ORIGINAL ARTICLE

The Brain-Derived Neurotrophic Factor Val66Met Polymorphism and Prediction of Neural Risk for Alzheimer Disease

Aristotle N. Voineskos, MD, PhD, FRCPC; Jason P. Lerch, PhD; Daniel Felsky, BSc; Sajid Shaikh, BSc; Tarek K. Rajji, MD, FRCPC; Dielle Miranda, MA; Nancy J. Lobaugh, PhD; Benoit H. Mulsant, MD, MS, FRCPC; Bruce G. Pollock, MD, PhD, FRCPC; James L. Kennedy, MD, MSc, FRCPC

Context: The brain-derived neurotrophic factor (*BDNF*) Val66Met (rs6265) polymorphism may predict the risk of Alzheimer disease (AD). However, genetic association studies of the *BDNF* gene with AD have produced equivocal results. Imaging-genetics strategies may clarify the manner in which *BDNF* gene variation predicts the risk of AD via characterization of its effects on at-risk structures or neural networks susceptible in this disorder.

Objective: To determine whether the *BDNF* Val66Met gene variant interacts with age to predict brain and cognitive measures in healthy volunteers across the adult lifespan in an intermediate phenotype pattern related to AD by examining (1) cortical thickness, (2) fractional anisotropy of white matter tracts (ie, white matter integrity), and (3) episodic memory performance.

Design: A cross-sectional study using genetics, high-resolution magnetic resonance imaging, diffusion tensor imaging, and cognitive testing in healthy individuals spanning the adult lifespan.

Setting: University hospital.

Participants: A total of 69 healthy volunteers ranging from 19 to 82 years of age.

Main Outcome Measures: The *BDNF* Val66Met genotype, apolipoprotein E genotype, cortical thickness, microstructural integrity of white matter tracts, and episodic memory performance were evaluated.

Results: The *BDNF* Val66Met polymorphism interacted with age to predict (1) cortical thickness (prominently at the entorhinal cortex and temporal gyri), (2) fractional anisotropy of white matter tracts (prominently at white matter tracts connecting to the medial temporal lobe), and (3) episodic memory performance. For each of these findings, the pattern was similar: valine/valine individuals in late life were susceptible, and in early adult life, methionine allele carriers demonstrated susceptibility.

Conclusions: The *BDNF* gene confers risk in an age-dependent manner on the brain structures and cognitive functions that are consistent with the neural circuitry vulnerable in the earliest stages of AD. Our novel findings provide convergent evidence in vivo for a *BDNF* genetic mechanism of susceptibility in an intermediate phenotype related to AD.

Arch Gen Psychiatry. 2011;68(2):198-206

Sporadic or late-onset Alzheimer disease (AD) constitutes 90% to 95% of AD cases and is a complex, heterogeneous disorder with increasing prevalence.[1] Although some notable examples of genetic risk in AD have been established[2] and some promising data from genome-wide studies have emerged,[3,4] genetic investigations in this disorder have been fraught with many of the same complexities and conundrums as those of other neuropsychiatric disorders.[5] The brain-derived neurotrophic factor (*BDNF*) gene represents an intriguing potential genetic mechanism for risk of late-onset AD.[6] Brain-derived neurotrophic factor is critical for neuronal plasticity and facilitates hippocampal and cortical long-term potentiation,[7] processes that are especially important for learning and memory. Learning and memory processes are substantially affected in AD, arising largely from impaired neuronal plasticity.[8] In AD patients, *BDNF* expression is prominently reduced in the hippocampus and the entorhinal cortex,[9] and these regions are consistently affected in the earliest stages of the disease.[10,11] Variation in the *BDNF* Val66Met (rs6265, G>A) polymorphism has been shown to be related to episodic memory performance in younger adults via the hippocampal formation, where methionine (Met) allele carriers had poorer episodic memory performance.[12] In addition, this polymorphism predicts cognitive performance in elderly individuals[13] and may confer risk for AD,[14] where valine/valine (Val/Val) individuals in these 2 studies were at risk. Recent animal model find-

Author Affiliations are listed at the end of this article.

©2011 American Medical Association. All rights reserved.
Downloaded From: http://archpsyc.jamanetwork.com/ on 05/24/2016

ings suggest a compelling potential role for *BDNF* as a therapeutic agent in AD.[15] Taken together, these findings suggest that *BDNF* gene variation may be a genetic susceptibility mechanism for AD.

The combination of neuroimaging and genetics (ie, imaging-genetics) offers the potential to characterize the effects of *BDNF* risk variants on at-risk neural structures relevant to AD via the intermediate phenotype approach. Such an approach may evince greater penetrance of the effects of the gene on the vulnerable neural structure or function and is not subject to confounds present in disease populations.[16] In AD, a prominent at-risk neural feature in gray matter is reduced cortical thickness in temporal lobe structures, demonstrated via structural magnetic resonance imaging. Reduced thickness is most prominent in the entorhinal cortex,[17,18] a finding present at the earliest stages of disease, which aligns directly with neuropathologic studies that show that the earliest and greatest neurodegenerative changes occur in the entorhinal cortex and then in the hippocampus.[11] However, structural brain changes in AD are not limited to gray matter; more recently, white matter abnormalities have become a focus of investigation.[19,20]

Diffusion tensor imaging (DTI) is a powerful tool that can differentiate between normal and abnormal white matter.[21] In patients with AD, DTI has demonstrated disruption of white matter fibers in AD in corticocortical association fiber tracts.[22] Recent work[23] has identified that disruption of the cingulum bundle is highly correlated with hippocampal atrophy, represents the source of disconnection between the hippocampus and the posterior cingulate cortex, and is the primary factor in posterior cingulate cortex hypometabolism, a characteristic feature of this disorder.[24] White matter findings in AD align with neuropathologic studies, in which individuals with AD exhibit more severe oligodendroglial loss and myelin breakdown,[25] as well as axonal loss,[26] compared with matched control individuals. Brain-derived neurotrophic factor plays a role in mediating myelination,[27] provides trophic support for oligodendrocytes, and influences levels of myelin basic protein,[28] the major protein in the myelin sheath.

