Published in final edited form as:

Neurobiol Aging. 2010 April ; 31(4): 542–548. doi:10.1016/j.neurobiolaging.2008.05.026.

COMMENTARY: Molecular polymorphism of A β in Alzheimer's disease

Harry LeVine III^{1,*} and Lary C. Walker²

¹Department of Molecular and Cellular Biochemistry, Chandler School of Medicine and the Center on Aging, University of Kentucky, Lexington, KY

²Yerkes National Primate Research Center and Department of Neurology, Emory University, Atlanta, GA

Keywords

PIB; transgenic mice; stoichiometry; high affinity binding; Thioflavine T; Congo Red; polythiophene

1. Introduction

Alzheimer's disease is defined pathologically by the presence of senile plaques, which consist primarily of extracellular aggregates of fibrillar A β peptide, and neurofibrillary tangles, which are abnormal, intracellular bundles of fibrillar tau protein. The advent of amyloid binding agents as diagnostic imaging probes for Alzheimer's disease (AD) has made it imperative to understand at a molecular and disease level what these ligands are reporting. In addition to improving the accuracy of diagnosis, we argue that these selective ligands can serve as probes for molecular polymorphisms that may govern the pathogenicity of abnormal protein aggregates.

2. There is a disconnect between amyloid pathology and neurodegeneration/clinical symptoms

A definite histopathologic diagnosis of AD requires the presence of senile plaques. Whether they are directly linked to neurodegeneration or are an epiphenomenon of the pathogenic process, the presence of plaques must be explained by any proposed disease mechanism. Their utility as biomarkers, though, is hampered by the weak correlation of the amount and regional distribution of A β deposition with the extent of neurodegeneration that results in functional deficits [5,8,50]. Although some investigators interpret the poor correlation to mean that the senile plaques are secondary to the essential disease process, a great deal of genetic and biochemical data indicate that A β is a prime-mover in the pathogenic cascade [12]. The amended Amyloid Cascade Hypothesis (perhaps better designated the A β Cascade Hypothesis) holds that soluble, multimeric forms of misfolded A β (i.e., A β -oligomers) are the most toxic

© 2008 Elsevier Inc. All rights reserved.

*Corresponding Author: Harry LeVine, III, ADDRESS: 209 Sanders-Brown Building, 800 S. Limestone Street, Lexington, KY 40536-0230 USA, hlevine@email.uky.edu, PHONE: 859-257-1412 X224, FAX: 859-323-2866.

The authors declare that there are no actual or potential conflicts of interest for the views expressed in this manuscript

Publisher's Disclaimer: This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final citable form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

species, whereas insoluble fibrils, which comprise the bulk of senile plaque cores, serve as either a less toxic depot or as a sink in equilibrium with monomers and/or oligomers.

3. Animal models have substantial A β deposition but relatively little neurodegeneration

Any postulated Alzheimer's disease *mechanism* must be reconciled with the stereotypical A β deposition disease *hallmarks*. It is critical to determine the association of A β deposits with the disease to validate the utility of imaging senile plaques in diagnosing AD and/or monitoring disease progression. Nonhuman species have proven to be only partial models of AD. Although APP-transgenic mice, nonhuman primates, and dogs all manifest age-related CNS A β deposition that is similar in distribution and quantity to that in humans with AD, these models lack the massive neurodegeneration and neurofibrillary tangles of AD [9,29]. Is it possible that the A β deposits in nonhuman animals are structurally different on a molecular/conformational level from AD A β deposits, and is this difference directly or indirectly associated with disease mechanism? Understanding the molecular diversity of A β multimers could provide important clues to the pathogenesis of AD, and could thereby suggest new therapeutic approaches to the disorder.

