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ORIGINAL ARTICLE

Gender Differences in Alzheimer Disease: Brain Atrophy, Histopathology Burden, and Cognition

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Abstract

Multiple studies suggest that females are affected by Alzheimer disease (AD) more severely and more frequently than males. Other studies have failed to confirm this and the issue remains controversial. Difficulties include differences in study methods and male versus female life expectancy. Another element of uncertainty is that the majority of studies have lacked neuropathological confirmation of the AD diagnosis. We compared clinical and pathological AD severity in 1028 deceased subjects with full neuropathological examinations. The age of dementia onset did not differ by gender but females were more likely to proceed to very severe clinical and pathological disease, with significantly higher proportions having a Mini-Mental State Examination score of 5 or less and Braak stage VI neurofibrillary degeneration. Median neuritic plaque densities were similar in females and males with AD but females had significantly greater tangle density scores. In addition, we found that AD-control brain weight differences were significantly greater for females, even after adjustment for age, disease duration, and comorbid conditions. These findings suggest that when they are affected by AD, females progress more often to severe cognitive dysfunction, due to more severe neurofibrillary degeneration, and greater loss of brain parenchyma.

Key Words: Amyloid plaque; Brain weight; Cognition; Neuritic plaque; Neurofibrillary tangle; Phosphorylated tau.

INTRODUCTION

For generations, clinical and basic science research often neglected to analyze for male–female differences, and often have failed to include females at all. In response to this, the United States Congress approved the Revitalization Act in the early 1990s, requiring the inclusion of women in National Institutes of Health-funded clinical research (1). The ideal today in clinical research is to have equal distributions of males and females, and many studies are trying to understand how health conditions and medications might differentially affect females and males.

Alzheimer disease (AD) is the most common cause of dementia in the elderly. Many studies have reported that females have a higher risk of developing AD (2–6), and that they are more severely affected (2, 6, 7). Yet, this concept is mired in controversy due to conflicting published results (8–13). Differences in experimental design and study populations may be responsible for some of the discrepant results, while, additionally, most studies have lacked pathological confirmation of the diagnosis of AD, thereby creating additional uncertainty. In this study, we used clinical and autopsy data to explore differences between male and females in terms of cognition, AD-specific histopathology and its impact on the neural parenchyma.

MATERIALS AND METHODS

Subjects were all volunteers in the Arizona Study of Aging and Neurodegenerative Disorders (AZSAND), a longitudinal clinicopathological study of normal aging, cognition, and movement in the elderly since 1996 in Sun City, Arizona. Autopsies are performed by the Banner Sun Health Research

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TABLE 1. General Characteristics of Study Subjects

Diagnosis/Gender (N)	Age at Death (SD)	MMSE (SD)	Cognitive Sx, Age Onset (SD)	Cognitive Sx, Years Duration (SD)	Education Years (SD)
ND-Female (126)	85.8 (9.3)#	29.0 (1.0)#	NA	NA	15.0 (2.4)#
ND-Male (166)	83.1 (9.2)	27.8 (2.0)	NA	NA	14.8 (2.9)
AD-Female (345)	83.5 (9.6)	12.2 (9.1)	74.7 (10.6)	8.8 (5.0)	14.0 (2.5)
AD-Male (391)	81.3 (7.6)*	13.4 (8.4)	73.7 (8.9)	7.7 (4.1)*	15.2 (2.8) *

MMSE, Mini-Mental State Examination; Sx, symptoms; NA, not applicable; ND, nondemented controls; AD, Alzheimer disease.
*$p < 0.01$ for gender comparisons within AD subjects.
#$p < 0.05$ for comparison of all groups.

