Chapter 3
Role of Cellular Prion Protein in the Amyloid-β Oligomer Pathophysiology of Alzheimer's Disease

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Abstract Alzheimer's disease (AD) is the most common form of dementia affecting millions worldwide. The primary histopathological features of AD are amyloid-beta (Aβ) plaques and neurofibrillary tangles. Aβ oligomers (Aβo) are believed to be essential mediators of the synaptotoxicity and cell death that are characteristic of this illness. For decades, the exact mechanism for how Aβ exerted its toxic effect remained unknown. Recently, it has been shown that the cellular Prion Protein (PrP^C) acts as a high-affinity binding partner for Aβo. Moreover, it has been demonstrated that PrP^C is necessary for memory loss, impaired long-term potentiation, and neuronal dysfunction in transgenic mouse models of AD. Antagonizing PrP^C in AD mouse models has also been shown to reverse memory deficits, so targeting PrP^C is a potential avenue for treatment. This chapter will review the evidence connecting PrP^C to Aβo pathophysiology.

Keywords Alzheimer • Amyloid beta peptide • Oligomer • Neurodegeneration • Signal transduction • Transgenic • Spatial memory • Long-term potentiation • Synaptic plasticity

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3.1 Introduction

Alzheimer's disease (AD) is a chronic and progressive neurodegenerative disease estimated to affect approximately 35 million individuals worldwide (Prince et al. 2009). AD is responsible for 50–70% of all cases of dementia. As the population continues to age, its prevalence is expected to quadruple by the year 2050 (Brookmeyer et al. 2007). The classical clinical manifestations of AD are an amnestic memory impairment, language deterioration, and visuospatial deficits, eventually leading to death (Cummings 2004). Patients with AD have a post-diagnosis median survival ranging from 3 to 8 years (Helzner et al. 2008). It is now the sixth most common cause of death in the USA (Thies and Bleiler 2011). Current treatment options for AD are limited to partial efficacy and to symptomatic control. There is no disease-modifying therapy for AD in clinical practice today. Due to these factors, AD places a tremendous burden on individuals and families, with societal costs of 100 billion dollars each year (Meek et al. 1998).

The disease was first described in 1907 as a condition with progressive memory loss, atrophic brain, visible plaques, and intraneuronal fibrils (Alzheimer et al. 1995). The specific histological pattern is, to this day, the definitive way to diagnose AD (1997). The National Institute of Aging has proposed a criterion based on biomarkers that may broaden diagnoses (McKhann et al. 2011). The classical histological lesions have since been determined to be composed of extracellular insoluble plaques of polymeric beta-amyloid (Aβ) peptide (Glenner and Wong 1984) and intraneuronal fibrillary tangles of the hyperphosphorylated microtubule-associated protein, tau (Kosik et al. 1986). Efforts to understand the pathophysiology of AD focus on these proteins and lesions.

3.2 Amyloid Hypothesis

Over the past decade, there has been a growing consensus that the key mediator of the memory loss associated with AD is the 38–43 amino acid peptide Aβ. The "amyloid hypothesis" states that Aβ is not just the main constituent of plaques but also causes neuronal toxicity (Fig. 3.1). There are numerous genetic and biochemical avenues of research that support this premise, and this topic is reviewed in detail elsewhere (Selkoe and Schenk 2003). Key findings in support of this theory initiated from the observation that the Aβ peptide is the main constituent of AD plaques. Aβ peptide is derived from the amyloid precursor protein (APP) by sequential protease action of a β-secretase and a γ-secretase (Mills and Reiner 1999; Goldgaber et al. 1987). The genetics of the rare cases of early onset autosomal dominant AD support the Aβ hypothesis. Genetic analysis of certain families has uncovered mutations in the APP gene itself (Citron et al. 1992). The familial AD mutations were found to cluster in or around the sites of cleavage activity and to promote a greater $A\beta_{42}$ to $A\beta_{40}$ ratio, where $A\beta_{42}$ is more prone to oligomerization and fibrillization than $A\beta_{40}$ (Hardy and Selkoe 2002). Rare AD inducing mutations within the APP gene did not