4. When is a misfolded protein deposit pathologic?

Insoluble protein deposits have classically been characterized by the presence or absence of fibrillar morphology and a condensed core, as well as by their staining properties with amyloid dyes such as Congo Red (Figure 1 [6]) and the Thioflavines (T and S, Figure 1 [1, 5]). Similar characteristics have been observed in deposits generated by animal models of A β deposition, yet neuronal loss is minimal in these instances, suggesting that some as yet unidentified property of the multimeric protein renders the molecule uniquely pathogenic in humans.

The prion diseases are the best characterized example of how conformational variants of the same primary amino acid sequence can lead to different disease phenotypes. During the pathogenesis of prionosis, the normal cellular conformation of the prion protein (PrP^C) is converted into β -sheet-rich, pathogenic conformations (PrP^{Sc}) that are resistant to denaturation and proteolysis. Distinct polymorphic forms of this multimeric PrP^{Sc}, also called *strains*, are differently folded molecules that present pathologically different phenotypes and morphologies [1,7]. Diverse structural variants of prions also can be generated *in vitro* from the same amino acid sequence and starting conformation of prion protein by adjusting the assembly conditions (temperature, pH, agitation) [30,46]. Each strain of prion protein tends to elicit a particular lesion phenotype and clinical course of the individual prion diseases that is dependent on characteristics of both the host and the protein [44,47,48].

5. Polymorphic forms of A β

Although the story is not as fully developed for A β , polymorphic forms of A β peptide fibrils also have been produced *in vitro* [40,49,52]. Distinct folded structures of two forms differing in hydrogen bonding across the β -turn between residues 25 and 28 have been determined by solid state NMR [40,41]. There is also evidence that pH similar to that in endosomal compartments influences epitope exposure in residues 8-14 of soluble monomeric A β peptide, reflecting a distinct, partially folded conformation [27,34]. *In vivo*, heterogeneous A β [3,51] deposits suggestive of A β strains have been induced by different exogenous A β seeds in APP-transgenic mice [35]. The molecular relationship between any of these molecular forms and the A β fibrils and oligomers found in AD brain remains to be determined. Another important question, given the strong correlation between neurofibrillary tangles and degree of dementia

in AD [5,8,50], is whether certain A β strains might preferentially promote the development of neurofibrillary tangles.

Polymorphism in the molecular structure of A β multimers is a potential explanation for the observation that only humans have the severe neuronal loss, neurofibrillary tangles and cognitive decline of AD, and nonhuman species do not. Strain differences might also account for the fact that some cognitively normal humans have large amounts of cerebral A β [4], and for other features of the disorder such as inflammation and cerebral β -amyloid angiopathy (CAA) [3,56]. Furthermore, morphological and distributional variants of A β deposits coexist within an individual AD brain [51]. These lesions include dense-core senile plaques, CAA, and a multiplicity of diffuse, granular and ribbon-like deposits [13,45,51,53]. Whether these morphologically diverse lesions are the supramolecular manifestations of different A β strains remains to be established.

6. Recognizing A β Polymorphisms

Classical methods for detecting structural differences in cerebral A β deposits do not make fine distinctions at the molecular level. Widely used A β sequence-specific antibodies such as 6E10 and 4G8, and techniques such as amyloidotrophic Thioflavine- and Congo Red-staining are of limited utility in distinguishing different types of amyloid deposits. Conformation-specific antibodies that distinguish amyloid fibrils [18,26,36,39] and oligomers are not primary amino acid sequence-dependent (except see [26,36]). However, antibodies that distinguish among fibrillar A β strains, similar to those available for PrP, are not yet available. Some of the anti-oligomer antibodies may indeed recognize conformational variants, notably the A11, I11 and OC (officer) antibodies [17,26,55].

Putative structural varieties of A β can be distinguished biochemically by differential extraction of AD brain tissue. As AD progresses, it is possible to demonstrate characteristic shifts of A β into increasingly insoluble multimers, beginning with a buffer-soluble state, followed by alkali-soluble, SDS-soluble, and finally requiring formic acid to solubilize the peptide. Similar distributions of peptide solubility also are found in the animal models [16,25,37].

