Chapter 8
Neuroprotection in Alzheimer's Disease

Introduction

Alzheimer's disease (AD) is a progressive degenerative disorder of the brain that begins with memory impairment and eventually progresses to dementia, physical impairment, and death. Patients develop various psychiatric and neurological signs during the course of the disease. The prevalence rates of dementia vary significantly in different countries, but range from 2.1 to 10.5%. AD is the most common type of dementia, accounting for 50–60% of all cases, and is described in detail in a special report on AD (Jain 2010).

Several other types of dementias are considered in the differential diagnosis of AD and sometimes all the dementias are lumped together if the type is not known. Other well-known types of dementias are vascular dementia, dementia of aging, and dementia associated with HIV infection. The focus of this section is on AD and some other dementias will be described in Chap. 11.

Pathomechanism of Alzheimer's Disease

Several factors that play a role in the etiology and pathogenesis of AD include the following:

- Aging
- Genetic risk factors
- Amyloid precursor protein (APP) and beta-amyloid (Aβ) accumulation with neural and vascular sequelae
- Tau hyperphosphorylation
- Membrane disturbances, phospholipid metabolism, and disruption of signal transduction
- Inflammatory reactions and immunological disturbances
- Environmental toxins: trace metals
- Neurotransmitter defects and imbalances

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- Neuroendocrine disturbances
- Oxidative injury and free radicals
- Disturbances in regulation and receptors of neurotrophic factors (NTFs)

Such a large number of factors in the etiology of AD are responsible for the plethora of theories of cause of AD. AD is certainly not the result of a single operative mechanism, but more likely comprises one or more processes that lead to intrinsic neuronal destruction. A complex disease like AD is difficult to attack because no single approach is adequate and the development of a single universal therapy is unlikely.

The most characteristic finding in the brains of patients with AD is a profusion of deposits of Aβ, a protein fragment of unknown function. Aβ is found in small quantities in normal brains. Amyloid deposits by themselves do not damage the brain, but in the presence of apoE, amyloid forms hair-shaped fibrils and neuritic plaques. The fact that apoE4 can increase both the amount of Aβ and the formation of amyloid fibrils seems to indicate that this version of the lipoprotein is a genetic risk factor for AD.

Although extensive data support a central pathogenic role for Aβ in AD, the amyloid hypothesis remains controversial, in part because a specific neurotoxic species of Aβ and the nature of its effects on synaptic function have not been defined in vivo. Natural oligomers of human Aβ are formed soon after the generation of the peptide within specific intracellular vesicles and are subsequently secreted from the cell. Clumps of just a few molecules of misfolded Aβ protein hinder memory processes in rat brains. Cerebral microinjection of cell medium containing these oligomers and abundant Aβ monomers but no amyloid fibrils markedly inhibited hippocampal long-term potentiation in rats in vivo. Finally, treatment of cells with γ-secretase inhibitors prevented oligomer formation at doses that allowed appreciable monomer production, and such medium no longer disrupted LTP, indicating that synaptotoxic Aβ oligomers can be targeted therapeutically.

Role of Glutamate Transport Dysfunction in AD

Glutamate transport dysfunction may increase susceptibility to glutamate toxicity, thereby contributing to neuronal cell injury and death observed in AD. The glial glutamate transporter, excitatory amino acid transporter 1 (EAAT1), is strongly expressed in a subset of cortical pyramidal neurons in dementia cases showing AD-type pathology. In addition, tau (a marker of neurofibrillary pathology) is colocalized to the same pyramidal cells that expressed EAAT1. These findings suggest that EAAT1 changes are related to tau expression (and hence neurofibrillary tangle formation) in dementia cases showing AD-type pathology. This study implicates aberrant glutamate transporter expression as a mechanism involved in neurodegeneration in AD.

Abnormal processing of APP may be associated with deficient functioning of the glutamate transporter system. A fragment of Aβ has been shown to inhibit tritium-labeled glutamate uptake in cultured astrocytes. It is conceivable that among other effects, Aβ produces glutamate transporter oxidation and dysfunction.

Role of Neurotrophic Factors in the Pathomechanism of AD

Lack of NTF production may constitute the common underlying mechanism of various degenerative disorders of the nervous system. However, no convincing experimental data have been produced as yet to prove the reduced bioavailability of NTFs in AD. Neuropil threads, spread throughout the AD cortex, are considered to be an evidence for neuronal sprouting. Dystrophic neurites are a common element of senile plaques, which are also considered to stimulate sprouting. Hippocampal cholinergic sprouting occurs in AD as a response to gradual denervation. NGF, PDGF, BDNF, and other NTFs have been implicated in the pathogenesis of AD.

The use of NTFs for the treatment of degenerative disorders of the nervous system is based on experimental evidence that these molecules have the capability of protecting and also restoring impaired function of the neurons caused by a variety of agents. This effect can be considered nonspecific because the relation between the mechanisms of these diseases and the effect of NTFs is not quite clear. There is no evidence that AD is caused by deficiency of NTFs. Reported deficiency of NTFs may be a consequence rather than a cause of these diseases. For the causal relationship to be established, deficiency of NTFs should be demonstrated at the onset of the disease or prior to functional manifestations of the disease.

Management of Alzheimer's Disease

Treatment of AD requires attention to the following aspects of the disease:

- Treatment of primary cognitive symptoms
- Management of secondary problems
- Slowing of the progression of the disease. This requires neuroprotective strategies

Several drugs have been used or investigated in the treatment of AD; many of these have been abandoned and others are still in development. Cholinergic approaches are shown in Table 8.1. The mainstays of management of AD currently are cholinesterase (ChE) inhibitors: rivastigmine, donepezil, and galantamine. Although not classified as neuroprotective, there is evidence that long-term use of these drugs slows the progression of disease.

Neuroprotection in Alzheimer's Disease

Multiple mechanisms are involved in the pathogenesis of AD. Current therapies, based on cholinergic augmentation, target one of the several disturbances in AD. Free radical scavengers aim at eliminating only one type of culprit. One of the problems in designing rational therapies is disagreement on the cellular events that cause brain-cell death in AD and lead to dementia. One view is that amyloid plaques, composed primarily of Aβ, accumulate outside brain neurons, growing

Table 8.1 Cholinergic approaches to the treatment of Alzheimer's disease

Presynaptic: increasing ACh production and release
Choline
Lecithin
L-Acetylcarnitine
Synaptic: increasing ACh by decreasing its breakdown (AChE inhibitors)
Physostigmine (Synapton)
Tacrine (Cognex)
Rivastigmine (Novartis' Exelon)
Donepezil (Eisai/Pfizer's Aricept)
Galantamine (Janssen's Reminyl)
Zanapezil (Takeda) in phase III clinical trials

larger and larger until they rupture the cells and kill them. Another view is that neurofibrillary tangles (NFTs) kill the cell. Therapies relevant to neuroprotection are shown in Table 8.2. Of these, those aimed at Aβ protein appear to be more important than others. Some of these agents have been described in other chapters and some will be described in the following text.

Inhibition of Aβ Aggregation

Proteolytic processing of APP generates Aβ, which is thought to be causal for the pathology and subsequent cognitive decline in AD. The reduction in levels of the potentially toxic Aβ peptide has emerged as one of the most important therapeutic goals in AD. Key targets for this goal are factors that affect the expression and processing of the β-amyloid precursor protein (βAPP).

Secretase Inhibitors

Aβ proteins are composed of 40–42 amino acid peptides that are proteolytically cleaved from βAPP. The deposition of a species of Aβ ending at the 42nd residue as diffuse plaques is an early and essential feature of the disease. This knowledge has led to the design of inhibitors of key enzymes (α-, β-, and γ-secretases) involved in the production of Aβ peptide as therapies for AD. A number of immunological approaches (also called vaccines) are in development based on the current concepts of cell biology of AD. In transgenic mouse models of AD, chronic mucosal administration of Aβ peptide might induce an anti-inflammatory process in brain tissue that could beneficially affect the neuropathological findings of progressive cerebral deposition of Aβ. Future therapeutic approaches to reduce amyloid deposition also include inhibitors for β-secretase. An aspartate protease named BACE is considered

Table 8.2 Neuroprotective agents for Alzheimer's disease

Anti-apoptotic agents
Dimebon
Anti-inflammatory drugs
Indomethacin
Prednisone
COX-2 inhibitors
PPARgamma (peroxisome proliferator-activated receptor gamma) agonists
Calcium channel blockers
Nimodipine
Free radical scavenger/antioxidants
Vitamin E
Melatonin
Idebenone (synthetic analog of coenzyme Q)
Gene therapy
Delivery of NGF by genetically engineered cells
Glutamate antagonists
Memantine
Inhibition of amyloid plaque formation
γ-Secretase inhibitors
Inhibition of amyloid-β protein aggregation by immunization: amyloid vaccine
Prevention of the transformation of spherons into amyloid plaques
Prevention of fibrillogenesis and plaque formation: 3APS (Bellus)
Clioquinol
Phenserine: inhibition of AChE and the accumulation of the toxic β-amyloid
Monoamine oxidase (type B) inhibitors
L-Deprenyl (selegiline)
Calcium channel blocker
Nimodepine
Neurotrophic factors/enhancers
Neotrofin (AIT-082): enhances nerve growth factor-mediated neurite outgrowth
Phosphodiesterase inhibitors
Denbufylline
Steroids
Dehydroepiandrosterone
Estrogen
Statins
Lovastatin
Pravastatin
Simvastatin
Nootropics
Piracetam and its congeners; oxiracetam, entiracetam, prameracetam, and aniracetam
Vincamine (Cetal), vinpocetine, and vinconate
Ergoloid mesylates; Hydergine, Orphol, nicerogoline (Sermion), and Ergoplus
G. biloba extract (Tebonin, Valverde Vital)
Cerebrolysin

to be β-secretase and is a drug target for the development of therapy aiming to lower the amyloid burden in AD. Such drugs, in combination with vaccine therapies, may lead to a cure of this disease.

