Biomarkers for Alzheimer’s Disease and Potential Future Directions

Usman Zafar Paracha1*, Ishtiaq Qadri2, Khezar Hayat1, Talib Hussain1

Abstract: Alzheimer’s disease (AD) is a degenerative disorder causing dementia in the later stages of life. Several biomarkers have been established for the diagnosis and prognosis of AD. Among the significant biomarkers are amyloid beta (Aβ) plaques and tau proteins. However, many biomarkers are still controversial in their findings of the level in the AD progression and severity. In this paper, we have discussed the biomarkers that can help in the diagnosis and prognosis of AD. Our primary emphasis was on to work on the future directions that can strengthen research.

Keywords: Alzheimer’s disease, Biomarkers, diagnosis and prognosis

1. Introduction

Alzheimer’s disease (AD) is a degenerative disorder that affects the brain and causes dementia, especially in the later stages of life [1]. Progressive memory loss, language and cognitive disorders in both visuospatial skills and executive functions are usually found in the patients of AD [2]. About 5.2 million Americans have AD including 5 million people above the age 65 and about 200,000 individuals below 65 years of age [3].

Bateman et al. [4] found that pathophysiological changes related to AD could start about 25 years before the start of dementia. Risk genes for AD have also been found to be linked to brain changes at birth, i.e. for some variations in AD related genes such as apolipoprotein E (ApoE) gene, the brain changes in infants were found to be very similar to the brain changes found in adults having the same variants [5].

Usually, measurements of tau, phospho-tau (p-tau), and Aβ levels in cerebrospinal fluid (CSF) through a lumbar puncture and plasma are done to get information about the disease in the patients [6, 7]. With the advancement of age, these proteins either disturb the signaling between the neurons or completely destroy them. Not only single protein but the combinations or ratios of one protein to another can also help in determining the deteriorations caused in the brain. Raven et al. have recently found that besides Aβ and tau, iron accumulation in hippocampus is also the cause of tissue damage [8].

2. Biomarkers

Many structural and functional biomarkers have been identified and various novel techniques have helped scientists to work on the problem of early diagnosis of AD.

Slight cognitive changes/mild cognitive impairment (MCI) shows higher risk of developing AD as it is considered as a transition phase between normal aging and AD [9].

Vos et al. [10] have proposed a classification system according to which preclinical AD can be represented by the initial decline of Aβ in the spinal fluid showing the formation of plaques in the brain. While the levels of Aβ continue to decline, levels of tau protein begin to rise in the spinal fluid showing the start of death of brain cells. During these disturbed amyloid and tau biomarkers, slight cognitive changes can be detected by neuropsychological testing.

2.1 Structural Markers

Studies showed that whole brain atrophy is elevated in early AD, from two times [11] to five times [12]. Neuropathological studies suggested an increase in neurofibrillary tangles [13] and huge loss of neurons in the hippocampus and entorhinal cortex of AD patients [14]. De Leon et al. [15] have reported atrophy of medial temporal regions, where AD can be diagnosed early in the disease, by using computed tomography (CT).

Deeper look into the finding shows that the accumulation of plaques is promoted by the disruption of neuronal communication that is the result of destruction of myelin, which is the fatty tissue covering the nerve fibers in the brain. Myelin is made up of oligodendrocytes that have a huge amount of iron and this is one of the reasons that the disturbance in iron levels is an important factor in AD. Magnetic resonance imaging (MRI) helped researchers in determining the hippocampal damage as a result of iron accumulation [8].

Zhu et al. [16] have also found an increase in myo-inositol levels in the parietal lobe gray matter (GM) regions.

2.2 Functional Markers

Many of the biomarkers and functional changes in the brain have been identified. Among the novel techniques Magnetic resonance imaging (MRI), functional MRI (fMRI), electroencephalography (EEG), Positron emission tomography (PET) and CT have helped a lot.

EEG is helpful in determining the functional connectivity in the brain and Keeser et al. [17] found that it could be an important marker for determining the cognitive decline in the very early stages of neurodegenerative disorders. fMRI showed disturbances in the brain activation of different parts in AD. Dickerson et al. found decreased activation of hippocampal and entorhinal regions in the patients of AD using fMRI [18]. Kato et al. [19] found lack

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of activation of temporal lobe or prefrontal regions in AD. Sperling et al. [20] found elevated activation in the medial parietal and posterior cingulate regions. With the help of 2-[18F] fluoro-2-deoxy-D-glucose (FDG) PET, researchers found severe reduction of glucose metabolism in bilateral parietal and posterior cingulate cortices and precuneus region in early onset AD patients [21, 22].

Doecke et al. [23] have reported a number of biomarkers for the diagnosis of AD, which are presented in the table 1. Some of the significant biomarkers are described below.