We conducted a study in healthy volunteers spanning the adult lifespan to assess the effect of the *BDNF* gene and age on neural structures and cognitive functions that are disrupted in AD. We hypothesized that the *BDNF* Val66Met polymorphism would interact with age to predict variation in (1) cortical thickness in temporal lobe structures, (2) microstructural integrity of white matter tracts that connect to the medial temporal lobe, and (3) episodic memory performance.

METHODS

STUDY PARTICIPANTS

Sixty-nine healthy volunteers (44 men and 25 women; mean [SD] age, 46 [18] years; age range, 19-82 years) met the inclusion criteria (age between 18 and 85 years; right handedness) and none of the exclusion criteria (any history of a mental disorder, including dementia; current substance abuse or a history of substance dependence; a positive urine toxicologic screen result; a history of head trauma with loss of consciousness, seizure, or another neurologic disorder; or a first-degree relative with a history of psychotic mental disorder). The ethnic distribution was 67 whites and 2 Asians. All participants were assessed with the Edinburgh handedness inventory,[29] were interviewed by a psychiatrist, and completed the Structured Clinical Interview for *DSM-IV* Disorders[30] and the Mini-Mental State Examination.[31] They also completed a urine toxicology screen. Participants were characterized using the following instruments (**Table**): the Wechsler Test for Adult Reading, the Hollingshead index,[32] the Cumulative Illness Rating Scale for Geriatrics,[33] and body mass index (calculated as weight in kilograms divided by height in meters squared) and blood pressure measurement. The study was approved by the Research Ethics Board of the Centre for Addiction and Mental Health (Toronto, Ontario, Canada), and all participants provided informed, written consent.

Table. Characteristics of the Study Participants

BDNF Genotype	Mean (SD) Met Carriers (n=28)	Mean (SD) Val/Val (n=41)	*t* test (*df*=67)	*P* Value
Age, y	45 (19)	47 (18)	−0.7	.49
Educational level, y	16 (2)	15 (2)	0.8	.43
Socioeconomic status[a]	52 (9)	48 (10)	1.4	.15
IQ (WTAR)	118 (9)	119 (6)	−0.6	.54
MMSE	29 (1)	29 (1)	1.1	.30
BMI	25 (3)	26 (5)	−1.3	.18
Systolic BP	124 (14)	123 (13)	0.2	.85
Diastolic BP	77 (6)	74 (9)	1.7	.10
CIRS-G (ratio score)	1 (1)	1 (1)	0.3	.78

Abbreviations: *BDNF*, brain-derived neurotrophic factor; BMI, body mass index (calculated as weight in kilograms divided by height in meters squared); BP, blood pressure; CIRS-G, Cumulative Illness Rating Scale–Geriatrics; Met, methionine; MMSE, Mini-Mental State Examination; Val, valine; WTAR, Wechsler Test of Adult Reading.

[a] The 4 factors, as designated by the Hollingshead Index of Socioeconomic Status, are educational level, occupation, sex, and marital status.

NEUROIMAGING

Image Acquisition

High-resolution magnetic resonance images were acquired as part of a multimodal imaging protocol using an 8-channel head coil on a 1.5-T GE Echospeed system (General Electric Medical Systems, Milwaukee, Wisconsin), which permits maximum gradient amplitudes of 40 mT/m. Axial inversion recovery–prepared spoiled gradient recall images were acquired: echo time, 5.3; repetition time, 12.3; time to inversion, 300.0; flip angle, 20°; and number of excitations, 1 (for a total of 124 contiguous images, 1.5-mm thickness). For DTI, a single-shot spin echo planar sequence was used with diffusion gradients applied in 23 noncollinear directions and $B=1000$ s/mm^2. Two $B=0$ images were obtained. Fifty-seven sections were acquired for whole brain coverage oblique to the axial plane. Section thickness was 2.6 mm, and voxels were isotropic. The field of view was 330 mm, and the size of the acquisition matrix was 128×128 mm, with an echo time of 85.5 milliseconds and a repetition time of 15 000 milliseconds. The entire sequence was repeated 3 times to improve signal to noise ratio.

©2011 American Medical Association. All rights reserved.
Downloaded From: http://archpsyc.jamanetwork.com/ on 05/24/2016

