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Immunotherapy Targeting Amyloid-β Peptides in Alzheimer's Disease

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Abstract: Neurodegenerative diseases, in particular Alzheimer's disease, represent significant unmet medical needs due to a lack of effective therapeutic treatment options and cause a substantial burden for health care systems. Accumulation of β-amyloid peptides within the brain is believed to be an initial trigger of the disease process. In the last 20 years, immunotherapy has emerged as a promising target-directed strategy to develop efficient treatment options with disease-modifying potential. Unfortunately, either active vaccination against β-amyloid or its fragments, as well as passive immunization using monoclonal antibodies, have largely failed to show a clinical benefit in a variety of clinical trials. This chapter addresses progress and developments with regard to active and passive immunization against Aβ and summarizes the current state of clinical trials.

Keywords: Alzheimer's disease; amyloid; immunization; immunotherapy; monoclonal antibodies

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INTRODUCTION

With an estimated 50 million people affected worldwide, Alzheimer's disease (AD) is the most frequent cause of dementia, accounting for about 60–80% of all cases (1). The incidence is expected to increase in the next decades, due to the rapid increase of age in the population of the developing nations, possibly reaching 152 million cases by 2050 (1). Despite numerous and continuous efforts to find an effective cure, no drug has been approved for AD in the last 17 years (2). Additionally, the currently available therapies, comprising cholinesterase inhibitors and N-methyl-D-aspartate receptor agonist, do not modify the underlying pathophysiology of the disease and offer only modest, symptomatic and transient effects (3, 4). The amyloid cascade hypothesis is still widely considered the main theory for the pathology of AD (5), supported by the discovery of genetic autosomal dominant mutations in the amyloid precursor protein (*APP*), presenilin 1 (*PSEN1*) or presenilin 2 (*PSEN2*) genes in patients with early onset AD (EOAD), resulting in an enhanced formation and accumulation of amyloid-β (Aβ) peptides in plaques (6). Aβ accumulation in the brain, which starts 15–20 years before the manifestation of clinical symptoms, is believed to be the starting point for the progression of AD, driving tau phosphorylation and leading to synaptic and neuronal loss, which ultimately translates to cognitive impairment. The cascade hypothesis has been revised and modified due to, among other reasons, lack of correlation between fibrillary Aβ aggregates and AD severity (7). The focus shifted to intraneuronal Aβ accumulations as a site of Aβ toxicity (8) or oligomeric forms of Aβ, which are considered the toxic and pathogenic driving force in AD (9). The cascade hypothesis is the rationale for the development of passive and active anti-Aβ immunotherapy strategies, targeting both fibrillary aggregates and soluble forms of Aβ. Reducing Aβ burden by employing monoclonal antibodies (mAb) appears a straightforward and appealing strategy to slow or prevent the progression of the disease. Numerous antibodies have been tested so far and are currently under investigation in clinical trials; however, the outcomes of the past two decades have been disappointing. Though some antibodies, such as bapineuzumab and aducanumab, appeared to clear parenchymal amyloid (10, 11), failure to meet the primary endpoints or the occurrence of adverse side effects such as vasogenic edema and/or microbleeding (12) caused the termination of the ongoing trials for most of the tested mAb. The reasons for the disappointing outcomes could also be imputable to factors independent from the actual mode of action of the tested mAbs. An inaccurate selection of trial patients, leading to huge variations in cognitive and clinical decline during the trial period, as well as a late intervention and insensitive efficacy measures are potentially confounding factors. Proper target engagement (e.g. soluble, monomeric, dimeric, oligomeric, fibrillary Aβ) is also a critical aspect that needs to be addressed.

Aβ GENERATION AND AMYLOID CASCADE HYPOTHESIS

The vast majority of AD cases are of sporadic origin, occurring beyond 65 years of age with an unknown cause. While mutations in *APP* or the *PSEN* genes have

been linked to early-onset autosomal dominant forms of familial AD (FAD) with an early disease onset (13, 14), so far, only genetic risk loci have been identified as potentially involved in APP processing or β-amyloid peptide generation in sporadic cases (15). APP is a single-pass transmembrane protein, and Aβ peptides are generated via a series of consecutive proteolytical cleavage steps from this larger precursor protein (16). The generation of Aβ peptides from its precursor APP is linked to the so-called amyloidogenic processing pathway, which is initiated by β-secretase cleavage. This cleavage is predominantly carried out by an aspartic protease named β-site APP cleaving enzyme (BACE1) (17), resulting in the release of a soluble APP fragment (sAPP-β) and a slightly longer APP C-terminal fragment of 99 amino acids (CTF-β). Further cleavage by γ-secretase, a protein complex consisting of PSEN1/2 among others (18), releases Aβ peptides. This complex is able to cut APP at slightly different positions, mainly resulting in the production of ~90% of $A\beta_{1-40}$ and less than 10% of $A\beta_{1-42}$ under basal conditions (19), but also shorter as well as slightly elongated Aβ peptides ($A\beta_{37} - A\beta_{43}$) (20). Processing by BACE1 and γ-secretase generates full-length Aβ peptides starting with an aspartic acid residue at position 1 (mainly $A\beta_{1-40}$ and $A\beta_{1-42}$). While most research efforts have concentrated on the full-length peptide species $A\beta_{1-40}$ and $A\beta_{1-42}$, there is accumulating evidence that a variety of other N- and C-terminally modified Aβ peptides may play an important role in the disease process (21–23).

The accumulation of Aβ peptides is regarded as one of the central processes underlying the neuropathological changes in AD. Almost 30 years ago, the amyloid cascade hypothesis was formulated, theorizing that Aβ accumulation is the initial event triggering further pathological alterations such as tau phosphorylation and neurofibrillary tangle formation, neuron and synapse loss, as well as cognitive impairment (5). While research efforts initially focused on fibrillar Aβ deposits in the form of extracellular plaques, the significance of soluble Aβ species (24, 25), mainly in the form of oligomers, became more and more recognized. They may directly injure synapses and neurites of brain neurons (26, 27), in addition to activating microglia and astrocytes (9). These metastable oligomeric forms likely exist in an equilibrium with amyloid plaques and consist of cross-β-sheet Aβ peptide units of variable size, including protofibrillar intermediates (28, 29) (Figure 1).

MECHANISM AND PRINCIPLES OF (AMYLOID-β) IMMUNOTHERAPY

Immunotherapy focuses on the generation (in case of active) or use (in case of passive) of antibodies targeting a specific antigen, Aβ in this specific context, counteracting the disease by activation of the immune system. In active immunizations, a vaccine containing the Aβ-antigen is administered usually intramuscularly. Depending on the type of antigen used, a humoral response with B-cell and helper T-cell (T_H) involvement, cytokine secretion and production of polyclonal antibodies, and/or a cell-mediated immunity response with the activation of phagocytes (antigen-specific cytotoxic T-lymphocytes) is induced. T-cell populations can be further divided into cytotoxic T-cells, which kill target cells by

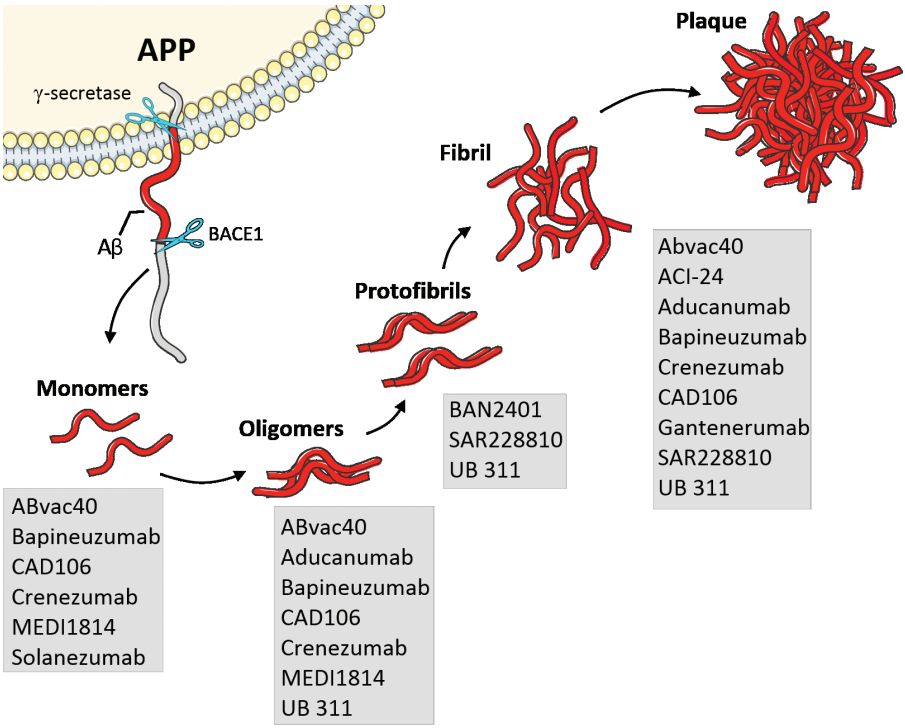

Figure 1. Proposed targets of anti-amyloid-β (Aβ) drugs used in active and passive immunization approaches (modified from (2)).

inducing apoptosis, macrophage-activating proinflammatory Th1 cells, and Th2 cells that stimulate B-cells into antibody-producing cells (30).

In passive immunization, monoclonal antibodies (mAb) against specific Aβ forms are administered by intravenous infusions or subcutaneous injection. In both cases, the antibodies are at first peripherally located and are required to pass the blood–brain barrier (BBB), greatly restricting the transport of antibodies, in order to reach the brain parenchyma. The access route for immunoglobulins has not been clearly identified yet, but could comprise passive diffusion, the lymphatic system, and perivascular spaces. The absence of active transport systems for antibodies, the presence of receptors (such as the neonatal Fc receptor) acting as a pump to remove antibodies in the central nervous system (CNS), as well as other not yet understood clearance mechanisms, are reasons why only a small fraction of antibodies (approximately 0.1%) introduced into the peripheral circulation can be detected in the brain or cerebrospinal fluid (CSF) (31). The presence of a large number of antibodies in the periphery could also act as a driving force for the efflux of Aβ out of the CNS, likely by changing the dynamic equilibrium between Aβ in the blood and the brain. Antibodies might therefore act as a peripheral Aβ "sink," creating a concentration gradient that attracts monomeric Aβ out of the CNS via passive diffusion mechanisms (32). In the brain parenchyma there

are several mechanisms, that are not mutually exclusive, by which the humoral response could exert its effects (33), and the Aβ epitope against which the antibody is directed (monomeric, oligomeric, fibrillary Aβ) may lead to a preferred mechanism over another. The antibodies could directly be responsible for the disassembly of Aβ deposits in the brain (34) or prevention of reassembly and inhibition of toxicity, as shown by *in-vitro* experiments (35, 36). Direct binding to Aβ oligomers, thus neutralizing their toxicity, is also a putative mechanism (37). The clearance of Aβ could also be enhanced by the antibodies through microglial activation, leading to Fc-mediated (32) or Fc-independent phagocytosis (38) (Figure 2). Peripherally, large immunoglobulin IgM, which is able to cross the BBB

