Review

The Immunological Aspects and Anti-Amyloid Strategy for Alzheimer's Dementia

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Alzheimer’s dementia (AD) is the most common form of dementia among the elderly, accounting for at least two-thirds of all dementia cases. Its global prevalence is estimated at 24 million cases, which certainly represents a costly burden. Amyloid beta or Ab plaques and neurofibrillary tangles define AD pathologically but do not fully explain this disease; it also involves inflammation as well as neuronal, axonal, and synaptic loss and dysfunction. Amyloid plaques are surrounded by activated microglia, cytokines, and complement components, which is suggestive of inflammation, an important component of AD pathophysiology. For the diagnosis of AD, cerebrospinal fluid markers, especially in vivo amyloid measurements, contribute to an accurate assessment of AD pathology and differential diagnosis. Aβ levels are a very good marker for the presence of amyloid deposits in the brain, while total tau and phosphorylated tau are useful for the detection of neurodegeneration. The implementation of anti-amyloid therapy and other disease-modifying interventions may have immense clinical impact if initiated at an early or presymptomatic stage of AD, i.e. before significant brain damage occurs. This paper briefly reviews the abovementioned topic and provides recommendations for future studies.

KEY WORDS:
Alzheimer's disease; amyloid-beta (Aβ) plaques; inflammation; microglia; anti-amyloid therapy
Alzheimer’s dementia (AD) is a progressive degenerative disease of the brain and the most common cause of dementia among elderly persons, accounting for at least two-thirds of all dementia cases (1, 2). The global prevalence of dementia is estimated to be as high as 24 million cases, and is predicted to double every 20 years by 2040, which will undoubtedly lead to a high and costly burden (3). AD is characterized as a progressive decline in cognitive functions, which typically begins with memory impairment and a characteristic change in personality and executive functions. Before death, individuals usually become dependent on caregivers. There is no available cure.

The neuropathological hallmarks of an AD brain include diffuse and neuritic extracellular plaques composed of amyloid beta (Aβ) peptides (Fig. 1), frequently surrounded by dystrophic neurites and intraneuronal neurofibrillary tangles (NFT), which are in turn composed of paired helical filaments or phosphorylated tau proteins (4). These hallmark pathologies are often accompanied by reactive microgliosis and neuron and synapse loss. The etiological mechanisms underlying neuropathological changes in AD still remain unclear, but are probably affected by environmental, genetic, and neuroinflammatory factors (3).

Up to 3% of AD cases are caused by an autosomal dominant mutation with three genes identified thus far: the amyloid precursor protein (APP), presenilin 1 (PSEN 1), and presenilin 2 (PSEN 2) (5). The Aβ peptide is cleaved from the APP by the sequential activities of β-secretase and γ-secretase enzymes. Aβ occurs in multiple forms, including those ranging from 37 to 43 amino acids in lengths. Among these, Ab$_{42}$ seems to be essential for initiating Aβ aggregation and is considered central to the amyloid cascade hypothesis of AD (6, 7). The APOE ε4 allele is the most important genetic risk factor for sporadic AD (8).
Inflammation mediated by activated microglia is an important component of AD pathophysiology (9) and neuritic plaques are the foci of the local inflammatory response (10). The Aβ neuritic plaques are surrounded by activated microglia, cytokines, and complement components, which may serve to indicate the „inflammatory foci”. In contrast, diffuse plaques without reactive microglia are considered clinically benign Aβ deposits. These objectively measured AD biomarkers are therefore useful for the diagnosis, longitudinal assessment, and evaluation of the subsequent therapeutic response. One of the most frequently used biomarker Aβ is produced in the brain and secreted into the brain’s extracellular space. Measurement of cerebrospinal fluid (CSF) amyloid peptide levels can indicate the extent of peptide generation and clearance in the brain, particularly in radio-labelled amino acid infusion studies (11). The mean concentration of Aβ42 in the CSF was reduced by as much as 50% in AD subjects relative to age-matched controls (12), as the result of Aβ42 deposition in amyloid plaques, preventing its transit from the brain into the CSF. In support of this hypothesis, all individuals with Aβ deposits show low concentrations of Aβ42 in the CSF regardless of their cognitive status (13-15). However, CSF Aβ42 does not correlate well with disease duration or severity (6). This is consistent with 11C-labeled Pittsburgh Compound B (11C-PIB) results from a study that showed that amyloid retention does not change significantly during the symptomatic stages of AD (16), and further supports the finding that amyloid pathology occurs very early in the process of this disease(6). Thus, CSF Aβ42 can serve as a diagnostic and surrogate biomarker for Aβ deposition in the brain. The decrease in CSF Aβ42 appears to precede amyloid retention as detected by amyloid imaging using 11C-PIB , signifying what was perhaps the first evidence of AD pathology in cognitively normal individuals (13, 15, 17).