AN-1792

AN-1792, a synthetic version of the protein fragment Aβ42, represents one of a wave of vaccines being developed for noninfectious conditions. In the case of AD, the treatment is based on the theory that injections of Aβ might activate the immune system to recognize and attack pathological accumulations; it is Aβ42 that causes most of the amyloid plaques, which are a hallmark of AD abnormality. Ideally, anti-Aβ antibodies would form and bind to the amyloid plaques. Simultaneously, microglial cells would be activated and begin engulfing the amyloid plaques. Immunization with AN-1792 significantly reduced preexisting amyloid plaques and inhibited further plaque formation in the brains of transgenic mouse models of AD. In another study, prophylactic immunization with AN-1792 prevented the majority of treated mice from developing virtually any amyloid plaque. Initial phase I clinical studies with AN-1792 indicated that it halted the progress of AD, and was found to be safe and well tolerated. Further development was discontinued in phase II in 2002 because of the complication of encephalitis in patients treated with AN-1792. Further studies suggest that immunization with non-amyloidogenic Aβ derivatives represents a potentially safer therapeutic approach to reduce amyloid burden in AD, instead of using toxic Aβ fibrils.

Monoclonal Antibody m266

Chronic treatment with the MAb m266, which is specific for therapeutic approach to reduce amyloid burden in AD, instead of using toxic Aβ, increases plasma concentrations of Aβ and reduces Aβ burden in the PDAPP transgenic mouse model of AD. Thus passive immunization with this anti-Aβ MAb can very rapidly reverse memory impairment in certain learning and memory tasks in the PDAPP mouse model of AD, owing perhaps to enhanced peripheral clearance and/or sequestration of a soluble brain Aβ species. Memory deficits in mice improved without reduction of the Aβ plaque.

Clioquinol

Inhibition of neocortical Aβ accumulation may be essential in an effective therapeutic intervention for AD. Cu and Zn are enriched in Aβ deposits in AD, which are solubilized by Cu/Zn-selective chelators in vitro. Clioquinol is a chelator that crosses the blood–brain barrier (BBB) and has greater affinity for zinc and copper

ions than for calcium and magnesium ions. Clioquinol acts on Aβ by perturbing its metallochemistry, and clioquinol treatment has been shown to decrease Aβ accumulation in a mouse model of AD.

There is another mechanism for the efficacy of Cu chelation treatment with the drug clioquinol. In a yeast model system, adding clioquinol to the yeast culture medium drastically increased the intracellular Cu concentration, but there was no significant effect observed on Zn levels (Treiber et al. 2004). This suggests that clioquinol can act therapeutically by changing the distribution of Cu or facilitating Cu uptake rather than by decreasing Cu levels. The overexpression of the human APP or APLP2 extracellular domains, but not the extracellular domain of APLP1, decreased intracellular Cu levels. The expression of a mutant APP deficient for Cu binding increased intracellular Cu levels several fold. These data uncover a novel biological function for APP and APLP2 in Cu efflux and provide a new conceptual framework for the formerly diverging theories of Cu supplementation and chelation in the treatment of AD.

Treatment with clioquinol was tested in patients with AD. The levels of CSF-tau protein correlated positively and significantly with the serum levels of copper and also with the serum copper/zinc ratio. Clinical ratings showed slight improvement after 3 weeks' treatment. A phase II clinical trial was conducted using PBT1 (clioquinol) on patients with AD. The two major findings of the study are as follows (Ritchie et al. 2003):

1. The Aβ42, which was the target of the drug's activity, was significantly reduced in the blood of patients in the treatment group compared to an increase in the placebo group.
2. The progression of the disease was slowed down in the more severely affected patients in the treatment group compared to that in the placebo group. The initial findings of the study indicate that the rate of cognitive deterioration, as measured by the ADAS-Cog assessment scale, was slowed in these patients.

However, various impurities were found to occur in the synthetic process with unacceptably high levels of a di-iodo (toxic) form of PBT1, which was responsible for an increased risk of side effects and mutagenicity. PBT2, the backup compound, is a small-molecular-weight chemical entity and has a structure that does not contain iodine. Therefore, PBT2 is not capable of forming the di-iodo impurity. PBT2, a significant pharmacokinetic improvement on PBT1, targets the interaction between oxidized metal proteins and Aβ. It completed phase II clinical trials successfully in 2008.

FKBP52 for Neuroprotection from Cu Toxicity in AD

Copper is essential for some of the enzymes that have a role in brain metabolism. Sophisticated mechanisms balance copper import and export to ensure proper nutrient levels (homeostasis), while minimizing toxic effects. Several neurodegenerative

diseases including AD are characterized by modified copper homeostasis. This change seems to contribute either directly or indirectly to increased oxidative stress, an important factor in neuronal toxicity. When coupled to misfolded proteins, this modified copper homeostasis appears to be an important factor in the pathological progression of AD (Donnelly et al. 2007).

FK506-binding protein 52 (FKBP52, Johnson & Johnson) is an immunophilin that possesses peptidylprolyl *cis*/*trans*-isomerase (PPIase) activity and is a component of a subclass of steroid hormone receptor complexes. Overexpression of FKBP52 increased rapid copper efflux in ^{64}Cu-loaded cells, suggesting that FKBP52 is a component of the copper efflux machinery, and in so, may also promote neuroprotection from copper toxicity (Sanokawa-Akakura et al. 2004). It is a potential neuroprotective agent for AD.

Phenserine

Phenserine is a potent, brain-targeted, reversible, and highly selective inhibitor of the ACh. In preclinical studies, inhibition of this enzyme has been shown to improve memory and cognitive performance. It can also slow or halt cell death and disease progression. Phenserine works through two mechanisms: by inhibiting (1) the degradation of the neurotransmitter ACh in the brains of animals and (2) the production of a toxic form of the Aβ protein in the brain, which is thought to be a cause of death of brain cells in AD. Unlike other AChE inhibitors that simply suppress the activity of the enzyme, Phenserine's dual mechanism of action suggests that it has the potential to not only improve memory and cognition but also slow the progression of the disease.

The regulation of βAPP expression by phenserine is posttranscriptional as it suppresses βAPP expression without altering βAPP mRNA levels. However, phenserine's action is neither mediated through classical receptor signaling pathways, involving extracellular signal-regulated kinase or phosphatidylinositol 3-kinase activation, nor is it associated with the AChE activity of the drug. Furthermore, phenserine reduces the expression of a chloramphenicol acetyltransferase reporter fused to the 5′-mRNA leader sequence of βAPP without altering the expression of a control chloramphenicol acetyltransferase reporter. These findings suggest that phenserine reduces Aβ levels by regulating βAPP translation via the iron regulatory element in the 5′-untranslated region of βAPP mRNA, which has been previously shown to be upregulated in the presence of IL-1. This study identifies an approach for the regulation of βAPP expression that can result in a substantial reduction in the level of Aβ.

A preclinical study has examined the structure of the 5′ UTR of APP mRNA and revealed how the 5′ UTR is acted on by different cytokines, such as IL-1 and TNF, that are involved in the neuroinflammatory cascade of events leading to AD pathogenesis (Lahiri et al. 2003). The drug phenserine is significant in this context because it has been shown to reduce APP through the same 5′ UTR element.

The safety results from the 72-patient, phase II clinical trial of phenserine in mild to moderate AD patients substantiate that the drug is safe and well tolerated. Phase III trials were conducted but did not show efficacy. There is no active development currently.

Colostrinin

Colostrinin (ReGen Therapeutics) is a proline-rich polypeptide complex produced from ovine colostrum (ewes' first milk after lambing) and is being evaluated as a treatment for AD. Clostrinin represents a diverse group of peptides produced in the mammary gland of mammals for the development of the optimal physiological responses in offspring. Colostrinin may act as an antioxidant, may prevent the formation of or dissolve amyloid-β plaques, and may modulate the immune system. A double-blind, placebo-controlled study showed that oral administration of colostrinin was well tolerated and improves the outcome of patients with mild to moderate AD.

Inhibition of Neuroinflammation

Inflammation of the nervous system is implicated in several neurological disorders including AD. A number of anti-inflammatory agents have been investigated and a few examples are described here.