A series of fluorescent polythiophene anions and cations have recently been described [6] that report on the spatial (molecular?) heterogeneity of multimeric A β . Under appropriate conditions these agents bind the stacks of assembled β -sheets in A β and other amyloid fibrils in a sequence-independent fashion [2]. Their fluorescence emission spectra reflect intra- and inter-polythiophene chain contacts which shift to green for tightly packed, regularly oriented fibrils and to red for less organized, less compact fibrils [38]. Synthetic A β -amyloid fibrils formed under quiescent conditions fluoresce green with these reagents, while fibrils formed with agitation emit red. These compounds also recognize molecular heterogeneity in A β deposits formed *in vivo* [34]. The fibrillar Thioflavine-positive amyloid deposits in Tg APP/PS1 mice and AD brain are mosaics of the two fibril conformations, indicating that individual fibril deposits consist of polymorphic domains [38]. These compact lesions also are distinct from diffuse or non-fibrillar deposits, which are Thioflavine- and Congo Red-negative. Structural evidence supporting the potential for differential fibril organization comes from solid state NMR, which shows that the synthetic A β peptide folds differently in fibrils prepared under quiescent vs. agitated conditions [40].

7. Amyloid probes and polymorphic A β

A probe of fibrillar A β , the N- ^{11}C -labeled benzothiazole ligand 6-OH-BTA-1 (Pittsburgh compound B; ^{11}C -PIB) (Figure 1 [2]), a brain-penetrant analog of BTA-1 (Figure 1, [3]), is retained in A β -rich brain regions of living MCI and AD subjects at a high affinity binding site, but it washes rapidly out of unaffected regions or appropriate control brains [20]. Positron-

Emission-Tomography (PET) images of these subjects can be processed into a map of putative A β -deposition in the living brain. Neurofibrillary tangles (hyperphosphorylated tau) [22] and Lewy Bodies (α -synuclein) [10], which also contain β -sheet structure, have low affinity for PIB, so they are not expected to contribute significantly to the signal at imaging concentrations of PIB used in AD subjects. Frontotemporal dementia (a primary tauopathy) gives detectable, but much weaker, PIB signals in some subjects [42], but the retention of label is distinguishable by its regional distribution. The possible contribution of a small amount of A β plaque pathology in these cases has not been excluded. PIB uptake is increased in disease-relevant brain regions in MCI subjects [11,19]. CAA, which is found to varying degrees in most AD subjects, also binds PIB [15]. When ^3H -PIB is applied to AD histological sections, dense-core plaques, CAA and some diffuse A β deposits bind the agent, but NFTs do not [14,31]. Studies are underway to determine whether PIB retention can be used to follow the progression of AD and whether it will be useful for monitoring the effectiveness of therapeutic interventions.

8. PIB detects differences between animal models and AD

The sulfonated Congo Red [6] and Thioflavine S [5] dyes are used routinely to analyze AD brain pathology. Congo Red and its congeners (Chrysamine G, X-34, BSB; Figure 1 [7-9]), as well as Thioflavine S, bind to sites distinct from the high affinity site that is occupied by PIB [32,54]. In fibrils made with synthetic A β 1-40 or A β 1-42 peptide, and in plaque-rich transgenic mouse and AD brain, Congo Red binds fibrils in a 1:1 stoichiometry with A β [21,28]. PIB binding may be more selective on a molecular level than are Congo Red and Thioflavine S. The Klunk group made the seminal observation that when ^{11}C -PIB of the same specific radioactivity and concentration ($\sim\text{nM}$) used for human studies was used to image APP/PS1 transgenic mice (Tg2576 X M146L-PS1) with an A β plaque load equal to or exceeding that in AD brain, the plaques did not bind enough radioligand to produce an image [21]. However, the murine A β plaque pathology was revealed by staining sections with 100 nM 6-CN-BTA-1 (a close analog of PIB [6-OH-BTA-1]), X-34, or with anti-A β antibodies. Subsequent quantitative binding studies comparing brain homogenates from AD, non-demented humans, and plaque-bearing transgenic mice revealed that the transgenic mice had a greatly reduced number of high affinity (nM) PIB binding sites per mole of A β peptide ($< 1:100$ compared to 1:2 in AD). These high affinity sites would be the only sites significantly labeled at the concentrations of ^{11}C -PIB usually achieved *in vivo*, hence the weak signal in transgenic mice. The high affinity binding stoichiometry was even lower (1:370) on fibrils of A β 1-40 or A β 1-42 formed from synthetic peptide [21]. The structural basis at the molecular level for these differences remains to be elucidated.

