Multiple Biological Pathways Link Cognitive Lifestyle to Protection from Dementia

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Background: An active cognitive lifestyle is linked to diminished dementia risk, but the underlying mechanisms are poorly understood. Potential mechanisms include disease modification, neuroprotection, and compensation. Prospective, population-based brain series provide the rare opportunity to test the plausibility of these mechanisms in humans.

Methods: Participants came from the United Kingdom Medical Research Council Cognitive Function and Ageing Study, comprising 13,004 individuals aged over 65 years and followed for 14 years. In study 1, a Cognitive Lifestyle Score (CLS) was computed on all Cognitive Function and Ageing Study subjects to define low, middle, and high groups. By August 2004, 329 individuals with CLS data had come to autopsy and underwent Consortium to Establish a Registry of Alzheimer's Disease assessment. Study 2 involved more detailed quantitative histology in the hippocampus and Brodmann area 9 in 72 clinically matched individuals with high and low CLS.

Results: CLS groups did not differ on several Alzheimer disease neuropathologic measures; however, high CLS men had less cerebrovascular disease after accounting for vascular risk factors, and women had greater brain weight. No group differences were evident in hippocampal neuronal density. In Brodmann area 9, cognitively active individuals had significantly greater neuronal density, as well as correlated increases in cortical thickness.

Conclusions: An active cognitive lifestyle was associated with protection from cerebrovascular disease in men, but there was no evidence for Alzheimer disease modification or hippocampal neuroprotection. Men and women both exhibited neurotrophic changes in the prefrontal lobe linked to cognitive lifestyle, consistent with a compensatory process. Lifespan complex cognitive activity may therefore protect against dementia through multiple biological pathways.

Key Words: Cortical thickness, dementia, mental activity, neuronal density, neuropathology, protective factors

Effective dementia-prevention strategies are an essential global health priority because the aging of modern societies will see the number of affected individuals rise to over 100 million by 2050 (1,2). Cultivation of an active cognitive lifestyle through lifespan engagement in a rich array of mentally stimulating activities may help reduce dementia risk and is hence a promising public health measure (3,4). Yet, the neurobiological mechanisms underlying this protective relationship are not well understood, particularly in humans.

Three classes of mechanism have been suggested that could plausibly mediate these positive dementia outcomes (5). Environmental enrichment produces a disease-modifying effect in transgenic mice models of Alzheimer's disease (AD), whereby increased social, cognitive, and physical stimulation results in reduction of β-amyloid plaque load (6,7). Enrichment studies also implicate a possible neuroprotective effect, evident by attenuated neuronal loss in animal models of neurodegenerative disease (8).

Human neuroimaging studies point to the role of a third important compensatory mechanism (9). Older individuals with more years of education or history of occupational complexity require a greater degree of neuropathologic damage to manifest a particular level of clinical symptoms (10,11). Older individuals with superior memory performance (in the range of younger individuals) also utilize additional prefrontal cortical areas in comparison with those with age-related memory dysfunction (12,13). In particular, mid-prefrontal lobe regions, such as Brodmann areas 9 and 10, have been implicated in successful memory performance (13,14). Because compensation includes increased functional activity of discrete brain networks, this long-term neuroplastic adaptation is likely to have microstructural correlates in the form of neurotrophic changes to underlying synaptic and neuronal populations (5).

Disentangling the primacy or relative importance of these three possible mechanisms in humans is very difficult; however, distinct predictions can be proposed. All things being equal, disease modification predicts that those individuals with a more active cognitive lifestyle should have less AD pathology at death; neuroprotection predicts that for a given level of AD pathology, an active cognitive lifestyle should translate to a greater number of healthy neurons in the brain regions most affected by AD; and compensation predicts that cognitively active individuals will manifest neurotrophic microstructural changes in the mid-prefrontal lobe.

Prospective, population-based studies of elderly individuals who have progressed to brain autopsy provide us with the rare chance to test the relative merits of these predictions (15). One such large and well-documented study is the Cognitive Function and Ageing Study (CFAS), a longitudinal multicenter study based in the United Kingdom now in its 14th year of follow-up of over 13,000

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individuals from around the country (16). Our objective was to assess at postmortem predictions based on three putative mechanisms in individuals with a broad history of cognitive lifestyle patterns.

Methods and Materials

CFAS Subjects

Data were taken from the United Kingdom Medical Research Council Cognitive Function and Ageing Study (http://www.cfas.ac.uk). This is a large multicenter population-based prospective cohort study of individuals aged 65 years and older, including community and residential settings (17). Individuals were randomly selected from the Family Health Service Authority lists in five areas in England and Wales, including two rural (Cambridgeshire and Gwynedd) and three urban (Newcastle, Nottingham, and Oxford). Baseline interviews were undertaken from 1991 to 1992. All Medical Research Council CFAS interviewing and brain donation were covered by local and multicenter research ethics approval. In addition, postmortem histologic brain studies carried out in Sydney were authorized by the University of New South Wales Human Research Ethics Committee (HREC #6325). Written consent was obtained from all participants. Further details about the CFAS study are available in Supplement 1.