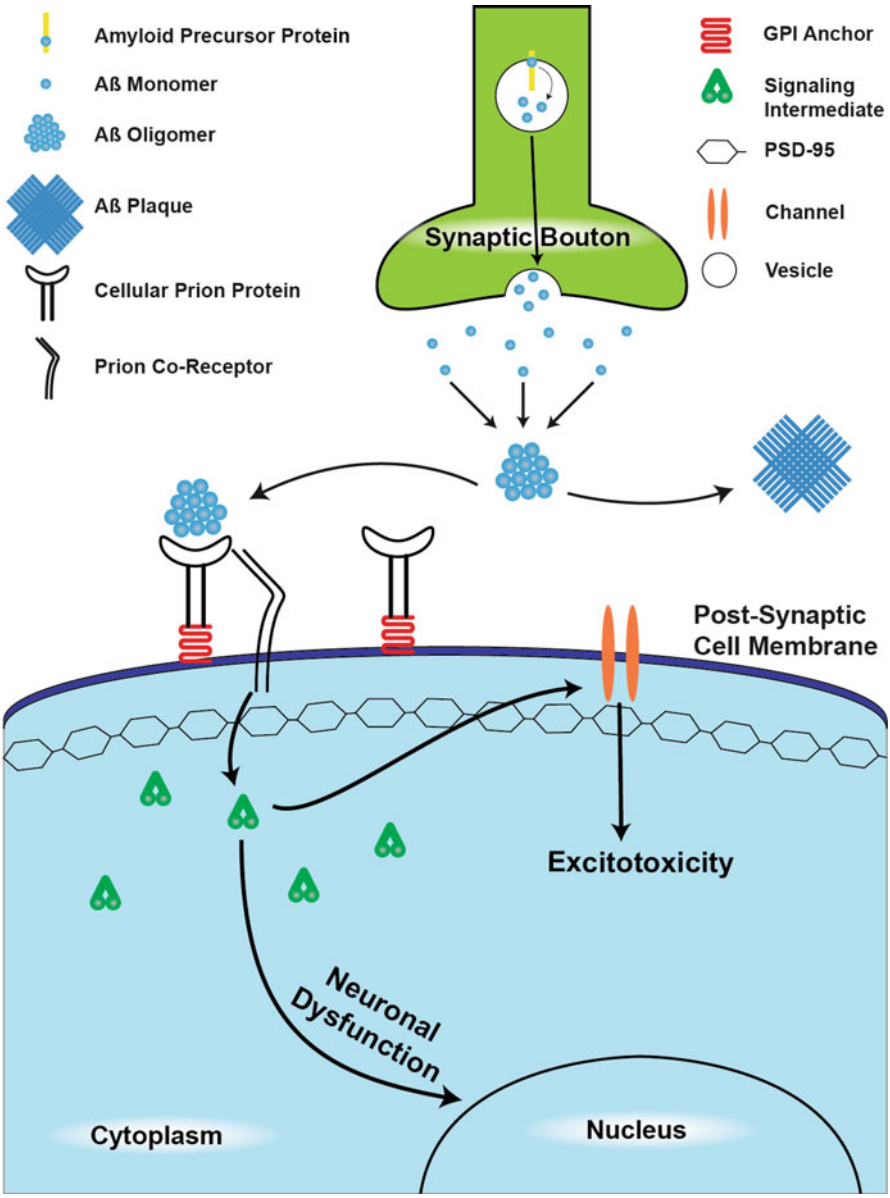

Fig. 3.1 Aβ Oligomers Bind to Neuronal PrP^C: amyloid precursor protein is cleaved by β- and γ-secretases within the presynaptic neuron to form 38–43 amino acid amyloid beta (Aβ) monomers. These monomers are then released into the synaptic cleft where they can oligomerize to form soluble Aβ oligomers. Alternatively, the monomers can continue to polymerize and form larger insoluble Aβ plaques. PrP^C has a high affinity for soluble Aβ oligomers while having limited affinity for both the monomers and the plaques. PrP^C on the postsynaptic neuron avidly binds Aβ oligomers and with the help of an unknown coreceptor initiates an intracellular cascade of events leading to neuronal dysfunction and excitotoxicity

affect Aβ processing directly, but increased rate of self-aggregation, leading indirectly to higher levels of Aβ plaques and fibrils (Wisniewski et al. 1991). Other cases of dominantly inherited early onset AD are caused by mutations in Presenilin-1 or 2, which are components of the γ-secretase. These AD mutations alter the enzymatic specificity of APP cleavage, leading to the same increase in Aβ42/Aβ40 ratio. Importantly, transfer of a human APPswe mutant transgene to mice was shown to recapitulate some aspects of human AD, including Aβ plaque and progressive memory deficits (Chapman et al. 1999).

It is also noteworthy that the APP gene is located on chromosome 21, and the Down's syndrome of trisomy 21 includes dementia and Aβ plaque deposition similar to non-syndromic AD (Masters et al. 1985). Moreover, a rare patient with Down's syndrome who did not develop memory loss was located and she had only a partial trisomy possessing the standard complement of two copies of APP (Prasher et al. 1998). Apart from Mendelian inheritance of early onset AD, genetic factors contribute to risk of late onset AD. Isoforms of the ApoE strongly affect risk, and these have been shown to alter Aβ clearance and aggregation (Kim et al. 2009). Variation at another risk genetic risk locus, clusterin, may cooperate ApoE to modulate Aβ levels (DeMattos et al. 2004).

More recently, consortium-based biomarker studies of aging and impaired cognition have demonstrated that Aβ alterations detected by either PET imaging or by CSF sampling are the first markers of AD, and that individuals with mild cognitive impairments (MCI) and Aβ marker abnormality have a very high likelihood of advancing to AD (Jack et al. 2010; Petersen et al. 2010; Sperling et al. 2011; Shaw et al. 2009; Heister et al. 2011). Thus, both genetic and disease progression studies support the amyloid hypothesis of AD.