Table 1: Noteworthy markers of Alzheimer’s disease

<table>
<thead>
<tr>
<th>Structural Markers</th>
<th>Functional Markers</th>
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<tbody>
<tr>
<td>Elevated brain atrophy</td>
<td>Aβ40 and Aβ42 peptides that make up the plaques are present in AD pathogenesis.</td>
</tr>
<tr>
<td>Neurofibrillary tangles</td>
<td>Aggregation of microtubule-associated tau proteins</td>
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<tr>
<td>Loss of neurons in the hippocampus and entorhinal cortex of AD</td>
<td>Increased myo-inositol levels in the parietal lobe gray matter (GM) regions</td>
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<td></td>
<td>Increased Isoprostanes levels, especially F2-isoprostanes, in the frontal and temporal cortex of AD</td>
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<tr>
<td>ACT levels increase in plasma and CSF</td>
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Aβ fragment signature consisting of Aβ1-16, Aβ1-33, Aβ1-39, and Aβ1-42 as analyzed by Matrix-Assisted Laser Desorption/Ionization Time-of-Flight MS (MALDI-TOF-MS) are found to distinguish sporadic AD patients from non-demented controls with a sensitivity of 89%, specificity of 83%, and accuracy of 86% [33].

Hyperphosphorylation and aggregation of microtubule-associated tau proteins with formation of tangles also represent the progression towards AD [34, 35]. Fagan et al. [29] have found increased levels of CSF tau and phosphorylated tau in AD.

Ratios of the potential markers such as tau/Aβ [32], tau/Aβ42 [29, 32], p-tau181/Aβ42 [29] and the CSF tau/Aβ42 ratio can also be used to predict the future dementia [36]. McMillan et al. [37] have recently found that MRI can predict the tau / Aβ42 ratio with 75% accuracy and therefore, it can be used as a substitute for CSF biomarkers selected from each section. In the randomly selected fields of vision, the numbers of the positive nerve cells were counted by the high power lens.

2.2.2 Isoprostanes

Researchers reported increased levels of isoprostanes, the end-products of lipid peroxidation, specifically F2-isoprostanes, in the frontal and temporal cortex of AD [38, 39]. Researchers have also found increased CSF F2-isoprostanes in MCI and AD patients [40-42]. Its measurements could accurately distinguish individuals, who can progress to MCI or AD from the people, who are stable [42]. Montine et al. used the combined analysis of CSF Aβ42, tau, and F2-isoprostanes and found the sensitivity of 84% and specificity of 89% in the diagnosis of AD [43].

2.2.3 α1-antichymotrypsin

α1-antichymotrypsin (ACT) is a serine protease inhibitor found to be colocalized with Aβ in senile/neuritic plaques [44]. DeKosky et al. found that ACT levels increase in plasma and CSF, and the levels are negatively correlated with dementia severity [45].

2.2.4 Neuronal system disturbances

Neuronal network and neurotransmitter systems failure are among the most important disturbances found in AD [7]. Selkoe et al. [46] found that synaptic disturbance and its loss results in deterioration of synaptic strength and plasticity, and these synaptic disturbances...
occur before the development of Aβ plaques and neurofibrillary tangles.

Disturbances in acetylcholine, noradrenaline, dopamine and serotonin neurotransmitter systems have primarily been found in AD [47]. Tryptophan biosynthesis has also been found to be disturbed in the plasma and CSF of the patients of AD resulting in changes in serotonin/melatonin pathway in CSF of AD patients [7].

Acetylcholine pathway in CSF of AD patients has also been found to be changed. Among the other pathways affected equally in CSF and plasma of AD patients are beta-alanine, aspartate and asparagine, alanine, L-cysteine, L-methionine, methionine-cysteine-glutamate along with L-arginine and lysine metabolism. Moreover, disturbances in the lysine metabolic pathways could accurately help us in differentiating cognitively normal people and the patients of AD [7].

Neurotransmitter norepinephrine (NE) and its major metabolite 3-methoxy-4-hydroxy phenylglycol (MHPG) are also changed in the disease [48]. One of the reasons for this change of NE is the neuronal loss of locus ceruleus in the patients of AD [49]. In one of the studies, Czech et al. [50] reported increased levels of NE in the CSF of AD patients while in another study, Kaddurah-Daouk et al. [51] found a decrease in NE levels and an increase in MHPG levels in the CSF of AD patients.

Amino acid N-acetyl aspartate (NAA) is present in neuron bodies, axons and dendrites, therefore it can be an important marker for neuronal disturbances [52]. Zhu et al. [16] have found the decrease in NAA levels independent of brain atrophy in the GM regions.

2.2.5 Lipids and Sterols

Lipid biosynthesis and metabolism related pathways could help us in distinguishing MCI from AD as these are increasingly disturbed in AD patients relative to MCI [7]. Gamba et al. [53] found that disturbances in the cholesterol metabolism and hypercholesterolemia could have an important role in amyloid plaque formation and tau hyperphosphorylation.