Etanercept

Etanercept is an FDA-approved TNF-α inhibitor that is now widely used for treatment of rheumatoid arthritis and other systemic diseases associated with inflammation. Complex influences of TNF-α on neural health suggest that manipulation of this cytokine might have important impacts on diseases characterized by glial activation, cytokine-mediated neuroinflammation, and synaptic dysfunction. In a 6-month study, 15 probable-AD patients were treated weekly with perispinal injection of etanercept and produced sustained cognitive improvement during the study period (Griffin 2008). In another prospective, single-center, open-label, 6-month pilot study, 12 patients with mild-to-severe AD disease were administered etanercept, 25–50 mg, weekly by perispinal administration (Tobinick and Gross 2008). Two-tailed, paired *t*-tests were conducted comparing baseline performance to 6-month performance on all neuropsychological measures. Test batteries included the California Verbal Learning Test-Second Edition, Adult Version; Logical Memory I and II from the Wechsler Memory Scale-Abbreviated;

the Comprehensive Trail Making Test; Boston Naming Test; and letter/category verbal fluency. All measures revealed a significant effect except for the Boston Naming Test and the TMT-4, with WMS-LM-II being marginally significant. Rapid improvement in verbal fluency and aphasia was seen in two patients with dementia, beginning minutes after perispinal etanercept administration.

Because of the inability of large molecules such as etanercept to cross the BBB following conventional systemic administration, it is likely that the more direct drug delivery system by perispinal injection also contributed to the effectiveness of the treatment. Perispinal administration of etanercept may enable rapid delivery to the CNS via the cerebrospinal venous system, resulting in improvement in synaptic mechanisms that have been dysregulated by excess TNF-α. TNF-α modulation in AD may also act by influencing glutamate, NMDA, amyloid, and other inflammatory pathways (Tobinick 2009). Larger-scale studies of this therapeutic intervention, including phase III trials, are warranted in dementias.

Neurotrophic Factors/Gene Therapy

NGF Gene Therapy

The first AD patient treated with intracerebroventricular NGF showed improvement in episodic memory but not of cognitive impairment and had no adverse effects (Olson et al. 1992). However, neuropathic pain was reported as a complication of this therapy. Intraventricular NGF infusion in the rat was shown to cause sympathetic sprouting in dorsal root ganglia, which can explain neuropathic pain. No further clinical trials of NGF are being carried out using intracerebroventricular administration in patients with AD.

There are differences as well as some parallels in cognitive impairment with aging and AD regarding the role of NGF. Extensive cell loss does not occur as a consequence of normal aging in human and nonhuman primate species. More recent studies have demonstrated significant reductions in functional neuronal markers in subcortical brain regions in primates as a consequence of aging, including dopaminergic and cholinergic systems, although corresponding losses in cortical innervation from these neurons have not been investigated. In one study, aging was associated with a significant 25% reduction in cortical innervation by cholinergic systems in rhesus monkeys. Furthermore, these age-related reductions are ameliorated by cellular delivery of human NGF to cholinergic somata in the basal forebrain, restoring levels of cholinergic innervation in the cortex to those of young monkeys. Thus, (1) aging is associated with a significant reduction in cortical cholinergic innervation; (2) this reduction is reversible by growth-factor delivery; and (3) growth factors can remodel axonal terminal fields at a distance, representing a nontropic action of growth factors in modulating adult neuronal structure and function (i.e., administration of growth factors to cholinergic somata significantly increases axon density in terminal fields). These findings are relevant to potential

clinical uses of growth factors to treat neurological disorders. In addition, extensive dose escalation/toxicity studies in primates demonstrated the safety of NGF gene delivery.

Based on these findings, a phase I trial of NGF gene therapy for AD was completed (Tuszynski et al. 2005). This trial, sponsored by the University of California (San Diego, CA) and Ceregene Inc., involved surgical implantation of cells from the patients' skin that were genetically modified to produce NGF into the brain of patients with AD. The primary goal of this study was to determine that the procedure is safe. The secondary goal was to determine whether NGF produced by the cells implanted into the brain can prevent the death of some nerve cells that are affected by AD and whether it can enhance the function of certain remaining brain cells. During the study, the cells, which originally are extracted from the patients' skin, were genetically modified to express NGF and then administered by stereotactic injection, a standard neurosurgical procedure, into the region of the brain called the nucleus basalis of Meynert – an area of the brain known to undergo serious degeneration in AD.

After a mean follow-up of 22 months in six subjects, no long-term adverse effects of NGF occurred. Evaluation of the Mini-Mental Status Examination and Alzheimer Disease Assessment Scale – Cognitive subcomponent suggested improvement in the rate of cognitive decline. Serial PET scans showed significant increases in cortical 18-FDG after treatment. Brain autopsy from one subject suggested robust growth responses to NGF. In the six evaluable, early-stage AD patients, an approximately 40–50% reduction was observed in their annual rate of decline on the measured cognitive function scales compared to that in their preoperative function. The subjects showed a reduced rate of decline that persisted throughout the period of the study. With currently available pharmacological therapy, the average rate of decline in cognitive function is up to 6% with a median duration of 3–6 months. The procedure was initially performed while the patients were awake but lightly sedated. Two patients moved as the cells were being injected, resulting in bleeding in the brain. Following these events, the protocol was redesigned with patients given general anesthesia during the procedure, and subsequent procedures were performed without complication. Based on these results, the NGF implants, when performed with patients fully anesthetized, appear to be safe and well tolerated by patients. Ceregene is now developing this product as CERE-110, which carries the NGF gene encased in an AAV viral coating to protect the gene and facilitates its delivery to brain cells. In 2004, a phase I trial was initiated using CERE-110 delivered by an AAV-2 vector to deliver NGF selectively into basal forebrain region of the brain in AD where neurons are degenerating, in order to prevent cell death and reverse cell atrophy. The delivery avoids other parts of the brain where it may cause side effects.

Another study has shown that the neurodegeneration induced by the expression of anti-NGF antibodies in AD11 mice (model of AD) can be largely reversed by NGF delivery through an olfactory route (Capsoni et al. 2002). Even if the NGF proves to be an effective therapy for AD, it may not provide definitive treatment. Pathogenetic mechanisms other than NGF deficiency are involved and the pathology is not restricted to NGF-responsive neurons.

AL-108

AL-108 (Allon Therapeutics Inc.), an 8-amino acid peptide discovered to be the smallest active element of activity-dependent neuroprotective protein (ADNP), a glial cell mediator of vasoactive intestinal peptide (VIP), induced neuroprotection. Its action is similar to that of the activity-dependent neurotrophic factor (ADNF), which is a novel glial-derived polypeptide known to protect neurons from a variety of toxic insults. AL-108 promotes the formation of synapses after CNS cells have been impacted by disease or injury. Outgrowth of neurites increases as the administered dose of AL-108 is increased.

Preclinical experiments show that AL-108 has potent neuroprotective, memory-enhancing, and neurotrophic properties. Toxicology studies indicate that AL-108 is safely tolerated at doses over three orders of magnitude above the effective concentration. Furthermore, the neuroprotective efficacy of AL-108 is validated in a comprehensive battery of in vitro and in vivo studies. AL-108 has been tested in numerous in vivo and in vitro models of AD, stroke, TBI, multiple sclerosis, and neuropathies and is safely tolerated, bioavailable, and suitable for drug development. There is considerable evidence that AL-108 will be an effective neuroprotectant in both chronic and acute indications. In 2005, Allon completed phase I human clinical trial evaluating intranasal AL-108 as a treatment for AD. It is currently in phase II clinical trials in diagnosed AD patients.

Targeting Plasminogen Activator Inhibitor Type-1 Gene

The plasmin-generating cascade appears to serve a protective role in the CNS, since plasmin-mediated proteolysis of APP utilizes the alpha site, eventually generating nontoxic peptides, and plasmin also degrades Aβ. The conversion of plasminogen to plasmin by tissue plasminogen activator in the brain is negatively regulated by plasminogen activator inhibitor type-1 (PAI-1), resulting in the attenuation of plasmin-dependent substrate degradation with resultant accumulation of Aβ. PAI-1 and its major physiological inducer TGF-β1, moreover, are increased in models of AD and have been implicated in the etiology and progression of human neurodegenerative disorders (Higgins 2006). Targeting of PAI-1 gene expression has the potential as a molecular approach to the therapy of AD.

Estrogen and AD

Estrogen enhances cholinergic activity and significantly increases the expression of the anti-apoptotic protein Bcl-xL. Estrogen-induced enhancement of Bcl-xL is associated with a reduction in measures of β-amyloid-induced apoptosis, including the inhibition of both caspase-mediated proteolysis and neurotoxicity. Brain areas

known to support memory were found to have high densities of estrogen receptors. Estrogen promotes the growth and survival of cholinergic neurons, and it also modulates serotonergic and catecholaminergic neurotransmission. It increases cerebral blood flow and has antioxidant, anti-inflammatory, and general neuroprotective activity. The role of estrogen action in the brain seems to extend beyond the confines of sexual differentiation and reproductive neuroendocrine function, and estrogen might act as a NTF, having important influences on the development, survival, plasticity, regeneration, and aging of the mammalian brain. Furthermore, the antioxidant and neuroprotective effects of estradiol might be independent from estrogenic properties. Another concern about estrogens is the risk of breast and uterine cancer.

