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Amyloid burden in cognitively normal elderly is associated with preferential hippocampal subfield volume loss

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Abstract

The earliest sites of brain atrophy in Alzheimer's disease are in the medial temporal lobe, following widespread cerebral cortical amyloid deposition. We assessed 74 cognitively normal participants with clinical measurements, A β -PET imaging, MRI, and a newly developed technique for MRI-based hippocampal subfield segmentation to determine the differential association of amyloid deposition and hippocampal subfield volume. Compared to amyloid-negative participants, amyloid-positive participants had significantly smaller hippocampal tail, presubiculum, subiculum, and total hippocampal gray matter volumes. We conclude that, prior to the development of cognitive impairment, atrophy in particular hippocampal subfields (HS) occurs preferentially with A β accumulation.

Keywords

Amyloid accumulation; hippocampal subfield volumes; cognitively normal elderly; subiculum; presubiculum

Introduction

According to the amyloid hypothesis, a predominant feature of Alzheimer's disease (AD) pathogenesis is the deposition of amyloid-beta (A β) peptide in the form of senile plaques in brain tissue [1–3]. The accumulation of A β appears to precede other AD pathophysiology, including cortical volume loss [4–6]. However, thus far clearance of A β plaques in

symptomatic AD patients is not associated with improved survival [7]. The failure of such A β clearance therapies may be due to their initiation late in the overall course of AD, as it is now recognized that the accumulation of A β deposits and the resulting pathophysiological process is ongoing years prior to any clinical manifestations [8]. The concept of preclinical AD initially was based on brain autopsy studies that found substantial numbers of A β plaques and other lesions characteristic of AD in cognitively normal individuals [9–11]. More recently, studies using the PET tracer ^{11}C Pittsburgh compound B (PIB), which specifically binds to A β plaques, have provided insights into the nature of amyloid accumulation [12]. PIB retention in cognitively normal older adults predicts progression to early symptomatic AD and AD dementia and is greater in individuals with symptomatic AD than in cognitively normal individuals [13–15].

Although A β accumulation begins in the neocortex, cortical atrophy – another hallmark of AD pathogenesis – first appears in the medial temporal regions [10,16–20]. Atrophy of the whole hippocampus has been correlated with A β accumulation as detected by PIB, but shows moderate sensitivity and low specificity to AD [14,21–26]. Van Leemput *et al.* have developed a fully automated method of hippocampal subfield (HS) segmentation in ultra-high resolution MRI data [27]. Investigation of the relationship between A β accumulation and differential hippocampal atrophy at the subfield level may be informative in the understanding the progression of AD pathophysiology. By examining both HS volumes and A β binding in the same participants, we can better define their relationships to determine the differential effect of A β accumulation on individual HS volumes.

Materials and Methods

74 community dwelling volunteers (mean age 67.9 years, 52 males) enrolled in studies at the Knight Alzheimer Disease Research Center at Washington University were assessed with the clinical dementia rating (CDR) scale. All participants were cognitively normal (CDR=0). All procedures were approved by Washington University's Human Research Protection Office. Written consent was obtained from each participant and their informant.

Volumetric MRI was obtained on a Siemens (Erlangen, Germany) TIM Trio 3T MRI scanner with 12-channel head coil. Voxel size=1×1×1mm was obtained sagittally. All MRI sessions were processed through the FreeSurfer 5.1 image analysis suite, which is documented as follows: (<http://surfer.nmr.mgh.harvard.edu/>). The technical details of these procedures are described in prior publications [28–39]. HS segmentation used the FreeSurfer pipeline, yielding 10 segments for both left and right hippocampus: CA1, CA2/3, CA4/DG, subiculum, presubiculum, hippocampal tail, fimbria, inferior lateral ventricle, choroid plexus and hippocampal fissure [27]. Certified raters examined all segmentations and subsegmentations for problems and outliers. All scans that passed segmentation examination had no issues with subsegmentations. Additional information surrounding the FreeSurfer quality control process may be found at: (<http://surfer.nmr.mgh.harvard.edu/fswiki/FsTutorial/TroubleshootingData>).