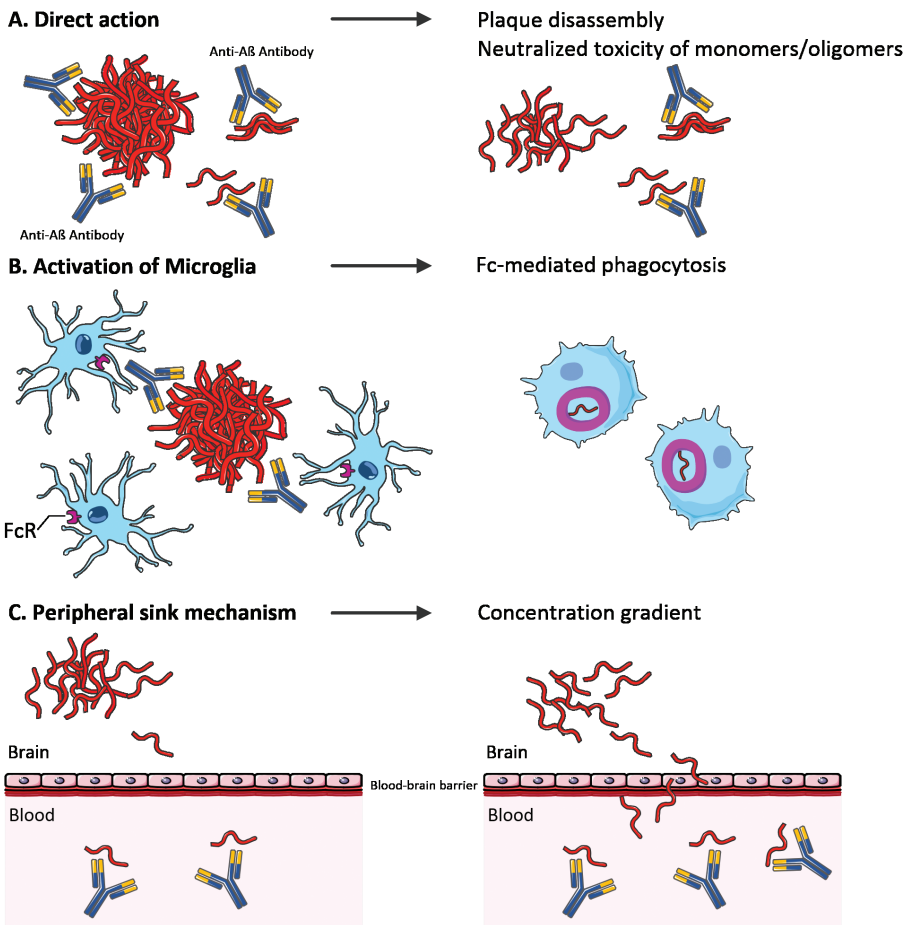

Figure 2. Proposed mechanisms of anti-amyloid-β (Aβ) antibodies. Antibodies might either directly target Aβ assemblies, leading to a neutralization of Aβ toxicity (A), or activate microglia, resulting in Fc-receptor (FcR) mediated phagocytosis (B). Alternatively, antibodies might not enter the brain but create a concentration gradient between the brain and the blood, leading to Aβ removal via a peripheral sink mechanism (C) (modified from (121)).

to a lesser extent compared to IgG but is likely involved in the already-mentioned peripheral-sink effect, is believed to directly hydrolyze Aβ (39). The specific advantages and disadvantages of active and passive immunization are described in the next paragraphs.

ACTIVE AMYLOID-β-DIRECTED IMMUNOTHERAPIES

In active immunization, immunity is achieved following exposure to an Aβ antigen that causes the generation of antibodies in the recipient. It engages the cellular and humoral immune system, including T and B cells. Typically, an active vaccine is comprised of an antigen (alone or conjugated to a non-self T helper cell epitope) combined with an immune boosting adjuvant to ensure high antibody production. An advantage of active immunization is that with few vaccinations the patient should be able to produce a prolonged antibody response. The variability of the induced response across patients is, on the other hand, a problematic aspect, especially when dealing with elderly individuals. Adverse side effects may occur after active immunization: when a T-cell response is induced, the risk of an abnormal immune response increases. With age, the competency of the immune system reduces and the probability of developing autoimmune responses is enhanced. Additionally, vaccines lead to the formation of polyclonal antibodies, which can recognize multiple and possibly overlapping epitopes on the target protein. Polyclonal antibodies may be problematic in case the goal is the recognition of a specific form of the antigen.

The first effort to explore active immunization as a possible therapy for AD was made in 2001 with a vaccine called AN1792, consisting of synthetic full-length $A\beta_{1–42}$ peptide with QS-21 adjuvant. Despite initial positive findings in an APP-overexpressing mouse model (40), the phase II clinical trial in individuals with mild-to-moderate AD was interrupted, as 6% of the treated patients developed a T-cell-mediated meningoencephalitis (41). Additionally, only 20% of patients produced antibodies above the preset therapeutic cut-off titration level and clinical outcomes were no better than those of the placebo-treated control subjects (42). Despite the cessation of the trial, several follow-up studies were carried out as post-mortem brain samples from trial participants became available. Neuropathological analyses from AN1792 recipients in general showed a lower mean Aβ load compared to an age-matched unimmunized control group. The degree of plaque removal varied among immunized patients along with mean antibody response, and no evidence of improved survival or delay in the development of severe dementia was observed (43). It was further reported that immunized patients showed several-fold increases in $A\beta_{42}$-containing blood vessels in the cerebral cortex and leptomeninges, as well as a higher density of micro-hemorrhages. However, no major cerebral amyloid angiopathy (CAA)-related intracerebral hemorrhages were noted and, interestingly, two of the longest survivors showed a virtually complete absence of both plaques and CAA (44). Further studies revealed that active immunotherapy with AN1792 was associated with wall splitting in leptomeningeal vessels (45) and an accelerated loss of damaged degenerating neurons, an observation consistent with imaging data indicating an increased rate of cerebral atrophy among immunized AD individuals (46). A recent study reporting on post-mortem data from two AD patients who died

14 years after immunization revealed that these patients remained virtually plaque-free, however, an extensive overall distribution of neurofibrillary tangles (Braak stage V/VI) was observed (47).

In order to control the immune response by eliciting a strong antibody production but avoiding inflammatory T-cell activation, second-generation vaccines were designed to target more specific epitopes (48). One of these second-generation Aβ vaccines, ACC-001 (vanutide cridificar), studied by Janssen Immunotherapy and Pfizer, was discontinued in phase II clinical trials, as the primary efficacy-biomarker endpoints were found not statistically significant in the considered dosage groups (49, 50). The vaccine was composed of $A\beta_{1-7}$ with QS-21 adjuvant, designed to avoid the autoimmune meningoencephalitis caused by Th1 lymphocyte activation seen with AN1792, attributed to Aβ residues 15–42. CAD106 (Amilomotide) was another second-generation Aβ vaccine that reached phase II clinical trials involving patients with mild AD. CAD106 is composed of multiple copies of $A\beta_{1-6}$ peptide, coupled to a Qβ virus-like particle. Phase II trials in the United States and Europe ended in 2010 and 2011, supporting the favorable safety profile found in phase I trials and reporting prolonged antibody titers in responders (51). In a phase IIb trial, 120 patients suffering from mild AD received up to 7 intramuscular injections of CAD106 or placebo over 60 weeks. The vaccine was generally well tolerated and elicited an Aβ-specific immune response with an acceptable safety profile and preliminary evidence of target engagement by amyloid positron emission tomography (PET) (52). Despite a phase II/III trial began in November 2015, set to run until 2023, in September 2019 Novartis noted in its quarterly financial report that it had "retired" the CAD106 program. Several other candidates have been investigated and reached different stages of clinical development (Table 1).

ABvac40

ABvac40 is an investigational vaccine targeting the C-terminus of $A\beta_{40}$. The agent comprises multiple repeats of a short C-terminal fragment of the Aβ peptide ($A\beta_{33-40}$), conjugated to the keyhole limpet cyanine (KHL) carrier protein and formulated with the adjuvant alum hydroxide. The phase I clinical trials demonstrated a favorable safety and tolerability profile with no incidence of vasogenic edema nor microhemorrhage (53). A phase II clinical trial by Araclon Biotech S.L. is ongoing in several European countries to confirm the results and explore the clinical efficacy of ABvac40 in patient with amnestic MCI and very mild AD (Clinical Trial: NCT03461276) and is due to be completed in February 2022.

ACI-24

ACI-24 is a liposome vaccine that is designed to elicit an antibody response against aggregated Aβ peptides. ACI-24 is based on the truncated $A\beta_{1-15}$ sequence, thus avoiding the T-cell epitopes. At each end of the peptide, a palmitoylated lysine residue was attached, enabling anchoring the peptide in the lipid bilayer of a liposome adjuvant thus adopting an aggregated β-sheet structure and forming a conformational epitope. After promising preclinical results (54), a phase I/II trial to assess safety, tolerability, immunogenicity as well as efficacy of the vaccine in patients with mild-to-moderate AD began in 2009 in Denmark, Finland and

TABLE 1 **Principal active amyloid-β-directed immunotherapy vaccines**

Drug	Main ref.	Peptide	Adjuvant	Subjects	Phase	Clinical trial #	Outcome
ABvac40	53	$A\beta_{33-40}$ (multiple copies) conjugated to KHL	Alum hydroxide	a-MCI or very mild AD	Phase II	NCT03461276	Ongoing trial
ACC-001	49	$A\beta_{1-7}$	QS-21	Mild-to-moderate AD	Phase II	NCT00479557 NCT00498602	Failed
ACI-24	54	$A\beta_{1-15}$ with palmitoylated lysine residues	Liposome adjuvant	AD in Down Syndrome	Phase II	NCT04373616	Trial scheduled for late 2020
Affitope AD02	55, 56	$A\beta_{1-6}$ conjugated to KHL	Aluminum	Early AD	Phase II	NCT01117818	Failed
AN1792	42	$A\beta_{1-42}$	QS-21	Mild-to-moderate AD	Phase II	NCT00021723	Failed
CAD106	51, 52	$A\beta_{1-6}$ (multiple copies) conjugated to Qβ	–	Mild AD	Phase II	NCT02565511	Failed
UB 311	57	two $A\beta_{1-14}$–targeting peptides	Alum-containing Th2-biased delivery system	Mild AD	Phase II	NCT02551809	Completed

The list is presented in alphabetical order. The table shows the status of studies on 31 July 2020, as reported in ClinicalTrials.gov.
Aβ, amyloid-β; AD, Alzheimer disease; a-MCI, amnesic mild cognitive impairment.

Sweden. In 2016, ACI-24 became the first anti-Aβ vaccine to be evaluated for the treatment of Alzheimer's disease in Down's syndrome, and in late 2020 a double-blind, randomized, placebo-controlled phase II trial to assess the safety, tolerability and target engagement in adults with Down syndrome is scheduled to start (Clinical Trial: NCT04373616).

Affitope AD02

Affitope AD02 consists of a synthetic peptide of six amino acids mimicking the N-terminus of Aβ, lacking the most common T-cell epitopes, but including the B cell epitope. This peptide induced an anti-Aβ antibody response when conjugated to Keyhole Limpet Hemocyanin and adjuvanted with aluminum (55). In 2009, a phase I study was conducted in Austria by AFFiRiS AG and showed a favorable safety and tolerability profile 1 year after treatment. A phase II trial of AD02 was conducted in Europe between 2010 and 2013 in patients with early AD, but no significant treatment effects were seen with AD02. Surprisingly, the placebo group receiving a dose of the immunomodulator aluminum oxihydroxide which was part of the formulation, then called AD04, showed a significantly reduced cognitive decline correlating with a reduced hippocampal shrinkage (56). The company declared to be interested in further investigating the potential therapeutic effects of AD04; however no further data have been disclosed yet and no further activities with regard to AD are listed on the company website.

UB 311

UB 311 is a synthetic peptide vaccine developed by United Neuroscience, coupling a helper T-cell epitope to the $A\beta_{1–14}$ sequence. The approach aims to stimulate a T helper type 2 regulatory immune response over a T helper type 1 proinflammatory response (57). In a transgenic AD mouse model (hAPP751), UB-311 reduced levels of $A\beta_{1–42}$ oligomers and protofibrils, as well as extracellular amyloid plaque load (57). In a first-in-human clinical trial in patients with mild-to-moderate AD, each participant received three immunizations (300 μg/dose) by intramuscular injection. The vaccine was well tolerated and showed encouraging improvement in ADAS-Cog scores in the subgroup of mild AD patients (57). As a result, a phase-II clinical trial started in Taiwan in October 2015 enrolling people with a clinical diagnosis of mild AD, which was followed by a safety extension in 2018. A press release from United Neuroscience at the beginning of 2019 reported a favorable safety profile and promising, yet not statistically significant, changes in the secondary endpoints (amyloid PET burden, CDR-SB, ADCS-ADL, ADAS-Cog and MMSE [Mini-Mental State Examination]) (58). United Biomedical, as of July 2020, lists UB 311 as investigational vaccine but no current clinical trials are registered.