CFAS Brain Series

Individuals, families, and caregivers were approached by trained liaison officers and invited to participate in counseling around brain donation. Those who agreed to brain donation were provided with information to allow those involved in the final illness to notify the death and initiate brain donation. Donations still proceeded, wherever possible, for cases coming to autopsy under the coroner. By August 1, 2004, the complete brain donation sample included 456 individuals; however, the Liverpool center ($n = 101$) did not have the required social engagement information for calculating the Cognitive Lifestyle Score (CLS). Hence, there are 329 individuals in this analysis representing all completed brain donations with CLS data (total potential sample of 355, 93% completion rate).

Clinical Status

Dementia was assessed prospectively as a Geriatric Mental State-Automated Geriatric Examination for Computer Assisted Taxonomy (AGECAT) organicity rating case level of 3 or above, which incorporates limited cognitive testing and is comparable with dementia as diagnosed by DSM-III-R (15,16). Final dementia status was established using multiple information sources, including the AGECAT diagnosis as well as notification of dementia in death certificates, a retrospective informant interview (RINI; www.cfas.ac.uk) with relatives and caregivers after death (to capture the gap between last interview and death), and the probability of being demented before death from a Bayesian analysis of all individuals, modeling the prevalence and incidence of dementia in CFAS (16). Dementia was thereby established without reference to pathologic data. More details of this process can be found elsewhere (18,19).We could not assign dementia status in 30 individuals in whom the study diagnosis was "not dementia." These respondents were not included in the analysis because their last interview was more than 6 months before death, no RINI was available, and dementia was not mentioned on the death certificate. For the quantitative histologic substudy, those individuals without dementia were further split into normal cognition (Mini-Mental State Examination score 26–30) and cognitive impairment (Mini-Mental State Examination score <26).

Cognitive Lifestyle Score

The CLS asks detailed information about an individual's range of educational, occupational, and social activities and has been shown to independently predict incident dementia in the larger CFAS sample (20). Briefly, years of education, occupational complexity coded according to social class and socioeconomic grouping, and social engagement based on frequency of contact with relatives, neighbors, and social events are combined to produce a score for the total CLS. Sex-specific tertiles were then calculated to classify individuals as low, middle, or high CLS.

Study 1: Pathologist Ratings

The brain sample of 329 individuals underwent standardized neuropathologic assessment based on paraffin-embedded tissues and an extension of the Consortium to Establish a Registry of Alzheimer's Disease protocol (developed by P.I.) (21). Consortium to Establish a Registry of Alzheimer's Disease data were augmented by a strategy for evaluating white matter lesions in the postmortem brain previously validated against histopathology (22). Neuropathology was assessed without knowledge of clinical, interview, or RINI data. Acceptable interrater reliability ($<5\%$ with scores more than one grade difference) was achieved for cerebral cortical atrophy, neurofibrillary tangles, β-amyloid plaques (diffuse and neuritic), Lewy bodies, and cerebral amyloid angiopathy by circulation of macroscopic brain photographs and microscopic slides.

Study 2: Quantitative Histology

A subsample of the CFAS brain series with high and low CLS scores was selected for more detailed quantitative histologic analysis ($n = 72$; Table S1 in Supplement 1). Selection was based on an exhaustive search for cases with a postmortem delay of less than 48 hours to ensure high-quality tissue and that cases were matched between CLS groups for age, sex, and cognitive status. Normal cognition, cognitive impairment, and dementia at death were equally represented in both CLS groups to assess putative biological mechanisms across the clinical spectrum and avoid biases inherent in choosing individuals with a given cognitive status. Initially, a pilot study of sections taken from 10 individuals was completed to validate the transportation and testing protocols for use in the main study. These individuals were representative of both high and low CLS scores and were matched cognitively. Selection for the main histologic substudy identified 66 individuals; however, 3 individuals were inadvertently in both the pilot and main study (blinded to investigators). Tissue from one individual could not be used. Since protocols for the pilot and main study were identical and the results were robust to inclusion or exclusion of the pilot cases (data not shown), the complete subsample of $n = 72$ individuals has been used here (i.e., $10 + 66 - 3 - 1$).

Paraffin-embedded coronal sections (10 μm) from the hippocampus and Brodmann area 9 were cut and mounted at the Sheffield Hospital Brain Bank and sent to Sydney for analysis. Immunostaining was adapted from a standard DAB peroxidase staining procedure (23). Details of immunohistologic procedures are available in Supplement 1.

Neuron Density Quantitation

Nissl staining was used to estimate neuronal density per cubic millimeter in Brodmann area 9 and cornu ammonis 1 region using our previously published procedures, which are based on stereological principles (24,25). Criteria for counting neurons were based on cell size and morphologic characteristics, including the presence of a nucleolus. For Brodmann area 9 sections, three to five sample

zones located at the base of sulci were analyzed. Further details are available in Supplement 1.