A β PET imaging was conducted following bolus injection of ≈ 12 mCi of ^{11}C PIB and 60 minute dynamic PET scan in 3D mode (septa retracted). Mean cortical binding potential

(MCBP) was calculated for each scan using regions obtained by sampling the precuneus, gyrus rectus, prefrontal, and lateral temporal cortex as previously described [16]. Values were dichotomized to MCBP ≥ 0.16 (n=18, “PIB positive” (PIB+)) or <0.16 (n=56, “PIB negative” (PIB-)), and results for subvolumes were compared between these two groups. Because we were interested in identifying the earliest changes in amyloid to compare with hippocampal subvolumes, we chose a level of 0.16, slightly lower than has been used in other dichotomizations [16].

Mean and standard deviation for age, education, gender, intracranial volume (ICV) and mini-mental state examination (MMSE) for PIB+ and PIB- individuals were determined. Total hippocampal gray matter volume was determined by totaling the volumes of hippocampal tail, presubiculum, subiculum, CA1, CA2/3, and CA4/DG. Unpaired t-tests compared demographic variables and hippocampal subfields (HS) and total volumes. A Pearson correlation was used to correlate MCBP and subfield volumes. A Benjamini-Hochberg procedure was used to control for the false discovery rate.

Results

Demographic variables did not differ in age, gender or intracranial volume (ICV) between PIB+ and PIB- participants, but PIB+ mean MMSE was significantly lower ($p = 0.03$) and PIB+ education was significantly higher ($p = 0.04$) (Table 1). Compared with the PIB- group (mean MCBP=0.02), the PIB+ group (mean MCBP=0.49) had significantly smaller hippocampal tail, presubiculum, subiculum, and total hippocampal gray matter volumes after Benjamini-Hochberg correction, whereas the groups did not differ in CA1, CA2/3, or CA4/dentate gyrus (Table 1, Figure 1). Examining MCBP as a continuous variable across all subjects it was correlated with all of the HS volumes. However, examining PIB+ and PIB- groups separately, there was no correlation of MCBP with hippocampal subfield volumes in PIB- subjects (all $p > 0.5$) whereas in PIB+ subjects MCBP and all HS volumes remained inversely correlated although not significant under the 95% confidence level due to small sample size (n= 18): (hippocampal tail -0.34 , $p = 0.17$; presubiculum -0.33 , $p = 0.19$; CA-1 -0.27 , $p = 0.27$; CA2-3 -0.43 , $p = 0.07$; subiculum -0.43 , $p = 0.07$; CA4 (DG) -0.45 , $p = 0.09$; total hippocampal gray matter -0.43 , $p = 0.07$).

Discussion

We found that hippocampal tail, presubiculum, subiculum, and total hippocampal gray matter volumes normalized for age, gender, ICV, and education were significantly lower in cognitively normal PIB+ compared with cognitively normal PIB- older adults. This provides evidence that in the early stages of the preclinical AD within the hippocampus there is evidence of preferential involvement of certain subregions over others. Interestingly, we show that even in the preclinical stage of Alzheimer’s disease with normal MMSE scores, those subjects who are PIB+ differed both in hippocampal subfield volumes and in MMSE scores. While the mean MMSE in PIB negative of 29.18 and mean MMSE of 28.11 in PIB+ are both normal, they do differ significantly ($p = 0.03$), likely resulting from the early stages of hippocampal atrophy. This is supported by the lack of correlation between

MCBP and hippocampal subfield volumes in PIB– subjects and the inverse correlation of MCBP with hippocampal subfield volumes in PIB+ subjects.

Although to our knowledge this is the first documented investigation of associations between HS atrophy and A β accumulation in humans, the topic has previously been studied in mice. Simon et al. found signs of neurodegeneration in the CA1 and entorhinal cortex in amyloid precursor protein transgenic mice, while Beauquis et al. discovered that these mice had reduced hippocampal volume with fewer pyramidal and granular neurons at 5 months of age [40,41]. Studies have also found degeneration of specific types of neurons in transgenic mice, including GABAergic septo-hippocampal neurons and calretinin interneurons [42,43].