Passive Immunotherapy with Monoclonal Antibodies

In passive immunization, externally produced antibodies are administered through intravenous infusions or subcutaneous injections. They can be humanized versions of murine mAb evaluated in previous preclinical trials (such as Bapineuzumab) or fully human mAbs (like Gantenerumab). In the first group,

murine mAbs are modified so that a large part of their protein sequences is similar to naturally produced human antibody variants, in order to reduce the immunogenicity that (foreign) murine antibodies would cause. Fully human mAbs are produced for example with transgenic mice that have been genetically engineered with the human immunoglobulin locus, while in contrast, humanized mAbs are initially generated in wild type mice with the endogenous murine immunoglobulin locus (59). Avoiding some of the side effects that the humanized murine mAb still possess, fully human mAb are considered safer and more effective (60).

The passive immunization strategy allows for a precise titration of the administered antibodies and a possible rapid clearance in case adverse effects develop, but has the disadvantage that repeated infusions/injections over time are required to maintain a constant amount of therapeutic antibodies. Passive immunization might allow for targeting specific conformations of the Aβ peptide, presumably leading to the specific removal of distinct Aβ assemblies such as monomers, oligomers, or fibrils (61). The employment of mAbs against Aβ has been associated with the risk of developing amyloid-related imaging abnormalities (ARIAs) as severe adverse effects. These abnormalities seen in neuroimaging of AD patients comprise "vasogenic edema" and/or sulcal effusion (ARIA-E) and hemosiderin deposits (ARIA-H) including microhemorrhage and cortical superficial siderosis (62), and are believed to be the consequence of the removal of vascular amyloid leading to increased vascular permeability. The development of ARIAs after mAb treatment appears to be compound-dependent and dose-related. ARIAs represent a core safety issue in immunotherapy trials and challenged the progress of mAbs as a treatment for AD. Magnetic resonance imaging (MRI) imaging is used to detect active stages of ARIAs in clinical trials, but is not appropriate for predicting the risk of developing ARIAs during treatment (63). Currently, efforts to discover and use specific biomarkers for ARIAs in clinical trials are being made to better manage these severe side effects and reduce the delay caused by this side effect often seen in clinical trials (64).

Amyloid clearance in immunotherapy is largely correlated with IgG Fcγ Receptor (FcγR)-mediated activation of microglia and antibody-mediated phagocytosis, however, these same effects are probably responsible for an increased inflammatory response and vascular side effects (ARIAs) observed in a variety of studies (65). FcγRs are activated by human IgG1 and mouse IgG2a with higher affinity compared to other IgG subclasses. Using a different class of immunoglobulin G (e.g. IgG4) could help prevent an excessive microglial activation, reducing the risk of vascular damage (66). Modification of the effector function, such as de-glycosylation, by antibody engineering, was also used as a strategy to reduce the incidence of adverse ARIAs (67). Even though the role of the antibody effector function in the development of vascular side effects is clear, the engaged epitope is also crucial. A comparative study of murine versions of therapeutic Aβ antibody candidates with a constant IgG2a region showed strong differences in their plaque-removing potential, demonstrating that the ability of an antibody to remove plaques and activate inflammation is critically dependent on its epitope and affinity (68). A variety of antibodies have been evaluated in passive immunotherapy approaches and reached different stages of clinical trials (Table 2).

TABLE 2 Principal passive amyloid-β-directed immunotherapy drugs

Drug	Main ref.	Target	Antibody - subtype	Subjects	Phase	Clinical trial #	Outcome
Aducanumab	122	$A\beta_{3-7}$	Humanized mAb - IgG1	Early AD	Phase III (ENGANGE and EMERGE)	NCT02477800 NCT02484547	Terminated
BAN2401	115	Soluble Aβ protofibrils	Humanized mAb - IgG1	Early AD Preclinical AD	Phase III Phase III	NCT03887455 NCT04468659	Ongoing Trial Ongoing Trial
Bapineuzumab	71	$A\beta_{1-5}$ (fibrillary and soluble Aβ)	Fully human mAb - IgG1	Mild-to-moderate AD	Phase III	NCT00676143 NCT00667810 NCT00575055 NCT00574132	Failed
Crenezumab	81 82	$A\beta_{13-24}$ (pentameric Aβ oligomers and fibrils)	Humanized mAb - IgG4	Prodromal-to-mild AD Preclinical AD with PSEN1 E280A mutation	Phase III (CREAD 1 and 2) Phase II	NCT02670083 NCT03114657 NCT01998841 NCT03977584	Failed Ongoing Trials
Gantenerumab	85, 86	Aβ fibrils	Humanized mAb - IgG1	Early AD	Phase III (GRADUATE 1 and 2)	NCT03443973 NCT03444870	Ongoing Trials
MEDI1814	129	$A\beta_{42}$ C-terminus (Aβ monomers and low-n oligomers)	Fully human mAb - IgG1λ	Mild-to-moderate AD	Phase I	NCT02036645	Completed
Ponezumab	95	$A\beta_{40}$ C-terminus (Aβ monomers)	Humanized mAb - IgG2Δa	Mild-to-moderate AD	Phase II	NCT00722046 NCT00945672	Failed
SAR228810	130	Protofibrillary and fibrillary Aβ	Humanized mAb - IgG4	AD	Phase I	NCT01485302	Completed
Solanezumab	105, 106	$A\beta_{16-26}$ (Aβ monomers)	Humanized mAb - IgG1	Mild-to-moderate AD Individuals with EOAD-associated mutations	Phase III (EXPEDITION 1,2 and 3) Phase II/III	NCT00905372 NCT00904683 NCT01900665 NCT01760005	Failed Ongoing Trial

The list is presented in alphabetical order. The table shows the status of studies on 31 July 2020, as reported in ClinicalTrials.gov.
Aβ, amyloid-β; AD, Alzheimer disease; EOAD, early onset Alzheimer disease; mAb, monoclonal Aβ.

Bapineuzumab

Bapineuzumab is a humanized form of the murine monoclonal antibody 3D6, directed specifically towards the N-terminus of the Aβ sequence starting at Asp1 (69, 70). This antibody of the IgG1 subclass binds fibrillary and soluble Aβ and activates microglial phagocytosis as well as cytokine production, aiming to reduce plaque formation and promote Aβ clearance (71). Preclinical studies and phase I–II clinical trials gave initial promising results. When 3D6 mAb was administered to 4-month-old PDAPP mice with i.p. injections of 10 mg/kg/week for 12 months, total Aβ deposition was reported to be almost completely reduced (72). Although the translatability of these preclinical studies was later questioned (73), bapineuzumab was tested in a phase I clinical trial where a single ascending dose was administered to patients with mild-to-moderate Alzheimer's disease in order to determine the safety, tolerability, and pharmacokinetics of the mAb (74). MRI abnormalities, consistent with vasogenic edema, were observed in 3 out of 10 patients receiving the higher dose of 5 mg/kg, but this resolved with time. MMSE scores improved at the lower doses (0.5 and 1.5 mg/kg) of bapineuzumab compared to the placebo, a finding not observed with the highest dose.

In a phase II clinical trial, patients with mild-to-moderate AD were randomly assigned to one of four dose cohorts (0.15, 0.5, 1.0, or 2.0 mg/kg) and received six infusions 13 weeks apart. The final assessments were performed at week 78 but no significant differences were found in co-primary efficacy endpoints, the ADAS-cog and Disability Assessment for Dementia (DAD). Exploratory analyses showed potential treatment differences on cognitive and functional endpoints. Differences based on APOE ε4 carrier status were also observed. ARIA-E was found in 12/124 treated patients, with a dose and APOE ε4 carrier-dependent incidence increase (71). Additional phase II studies reported a reduction in exploratory CSF biomarkers T-Tau and p-Tau, the latter being significantly different between treated and placebo groups (75). A reduced cortical ^{11}C-Pittsburgh compound B (PiB) average uptake, visualized by PET, was also found after 78 weeks of treatment with bapineuzumab (76). The feasible and tolerable administration of bapineuzumab, together with evidence that the mAb could be disease modifying, led to the actualization of phase III clinical trials.

A four-trial phase III program was launched in North America and Europe. The first two double-blind, randomized, placebo-controlled 18-month phase III trials tested bapineuzumab in patients with mild-to-moderate Alzheimer's disease, divided into APOE ε4 carriers and non-carriers (71). Bapineuzumab was administered by intravenous infusion every 13 weeks for 78 weeks at a dose of 0.5 mg/kg in APOE ε4 carriers and at 0.5 mg/kg, 1 mg/kg, and 2 mg/kg doses in non-carriers, even though the highest dose was soon discontinued due to ARIA-E and ARIA-H development. No significant differences were found in the primary outcome measures (ADAS-cog11 and DAD) between groups. The APOE ε4 carriers group showed a modest reduction in PiB PET binding as well as a significant reduction of CSF p-Tau when compared to the placebo group. Consistent with the phase II data, a dose-related and APOE ε4 carriers-dependent increase in ARIA-E was observed. The failure to meet the primary endpoints led to the discontinuation of two additional phase III clinical trials and the further evaluation of bapineuzumab as treatment for AD.

Crenezumab

Crenezumab is a humanized mAb designed on an IgG4 backbone targeting multiple species of Aβ. Its epitope is located in the central part (~ $A\beta_{13–24}$) of the peptide and it shows particular affinity for pentameric oligomeric and fibrillary 16mer assemblies of aggregated Aβ (77, 78). A recent study confirmed that it detects a variety of full-length and N-terminal truncated Aβ variants in post-mortem human AD brain samples (70). Limited preclinical data are currently published on the efficacy of chronic treatments with crenezumab. The murine version of the antibody (mC2) was tested in 18-month Tg2576 transgenic mice with a single intracerebral injection of 2 µg of antibody, which did not cause significant inflammatory changes (68). *In vivo* imaging of 10-month-old transgenic $hAPP^{(V717I)}$/PS1 mice showed decreased plaque volumes over a period of 3 weeks after an intraperitoneal injection of 60 mg/kg antibody (77). The same study reported the results of a phase I clinical trial, performed in patients with mild-to-moderate AD. No ARIAs were observed either with a single or multiple ascending dosage.

Crenezumab was further tested in phase II clinical trials in patients with mild-to-moderate AD. A total of 431 patients were enrolled in the ABBY study, receiving either a low subcutaneous dose (300 mg) or placebo every 2 weeks, or an intravenous high dose (15 mg/kg) or placebo every 4 weeks, for a total period of 68 weeks (79). The primary endpoints (changes in ADAS-Cog12 and CDR-SB scores), measured at week 73, were not met. Exploratory analyses pointed towards a reduction in decline on the ADAS-Cog12 in the high-dose group, and the patients with mild AD showed the greatest deviation from the placebo group. This difference became significant in the group with MMSE scores ranging from 22 to 26. These trends were also observed in a smaller phase II brain imaging study (BLAZE), enrolling 91 patients. Even though no significant differences were observed in the primary outcome measures, non-significant trends toward ADAS-Cog12 and CDR-SB score improvements were observed in the mild AD group receiving the higher dose of antibody (80). Throughout these studies, no ARIAs adverse effects were reported.

Two large phase III clinical trials, CREAD1 and CREAD2, started in 2016 and 2017 respectively, and enrolled patients with prodromal-to-mild AD. These double-blind, placebo-controlled global studies recruited overall more than 1500 patients, testing a 60 mg/kg dose by intravenous infusion every 4 weeks for a period of 100 weeks with the primary endpoint being changes in the CDR-SB score at 2 years (81). In January 2019 the company Roche announced the decision to discontinue both trials, based on preliminary analyses suggesting that the primary endpoint would unlikely be met. Crenezumab is, to date, being tested as a preventive treatment as part of the Alzheimer Prevention Initiative (API) in a randomized, double-blind, placebo-controlled phase II study by Genentech, estimated to end in 2022 (Clinical Trial: NCT01998841). The 5-year trial started in 2013 and recruited patients who carry the PSEN1 E280A autosomal-dominant mutation and are still in a preclinical phase of AD (82). In a subgroup of participants (carriers and non-carriers) the longitudinal tau burden will be evaluated with a tau positron emission tomography (PET) scan after IV injection of the probe [18F]GTP1 (Clinical Trial: NCT03977584).