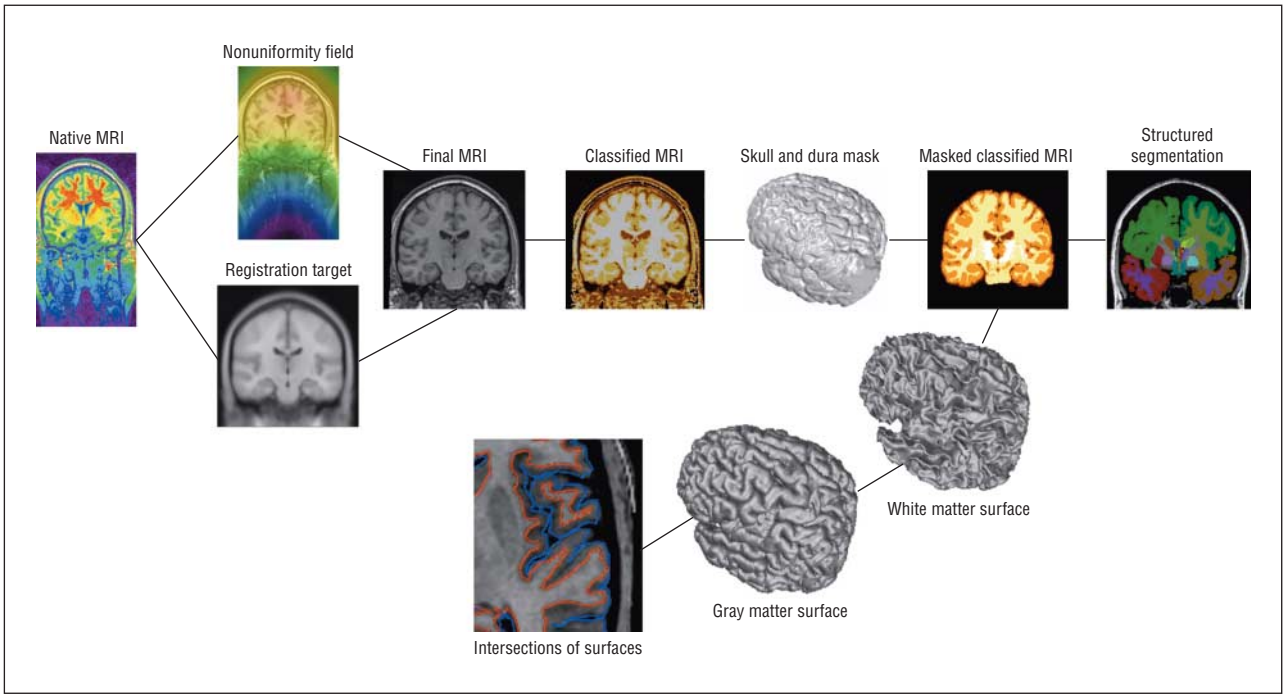

Figure 1. Processing pipeline for extraction of cortical thickness measures used to derive anatomical information from T1-weighted magnetic resonance images (MRIs). Each image is aligned to stereotaxic space, corrected for nonuniformity artifacts, tissue classified, and masked, and inner and outer cortical surfaces are extracted.

Image Processing

Cortical Thickness Mapping. All magnetic resonance images were submitted to the CIVET processing pipeline (version 1.1.9; Montreal Neurological Institute at McGill University, Montreal, Quebec, Canada). T1-weighted images were registered to the ICBM152 nonlinear sixth-generation template with a 9-parameter linear transformation, inhomogeneity corrected[34] and tissue classified.[35,36] Deformable models were then used to create white and gray matter surfaces for each hemisphere separately, resulting in 4 surfaces of 40 962 vertices each.[37,38] From these surfaces, the t-link metric was derived for determining the distance between the white and gray surfaces.[39] The thickness data were subsequently blurred using a 20-mm surface–based diffusion blurring kernel in preparation for statistical analyses. Unnormalized, native-space thickness values were used in all analyses owing to the poor correlation between cortical thickness and brain volume. Normalizing for global brain size when it has little pertinence to cortical thickness risks introducing noise and reducing power[40] (**Figure 1**).

DTI Image Analysis, Whole-Brain Tractography, and Clustering Segmentation. The 3 repetitions were coregistered to the first $B=0$ image in the first repetition using the Functional Magnetic Resonance Imaging of the Brain Software Library (version 4.0; Functional Magnetic Resonance Imaging of the Brain Centre, University of Oxford, Department of Clinical Neurology, Oxford, England; www.fmrib.ox.ac.uk) to produce a new averaged image, with gradients reoriented using a weighted least squares approach. Registration corrects eddy current distortions and subject motion, important artifacts that can affect the data, and averaging improves the signal to noise ratio. A brain mask was then generated. Points were seeded throughout each voxel of the brain. Whole-brain tractography was performed with a deterministic (streamline) approach (Runge-Kutta order 2 tractography with a fixed step size of 0.5 mm). More detailed descriptions of our tractography approach and our clustering segmentation algorithm have been recently published[41,42] and are summarized here. Threshold parameters for tractography were based on the linear anisotropy measure C_L, which provides specific advantages compared with thresholding using fractional anisotrophy.[43,44] The parameters chosen for this study were as follows: T_{seed}, $C_L=0.3$; T_{stop}, 0.15; and T_{length}, 20 mm. Tractography and creation of white matter fiber tracts were performed using the 3D Slicer (www.slicer.org) and MATLAB 7.0 (The Mathworks Inc, Natwick, Massachusetts; www.mathworks.com).

A pairwise fiber trajectory similarity was quantified and the directed distances between fibers A and B were converted to a symmetric pairwise fiber distance. A spectral embedding of fibers was then created based on the eigenvectors of the fiber affinity matrix, and shape similarity information for each fiber was calculated using a *k*-way normalized cuts clustering alogorithm.[41]

Once the whole brain cluster model was produced, a trained operator (A.N.V.) combined clusters corresponding to a given fiber tract. Left and right association fiber tracts connecting to the temporal lobe were selected[42]: uncinate fasciculus, inferior occipitofrontal fasciculus, cingulum bundle, inferior longitudinal fasciculus, and arcuate fasciculus. The genu of the corpus callosum was selected for comparative purposes because this structure is highly susceptible to age-related fractional anistrophy (FA) change in healthy aging populations[45] and is not preferentially disrupted at the earliest stages of AD[19,46] (**Figure 2**) (although it may be affected in later stages of AD[25,47]). As reported elsewhere,[42] excellent spatial and quantitative reliability using this clustering method (ie, voxel overlap and scalar measures of the tensor showed high agreement) has been demonstrated. For each white matter tract, MATLAB (version 7.0) was used to calculate a mean FA[48] value along the selected tract.

GENETICS

The *BDNF* Val66Met polymorphism (rs6265) was genotyped in each study participant. This polymorphism lies in the 5′ region of the *BDNF* gene and affects intracellular packaging and secretion of *BDNF*.[12] Genotyping of this polymorphism was performed using a standard (Applied Biosystems Inc, Foster City,

©2011 American Medical Association. All rights reserved.
Downloaded From: http://archpsyc.jamanetwork.com/ on 05/24/2016