Studies of humans have observed whole hippocampal volume loss in individuals in the preclinical, mild cognitive impairment (MCI), and AD dementia stages of AD [23,44,45]. Smaller hippocampal volume is also associated with conversion from asymptomatic and MCI stages to AD dementia [46–49]. Moreover, hippocampal atrophy has been associated with A β accumulation, as detected using PIB: asymptomatic PIB+ individuals have increased rates of hippocampal atrophy, and correlations between inferior temporal region PIB retention and hippocampal volume have been found [21,22]. Subfields of the hippocampus, however, appear to be preferentially vulnerable to AD, as their volumes distinguish individuals with MCI from controls more accurately than global hippocampal volumes [26].

Previous studies of HS atrophy across stages of AD have discovered varied results. Studies of preclinical AD have found no changes in neuronal number or volume [50,51]. Two studies of MCI found greater subicular atrophy, though studies have also found CA1 and CA2/3 atrophy [26,52,53]. In AD, various studies have found atrophy predominantly in the CA1 and subiculum [50,53–56], but also in the entorhinal cortex [55]. Each of these studies utilized different methods to determine subfield volumes. Some relied on manual HS delineation [26,54–56] – an accurate but severely time consuming method – while others used a faster automated method [52]. Our study utilized a fully automated method of HS segmentation developed by Van Leemput *et al.*, which primarily uses geometrical rules to delineate each HS [27]. Van Leemput *et al.* validated their technique against manual hippocampal segmentations of 10 individuals and found that CA1, CA2/3, CA4/DG, presubiculum, and subiculum all had reasonable Dice overlap coefficients (>0.62) (Dice overlap coefficients measure the similarity of two samples by comparing the number of species shared by two samples to the number of species in each unique sample). Pearson's correlations, however, were only significant for CA2/3 and CA4/DG, though subiculum also correlated to some degree.

Limitations of the study

An intrinsic limitation of using the Van Leemput methods is that the internal boundaries of the hippocampus are not visible in 1mm T1 data. Further, the DICE overlap data comes from much higher resolution T1 data ($0.38\times0.38\times0.8\text{mm}^3$) than applied to in this study. Thus, the method was not validated on more standard T1 imaging protocols. Large parts of CA1 are actually labeled as CA23 by Van Leemput's software. Comparisons of automated segmentations with manual delineations show that this method underestimates the volume of

CA1; CA1, defined histologically, would include most if not all of the subiculum as defined by Van Leemput *et al.* [57]. Thus, further studies with a more anatomically accurate segmentation tool may be required. An automated method recently developed by Yushkevich *et al.* using MRI data acquired with a shorter scan time generally produces better DICE overlap coefficients [58]. Instead of using geometrical rules, this method uses multi-atlas segmentation to propagate subfield labels from a set of manually segmented training MRI scans to the target MRI scan. The manual segmentations of the training data were derived from comparison to labeled postmortem data in published atlases. The method also requires the use of a high resolution T2 scan.

An alternative method, delineating hippocampal shape, may also be closely associated with A β accumulation in cognitively normal individuals. Previous studies using hippocampal shape analysis have found differences in hippocampal deformation between individuals with symptomatic AD and asymptomatic individuals, especially in the CA1 and subiculum [49,59]. Moreover, automated hippocampal shape determination can be used to discriminate between individuals with AD dementia, individuals with MCI, and asymptomatic individuals with more accuracy than hippocampal volumetry [60,61]. Most importantly, variation of hippocampal shape – determined both manually and automatically – is predictive for AD in asymptomatic individuals [62,63]. These studies highlight the potential usefulness of hippocampal shape as a measure of early AD pathology.

In summary, we have shown that some hippocampal subfields (and not others) significantly differ in volume between cognitively normal PIB+ and PIB– participants. Moreover, given the uncertainty of the exact HS boundaries as measured using the automated process described, our study also highlights the importance of current efforts to develop a standardized protocol for efficiently and accurately determining subfield volumes.