Gantenerumab

Gantenerumab is a recombinant human IgG1 antibody, designed to recognize a conformational epitope present on Aβ fibrils, in order to disassemble and degrade aggregated Aβ peptides via recruiting microglia and activating phagocytosis (83). Using peptide mapping, N-terminal as well as central portions of Aβ were recognized and no evidence of altered plasma Aβ was detected. In a preclinical study, gantenerumab bound cerebral Aβ and significantly reduced small amyloid-β plaques in APP/PS2 transgenic mice with chronic treatment (83). An initial randomized study of AD patients receiving either 60 mg or 200 mg intravenous gantenerumab or placebo, showed a ~16% or ~36% reduction in Pittsburgh Compound B retention in the 60 mg and 200 mg gantenerumab group respectively. However, two patients in the 200 mg group showed vasogenic edema and focal areas of inflammation on MRI scans at sites with the highest level of amyloid removal (84).

The Scarlet RoAD trial assessed the efficacy and safety of gantenerumab in prodromal AD patients. Participants enrolled in this 2-year randomized double-blind phase III study received 105 mg, 225 mg or placebo every 4 weeks subcutaneously. A dose- and APOE ε4 genotype-dependent increase of generally asymptomatic ARIAs was noticed and the study was terminated for futility when no differences in primary or secondary endpoints were observed (85). Of note, significant reductions in total and phosphorylated tau in the CSF, as well as a dose-dependent reduction in brain amyloid on PET scans were observed in an exploratory biomarker analysis (85). A 2-year PET sub-study evaluating the effect of up to 1200 mg of gantenerumab every 4 weeks in patients with prodromal-to-moderate AD, revealed a 3.5-times greater reduction in amyloid-PET signal than seen after 2 years at a dose of 225 mg, with 51% of patients having amyloid-β plaque levels below the positivity threshold (86).

A phase I randomized, open-label study including healthy volunteers aged 40–80 years, evaluated different subcutaneous injection regimens of gantenerumab, with regard to pharmacokinetic properties and tolerability. The results of this study suggest that subcutaneous injections at speeds of 5 and 15 s were well-tolerated and might enable at-home administrations by AD patients or their caregivers (87). Gantenerumab is currently under investigation in two large phase III trials (GRADUATE 1 and 2), which started enrolling patients with early AD in 2018 with the goal of more than 1500 patients in up to 350 study centers with a data read-out expected in 2022 (Clinical Trial: NCT03443973 and NCT03444870).

Ponezumab

Ponezumab is a humanized monoclonal IgG2Δa anti-Aβ antibody reported to bind to the C-terminus of the most abundant $A\beta_{1–40}$ peptide. It contains two mutations that eliminate effector function and therefore potential cell toxicity depending on the antibody. Structural analyses revealed extensive contacts of ponezumab with the carboxyl moiety of $A\beta_{40}$ (88). Preclinical analyses using the murine antibody 2H6, similarly binding to the C-terminal of $A\beta_{1–40}$, demonstrated a robust reduction of amyloid deposits in aged Tg2576 (89). Intraperitoneal

injections of ponezumab increased plasma $A\beta_{1-x}$ and $A\beta_{x-40}$ levels in PS1xAPP mice in a concentration-dependent manner, while $A\beta_{x-42}$ plasma concentrations remained unchanged. This led to the suggestion that ponezumab removes brain Aβ via a peripheral sink mechanism (88). Another preclinical study in cynomolgus monkeys, sharing the same Aβ peptide sequence with humans, confirmed increased plasma $A\beta_{1-40}$ and $A\beta_{1-x}$ levels in treated animals versus controls (90).

An initial randomized, double-blind, single-dose-escalation study evaluated safety, pharmacokinetics and pharmacodynamics using doses of 0.1 mg/kg up to 10 mg/kg. The 2-h infusion was well-tolerated, and in individuals receiving the highest dose increases in CSF Aβ were observed, which is suggestive of altered central Aβ levels (91). A related study in a cohort of Japanese subjects yielded comparable results (92). A different administration protocol of a single 10-min intravenous infusion was evaluated and produced comparable effects on plasma Aβ species (93). Individuals aged 50 and older with a diagnosis of mild-to-moderate AD and a MMSE score of 16 to 26 were enrolled in a placebo-controlled, multiple dose study (0.1 mg/kg up to 8.5 mg/kg) of ponezumab. The treatment was administered as 10 2-h infusion every 2 months, and was generally well tolerated with an acceptable safety profile and robust plasma Aβ increases but no evidence of a dose response with regard to CSF biomarkers (94). Effects on peripheral and central Aβ were characterized in small Swedish cohorts suffering from mild-to-moderate AD. One cohort received ponezumab (10 mg/kg) or placebo quarterly over 1 year, whereas a second cohort started with an initial dose of 10 mg/kg or placebo, followed by monthly infusions of 7.5 mg/kg or placebo respectively. This phase II study again showed that ponezumab was generally safe and well tolerated, with dose-dependent increases in plasma Aβ. However, no apparent differences in brain amyloid burden assessed by PiB-PET were detected and changes in both cognitive and functional decline were observed during the course of the study without, however, differences between treatment arms (95). The potential effect of intravenous ponezumab was also investigated in patients with probable cerebral amyloid angiopathy (CAA) (96), a disease condition with amyloid deposition in the walls of leptomeningeal and intracortical blood vessels of the CNS (97, 98). In this study, again, ponezumab was safe and well tolerated; however, this antibody has been discontinued as prespecified efficacy criteria were not met in the majority of the trials.

Solanezumab

Solanezumab is a humanized monoclonal IgG1 antibody (mouse version m266), targeting the mid-region of Aβ. Co-crystallization studies revealed that solanezumab accommodates a large Aβ epitope (residues 16–26), forming extensive contacts and hydrogen bonds with the antibody (99). As administration of solanezumab as well as its murine precursor m266 cause substantial dose-dependent increases in plasma antibody-bound Aβ levels (100–102), it has been suggested that this antibody primarily targets soluble monomeric forms of Aβ. On the contrary, neuropathological studies employing human brain samples indicated that a recombinant biosimilar antibody of solanezumab showed a strong binding affinity to amyloid plaques (103), calling its assumed selectivity for monomeric Aβ into question. In transgenic PDAPP mice, administration of m266 resulted in a rapid

reversal of memory deficits in the absence of amyloid plaque reductions (102); however, a more recent study in the J20 mouse model of AD reported no improvement of behavioral deficits and even a strongly increased mortality rate following m266 immunization (101).

Solanezumab has been investigated in several clinical trials in order to evaluate its disease-modifying potential. Following a phase II trial with 52 patients suffering from mild-to-moderate AD evaluating diverse dose regimens (104), two large phase III studies (EXPEDITION-1, EXPEDITION-2) were launched. These studies recruited 2,052 mild-to-moderate AD patients, who received monthly 400 mg infusions. However, both showed a lack of efficacy with regard to cognitive performance, the primary outcome measure of both studies (105). Pooled analyses of both studies suggested less functional and cognitive decline in the mild AD population; however, no significant differences in baseline-to-endpoint changes were found for a variety of secondary outcome measures such as activities of daily living (106). Following the review of the data obtained from the pooled mild AD population, a third phase III trial (EXPEDITION-3) was initiated. This trial enrolled 2129 patients with mild dementia and evidence of amyloid deposition, shown by either florbetapir PET or $A\beta_{1\text{-}42}$ measurements in CSF, and patients received 400 mg solanezumab or placebo every 4 weeks for 76 weeks. As a result, the secondary analyses of the previous EXPEDITION trials were not reproduced and solanezumab showed no benefit with regard to cognitive decline in patients with mild AD (107).

Solanezumab is being tested within the Dominantly Inherited Alzheimer Network (DIAN) trial in a phase II/III study as a potential disease-modifying treatment, together with gantenerumab, in individuals at risk for or with a mutation associated with EOAD. The trials are estimated to be completed by March 2021 (Clinical trial: NCT017660005).

BAN2401

BAN2401 is the humanized version (IgG1) of the mouse monoclonal antibody mAb158, which has been shown to primarily bind to large soluble Aβ protofibrils (108). Selectivity for this type of aggregate has been described to be at least 1000-fold higher than for monomers and 10–15 times better than for Aβ fibrils (109, 110). Administration of mAb158 to plaque-bearing AD transgenic mice carrying both the Arctic and Swedish APP mutations (tg-ArcSwe) resulted in lowered Aβ protofibrils, albeit unchanged insoluble Aβ levels. When treatment was started prior to extracellular plaque onset, a prevention of amyloid deposition and a reduction in protofibril levels was observed (111). Interestingly, individual performance of young tg-ArcSwe mice in a spatial memory test (Morris water maze) was inversely correlated with protofibril but not total Aβ levels (112). This antibody, as well as its humanized version BAN2401, efficiently precipitated soluble Aβ aggregates from the human brain, and more than 50% reduction of protofibrils/oligomers was observed after long-term mAb158 treatment in the CSF of tg-ArcSwe mice (113). A radiolabeled version of mAb158 conjugated to a transferrin receptor antibody has been shown to effectively visualize Aβ in the brain of two AD mouse models, enabled via receptor-mediated transcytosis across the BBB (114).

Safety and tolerability of BAN2401 were investigated in an ascending dose study (0.1 mg/kg up to 10 mg/kg biweekly) for 4 months in mild-to-moderate AD cases. The treatment was well-tolerated across all doses and a slight elevation of plasma $A\beta_{1-40}$ was noted, albeit in the absence of measurable effects on CSF biomarkers (115). A subsequent placebo-controlled, double-blind, randomized phase IIb study enrolling 856 patients with mild cognitive impairment (MCI) caused by AD or mild AD-dementia evaluated several doses in a Bayesian adaptive design. A statistically significant reduction in amyloid PET standard uptake value ratio (SUVR) was observed after 18 months at the highest dose, together with a significant clinical benefit measured by ADCOMS at 6 and 12 months. The drug was well-tolerated with an incidence of ARIA-E of not more than 10% in any treatment arm and less than 15% in APOE ε4 carriers at the highest dose (116). Assessment of amyloid PET status in patients in an ongoing open-label extension (OLE) of BAN2401-G000-201 revealed that all amyloid-negative, BAN2401-treated individuals entering the OLE were also amyloid negative at OLE baseline, despite subjects being off treatment for 9–52 months (117).

A phase III trial for individuals with preclinical AD and elevated amyloid (AHEAD 3–45 study) is currently underway and participants are being recruited and is expected to be completed in October 2027 (Clinical Trial: NCT04468659).

Aducanumab

Aducanumab (BIIB037) is a recombinant human IgG1 antibody that has been isolated from blood lymphocytes of a healthy donor population of elderly subjects with unusually slow cognitive decline and lack of symptoms of cognitive impairment. Preclinical studies in the Tg2576 mouse model employing chronic dosing of a murine IgG2a/k chimeric aducanumab analogue showed significant reductions of Aβ in both soluble and insoluble protein extracts, as well as significantly reduced Aβ deposits in both the cortex and hippocampus; however, no data on behavioral performance was provided (11). Structural and biochemical analyses revealed that aducanumab binds a linear Aβ epitope comprised of amino acids 3–7 in an extended conformation, discriminating between monomers and higher molecular weight peptide assemblies, based on a strong avidity for epitope-rich aggregates and very weak monomer affinity (118). The linear sequence recognized by aducanumab substantially overlaps with other Aβ antibodies (such as bapineuzumab or gantenerumab), while specific interactions such as critical contacts formed with Phe-4 and His-6, are different and the interaction of Aβ and aducanumab is quite shallow (118).

An initial phase I study investigated the safety, tolerability, and pharmacokinetics of a single ascending aducanumab dose (0.3–60 mg/kg) or placebo in mild-to-moderate AD patients. While doses up to 30 mg/kg were generally well-tolerated, all three patients receiving 60 mg/kg developed serious adverse events (SAEs) of symptomatic ARIA, which completely resolved after several weeks (119).