3.3 The Importance of Oligomeric Aβ

One of the arguments against the amyloid hypothesis has been that the level of memory impairment and brain atrophy found in patients with AD correlates poorly with the number of plaques found in the brain (Terry et al. 1991; Josephs et al. 2008; Katzman 1986). Additionally, when neurons are exposed to fibrillar Aβ, the concentrations necessary to induce cell death were not consistent with physiologic levels. There has been increasing interest in Aβ oligomers (Aβo) as the solution to this conundrum (Fig. 3.1) (Walsh and Selkoe 2007). Oligomers are smaller soluble peptide polymers of Aβ monomers ranging in size from dimers up to 100-mers (Gunther and Strittmatter 2009). Consistent with a role in human neurodegeneration, nanomolar concentrations of Aβo derived from the cortices of patients with AD have been shown to inhibit long-term potentiation (LTP), reduce dendritic spine density, and impair recall of learned behavior (Wang et al. 2002; Walsh et al. 2002; Shankar et al. 2008). In the same assays, monomeric and polymeric Aβ had limited to no impact. Synthetically produced oligomers, also referred to as Aβ-derived diffusible ligands (ADDL), have been shown to induce memory dysfunction in the

AD mouse model independent of the presence of Aβ plaques (Lesne et al. 2006). Antibodies developed against the N-terminus of ADDL have been shown to block memory impairment.

While gathering evidence supports a crucial role for oligomeric Aβ, this begs the mechanistic question of how Aβo mediates its synaptotoxic and neurotoxic effects. Knowing that the effects of oligomeric Aβ are rapid, specific, and reversible all point to the existence of a high-affinity receptor. The existence of such a receptor would bring together many disparate facets within the field. Antagonizing this receptor would also represent a novel strategy for intervening in the progression of AD.

3.4 PrP^C Is the Binding Site for Oligomeric Aβ

It has been recently shown that cellular Prion Protein (PrP^C) acts as a high affinity binding site for Aβo (Laurén et al. 2009; Balducci et al. 2010). PrP^C has also been shown to transmit the synaptotoxic effect of Aβo (Laurén et al. 2009; Freir et al. 2011a; Barry et al. 2011; Chung et al. 2010). The original identification of PrP^C as an Aβo binding site utilized biotin-conjugated ADDLs (Laurén et al. 2009). Tagged ligand was then exposed to COS-7 cells that were expressing cDNA from an unbiased genome-wide adult mouse brain library in order to determine what gene products, if any, could bind ADDLs. COS-7 cells were chosen for this screening procedure because they bind less than 5% of the level of ADDL that endogenous receptors on hippocampal neurons bind. From within the 225,000 clones, there were only two positive hits, which both encoded a full-length version of PrP^C. The apparent dissociation constant for these clones was identical to that of hippocampal neurons, with nM affinity for ADDL. Depending on how the dissociation constant was calculated, it was found to be somewhere between 0.4 nM and 92 nM. PrP^C showed high selectivity for oligomerized Aβ versus monomeric Aβ, with a Kd difference of two orders of magnitude. Strong binding and specificity was also evident when PrP^C-Fc fragments are immobilized on resin and are exposed to ADDL.

A second library of 352 clones expressing transmembrane proteins was screened individually to identify hits with weaker interactions (Laurén et al. 2009). This produced a few hits; nonetheless the lowest dissociation constant found for any of these hits was 660 nM and there was minimal selectivity for oligomers over monomers. Previous papers had reported a possible interaction between monomeric Aβ and the receptor for advanced glycation products (Yan et al. 1996) or the α7 nicotinic acetylcholine receptor (Wang et al. 2000), but even with this lower stringency, direct Aβo binding did not indicate significant affinity.

E18 neurons have minimal affinity for Aβo immediately upon plating in vitro; however, the affinity for Aβo dramatically increases over a 15–20-day period that is contemporaneous with an equivalent increase in PrP^C expression levels in these cells (Laurén et al. 2009). There is broad colocalization of the immunoreactivity of bound Aβo and PrP^C. Neurons from *PRNP*$^{-/-}$ mice, which are PrP^C null, showed a

50% reduction in binding. Taken together, these data indicate that PrP^C contributes considerably to oligomeric Aβ binding, although there are likely other players or redundancy within the system.

3.5 Aβ Oligomers Bind to the Unstructured Central Domain of PrP^C

The specific domain of PrP^C that acts as the high affinity binding partner for Aβo was established by several methods. Mutant forms of PrP^C with different domains deleted were expressed on COS-7 cells to gauge the contributions of each domain to overall binding of ADDLs (Laurén et al. 2009). Removing the octapeptide repeat domain or the hydrophobic domain did not decrease the binding capacity, while cells expressing solely the globular domain were unable to bind oligomers. However, removing the unstructured central region, amino acids 95–110, dramatically lowered binding capability by 80%. In a DELFIA assay, human PrP^C fragments of amino acids 91–231 exhibited identical binding to ADDLs compared to that of full-length PrP^C, while fragments of amino acids 119–231 displayed almost no interaction (Freir et al. 2011a). This further emphasized the essential role that the amino acids 95–110 have for binding oligomers. Interestingly, the unstructured central domain has been implicated in contributing to neurodegeneration in mice (Baumann et al. 2007). Surface plasmon resonance studies demonstrated Aβo binding to both the 95–110 region and the extreme amino terminus 23–27, but not other regions of PrP^C (Chen et al. 2010).

The 6D11 antibody has as its epitope the amino acids 95–110 of the PrP^C protein. Preincubating the PrP^C expressing cells with 6D11 antibody effectively blocked the cells from interacting with oligomers (Laurén et al. 2009). The antibodies 8 G8 and ICSM-35 which both have epitopes that overlap with the epitope of 6D11 showed similar reduction in binding in a standard dose–response fashion (Laurén et al. 2009; Freir et al. 2011a). Anti-PrP^C antibodies that did not bind to this integral area did not impact binding, with one exception. Antibodies directed against the helix-1 domain appeared to lower affinity for ADDLs by up to 60%, which is surprising as this domain is quite far from the putative primary binding region (Freir et al. 2011a). It is possible that the antibodies at this region block a conformational shift within the PrP^C molecule that normally allows a stabilization of the binding of the oligomer, or it could potentially be a secondary binding site which could be consistent with the repetitive structure of Aβ.