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References

- [1]. Hardy J, Selkoe DJ. The amyloid hypothesis of Alzheimer's disease: progress and problems on the road to therapeutics. *Science*. 2002; 297:353–356. [PubMed: 12130773]
- [2]. Gravina SA, Ho L, Eckman CB, Long KE, Otvos L, Younkin LH, Suzuki N, Younkin SG. Amyloid β Protein (A β) in Alzheimer's Disease Brain. *Nature*. 1995; 270:7013–7016.
- [3]. Yates, P a; Sirisriro, R.; Villemagne, VL.; Farquharson, S.; Masters, CL.; Rowe, CC. Cerebral microhemorrhage and brain β -amyloid in aging and Alzheimer disease. *Neurology*. 2011; 77:48–54. [PubMed: 21700585]
- [4]. Jack CR, Knopman DS, Jagust WJ, Shaw LM, Aisen PS, Weiner W, Petersen RC, Trojanowski JQ. Hypothetical model of dynamic biomarkers of the Alzheimer's pathological cascade. *Lancet Neurol*. 2010; 9:1–20. [PubMed: 20083020]
- [5]. Jack CR, Knopman DS, Jagust WJ, Peterson RC, Weiner MW, Aisen PS, Shaw LM, Vemuri P, Wiste HJ, Weigand SD, Lesnick TG, Pankratz VS, Donohue MC, Trojanowski JQ. Update on

- hypothetical model of Alzheimer's disease biomarkers. *Lancet Neurol.* 2013; 12:207–216. [PubMed: 23332364]
- [6]. Perrin RJ, Fagan AM, Holtzman DM. Multimodal techniques for diagnosis and prognosis of Alzheimer's disease. *Nature.* 2009; 461:916–922. [PubMed: 19829371]
- [7]. Holmes C, Boche D, Wilkinson D. Long-term effects of A β 42 immunisation in Alzheimer's disease: follow-up of a randomised, placebo-controlled phase I trial. *Lancet.* 2008; 372:11–14. [PubMed: 18603144]
- [8]. Sperling, P a; Aisen, PS.; Beckett, P a; Bennett, P a; Craft, S.; Fagan, AM.; Iwatsubo, T.; Jack, CR.; Kaye, J.; Montine, TJ.; Park, DC.; Reiman, EM.; Rowe, CC.; Siemers, E.; Stern, Y.; Yaffe, K.; Carrillo, MC.; Thies, B.; Morrison-Bogorad, M.; Wagster, MV.; Phelps, CH. 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.* 2011; 7:280–292. [PubMed: 21514248]
- [9]. Katzman R, Terry R, Deteresa R, Brown T, Davies P, Fuld P, Renbing X, Peck A. Neurochemical Changes in Dementia: A Subgroup with Preserved Mental Status and Numerous Neocortical Plaques. *Ann. Neurol.* 1988; 23:138–144. [PubMed: 2897823]
- [10]. Price JL, Morris JC. Tangles and plaques in nondemented aging and "preclinical" Alzheimer's disease. *Ann. Neurol.* 1999; 45:358–368. [PubMed: 10072051]
- [11]. 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; 38:1682–1687. [PubMed: 3185902]
- [12]. Klunk WE, Engler H, Nordberg A, Wang Y, Blomqvist G, Holt DP, Bergstro M, Savitcheva I, Debnath ML, Barletta J, Price JC, Sandell J, Lopresti BJ, Wall A, Koivisto P, Antoni G, Mathis CA, Långstro B. Imaging Brain Amyloid in Alzheimer's Disease with Pittsburgh Compound-B. *Ann. Neurol.* 2004; 55:306–319. [PubMed: 14991808]
- [13]. Grimmer T, Henriksen G, Wester H-J, Förstl H, Klunk WE, Mathis P a, Kurz A, Drzezga A. Clinical severity of Alzheimer's disease is associated with PIB uptake in PET. *Neurobiol. Aging.* 2009; 30:1902–1909. [PubMed: 18346821]
- [14]. Jack CR, Lowe VJ, Senjem ML, Weigand SD, Kemp BJ, Shiung MM, Knopman DS, Boeve BF, Klunk WE, Mathis P a, Petersen RC. 11C PiB and structural MRI provide complementary information in imaging of Alzheimer's disease and amnesic mild cognitive impairment. *Brain.* 2008; 131:665–680. [PubMed: 18263627]
- [15]. Morris JC, Roe CM, Grant P a, Head D, Storandt M, Goate AM, Fagan AM, Holtzman DM, Mintun P a. Pittsburgh compound B imaging and prediction of progression from cognitive normality to symptomatic Alzheimer disease. *Arch. Neurol.* 2009; 66:1469–1475. [PubMed: 20008650]
- [16]. Mintun, P a; Larossa, GN.; Sheline, YI.; Dence, CS.; Lee, SY.; Mach, RH.; Klunk, WE.; Mathis, P a; DeKosky, ST.; Morris, JC. [11C]PiB in a nondemented population: potential antecedent marker of Alzheimer disease. *Neurology.* 2006; 67:446–452. [PubMed: 16894106]
- [17]. Killiany RJ, Moss MB, Albert MS, Sandor T, Tieman J, Jolesz F. Temporal Lobe Regions on Magnetic Resonance Imaging Identify Patients With Early Alzheimer's Disease. *Arch. Neurol.* 1993; 50:949–954. [PubMed: 8363449]
- [18]. Braak H, Braak E. Neuropathological staging of Alzheimer-related changes. *Acta Neuropathol.* 1991; 82:239–259. [PubMed: 1759558]
- [19]. Hyman B, Van Hoesen G, Damasio A, Barnes C. Alzheimer's disease: cell-specific pathology isolates the hippocampal formation. *Science* (80-.). 1984; 225:1168–1170.
- [20]. Ball M, Hachinski V, Fox A, Kirshen A. A new definition of Alzheimer's disease: a hippocampal dementia. *Lancet.* 1985; 325:14–16. [PubMed: 2856948]
- [21]. Bourgeat P, Chételat G, Villemagne VL, Frapp J, Raniga P, Pike K, Acosta O, Szoeke C, Ourselin S, Ames D, Ellis P a, Martins RN, Masters CL, Rowe CC, Salvado O. Beta-amyloid burden in the temporal neocortex is related to hippocampal atrophy in elderly subjects without dementia. *Neurology.* 2010; 74:121–127. [PubMed: 20065247]
- [22]. Andrews K, Modat M, Macdonald K. Atrophy rates in asymptomatic amyloidosis: implications for Alzheimer prevention trials. *PLoS One.* 2013; 8:e58816. [PubMed: 23554933]