Statistical Analysis

The complete CFAS brain series was analyzed using univariate logistic regression, as well as after adjustment for age, dementia at death, apolipoprotein E, and vascular risk factors. The association between CLS and dementia adjusted for neuropathologic factors used multivariable logistic regression. For the quantitative histologic study, relationships between neuropathologic variables and CLS group were examined using analysis of covariance, after adjusting just for matching variables (sex, age at death, and cognitive group) and then after additional adjusting for brain weight, apolipoprotein E4 (APOE4) allele status, and vascular risk factors. History of vascular risk factors (diabetes, medicated high blood pressure, heart attack, angina, stroke) was asked at each interview and smoking history at baseline. These factors were combined to produce a vascular risk factor score: individuals were given 1 point for the presence of each risk factor, except for smoking where individuals were coded as 0 for nonsmokers, 1 for ex-smokers (more than 5 years ago), and 2 for current smokers. Individuals could therefore score between 0 and 8, and the median score was 2.

Differences between the two CLS groups were estimated from the adjusted marginal totals. Correlations between the continuous variables were evaluated using Spearman's rho. Brain weight was missing in 12 individuals, and hence, results presented have used multiple imputation (100 replications). A sensitivity analysis to the missing data was performed, excluding those with missing brain weight and additionally after including them in either the lowest and highest CLS groups with no differences in the results (data not shown). The relationship between the variables and clinical status has been investigated using ordered logistic regression. STATA 11.0 (StataCorp LP, College Station, Texas) was used for the analysis.

Results

Sample Characteristics

The CFAS brain series comprised 329 individuals with CLS data who had come to autopsy by August 1, 2004. Their clinical characteristics in comparison with the whole CFAS sample and those who had died by the same date without brain donation consent (comparison group) are presented in Table 1. In general, there were only minor differences between the CFAS brain series and comparison group.

No Evidence for Alzheimer's Disease Modification by Cognitive Lifestyle

In study 1, the disease modification hypothesis was assessed in the CFAS brain series by comparing a broad panel of neuropathology measures between CLS groups. There were no significant differences on any AD marker, including diffuse and neuritic plaques, neurofibrillary tangles, hippocampal atrophy, and cerebral amyloid angiopathy (Table S2 male subjects and Table S3 female subjects in Supplement 1). This absence of group CLS differences persisted

Table 1. Cohort Characteristics of CFAS Brain Series and Comparison with the Sample of Individuals in the Epidemiologic Cohort Who Died Within the Same Time Period

	CFAS Brain Series Men (n = 138)		CFAS Cohort Deceased Men (n = 3414)		CFAS Brain Series Women (n = 191)		CFAS Cohort Deceased Women (n = 4652)	
	n	%	*n*	%	*n*	%	*n*	%
Center								
Cambridgeshire	34	25	737	22	59	31	870	19
Gwynedd	3	2	647	19	3	2	822	18
Newcastle	21	15	636	19	27	14	1026	22
Nottingham	54	39	720	21	66	35	935	20
Oxford	26	19	674	20	36	19	999	21
Median Time Between Last Interview and Death, IQR (Years)	1.4	.8–2.2	5.2	2.6–8.2	1.3	.7–2.3	5.8	3.0–8.8
Age at Death								
<80 years	42	30	1323	39	32	17	993	21
80–89 years	70	51	1653	48	82	43	2396	52
90+ years	26	19	438	13	77	40	1263	27
Mean Age at Death, SD (Years)	83.5	7.1	82.1	6.7	87.6	7.3	85.5	6.9
Dementia Status at Death								
Dementia	46	33	Not known		102	53	Not known	
No dementia	77	56	Not known		76	40	Not known	
Unclear	15	11			13	7		
Vascular Risk Factors								
Stroke	40	29	909	27	65	34	1359	30
Angina	33	24	767	22	41	21	870	19
Diabetes	17	12	381	11	21	11	445	10
Hypertension (medicated)	44	32	950	28	71	37	1576	35
Heart attack	37	27	1030	30	36	19	1025	22
Cognitive Lifestyle Score								
Low	53	38	1169	35	80	42	1671	38
Medium	44	32	1037	31	61	32	1420	32
High	41	30	1138	34	50	26	1347	30

n refers to number of individuals and % to proportion of relevant total sample (*N*).
CFAS, Cognitive Function and Ageing Study; IQR, interquartile range; SD, standard deviation.

after multivariable adjustment for age, dementia status, APOE4, and vascular risk factors. There was therefore no support for the hypothesis that an active cognitive lifestyle modifies the pathogenesis of AD.