9. Implications

An intriguing possibility is that the high affinity PIB binding site represents a molecular polymorph of A β that constitutes a high proportion of deposits in AD brain, but a low proportion in APP transgenic mouse brain and in fibrils of synthetic peptide. Support for this hypothesis comes from imaging studies with ten-fold higher specific activity ^{11}C -PIB, which is sufficient to detect the lower percentage of high affinity binding sites in the APP23 Tg mouse and in histological sections of Tg2576 and Tg2576 X A260V-PS1 brains [33]. *In vivo* binding, homogenate binding, and autoradiographic studies of the Tg2576 X M146L-PS1 mice with ^{125}I -IMPY ([4], an analog of PIB) of similar high specific radioactivity also detect a low abundance, high affinity binding site that is associated with plaques and cerebrovascular amyloid fibrils, but only minimally with neurofibrillary tangles [23,24].

Whether the material in brain that is selectively labeled by PIB is a cause or effect of the pathogenic process in AD, it appears to be a marker for an aspect of the disease that is only minimally recapitulated by synthetic peptides and current APP transgenic mouse models. Aged

nonhuman primates and dogs naturally deposit human-sequence A β in the brain and display some behavioral dysfunction, but, like APP-transgenic mice, they do not exhibit extensive neuronal loss or dementia. Our preliminary studies of aged nonhuman primates with heavy A β -amyloidosis [43] support the hypothesis that PIB recognizes an enriched human molecular morphotype of A β . PIB binding, along with other new molecular probes, could be useful tools for assessing efforts to expand current models beyond recreating Alzheimer's histopathology to more accurately phenocopying Alzheimer's disease.

Acknowledgments

Support is acknowledged from the Woodruff Foundation, NIH RR-00165, P01 AG026423-01A2, and by the Sanders-Brown Center on Aging and Chandler Medical Center of the University of Kentucky.