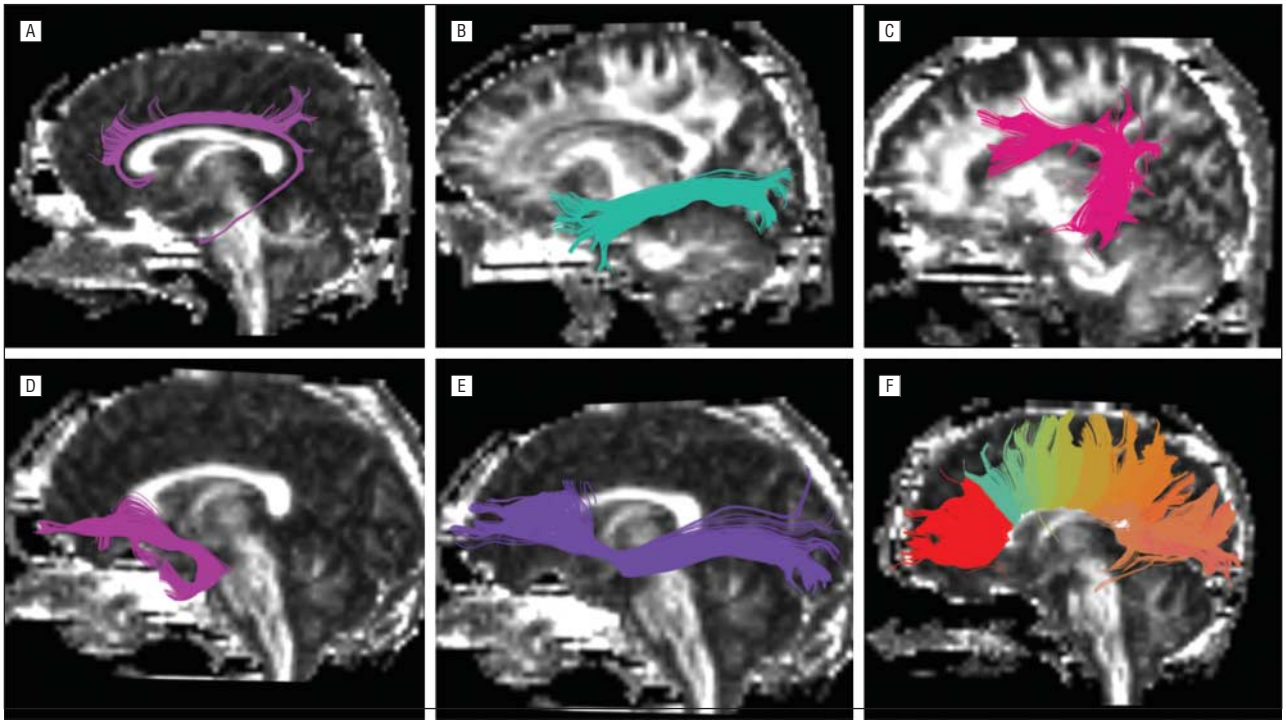

Figure 2. Models of white matter tracts measured. A, Left cingulum bundle; B, left inferior longitudinal fasciculus; C, left arcuate fasciculus; D, left uncinate fasciculus; E, left inferior occipitofrontal fasciculus; F, genu of corpus callosum (in red).

California) 5′ nuclease TaqMan assay-on-demand protocol in a total volume of 10 µL. Postamplification products were analyzed on the ABI 7500 Sequence Detection System (Applied Biosystems), and genotype calls were performed manually. Results were verified independently by 2 laboratory personnel masked to demographic and phenotypic information. Quality control analysis was performed on 10.0% of the sample. All participants also underwent genotyping at the apolipoprotein E (*APOE*) gene to determine *APOE4* allele status. Apolipoprotein E was genotyped by combining allelic results from 2 single-nucleotide polymorphism assays (also assay-on-demand protocol) for rs429358 (T/C) and rs7412 (T/C). The combination of these 2 polymorphisms result in cysteine-to-arginine amino acid substitutions in *APOE* at positions 130 and 176. The *E2* allele is represented by the Cys-Cys combination, *E3* by the Cys130-Arg176 combination, and *E4* by the Arg-Arg combination.

COGNITIVE MEASURES

Sixty-five of the study participants completed cognitive testing that included the Repeatable Battery for the Assessment of Neuropsychological Status (RBANS). Verbal episodic memory performance and visuospatial episodic memory performance were measured using the list recall and figure recall tests of the RBANS, respectively.

STATISTICAL ANALYSIS

Three separate analyses were performed according to the general linear model to examine the effects of the *BDNF* gene and age on (1) cortical thickness, (2) white matter tract integrity, and (3) cognitive performance. Two genotypic groups were created: Met allele carriers and Val/Val individuals. Genotypic group served as the between-group factor in each model.

The first model examined an analysis of covariance (ANCOVA) relating *BDNF* genotype and age to cortical thickness. Statistical thresholds were determined by application of a 5% false discovery rate correction, where $q<0.05$ was considered significant.[49]

The second model used a repeated-measures ANCOVA with *BDNF* genotype group as the between-group factor and age as the covariate to examine white matter tract FA (all tract FA values were within-group measures) of association fiber tracts and of the genu of the corpus callosum. For episodic memory performance, a repeated-measures ANCOVA was conducted with *BDNF* genotype group as the between-group factor and age as the covariate. Scores on the list recall and figure recall tests of the RBANS were the 2 within-group measures. Because recent evidence suggests that risk conferred by the *BDNF* Val66Met for AD may be dependent on sex,[50] we conducted a separate analysis on brain measures and cognitive performance stratified for sex.

RESULTS

The 2 genotypic groups did not differ in terms of age, sex, IQ, years of education, ethnicity, socioeconomic status, systolic blood pressure, diastolic blood pressure, or body mass index (Table). Of the 69 healthy volunteers, there were 28 Met allele carriers (including 5 Met homozygotes), and 41 individuals who were Val/Val homozygotes ($\chi^2_1=0.34$; $P=.56$). Of the Met allele carriers, 3 were *APOE4* allele carriers, and of the Val/Val individuals, 9 were *APOE4* allele carriers. A 100% genotyping success rate was achieved. The sample did not deviate from Hardy-Weinberg equilibrium ($\chi^2_1=0.487$, 2-tailed $P=.48$). No individual had 2 *APOE4* alleles, and 12 individuals were carriers of 1 *APOE4* allele.

A *BDNF* genotype by age interaction predicted cortical thickness at several regions in the temporal lobe, with large effect sizes, prominently at the entorhinal cortex ($F_{1,65}=12.5$, $q=0.03$, partial $\eta^2=0.15$) and inferior temporal gyrus ($F_{1,65}=13.9$, $q=0.016$, partial $\eta^2=0.18$) after false discov-

©2011 American Medical Association. All rights reserved.
Downloaded From: http://archpsyc.jamanetwork.com/ on 05/24/2016

ery rate correction (**Figure 3**A). Cortical thickness at the middle temporal gyrus and superior temporal gyrus, along with the parietooccipital sulcus, also met the false discovery rate threshold for the *BDNF* genotype by age interaction (eAppendix and eTable 1; available at http://www.archgenpsychiatry.com). No main effects of the *BDNF* genotype but significant effects of age were present (all $q < 0.05$, except for right inferior temporal gyrus).