Reduced Cerebrovascular Disease in Male Subjects

In men ($n = 138$), we found a significant reduction in microvascular disease in the active cognitive lifestyle group, especially an 80% relative reduction in deep white matter lesions (13% in the high CLS group vs. 43% in the low CLS group, odds ratio [OR] = .2, confidence interval [CI]: .1–.6, $p = .003$; see Figure 1 and Table S4 in Supplement 1 for further details). After adjusting for age, dementia status, cardiovascular risk factors, and APOE4, the protective effect of active cognitive lifestyle remained undiminished (OR = .2, CI: .0–.5, $p = .002$), along with protective effects on frequency of lacunar infarcts (OR = .3, CI: .1–.9, $p = .04$) and microscopic atherosclerosis (OR = .3, CI: .1–1.0, $p = .04$). A continuous outcome variable was also calculated based on presence or absence of any cerebrovascular disease features (maximum = 11, minimum = 0). Multivariable regression analysis replicated this protective effect: membership of the high CLS group was associated with 1.2 fewer cerebrovascular disease features in men (CI: .4–2.0, $p = .005$, adjusted for background variables of age, dementia status, and vascular risk factors).

Parallel analyses in women ($n = 191$) did not reveal any associations between cognitive lifestyle and any vascular or pathologic lesion (see Table S5 in Supplement 1 for further details), despite men and women exhibiting comparable levels of vascular pathology (mean vascular disease summary score in men was 3.5, SD = 2.1, and in women 4.1, SD = 2.0). Higher than average brain weight, however, was more common in women with high CLS (46%) compared with low CLS groups (20%, OR = 3.0, CI: 1.1–7.3 $p = .002$ adjusted). In women, therefore, an active cognitive lifestyle was associated with greater brain weight but no such relationship was observed in male subjects.

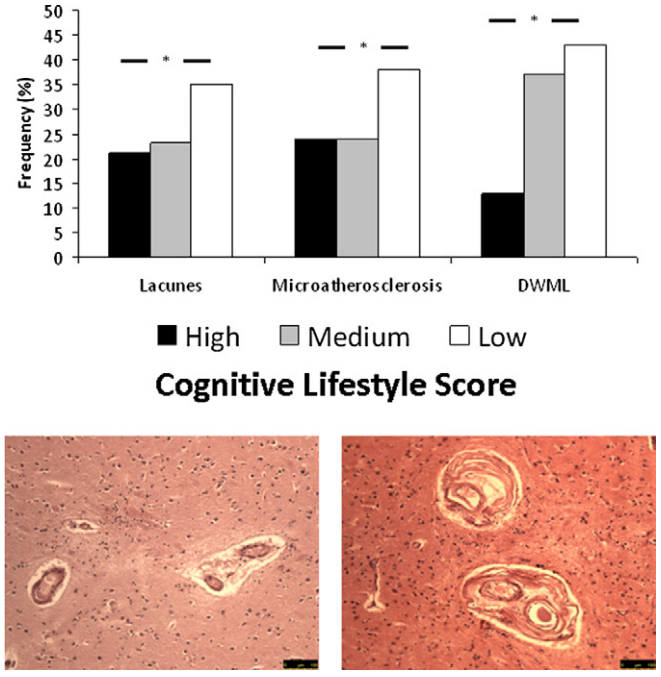

Figure 1. Decreased frequency of microvascular disease in men with a high Cognitive Lifestyle Score ($n = 138$). Graph shows frequency of lacunes, microatherosclerosis, and deep white matter lesions in the different Cognitive Lifestyle Score tertile groups (high, medium, and low). Exemplar images of individuals with normal microvasculature (left) and severe vessel atherosclerosis. Further details available in Tables S3 and S4 in Supplement 1. DWML, deep white matter lesions. *Significant results, $p < .05$.

Cognitive Lifestyle Effect Remains After Accounting for Neuropathology

A logistic regression analysis was tested with dementia status as the dependent variable, along with predictor variables: CLS group membership, age, brain weight, cerebrovascular summary score, and six different neuropathologic AD and non-AD variables (Table 2). In male subjects, an active cognitive lifestyle remained a protective factor for dementia at death compared with those with a low cognitive lifestyle after accounting for these neuropathologic variables (OR = .2, CI: .0–1.0, $p = .04$). There was no residual protective effect of cognitive lifestyle on dementia at death in women; however, significant predictors included neurofibrillary tangles, vascular disease, and hippocampal atrophy.

No Evidence of Neuroprotective Effect in Hippocampus

Study 2 aimed at testing predictions based on the neuroprotection and compensation hypotheses and therefore required detailed quantitative measures of neuronal density, as well as cortical thickness. A subsample ($n = 72$) of individuals from high and low CLS groups was selected from the CFAS brain series, with criteria based on adequate tissue quality (postmortem interval <48 hours), as well as age, sex, and vascular risk factor matched across a broad clinical spectrum.