References

1. Aguzzi A, Heikenwalder M, Polymenidou M. Insights into prion strains and neurotoxicity. *Nat Rev Mol Cell Biol* 2007;8(7):552–61. [PubMed: 17585315]
2. Aslund A, Herland A, Hammarstrom P, Nilsson KP, Jonsson BH, Inganas O, Konradsson P. Studies of Luminescent Conjugated Polythiophene Derivatives: Enhanced Spectral Discrimination of Protein Conformational States. *Bioconjug Chem*. 2007
3. Attems J, Lintner F, Jellinger KA. Amyloid beta peptide 1-42 highly correlates with capillary cerebral amyloid angiopathy and Alzheimer disease pathology. *Acta Neuropathol (Berl)* 2004;107(4):283–91. [PubMed: 14986026]
4. Bennett DA, Schneider JA, Arvanitakis Z, Kelly JF, Aggarwal NT, Shah RC, Wilson RS. Neuropathology of older persons without cognitive impairment from two community-based studies. *Neurology* 2006;66(12):1837–44. [PubMed: 16801647]
5. Bierer LM, Hof PR, Purohit DP, Carlin L, Schmeidler J, Davis KL, Perl DP. Neocortical neurofibrillary tangles correlate with dementia severity in Alzheimer's disease. *Arch Neurol* 1995;52(1):81–8. [PubMed: 7826280]
6. Chandra R, Kung MP, Kung HF. Design, synthesis, and structure-activity relationship of novel thiophene derivatives for beta-amyloid plaque imaging. *Bioorg Med Chem Lett* 2006;16(5):1350–2. [PubMed: 16325402]
7. Collinge J, Clarke AR. A general model of prion strains and their pathogenicity. *Science* 2007;318(5852):930–6. [PubMed: 17991853]
8. Crystal H, Dickson D, Fuld P, Masur D, Scott R, Mehler M, Masdeu J, Kawas C, Aronson M, Wolfson L. Clinico-pathologic studies in dementia: nondemented subjects with pathologically confirmed Alzheimer's disease. *Neurology* 1988;38(11):1682–7. [PubMed: 3185902]
9. Eriksen JL, Janus CG. Plaques, tangles, and memory loss in mouse models of neurodegeneration. *Behavior genetics* 2007;37(1):79–100. [PubMed: 17072762]
10. Fodero-Tavoletti MT, Smith DP, McLean CA, Adlard PA, Barnham KJ, Foster LE, Leone L, Perez K, Cortes M, Culvenor JG, Li QX, Laughton KM, Rowe CC, Masters CL, Cappai R, Villemagne VL. In vitro characterization of Pittsburgh compound-B binding to Lewy bodies. *J Neurosci* 2007;27(39):10365–71. [PubMed: 17898208]
11. Forsberg A, Engler H, Almkvist O, Blomquist G, Hagman G, Wall A, Ringheim A, Langstrom B, Nordberg A. PET imaging of amyloid deposition in patients with mild cognitive impairment. *Neurobiol Aging*. 2007
12. Hardy J, Selkoe DJ. The amyloid hypothesis of Alzheimer's disease: progress and problems on the road to therapeutics. *Science* 2002;297(5580):353–6. [PubMed: 12130773]
13. Ikeda S, Allsop D, Glenner GG. Morphology and distribution of plaque and related deposits in the brains of Alzheimer's disease and control cases. An immunohistochemical study using amyloid beta-protein antibody. *Lab Invest* 1989;60(1):113–22. [PubMed: 2642985]
14. Ikonomic MD, Klunk WE, Abrahamson EE, Mathis CA, Price JC, Tsopelas ND, Lopresti BJ, Ziolkowski S, Bi W, Paljug WR, Debnath ML, Hope CE, Isanski BA, Hamilton RL, Dekosky ST. Post-

mortem correlates of in vivo PiB-PET amyloid imaging in a typical case of Alzheimer's disease. *Brain*. 2008