Tau is a major protein component of, at least initially, intraneuronal NFT and is elevated in the CSF of most AD patients. In addition, it has been shown that tau levels in the CSF can
rapidly increase following neuronal injury, indicating the severity of the underlying neurodegeneration (18). Tau, as a biomarker of neuronal injury, can be transiently increased after any acute brain injury, such as a stroke or physical trauma (19). Levels of abnormal phosphorylated tau are being effective in differentiating AD from nondemented controls (20) and other dementia, e.g. fronto temporal dementia (21) and dementia with Lewy body or DLB (2, 15). Distinguishing DLB from AD is a major clinical challenge due to different optimal management, allowing initiation of effective pharmacotherapy, and avoiding the consequences of neuroleptic sensitivity.

The deposition of Aβ peptides drives cerebral neuroinflammation by activating microglia. Indeed, Aβ activation of the NLRP3 inflammasome in microglia is fundamental for interleukin-1β (IL-1B) maturation and subsequent inflammatory events. However, it remains unknown whether the NLRP3 inflammasome, which leads to the production of the pro-inflammatory IL-1B, thus indicating an inflammatory process, plays a role in AD progression (22). On an animal model, behavioural and cognitive functions were improved by reducing the signalling from this particular inflammasome, making inflammasomes a promising therapeutic target for AD therapy (22). Chronic inflammation coupled with neuronal ageing induces cellular stress and concomitant impairments in basic neuronal functions. The beneficial side of the inflammation immune system against amyloids is the reduction of Aβ and the immunization against Aβ.

Today’s anti-amyloid strategies include immune therapies and vaccines to clear amyloids out, plaque busters, and amyloid clearance enhancers. These drugs are designed as an antibody to bind Aβ. The stimulation of Aβ clearance from the brain of AD patients via the administration of Aβ antigens (active vaccination) or anti-Aβ monoclonal amyloid antibodies (passive vaccination) has already been applied. In 2001, the first clinical trial with the Aβ peptide active vaccine (AN1792, consisting of preaggregate Aβ and an immune adjuvant, QS-21) was initiated. After the second dose, however, meningoencephalitis occurred in 5-6% of immunized patients, and the trial was stopped.
The clearance of Aβ from the human brain, however, was successful (23), suggesting that the immune response generated against the peptide elicited the clearance of Aβ plaques. Nowadays, passive immunotherapy with anti-amyloid substances is in development. **Bapineuzumab** (**β-amyloid peptide** (**i**) **neutralizing** (**u**) **monoclonal antibody**), was designed to bind to Aβ in patients with mild-to-moderate forms of AD, but all phase III clinical trials of bapineuzumab have been halted in 2012, because the drug did not meet the endpoints for cognition and global function ([http://www.mmm-online.com](http://www.mmm-online.com)). Also, the occurrence of vasogenic oedema after bapineuzumab, and more rarely brain microhemorrhages (especially in Apo E ε4 carriers), has raised concerns about the safety of these antibodies directed against the N-terminus of Aβ. Recently, **solanezumab** (**soluble amyloid neutralizing** (**u**) **monoclonal antibody**), a humanized anti-Aβ monoclonal antibody directed against the Aβ peptide, was shown to neutralize soluble Aβ species, thus becoming the first therapeutic drug to be evaluated in the anti-amyloid treatment in asymptomatic AD ([http://www.alzforum.org](http://www.alzforum.org)). This was the first application of an Aβ clearing drug in older people thought to be in the presymptomatic stage of AD with evidence of amyloid in their brains shown by 11C-labeled PIB imaging (24), but with no clinical symptoms of the disease yet. The clearance of plaques, however, has not yet been shown to reverse memory in clinical trials, even though many scientists in the field feel that immunotherapy holds promise. The failure of bapineuzumab to produce benefit has aroused some scepticism about the amyloid cascade hypothesis (3), which holds that toxic amyloid protein cleaved from the APP initiates AD (Fig. 2).