Institute Brain and Body Donation Program (BBDP; www.brainandbodydonation program.org). Subsets of the Program are funded by the US National Institute on Aging Arizona Alzheimer's Disease Core Center and the US National Institute of Neurological Disorders and Stroke National Brain and Tissue Resource for Parkinson's Disease and Related Disorders. Cognitively normal volunteer subjects are all recruited from the communities of greater Phoenix, Arizona through public relations activities. Many AZSAND subjects with cognitive impairment, dementia, or movement disorders are also community-derived while some are enrolled through neurologists' practices or dementia clinics. All subjects sign Institutional Review Board-approved informed consents allowing both clinical assessments during life and several options for brain and/or body organ donation after death. Most subjects are clinically characterized with annual standardized test batteries consisting of general neurological, cognitive, and movement disorders components, including the Mini-Mental State Examination (MMSE). Subjects for the current study were chosen by searching the BBDP database for cases with a clinicopathological diagnosis of AD (n = 736) or control (n = 292) (total = 1028, Table 1). Control (ND) was defined clinically as those lacking dementia; these patients could have mild cognitive impairment or incidental pathology but did not meet clinical or neuropathological criteria for a defined neurodegenerative disease. Clinically defined measures used in this study included age at cognitive symptom onset and cognitive symptom duration, the latter defined as the number of years between cognitive symptom onset and death (14). The influence of comorbid (non-AD) brain disease was explored by defining a "multiple diagnoses" group, within which AD subjects had at least one other major neurodegenerative or cerebrovascular condition, or a movement disorder; these included subjects with clinicopathologically defined AD as well as Parkinson disease, vascular dementia, progressive supranuclear palsy, corticobasal degeneration, multiple system atrophy, dementia with Lewy bodies, hippocampal sclerosis, frontotemporal lobar degeneration with TDP-43 proteinopathy (FTLD-TDP), Pick disease, and Huntington disease.

The complete neuropathological examination was performed using standard AZSAND methods (15, 16). Brain weights were determined at autopsy, after removal of 10–30 cc of ventricular cerebrospinal fluid but prior to fixation. The gender-specific AD-control brain weight difference was expressed for each AD subject as the ratio (converted to a percentage) of that individual's brain weight as compared with the mean brain weight of all control subjects of the same gender. Neuropathological examinations were performed in a standardized manner and consisted of gross and microscopic observations, the latter including assessment of frontal, parietal, temporal, and occipital lobes, all major diencephalic nuclei and major subdivisions of the brainstem, cerebellum, and spinal cord (the latter only for those with whole-body autopsy). Following fresh slicing and subsequent fixation in cold 10% neutral-buffered formalin for 36–60 hours, histological preparations included large-format (3 × 5 cm), 40–80 μm-thick, cryoprotected frozen sections as well as paraffin-embedded 6-μm sections. Both sets were stained with hematoxylin and eosin; the former set was also stained for senile plaques, neurofibrillary changes, and other neuronal and glial tauopathies using thioflavin S, Gallyas and Campbell-Switzer methods (15, 17, 18). In all cases, an additional set of paraffin sections was immunohistochemically stained for phosphorylated α-synuclein (p-syn), while staining for phosphorylated TDP-43 (p-TDP43) was done only for subjects judged after initial neuropathological examination to be at risk for FTLD-TDP (19–23). Neuritic plaque and neurofibrillary tangle densities were graded blindly as recommended by The Consortium to Establish a Registry for Alzheimer's Disease (CERAD) with separate semiquantitative density estimates of none, sparse, moderate, or frequent (24). All scores were converted to a 0–3 scale for statistical purposes. Regions scored included cortical gray matter from frontal (F), temporal (T), parietal (P), hippocampal CA1 (H), and entorhinal (E) regions. Neurofibrillary degeneration was staged on the thick frozen sections by the original method of Braak (18, 25), and neuropathological AD diagnoses were made when neuritic plaque densities and Braak stage met "intermediate" or "high" criteria according to National Institute on Aging/Reagan Institute criteria (26–28). Non-AD conditions were diagnosed using standard clinicopathological criteria with international consensus criteria for those disorders where these were available.

Statistical Methods

Univariate analyses were used as an initial screen to indicate which variables might be significantly affected by gender and also have significant relationships with AD-control brain weight difference and/or MMSE score. For comparing group measures, the Mann–Whitney U-Test, 1-way analysis

TABLE 2. Pathological Characteristics of Study Subjects

Diagnosis-Gender (N)	Brain Weight (SD)	Within-Gender AD-Control Brain Weight Difference (%)	Plaque Density (SD)	Braak NF (SD)	Total Infarct Volume in mm^3 (SD)	% Cases Multiple Dx (N)	Postmortem Interval, in Hours (SD)
ND-Female (126)	1126.3 (98.0) #	9.0 (11.6)&	1.4 (1.2) #	3.0 (1.0) #	12.7 (66.7)	NA	3.7 (3.1) #
ND-Male (166)	1255.8 (111.1)	5.6 (11.0)	1.1 (1.1)	2.7 (1.2)	4.1 (23.3)	NA	4.7 (11.5)
AD-Female (345)	1024.2 (124.4)	9.0 (11.6)	2.8 (0.4)	5.1 (1.0)	11.6 (51.7)	28.7 (98)	4.7 (6.7)
AD-Male (391)	1188.6 (133.5)	5.6 (11.0)	2.7 (0.4)	4.7 (1.2)*	4.2 (25.4)	32.0 (125)	6.5 (10.7) *