We first collapsed across CLS groups and observed the expected relationship between cognitive impairment and AD pathology: demented individuals had greater β-amyloid and tau pathology compared with cognitively intact individuals. Significant differences were seen for hippocampal β-amyloid (OR = 2.2, 95% CI: 1.1–4.5, $p = .03$), frontal β-amyloid (OR = 7.6, 95% CI: 1.6–34.9, $p = .009$), hippocampal tau (OR = 2.8, 95% CI: 1.3–6.3, $p = .01$), and frontal tau (OR = 177, 95% CI: 5.4–5748, $p = .004$). Demented individuals also had higher hippocampal glial fibrillary acidic protein (OR = 3.0, 95% CI: 1.1–7.7, $p = .027$) but not frontal glial fibrillary acidic protein or human leukocyte antigen-DR in either region ($p > 0.1$). In addition, we replicated the absence of any disease burden differences between CLS groups noted above, on this occasion using areal fractions of β-amyloid, tauopathy, and activated microglia (Figure 2F**).**

Next, to specifically test the neuroprotection hypothesis, three covariance analyses compared CLS groups after controlling for either β-amyloid, tau pathology, or activated microglia in the hippocampus and found no significant differences in hippocampal neuronal number per unit area. These findings did not change after further controlling for age, sex, brain weight, cardiovascular risk factors, and APOE4 status. Moreover, estimated adjusted marginal means were in the opposite direction to that predicted by the neuroprotection hypothesis (difference = 15.7, 95% CI: −34.4–3.0, $p = .10$). There was, hence, no evidence for neuronal protection in the hippocampus in association with differences on the CLS.

Neurotrophic Effect in Brodmann Area 9

As shown in Figure 3A–E, the high CLS group had significantly higher neuronal densities and a thicker cortical ribbon. The neuronal density difference was 13.1 neurons per unit area (95% CI: 2.6–23.6, $p = .01$) and the cortical thickness difference was 142 microns (95% CI: 0–284, $p = .05$); both these findings were significant after correction for age, sex, brain weight, APOE4, and cardiovascular risk factors (adjusted neuronal density difference 13.8, 95% CI: 2.8–25.4, $p = .02$; cortical thickness difference167 microns, 95% CI: 13–321,

Table 2. Multivariable OR Predictors of Clinical Dementia at Death

	Men ($n = 103$)				Women ($n = 150$)			
Variable	*n*	OR	95% CI	*p*	*n*	OR	95% CI	*p*
Cognitive Lifestyle Score								
Low	42	1.0		.04[a]	64	1.0		.24
Medium	35	.3	.1–1.3		45	1.0	.4–3.0	
High	26	.2	.0–1.0		41	.5	.1–1.5	
Age at Death								
<80 years	31	1.0		.10	27	1.0		.22
80–89 years	51	1.3	.3–5.8		64	2.9	.7–11.6	
≥90 years	21	5.9	.8–43.2		59	2.9	.7–12.5	
Brain Weight for Sex								
Low	33	1.0		.55	53	1.0		.12
Average	35	.7	.2–2.8		48	.5	.2–1.4	
High	35	.6	.1–3.1		49	.4	.1–1.3	
Neuritic Plaques in Neocortex								
None/mild/moderate	90	1.0		.06	136			
Severe	13	12.4	.9–176.5		14	NA[b]		
Tangles in Neocortex								
None/mild	83	1.0		.12	115	1.0		.001[a]
Moderate/severe	20	5.6	.6–49.6		35	41.7	4.5–387.3	
Congophilic Angiopathy								
None	66	1.0		.02[a]	89	1.0		.58
Mild	22	10.9	1.9–61.9		28	.8	.2–2.9	
Moderate/severe	15	4.8	.9–25.1		33	1.6	.4–5.6	
Lewy Bodies								
No	97	1.0		.09	140	1.0		.08
Yes	6	9.3	.7–125.4		10	7.8	.8–77.7	
Overall Vascular Pathology								
None/infarcts/hemorrhage	16	1.0		.26	21	1.0		.03[a]
SVD/DWML/lacunes	58	8.5	.8–90.8		78	1.4	.4–5.2	
Both	29	7.5	.6–89.6		51	4.1	1.0–16.7	
Hippocampal Atrophy								
None	61	1.0		.03[a]	64	1.0		.004[a]
Mild	23	10.0	1.8–55.9		33	2.3	.7–6.9	
Moderate/severe	19	4.7	.7–32.7		53	5.8	1.8–19.0	

High Cognitive Lifestyle Score was a protective factor in men after adjustment for all other listed variables. *p* values refer to test of trend effect.
CI, confidence interval; DWML, deep white matter lesions; NA, not applicable; OR, odds ratio; SVD, small vessel disease.
[a]Significant *p* values.
[b]Neocortical neuritic plaques not included in the multivariable model for women due to multicollinearity.

$p = .03$). These results were independent of the cognitive or clinical characteristics of our sample, since both high and low CLS groups were intentionally matched to include equivalent numbers of individuals with dementia and borderline and intact cognition. Neuronal density and cortical thickness in Brodmann area 9 were also positively correlated (Spearman's rho = .38, 95% CI: .17–.57).

Discussion

Our findings suggest that the protective relationship between cognitive lifestyle and dementia risk could be mediated by a number of mechanisms. In men, reduced development of microvascular disease was implicated, while in women, greater brain weight. In both sexes, we found convergent evidence of a neurotrophic effect in Brodmann area 9, including correlated increases in cortical thickness and neuronal density.