Finally, although both Aβ and the octapeptide repeat domain of PrP^C are capable of binding copper ions with high affinity, the chelation of copper ions does not appear to contribute to their interaction. There was no change in binding affinity between COS-7 cells expressing PrP^C in copper-free F12 medium or in F12 medium with 1 mM of copper sulfate added (Laurén et al. 2009). The addition of up to 10 mM of EDTA, which would sequester any copper ions away from PrP^C and Aβ, had no impact on binding in hippocampal neuronal cultures (Freir et al. 2011a).

3.6 Aβ Oligomers Inhibit LTP Through PrP^C

LTP is a persistent increase in synaptic strength represented primarily by an increase in excitatory postsynaptic potentials (EPSP) that can last for hours in response to a high frequency train of electrical stimuli. It is believed to be a form of synaptic plasticity that likely forms the cellular and molecular basis for learning. Hippocampal LTP has been shown as necessary to form persistent spatial memories (Morris et al. 1986). In particular, Schaffer collateral LTP has been shown to be strongly inhibited by nanomolar concentrations of Aβo (Wang et al. 2002; Walsh et al. 2002). This makes LTP an excellent method to probe whether PrP^C participates in the pathogenicity of Aβo.

Hippocampal slices of brain from wild-type and *PRNP*$^{-/-}$ mice on a C57Bl6 background were stimulated to induce LTP in the presence of 2 nM Aβo in vitro (Laurén et al. 2009). Wild-type brain slices only had a 20% augmentation of the slope of the EPSP, a significant reduction in what would normally be expected. In contrast, the slope of the EPSP for the treated knockout brain slices had an 80% augmentation, which is identical to the EPSP of untreated brain slice. In order to rule out that congenital loss of PrP^C could induce some compensatory effects that could explain the unaffected LTP of the knockout brain slices in the face of Aβo challenge, wild-type brain slices were incubated with the 6D11 antibody followed by exposure to Aβo. These 6D11 antibody pretreated slices were completely protected from the expected loss of EPSP from Aβo.

The Malinow group also exposed hippocampal neurons of *PRNP*$^{+/+}$ or *PRNP*$^{-/-}$ genotypes to an Aβ42 preparation and monitored LTP (Kessels et al. 2010). In contrast to the findings described above, neither genotype had any augmentation of EPSP, even briefly, after LTP induction. These findings are also distinct from several previous studies of Aβo activity in wild-type neurons (Wang et al. 2002; Walsh et al. 2002), in which the peptide failed to abrogate short-term induction, but caused a diminution of long-term maintenance. The PrP-negative study (Kessels et al. 2010) also reported baseline inhibition by Aβ prior to induction. These two findings suggest that a general cytotoxic response was elicited by this incompletely characterized Aβ preparation. This led Collinge's group to demonstrate that a biochemically well-characterized Aβo preparation inhibited LTP in a PrP^C-dependent fashion (Freir et al. 2011b), replicating the original observation (Laurén et al. 2009).

PRNP$^{-/-}$ mice were crossed with APP-PS$^+$ mice, which express human mutant forms of APP and PSen-1, to further evaluate the in vivo effects of Aβ on LTP (Calella et al. 2010). The Aguzzi group showed a deficiency in the augmentation of LTP at 4 months of age, regardless of PrP^C expression. Of note, this mouse model of AD is known for rapidly producing Aβ amyloid at an early age. The rapid production of Aβ might overwhelm PrP^C binding and bind to secondary receptors leading to irreversible damage. Intriguingly, APP-PS$^+$ mice overexpressing an anchorless version of PrP^C were protected from LTP impairment. The secreted PrP^C likely bound to the soluble Aβ oligomers and protected the hippocampal neurons. This finding supports the hypothesis that PrP^C is the high affinity binding partner for relevant Aβo species.

Although synthetically produced Aβo is a potent synaptic toxin, it may not be identical to naturally occurring Aβo found within the brains of patients with AD. Importantly, water-soluble extracts derived from the brains of patients with AD have similar synaptotoxic effects to that of synthetic Aβo. Such AD extracts were preincubated with hippocampal slices from wild-type and *PRNP*$^{-/-}$ mice, followed by high frequency stimulation (HFS) (Freir et al. 2011a). The wild-type slices exposed to AD extracts had impaired LTP, while the knockout slices were resistant to LTP impairment. Furthermore, water-soluble extract from a non-demented individual was incubated on wild-type and *PRNP*$^{-/-}$ brain slices followed by stimulation, but had no impact on LTP for either genotype. Pretreating wild-type slices with an anti-PrP^{C} antibody directed against the unstructured central domain was also found to be protective against the loss of LTP from AD brain-derived extract. Therefore, PrP^{C} likely is necessary for human disease-derived Aβo to exert their plasticity-impairing effects.