15. Johnson KA, Gregas M, Becker JA, Kinnecom C, Salat DH, Moran EK, Smith EE, Rosand J, Rentz DM, Klunk WE, Mathis CA, Price JC, Dekosky ST, Fischman AJ, Greenberg SM. Imaging of amyloid burden and distribution in cerebral amyloid angiopathy. *Ann Neurol*. 2007
16. Kalback W, Watson MD, Kokjohn TA, Kuo YM, Weiss N, Luehrs DC, Lopez J, Brune D, Sisodia SS, Staufenbiel M, Emmerling M, Roher AE. APP transgenic mice Tg2576 accumulate Abeta peptides that are distinct from the chemically modified and insoluble peptides deposited in Alzheimer's disease senile plaques. *Biochemistry* 2002;41(3):922–8. [PubMed: 11790115]
17. Kaye R, Glabe CG. Conformation-dependent anti-amyloid oligomer antibodies. *Methods Enzymol* 2006;413:326–44. [PubMed: 17046404]
18. Kaye R, Head E, Sarsoza F, Saing T, Cotman CW, Necula M, Margol L, Wu J, Breydo L, Thompson JL, Rasool S, Gurlo T, Butler P, Glabe CG. Fibril specific, conformation dependent antibodies recognize a generic epitope common to amyloid fibrils and fibrillar oligomers that is absent in prefibrillar oligomers. *Mol Neurodegener* 2007;2(1):18. [PubMed: 17897471]
19. Kempainen NM, Aalto S, Wilson IA, Nagren K, Helin S, Bruck A, Oikonen V, Kailajarvi M, Scheinin M, Viitanen M, Parkkola R, Rinne JO. PET amyloid ligand [11C]PIB uptake is increased in mild cognitive impairment. *Neurology* 2007;68(19):1603–6. [PubMed: 17485647]
20. Klunk WE, Engler H, Nordberg A, Wang Y, Blomqvist G, Holt DP, Bergstrom M, Savitcheva I, Huang GF, Estrada S, Ausen B, Debnath ML, Barletta J, Price JC, Sandell J, Lopresti BJ, Wall A, Koivisto P, Antoni G, Mathis CA, Langstrom B. Imaging brain amyloid in Alzheimer's disease with Pittsburgh Compound-B. *Ann Neurol* 2004;55(3):306–19. [PubMed: 14991808]
21. Klunk WE, Lopresti BJ, Debnath ML, Holt DP, Wang Y, Huang G-F, Shao L, Lefterov I, Koldamova R, Ikonovic M, DeKosky ST, Mathis CA. Amyloid deposits in transgenic PS1/APP mice do not bind the amyloid PET tracer, PIB, in the same manner as human brain amyloid. *Neurobiology of Aging* 2004;25(S2):232.
22. Klunk WE, Wang Y, Huang GF, Debnath ML, Holt DP, Shao L, Hamilton RL, Ikonovic MD, DeKosky ST, Mathis CA. The binding of 2-(4'-methylaminophenyl)benzothiazole to postmortem brain homogenates is dominated by the amyloid component. *J Neurosci* 2003;23(6):2086–92. [PubMed: 12657667]
23. Kung MP, Hou C, Zhuang ZP, Cross AJ, Maier DL, Kung HF. Characterization of IMPY as a potential imaging agent for beta-amyloid plaques in double transgenic PSAPP mice. *Eur J Nucl Med Mol Imaging*. 2004;10.1007/S00259-004-1487-z
24. Kung MP, Hou C, Zhuang ZP, Zhang B, Skovronsky D, Trojanowski JQ, Lee VM, Kung HF. IMPY: an improved thioflavin-T derivative for in vivo labeling of beta-amyloid plaques. *Brain Res* 2002;956(2):202–10. [PubMed: 12445687]
25. Kuo YM, Kokjohn TA, Beach TG, Sue LI, Brune D, Lopez JC, Kalback WM, Abramowski D, Sturchler Pierrat C, Staufenbiel M, Roher AE. Comparative analysis of amyloid-beta chemical structure and amyloid plaque morphology of transgenic mouse and Alzheimer's disease brains. *J Biol Chem* 2001;276(16):12991–8. [PubMed: 11152675]
26. Lambert MP, Velasco PT, Chang L, Viola KL, Fernandez S, Lacor PN, Khuon D, Gong Y, Bigio EH, Shaw P, De Felice FG, Krafft GA, Klein WL. Monoclonal antibodies that target pathological assemblies of Abeta. *J Neurochem* 2007;100(1):23–35. [PubMed: 17116235]
27. LeVine H III. Y10W beta(1-40) fluorescence reflects epitope exposure in conformers of Alzheimer's beta-peptide. *Arch Biochem Biophys* 2003;417(1):112–22. [PubMed: 12921787]
28. LeVine H III. Multiple ligand binding sites on A beta(1-40) fibrils. *Amyloid* 2005;12(1):5–14. [PubMed: 16076606]
29. LeVine, H., III; Walker, LC. Chapter 11. Models of Alzheimer's Disease. In: Conn, MP., editor. *Handbook of Models for Human Aging*. New York, NY: Academic Press/Elsevier; 2006. p. 121-34.
30. Lim KH, Nguyen TN, Damo SM, Mazur T, Ball HL, Prusiner SB, Pines A, Wemmer DE. Solid-state NMR structural studies of the fibril form of a mutant mouse prion peptide PrP89-143(P101L). *Solid state nuclear magnetic resonance* 2006;29(13):183–90. [PubMed: 16256316]