Braak NF, Braak neurofibrillary tangle stage; Dx, diagnoses; NA, not applicable; ND, no dementia.
*$p < 0.05$ for gender comparisons within AD subjects.
#$p < 0.05$ for comparison of all groups; analysis of variance.
&$p < 0.001$ for % brain weight difference; contrast analysis.

of variance (ANOVA), Kruskal–Wallis ANOVA, and contrast analysis were used as appropriate. The chi-squared test was used to compare proportions and Spearman's method was used to test univariate correlations. Variables that were significantly affected on this initial screen by gender, or that correlated significantly with AD-control brain weight difference and/or MMSE score, were included in multivariable logistic regression models.

RESULTS

Of deceased AZSAND subjects, 28% of women versus 30% of men had a final clinicopathological diagnosis of AD (not significantly different [ns]). The group age means differed significantly with the youngest group (males with AD) having a mean age of 81.3 years, whereas for the oldest group (control females), the mean age was 85.8 years ($p < 0.05$) (Table 1). Both AD and control female groups were significantly older than their respective male groups ($p < 0.01$); however, in both genders the age difference between control and AD subjects was of the same magnitude (ns). The mean age of onset of cognitive symptoms was 74.7 for females and 73.7 for males (ns); females with AD survived significantly longer after symptom onset (8.8 vs 7.7 years; $p < 0.01$).

The CERAD neuritic plaque density score showed a significant gender-related difference only within the control group, with significantly greater densities in females (Table 2; $p < 0.05$); this difference was significant even after adjustment for age and cognitive symptom duration. As compared with males with AD, females with AD had higher Braak stage ($p < 0.05$) even after adjustment for age and cognitive symptom duration. Furthermore, females as a group were significantly more likely to reach the highest Braak neurofibrillary stage (Fig. 1, $p < 0.001$) and significantly less likely to be in the lowest Braak stage (Fig. 1, $p < 0.05$). These differences also persist after adjusting for age and cognitive symptom duration ($p < 0.05$).

As expected, females had significantly lower brain weights, and AD brain weights were significantly lower than those of controls (Fig. 2, Table 2; $p < 0.05$). A contrast analysis indicated that the AD-control brain weight difference was significantly greater for females ($p < 0.001$). Spearman's correlations indicated that some variables, including years of education, cognitive symptom age of onset, and total infarct

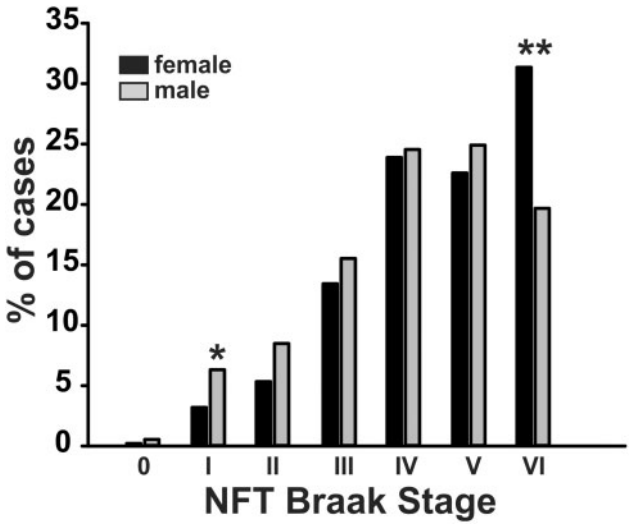

FIGURE 1. Distribution of neurofibrillary degeneration severity by Braak stage. A higher proportion of females (black) were classified as having the highest (stage VI) Braak neurofibrillary stage (**$p < 0.001$). A higher proportion of males (gray) were classified as having the lowest (stage I) nonzero stage (*$p < 0.05$).

volume, did not correlate significantly with AD-control brain weight difference (Table 3). The 7 variables that correlated significantly with AD-control brain weight difference, including gender, age at death, years since cognitive symptom onset, postmortem interval, multiple diagnoses, neuritic plaque density, and Braak stage (Table 3), were progressively added to logistic regression models, with all models including up to 6 variables (when the 6-variable equation omitted Braak stage) achieving significance (Table 4).