- [23]. Frisoni G, Fox N, Jack C, Scheltens P, Thompson P. The clinical use of structural MRI in Alzheimer disease. *Nat. Rev. Neurol.* 2010; 6:67–77. [PubMed: 20139996]
- [24]. Lindberg O, Walterfang M, Looi JCL, Malykhin N, Ostberg P, Zandbelt B, Styner M, Paniagua B, Velakoulis D, Omdahl E, Wahlund L-O. Hippocampal shape analysis in Alzheimer's disease and frontotemporal lobar degeneration subtypes. *J. Alzheimer's Dis.* 2012; 30:355–365. [PubMed: 22414571]
- [25]. Tondelli M, Wilcock GK, Nichelli P, De Jager P a, Jenkinson M, Zamboni G. Structural MRI changes detectable up to ten years before clinical Alzheimer's disease. *Neurobiol. Aging.* 2012; 33:825.e25–825.e36. [PubMed: 21782287]
- [26]. La Joie R, Perrotin A, de La Sayette V, Egret S, Doeuve L, Belliard S, Eustache F, Desgranges B, Chételat G. Hippocampal subfield volumetry in mild cognitive impairment, Alzheimer's disease and semantic dementia. *NeuroImage. Clin.* 2013; 3:155–162. [PubMed: 24179859]
- [27]. Van Leemput K, Bakkour A, Benner T. Automated segmentation of hippocampal subfields from ultra-high resolution in vivo MRI. *Hippocampus.* 2009; 19:549–557. [PubMed: 19405131]
- [28]. Dale AM, Fischl B, Sereno MI. Cortical Surface-Based Analysis. I: Segmentation and surface reconstruction. *Neuroimage.* 1999; 9:179–194. [PubMed: 9931268]
- [29]. Dale A, Sereno M. Improved Localization of Cortical Activity By Combining EEG and MEG with MRI Cortical Surface Reconstruction. *J. Cogn. Neurosci.* 1993; 5:162–176. [PubMed: 23972151]
- [30]. Fischl B, Dale a M. Measuring the thickness of the human cerebral cortex from magnetic resonance images. *Proc. Natl. Acad. Sci. U. S. A.* 2000; 97:11050–11055. [PubMed: 10984517]
- [31]. Fischl B, Liu a, Dale a M. Automated manifold surgery: constructing geometrically accurate and topologically correct models of the human cerebral cortex. *IEEE Trans. Med. Imaging.* 2001; 20:70–80. [PubMed: 11293693]
- [32]. Fischl B, Salat DH, Busa E, Albert M, Dieterich M, Haselgrove C, van der P a, Killiany R, Kennedy D, Klaveness S, Montillo A, Makris N, Rosen B, Dale AM. Whole brain segmentation: automated labeling of neuroanatomical structures in the human brain. *Neuron.* 2002; 33:341–355. [PubMed: 11832223]
- [33]. Fischl B. Automatically Parcellating the Human Cerebral Cortex. *Cereb. Cortex.* 2004; 14:11–22. [PubMed: 14654453]
- [34]. Fischl B, Sereno MI, Dale AM. Cortical Surface-Based Analysis. II: Inflation, flattening, and a surface-based coordinate system. *J. Cogn. Neurosci.* 1999; 9:195–207.
- [35]. Fischl B, Sereno MI, Tootell RB, Dale a M. High-resolution intersubject averaging and a coordinate system for the cortical surface. *Hum. Brain Mapp.* 1999; 8:272–284. [PubMed: 10619420]
- [36]. Fischl B, Salat DH, van der Kouwe AJW, Makris N, Ségonne F, Quinn BT, Dale AM. Sequence-independent segmentation of magnetic resonance images. *Neuroimage.* 2004; 23(Suppl 1):S69–84. [PubMed: 15501102]
- [37]. Han X, Jovicich J, Salat D, van der P a, Quinn B, Czanner S, Busa E, Pacheco J, Albert M, Killiany R, Maguire P, Rosas D, Makris N, Dale A, Dickerson B, Fischl B. Reliability of MRI-derived measurements of human cerebral cortical thickness: the effects of field strength, scanner upgrade and manufacturer. *Neuroimage.* 2006; 32:180–194. [PubMed: 16651008]
- [38]. Jovicich J, Czanner S, Greve D, Haley E, van der P a, Gollub R, Kennedy D, Schmitt F, Brown G, Macfall J, Fischl B, Dale A. Reliability in multi-site structural MRI studies: effects of gradient non-linearity correction on phantom and human data. *Neuroimage.* 2006; 30:436–443. [PubMed: 16300968]
- [39]. Ségonne F, Dale a M, Busa E, Glessner M, Salat D, Hahn HK, Fischl B. A hybrid approach to the skull stripping problem in MRI. *Neuroimage.* 2004; 22:1060–1075. [PubMed: 15219578]
- [40]. Simón A-M, Schiapparelli L, Salazar-Colocho P, Cuadrado-Tejedor M, Escribano L, López de Maturana R, Del Rfo J, Pérez-Mediavilla A, Frechilla D. Overexpression of wild-type human APP in mice causes cognitive deficits and pathological features unrelated to Abeta levels. *Neurobiol. Dis.* 2009; 33:369–378. [PubMed: 19101630]