A subsequent phase Ib, 12-month, double-blind placebo-controlled, multiple ascending-dose (1 – 10 mg/kg) study (PRIME) enrolled 165 patients with a clinical diagnosis of prodromal or mild AD (11). Amyloid PET imaging using florbetapir was used as an adjunct tool to identify and select patients for enrollment (120). Of the 165 dosed patients, 40 discontinued treatment, mainly due to adverse events

or withdrawal of consent. Aducanumab reduced brain Aβ plaques as quantified by florbetapir PET in a dose- and time-dependent manner, with significantly reduced SUVR composite scores in the 3, 6 and 10 mg/kg dose groups after 54 weeks of treatment. A slowing of clinical progression as measured by both the MMSE as well as the CDR-SB was observed in patients receiving the highest dose after 1 year of treatment (11). Although this represents the first study reporting an effect of lowering the brain Aβ load coupled to beneficial effects on cognitive outcomes, the small sample size, a staggered parallel-group design and potential unblinding due to ARIA-E in the treatment groups receiving higher antibody doses, impede interpretation of the results. In addition, the clinical stage of dropouts might bear a potential interpretation bias. Similar discontinuation rates were reported among prodromal and mild AD patients in the placebo group; however, more mild than prodromal AD patients at baseline dropped out in the 10 mg/kg group, with a potential impact on the observed slower cognitive decline (121).

Two large 18-month, randomized, double-blind, placebo-controlled phase III trials (ENGAGE & EMERGE) evaluated aducanumab in patients with early AD and MCI due to AD with PET-confirmed amyloid pathology (122). The participants were randomized to receive a low dose (3 mg/kg for ApoE ε4 carriers, 6 mg/kg for non-carriers) or a high-dose of 6 or 10 mg/kg for 78 weeks. The protocol has been amended during the course of the study, allowing ApoE ε4 carriers to receive up to 10 mg/kg and increasing the sample size of each trial to 1650 to compensate for larger than expected standard deviation. A planned futility analysis indicated little chance of treatment efficacy and the trials were terminated in March 2019 (123). Later in 2019, analyses of a more complete data set from both studies were presented, with 29% of patients in EMERGE and 22% in ENGAGE receiving the full possible 14 doses of 10 mg/kg and final participant numbers of 982 and 1084 respectively (124). In EMERGE, the high dose aducanumab group showed a significant 23% reduction in decline on the CDR-SB and 27% reduction on the AD Assessment Scale-Cognitive Subscale 13 items (ADAS-Cog 13) compared to placebo; however, only a 2% reduction on CDR-SB and a 12% reduction in ADAS-Cog 13 were observed in the high-dose group in the ENGAGE sister trial (124). This was explained by the greater exposure to high-dose aducanumab in the EMERGE trial; however, other possibilities such as greater worsening in the placebo group are conceivable as well (125). On July 8, 2020 Biogen announced that it had completed the submission of a Biologics License Application (BLA) to the U.S. Food and Drug Administration (FDA) for the approval of aducanumab (126). On August 7, 2020, Biogen announced that the agency accepted this BLA granting priority review, which means that the time to review is cut down to 6 months. In case of successful approval, aducanumab will be the first approved biological capable of removing amyloid plaques.

MEDI1814

MEDI1814 is a fully human monoclonal IgG1λ antibody targeting the C-terminus of $A\beta_{42}$, with a triple mutation in the Fc tail to reduce its effector function. It aims to bind and remove monomers and low n-oligomers from circulation, thus preventing further aggregation of the peptide (127). MEDI1814 showed a dose-dependent suppression of up to 90% of free $A\beta_{42}$ in the CSF of V717I transgenic mice, naıve rats and cynomolgus monkeys (128).

AstraZeneca started a clinical trial in the United States in 2014 testing single and multiple ascending dose in subjects with mild-to-moderate AD. Safety, tolerability, pharmacokinetics, and pharmacodynamics were analyzed and none of the participants on the drug showed signs of ARIAs. In addition, pharmacokinetics and pharmacodynamics data provided evidence of dose-dependent and selective $A\beta_{42}$ target engagement in the CNS (129).

SAR228810

SAR228810 is a humanized version of murine IgG1 SAR255952 antibody with an engineered human IgG4 backbone with two amino-acid substitutions to reduce the Fc effector function-dependent risk of ARIAs. It binds specifically to soluble protofibrillar and fibrillar forms of Aβ and it is relatively inactive against Aβ monomers and small oligomeric aggregates (130). Co-application of SAR228810 and oligomeric $A\beta_{42}$ preparations significantly inhibited Aβ-induced neurotoxicity in primary neurons (131). Preclinical pharmacological studies of SAR255952 in APPSL mice showed that a chronic 4-month treatment dose-dependently prevented brain amyloid plaque formation. Even with high doses (up to 50 mg/kg/week intravenously), SAR255952 did not increase brain micro-hemorrhages in old mice. In immunotolerized APPSL mice, in which $CD4^+$ T lymphocytes have been transiently depleted, SAR228810 demonstrated the same efficacy as its murine precursor (130, 132).

A multi-center, double-blind, placebo-controlled phase I clinical trial testing escalating single and multiple doses by Sanofi has been completed and no further clinical trials are ongoing at the moment. SAR228810 has been administered by intravenous infusion or subcutaneous injection in patients with mild-to-moderate AD.

CONCLUSION

A multitude of preclinical biochemical, histopathological and animal studies, as well as a large number of genetic, biomarker and clinical reports support the central role of Aβ in AD pathogenesis. While the amyloid cascade hypothesis, with all its modifications, is still considered relevant, the continuous failures of late stage clinical trials with immunotherapy approaches raise questions about considering the right target. There is increasing evidence that Aβ peptides might also play important physiological roles, as neurotrophic effects (133) or improved synaptic function after application of picomolar Aβ concentrations in mice depleted of endogenous Aβ have been described (134, 135). The observation that Aβ is elevated in the CSF after sleep deprivation in healthy adults (136), together with its increased brain levels in a variety of other neurologic disease conditions such as traumatic brain injury (137) or cerebrovascular lesions (138) may indicate, that Aβ production in the case of neuronal stress or damage might represent response rather than origin. While immunotherapy trials targeting Aβ have been regarded as the final proof of the validity of the amyloid cascade hypothesis, the aforementioned studies still paint a nebulous picture and alternative therapeutic strategies and approaches should be vigorously investigated.

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REFERENCES

1. World Alzheimer Report 2019. Attitudes to dementia. London: Alzheimer's Disease International; 2019.
2. Panza F, Lozupone M, Logroscino G, Imbimbo BP. A critical appraisal of amyloid-β-targeting therapies for Alzheimer disease. Nat Rev Neurol. 2019;15(2):73–88. http://dx.doi.org/10.1038/s41582-018-0116-6
3. Hansen RA, Gartlehner G, Webb AP, Morgan LC, Moore CG, Jonas DE. Efficacy and safety of donepezil, galantamine, and rivastigmine for the treatment of Alzheimer's disease: A systematic review and meta-analysis. Clin Interv Aging. 2008;3(2):211–25.
4. Kaduszkiewicz H, Zimmermann T, Beck-Bornholdt H-P, van den Bussche H. Cholinesterase inhibitors for patients with Alzheimer's disease: Systematic review of randomised clinical trials. BMJ. 2005;331(7512):321–7. http://dx.doi.org/10.1136/bmj.331.7512.321
5. Hardy J, Allsop D. Amyloid deposition as the central event in the aetiology of Alzheimer's disease. Trends Pharmacol Sci. 1991;12(10):383–8. http://dx.doi.org/10.1016/0165-6147(91)90609-V
6. Hardy J. The amyloid hypothesis for Alzheimer's disease: A critical reappraisal. J Neurochem. 2009;110(4):1129–34. http://dx.doi.org/10.1111/j.1471-4159.2009.06181.x
7. Giannakopoulos P, Herrmann FR, Bussiere T, Bouras C, Kovari E, Perl DP, et al. Tangle and neuron numbers, but not amyloid load, predict cognitive status in Alzheimer's disease. Neurology. 2003;60(9):1495–500. http://dx.doi.org/10.1212/01.WNL.0000063311.58879.01
8. Wirths O, Multhaup G, Bayer TA. A modified beta-amyloid hypothesis: Intraneuronal accumulation of the beta-amyloid peptide – The first step of a fatal cascade. J Neurochem. 2004;91(3):513–20. http://dx.doi.org/10.1111/j.1471-4159.2004.02737.x
9. Selkoe DJ, Hardy J. The amyloid hypothesis of Alzheimer's disease at 25 years. EMBO Mol Med. 2016;8(6):595–608. http://dx.doi.org/10.15252/emmm.201606210
10. Liu E, Schmidt ME, Margolin R, Sperling R, Koeppe R, Mason NS, et al. Amyloid-β 11C-PiB-PET imaging results from 2 randomized bapineuzumab phase 3 AD trials. Neurology. 2015;85(8):692–700. http://dx.doi.org/10.1212/WNL.0000000000001877
11. Sevigny J, Chiao P, Bussière T, Weinreb PH, Williams L, Maier M, et al. The antibody aducanumab reduces Aβ plaques in Alzheimer's disease. Nature. 2016;537(7618):50–6. http://dx.doi.org/10.1038/nature19323
12. Penninkilampi R, Brothers HM, Eslick GD. Safety and efficacy of anti-amyloid-β immunotherapy in Alzheimer's disease: A systematic review and meta-analysis. J Neuroimmune Pharmacol. 2017;12(1):194–203. http://dx.doi.org/10.1007/s11481-016-9722-5
13. Schellenberg G, Montine T. The genetics and neuropathology of Alzheimer's disease. Acta Neuropathol. 2012;124(3):305–23. http://dx.doi.org/10.1007/s00401-012-0996-2