As expected, both females and males with AD had significantly lower MMSE scores than controls (Table 1; $p < 0.05$). The male–female AD difference between mean MMSE scores approached the significance level (13.4 vs 12.2, respectively; $p = 0.07$) and a higher proportion of females had very severe cognitive impairment as defined by MMSE scores of 5 or less (Fig. 3; $p < 0.05$). MMSE scores did not correlate with education years, neuritic plaque density, or multiple diag-

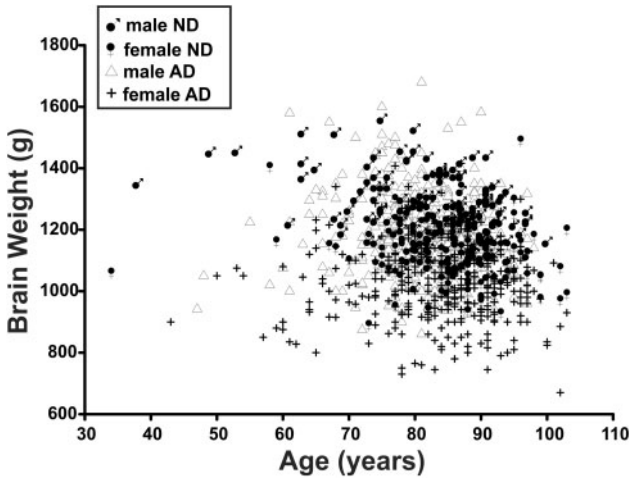

FIGURE 2. Brain weights in males and females, subdivided by having Alzheimer disease (AD) or no dementia, by age. The differences between AD and control brain weights, within females and males, are maintained in a fairly uniform way across different ages. There is no suggestion of an increasing gap between AD and controls with increasing age, as might be expected if there were an AD-aging synergy acting upon brain atrophy.

TABLE 3. Screening of Factors for Effects on Alzheimer Disease-Control Brain Weight Loss

Variable	Spearman Rho (ρ)	p Value
Gender	0.153	0.0000316
Age	0.0995	0.00692
Cognitive symptoms, years duration	0.272	0.00001
Braak stage	0.280	0.00001
Neuritic plaque density	0.113	0.002
PMI	−0.211	0.00000008
Multiple diagnoses	−0.113	0.002
Total infarct volume	−0.005	0.894
Education	−0.0549	0.223
Cognitive symptoms, age onset	−0.1	0.767

AD, Alzheimer disease; ND, nondemented; PMI, postmortem interval.

noses, while 6 variables, including gender, age, age of cognitive symptom onset, cognitive symptom duration, brain weight, and Braak stage, all correlated significantly with MMSE (Table 5; $p < 0.05$). These 6 variables were progressively added to multiple logistic regression models, with all models including up to 5 variables (when the 5-variable equation omitted Braak stage) achieving significance (Table 6).

DISCUSSION

Over the last decade it has become almost dogma that females have a greater risk of developing AD and are more affected by the disease than males, but upon comprehensive review, it is evident that this view is not without reasonable doubt. Several studies suggest that females are at higher risk of developing AD (29–31), whereas other analyses, such as that done in the Rotterdam study, propose that only the very oldest (>90 years) females have a higher risk (32). Furthermore, Kukull et al (8), Rocca et al (9), and Knopman et al (33) did not find any incidence differences between males and females. It is well accepted that females have a longer life expectancy than males; therefore, some results finding higher AD severity, prevalence, and incidence rates in females might be due only to a higher prevalence of very old females in the population. Additionally, analysis based only on the oldest of subjects may be idiosyncratic and/or limited by small sample size.

The AZSAND is not a random sample of the local or US population and, therefore, results may not be readily generalizable. All autopsy studies (except those of court-ordered autopsies) are subject to volunteer bias (34–36). While the control subjects are all derived from people living independently in the communities of greater Phoenix, those with cognitive impairment and dementia are a more selected subset because they are often referred from neurologists' offices or dementia clinics. It is expected, however, that the increased diagnostic precision and specific histopathological lesion measurement obtained through neuropathological examination confer distinct advantages to autopsy studies such as this one (37–39).

It is well known that cognition declines with aging, and while some studies suggest that this decline is more noticeable in females (30, 40–44), this has not been replicated by all studies (45–47). Barnes et al (6, 7) reported no gender differences in the rate of cognitive decline but suggest that AD pathology is more likely to result in cognitive dysfunction in females than in males. Similarly, Salehi et al (48) found gender differences in AD-related pathology, but others reported no pathological differences between women and men (49–51).