To further evaluate the essential role of PrP^{C} for Aβo action in vivo, Wistar rats had one of their lateral ventricles cannulated. Through this cannula, water-soluble extract derived from the brains of patients with AD was infused. There was no change in baseline potentiation prior to induction, but there was a significant depression of LTP (Barry et al. 2011). A cohort of rats was infused with anti-PrP^{C} antibodies, D13 and ICSM-18, prior to receiving the brain-derived extract and HFS. These pretreated rats were fully protected from Aβo-induced loss of plasticity and LTP (Freir et al. 2011a; Barry et al. 2011). This strongly shows that the requirement of PrP^{C} for Aβo binding is relevant to AD.

3.7 PrP^{C} Is Necessary for Memory Impairment In Vivo

Until recently, the joint impact of Aβo and PrP^{C} on the performance of an in vivo learning and memory task had been unknown. Age-dependent memory loss is among the cardinal features of AD and can be tested in mice with a Morris water maze. In such a task, mice are placed in a large tank of water with a platform that is hidden from their view. Mice, being naturally averse to water, undertake a coordinated search strategy to find an exit. Over the course of repeated trials, the mice eventually learn the location of the hidden platform and escape quickly. Mice with spatial memory deficits take a significantly longer time in locating the platform to escape. This task is especially appropriate for better understanding AD as it has been shown that successfully completing the task relies on having a functioning hippocampus (Redish and Touretzky 1998).

A Morris water maze swim task was performed with wild-type mice, *PRNP*$^{-/-}$ mice, APPswe/Psen1 ΔE9 mice (an AD transgenic model), and APPswe/Psen1 ΔE9 *PRNP*$^{-/-}$ mice at 3 months and at 12 months (Gimbel et al. 2010). At 3 months, there was no apparent difference between any of the groups. At 12 months, the APPswe/Psen1 ΔE9 mice demonstrated significant impaired latencies to escape, while the APPswe/Psen1 ΔE9 mice lacking PrP^{C} had much faster latencies to escape,

and were equivalent to the wild-type mice. To test retention of the learned location, the hidden platform was removed. At 12 months, APPswe/Psen1 ΔE9 mice crossed over the area where the platform had been significantly fewer times than the APPswe/Psen1 ΔE9 mice lacking $PrP^{C.}$ The AD mice without PrP^{C} crossed the target area as many times as the wild-type group. The mice were also trained to avoid entering a darkened chamber by administration of an aversive shock. The APPswe/Psen1 ΔE9 mice did not remember this passive avoidance training and quickly went into the darkened chamber. In contrast, the PrP^{C} knockout APPswe/Psen1 ΔE9 mice demonstrated better learning by more prolonged avoidance of the darkened chamber (Gimbel et al. 2010). The levels of APP and Aβ were the same independent of genotype. These results are consistent with PrP^{C} being crucial for transgenic AD memory impairment.

Further support for the role of PrP^{C} in AD-related memory impairment comes from a study showing that short-term treatment with the 6D11 could reverse memory impairment in the APP/PS1 transgenic AD model (Chung et al. 2010). Transgenic mice received 10 high dose injections of the 6D11 antibody into their peritoneum over the course of 2 weeks. High doses were utilized so that a non-negligible amount of antibody would be able to successfully cross the blood–brain barrier. The mice were tested with a radial arm maze, and the number of errors that were made while completing the maze was counted. The number of errors that the treated APP/PS1 mice made was significantly fewer than that of the untreated APP/PS1 mice, and was not different from the error rate of wild-type mice. Again, treatment had no impact on amyloid burden, making a simple antagonism of the receptor the most likely mechanism for memory improvement.

Normally, when presented with a noveland familiar object, mice spend more time exploring the novel object compared to the familiar object. This forms the basis for the novel object recognition test, in which a memory-impaired mouse will not remember which object is novel and will show no preference for either object. $PRNP^{+/+}$ and $PRNP^{-/-}$ had a 100 μM solution of synthetic Aβo infused into their ventricles prior to testing over several days (Balducci et al. 2010). The pharmacokinetics of Aβo in this experiment are complicated because the starting dose is high, but the half-life of Aβ in the brain is very short, on the order of 1 h (Cirrito et al. 2003). The Aβ-injected $PRNP^{+/+}$ mice showed no preference for either object, consistent with memory impairment during some segments of the time. The Aβ-injected $PRNP^{-/-}$ mice did not show a preference for the novel object, but exhibited a preference for the familiar object. The authors interpreted these results to imply that PrP^{C} was not essential for Aβo-induced memory impairment. However, a preference for the familiar object rather than the novel object by the injected PrP knockout mice suggests intact memory, but altered novelty seeking. For transgenic AD mice, novel object recognition is less consistently impaired than is spatial memory (Chen et al. 2000).

Complicating the analysis further, hAPPJ20 mice, another transgenic AD model, had no preference for either object in the novel object recognition test with or without PrP^{C} (Cisse et al. 2011a). The hAPPJ20 mice also performed worse than the wild-type mice in a Morris water maze task, independent of PrP^{C} status. In fact, the

PRNP$^{-/-}$ hAPPJ20 mice did slightly worse than any other group in latency to escape and in the number of crosses over the platform area when the platform was removed. It has been previously shown however that the hAPPJ20 mice develop deficits at an early age that are not progressive (Harris et al. 2010). It can be hypothesized that PrP^{C} is necessary for the age-dependent loss of spatial memory seen in transgenic AD-like progression, but that juvenile-onset, age-independent impairment in hAPPJ20 mice occurs through a PrP^{C}-independent mechanism, perhaps involving EphB2 (Cisse et al. 2011b).