14. Tanzi RE. The genetics of Alzheimer disease. Cold Spring Harb Perspect Med. 2012;2(10):a006296. http://dx.doi.org/10.1101/cshperspect.a006296
15. Dourlen P, Kilinc D, Malmanche N, Chapuis J, Lambert J-C. The new genetic landscape of Alzheimer's disease: From amyloid cascade to genetically driven synaptic failure hypothesis? Acta Neuropathol. 2019;138(2):221–36. http://dx.doi.org/10.1007/s00401-019-02004-0
16. Nhan HS, Chiang K, Koo EH. The multifaceted nature of amyloid precursor protein and its proteolytic fragments: Friends and foes. Acta Neuropathol. 2015;129(1):1–19. http://dx.doi.org/10.1007/s00401-014-1347-2
17. Vassar R, Bennett BD, Babu-Khan S, Kahn S, Mendiaz EA, Denis P, et al. Beta-secretase cleavage of Alzheimer's amyloid precursor protein by the transmembrane aspartic protease BACE. Science. 1999;286(5440):735–41. http://dx.doi.org/10.1126/science.286.5440.735
18. Golde TE, Koo EH, Felsenstein KM, Osborne BA, Miele L. Gamma-secretase inhibitors and modulators. Biochim Biophys Acta. 2013;1828(12):2898–907. http://dx.doi.org/10.1016/j.bbamem.2013.06.005
19. Dovey HF, Suomensaari-Chrysler S, Lieberburg I, Sinha S, Keim PS. Cells with a familial Alzheimer's disease mutation produce authentic beta-peptide. Neuroreport. 1993;4(8):1039–42. http://dx.doi.org/10.1097/00001756-199308000-00011
20. Steiner H, Fukumori A, Tagami S, Okochi M. Making the final cut: Pathogenic amyloid-β peptide generation by γ-secretase. Cell Stress. 2018;2(11):292–310. http://dx.doi.org/10.15698/cst2018.11.162
21. Bayer TA, Wirths O. Focusing the amyloid cascade hypothesis on N-truncated Aβ peptides as drug targets against Alzheimer's disease. Acta Neuropathol. 2014;127(6):787–801. http://dx.doi.org/10.1007/s00401-014-1287-x
22. Dunys J, Valverde A, Checler F. Are N- and C-terminally truncated Aβ species key pathological triggers in Alzheimer's disease? J Biol Chem. 2018;293(40):15419–28. http://dx.doi.org/10.1074/jbc.R118.003999
23. Wirths O, Zampar S. Emerging roles of N- and C-terminally truncated Aβ species in Alzheimer's disease. Expert Opin Ther Targets. 2019;23(12):991–1004. http://dx.doi.org/10.1080/14728222.2019.1702972
24. Chabrier MA, Blurton-Jones M, Agazaryan AA, Nerhus JL, Martinez-Coria H, Laferla FM. Soluble abeta promotes wild-type tau pathology in vivo. J Neurosci. 2012;32(48):17345–50. http://dx.doi.org/10.1523/JNEUROSCI.0172-12.2012
25. McLean CA, Cherny RA, Fraser FW, Fuller SJ, Smith MJ, Beyreuther K, et al. Soluble pool of Abeta amyloid as a determinant of severity of neurodegeneration in Alzheimer's disease. Ann Neurol. 1999 Dec;46(6):860–6. http://dx.doi.org/10.1002/1531-8249(199912)46:6%3C860::AID-ANA8%3E3.0.CO;2-M
26. Shankar GM, Bloodgood BL, Townsend M, Walsh DM, Selkoe DJ, Sabatini BL. Natural oligomers of the Alzheimer amyloid-beta protein induce reversible synapse loss by modulating an NMDA-type glutamate receptor-dependent signaling pathway. J Neurosci. 2007;27(11):2866–75. http://dx.doi.org/10.1523/JNEUROSCI.4970-06.2007
27. Selkoe DJ. Soluble oligomers of the amyloid beta-protein impair synaptic plasticity and behavior. Behav Brain Res. 2008;192(1):106–13. http://dx.doi.org/10.1016/j.bbr.2008.02.016
28. Benilova I, Karran E, De Strooper B. The toxic Abeta oligomer and Alzheimer's disease: An emperor in need of clothes. Nat Neurosci. 2012;15(3):349–57. http://dx.doi.org/10.1038/nn.3028
29. Kayed R, Pensalfini A, Margol L, Sokolov Y, Sarsoza F, Head E, et al. Annular protofibrils are a structurally and functionally distinct type of amyloid oligomer. J Biol Chem. 2009;284(7):4230–7. http://dx.doi.org/10.1074/jbc.M808591200
30. Janeway CAJ, Travers P, Walport M, Shlomchik MJ. Immunobiology: The immune system in health and disease. 5th ed. New York: Garland Science; 2001.
31. Lannfelt L, Relkin NR, Siemers ER. Amyloid-β-directed immunotherapy for Alzheimer's disease. J Int Med. 2014;275(3):284–95. http://dx.doi.org/10.1111/joim.12168
32. DeMattos RB, Bales KR, Cummins DJ, Dodart JC, Paul SM, Holtzman DM. Peripheral anti-Abeta antibody alters CNS and plasma Abeta clearance and decreases brain Abeta burden in a mouse model of Alzheimer's disease. Proc Natl Acad Sci U S A. 2001;98(15):8850–5. http://dx.doi.org/10.1073/pnas.151261398
33. Wisniewski T, Konietzko U. Amyloid-beta immunisation for Alzheimer's disease. Lancet Neurol. 2008;7(9):805–11. http://dx.doi.org/10.1016/S1474-4422(08)70170-4

34. Bacskai BJ, Kajdasz ST, Christie RH, Carter C, Games D, Seubert P, et al. Imaging of amyloid-β deposits in brains of living mice permits direct observation of clearance of plaques with immunotherapy. Nat Med. 2001;7(3):369–72. http://dx.doi.org/10.1038/85525
35. Solomon B, Koppel R, Hanan E, Katzav T. Monoclonal antibodies inhibit in vitro fibrillar aggregation of the Alzheimer beta-amyloid peptide. Proc Nat Acad Sci. 1996;93(1):452–5. http://dx.doi.org/10.1073/pnas.93.1.452
36. Frenkel D, Solomon B, Benhar I. Modulation of Alzheimer's β-amyloid neurotoxicity by site-directed single-chain antibody. J Neuroimmunol. 2000;106(1):23–31. http://dx.doi.org/10.1016/S0165-5728(99)00232-5
37. Brody DL, Holtzman DM. Active and passive immunotherapy for neurodegenerative disorders. Annu Rev Neurosci. 2008;31:175–93. http://dx.doi.org/10.1146/annurev.neuro.31.060407.125529
38. Bacskai BJ, Kajdasz ST, McLellan ME, Games D, Seubert P, Schenk D, et al. Non-Fc-mediated mechanisms are involved in clearance of amyloid-beta in vivo by immunotherapy. J Neurosci. 2002;22(18):7873–8. http://dx.doi.org/10.1523/JNEUROSCI.22-18-07873.2002
39. Taguchi H, Planque S, Nishiyama Y, Symersky J, Boivin S, Szabo P, et al. Autoantibody-catalyzed hydrolysis of amyloid β peptide. J Biol Chem. 2008;283(8):4714–22. http://dx.doi.org/10.1074/jbc.M707983200
40. Schenk D, Barbour R, Dunn W, Gordon G, Grajeda H, Guido T, et al. Immunization with amyloid-beta attenuates Alzheimer-disease-like pathology in the PDAPP mouse. Nature. 1999;400(6740):173–7. http://dx.doi.org/10.1038/22124
41. Orgogozo JM, Gilman S, Dartigues JF, Laurent B, Puel M, Kirby LC, et al. Subacute meningoencephalitis in a subset of patients with AD after Abeta42 immunization. Neurology. 2003;61(1):46–54. http://dx.doi.org/10.1212/01.WNL.0000073623.84147.A8
42. Gilman S, Koller M, Black RS, Jenkins L, Griffith SG, Fox NC, et al. Clinical effects of Aβ immunization (AN1792) in patients with AD in an interrupted trial. Neurology. 2005;64(9):1553–62. http://dx.doi.org/10.1212/01.WNL.0000159740.16984.3C
43. Holmes C, Boche D, Wilkinson D, Yadegarfar G, Hopkins V, Bayer A, et al. Long-term effects of Abeta42 immunisation in Alzheimer's disease: Follow-up of a randomised, placebo-controlled phase I trial. Lancet. 2008;372(9634):216–23. http://dx.doi.org/10.1016/S0140-6736(08)61075-2
44. Boche D, Zotova E, Weller RO, Love S, Neal JW, Pickering RM, et al. Consequence of Abeta immunization on the vasculature of human Alzheimer's disease brain. Brain. 2008;131(Pt 12):3299–310. http://dx.doi.org/10.1093/brain/awn261
45. Sakai K, Boche D, Carare R, Johnston D, Holmes C, Love S, et al. Aβ immunotherapy for Alzheimer's disease: Effects on apoE and cerebral vasculopathy. Acta Neuropathol. 2014;128(6):777–89. http://dx.doi.org/10.1007/s00401-014-1340-9
46. Paquet C, Amin J, Mouton-Liger F, Nasser M, Love S, Gray F, et al. Effect of active Aβ immunotherapy on neurons in human Alzheimer's disease. J Pathol. 2015;235(5):721–30. http://dx.doi.org/10.1002/path.4491
47. Nicoll JAR, Buckland GR, Harrison CH, Page A, Harris S, Love S, et al. Persistent neuropathological effects 14 years following amyloid-β immunization in Alzheimer's disease. Brain. 2019;142(7):2113–26. http://dx.doi.org/10.1093/brain/awz142
48. Sterner RM, Takahashi PY, Yu Ballard AC. Active vaccines for Alzheimer disease treatment. J Am Med Dir Assoc. 2016;17(9):862.e11–15. http://dx.doi.org/10.1016/j.jamda.2016.06.009
49. Ketter N, Liu E, Di J, Honig LS, Lu M, Novak G, et al. A randomized, double-blind, phase 2 study of the effects of the vaccine vanutide cridificar with QS-21 adjuvant on immunogenicity, safety and amyloid imaging in patients with mild to moderate Alzheimer's disease. J Prev Alzheimers Dis. 2016;3(4):192–201.
50. Pasquier F, Sadowsky C, Holstein A, Leterme GLP, Peng Y, Jackson N, et al. Two phase 2 multiple ascending–dose studies of vanutide cridificar (ACC-001) and QS-21 adjuvant in mild-to-moderate Alzheimer's disease. J Alzheimers Dis. 2016;51:1131–43. http://dx.doi.org/10.3233/JAD-150376
51. Farlow MR, Andreasen N, Riviere ME, Vostiar I, Vitaliti A, Sovago J, et al. Long-term treatment with active Aβ immunotherapy with CAD106 in mild Alzheimer's disease. Alzheimers Res Ther. 2015;7(1):23. http://dx.doi.org/10.1186/s13195-015-0108-3
52. Vandenberghe R, Riviere M-E, Caputo A, Sovago J, Maguire RP, Farlow M, et al. Active Aβ immunotherapy CAD106 in Alzheimer's disease: A phase 2b study. Alzheimers Dementia. 2016;3(1):10–22. http://dx.doi.org/10.1016/j.trci.2016.12.003

53. Lacosta A-M, Pascual-Lucas M, Pesini P, Casabona D, Pérez-Grijalba V, Marcos-Campos I, et al. Safety, tolerability and immunogenicity of an active anti-Aβ40 vaccine (ABvac40) in patients with Alzheimer's disease: A randomised, double-blind, placebo-controlled, phase I trial. Alzheimers Res Ther. 2018;10(1):12. http://dx.doi.org/10.1186/s13195-018-0340-8
54. Muhs A, Hickman DT, Pihlgren M, Chuard N, Giriens V, Meerschman C, et al. Liposomal vaccines with conformation-specific amyloid peptide antigens define immune response and efficacy in APP transgenic mice. Proc Nat Acad Sci. 2007;104(23):9810–15. http://dx.doi.org/10.1073/pnas.0703137104
55. Mandler M, Santic R, Gruber P, Cinar Y, Pichler D, Funke SA, et al. Tailoring the antibody response to aggregated Aß using novel Alzheimer-vaccines. PLoS One. 2015;10(1):e0115237. http://dx.doi.org/10.1371/journal.pone.0115237
56. Schneeberger A, Hendrix S, Mandler M, Ellison N, Burger V, Brunner M, et al. Results from a Phase II study to assess the clinical and immunological activity of AFFITOPE(R) AD02 in patients with early Alzheimer's disease. J Prev Alzheimers Dis. 2015;2(2):103–14.
57. Wang CY, Wang P-N, Chiu M-J, Finstad CL, Lin F, Lynn S, et al. UB-311, a novel UBITh(®) amyloid β peptide vaccine for mild Alzheimer's disease. Alzheimers Dementia. 2017;3(2):262–72. http://dx.doi.org/10.1016/j.trci.2017.03.005
58. United Neuroscience announces positive top-line results from Phase 2a clinical study of UB-311 vaccine in patients with Alzheimer's disease. United Neuroscience, Press Release. 2019:Jan 16, 2019.
59. Mallbris L, Davies J, Glasebrook A, Tang Y, Glaesner W, Nickoloff BJ. Molecular insights into fully human and humanized monoclonal antibodies: What are the differences and should dermatologists care? J Clin Aesthet Dermatol. 2016;9(7):13–15.
60. Nelson AL, Dhimolea E, Reichert JM. Development trends for human monoclonal antibody therapeutics. Nature Rev Drug Discov. 2010;9(10):767–74. http://dx.doi.org/10.1038/nrd3229
61. van Dyck CH. Anti-amyloid-β monoclonal antibodies for Alzheimer's disease: Pitfalls and promise. Biol Psychiatry. 2018;83(4):311–19. http://dx.doi.org/10.1016/j.biopsych.2017.08.010
62. Sperling RA, Jack CR, Black SE, Frosch MP, Greenberg SM, Hyman BT, et al. Amyloid-related imaging abnormalities in amyloid-modifying therapeutic trials: Recommendations from the Alzheimer's Association Research Roundtable Workgroup. Alzheimers Dementia. 2011;7(4):367–85. http://dx.doi.org/10.1016/j.jalz.2011.05.2351
63. DiFrancesco J, Longoni M, Piazza F. Anti-Aβ autoantibodies in amyloid related imaging abnormalities (ARIA): Candidate biomarker for immunotherapy in Alzheimer's disease and cerebral amyloid angiopathy. Front Neurol. 2015;6:207. http://dx.doi.org/10.3389/fneur.2015.00207
64. Piazza F, Winblad B. Amyloid-Related Imaging Abnormalities (ARIA) in immunotherapy trials for Alzheimer's disease: Need for prognostic biomarkers? J Alzheimers Dis. 2016;52(2):417–20. http://dx.doi.org/10.3233/JAD-160122
65. Wilcock DM, Colton CA. Immunotherapy, vascular pathology, and microhemorrhages in transgenic mice. CNS Neurol Disord Drug Targets. 2009;8(1):50–64. http://dx.doi.org/10.2174/187152709787601858
66. Bruhns P. Properties of mouse and human IgG receptors and their contribution to disease models. Blood. 2012;119(24):5640–9. http://dx.doi.org/10.1182/blood-2012-01-380121
67. Freeman GB, Brown TP, Wallace K, Bales KR. Chronic administration of an aglycosylated murine antibody of ponezumab does not worsen microhemorrhages in aged Tg2576 mice. Curr Alzheimer Res. 2012;9(9):1059–68. http://dx.doi.org/10.2174/156720512803569064
68. Fuller JP, Stavenhagen JB, Christensen S, Kartberg F, Glennie MJ, Teeling JL. Comparing the efficacy and neuroinflammatory potential of three anti-abeta antibodies. Acta Neuropathol. 2015;130(5):699–711. http://dx.doi.org/10.1007/s00401-015-1484-2
69. Miles LA, Crespi GA, Doughty L, Parker MW. Bapineuzumab captures the N-terminus of the Alzheimer's disease amyloid-beta peptide in a helical conformation. Sci Rep. 2013;3:1302. http://dx.doi.org/10.1038/srep01302
70. Zampar S, Klafki HW, Sritharen K, Bayer TA, Wiltfang J, Rostagno A, et al. N-terminal heterogeneity of parenchymal and vascular amyloid-β deposits in Alzheimer's disease. Neuropathol Appl Neurobiol. 2020;46:673–85. http://dx.doi.org/10.1111/nan.12637
71. Salloway S, Sperling R, Fox NC, Blennow K, Klunk W, Raskind M, et al. Two phase 3 trials of bapineuzumab in mild-to-moderate Alzheimer's disease. New Engl J Med. 2014;370(4):322–33. http://dx.doi.org/10.1056/NEJMoa1304839