Contradictory results between studies could be explained by differences in experimental design, experimental analysis, and study populations. Relatively few studies have looked at gender differences in AD pathology (6, 7, 48–51) and many that explored cognitive function and incidence rate lacked pathological confirmation of the diagnosis of AD (3–5, 8–10, 30, 43, 44, 52). The accuracy of the clinical diagnosis of AD is low enough, with sensitivity ranging from 71% to 87% and specificity ranging from 44% to 71% (37), to increase the likelihood of false negative findings in studies lacking neuropathological diagnosis confirmation.

This study supports previous published investigations that have suggested that females may be more severely affected by AD than males. In AZSAND subjects, age of cognitive symptom onset did not significantly differ by gender but females were more likely to proceed to very severe disease, both clinically and pathologically, with a significantly higher proportion of females progressing to MMSE scores of 5 or less and to the highest Braak stage of neurofibrillary degeneration (Figs. 1, 3). Neuritic plaque density also tended to be greater in females but the statistical significance of this was limited to control subjects. These results closely parallel those of Barnes et al (6) who also found a higher female tangle burden.

While plaques and tangles may be specific markers of AD pathology, the most proximal cause of cognitive dysfunc-

TABLE 4. Factors Affecting Alzheimer Disease-Control Brain Weight Difference, Multivariable Analysis

Variable	Complete Equation OR; 95% CI; p Value	Gender Values OR; 95% CI; p Value
Gender	1.3; 1.1–1.6; 0.01	0.6; 0.5–0.8; 0.001
Gender + Braak stage	0.2; 0.4–0.4; <0.001	0. 7; 0.5–0.9; 0.019
Gender+ Expired age + PMI + Neuritic plaques+ Cognitive symptoms, years duration + Multiple diagnoses	0.1; 0.01–0.07; 0.02	0.7; 0.6–1.0; 0.05
Gender+ Expired age + PMI + Neuritic plaques + Cognitive symptoms, years duration + Multiple diagnoses + Braak stage	0.02; 0.003–0.2; <0.001	0.8; 0.6–1.1; 0.198

CI, confidence interval; for gender, females had an assigned value; OR, odds ratio.

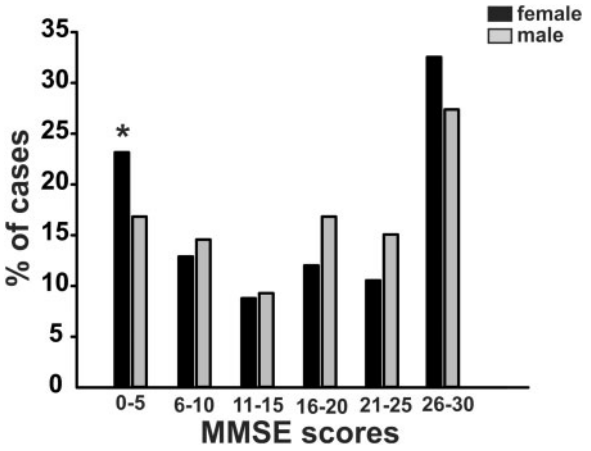

FIGURE 3. Mini-Mental State Examination (MMSE) scores distribution. A higher proportion of females (black) had very severe cognitive impairment as defined by MMSE scores of 5 or less (*$p < 0.05$). All other paired comparisons are not significantly different.

TABLE 5. Screening of Factors for Effects on Lower Mini-Mental State Examination Score

Variable	Spearman rho (ρ)	p Value
Gender	0.102	0.0172
% brain weight difference	0.200	0.000001
Age	0.178	0.0000287
Braak stage	−0.230	0.0000000564
Cognitive symptoms, years	−0.266	0.000000000308
Cognitive symptoms, age onset	−0.279	0.0000000000362
Neuritic plaque density	−0.0723	0.0910
Multiple diagnoses	0.0588	0.170
Education	0.0144	0.763