Several longitudinal studies show an inverse relationship between estrogen replacement therapy and the development of AD. However, not all the studies support neuroprotective effect of estrogen. Results from controlled trials are conflicting. In addition, estrogen plus progestin therapy did not prevent mild cognitive impairment (MCI) in these women. These findings, coupled with previously reported risk of cancer, support the conclusion that the risks of estrogen plus progestin outweigh the benefits.

Antioxidants

NSAIDS

Studies done in the laboratory and animal models have suggested that anti-inflammatory drugs prevented AD or delayed its onset by reducing inflammation in the brain. A prospective study has shown that long-term use of anti-inflammatory drugs such as ibuprofen and naproxen can lower the risk of AD but not of vascular dementia (in t' Veld et al. 2001). HCT 1026 and NCX 2216 (NicOx SA), nitric oxide-donating derivatives of flurbiprofen with antioxidant properties, were in clinical studies for the prevention of AD, none of which were successful.

Memantine

Memantine is approved for the treatment of moderate to severe AD. It has been marketed in Europe for the treatment of dementia for several years and has a good safety record. It is a noncompetitive, low-affinity NMDA antagonist with high-voltage dependency and fast receptor kinetics. It has anti-excitotoxic action and neuroprotective properties. The rationale for use is excitotoxicity as a pathomechanism of neurodegenerative disorders. Mechanisms of action of memantine are as follows:

- Memantine and Mg^{2+} occupy the same NMDA receptor channel and are mutually exclusive.
- Unlike Mg^{2+}, memantine does not leave the channel so easily.

- Blocking of NMDA receptor channels is partial and 15–20% of the channels unblock in the absence of an agonist and are available for subsequent physiological activation.
- It increases brain-derived NTF mRNA levels in the limbic cortex.

Memantine acts as a neuroprotective agent against this pathomechanism, which is also implicated in vascular dementia and dementia associated with HIV infection. Memantine at a clinically relevant dose also markedly increased BDNF mRNA levels in the limbic cortex, and this effect is more widespread and pronounced at higher doses. Thus the neuroprotective properties of memantine could be mediated by the increased endogenous production of BDNF in the brain. Memantine has been investigated extensively in animal studies, and its efficacy and safety have been established and confirmed by clinical experience in humans. Advantages of memantine are as follows:

- Efficacy and safety established by extensive investigations in animals.
- It shows none of the undesirable effects associated with competitive NMDA antagonists such as dizocilpine.
- Has been in clinical use for dementia in Germany for more than 15 years.
- It can be combined with acetylcholinesterase inhibitors, which are the mainstay of current symptomatic treatment of AD.

The efficacy of memantine in a variety of dementias has been shown in clinical trials. Memantine is considered to be a promising neuroprotective drug for the treatment of dementias, particularly AD, for which there is no other neuroprotective therapy available (Jain 2000). A double-blind, placebo-controlled, phase II clinical trial of memantine was conducted for AIDS-related dementia. Two other placebo-controlled phase III trials in mild to moderate vascular dementia have been conducted in Europe (France and the UK).

In the future management of AD, there would be an increasing emphasis on neuroprotective and regenerative therapies. Currently, memantine is the only clinically available neuroprotective drug that can be combined with other current therapies such as cholinergic agents and will continue to be in clinical use for several years.

Dimebon

Dimebon™ (Medivation Inc.) is an oral drug with anti-apoptotic and neuroprotective action. It has a strong safety record for over more than 20 years of human use; it has been on the market in Russia since 1983, where it is approved for the treatment of allergic rhinitis and allergic dermatitis. Evidence of neuroprotective action is as follows (Bachurin et al. 2001):

- Dimebon demonstrated cognition- and memory-enhancing properties in the active avoidance test in rats treated with the neurotoxin AF64A, which selectively destroys cholinergic neurons.

- Dimebon protected neurons in the cerebellum cell culture against the neurotoxic action of Aβ25–35.
- Dimebon inhibited signs of aging in normally aging rodents after 1 year of dosing.
- Dimebon causes significant improvements in performance in memory- and cognition-impaired rodents in the Rat Water Maze Test and Active Avoidance Tests, tests of memory and cognition.

Dimebon was compared with memantine, an approved drug for AD with neuroprotective action (Grigorev et al. 2003). Dimebon in low concentrations potentiated the activity of AMPA receptors in rat cerebellar Purkinje neurons, while memantine produced only an insignificant potentiation in a small group of these cells. In cortical neurons of rat brain, memantine efficiently blocked NMDA-induced currents in dimebon-insensitive neurons. By contrast, its effect was far weaker in neurons where the blocking action of dimebon on NMDA receptors was most pronounced. It was hypothesized that the differences in the effects of memantine and dimebon are determined by their interaction with different sites of NMDA receptors.

Dimebon appeared to improve some aspects of memory, cognitive, and global function in a pilot clinical study of 14 AD patients conducted at the Moscow Center of Gerontology in Russia. The patients were treated with oral dimebon three times daily for 2 months. Patients' memory, cognitive, and global function were assessed using two distinct psychiatric scales. Baseline scores for individual patients were determined prior to drug treatment. After 2 months, the treatment was stopped and patient psychiatric assessments for memory, cognitive, and global function continued for an additional 2 months. Patients in this open-label study experienced an improvement in memory and cognition after 2 months of therapy. Furthermore, after dimebon was discontinued at week 8, investigators observed deterioration in patients' cognitive function.

Dimebon is in development for both AD and HD. In 2005, Medivation began a randomized, double-blind, placebo-controlled, phase II study of dimebon in patients with mild to moderate AD in Russia, and completed enrollment in 2006 with 183 patients. Medivation designed the trial with the same study end points as those used in pivotal phase III registration studies for drugs previously approved by the FDA to treat AD. The results of this trial are not available as yet.

Cerebrolysin

Cerebrolysin (FPF 1070, Ebewe Pharmaceuticals) is a porcine-derived peptide preparation that has been claimed to regulate neuronal energy metabolism, influence behavior by neuromodulation, and protect nerve cells through neurotrophic stimulation. It has been shown to improve cognitive function, noncognitive psychiatric symptoms, and behavior in patients with AD in several uncontrolled studies in Russia and China. The Cerebrolysin Study Group has compared cerebrolysin with placebo, both given as intravenous infusions 5 days a week for 4 weeks, in the

treatment of 157 patients with mild to moderate AD. Overall, 76% of the cerebrolysin-treated patients showed some improvement at the 4-week assessment, compared with only 60% of the control group. There was a trend toward improvement of activities of daily living among the cerebrolysin-treated patients, though the results indicated no statistically significant difference between the treatment groups. Cerebrolysin is currently approved for marketing in 28 countries. Clinical research with cerebrolysin is ongoing with the aim of obtaining marketing approval in the USA and other countries.

Ginkgo biloba

G. biloba is a tree with a history of use in traditional Chinese medicine. Although the seeds are most commonly employed in traditional Chinese medicine, in recent years standardized extracts of the leaves have been widely sold as a phytomedicine in Europe and as a dietary supplement in the USA. The primary active constituents of the leaves include flavonoid glycosides and unique diterpenes known as ginkgolides; the latter are potent inhibitors of platelet activating factor. The mechanism of action of *G. biloba* extract (GBE) is not well understood, although it is considered to have antioxidant properties. In aging rats, GBE treatment lowers circulating free cholesterol levels and inhibits the production of brain APP and Aβ (Yao et al. 2004). Clinical studies have shown that GBE exhibits therapeutic activity in a variety of disorders including AD, failing memory, age-related dementias, poor cerebral and ocular blood flow, and congestive symptoms of premenstrual syndrome, and in the prevention of altitude sickness. Thousands of patients have been treated in various clinical trials. Although not approved by the regulatory authorities, commercial products of GBE are available as over-the-counter products for age-related cognitive decline, AD, and various eye disorders related to vascular insufficiency.

Some clinical trials, based on simple comparisons of drug–placebo differences, concluded that EGb 761 is less effective in the treatment of AD than ChE inhibitors. The methods of these clinical trials have been questioned (Hoerr 2005). The data of studies with both types of drugs show that drug–placebo differences in cognitive outcomes are more influenced by the degrees of deterioration in the placebo groups than by changes in the actively treated groups. Since the deterioration in the placebo group is determined by characteristics of the patients and not by drug effects, direct comparisons of drug–placebo differences are inappropriate to compare efficacies of anti-dementia drugs. Comparisons should take into account the different unspecific factors as well. The currently available data do not support the superiority of ChE inhibitors over EGb 761.

Controversy about the role of GBE as a neuroprotectant against decline of mental function with aging continues. A randomized, placebo-controlled, double-blind, 42-month, pilot study was carried out to assess the feasibility, safety, and efficacy of GBE on delaying the progression to cognitive impairment in normal elderly people aged 85 years and older (Dodge et al. 2008). In unadjusted analyses,

GBE neither altered the risk of progression from normal to Clinical Dementia Rating (CDR) scale of 0.5, nor protected against a decline in memory function. Secondary analysis taking into account medication adherence showed a protective effect of GBE on the progression to CDR and memory decline. Ginkgo Evaluation of Memory study, a randomized, double-blind, placebo-controlled clinical trial of community-dwelling participants aged 72–96 years with a median follow-up of 6 years, showed that the use of *G. biloba* did not reduce cognitive decline in older adults with normal cognition or with MCI (Snitz et al. 2009).