3.8 Neuronal Degeneration and Dysfunction Are Dependent upon PrP^{C}

Neurodegeneration is classically seen in AD, but most AD mouse models show limited neurodegeneration even in the face of significant amyloid burden. There have been reports however of monoamine neuronal degeneration in the AD model (Liu et al. 2008). Brains slices from APPswe/Psen1 ΔE9 show signs of axonal degeneration as evidenced by having fewer serotonin axons in the cerebral cortex than wild-type mice. The APPswe/Psen1 ΔE9/*PRNP*$^{-/-}$ brain have indistinguishable levels of serotonin-positive axons compared to wild-type mice, consistent with PrP^{C} being required for this form of AD transgene-induced degeneration (Gimbel et al. 2010).

Synaptophysin is a presynaptic marker and its level can be used to assay synaptic health. A loss of synapses is documented in AD, and APPswe/Psen1 ΔE9 mice show a decrease in levels of synaptophysin in the cortex (Gimbel et al. 2010). APPswe/Psen1 ΔE9 mice lacking PrP^{C} had similar levels of synaptophysin to that of wild-type mice (Gimbel et al. 2010). The postsynaptic marker PSD-95 was also preserved in APPswe/Psen1 ΔE9 PrP^{C} null mice (Gimbel et al. 2010). Excitingly, acute treatment with 6D11 anti-PrP antibody raises synaptophysin levels in the hippocampus of APPswe/Psen1 ΔE9 mice (Chung et al. 2010).

Transgenic AD mice have reduced survival with sudden unexplained deaths. It has been hypothesized that the sudden death may be mediated by hyperexcitability or status epilepticus (Minkeviciene et al. 2009). Over the course of 1 year, 40% of the APPswe/Psen1 ΔE9 mice died, while less than 4% of the APPswe/Psen1 ΔE9 *PRNP*$^{-/-}$ mice died (Gimbel et al. 2010). Wild-type mice experienced a less than 4% death rate as well. For this AD strain, PrP^{C} is essential for the early death phenotype.

Related to the sudden death phenotype, epileptiform discharges have been examined in hAPPJ20 mice with and without PrP^{C}. Knocking out PrP^{C} in this mouse strain slightly increased epileptiform spikes to about 15 per hour, although there were no convulsive seizures (Cisse et al. 2011a). Importantly, historical standards for hAPPJ20 have reported 100–1,000 spikes per hour (Roberson et al. 2011; Palop et al. 2007). Due to variability, single spikes may not be a robust phenotype. Consistent with the possible increase in spike discharges, the same group reported an increase in sudden death from the age of 30 days to 270 days for the hAPPJ20

mice without PrP^C compared to those with PrP^C (Cisse et al. 2011a). If the deaths during the first 30 days of life are included, the difference between the groups is nil. Either way, PrP^C does not appear to improve mortality in the hAPPJ20 mice, in contrast to the APPswe/Psen1 ΔE9 mice. This highlights the need for more research into the difference between the strains to explain the relevant factors. These studies simultaneously emphasize the importance and difficulty of modeling AD behavior in laboratory animals.

3.9 Human *PRNP* Genetics in AD

The possibility of an association between PrP^C genetic variation and AD has been considered in several studies. Most studies have focused on a common coding region variant, the presence of Met vs. Val at codon 129 (rs1799990). In particular, four studies found that the minor Val allele is underrepresented in the AD population (Gacia et al. 2006; Riemenschneider et al. 2004; Golanska et al. 2004; Dermaut et al. 2003). These studies also observed that M/V heterozygous state is less common among AD cases, suggesting that the homozygous state at residue 129 is a risk for AD. The interaction of the residue 129 status with age of onset and with ApoE genotype has not been consistent across these studies. A meta-analysis of published studies is available at AlzGene, and suggests limited, if any, association of the Val allele with AD http://www.alzforum.org/res/com/gen/alzgene/. In a genome-wide SNP study, Roses and colleagues confirmed a role of ApoE and identified new candidate risk loci for late onset Alzheimer's disease (LOAD) (Li et al. 2008). As part of that genome-wide study, a focused analysis of some 25 previously reported LOAD risk genes was completed and only *PRNP* achieved statistical significance in this large-scale genomic study (Li et al. 2008). The strongest association was with an intronic SNP of the *PRNP* gene. Altogether, the contribution of common genetic variants at the *PRNP* locus to AD does not appear to be strong. The potential presence of rare *PRNP* variants having a large effect for AD risk has not yet been explored.

3.10 Conclusion

A range of molecular, proteomic, electrophysiology, and behavioral data supports the hypothesis that PrP^C binding mediates a significant fraction of Aβo-specific pathophysiology in AD models. Additional work is required to understand the relative role of PrP^C in various mouse AD models, to elucidate coreceptors that function with PrP^C to mediate toxic effects, and to characterize the downstream signal transducers of PrP^C activation by amyloid oligomers (Fig. 3.1). Nonetheless, PrP^C remains an enticing target for pharmaceutical blockade, since deleting or antagonizing PrP^C function does not have substantial adverse effects in mice. Targeting PrP^C constitutes a unique strategy for rational disease-modifying AD therapy.