For white matter tract integrity, a significant *BDNF* genotype by age interaction ($F_{1,65}=14.0$, $P<.001$) and main effects of genotype and age ($F_{1,65}=9.0$, $P=.004$, and $F_{1,65}=53.7$, $P<.001$, respectively) were seen. Because the overall model for white matter integrity was statistically significant, follow-up univariate ANCOVAs were used with a Bonferroni corrected threshold *P* value for 11 comparisons at $P=.004$. The interaction was most notable, with large effect sizes, at the left cingulum bundle ($F_{1,65}=10.8$, $P=.002$, partial $\eta^2=0.14$) (Figure 3B) and left inferior longitudinal fasciculus ($F_{1,65}=10.2$, $P=.002$, partial $\eta^2=0.14$), which are white matter tracts connecting to the medial temporal lobe, and the left arcuate fasciculus ($F_{1,65}=10.0$, $P=.002$, partial $\eta^2=0.13$), a white matter tract with temporoparietal and temporofrontal fibers. There was no significant interaction between *BDNF* genotype and the integrity of any of the other white matter tracts studied. In particular, there was no interaction for the genu of the corpus callosum, the white matter tract typically most vulnerable in healthy aging studies ($F_{1,65}=4.0$, $P=.05$) (eTable 2).

Finally, for episodic memory performance, a *BDNF* genotype by age interaction ($F_{1,61}=6.2$, $P=.02$) (Figure 3C) and main effects of *BDNF* genotype and age ($F_{1,61}=4.5$, $P=.04$, and $F_{1,61}=18.6$, $P<.001$, respectively) were also present. Because the overall model for episodic memory was statistically significant, follow-up univariate ANCOVAs were used with a Bonferroni-corrected *P* value for 2 comparisons, at threshold $P=.02$, to investigate each episodic memory task separately. The *BDNF* genotype by age interaction revealed only small to modest effect sizes for visuospatial episodic memory performance ($F_{1,61}=4.7$, $P=.03$, partial $\eta^2=0.07$) and verbal episodic memory performance ($F_{1,61}=3.2$, $P=.08$, partial $\eta^2=0.05$) (eTable 2). Cortical thickness, white matter tract integrity, and episodic memory performance results remained significant after reanalysis of the data without the 2 participants of Asian ethnicity or the 12 *APOE4* carriers.

After stratification of our analyses for sex, similar patterns in men and women, as in the overall analysis, were observed for Met allele carriers and for Val/Val individuals, in which Met allele carriers were at risk in earlier life and Val/Val individuals in later life for reduced cortical thickness, white matter integrity, and episodic memory performance.

COMMENT

We found that the *BDNF* Val66Met polymorphism interacts with age in a biologically convergent manner to predict variation in at-risk neural structures and cognitive functions of AD in healthy humans. Our findings support *BDNF* as a genetic susceptibility mechanism in an intermediate phenotype related to AD via its effect on thickness of temporal lobe structures, including the entorhinal cortex,[10,18] white matter integrity of association fiber tracts connecting to the medial temporal lobe,[19,20,51] and episodic memory formation.[52]

Multiple lines of evidence implicate *BDNF* in the AD process: *BDNF* expression is reduced in the hippocampus and the entorhinal cortex in AD[9]; neurons containing neurofibrillary tangles, the hallmark finding of AD, do not have detectable levels of *BDNF* immunoreactive material, whereas neurons more intensely labeled with *BDNF*-specific antibodies are free of tangles[53]; and altered levels of *BDNF* in serum and cerebrospinal fluid have been found in AD in vivo[54,55] and have been associated with disease severity and episodic memory performance. Furthermore, recent data suggest potentially substantial effects of *BDNF* as a therapeutic agent: hippocampal neural stem cell transplantation restored spatial learning and memory deficits in aged triple transgenic mice, expressing pathogenic forms of amyloid precursor protein, presenilin, and tau, without altering Aβ or tau pathologic findings,[56] but rather mediated via *BDNF*. In another study,[15] *BDNF* gene delivery to the entorhinal cortex in amyloid transgenic mice reversed synaptic loss, improved cell signaling, and restored learning and memory without altering amyloid plaque load. Therefore, *BDNF* can exert substantial protective effects on crucial neuronal circuitry in AD by acting through amyloid-independent mechanisms.

Considerable evidence implicates *BDNF* in AD. However, results of genetic association studies examining the *BDNF* gene with AD have not been consistently replicated. Early genetic studies of the *BDNF* Val66Met polymorphism demonstrated that the Val/Val genotype was associated with AD.[14] Prospective data from the large Lothian Birth Cohort demonstrated that *BDNF* Val/Val individuals in late life experience a greater age-related decline in reasoning skills than Met carriers.[13] However, these findings have not been consistently replicated.[57,58] Such difficulties in genetic association studies of complex disorders have been well characterized, and a number of explanations have been put forward.[59,60] One challenge may be that the rate-limiting step in gene identification in complex behavioral disorders can be the effect size of the risk allele on phenotypic variance.[16]

Imaging genetics offers an alternative strategy to conventional genetic association studies by delineating neural systems that are affected by genetic variation via the intermediate phenotype strategy.[16] Genotype to brain phenotype associations can be shown in carriers of risk alleles even if the carriers do not exhibit the clinical phenotype. Our findings are most robust at the level of brain structure and least robust at the level of observable behavior (ie, cognition), consistent with the intermediate phenotype concept. Importantly, *BDNF* variation is not related in our study to structures prominently affected in healthy aging, namely, frontal gray matter[61] or white matter tracts that are frontally based, such as the genu of the corpus callosum.[45] Rather, the structures affected by *BDNF* in our healthy study participants are the structures affected in the preclinical and earliest clinical stages of AD. In gray matter, medial and then lateral temporal areas are affected first, before extending to cingulate cortex and temporoparietal regions[62]; in white matter, corticocortical association path-

©2011 American Medical Association. All rights reserved.
Downloaded From: http://archpsyc.jamanetwork.com/ on 05/24/2016