Tetrahydrocannabinol for Neuroprotection in AD

Cannabinoids are neuroprotective agents against excitotoxicity in vitro and acute brain damage in vivo. This background has prompted the study of localization, expression, and function of cannabinoid receptors in AD and the possible protective role of cannabinoids after exposure to Aβ, both in vivo and in vitro. One study showed that senile plaques in AD patients express cannabinoid receptors CB1 and CB2, together with markers of microglial activation, and that CB1-positive neurons, present in high numbers in control cases, are greatly reduced in areas of microglial activation (Ramirez et al. 2005). Pharmacological experiments show that GPCR and CB1 receptor protein expression are markedly decreased in AD brains. Additionally, in AD brains, protein nitration is increased and, more specifically, CB1 and CB2 proteins show enhanced nitration. Intracerebroventricular administration of the synthetic cannabinoid WIN55,212-2 to rats prevents Aβ-induced microglial activation, cognitive impairment, and loss of neuronal markers. Cannabinoids block Aβ-induced activation of cultured microglial cells, as judged by mitochondrial activity, cell morphology, and TNF-α release. These effects are independent of the antioxidant action of cannabinoid compounds and are also exerted by a CB2-selective agonist. Moreover, cannabinoids abrogate microglia-mediated neurotoxicity after Aβ addition to rat cortical cocultures. These findings indicate that cannabinoid receptors are important in the pathology of AD and that cannabinoids prevent the neurodegenerative process occurring in the disease.

The active component of marijuana, Δ^9-tetrahydrocannabinol (THC), competitively inhibits the enzyme AChE as well as prevents AChE-induced Aβ aggregation (Eubanks et al. 2006). In a test against propidium, one of the most effective inhibitors reported to date, THC blocked AChE-induced aggregation completely, while propidium did not. Computational modeling of the THC–AChE interaction reveals that THC binds in the peripheral anionic site of AChE, the critical region involved in amyloidogenesis. Compared to currently approved drugs prescribed for the treatment of AD, THC is a considerably superior inhibitor of Aβ aggregation, and this study provides a previously unrecognized molecular mechanism through which cannabinoid molecules may directly impact the progression of AD. This could be the basis of new drug discovery for AD.

Ladostigil Tartrate

Ladostigil tartrate (TV3326, Teva Pharmaceutical), or (*N*-propargyl-(3*R*) aminoindan -5-yl)-ethyl methyl carbamate, is a ChE inhibitor derived from rasagiline. Rasagiline is a selective, irreversible monoamine oxidase B (MAO B) inhibitor that has been developed as an anti-PD drug. TV3326 inhibits MAO-A and -B, stimulates the release of the non-amyloidogenic α-secretase form of soluble APP (sAPPalpha), and directly induces the phosphorylation of p44 and p42 MAP kinase (Yogev-Falach et al. 2002). These data suggest a novel pharmacological mechanism whereby these AChE inhibitors regulate the secretory processes of APP via the activation of the MAP kinase pathway. In addition to this, the multimodal effects of ladostigil in terms of neuroprotective molecular mechanism also include regulation of the Bcl-2 family members, inhibition of cell death markers, and upregulation of NTFs (Weinreb et al. 2008). Therefore, ladostigil has potential for the treatment of AD. Phase I studies with ladostigil have been completed and a phase IIa study has been recently initiated in Europe.

Phosphodiesterase Inhibitors as Neuroprotectives

Sildenafil (Viagra), phosphodiesterase (PDE) 5 inhibitor, is a commonly prescribed drug for erectile dysfunction, but also has neuroprotective effect in stroke (see Chap. 3). PDE4 is an enzyme that breaks down cAMP levels in the brain. PDE4 inhibitors such as denbufylline have shown efficacy in raising cAMP levels across multiple biochemical and physiological assays and in reversing memory deficits in animal behavior models of cognitive impairment. Ibudilast, a PDE4 inhibitor, which was originally developed as a bronchodilator for asthma in Japan, has also been investigated in stroke as a vasodilator with neuroprotective effects. Ibudilast crosses the BBB and suppresses glial cell activation. It may also be useful in the treatment of multiple sclerosis (see Chap. 11). Ibudilast is also the active constituent in AV-411 (Avigen Inc.), which is being developed for the treatment of neuropathic pain.

PPAR-γ Agonists

NSAIDs are efficacious in reducing the incidence and risk of AD and significantly delaying disease progression. A recently appreciated target of NSAIDs is the ligand-activated nuclear receptor PPAR-γ. NSAIDs, drugs of the thiazolidinedione class, and the natural ligand prostaglandin J2 act as agonists for PPAR-γ and inhibit the Aβ-stimulated secretion of proinflammatory products by microglia and monocytes responsible for neurotoxicity and astrocyte activation. PPAR-γ agonists were shown to inhibit the Aβ-stimulated expression of the cytokine genes IL-6 and

TNF-α. Furthermore, PPAR-γ agonists inhibited the expression of COX-2. These data provide direct evidence that PPAR-γ plays a critical role in regulating the inflammatory responses of microglia and monocytes to Aβ. Efficacy of NSAIDs in the treatment of AD may be a consequence of their actions on PPAR-γ rather than on their usually described COX targets. The efficacy of these agents in inhibiting a broad range of inflammatory responses suggests that PPAR-γ agonists may provide a novel therapeutic approach to AD.

Role of Statins in Reducing the Risk of AD

Increased level of circulating cholesterol has long been linked to an increased risk of coronary artery disease, and is now linked to an increased risk of developing AD. Statins (HMG-CoA reductase inhibitors) lower elevated serum cholesterol levels. Epidemiological data strongly suggest a protective effect of statins in dementia, particularly AD. Statins were described as neuroprotective agents in Chap. 2. The statin treatment of dyslipidemia is associated with a reduced risk of development of AD, but the mechanism is not clear. The effect may be mediated by a reduction in cholesterol biosynthesis in the brain, by lowering levels of ApoE-containing lipoproteins, or by reduction in Aβ production. In the brain, cholesterol from damaged or dying neurons is converted to 24S-hydroxycholesterol by cholesterol 24-hydroxylase (CYP46). The oxysterol is subsequently transferred across the BBB, transported to the liver by low-density lipoproteins (LDLs), and excreted as bile acids. Most of plasma 24S-hydroxycholesterol is derived from brain cholesterol; consequently, plasma levels of the oxysterol reflect brain cholesterol catabolism. In one study, statins lowered levels of plasma 24S-hydroxycholesterol without affecting levels of ApoE (Vega et al. 2003). The LDL lowering was more pronounced than 24S-hydroxycholesterol reductions. The effect of statins on LDL partially explains the reduction of plasma oxysterol level.

Examination of data from the Decision Support System database of the US Veterans Affairs Medical System, which contains information on approximately 4.5 million people, shows that simvastatin reduces the incidence of AD and PD by 50% (Wolozin et al. 2007). Although other statins were also examined, the selective benefit of simvastatin might be due to the combination of high potency and the ability to enter the brain.

Four controlled clinical trials have attempted to assess a direct cognitive-enhancing effect of statins in normal elderly people but all these were negative. Cholesterol-lowering drugs other than statins are not significantly associated with a reduced risk of developing AD. A growing body of evidence suggests that statins exhibit additional benefits that are independent of their cholesterol-lowering actions. Statin treatment also has considerable effect in the prevention of ischemic stroke. In animal models of ischemic stroke, statins have been proven to reduce infarct size through the upregulation of eNOS.

Combined Therapeutic Approaches to AD

From a therapeutic point of view, the most efficient strategies of therapy for AD might involve simultaneous combined administration of drugs interfering with distinct cellular signaling pathways elicited by noxious extracellular concentrations of Aβ in the brain. It is possible that effective neuroprotection achieved by simultaneous administration of a noncompetitive NMDA receptor antagonist and a Ca^{2+} channel blocker might exert protection against Aβ toxicity.

Clinical Trials in AD

As of October 2010, 829 clinical trials of AD are listed on the US Government web site http://www.clinicaltrials.gov/ct2/results?term=Alzheimer%27s+disease. These deal with all aspects of AD, including diagnosis and management. Of these, 120 that are relevant to neuroprotection and in progress are shown in Table 8.3. Discontinued or failed 50 clinical trials are discussed in a special report on AD (Jain 2010).

Future Prospects of Neuroprotection in AD

Several of the clinical trials have shown disappointing results for any neuroprotective benefit in AD. Fortunately, new strategies are emerging and provide hope for the future. These strategies are described in a special report on AD (Jain 2010). Some of these strategies are listed in Table 8.4.

Mild Cognitive Impairment

MCI has evolved in recognition of the fact that measurable memory deficits, more severe than can be accounted for by simple aging, are identified without functional decline sufficient for a diagnosis of dementia. MCI is quite frequent. Approximately one-fourth of persons over 65 years of age in the USA have some degree of cognitive impairment, not including elderly persons with dementia. Furthermore, the prevalence of non-dementia cognitive impairment increased with age, from 19.2% for people 65–74 years of age to 38% for people 85 years of age and older. Most cases of non-dementia cognitive impairment in this study were due to medically unexplained memory loss. The next most common cause was medical illness-associated impairment, followed by stroke and alcohol abuse. Non-dementia cognitive impairment is a major risk factor for later development of dementia. More than 80% of patients with MCI develop AD within 10 years, at a rate of about 10–15% of patients per year.