31. Lockhart A, Lamb JR, Osredkar T, Sue LI, Joyce JN, Ye L, Libri V, Leppert D, Beach TG. PIB is a non-specific imaging marker of amyloid-beta (A β) peptide-related cerebral amyloidosis. *Brain*. 2007
32. Lockhart A, Ye L, Judd DB, Merritt AT, Lowe PN, Morgenstern JL, Hong G, Gee AD, Brown J. Evidence for the presence of three distinct binding sites for the thioflavin T class of Alzheimer's disease PET imaging agents on beta-amyloid peptide fibrils. *J Biol Chem* 2005;280(9):7677–84. [PubMed: 15615711]
33. Maeda J, Ji B, Irie T, Tomiyama T, Maruyama M, Okauchi T, Staufenbiel M, Iwata N, Ono M, Saido TC, Suzuki K, Mori H, Higuchi M, Suhara T. Longitudinal, quantitative assessment of amyloid, neuroinflammation, and anti-amyloid treatment in a living mouse model of Alzheimer's disease enabled by positron emission tomography. *J Neurosci* 2007;27(41):10957–68. [PubMed: 17928437]
34. Matsunaga Y, Saito N, Fujii A, Yokotani J, Takakura T, Nishimura T, Esaki H, Yamada T. A pH-dependent conformational transition of A β peptide and physicochemical properties of the conformers in the glial cell. *Biochem J* 2002;361(Pt 3):547–56. [PubMed: 11802784]
35. Meyer-Luehmann M, Coomaraswamy J, Bolmont T, Kaeser S, Schaefer C, Kilger E, Neuenschwander A, Abramowski D, Frey P, Jaton AL, Vigouret JM, Paganetti P, Walsh DM, Mathews PM, Ghiso J, Staufenbiel M, Walker LC, Jucker M. Exogenous induction of cerebral beta-amyloidogenesis is governed by agent and host. *Science* 2006;313(5794):1781–4. [PubMed: 16990547]
36. Miller DL, Currie JR, Mehta PD, Potempska A, Hwang YW, Wegiel J. Humoral Immune Response to Fibrillar beta-Amyloid Peptide. *Biochemistry* 2003;42(40):11682–92. [PubMed: 14529278]
37. Murphy MP, Beckett TL, Ding Q, Patel E, Markesbery WR, Clair DKS, LeVine H, Keller JN. A β solubility and deposition during AD progression and in APPXPS1 Knock-in mice. *Neurobiology of Disease*. 2007
38. Nilsson KP, Aslund A, Berg I, Nystrom S, Konradsson P, Herland A, Inganas O, Stabo-Eeg F, Lindgren M, Westermarck GT, Lannfelt L, Nilsson LN, Hammarstrom P. Imaging distinct conformational states of amyloid-beta fibrils in Alzheimer's disease using novel luminescent probes. *ACS Chem Biol* 2007;2(8):553–60. [PubMed: 17672509]
39. O'Nuallain B, Wetzel R. Conformational Abs recognizing a generic amyloid fibril epitope. *Proceedings of the National Academy of Sciences of the United States of America* 2002;99(3):1485–90. [PubMed: 11818542]
40. Petkova AT, Leapman RD, Guo Z, Yau WM, Mattson MP, Tycko R. Self-propagating, molecular-level polymorphism in Alzheimer's beta-amyloid fibrils. *Science* 2005;307(5707):262–5. [PubMed: 15653506]
41. Petkova AT, Yau WM, Tycko R. Experimental Constraints on Quaternary Structure in Alzheimer's beta-Amyloid Fibrils. *Biochemistry* 2006;45(2):498–512. [PubMed: 16401079]
42. Rabinovici GD, Furst AJ, O'Neil JP, Racine CA, Mormino EC, Baker SL, Chetty S, Patel P, Pagliaro TA, Klunk WE, Mathis CA, Rosen HJ, Miller BL, Jagust WJ. 11C-PIB PET imaging in Alzheimer disease and frontotemporal lobar degeneration. *Neurology* 2007;68(15):1205–12. [PubMed: 17420404]
43. Rosen, RF.; LeVine, H.; Walker, LC. PIB Binding in Alzheimer's Disease and Nonhuman Primate Brain. 11th International Conference on Alzheimer's Disease and Related Disorders; Chicago, IL. 2008.
44. Somerville RA. Host and transmissible spongiform encephalopathy agent strain control glycosylation of PrP. *J Gen Virol* 1999;80(Pt 7):1865–72. [PubMed: 10423157]
45. Tagliavini F, Giaccone G, Frangione B, Bugiani O. Pre-amyloid deposits in the cerebral cortex of patients with Alzheimer's disease and nondemented individuals. *Neurosci Lett* 1988;93(23):191–6. [PubMed: 3241644]
46. Toyama BH, Kelly MJ, Gross JD, Weissman JS. The structural basis of yeast prion strain variants. *Nature* 2007;449(7159):233–7. [PubMed: 17767153]
47. Walker L, LeVine H, Jucker M. Koch's postulates and infectious proteins. *Acta Neuropathol* 2006;112(1):1–4. [PubMed: 16703338]
48. Weissmann C. The state of the prion. *Nat Rev Microbiol* 2004;2(11):861–71. [PubMed: 15494743]