tion is likely to be loss of neural parenchyma. Loss of neural parenchyma is commonly expressed, at the gross anatomical level, by brain weight and volume. Numerous studies over many decades have documented how brain volume and weight loss starts as early as in middle age, and how this phenomenon seems to be amplified by neurological disorders such as AD (53–56). To our knowledge, this is the first study that has compared gender-specific AD-control brain weight differences. Even after adjusting for several potential confounders, including age, neuritic plaque densities, disease duration, and presence or absence of multiple diagnoses, the AD-control brain weight difference in AZSAND subjects was found to be significantly greater for women than men. Although both control groups are similarly older than their counterpart AD groups, both female groups are older, on average, than both male groups. Age-related brain atrophy could be synergistic with AD-related brain atrophy, resulting in an accelerated loss of brain weight with increasing age in patients with AD. However, in this study we used multiple regression equations to demonstrate that age did not independently contribute to AD-control brain weight differences when gender was a covariable. Gender at least partially influences the brain weight loss observed in AD and its influence seems to be even stronger than age, in particular with AD. Likewise, it is well known that many dementia subjects who meet criteria for a diagnosis of AD also meet criteria for a diagnosis of other diseases, such as Parkinson disease, dementia with Lewy bodies, vascular dementia, and progressive supranuclear palsy. Therefore, we postulated that this variable could also affect AD-control brain weight differences even though the percentage of females with more than 1 diagnosis did not differ from that in males. On our 2-factor correlation analysis, the correlation of multiple diagnoses with brain weight difference is significant but failed to reach significance on our multivariable analysis.

The loss of significance in our multivariable models including Braak stage indicates that a greater severity of neurofibrillary changes in women may be at least partially responsible for females' greater AD-control brain weight difference as well as greater MMSE score decline. Nevertheless, logistic regression equations including only gender and Braak stage demonstrated that gender itself may have a significant and independent influence on the observed brain weight difference. Over the past few decades, a variety of studies have suggested that estrogen protects against synaptic loss. One can speculate that the sharp loss of estrogen observed at menopause could be accelerating the synaptic loss observed with aging and AD, consequently leading to a higher brain weight loss. Taken together, these results suggest that females not only develop more AD-specific pathology, espe-

TABLE 6. Factors Affecting Mini-Mental State Examination Difference, Multivariable Analysis

Variable	Complete Equation OR; 95% CI; p Value	Gender Values OR; 95% CI; p Value
Gender	0.623; 0.465–0.834; 0.001	0.6; 0.5–0.8; 0.001
Gender +Braak	0.2; 0.4–0.4; <0.001	0.7; 0.5–0.9; 0.019
Gender + Age + Cognitive symptoms, years + Cognitive symptoms, age onset + % brain weight difference	0.07; 0.01–0.5; 0.007	1.5; 1.0–2.3; 0.05
Gender + Age + Cognitive symptoms, years + Cognitive symptoms, age onset + % brain weight difference+ Braak	0.2; 0.01–2.1; 0.1	1.5; 1.0–2.2; 0.07

CI, confidence interval; for gender, females had an assigned value; OR, odds ratio.

cially neurofibrillary tangles, but as a result they also suffer a greater AD-related loss of brain parenchyma and cognitive function.