Table 8.3 Clinical trials for neuroprotection in Alzheimer's disease

Drug/sponsor	Mode of action	Status
3APS (3-amino-1-propanesulfonic acid)/Bellus	A synthetic GABA, analog	Phase III
ABT-560/Abbott	nACh modulator	Phase I
ACI-24/AC Immune Inc.	Active vaccine stimulates immune system to produce β-sheet-specific antibodies to prevent plaque deposition	Phase I/II
AC-3933/Dainippon	Inverse agonistic activity at GABA benzodiazepine receptor complex with cholinergic action	Phase I/II
AD-1/Affiris GmbH	A vaccine against AD based on the Affitope technology	Phase I
AL-108/Allon Therapeutics	Intranasal activity-dependent neuroprotective protein	Phase II/AD
AL-208	I/V activity-dependent neuroprotective protein	Phase II MCI
Anti-Aβ MAb/Genentech Inc.	Passive immunotherapy	Phase I
Antioxidant/NIA	Vitamin E + C + alpha-lipoic acid, and coenzyme Q	Phase I
Apan/Praecis Pharmaceuticals	Mobilizes Aβ in the brain and may be facilitating its clearance	Phase Ib
AZD0328/AstraZeneca	A selective neuronal nicotinic receptor agonist	Phase II
AZD3480/AstraZeneca	A neuronal nicotinic receptor agonist	Phase II
AV965/Avera Pharmaceuticals	A 5-HT_{1A} receptor antagonist	Phase I
Bapineuzumab (ACC-001 and QS-21)/Wyeth & Elan	A humanized monoclonal antibody to Aβ	Phase IIa
Bapineuzumab/Janssen	Evaluation in apolipoprotein Eε4 carriers	Phase III
Begacestat/Wyeth	A γ-secretase inhibitor	Phase I
BGC20-1259/BTG International	Combines inhibition of AChE and L-type Ca^{+} blocking	Phase I
BMS-708163/Bristol-Myers Squibb	γ-Secretase inhibitor	Phase II
Bryostatin-1/Blanchette Rockefeller Neurosciences Institute	PKC activator: enhances secretion of the α-secretase product sAPPα in fibroblasts from AD patients	Phase II
CAD-106/Cytos Biotechnology	Aβ immunization	Phase II
Celecoxib/Pfizer	To determine if a NSAID is efficacious in delaying progression in people with age-related cognitive losses who are at risk for developing AD	Phase III
CERE-110 (AAV-NGF)/Ceregene Inc., NIA, Emory University	Gene therapy: transplant of genetically engineered cells secreting nerve growth factor	Phase II
Cerebrolysin/Ebewe	Nootropic	Phase III
Cevimeline (AF102B)/Snow Brand Milk Products Co.	M1 agonist	Phase III

(continued)

Table 8.3 (continued)

Drug/sponsor	Mode of action	Status
COGNIShunt/Eunoe	Drainage of CSF from brain by a shunt system	Phase III
Colostrinin/ReGen Therapeutics	May act as an antioxidant and may prevent the formation of or dissolve Aβ plaques	Phase II
CPI-1189/Centaur	An orally bioavailable synthetic benzamide, which is an antagonist of TNF-α-mediated neurodegeneration	Phase II
Cripar (alpha-dihydroergocryptine)	A potent selective dopamine agonist which is marketed in Switzerland for the treatment of Parkinson's disease	Phase III in Italy
CTS-21166/CoMentis Inc.	β-Secretase inhibitor	Phase II
Curcumin/Institute for the Study of Aging	Curcumin, found in the spice turmeric, has antioxidant and cholesterol-lowering properties	Phase II
CX-717/Cortex	AMPA receptor activator: 30–100 times more potent than CX516	Phase IIa
CX-1739/Cortex	AMPA receptor activator	Phase I
Cyclophosphamide/NIH	To block the inflammatory response in AD	Phase II complete
DEBIO-9902 (prodrug of huperzine-A, ZT-1, Mimopezil) SR implant/Debiopharm AG	Dual action: inhibitor of AChE and NMDA antagonist. BRAINz' (Better Recollection for Alzheimer's patients with ImplaNts of ZT-1) trial: DEBIO-9902 vs. donepezil	Phase III
Dimebon/Motivation Inc./Pfizer	Neuroprotective agent	Phase III (Russia)
Docosahexaenoic acid/NIA and Martek Biosciences Corporation	To determine if treatment with DHA, an omega-3 fatty acid that reduces brain levels of Aβ, slows decline in AD	Phase I/II
DOMINO-AD (donepezil and/or memantine)	To determine the best option: donepezil or memantine, or a combination of both	Phase II
Donepezil (Aricept)/Eisai & Pfizer	For cognitive symptoms associated with Down's syndrome	Phase III
E2012/Eisai Company Ltd	A γ-secretase modulator that suppresses the production of Aβ	Phase I/current status not known
EHT 0202/Exonhit	$GABA_A$ receptor modulator that also weakly inhibits phosphodiesterase 4	Phase II
Estrogen/Wyeth-Ayerst	Double-blind, placebo-controlled trial to determine whether estrogen (or estrogen and progesterone) can delay the onset of memory loss or AD in elderly women with a family history of the disease	Phase III

(continued)

Table 8.3 (continued)

Drug/sponsor	Mode of action	Status
Estrogen/Pfizer	Response of female patients with mild to moderate AD who are concurrently treated with anticholinesterase therapy (donepezil) will be tested with drugs that block central cholinergic receptors (scopolamine)	Phase II
Etanercept given perispinally/ UCLA, Los Angeles	TNF-α inhibitor, approved for treatment of rheumatoid arthritis, reduces neuroinflammation in AD	Open study
EVP-6124/EnVivo	Selective α-7 nicotinic agonist	Phase IIa
EVT 302/Evotec AG	Selective MAO-B inhibitor: disease-modifying therapy	Phase I
Exebryl-1®/ProteoTech Inc.	Small-molecule drug targeting Aβ	Phase I
Far infrared radiation/GAAD Medical Research Institute Inc.	Hypothesis: far infrared radiation (5–15 μm wavelength) can cure AD	Phase I
G. biloba extract EGb 761/Schwabe	Trial to determine if it slows cognitive decline in the elderly people and prevents AD	Phase III in USA and marketed in EU
G. biloba extract/Imperial College, London & London School of Hygiene and Tropical Medicine	A double-blind trial will be added on to conventional medicines for age-associated memory loss. Subjects will receive 60 mg of gingko extract or a placebo twice daily for 6 months to see if it slows disease progression	Phase I/II
GSK 742457/GlaxoSmithKline	A 5-HT_6 antagonist that has been shown to enhance neurotransmitters and improve learning and memory	Phase II in 2005
GSK 239512/GlaxoSmithKline	A HT_3 receptor antagonist	Phase I
GSK 933776	Monoclonal antibody	Phase I
GTS21 (DMXBA)/CoMentis Inc.	Selective 7 nAChR partial agonist	Phase II
HCT 1026/Nicox	NO-donating derivative of the NSAID flurbiprofen	Phase I
HF0220/Hunter-Fleming	A neurosteroid, which acts by activation of endogenous 7-hydroxysteroid-driven neuroprotection pathways	Phase I
Huperzine-A/Savient Pharmaceuticals/NIA	An AChE inhibitor that decreases neuronal cell death caused by toxic levels of glutamate	Phase II
HT-0712/Helicon	PDE4 antagonist for age-associated memory disorders	Phase I
Isoprinicline: AZD3480/TC-1734/ Targacept	Selective $\alpha_4\beta_2$ neuronal nicotinic receptor agonist	Phase IIb
IVIG/Weill Cornell Medical Center, New York/Baxter	Antibodies in intravenous immunoglobulin (IVIG) derived from human plasma can capture Aβ	Phase II

(continued)

Table 8.3 (continued)