- [41]. Beauquis J, Vinuesa A, Pavia P, Galvan V, Saravia F. Neuronal and glial alterations, increased anxiety, and cognitive impairment before hippocampal amyloid deposition in PDAPP mice, model of Alzheimer's disease. *Hippocampus*. 2014; 24:257–269. [PubMed: 24132937]
- [42]. Loreth D, Ozmen L, Revel FG, Knoflach F, Wetzel P, Frotscher M, Metzger F, Kretz O. Selective degeneration of septal and hippocampal GABAergic neurons in a mouse model of amyloidosis and tauopathy. *Neurobiol. Dis.* 2012; 47:1–12. [PubMed: 22426397]
- [43]. Baglietto-Vargas D, Moreno-Gonzalez I, Sanchez-Varo R, Jimenez S, Trujillo-Estrada L, Sanchez-Mejias E, Torres M, Romero-Acebal M, Ruano D, Vizuet M, Vitorica J, Gutierrez A. Calretinin interneurons are early targets of extracellular amyloid-beta pathology in PS1/AbetaPP Alzheimer mice hippocampus. *J. Alzheimers. Dis.* 2010; 21:119–132. [PubMed: 20413859]
- [44]. Scahill RI, Schott JM, Stevens JM, Rossor MN, Fox NC. Mapping the evolution of regional atrophy in Alzheimer's disease: unbiased analysis of fluid-registered serial MRI. *Proc. Natl. Acad. Sci. U. S. A.* 2002; 99:4703–4707. [PubMed: 11930016]
- [45]. Fox N, Warrington E. Presymptomatic hippocampal atrophy in Alzheimer's disease A longitudinal MRI study. *Brain*. 1996; 119:2001–2007. [PubMed: 9010004]
- [46]. Apostolova LG, Dutton P a, Dinov ID, Hayashi KM, Toga AW, Cummings JL, Thompson PM. Conversion of mild cognitive impairment to Alzheimer disease predicted by hippocampal atrophy maps. *Arch. Neurol.* 2006; 63:693–699. [PubMed: 16682538]
- [47]. Den Heijer T, Geerlings MI, Hoebeek FE, Hofman A, Koudstaal PJ, Breteler MMB. Use of hippocampal and amygdalar volumes on magnetic resonance imaging to predict dementia in cognitively intact elderly people. *Arch. Gen. Psychiatry.* 2006; 63:57–62. [PubMed: 16389197]
- [48]. Jack CR, Petersen RC, Xu YC, O'Brien PC, Smith GE, Ivnik RJ, Boeve BF, Waring SC, Tangalos EG, Kokmen E. Prediction of AD with MRI-based hippocampal volume in mild cognitive impairment. *Neurology*. 1999; 52:1397–1397. [PubMed: 10227624]
- [49]. Scher, a I.; Xu, Y.; Korf, ESC.; White, LR.; Scheltens, P.; Toga, a W.; Thompson, PM.; Hartley, SW.; Witter, MP.; Valentino, DJ.; Launer, LJ. Hippocampal shape analysis in Alzheimer's disease: a population-based study. *Neuroimage*. 2007; 36:8–18. [PubMed: 17434756]
- [50]. West MJ, Kawas CH, Stewart WF, Rudow GL, Troncoso JC. Hippocampal neurons in pre-clinical Alzheimer's disease. *Neurobiol. Aging*. 2004; 25:1205–1212. [PubMed: 15312966]
- [51]. Price JL, Ko a I, Wade MJ, Tsou SK, McKeel DW, Morris JC. Neuron number in the entorhinal cortex and CA1 in preclinical Alzheimer disease. *Arch. Neurol.* 2001; 58:1395–1402. [PubMed: 11559310]
- [52]. Hanseeuw BJ, Van Leemput K, Kavec M, Grandin C, Seron X, Ivanoiu a. Mild cognitive impairment: differential atrophy in the hippocampal subfields. *AJNR. Am. J. Neuroradiol.* 2011; 32:1658–61. [PubMed: 21835940]
- [53]. Rössler M, Zarski R, Bohl J, Ohm TG. Stage-dependent and sector-specific neuronal loss in hippocampus during Alzheimer's disease. *Acta Neuropathol.* 2002; 103:363–369. [PubMed: 11904756]
- [54]. Frisoni GB, Ganzola R, Canu E, Rüb U, Pizzini FB, Alessandrini F, Zoccatelli G, Beltramello A, Caltagirone C, Thompson PM. Mapping local hippocampal changes in Alzheimer's disease and normal ageing with MRI at 3 Tesla. *Brain*. 2008; 131:3266–3276. [PubMed: 18988639]
- [55]. Mueller SG, Schuff N, Yaffe K, Madison C, Miller B, Weiner MW. Hippocampal atrophy patterns in mild cognitive impairment and Alzheimer's disease. *Hum. Brain Mapp.* 2010; 31:1339–1347. [PubMed: 20839293]
- [56]. Mueller SG, Weiner MW. Selective effect of age, Apo e4, and Alzheimer's disease on hippocampal subfields. *Hippocampus*. 2009; 19:558–564. [PubMed: 19405132]
- [57]. Amaral, DG. Hippocampal Formation. In: Paxinos, G., editor. *The Human Nervous System*. Academic Press, Inc.; San Diego: 1990. p. 711-755.
- [58]. Yushkevich PA, Pluta J, Wang H, Xie L, Ding S, Gertje E, Mancus0 L, Kliot D, Das SR, Wolk D. Automated volumetry and regional thickness analysis of hippocampal subfields and medial temporal cortical structures in mild cognitive impairment. *Human Brain Mapping*. 2014 Epub ahead of print.