Cerebrovascular disease (CVD) is a major determinant of cognitive dysfunction in late life (26), so it is revealing that a substantial reduction in CVD was observed in men with an active cognitive lifestyle, particularly on multiple measures of microvascular disease. Remarkably, rates of deep white matter lesions, microscopic atherosclerosis, and lacunes were reduced by between 70% to 80% in more cognitively active men, a finding that could not be explained by differences in vascular risk factors. Del Ser *et al.* (27) found higher rates of CVD in those with lower education level, but this study was based on a clinic-based sample and, paradoxically, also reported that those with lower education became demented later in life compared with those with higher education levels. Our population-based CFAS brain series helps clarify the issue—an inactive cognitive lifestyle was associated with both an increase in microvascular disease and increased frequency of dementia at death. Similar findings have been found in animal stroke models, with environmental enrichment leading to decreased infarct volume, increased spine density, and normalized neuronal and astrocyte counts. Interestingly, even after accounting for vascular disease and a number of neuropathologic variables in our multivariable modeling, in men there remained a protective association between cognitive lifestyle and reduced dementia at death. Modification of CVD is therefore not likely to be the only implicated biological pathway.

In women, an active cognitive lifestyle was associated with greater brain weight, which in the context of a clinicopathologic study is difficult to interpret. These women may have had a rela-

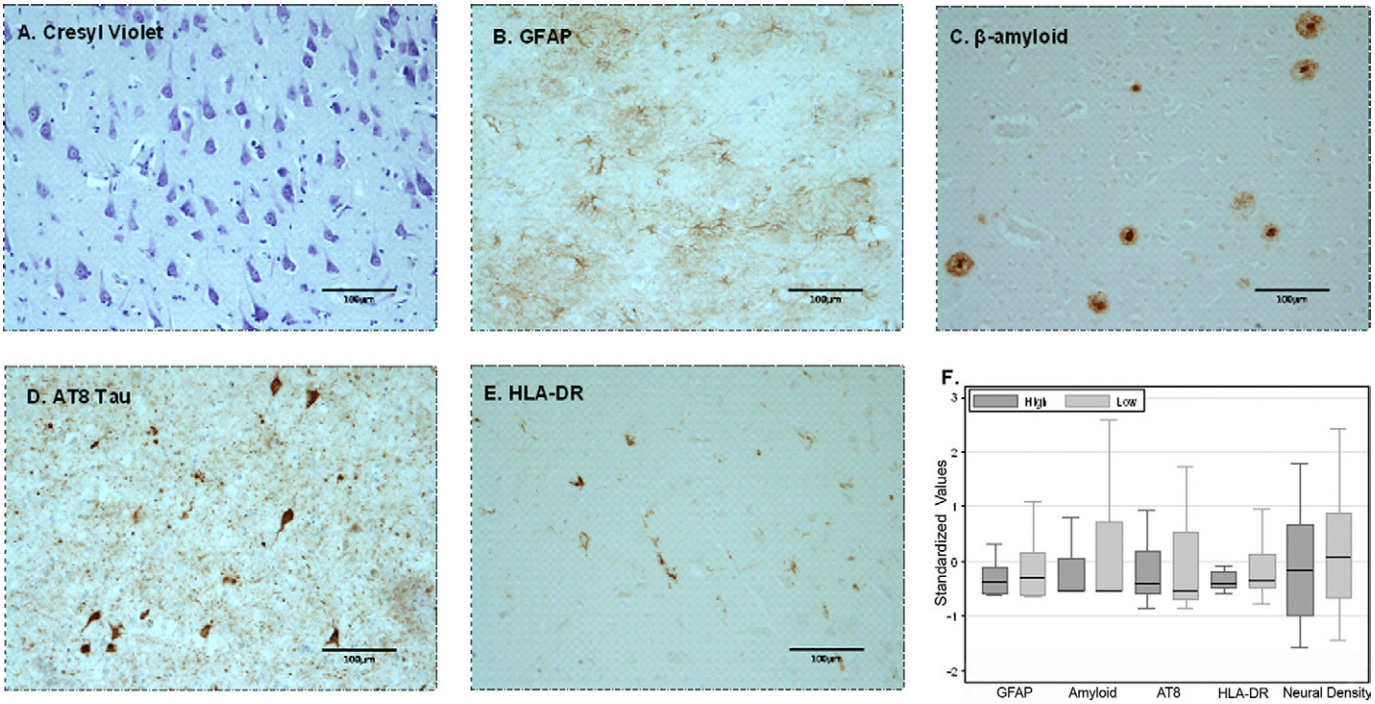

Figure 2. No evidence of Alzheimer disease modification or pan-neuronal neuroprotection in the hippocampus. Exemplar images and results of histologic analyses in the cornu ammonis 1 subregion: **(A)** neurons, **(B)** glial cells, **(C)** β-amyloid, **(D)** tauopathy, and **(E)** activated microglia. Graph in **(F)** shows there were no significant differences between the Cognitive Lifestyle Score groups on any histologic measure (all variables have been normalized for comparison purposes). Raw values available in Tables S1 and S2 in Supplement 1. GFAP, glial fibrillary acidic protein; HLA-DR, human leukocyte antigen-DR.