72. Seubert P, Barbour R, Khan K, Motter R, Tang P, Kholodenko D, et al. Antibody capture of soluble Aβ does not reduce cortical Aβ amyloidosis in the PDAPP mouse. Neurodegen Dis. 2008;5(2):65–71. http://dx.doi.org/10.1159/000112834
73. Karran E, Hardy J. A critique of the drug discovery and phase 3 clinical programs targeting the amyloid hypothesis for Alzheimer disease. Ann Neurol. 2014;76(2):185–205. http://dx.doi.org/10.1002/ana.24188
74. Black RS, Sperling RA, Safirstein B, Motter RN, Pallay A, Nichols A, et al. A single ascending dose study of bapineuzumab in patients with Alzheimer disease. Alzheimer Dis Assoc Disord. 2010;24(2):198–203. http://dx.doi.org/10.1097/WAD.0b013e3181c53b00
75. Blennow K, Zetterberg H, Rinne JO, Salloway S, Wei J, Black R, et al. Effect of immunotherapy with bapineuzumab on cerebrospinal fluid biomarker levels in patients with mild to moderate Alzheimer disease. Arch Neurol. 2012;69(8):1002–10. http://dx.doi.org/10.1001/archneurol.2012.90
76. Rinne JO, Brooks DJ, Rossor MN, Fox NC, Bullock R, Klunk WE, et al. 11C-PiB PET assessment of change in fibrillar amyloid-beta load in patients with Alzheimer's disease treated with bapineuzumab: A phase 2, double-blind, placebo-controlled, ascending-dose study. Lancet Neurol. 2010;9(4):363–72. http://dx.doi.org/10.1016/S1474-4422(10)70043-0
77. Adolfsson O, Pihlgren M, Toni N, Varisco Y, Buccarello AL, Antoniello K, et al. An effector-reduced anti-β-amyloid (Aβ) antibody with unique Aβ binding properties promotes neuroprotection and glial engulfment of Aβ. J Neurosci. 2012;32(28):9677–89. http://dx.doi.org/10.1523/JNEUROSCI.4742-11.2012
78. Zhao J, Nussinov R, Ma B. Mechanisms of recognition of Aβ monomer, oligomer, and fibril by homologous antibodies. J Biol Chem. 2017;292(44):18325–43. http://dx.doi.org/10.1074/jbc.M117.801514
79. Cummings JL, Cohen S, van Dyck CH, Brody M, Curtis C, Cho W, et al. ABBY – A phase 2 randomized trial of crenezumab in mild to moderate Alzheimer disease. Neurology. 2018;90(21):e1889–e97. http://dx.doi.org/10.1212/WNL.0000000000005550
80. Salloway S, Honigberg LA, Cho W, Ward M, Friesenhahn M, Brunstein F, et al. Amyloid positron emission tomography and cerebrospinal fluid results from a crenezumab anti-amyloid-beta antibody double-blind, placebo-controlled, randomized phase II study in mild-to-moderate Alzheimer's disease (BLAZE). Alzheimers Res Ther. 2018;10(1):96. http://dx.doi.org/10.1186/s13195-018-0424-5
81. Blaettler T, Smith J, Smith J, Paul R, Asnaghi V, Fuji R, et al. P2-003: Clinical trial design of cread: A randomized, double-blind, placebo-controlled, parallel-group phase 3 study to evaluate crenezumab treatment in patients with prodromal-to-mild Alzheimer's disease. Alzheimers Dementia. 2016;12:P609. http://dx.doi.org/10.1016/j.jalz.2016.06.1207
82. Tariot PN, Lopera F, Langbaum JB, Thomas RG, Hendrix S, Schneider LS, et al. The Alzheimer's prevention initiative autosomal-dominant Alzheimer's disease trial: A study of crenezumab versus placebo in preclinical PSEN1 E280A mutation carriers to evaluate efficacy and safety in the treatment of autosomal-dominant Alzheimer's disease, including a placebo-treated noncarrier cohort. Alzheimers Dementia. 2018;4:150–60. http://dx.doi.org/10.1016/j.trci.2018.02.002
83. Bohrmann B, Baumann K, Benz J, Gerber F, Huber W, Knoflach F, et al. Gantenerumab: A novel human anti-abeta antibody demonstrates sustained cerebral amyloid-beta binding and elicits cell-mediated removal of human amyloid-beta. J Alzheimers Dis. 2011;28(1):49–69. http://dx.doi.org/10.3233/JAD-2011-110977
84. Ostrowitzki S, Deptula D, Thurfjell L, Barkhof F, Bohrmann B, Brooks DJ, et al. Mechanism of amyloid removal in patients with Alzheimer disease treated with gantenerumab. Arch Neurol. 2012;69(2):198–207. http://dx.doi.org/10.1001/archneurol.2011.1538
85. Ostrowitzki S, Lasser RA, Dorflinger E, Scheltens P, Barkhof F, Nikolcheva T, et al. A phase III randomized trial of gantenerumab in prodromal Alzheimer's disease. Alzheimers Res Ther. 2017;9(1):95. http://dx.doi.org/10.1186/s13195-017-0318-y
86. Klein G, Delmar P, Voyle N, Rehal S, Hofmann C, Abi-Saab D, et al. Gantenerumab reduces amyloid-β plaques in patients with prodromal to moderate Alzheimer's disease: A PET substudy interim analysis. Alzheimers Res Ther. 2019;11(1):101. http://dx.doi.org/10.1186/s13195-019-0559-z
87. Portron A, Jordan P, Draper K, Muenzer C, Dickerson D, van Iersel T, et al. A phase I study to assess the effect of speed of injection on pain, tolerability, and pharmacokinetics after high-volume

subcutaneous administration of gantenerumab in healthy volunteers. Clin Ther. 2020;42(1):108–20. http://dx.doi.org/10.1016/j.clinthera.2019.11.015
88. La Porte SL, Bollini SS, Lanz TA, Abdiche YN, Rusnak AS, Ho WH, et al. Structural basis of C-terminal beta-amyloid peptide binding by the antibody ponezumab for the treatment of Alzheimer's disease. J Mol Biol. 2012;421(4–5):525–36. http://dx.doi.org/10.1016/j.jmb.2011.11.047
89. Karlnoski RA, Rosenthal A, Kobayashi D, Pons J, Alamed J, Mercer M, et al. Suppression of amyloid deposition leads to long-term reductions in Alzheimer's pathologies in Tg2576 mice. J Neurosci. 2009;29(15):4964–71. http://dx.doi.org/10.1523/JNEUROSCI.4560-08.2009
90. Freeman GB, Lin JC, Pons J, Raha NM. 39-week toxicity and toxicokinetic study of ponezumab (PF-04360365) in cynomolgus monkeys with 12-week recovery period. J Alzheimers Dis. 2012;28(3):531–41. http://dx.doi.org/10.3233/JAD-2011-110869
91. Landen JW, Zhao Q, Cohen S, Borrie M, Woodward M, Billing CBJ, et al. Safety and pharmacology of a single intravenous dose of ponezumab in subjects with mild-to-moderate Alzheimer disease: A phase I, randomized, placebo-controlled, double-blind, dose-escalation study. Clin Neuropharmacol. 2013;36(1):14–23. http://dx.doi.org/10.1097/WNF.0b013e31827db49b
92. Miyoshi I, Fujimoto Y, Yamada M, Abe S, Zhao Q, Cronenberger C, et al. Safety and pharmacokinetics of PF-04360365 following a single-dose intravenous infusion in Japanese subjects with mild-to-moderate Alzheimer's disease: A multicenter, randomized, double-blind, placebo-controlled, dose-escalation study. Int J Clin Pharmacol Ther. 2013;51(12):911–23. http://dx.doi.org/10.5414/CP201816
93. Burstein AH, Zhao Q, Ross J, Styren S, Landen JW, Ma WW, et al. Safety and pharmacology of ponezumab (PF-04360365) after a single 10-minute intravenous infusion in subjects with mild to moderate Alzheimer disease. Clin Neuropharmacol. 2013;36(1):8–13. http://dx.doi.org/10.1097/WNF.0b013e318279bcfa
94. Landen JW, Cohen S, Billing CB, Cronenberger C, Styren S, Burstein AH, et al. Multiple-dose ponezumab for mild-to-moderate Alzheimer's disease: Safety and efficacy. Alzheimers Dementia. 2017;3(3):339–47. http://dx.doi.org/10.1016/j.trci.2017.04.003
95. Landen JW, Andreasen N, Cronenberger CL, Schwartz PF, Börjesson-Hanson A, Östlund H, et al. Ponezumab in mild-to-moderate Alzheimer's disease: Randomized phase II PET-PIB study. Alzheimers Dementia. 2017;3(3):393–401. http://dx.doi.org/10.1016/j.trci.2017.05.003
96. Leurent C, Goodman JA, Zhang Y, He P, Polimeni JR, Gurol ME, et al. Immunotherapy with ponezumab for probable cerebral amyloid angiopathy. Ann Clin Transl Neurol. 2019;6(4):795–806. http://dx.doi.org/10.1002/acn3.761
97. Revesz T, Ghiso J, Lashley T, Plant G, Rostagno A, Frangione B, et al. Cerebral amyloid angiopathies: A pathologic, biochemical, and genetic view. J Neuropathol Exp Neurol. 2003;62(9):885–98. http://dx.doi.org/10.1093/jnen/62.9.885
98. Attems J, Jellinger K, Thal DR, Van Nostrand W. Review: Sporadic cerebral amyloid angiopathy. Neuropathol Appl Neurobiol. 2011;37(1):75–93. http://dx.doi.org/10.1111/j.1365-2990.2010.01137.x
99. Crespi GAN, Hermans SJ, Parker MW, Miles LA. Molecular basis for mid-region amyloid-β capture by leading Alzheimer's disease immunotherapies. Sci Rep. 2015;5(1):9649. http://dx.doi.org/10.1038/srep09649
100. Siemers ER, Friedrich S, Dean RA, Gonzales CR, Farlow MR, Paul SM, et al. Safety and changes in plasma and cerebrospinal fluid amyloid β after a single administration of an amyloid β monoclonal antibody in subjects with Alzheimer disease. Clin Neuropharmacol. 2010;33(2):67–73. http://dx.doi.org/10.1097/WNF.0b013e3181cb577a
101. Mably AJ, Liu W, Mc Donald JM, Dodart J-C, Bard F, Lemere CA, et al. Anti-Aβ antibodies incapable of reducing cerebral Aβ oligomers fail to attenuate spatial reference memory deficits in J20 mice. Neurobiol Dis. 2015;82:372–84. http://dx.doi.org/10.1016/j.nbd.2015.07.008
102. Dodart J-C, Bales KR, Gannon KS, Greene SJ, DeMattos RB, Mathis C, et al. Immunization reverses memory deficits without reducing brain Aβ burden in Alzheimer's disease model. Nat Neurosci. 2002;5:452–7. http://dx.doi.org/10.1038/nn842
103. Bouter Y, Noguerola JS, Tucholla P, Crespi GA, Parker MW, Wiltfang J, et al. Abeta targets of the biosimilar antibodies of Bapineuzumab, Crenezumab, Solanezumab in comparison to an antibody