- [59]. Li S, Shi F, Pu F, Li X, Jiang T, Xie S, Wang Y. Hippocampal shape analysis of Alzheimer disease based on machine learning methods. *AJNR. Am. J. Neuroradiol.* 2007; 28:1339–1345. [PubMed: 17698538]
- [60]. Ferrarini L, Frisoni GB, Pievani M, Reiber JHC, Ganzola R, Milles J. Morphological hippocampal markers for automated detection of Alzheimer's disease and mild cognitive impairment converters in magnetic resonance images. *J. Alzheimers. Dis.* 2009; 17:643–659. [PubMed: 19433888]
- [61]. Gerardin E, Chételat G, Chupin M, Cuingnet R, Desgranges B, Kim H-S, Niethammer M, Dubois B, Lehericy S, Garnero L, Eustache F, Colliot O. Multidimensional classification of hippocampal shape features discriminates Alzheimer's disease and mild cognitive impairment from normal aging. *Neuroimage.* 2009; 47:1476–1486. [PubMed: 19463957]
- [62]. Achterberg HC, van der Lijn F, den Heijer T, Vernooij MW, Ikram MA, Niessen WJ, de Bruijne M. Hippocampal shape is predictive for the development of dementia in a normal, elderly population. *Hum. Brain Mapp.* 2014; 35:2359–2371. [PubMed: 24039001]
- [63]. Csernansky JG, Wang L, Swank J, Miller JP, Gado M, McKeel D, Miller MI, Morris JC. Preclinical detection of Alzheimer's disease: hippocampal shape and volume predict dementia onset in the elderly. *Neuroimage.* 2005; 25:783–792. [PubMed: 15808979]