REFERENCES

1. Clayton JA, Collins FS. Policy: NIH to balance sex in cell and animal studies. Nature 2014;509:282–3
2. Li R, Singh M. Sex differences in cognitive impairment and Alzheimer's disease. Front Neuroendocrinol 2014;35:385–403
3. Chene G, Beiser A, Au R, et al. Gender and incidence of dementia in the Framingham Heart Study from mid-adult life. Alzheimers Dement 2015; 11:310–20
4. Fillenbaum GG, Heyman A, Huber MS, et al. The prevalence and 3-year incidence of dementia in older Black and White community residents. J Clin Epidemiol 1998;51:587–95
5. Launer LJ, Andersen K, Dewey ME, et al. Rates and risk factors for dementia and Alzheimer's disease: Results from EURODEM pooled analyses. EURODEM Incidence Research Group and Work Groups. European Studies of Dementia. Neurology 1999;52:78–84
6. Barnes LL, Wilson RS, Schneider JA, et al. Gender, cognitive decline, and risk of AD in older persons. Neurology 2003;60:1777–81
7. Barnes LL, Wilson RS, Bienias JL, et al. Sex differences in the clinical manifestations of Alzheimer disease pathology. Arch Gen Psychiatry 2005;62:685–91
8. Kukull WA, Higdon R, Bowen JD, et al. Dementia and Alzheimer disease incidence: A prospective cohort study. Arch Neurol 2002;59: 1737–46
9. Rocca WA, Cha RH, Waring SC, et al. Incidence of dementia and Alzheimer's disease: A reanalysis of data from Rochester, Minnesota, 1975-1984. Am J Epidemiol 1998;148:51–62
10. Bachman DL, Wolf PA, Linn RT, et al. Incidence of dementia and probable Alzheimer's disease in a general population: The Framingham Study. Neurology 1993;43:515–9
11. Kawas C, Gray S, Brookmeyer R, et al. Age-specific incidence rates of Alzheimer's disease: The Baltimore Longitudinal Study of Aging. Neurology 2000;54:2072–7
12. Ganguli M, Dodge HH, Chen P, et al. Ten-year incidence of dementia in a rural elderly US community population: The MoVIES Project. Neurology 2000;54:1109–16
13. Hebert LE, Scherr PA, McCann JJ, et al. Is the risk of developing Alzheimer's disease greater for women than for men? Am J Epidemiol 2001; 153:132–6
14. McKhann GM, Knopman DS, Chertkow H, et al. 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 2011;7:263–9
15. Beach TG, Adler CH, Sue LI, et al. Arizona Study of Aging and Neurodegenerative Disorders and Brain and Body Donation Program. Neuropathology 2015;35:354–89
16. Beach TG, Sue LI, Walker DG, et al. The Sun Health Research Institute Brain Donation Program: Description and experience, 1987-2007. Cell Tissue Bank 2008;9:229–45
17. Gallyas F. An argyrophil III method for the demonstration of fibrous neuroglia. Acta Morphol Acad Sci Hung 1981;29:185–93
18. Braak H, Braak E. Demonstration of amyloid deposits and neurofibrillary changes in whole brain sections. Brain Pathol 1991;1:213–6
19. Hasegawa M, Arai T, Nonaka T, et al. Phosphorylated TDP-43 in frontotemporal lobar degeneration and amyotrophic lateral sclerosis. Ann Neurol 2008;64:60–70
20. Beach TG, White CL, Hamilton RL, et al. Evaluation of alpha-synuclein immunohistochemical methods used by invited experts. Acta Neuropathol 2008;116:277–88
21. Fujiwara H, Hasegawa M, Dohmae N, et al. alpha-Synuclein is phosphorylated in synucleinopathy lesions. Nat Cell Biol 2002;4:160–4
22. Obi K, Akiyama H, Kondo H, et al. Relationship of phosphorylated alpha-synuclein and tau accumulation to Abeta deposition in the cerebral cortex of dementia with Lewy bodies. Exp Neurol 2008;210:409-20
23. Walker DG, Lue LF, Adler CH, et al. Changes in properties of serine 129 phosphorylated alpha-synuclein with progression of Lewy-type histopathology in human brains. Exp Neurol 2013;240:190–204
24. Mirra SS, Heyman A, McKeel D, et al. The Consortium to Establish a Registry for Alzheimer's Disease (CERAD). Part II. Standardization of the neuropathologic assessment of Alzheimer's disease. Neurology 1991;41:479–86
25. Braak H, Braak E. Neuropathological stageing of Alzheimer-related changes. Acta Neuropathol 1991;82:239–59
26. Montine TJ, Phelps CH, Beach TG, et al. National Institute on Aging-Alzheimer's Association guidelines for the neuropathologic assessment of Alzheimer's disease: A practical approach. Acta Neuropathol 2012; 123:1–11
27. 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 1997;18:S1–2
28. Hyman BT, Trojanowski JQ. Consensus recommendations for the postmortem diagnosis of Alzheimer disease from the National Institute on Aging and the Reagan Institute Working Group on diagnostic criteria for the neuropathological assessment of Alzheimer disease. J Neuropathol Exp Neurol 1997;56:1095–7