References

(1997) Consensus recommendations for the postmortem diagnosis of Alzheimer's disease. The National Institute on Aging, and Reagan Institute Working Group on Diagnostic Criteria for the Neuropathological Assessment of Alzheimer's Disease. Neurobiol Aging 18(4 Suppl):S1–S2

Alzheimer A et al (1995) An english translation of Alzheimer's 1907 paper, "Uber eine eigenartige Erkankung der Hirnrinde". Clin Anat 8(6):429–431

Balducci C et al (2010) Synthetic amyloid- oligomers impair long-term memory independently of cellular prion protein. Proc Natl Acad Sci USA 107(5):2295–2300

Barry AE et al (2011) Alzheimer's disease brain-derived amyloid-mediated inhibition of ltp in vivo is prevented by immunotargeting cellular prion protein. J Neurosci 31(20):7259–7263

Baumann F et al (2007) Lethal recessive myelin toxicity of prion protein lacking its central domain. EMBO J 26(2):538–547

Brookmeyer R et al (2007) Forecasting the global burden of Alzheimer's disease. Alzheimers Dement 3(3):186–191

Calella AM et al (2010) Prion protein and Abeta-related synaptic toxicity impairment. EMBO Mol Med 2(8):306–314

Chapman PF et al (1999) Impaired synaptic plasticity and learning in aged amyloid precursor protein transgenic mice. Nat Neurosci 2(3):271–276

Chen G et al (2000) A learning deficit related to age and beta-amyloid plaques in a mouse model of Alzheimer's disease. Nature 408(6815):975–979

Chen S, Yadav SP, Surewicz WK (2010) Interaction between human prion protein and amyloid-{beta} (A{beta}) oligomers: role of N-terminal residues. J Biol Chem 285(34):26377–26383

Chung E et al (2010) Anti-PrPC monoclonal antibody infusion as a novel treatment for cognitive deficits in an Alzheimer's disease model mouse. BMC Neurosci 11(1):130

Cirrito JR et al (2003) In vivo assessment of brain interstitial fluid with microdialysis reveals plaque-associated changes in amyloid-beta metabolism and half-life. J Neurosci 23(26):8844–8853

Cisse M et al (2011a) Ablation of cellular prion protein does not ameliorate abnormal neural network activity or cognitive dysfunction in the J20 line of human amyloid precursor protein transgenic mice. J Neurosci 31(29):10427–10431

Cisse M et al (2011b) Reversing EphB2 depletion rescues cognitive functions in Alzheimer model. Nature 469(7328):47–52

Citron M et al (1992) Mutation of the beta-amyloid precursor protein in familial Alzheimer's disease increases beta-protein production. Nature 360(6405):672–674

Cummings JL (2004) Alzheimer's disease. N Engl J Med 351(1):56–67

DeMattos RB et al (2004) ApoE and clusterin cooperatively suppress Abeta levels and deposition: evidence that ApoE regulates extracellular abeta metabolism in vivo. Neuron 41(2):193–202

Dermaut B et al (2003) PRNP Val129 homozygosity increases risk for early-onset Alzheimer's disease. Ann Neurol 53(3):409–412

Freir DB et al (2011) Interaction between prion protein and toxic amyloid β assemblies can be therapeutically targeted at multiple sites. Nat Commun 2:336

Gacia M et al (2006) Prion protein gene M129 allele is a risk factor for Alzheimer's disease. J Neural Transm 113(11):1747–1751

Gimbel DA et al (2010) Memory impairment in transgenic alzheimer mice requires cellular prion protein. J Neurosci 30(18):6367–6374

Glenner GG, Wong CW (1984) Alzheimer's disease and Down's syndrome: sharing of a unique cerebrovascular amyloid fibril protein. Biochem Biophys Res Commun 122(3):1131–1135

Golanska E et al (2004) Polymorphisms within the prion (PrP) and prion-like protein (Doppel) genes in AD. Neurology 62(2):313–315

Goldgaber D et al (1987) Characterization and chromosomal localization of a cDNA encoding brain amyloid of Alzheimer's disease. Science 235(4791):877–880