Drug/sponsor	Mode of action	Status
Ketasyn™ (AC1202)/Accera Inc.	Nutritional: ketone body modulation. Improvement in short-term memory and cognition in AD	Phase IIa/fast track
Lecozotan (SRA-333)/Wyeth	A serotonin 1a receptor antagonist to treat cognitive deficits seen in patients with mild to moderate AD	Phase I/II
Lipitor (atorvastatin)/Pfizer	Assessment of the clinical benefit of a cholesterol-lowering drug vs. placebo in the treatment of AD	Phase II completed
Lithium and divalproex/NIH	Glycogen synthetase kinase 3 inhibition of tau proteins	Phase II
Marinol® (dronabinol)/Solvay Pharmaceuticals	Cannabinoids may be neuroprotective due to antioxidant and anti-inflammatory properties	Phase I
MCD-386CR/Mithridion Inc.	Selective activator of the M1-type muscarinic receptors	Phase I
Memryte/Voyager Pharmaceutical Corporation	ALADDIN (Anti-gonadotropin in AD Drug INvestigation) to determine if it will slow the progress of AD. Effective in women but not in men	Phase III
MITO-4509/Migenix Inc.	Neuroprotective at mitochondrial level disturbances in AD	Phase II
MK-0249/Merck & Co.	Improves cognition but structure not disclosed	Phase II
MK-0952/Merck & Co.	PDE4 inhibitor to improve cognitive impairment in mild-to-moderate AD	Phase II
MK-0572/Merck & Co.	γ-Secretase inhibitor	Phase I
MKC-231 (coluracetam)/Mitsubishi	Choline uptake enhancer	Phase II/III
Motiva™ (nefiracetam)/Neuren Pharmaceuticals	A novel cyclic GABA derivative for improving cognitive deficits in AD and Parkinson's disease	Phase IIb
Neramexane/Forest Labs	NMDA receptor antagonist, structurally similar to memantine, was combined with ACh inhibitors and tested against patients who received only AChE inhibitors	Phase III showed no difference between two groups
NIC 5-15/Humanetics Pharmaceuticals	Prevents the formation of Aβ plaques	Phase II
NsG0202/NsGene AS	An implantable encapsulated cell biodelivery device that secretes NGF and is aimed at treating the progressive dementia associated with AD	Phase Ib, implants completed in 6 study patients
NGX267/Raptor Pharmaceuticals	Muscarinic receptor agonist. Lowers Aβ42 without inhibiting protein processes for normal function	Phase I

(continued)

Table 8.3 (continued)

Drug/sponsor	Mode of action	Status
Nicotine/NIH	Reduces Aβ aggregation in animal models. To improve symptoms of memory loss in MCI	Phase I
NP-12/Noscira	Microtubule and tau modulator	Phase II
NRM8499/Bellus Health	Prodrug of tramiprosate, which failed earlier trials	Phase I
Nutritional supplement/Harrison Chemists	To determine if the nutritional supplement would improve the function of the AD brain by increasing its ability to use sugar effectively	Phase II
Omega-3 fatty acids (fish oil) + alpha lipoic acid/National Center for Complementary and Alternative Medicine, USA	A double-blind, placebo-controlled study in AD subjects with mild cognitive decline to slow the progression of AD by reducing oxidative stress	Phase I/II
Oxygon™/Intellect Neurosciences	Synthetic preparation of a naturally occurring, small molecule protects neurons against Aβ-induced toxicity	Phase I
PAZ-417/Wyeth	A plasminogen activator inhibitor that tries to enhance clearance of Aβ	Phase I
PBT2/Prana Biotechnology	A small molecule that binds zinc and copper, reducing Aβ protein so that it is cleared from the brain	Phase IIa
PRX-03140/EPIX/Glaxo SmithKline	Selective 5-HT_4 receptor partial agonist: dual cholinergic/ neuroprotective mechanism	Phase IIa
PTI-00703/ProteoTech	Aβ protein fibrillogenesis inhibitor in patients with mild-to-moderate AD	Phase I/II
PYM50028/Phytopharm plc	A synthetic neuroprotective and neuroregenerative agent is being developed as a prescription drug for neurodegenerative conditions including AD	Phase II
Raloxifene/Eli Lilly	SERM approved for osteoporosis: improves memory and independent living in postmenopausal women with AD	Phase III
RG1450 (gantenerumab)/Roche & Morphosys	MAb to Aβ	Phase I
RG7412/Roche & AC Immune	MAb to Aβ	Phase I
RG3487/Roche	Nicotinic α7 selective agonist	Phase II
Rasagiline mesylate (Agilect)/Teva	Selective, irreversible MAO type-B inhibitor, which has shown beneficial activity in AD models	Phase II
rember™/TauRx Therapeutics	Tau aggregation inhibitor	Phase II
RN1219/Pfizer	Humanized MAb to Aβ	Phase II
S18986 (CX516)/Servier	AMPA receptor modulator	Phase II

(continued)

Table 8.3 (continued)

Drug/sponsor	Mode of action	Status
Salvia officinalis/National Center for Complementary and Alternative Medicine	The herb sage has been shown to enhance memory and mental function in healthy, young adults. This study will determine the effect of sage extract in early AD	Phase I
SAM-531/Wyeth	5-HT_6 antagonist	Phase II
SAN-61/Sanomune	A naturally occurring enzyme that cleaves soluble and fibril Aβ by activating MMP-9	Phase II pending
SAR110894/Sanofi-Aventis	H_3 antagonist	Phase I
Scyllocyclohexanehexol (AZD-103/ELND005)/Transition Therapeutics and Elan	Inhibits aggregation of Aβ	Phase II
Sermion® (nicergoline)/Pfizer	An ergot derivative known as nootropic agent	Phase III
Simvastatin/Merck & Co.	CLASP: a study to investigate the safety and effectiveness of simvastatin (a cholesterol-lowering drug) to slow the progression of mild-to-moderate AD	Phase III
Solanezumab (LY2062430)/Lilly	MAb that binds to Aβ and removes it from the blood and CSF	Phase III
T-817MA/Toyama Chemical Co.	Neuroprotective, prevents neurodegeneration induced by Aβ and promotes neurite outgrowth	Phase II
TAK-065/Takeda Pharmaceutical Corporation	An oral regeneration enhancer	Phase I (no information on current status)
Tanakan® (EGb 76)/Ipsen	*G. biloba* extract	Phase III
Testosterone/UCLA	Significant improvement in the caregiver version of the quality-of-life scale but minimal effects on cognition	Phase I/II
Transdermal 17-β-estradiol/Berlex	To determine the effectiveness of hormone replacement therapy in improving memory and the ability to live independently in postmenopausal women with AD	Phase III
TTP488 (TransTech Pharma)/now PF-4494700/Pfizer	A small molecule that targets the receptor for advanced glycation (RAGE) end products	Phase IIa completed
TTP4000 (TransTech Pharma)/now PF-3084014/Pfizer	A large molecule that targets the receptor for advanced glycation end products	Phase I
TV 3326 (ladostigil tartrate)/Teva	AChE inhibitor regulates the secretory processes of APP via activation of the MAP kinase pathway	Phase II

(continued)

Table 8.3 (continued)

Drug/sponsor	Mode of action	Status
United Biomedical	Vaccine UB 311 targeting Aβ	Phase I
V950/Merck & Co.	Vaccine: prevents the buildup of Aβ	Phase I
Vagal nerve stimulation/Cyberonics Inc.	Procedure already approved for epilepsy and depression (only in Europe)	Phase II
Valproate (Abbott)/NIH and Alzheimer's Disease Cooperative Study, UCLA	VALID: Valproate in Dementia. Valproate is an old antiepileptic drug that may have a neuroprotective effect and retard the progression of AD	Phase III
	Valproate Neuroprotection Trial, started in 2005 for 2 years in AD patients. Monitoring by MRI	Phase I/II
VITAL (VITamins to slow AD)/ National Institute on Aging	To determine if reduction of homocysteine levels with folate and vitamins B6 and B12 supplementation will slow the rate of cognitive decline in persons with AD	Phase III
Vitamin E and selenium/National Institute on Aging	PREADVISE (Prevention of Alzheimer's Disease by Vitamin E and Selenium) in elderly subjects	Phase III in progress

Table 8.4 Strategies for discovery of neuroprotective therapies for AD

Drugs acting on signaling pathways
JNK pathway as a target
Mitogen-activated protein kinase pathway as target
CPI-1189
Drugs to reverse inhibition of the PKA/CREB pathway in AD
Novels targets/receptors for AD drug discovery
Cyclin-dependent kinase-5
Inactivation of aph-1 and pen-2 reduces APP cleavage
Phosphodiesterase inhibitors
Pin 1 as a target in AD
Protein kinase C activators
Receptor for advanced glycation end products
Src homology-containing protein-1 inhibitors
Nitric oxide mimetics for direct activation of signal transduction cascades
Targeting GABAergic system

Relation of MCI to AD

MCI generally represents early-stage AD. Individuals currently characterized as having MCI progress steadily to greater stages of dementia severity at rates dependent on the level of cognitive impairment at entry, and they almost always have the neuropathological features of AD. Prevalence of MCI and predictive validities are highly dependent on the diagnostic criteria applied. Because of the lack of any uniform criteria for MCI, estimated prevalence rates range from 3 to 36% according to the concept applied. In a study in Germany, conversion rates to dementia over 2.6 years ranged from 23 to 47% (Busse et al. 2003). In addition, receiver operating characteristic curves indicated that all but one concept for MCI could predict dementia. Recent evidence also suggests that the etiological heterogeneity among individuals with MCI could be greater than previously reported. For example, cerebrovascular disease seems to be underestimated as a potential cause of MCI.