against N-truncated Abeta in sporadic Alzheimer disease cases and mouse models. Acta Neuropathol. 2015;130:713–29. http://dx.doi.org/10.1007/s00401-015-1489-x
104. Farlow M, Arnold SE, van Dyck CH, Aisen PS, Snider BJ, Porsteinsson AP, et al. Safety and biomarker effects of solanezumab in patients with Alzheimer's disease. Alzheimers Dementia. 2012;8(4):261–71. http://dx.doi.org/10.1016/j.jalz.2011.09.224
105. Doody RS, Thomas RG, Farlow M, Iwatsubo T, Vellas B, Joffe S, et al. Phase 3 trials of solanezumab for mild-to-moderate Alzheimer's disease. New Engl J Med. 2014;370(4):311–21. http://dx.doi.org/10.1056/NEJMoa1312889
106. Siemers ER, Sundell KL, Carlson C, Case M, Sethuraman G, Liu-Seifert H, et al. Phase 3 solanezumab trials: Secondary outcomes in mild Alzheimer's disease patients. Alzheimers Dementia. 2016;12(2):110–20. http://dx.doi.org/10.1016/j.jalz.2015.06.1893
107. Honig LS, Vellas B, Woodward M, Boada M, Bullock R, Borrie M, et al. Trial of solanezumab for mild dementia due to Alzheimer's disease. New Engl J Med. 2018;378(4):321–30. http://dx.doi.org/10.1056/NEJMoa1705971
108. Englund H, Sehlin D, Johansson AS, Nilsson LN, Gellerfors P, Paulie S, et al. Sensitive ELISA detection of amyloid-beta protofibrils in biological samples. J Neurochem. 2007;103(1):334–45.
109. Sehlin D, Englund H, Simu B, Karlsson M, Ingelsson M, Nikolajeff F, et al. Large aggregates are the major soluble Aβ species in AD brain fractionated with density gradient ultracentrifugation. PLoS One. 2012;7(2):e32014. http://dx.doi.org/10.1371/journal.pone.0032014
110. Sehlin D, Hedlund M, Lord A, Englund H, Gellerfors P, Paulie S, et al. Heavy-chain complementarity-determining regions determine conformation selectivity of anti-Aβ antibodies. Neurodegen Dis. 2011;8(3):117–23. http://dx.doi.org/10.1159/000316530
111. Lord A, Gumucio A, Englund H, Sehlin D, Sundquist VS, Soderberg L, et al. An amyloid-beta protofibril-selective antibody prevents amyloid formation in a mouse model of Alzheimer's disease. Neurobiol Dis. 2009;36(3):425–34. http://dx.doi.org/10.1016/j.nbd.2009.08.007
112. Lord A, Englund H, Soderberg L, Tucker S, Clausen F, Hillered L, et al. Amyloid-beta protofibril levels correlate with spatial learning in Arctic Alzheimer's disease transgenic mice. FEBS J. 2009;276(4):995–1006. http://dx.doi.org/10.1111/j.1742-4658.2008.06836.x
113. Tucker S, Möller C, Tegerstedt K, Lord A, Laudon H, Sjödahl J, et al. The murine version of BAN2401 (mAb158) selectively reduces amyloid-β protofibrils in brain and cerebrospinal fluid of tg-ArcSwe mice. J Alzheimers Dis. 2015;43:575–88. http://dx.doi.org/10.3233/JAD-140741
114. Sehlin D, Fang XT, Cato L, Antoni G, Lannfelt L, Syvänen S. Antibody-based PET imaging of amyloid beta in mouse models of Alzheimer's disease. Nat Commun. 2016;7(1):10759. http://dx.doi.org/10.1038/ncomms10759
115. Logovinsky V, Satlin A, Lai R, Swanson C, Kaplow J, Osswald G, et al. Safety and tolerability of BAN2401 – a clinical study in Alzheimer's disease with a protofibril selective Aβ antibody. Alzheimers Res Ther. 2016;8(1):14. http://dx.doi.org/10.1186/s13195-016-0181-2
116. Osswald G. BioArctic announces positive topline results of BAN2401 phase 2b at 18 months in early Alzheimer's disease. BioArctic Press Release. 2018:Jul 6, 2018.
117. Swanson CJ, Zhang Y, Dhadda S, Wang J, Kaplow J, Bradley H, et al. Persistence of BAN2401-mediated amyloid reductions post-treatment: A preliminary comparison of amyloid status between the core phase of BAN2401-G000-201 and baseline of the open-label extension phase in subjects with early Alzheimer's disease (1330). Neurology. 2020;94(15 Suppl.):1330.
118. Arndt JW, Qian F, Smith BA, Quan C, Kilambi KP, Bush MW, et al. Structural and kinetic basis for the selectivity of aducanumab for aggregated forms of amyloid-β. Sci Rep. 2018;8(1):6412. http://dx.doi.org/10.1038/s41598-018-24501-0
119. Ferrero J, Williams L, Stella H, Leitermann K, Mikulskis A, O'Gorman J, et al. First-in-human, double-blind, placebo-controlled, single-dose escalation study of aducanumab (BIIB037) in mild-to-moderate Alzheimer's disease. Alzheimers Dementia. 2016;2(3):169–76. http://dx.doi.org/10.1016/j.trci.2016.06.002
120. Sevigny J, Suhy J, Chiao P, Chen T, Klein G, Purcell D, et al. Amyloid PET screening for enrichment of early-stage Alzheimer disease clinical trials: Experience in a phase 1b clinical trial. Alzheimer Dis Assoc Disord. 2016;30(1):1–7. http://dx.doi.org/10.1097/WAD.0000000000000144

121. Panza F, Lozupone M, Seripa D, Imbimbo BP. Amyloid-β immunotherapy for alzheimer disease: Is it now a long shot? Ann Neurol. 2019;85(3):303–15. http://dx.doi.org/10.1002/ana.25410
122. Budd Haeberlein S, O'Gorman J, Chiao P, Bussière T, von Rosenstiel P, Tian Y, et al. Clinical development of aducanumab, an anti-Aβ human monoclonal antibody being investigated for the treatment of early Alzheimer's disease. J Prev Alzheimers Dis. 2017;4(4):255–63.
123. Howard R, Liu KY. Questions EMERGE as Biogen claims aducanumab turnaround. Nat Rev Neurol. 2020;16(2):63–4. http://dx.doi.org/10.1038/s41582-019-0295-9
124. Budd Haeberlein S, Von Hehn C, Tian Y, Chalkias S, Muralidharan KK, Chen T, et al. EMERGE and ENGAGE topline results: Two phase 3 studies to evaluate aducanumab in patients with early Alzheimer's disease [Internet]. San Diego, CA: CTAD; Dec 5, 2019. Available from: http://investors.biogen.com/static-files/ddd45672-9c7e-4c99-8a06-3b557697c06f
125. Schneider L. A resurrection of aducanumab for Alzheimer's disease. Lancet Neurol. 2020;19(2):111–12. http://dx.doi.org/10.1016/S1474-4422(19)30480-6
126. Caouette D. Biogen completes submission of biologics license application to FDA for aducanumab as a treatment for Alzheimer's disease. Biogen Press Release. 2020:July 8, 2020.
127. Bogstedt A, Groves M, Tan K, Narwal R, McFarlane M, Höglund K. Development of immunoassays for the quantitative assessment of amyloid-β in the presence of therapeutic antibody: Application to pre-clinical studies. J Alzheimers Dis. 2015;46:1091–101. http://dx.doi.org/10.3233/JAD-142988
128. Billinton A, Newton P, Lloyd C, Groves M, Welsh F, Bogstedt A, et al. Preclinical discovery and development of MEDI1814, a monoclonal antibody selectively targeting β-amyloid 42 (Aβ42). Alzheimers Dementia. 2017;13:P266. http://dx.doi.org/10.1016/j.jalz.2017.06.141
129. Ostenfeld T, Pomfret M, Billinton A, Chessell I, Chessell T, Lindqvist E, et al. Evaluation of safety, tolerability, pharmacokinetics and pharmacodynamics of MEDI1814, a β-amyloid 42 (Aβ42)-specific antibody, in patients with mild-moderate Alzheimer's disease. Alzheimers Dementia. 2017;13:P574. http://dx.doi.org/10.1016/j.jalz.2017.07.193
130. Pradier L, Blanchard-Brégeon V, Bohme A, Debeir T, Menager J, Benoit P, et al. SAR228810: An antibody for protofibrillar amyloid β peptide designed to reduce the risk of amyloid-related imaging abnormalities (ARIA). Alzheimers Res Ther. 2018;10(1):117. http://dx.doi.org/10.1186/s13195-018-0447-y
131. Taupin V, Vaucher N, Loux C, Le Parc J, Michoux F, Colonna C, et al. Anti-amyloid antibodies as a potential therapeutic for Alzheimer's disease: Neuroprotective activity of SAR228810 against amyloid-induced neurotoxicity in vitro. Alzheimers Dementia. 2017;13:P944. http://dx.doi.org/10.1016/j.jalz.2017.06.1851
132. Pradier L, Cohen C, Veronique B, Debeir T, Barneoud P, Canton T, et al. SAR228810: An antiprotofibrillar beta-amyloid antibody designed to reduce risk of amyloid-related imaging abnormalities (ARIA). Alzheimers Dementia. 2013;9:P808. http://dx.doi.org/10.1016/j.jalz.2013.05.1678
133. Whitson JS, Selkoe DJ, Cotman CW. Amyloid beta protein enhances the survival of hippocampal neurons in vitro. Science. 1989;243(4897):1488–90. http://dx.doi.org/10.1126/science.2928783
134. Puzzo D, Privitera L, Leznik E, Fa M, Staniszewski A, Palmeri A, et al. Picomolar amyloid-beta positively modulates synaptic plasticity and memory in hippocampus. J Neurosci. 2008;28(53):14537–45. http://dx.doi.org/10.1523/JNEUROSCI.2692-08.2008
135. Puzzo D, Privitera L, Fa M, Staniszewski A, Hashimoto G, Aziz F, et al. Endogenous amyloid-beta is necessary for hippocampal synaptic plasticity and memory. Ann Neurol. 2011;69(5):819–30. http://dx.doi.org/10.1002/ana.22313
136. Lucey BP, Hicks TJ, McLeland JS, Toedebusch CD, Boyd J, Elbert DL, et al. Effect of sleep on overnight cerebrospinal fluid amyloid β kinetics. Ann Neurol. 2018;83(1):197–204. http://dx.doi.org/10.1002/ana.25117
137. Szczygielski J, Mautes A, Steudel WI, Falkai P, Bayer TA, Wirths O. Traumatic brain injury: Cause or risk of Alzheimer's disease? A review of experimental studies. J Neural Transm. 2005 Nov;112(11):1547–64. http://dx.doi.org/10.1007/s00702-005-0326-0
138. Garcia-Alloza M, Gregory J, Kuchibhotla KV, Fine S, Wei Y, Ayata C, et al. Cerebrovascular lesions induce transient β-amyloid deposition. Brain. 2011;134(12):3697–707. http://dx.doi.org/10.1093/brain/awr300