tively larger brain throughout their lives, as suggested by epidemiologic studies linking greater intracranial volume (28) or skull size (29) to reduced dementia risk. However, this has not been entirely consistent (30) and, interestingly, may be more reliable in women than in men (31). Alternatively, women with a more active cognitive lifestyle may have benefited from less brain atrophy over the years (32) and hence died with relatively greater brain weight. Experience-dependent increases in white matter integrity (33) or brain volume as seen in human neuroimaging (34) and animal studies (35) may also have contributed. Further research is required to clarify these findings.

A putative AD modification mechanism was assessed on the basis of the largest population-based postmortem sample so far to examine cognitive lifestyle factors. Across nine different neuropathologic variables, no evidence for such a link was found, so it supports similar null results witnessed in some transgenic AD enrichment studies (36) but not others (7). Our null finding is also consistent with a recent multicenter study that found no relationship between education levels and AD pathology (37). Since an active cognitive lifestyle appeared to have a protective effect on dementia at death in the CFAS brain series but not on any AD-related pathologic measure, there was no support for an AD-modification mechanism.

More detailed quantitative histological analysis was required to assess the neuroprotection or compensation hypotheses, and we adopted a strategy of matching high and low CLS groups based on age, sex, vascular risk factors, and clinical status. This was important because high CLS decreases prospective risk for dementia (20). A cognitively unrestricted sample would therefore have been biased to selecting nondemented high CLS subjects and demented low CLS subjects. Our second study therefore minimized the role of cognitive selection bias and was specifically orientated toward estimating neuronal and cortical characteristics that varied with cognitive lifestyle. Neuroprotection was defined as a greater neuronal density after accounting for AD pathology in the local area, and we found no evidence for this in the hippocampus. In fact, when trying to explain the link between an active cognitive lifestyle and dementia risk reduction, our results trended in the opposite direction to that predicted, so there was no support for a pan-neuronal form of protection in the human hippocampus. Other relevant types of neuroprotection, however, cannot be excluded, such as protective effects on synaptic density (38), postsynaptic integrity (39), or dendritic arborization (40).

The superior frontal gyrus was examined because several functional neuroimaging studies have implicated this area in a potential compensatory network. Specifically, we tested for potential microstructural correlates of compensatory brain activity. Our analyses showed that an active cognitive lifestyle was linked to a thicker cortical ribbon and increased neuronal density in Brodmann area 9. These changes were positively correlated and not likely to be confounded, since a tissue shrinkage artifact would produce a negative correlation between cortical thickness and object counts. A degree of regional specificity was also implied, given these results were independent of brain weight at death or hippocampal neuronal density. Use of in vivo structural neuroimaging could further characterize the relationship between cognitive lifestyle and atrophy patterns across the whole brain. For example, using a similar measure to our CLS, one study has reported a positive association with frontal lobe gray matter volume (41). At a cellular level, these neurotrophic changes may reflect a combination of resistance to oxidative stress and apoptosis, neuronal soma growth, dendritic elaboration, and increased synaptic connectivity, processes that all require angiogenesis for metabolic regulation and could, in turn, lead to neuropil expansion. Indeed, individuals who remain cogni-

