is thought to increase membrane permeability to Ca^{2+}, Na^{+}, and K^{+} ions **(Müller et al., 1995)**. Regardless of how Aβ causes Ca^{2+} dyshomeostasis in AD, the disruption of intracellular Ca^{2+} signaling ultimately leads to synapse deterioration, cell death, and irreversible memory loss **(McLaurin and Chakrabartty, 1996)**.

3.6. Increased OS and mitochondrial dysfunction

Destabilization of cytosolic Ca^{2+} levels enhances the generation of ROS, which leads to lipid peroxidation and the oxidation of proteins in addition to altering neurotransmitter receptor characteristics and inducing apoptosis **(Bernardi et al., 2006)**. The production of ROS can be increased when the cytosolic Ca^{2+} level rises since this can result in an excessive Ca^{2+} flow into the mitochondria. Aβ has been discovered to accumulate in mitochondria, leading to increased hydrogen peroxide production and decreased cytochrome-c oxidase activity, which results in mitochondrial oxidative damage, dysfunction, reduced energy metabolism **(Manczak et al., 2006)** and cell death.

This accumulation of Aβ has been studied in transgenic mouse models and neuronal cell cultures. Importantly, it is well known that OS itself causes an increase in the production of Aβ **(Apelt et al., 2004; Tong et al., 2005)**, highlighting the fact that OS plays a significant role in the pathogenesis of AD.

3.7. Neuroinflammation

Another mechanism of Aβ neurotoxicity is the stimulation of immunocompetent brain cells (microglia and astrocytes) by Aβ, which causes neuroinflammation. Although it has been demonstrated that Aβ activates microglia, the precise mechanism of this activation is still unclear. **(Kauppinen et al., 2011)**.

Using microglial cultures, it was shown that PARP-1 activity is essential for Aβ-induced NF-κB activation, morphological transformation, nitric oxide release (NO), tumor necrosis factor-α (TNF-α) release and neurotoxicity. Treatment of astrocytes with oligomeric Aβ increased the generation of the major pro-inflammatory cytokine interleukin-1, NO and TNF-α in rat astrocyte cultures and led to increased astrocyte-mediated inflammation **(White et al., 2005)**. Furthermore, oligomeric Aβ correlates with neuronal loss and astrocyte inflammatory response in APP/Tau transgenic mice and astrocytes are important mediators of Aβ-induced neurotoxicity and tau phosphorylation in primary cultures **(Garwood et al., 2011)**.

Similar to the feed-forward cycle that Aβ induces OS and increased OS itself elevates Aβ generation, Aβ triggers neuroinflammation, which in turn increases Aβ generation **(Heneka et al., 2010)**. Additionally, OS triggers the release of proinflammatory cytokines, chemokines, and complements, which activate microglia and astrocytes **(Candore et al., 2010)**, highlighting the way in which the various mechanisms of Aβ toxicity interact and the fact that AD is a multifactorial neurodegenerative condition **(Figure 2)**.

4. Conclusion

Alzheimer disease is a neurological condition marked by memory loss and slow but constant

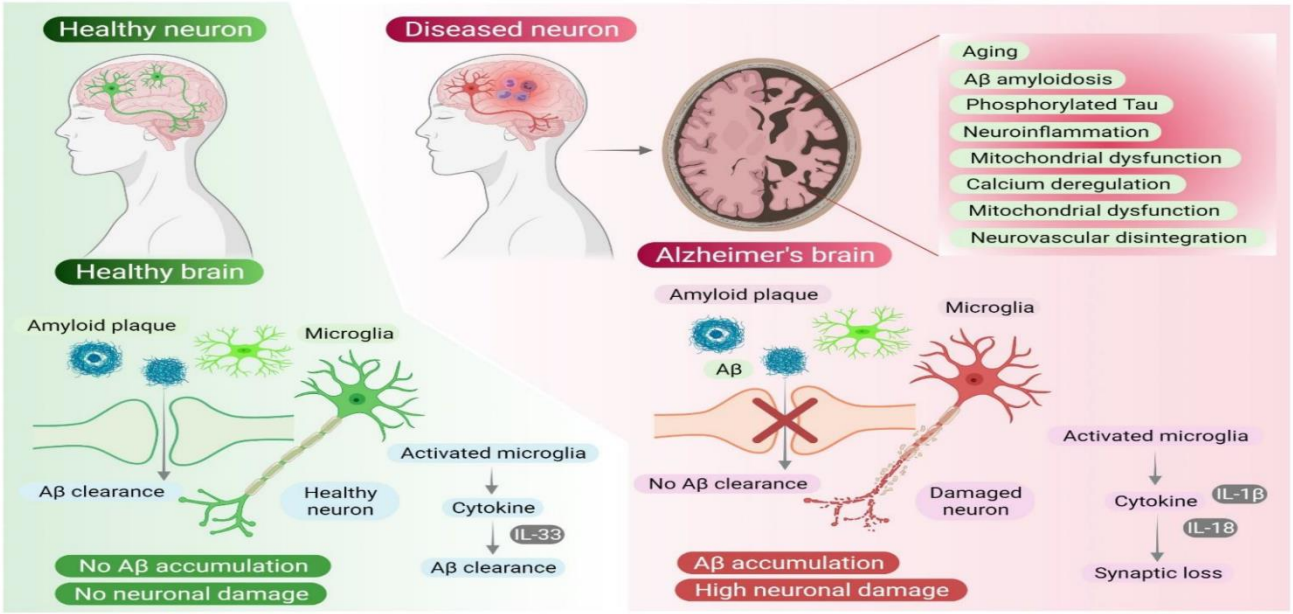

Figure 2: Major pathological hallmarks of AD **(Prasanna et al., 2021)**.

decline in cognitive function. It has a terrible impact on the patient and a significant socioeconomic effect. Extracellular Aβ deposits in senile plaques, NFTs, and specific neuronal and synaptic loss in cortical regions of the brain linked to cognitive and memory functions are the neuropathologic characteristics of the disease. The primary element of AP is Aβ. The breakdown of Aβ homeostasis plays a critical role in the etiology of AD. The pathogenesis and course of AD have been clarified through genetic investigations. Understanding the connection between tau and amyloid pathology and identifying the mechanism behind AD development will require more study into the cellular function of tau proteins, particularly APP.

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