In a postmortem study of the brains of those diagnosed with MCI during life, over half were found to have AD, a third had evidence of cerebral infarcts, and fewer than one-fourth showed no signs of either disease (Bennett et al. 2005). Analysis of results shows that the level of AD pathology in persons with MCI is intermediate between that of those without cognitive impairment and those with dementia. Furthermore, the relationship between cognition and AD pathology in persons with MCI does not differ significantly from the relationship between cognition and AD pathology in persons with dementia or those without cognitive impairment. These data suggest that MCI may be the earliest clinical manifestation of AD. A study from the Mayo Clinic showed that there are structural changes in the brains of patients who may develop AD (Jicha et al. 2006). However, neither demographic variables nor cognitive measures had predictive value in determining which patients diagnosed with MCI will develop the neuropathological features of AD. MCI is a clinical diagnosis that is similar to the diagnoses of dementia or AD. Although cognitive tests and functional measures are very useful, ultimately, the final determination relies on the clinician's judgment. A MRI volumetric study of MCI subgroups using 3D mesh reconstructions of the structure revealed that the hippocampi of the patients who developed AD were significantly atrophic relative to those of the healthy controls (Becker et al. 2006).

Following decline of memory, executive function is the next brain function to deteriorate in the progression from MCI, a pre-AD condition, to AD. A decline in executive function will cause people to become more impaired in their daily activities, as it is quite important for daily function. The disease progression is correlated with the areas of the brain affected: the medial temporal lobe is affected as memory declines and then the frontal regions of the brain are affected as executive function worsens. In future studies of MCI, measuring whether a patient has moved from memory impairment alone to weakened executive function could help determine whether a particular drug is successful in slowing or stopping the disease.

Neuroprotection in MCI

A number of strategies are being pursued to prevent the progression of MCI. Almost all the drugs used for AD have been considered for MCI. Potential therapeutic agents for MCI are as follows:

- AChE inhibitors
- Antioxidants
- Anti-inflammatory drugs
- Glutamate receptor modulators
- Nootropics
- Immunomodulators
- Secretase inhibitors

Clinical trials of a number of agents aimed at halting the advance of pathological brain lesions in AD are in progress. Some of these agents may prove effective in slowing the progression of MCI to AD. Numerous nootropic compounds have been studied for possible effects in AD, and most of these studies have been negative, but the possibility that these compounds could be beneficial in earlier stages of the disease process has prompted a trial of piracetam in MCI. Because MCI involves more substantial cognitive and memory decline than normal aging, it represents a significant opportunity to manage a risk factor for the development of AD (Levey et al. 2006). Further research into treatments is needed to delay the conversion from MCI to AD. New drugs such as secretase inhibitors, small molecules that disrupt amyloid aggregation, and immunotherapies are in development for AD and need to be tested in MCI as well.

An increased rate of brain atrophy is often observed in older subjects, in particular in those who suffer from cognitive decline. Homocysteine is a risk factor for brain atrophy, MCI, and dementia. A randomized, double-blind, controlled trial of high-dose folic acid and vitamins B6 and B12 has shown that accelerated rate of brain atrophy in elderly people with MCI can be slowed by treatment with homocysteine-lowering B vitamins (Smith et al. 2010). Since accelerated brain atrophy is a characteristic of subjects with MCI who convert to AD, trials are needed to see if the same treatment will delay the development of AD.

References

Bachurin S, Bukatina E, Lermontova N, et al. Antihistamine agent Dimebon as a novel neuroprotector and a cognition enhancer. Ann N Y Acad Sci 2001;939:425–35.

Becker JT, Davis SW, Hayashi KM, et al. Three-dimensional patterns of hippocampal atrophy in mild cognitive impairment. Arch Neurol 2006;63:97–101.

Bennett DA, Schneider JA, Bienias JL, et al. Mild cognitive impairment is related to Alzheimer disease pathology and cerebral infarctions. Neurology 2005;64:834–41.

Busse A, Bischkopf J, Riedel-Heller SG, et al. Mild cognitive impairment: prevalence and predictive validity according to current approaches. Acta Neurol Scand 2003;108:71–81.

Capsoni S, Giannotta S, Cattaneo A. Nerve growth factor and galantamine ameliorate early signs of neurodegeneration in anti-nerve growth factor mice. Proc Natl Acad Sci U S A 2002;99:12432–7.

Dodge HH, Zitzelberger T, Oken BS, et al. A randomized placebo-controlled trial of ginkgo biloba for the prevention of cognitive decline. Neurology 2008;70:1809–17.

Donnelly PS, Xiao Z, Wedd AG. Copper and Alzheimer's disease. Curr Opin Chem Biol 2007;11:128–33.

Eubanks LM, Rogers CJ, Beuscher AE, et al. A molecular link between the active component of marijuana and Alzheimer's disease pathology. Mol Pharm 2006;3:773–7.

Griffin WS. Perispinal etanercept: potential as an Alzheimer therapeutic. J Neuroinflammation 2008;5:3.

Grigorev VV, Dranyi OA, Bachurin SO. Comparative study of action mechanisms of dimebon and memantine on AMPA- and NMDA-subtypes glutamate receptors in rat cerebral neurons. Bull Exp Biol Med 2003;136:474–7.

Higgins PJ. The TGF-beta1/upstream stimulatory factor-regulated PAI-1 gene: potential involvement and a therapeutic target in Alzheimer's disease. J Biomed Biotechnol 2006;2006:15792.

Hoerr R. Treatment effects of EGb 761 and cholinesterase inhibitors – why available studies do not demonstrate superiority of the latter. Phytomedicine 2005;12:598–600.

in t' Veld BA, Ruitenberg A, Hofman A, et al. Nonsteroidal antiinflammatory drugs and the risk of Alzheimer's disease. N Engl J Med 2001;345:1515–21.

Jain KK. An evaluation of memantine in the management of dementia. Expert Opin Investig Drugs 2000;9:1397–406.

Jain KK. Alzheimer Disease: New Drugs, Markets, and Companies. Jain PharmaBiotech Publications, Basel, Switzerland, 2010.

Jicha GA, Parisi JE, Dickson DW, et al. Neuropathologic outcome of mild cognitive impairment following progression to clinical dementia. Arch Neurol 2006;63:674–81.

Lahiri DK, Chen D, Vivien D, et al. Role of cytokines in the gene expression of amyloid beta-protein precursor: identification of a 5′-UTR-binding nuclear factor and its implications in Alzheimer's disease. J Alzheimers Dis 2003;5:81–90.

Levey A, Lah J, Goldstein F, et al. Mild cognitive impairment: an opportunity to identify patients at high risk for progression to Alzheimer's disease. Clin Ther 2006;28:991–1001.

Olson L, Nordberg A, von Holst H, et al. Nerve growth factor affects ^{11}C-nicotine binding, blood flow, EEG, and verbal episodic memory in an Alzheimer's disease patient. J Neural Transm Park Dis Dement Sect 1992;4:79–95.

Ramirez BG, Blazquez C, Gomez del Pulgar T, et al. Prevention of Alzheimer's disease pathology by cannabinoids: neuroprotection mediated by blockade of microglial activation. J Neurosci 2005;25:1904–13.

Ritchie CW, Bush AI, Mackinnon A, et al. Metal-protein attenuation with iodochlorhydroxyquin (clioquinol) targeting a amyloid deposition and toxicity in Alzheimer disease: a pilot phase 2 clinical trial. Arch Neurol 2003;60:1685–91.

Sanokawa-Akakura R, Dai H, Akakura S, et al. A novel role for the immunophilin FKBP52 in copper transport. J Biol Chem 2004;279:27845–8.

Smith AD, Smith SM, de Jager CA, et al. Homocysteine-lowering by B vitamins slows the rate of accelerated brain atrophy in mild cognitive impairment: a randomized controlled trial. PLoS ONE 2010;5(9):e12244; DOI: 10.1371/journal.pone.0012244.

Snitz BE, O'Meara ES, Carlson MC, et al. *Ginkgo biloba* for preventing cognitive decline in older adults. JAMA 2009;302:2663–70.

Tobinick E. Perispinal etanercept for neuroinflammatory disorders. Drug Discov Today 2009;14:168–77.

Tobinick EL, Gross H. Rapid improvement in verbal fluency and aphasia following perispinal etanercept in Alzheimer's disease. BMC Neurol 2008;8:27; DOI: 10.1186/1471-2377-8-27.

Treiber C, Simons A, Strauss M, et al. Clioquinol mediates copper uptake and counteracts Cu efflux activities of the amyloid precursor protein of Alzheimer's disease. J Biol Chem 2004;279:51958–64.

Tuszynski MH, Thal L, Pay M, et al. A phase 1 clinical trial of nerve growth factor gene therapy for Alzheimer disease. Nat Med 2005;11:551–5.

Vega GL, Weiner MF, Lipton AM, et al. Reduction in levels of 24S-hydroxycholesterol by statin treatment in patients with Alzheimer disease. Arch Neurol 2003;60:510–5.

Weinreb O, Amit T, Bar-Am O, et al. The neuroprotective mechanism of action of the multimodal drug ladostigil. Front Biosci 2008;13:5131–7.

Wolozin B, Wang SW, Li NC, et al. Simvastatin is associated with a reduced incidence of dementia and Parkinson's disease. BMC Med 2007;5:20.

Yao ZX, Han Z, Drieu K, et al. *Ginkgo biloba* extract (Egb 761) inhibits beta-amyloid production by lowering free cholesterol levels. J Nutr Biochem 2004;15:749–56.

Yogev-Falach M, Amit T, Bar-Am O, et al. Involvement of MAP kinase in the regulation of amyloid precursor protein processing by novel cholinesterase inhibitors derived from rasagiline. FASEB J 2002;16:1674–6.