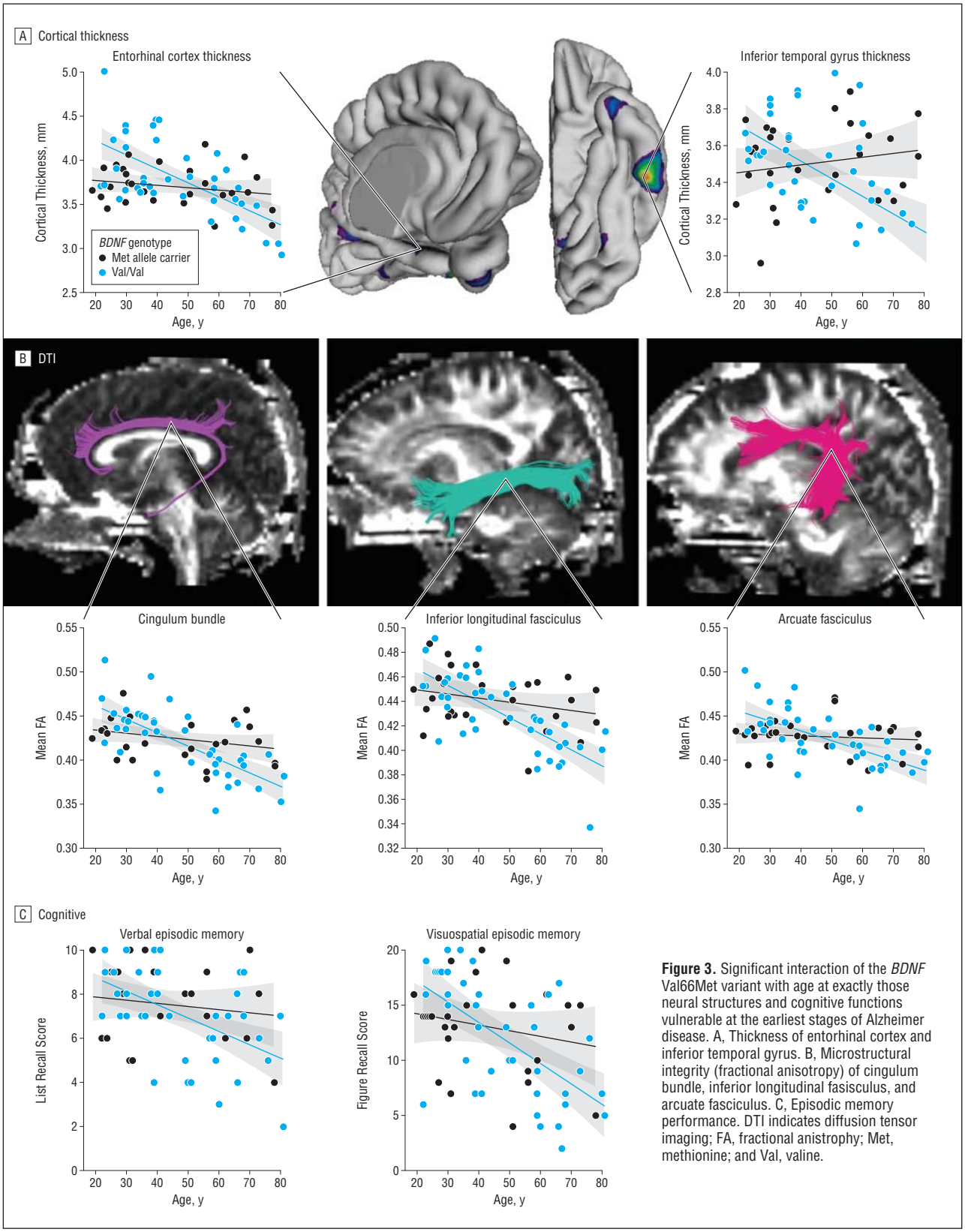

Figure 3. Significant interaction of the *BDNF* Val66Met variant with age at exactly those neural structures and cognitive functions vulnerable at the earliest stages of Alzheimer disease. A, Thickness of entorhinal cortex and inferior temporal gyrus. B, Microstructural integrity (fractional anisotropy) of cingulum bundle, inferior longitudinal fasisculus, and arcuate fasciculus. C, Episodic memory performance. DTI indicates diffusion tensor imaging; FA, fractional anistrophy; Met, methionine; and Val, valine.

ways (eg, cingulum bundle, inferior longitudinal fasciculus, and arcuate fasciculus), the latest-myelinating fiber pathways in the brain, are affected earliest in AD[22]; cognitively, episodic memory performance is also affected in preclinical and the earliest clinical stages of AD. Intermediate phenotypes in other neuropsychiatric disorders, such as schizophrenia[63] and depression,[64] have been previously characterized using similar approaches.

©2011 American Medical Association. All rights reserved.
Downloaded From: http://archpsyc.jamanetwork.com/ on 05/24/2016

Unlike the *APOE* gene, for which *APOE4* allele carriers are at a disadvantage even in early adult life,[65] the direction of the effects of the *BDNF* Val66Met on brain structure and cognitive function found in our study differ in an age-dependent manner. Previous investigations in young healthy individuals suggest that *BDNF* Met allele carriers demonstrate reduced hippocampal/parahippocampal complex volumes, function, and episodic memory performance.[12,66] One explanation for Met allele vulnerability in early adult life is based on findings that the Met allele may fail to localize *BDNF* to secretory granules or synapses,[12] altering activity-dependent processes of cortical development and plasticity in the process. Our results support these findings and add new evidence based on cortical thickness and tractography measures: young *BDNF* Met allele carriers are more likely to have reduced cortical thickness of medial and lateral temporal lobe structures and reduced microstructural integrity of white matter tracts connecting to medial and lateral temporal lobe regions.