Gunther EC, Strittmatter SM (2009) β-amyloid oligomers and cellular prion protein in Alzheimer's disease. J Mol Med 88(4):331–338
Hardy J, Selkoe DJ (2002) The amyloid hypothesis of Alzheimer's disease: progress and problems on the road to therapeutics. Science 297(5580):353–356
Harris JA et al (2010) Many neuronal and behavioral impairments in transgenic mouse models of Alzheimer's disease are independent of caspase cleavage of the amyloid precursor protein. J Neurosci 30(1):372–381
Heister D et al (2011) Predicting MCI outcome with clinically available MRI and CSF biomarkers. Neurology 77(17):1619–1628
Helzner EP et al (2008) Survival in Alzheimer disease: a multiethnic, population-based study of incident cases. Neurology 71(19):1489–1495
Jack CR Jr et al (2010) Hypothetical model of dynamic biomarkers of the Alzheimer's pathological cascade. Lancet Neurol 9(1):119–128
Josephs KA et al (2008) Beta-amyloid burden is not associated with rates of brain atrophy. Ann Neurol 63(2):204–212
Katzman R (1986) Alzheimer's disease. N Engl J Med 314(15):964–973
Kessels HW et al (2010) The prion protein as a receptor for amyloid-beta. Nature 466(7308):E3–E4, discussion E4–E5
Kim J, Basak JM, Holtzman DM (2009) The role of apolipoprotein E in Alzheimer's disease. Neuron 63(3):287–303
Kosik KS, Joachim CL, Selkoe DJ (1986) Microtubule-associated protein tau (tau) is a major antigenic component of paired helical filaments in Alzheimer disease. Proc Natl Acad Sci USA 83(11):4044–4048
Laurén J et al (2009) Cellular prion protein mediates impairment of synaptic plasticity by amyloid-β oligomers. Nature 457(7233):1128–1132
Lesne S et al (2006) A specific amyloid-beta protein assembly in the brain impairs memory. Nature 440(7082):352–357
Li H et al (2008) Candidate single-nucleotide polymorphisms from a genomewide association study of Alzheimer disease. Arch Neurol 65(1):45–53
Liu Y et al (2008) Amyloid pathology is associated with progressive monoaminergic neurodegeneration in a transgenic mouse model of Alzheimer's disease. J Neurosci 28(51):13805–13814
Masters CL et al (1985) Amyloid plaque core protein in Alzheimer disease and Down syndrome. Proc Natl Acad Sci USA 82(12):4245–4249
McKhann GM et al (2011) The diagnosis of dementia due to Alzheimer's disease: recommendations from the National Institute on aging-Alzheimer's Association workgroups on diagnostic guidelines for Alzheimer's disease. Alzheimers Dement 7(3):263–269
Meek PD, McKeithan K, Schumock GT (1998) Economic considerations in Alzheimer's disease. Pharmacotherapy 18(2 Pt 2):68–73, discussion 79–82
Mills J, Reiner PB (1999) Regulation of amyloid precursor protein cleavage. J Neurochem 72(2):443–460
Minkeviciene R et al (2009) Amyloid beta-induced neuronal hyperexcitability triggers progressive epilepsy. J Neurosci 29(11):3453–3462
Morris RG et al (1986) Selective impairment of learning and blockade of long-term potentiation by an N-methyl-D-aspartate receptor antagonist, AP5. Nature 319(6056):774–776
Palop JJ et al (2007) Aberrant excitatory neuronal activity and compensatory remodeling of inhibitory hippocampal circuits in mouse models of Alzheimer's disease. Neuron 55(5):697–711
Petersen RC et al (2010) Alzheimer's Disease Neuroimaging Initiative (ADNI): clinical characterization. Neurology 74(3):201–209
Prasher VP et al (1998) Molecular mapping of Alzheimer-type dementia in Down's syndrome. Ann Neurol 43(3):380–383
Prince M et al (2009) Alzheimer's disease international world alzheimer report 2009. In: International AsD (ed) pp 1–96
Redish AD, Touretzky DS (1998) The role of the hippocampus in solving the Morris water maze. Neural Comput 10(1):73–111

Riemenschneider M et al (2004) Prion protein codon 129 polymorphism and risk of Alzheimer disease. Neurology 63(2):364–366
Roberson ED et al (2011) Amyloid-beta/Fyn-induced synaptic, network, and cognitive impairments depend on tau levels in multiple mouse models of Alzheimer's disease. J Neurosci 31(2):700–711
Selkoe DJ, Schenk D (2003) Alzheimer's disease: molecular understanding predicts amyloid-based therapeutics. Annu Rev Pharmacol Toxicol 43:545–584
Shankar GM et al (2008) Amyloid-beta protein dimers isolated directly from Alzheimer's brains impair synaptic plasticity and memory. Nat Med 14(8):837–842
Shaw LM et al (2009) Cerebrospinal fluid biomarker signature in Alzheimer's disease neuroimaging initiative subjects. Ann Neurol 65(4):403–413
Sperling RA et al (2011) Toward defining the preclinical stages of Alzheimer's disease: recommendations from the National Institute on Aging-Alzheimer's Association workgroups on diagnostic guidelines for Alzheimer's disease. Alzheimers Dement 7(3):280–292
Terry RD et al (1991) Physical basis of cognitive alterations in Alzheimer's disease: synapse loss is the major correlate of cognitive impairment. Ann Neurol 30(4):572–580
Thies W, Bleiler L (2011) 2011 Alzheimer's disease facts and figures. Alzheimers Dement 7(2):208–244
Walsh DM, Selkoe DJ (2007) A? Oligomers ? a decade of discovery. J Neurochem 101(5): 1172–1184
Walsh DM et al (2002) Naturally secreted oligomers of amyloid beta protein potently inhibit hippocampal long-term potentiation in vivo. Nature 416(6880):535–539
Wang HY et al (2000) Beta-Amyloid(1–42) binds to alpha7 nicotinic acetylcholine receptor with high affinity. Implications for Alzheimer's disease pathology. J Biol Chem 275(8):5626–5632
Wang HW et al (2002) Soluble oligomers of beta amyloid (1–42) inhibit long-term potentiation but not long-term depression in rat dentate gyrus. Brain Res 924(2):133–140
Wisniewski T, Ghiso J, Frangione B (1991) Peptides homologous to the amyloid protein of Alzheimer's disease containing a glutamine for glutamic acid substitution have accelerated amyloid fibril formation. Biochem Biophys Res Commun 179(3):1247–1254
Yan SD et al (1996) RAGE and amyloid-beta peptide neurotoxicity in Alzheimer's disease. Nature 382(6593):685–691