Recently, a coding mutation (A673T) found in the **APP** gene confers strong protection against AD even in people who carry the APOE4 gene, therefore demonstrating the importance of keeping Aβ levels in the brain low (1). The discovery of the protective APP mutation in approx. 1% of Icelanders suggested that lowering amyloid in the brain, either with anti-amyloid therapy or some other treatment must begin long before the symptoms of AD set in. As a result, the levels of Aβ in the CSF began to decline 25 years before the expected onset of symptoms, which is a signal that the protein is being sequestered in the brain as insoluble plaque, as shown in the group of 128 participants in the prospective, longitudinal Dominantly Inherited Alzheimer Network (DIAN) study with early onset AD(25). Several grants for Alzheimer’s prevention studies were awarded by the N.I.H. in order to test anti-amyloid drugs on members of a Colombian family, the largest extended family in the world with a gene mutation that causes them to develop AD early, showing cognitive impairment by around age 45, and full dementia a few years later.
So far three investigational monoclonal antibodies against amyloid for clinical trials have been selected that will try to prevent dementia in people who are on the path to AD due to an inherited autosomal-dominant mutation (www.nia.nih.gov/alzheimers/clinical trials).

In the majority of cases (97%) involving late-onset AD caused by a number of risk-factor genes, identifying these genes will be the greatest advancement towards effective treatment. Based on the new diagnostic criteria of AD (24, 26), and recent experience with major failures of anti-Aβ drugs in mild-to-moderate AD patients, one could argue that clinical trials on potential disease-modifying drugs, including immunological approaches, should be performed in the early or preclinical stages of AD, years before the symptoms occur (27). For anti-amyloid therapeutic programs, it seems appropriate to select individuals for trials on the basis of amyloid PET imaging and/or low CSF Aβ42.

In summary, despite tremendous investments in basic and clinical research, no cure or preventive treatment for Alzheimer’s dementia (AD) exists. The amyloid-beta (Aβ) peptide has become a major therapeutic target in AD. The genetic mutations cause increased Aβ levels, followed by amyloidosis, tauopathy, brain atrophy, and decreased metabolism. Inflammatory processes are strongly correlated with the onset and progression of AD in humans, and could have a pivotal role in the AD aetiology. Based on the new diagnostic criteria and recent experiences with anti-Aβ drugs in early stage AD patients, one could argue that the treatment for AD patients should start years before the first onset of clinical symptoms.

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References:


**Sažetak**

**IMUNOLOŠKI ASPEKTI I ANTIAMILOIDNE STRATEGIJE ZA ALZHEIMEROVU DEMENCIJU**

Alzheimerova bolest (AB) je najučestaliji oblik demencija u starijih osoba i iznosi najmanje 2/3 sveukupnih slučajeva demencija. Prevalencija bolesti u svijetu se procjenjuje na 24 milijuna slučajeva, što predstavlja značajno financijsko opterećenje. Amiloidni beta (Aβ) plakovi (eng. amyloid plaques) i neurofibrilarni snopići (eng. neurofibrillary tangles) predstavljaju glavne patološke karakteristike AB, ali u cjelosti ne daju objašnjenje ove bolesti, već dodatno uključuju upalu, neuronalnu leziju, gubitak odnosno disfunkciju aksona i sinapsa. Amiloidni plakovi okružuju aktiviranu mikrogliju, citokine i komponente komplementa, što zajedno ukazuje na upalu, koja je važna karika u patofiziologiji AB. Za dijagnozu AB, važni ulogu imaju markeri cerebrospinalnog likvora i in vivo detekcija amiloida u mozgu doprinosu najoptimalnijoj procjeni patologije bolesti i diferencijalne dijagnoze. Za detekciju amiloida u mozgu koristi se Aβ1-42 u cerebrospinalnom likvoru, dok total-tau, fosforilirani-tau (p-tau) su značajni markeri za detekciju neurodegenerativnih promjena. Uvođenje anti-amiloidne terapije kao i ostalih tkz. disease modyfing terapija moglo bi imati značajne pozitivne kliničke učinke, u slučaju da bi se s terapijom započelo rano i/ili u presimptomatskoj fazi bolesti, prije nego što je došlo do značajnog gubitka neurona. U ovom radu dan je kratak prikaz gore navedene teme, uz osvrt na aktualna i buduća istraživanja i kliničke studije primjenom monoklonalnih antitijela protiv amiloida kao mogućih uzročnika AB-a u početnoj i/ili ranoj fazi bolesti.

**KLJUČNE RIJEČI:**
Alzheimerova bolest; amiloid-beta (Aβ) plak; upala; mikroglija; anti-amiloidna terapija

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Figure 1 The evolution of plaques in Alzheimer's disease. Diffuse plaques, also called benign plaques, occur much earlier than neuritic plaques, thus supporting the idea of a therapeutic intervention in the early stage of the disease. (Courtesy of Professor Nigel J Cairns, PhD, Washington University in St Louis, MO, USA)
Figure 2 The Amyloid Cascade Hypothesis (from Reitz C; Int J Alzheimers Dis 2012, 2012:369808)