In contrast to our findings in early adult life, Val/Val individuals in late adult life had diminished entorhinal cortex thickness, white matter tract integrity, and episodic memory performance. Here, our findings also align with previous findings, where others have shown that Val/Val individuals are at increased risk in later life for poor cognitive performance[13] and AD.[14] The substantial literature implicating *BDNF* in the pathophysiology of mood disorders provides an intriguing genetic mechanism for an overlapping clinical picture with AD. First, a history of depressive mood episodes is a risk factor for subsequent AD.[67] Second, the *BDNF* Val/66Met polymorphism has been associated with risk for mood disorders[68] and for neuroticism,[69] although the manner in which risk is determined (ie, Met carrier vs Val/Val) is under debate. A recent investigation highlighted the complexity of risk determined by the *BDNF* Val66Met on physiologic measures of depression and anxiety.[70] It is possible, therefore, that there is a lifetime burden with a Val/Val genotype, whereby effects of mood vulnerability, highly sensitive plasticity (eg, high stress sensitivity), or reduced resilience contribute to the intermediate phenotype found in our study.

One limitation of our study is its cross-sectional design. Specifically, we are only able to conclude that Val/Val individuals in late life and Met allele carriers in early adult life may be at a disadvantage, given the phenotypic measures used. A longitudinal study would have allowed us to examine progression across adult life of our phenotypic measures according to the *BDNF* genotype. However, such a study design carries its own set of challenges, including technical limitations of repeated imaging measures, attrition, and cost. Despite the cross-sectional nature of our sample, our finding is unlikely to be due to a sampling bias or cohort effect because our elderly individuals were not different from our younger individuals for IQ or educational levels. Furthermore, conclusions regarding AD severity, outcome, or treatment response, in relation to potential effects of the *BDNF* Val66Met polymorphism on brain and cognitive measures, cannot be drawn because AD patients were not included in the present study. Although we screened for dementia using the Mini-Mental State Examination, it is possible, given low scores on episodic memory testing, that 2 individuals in our study had mild cognitive impairment, and this might be considered a limitation of our study. Another limitation is that we did not include the fornix of the hippocampus, a commissural white matter tract, for study in our sample because of challenges in achieving high reliability[42] using streamline tractography for the fornix. Investigation of this fiber tract in relation to the *BDNF* genotype would be useful because the fornix is an important part of the hippocampal system, may be involved in learning and memory, and may be disrupted in AD.[51] Finally, although mean systolic and diastolic blood pressure results for our sample indicated that our participants were not characterized by high blood pressure as a group, 7 individuals had blood pressure results that fall within the range of stage I hypertension (as defined by the American Heart Association[71]). This could be considered a limitation of our study because hypertension has been associated with lower white matter integrity.[72]

Although others have investigated the effects of the *BDNF* gene on brain structure in healthy individuals across the adult lifespan,[73,74] none, to our knowledge, has investigated the effects of *BDNF* on cortical thickness or association fiber tracts as intermediate phenotype measures. The convergent pattern of our findings across gray matter, white matter, and cognitive performance provide a more convincing picture of the effect of the *BDNF* Val66Met polymorphism on an intermediate phenotype related to AD than any one of these findings alone. Our findings suggest that the *BDNF* gene may be a susceptibility mechanism for AD and highlight a critical alternative pathway in this neurodegenerative disorder.

Submitted for Publication: April 14, 2010; final revision received July 19, 2010; accepted August 10, 2010.
Author Affiliations: Department of Psychiatry, Geriatric Mental Health Program (Drs Voineskos, Rajji, Mulsant, and Pollock and Ms Miranda) and Department of Psychiatry and Neurogenetics Section and Department of Neuroscience, Centre for Addiction and Mental Health (Drs Voineskos and Kennedy and Messrs Felsky and Shaikh), and Department of Biomedical Imaging, Toronto Centre for Phenogenomics and Hospital for Sick Children (Dr Lerch), and Department of Cognitive Neurology, Sunnybrook Health Sciences Centre, Department of Medicine (Dr Lobaugh), and Rotman Research Institute, Baycrest Hospital, and Department of Psychiatry (Dr Pollock), University of Toronto, Toronto, Ontario, Canada; and Department of Psychiatry, University of Pittsburgh, Pittsburgh, Pennsylvania (Dr Mulsant).
Correspondence: Aristotle N. Voineskos, MD, PhD, FRCPC, Department of Psychiatry, Geriatric Mental Health Program, Centre for Addiction and Mental Health, 250 College St, Toronto, Ontario, Canada M5T 1R8 (aristotle_voineskos@camh.net).
Financial Disclosure: Dr Pollock receives research support from the National Institutes of Health and the Canadian Institutes of Health Research. Within the past 2 years, he has been a member of the advisory board of Lundbeck Canada (final meeting was May 15, 2009) and has served 1 time as a consultant for Wyeth Pharmaceuticals (October 4-5, 2008). He is currently a faculty member of the Lundbeck International Neuroscience Foundation. Dr Mulsant currently receives research support from the US National Institute of Mental Health, the Ca-

©2011 American Medical Association. All rights reserved.
Downloaded From: http://archpsyc.jamanetwork.com/ on 05/24/2016

nadian Institutes of Health Research, Bristol-Myers Squibb, and Wyeth Pharmaceuticals. During the past 5 years, he has also received research support or honoraria from AstraZeneca Pharmaceuticals, Eli Lilly and Company, Forest Laboratories Inc, GlaxoSmithKline PLC, Janssen, H. Lundbeck A/S, and Pfizer Inc.

Funding/Support: This work was supported by the Canadian Institutes of Health Research Clinician Scientist Award (Dr Voineskos), American Psychiatric Association/American Psychiatric Institute for Research and Education AstraZeneca Young Minds in Psychiatry Award (Dr Voineskos), Canadian Institutes of Health Research Fellowship (Dr Rajji), the Sandra A. Rotman Program of the Rotman Research Institute (Dr Pollock), and the Centre for Addiction and Mental Health.

Online-Only Material: The eTables are available at http://www.archgenpsychiatry.com.

Additional Information: Drs Voineskos and Lerch contributed equally to the manuscript. Drs Pollock and Kennedy are senior coauthors.

Additional Contributions: Tiffany Chow, MD, and Sandra Moses, PhD, provided thoughtful comments and suggestions in the writing of the manuscript and Faranak Farzan, PhD, contributed help.