49. Wetzel R, Shivaprasad S, Williams AD. Plasticity of Amyloid Fibrils. *Biochemistry* 2007;46(1):1–10. [PubMed: 17198370]
50. Wilcock GK, Esiri MM. Plaques, tangles and dementia. A quantitative study. *J Neurol Sci* 1982;56(23):343–56. [PubMed: 7175555]
51. Wisniewski HM, Bancher C, Barcikowska M, Wen GY, Currie J. Spectrum of morphological appearance of amyloid deposits in Alzheimer's disease. *Acta Neuropathol (Berl)* 1989;78(4):337–47. [PubMed: 2551122]
52. Yagi H, Ban T, Morigaki K, Naiki H, Goto Y. Visualization and Classification of Amyloid beta Supramolecular Assemblies. *Biochemistry* 2007;46(51):15009–17. [PubMed: 18044976]
53. Yamaguchi H, Hirai S, Morimatsu M, Shoji M, Ihara Y. A variety of cerebral amyloid deposits in the brains of the Alzheimer-type dementia demonstrated by beta protein immunostaining. *Acta Neuropathol* 1988;76(6):541–9. [PubMed: 3059748]
54. Ye L, Morgenstern JL, Gee AD, Hong G, Brown J, Lockhart A. Delineation of PET imaging agent binding sites on beta -amyloid peptide fibrils. *J Biol Chem* 2005;280(25):23599–604. [PubMed: 15855161]
55. Yoshiike Y, Kaye R, Milton SC, Takashima A, Glabe CG. Pore-forming proteins share structural and functional homology with amyloid oligomers. *Neuromolecular Med* 2007;9(3):270–5. [PubMed: 17914185]
56. Zhang-Nunes SX, Maat-Schieman ML, van Duinen SG, Roos RA, Frosch MP, Greenberg SM. The cerebral beta-amyloid angiopathies: hereditary and sporadic. *Brain Pathol* 2006;16(1):30–9. [PubMed: 16612980]

Figure 1.
Amyloid-Binding Ligands

- [1] Thioflavine T
- [2] PIB1
- [3] BTA-1
- [4] IMPY
- [5] Thioflavine S (mixture)

- [6] Congo Red
- [7] Chrysamine G
- [8] X-34
- [9] BSB