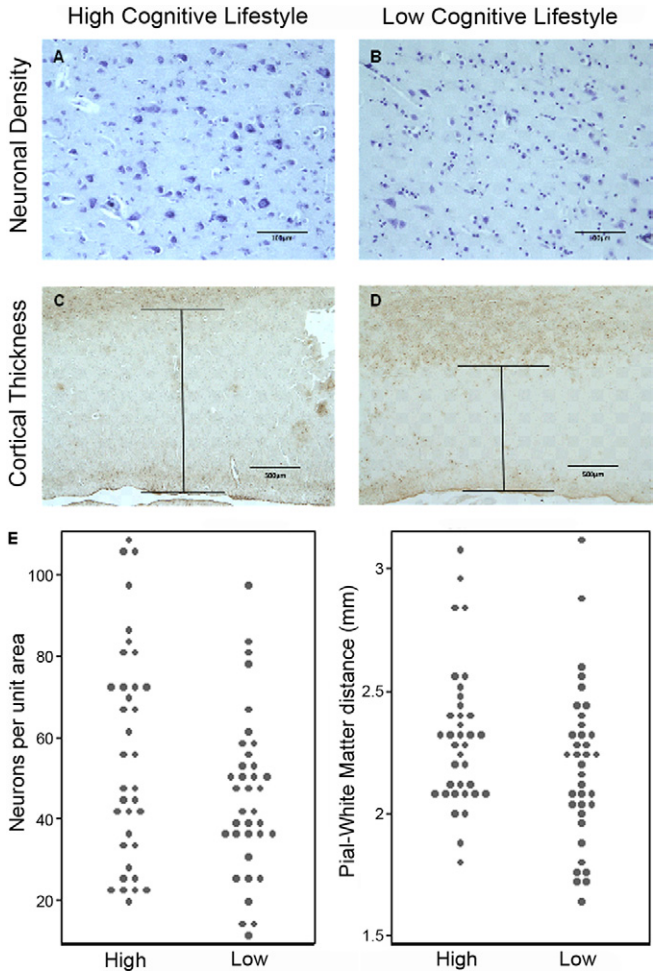

Figure 3. Neurotrophic effects of an active cognitive lifestyle in Brodmann area 9. Exemplar mid-frontal Brodmann area 9 sections from two individuals, showing that high Cognitive Lifestyle Score was associated with increased neuronal density **(A, B)** and greater neocortical thickness **(C, D)**. Part **(E)** depicts significant intergroup differences.

tively intact despite significant AD pathology exhibit widespread neuronal hypertrophy linked to greater linguistic ability in early life (42) or higher premorbid intelligence (43). These adaptations may reflect the influence of brain-derived neurotrophic factor (44) and other growth-signaling molecules, so further research linking cognitive lifestyle to molecular and intracellular signaling pathways is of considerable interest. At a functional level, microstructural changes in those who engage in a more active cognitive lifestyle may permit enhanced information processing in distributed prefrontal cortical networks known to mediate working memory (45) and executive function (46) and therefore combine to circumvent cognitive impairment.

These findings increase our understanding of the biological pathways linking cognitive lifestyle and brain function in later life and are hence relevant to discussions of brain and cognitive reserve (9,47). Prior studies have noted that education can modify the relationship between AD pathology and cognition (48–50), but our work suggests that a focus on education alone may be too narrow. In fact, cognitive activity beyond school was critical for maximal protection from dementia in the wider epidemiologic CFAS cohort (20). By employing such a lifespan perspective, a number of candidate mechanisms have been identified here that could plausibly mediate this relationship. The notion of a singular reserve may be inaccurate; rather, an active cognitive lifestyle may promote a number of reserve-related processes. A hypothetical model is proposed in Figure 4, which brings together these biological pathways—further research is required to tests the model's predictions. In particular, a population-based brain series with detailed neuropsychologic data, rather than the brief assessment included within the AGECAT diagnostic system, would help further understand the relationship between cognitive lifestyle and dementia risk. The broader significance of these findings is that they provide new biological support for strategies aimed at enriching the cognitive lifestyle of older individuals for the better prevention of dementia (9,47). Cognitive brain training can be effective when commenced in later life for delaying cognitive decline (4) and leads to detectable increases in cortical thickness (51); yet, whether these programs prevent or delay dementia incidence has yet to be established. Whether less structured, lifestyle-based cognitive engagement, such as volunteer work (52) or learning a new language (53), is equally effective is also unknown.

In conclusion, the connection between cognitive lifestyle and dementia risk is likely to be mediated by a number of biological mechanisms. These findings shed new light on our understanding of the brain's reserve processes and provide a firmer neurobiological basis for the development and testing of preventative cognitive lifestyle programs.

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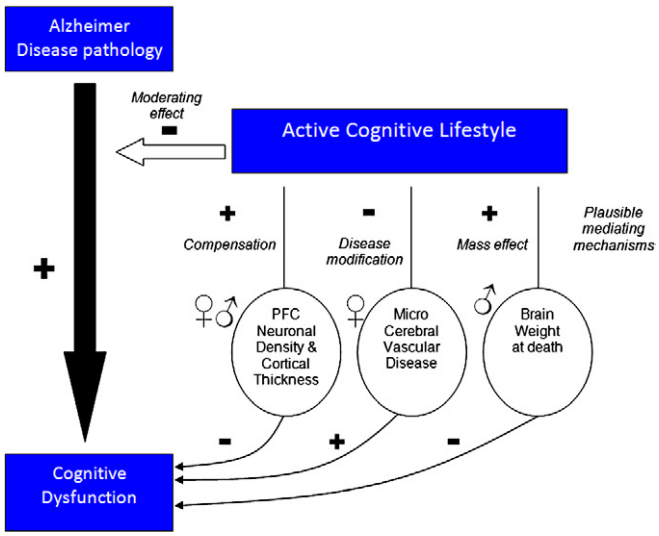

Figure 4. Model for how cognitive lifestyle modifies the link between Alzheimer pathology and cognitive function in later life. Three plausible mechanisms suggested: 1) modification of microcerebrovascular disease in men, 2) a whole brain mass effect in women, and 3) in both sexes, a neurotrophic effect in prefrontal cortex Brodmann area 9 involving a relative increase in neuronal density accompanied by enhanced cortical thickness. PFC, prefrontal cortex.

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