Figure 1.

(A) Sagittal, (B) Coronal, and (C) Axial views of hippocampus, as segmented using the method of Van Leemput *et al.* Hippocampal tail, subiculum, and presubiculum volumes were significantly lower in PIB+ participants compared to PIB- participants. Total hippocampal gray matter volume was also significantly lower in PIB+ participants compared to PIB- participants.

Table 1

Differences in demographic characteristics and hippocampal subfield volumes between PIB– and PIB+ participants; Pearson's correlation coefficients of hippocampal subfield volumes with MCBP. MMSE and educational level differed between PIB– and PIB+. After Benjamini-Hochberg correction, hippocampal tail, presubiculum, subiculum, and total hippocampal gray matter volumes were significantly lower in PIB+ participants.

Demographic/ Region volume mean (SD)	Structural Comparison				
	PIB– (N=56)	PIB+ (N=18)	% decrease	<i>p</i>	<i>p</i> hochberg
Age	67.4 (5.7)	69.6 (7.4)	-	0.26	
Education	15.5 (2.6)	16.7 (1.8)	-	0.04	
Gender (F, M)	41, 15	11, 7	-	0.33	
ICV	1377034.23 (207416.5)	1408313.06 (171999.5)	-	0.53	
MMSE	29.18 (1.19)	28.11 (1.78)	-	0.03	
Hippocampal Tail	875.3 (112.8)	773.2 (115.4)	11.7	0.000	0.002
Presubiculum	1054.5 (175.7)	943.7 (152.4)	10.5	0.009	0.035
CA-1	764.4 (98.6)	729.8 (114.4)	4.5	0.051	0.103
CA2, 3	2264.7 (297.3)	2138.5 (348.2)	5.6	0.105	0.105
Subiculum	1472.1 (202.4)	1321.2 (209.0)	10.3	0.002	0.014
CA4 (DG)	1273.6 (164.2)	1188.2 (188.5)	6.7	0.046	0.103
Total Hippocampal Gray Matter	7704.7 (944.9)	7094.5 (1019.3)	7.9	0.006	0.032