29. Andersen K, Launer LJ, Dewey ME, et al. Gender differences in the incidence of AD and vascular dementia: The EURODEM Studies. EURODEM Incidence Research Group. Neurology 1999;53:1992–7
30. Brayne C, Gill C, Paykel ES, et al. Cognitive decline in an elderly population–A two wave study of change. Psychol Med 1995;25:673–83
31. Miech RA, Breitner JC, Zandi PP, et al. Incidence of AD may decline in the early 90s for men, later for women: The Cache County study. Neurology 2002;58:209–18
32. Ruitenberg A, Ott A, van Swieten JC, et al. Incidence of dementia: Does gender make a difference? Neurobiol Aging 2001;22:575–80
33. Knopman DS. beta-Amyloidosis and neurodegeneration in Alzheimer disease: Who's on first? Neurology 2014;82:1756–7
34. Sofuoglu M, Dudish-Poulsen S, Nicodemus KK, et al. Characteristics of research volunteers for inpatient cocaine studies: Focus on selection bias. Addict Behav 2000;25:785–90
35. Mandel FS, Weiner M, Kaplan S, et al. An examination of bias in volunteer subject selection: Findings from an in-depth child abuse study. J Trauma Stress 2000;13:77–88
36. Gustavsson JP, Asberg M, Schalling D. The healthy control subject in psychiatric research: Impulsiveness and volunteer bias. Acta Psychiatr Scand 1997;96:325–8
37. Beach TG, Monsell SE, Phillips LE, et al. Accuracy of the clinical diagnosis of Alzheimer disease at National Institute on Aging Alzheimer Disease Centers, 2005-2010. J Neuropathol Exp Neurol 2012;71:266–73
38. Mayeux R, Saunders AM, Shea S, et al. Utility of the apolipoprotein E genotype in the diagnosis of Alzheimer's disease. Alzheimer's Disease Centers Consortium on Apolipoprotein E and Alzheimer's Disease. N Engl J Med 1998;338:506–11
39. Tsuboi Y, Josephs KA, Cookson N, et al. APOE E4 is a determinant for Alzheimer type pathology in progressive supranuclear palsy. Neurology 2003;60:240–5
40. Dealberto MJ, Mcavay GJ, Seeman T, et al. Psychotropic drug use and cognitive decline among older men and women. Int J Geriatr Psychiatry 1997;12:567–74
41. Rozzini R, Ferrucci L, Losonczy K, et al. Protective effect of chronic NSAID use on cognitive decline in older persons. J Am Geriatr Soc 1996;44:1025–9
42. Zhu L, Viitanen M, Guo Z, et al. Blood pressure reduction, cardiovascular diseases, and cognitive decline in the mini-mental state examination in a community population of normal very old people: A three-year follow-up. J Clin Epidemiol 1998;51:385–91
43. Piccinin AM, Muniz-Terrera G, Clouston S, et al. Coordinated analysis of age, sex, and education effects on change in MMSE scores. J Gerontol B Psychol Sci Soc Sci 2013;68:374–90
44. Matthews F, Marioni R, Brayne C. Examining the influence of gender, education, social class and birth cohort on MMSE tracking over time: A population-based prospective cohort study. BMC Geriatr 2012;12:45
45. Jacqmin-Gadda H, Fabrigoule C, Commenges D, et al. A 5-year longitudinal study of the Mini-Mental State Examination in normal aging. Am J Epidemiol 1997;145:498–506
46. Prince M, Lewis G, Bird A, et al. A longitudinal study of factors predicting change in cognitive test scores over time, in an older hypertensive population. Psychol Med 1996;26:555–68
47. Caselli RJ, Dueck AC, Locke DE, et al. Sex-based memory advantages and cognitive aging: A challenge to the cognitive reserve construct? J Int Neuropsychol Soc 2015;21:95–104
48. Salehi A, Gonzalez M,V, Swaab DF. A sex difference and no effect of ApoE type on the amount of cytoskeletal alterations in the nucleus basalis of Meynert in Alzheimer's disease. Neurobiol Aging 1998;19:505–10
49. Miller FD, Hicks SP, D'Amato CJ, et al. A descriptive study of neuritic plaques and neurofibrillary tangles in an autopsy population. Am J Epidemiol 1984;120:331–41
50. Ghebremedhin E, Schultz C, Thal DR, et al. Gender and age modify the association between APOE and AD-related neuropathology. Neurology 2001;56:1696–701
51. Sandberg G, Stewart W, Smialek J, et al. The prevalence of the neuropathological lesions of Alzheimer's disease is independent of race and gender. Neurobiol Aging 2001;22:169–75
52. Rocca WA. Frequency, distribution, and risk factors for Alzheimer's disease. Nurs Clin North Am 1994;29:101–11
53. Bondareff W. Age and Alzheimer disease. Lancet 1983;1:1447
54. Hatazawa J, Ito M, Yamaura H, et al. Sex difference in brain atrophy during aging; a quantitative study with computed tomography. J Am Geriatr Soc 1982;30:235–9
55. Hubbard BM, Anderson JM. A quantitative study of cerebral atrophy in old age and senile dementia. J Neurol Sci 1981;50:135–45
56. Scheff SW, Neltner JH, Nelson PT. Is synaptic loss a unique hallmark of Alzheimer's disease? Biochem Pharmacol 2014;88:517–28