Disturbances have also been found in the levels of phosphatidylcholine, plasmalogens, sphingomyelins and sterols in plasma of AD patients [9, 54-56]. Farooqui et al. [57] found that the level of ethanolamine plasmalogens is decreased while the level of serine glycerophospholipids is significantly elevated in the plasma membrane phospholipid in the brain in AD.

Mielke et al. [58] also evidenced that sphingomyelins/ceramide ratio and dihydrospingomyelin/dihydroceramides ratio could become one of the potential blood-based biomarkers for clinical progression of AD. Sato et al. [59] found a decline in the level of desmosterol and the desmosterol/cholesterol plasma ratio in AD. Desmosterol could be one of the sensitive biomarker for early and easy detection of AD.

Trushina et al. [7] found variations in the CSF PGE2 biosynthesis and metabolism with AD severity. In one of the studies, Combrinck et al. [60] showed a decrease in PGE2 levels with more advanced AD while PGE2 levels are higher in patients with mild memory impairment.

Czech et al. [50] found increased cortisol levels in CSF and plasma of patients with severe forms of AD. Trushina et al. [7] found that the pathways related to the cortisol biosynthesis from cholesterol disturb significantly, in both the CSF and plasma of AD patients.

3. Future Directions

Recent advances in AD biomarkers have increased the ability for early diagnosis of the disease. However, the relationship of presently known biomarkers with the cause of the disease needs further research as the molecular mechanism of the disease is not clear [7]. Moreover, Vos et al. [10] have found that the people with preclinical AD have about six times more chances of dying over the next decade than older adults without any conditions of preclinical AD. However, the reasons behind this phenomenon are not clear.

Loss of weight and lowered lipid levels are common in AD patients. Further investigations are required to determine the specific lipid subclasses that are affected with the progression of AD. Researchers found that obesity and type-II diabetes related pathways are uniquely affected in the patients of AD showing their potential ability to provide biomarkers for early detection of AD.

Avarahi et al. [1] have reported that lysosomal acidity restoration could help in diminishing the symptoms of AD as it would result in decreased accumulation of Aβ plaques. However, among other studies no decrease in cognitive and functional decline in the patients of AD has been found even with the maintenance of amyloid burden and decrease in p-tau levels in CSF [7]. As the accumulation of iron is found to be one of the important causes in the development of AD [8], research can be done on the dietary and environmental factors affecting iron levels in the brain such as the consumption of red meat and iron supplements in the diet and their possible role in the development of AD.

Suh et al. [26] are of opinion that ADAM10 activity is important in controlling AD risk factors. However, its activity, molecular structure and mechanism of ADAM10 enzyme to maintain AD risk factors need further research. Further investigations are required to understand the exact mechanisms involved in disturbed Aminoacyl-tRNA synthetases (AARS) pathway in AD [7].

Researchers have reported different findings of APP and its secreted forms in the patients of AD. Some studies showed increased levels [61, 62] of APP while the other studies showed decreased [63, 64] or unchanged levels [65] of APP. Further investigations have to be carried out to resolve this conflict.

There are many other controversial findings about the level of Aβ1-42 peptides, F2-isoprostanes in the plasma and urine, CSF NE and interleukin-6 (IL-6). Resolving these controversies could help in significant advancement in the research of AD diagnosis and therapy. Trushina et al. [7] have reported that the differences in findings of CSF NE levels could be due to post mortem changes, origin of CSF samples or the utilization of different metabolomics platforms.

β2-Microglobulin, the constant component of the class I major histocompatibility complex, has been found to be increased in some studies such as by Carrette et al. [66] and...
decreased in the other studies [67]. Further investigations are required in this regard. Similarly, mixed results have been obtained for transthyretin, requiring further research. Transthyretin is found to have a role in AD pathogenesis by preventing Aβ aggregation [68].

The ability of several metabolites such as choline containing compounds, creatine and phosphocreatine, and glutamate and glutamine as the biomarkers for AD needs further investigation. In research of AD, we can take help of combination of different techniques of brain scanning as for example the combination of PET and MRI [69] and/or the combination of fMRI and EEG [70].

The mechanism that results in AD due to reduced mitochondrial DNA (mtDNA) levels in CSF needs further investigation. Moreover, further research is required to find whether AD could be treated or not before the symptoms appear with the blockage of this decrease in mtDNA.

Oresic et al. have found that hypoxia and oxidative stress may show progression towards AD [9] and in other studies, we have also reported a clear link between oxidative stress resulting from Hepatitis-C virus (HCV) [71]. Investigations can be done in the viral linkage to Alzheimer’s disease.

As a conclusion, we can say that further biomarker investigations are required not only to better understand the mechanism of AD but also to treat the disease at an early stage as well as at an advanced stage.

Conflict of interest: None declared.

References