REFERENCES

1. Harman D. Alzheimer's disease pathogenesis: role of aging. *Ann N Y Acad Sci*. 2006;1067:454-460.
2. Corder EH, Saunders AM, Strittmatter WJ, Schmechel DE, Gaskell PC, Small GW, Roses AD, Haines JL, Pericak-Vance MA. Gene dose of apolipoprotein E type 4 allele and the risk of Alzheimer's disease in late onset families. *Science*. 1993; 261(5123):921-923.
3. Harold D, Abraham R, Hollingworth P, Sims R, Gerrish A, Hamshere ML, Pahwa JS, Moskvina V, Dowzell K, Williams A, Jones N, Thomas C, Stretton A, Morgan AR, Lovestone S, Powell J, Proitsi P, Lupton MK, Brayne C, Rubinsztein DC, Gill M, Lawlor B, Lynch A, Morgan K, Brown KS, Passmore PA, Craig D, McGuinness B, Todd S, Holmes C, Mann D, Smith AD, Love S, Kehoe PG, Hardy J, Mead S, Fox N, Rossor M, Collinge J, Maier W, Jessen F, Schürmann B, van den Bussche H, Heuser I, Kornhuber J, Wiltfang J, Dichgans M, Frölich L, Hampel H, Hüll M, Rujescu D, Goate AM, Kauwe JSK, Cruchaga C, Nowotny P, Morris JC, Mayo K, Sleegers K, Bettens K, Engelborghs S, De Deyn PP, Van Broeckhoven C, Livingston G, Bass NJ, Gurling H, McQuillin A, Gwilliam R, Deloukas P, Al-Chalabi A, Shaw CE, Tsolaki M, Singleton AB, Guerreiro R, Mühleisen TW, Nöthen MM, Moebus S, Jöckel K-H, Klopp N, Wichmann H-E, Carrasquillo MM, Pankratz VS, Younkin SG, Holmans PA, O'Donovan M, Owen MJ, Williams J. Genome-wide association study identifies variants at *CLU* and *PICALM* associated with Alzheimer's disease. *Nat Genet*. 2009;41(10):1088-1093.
4. Lambert J-C, Heath S, Even G, Campion D, Sleegers K, Hiltunen M, Combarros O, Zelenika D, Bullido MJ, Tavernier B, Letenneur L, Bettens K, Berr C, Pasquier F, Fiévet N, Barberger-Gateau P, Engelborghs S, De Deyn P, Mateo I, Franck A, Helisalmi S, Porcellini E, Hanon O, de Pancorbo MM, Lendon C, Dufouil C, Jaillard C, Leveillard T, Alvarez V, Bosco P, Mancuso M, Panza F, Nacmias B, Bossù P, Piccardi P, Annoni G, Seripa D, Galimberti D, Hannequin D, Licastro F, Soininen H, Ritchie K, Blanché H, Dartigues J-F, Tzourio C, Gut I, Van Broeckhoven C, Alpérovitch A, Lathrop M, Amouyel P; European Alzheimer's Disease Initiative Investigators. Genome-wide association study identifies variants at *CLU* and *CR1* associated with Alzheimer's disease. *Nat Genet*. 2009;41(10):1094-1099.
5. Kennedy JL, Farrer LA, Andreasen NC, Mayeux R, St George–Hyslop P. The genetics of adult-onset neuropsychiatric disease: complexities and conundra? *Science*. 2003;302(5646):822-826.
6. Zuccato C, Cattaneo E. Brain-derived neurotrophic factor in neurodegenerative diseases. *Nat Rev Neurol*. 2009;5(6):311-322.
7. Figurov A, Pozzo-Miller LD, Olafsson P, Wang T, Lu B. Regulation of synaptic responses to high-frequency stimulation and LTP by neurotrophins in the hippocampus. *Nature*. 1996;381(6584):706-709.
8. Tapia-Arancibia L, Aliaga E, Silhol M, Arancibia S. New insights into brain BDNF function in normal aging and Alzheimer disease. *Brain Res Rev*. 2008;59(1): 201-220.
9. Narisawa-Saito M, Wakabayashi K, Tsuji S, Takahashi H, Nawa H. Regional specificity of alterations in NGF, BDNF and NT-3 levels in Alzheimer's disease. *Neuroreport*. 1996;7(18):2925-2928.
10. Gómez-Isla T, Price JL, McKeel DW Jr, Morris JC, Growdon JH, Hyman BT. Profound loss of layer II entorhinal cortex neurons occurs in very mild Alzheimer's disease. *J Neurosci*. 1996;16(14):4491-4500.
11. Price JL, Ko AI, Wade MJ, Tsou SK, McKeel DW, Morris JC. Neuron number in the entorhinal cortex and CA1 in preclinical Alzheimer disease. *Arch Neurol*. 2001; 58(9):1395-1402.
12. Egan MF, Kojima M, Callicott JH, Goldberg TE, Kolachana BS, Bertolino A, Zaitsev E, Gold B, Goldman D, Dean M, Lu B, Weinberger DR. The BDNF val66met polymorphism affects activity-dependent secretion of BDNF and human memory and hippocampal function. *Cell*. 2003;112(2):257-269.
13. Harris SE, Fox H, Wright AF, Hayward C, Starr JM, Whalley LJ, Deary IJ. The brain-derived neurotrophic factor Val66Met polymorphism is associated with age-related change in reasoning skills. *Mol Psychiatry*. 2006;11(5):505-513.
14. Ventriglia M, Bocchio Chiavetto L, Benussi L, Binetti G, Zanetti O, Riva MA, Gennarelli M. Association between the BDNF 196 A/G polymorphism and sporadic Alzheimer's disease. *Mol Psychiatry*. 2002;7(2):136-137.
15. Nagahara AH, Merrill DA, Coppola G, Tsukada S, Schroeder BE, Shaked GM, Wang L, Blesch A, Kim A, Conner JM, Rockenstein E, Chao MV, Koo EH, Geschwind D, Masliah E, Chiba AA, Tuszynski MH. Neuroprotective effects of brain-derived neurotrophic factor in rodent and primate models of Alzheimer's disease. *Nat Med*. 2009;15(3):331-337.
16. Meyer-Lindenberg A, Weinberger DR. Intermediate phenotypes and genetic mechanisms of psychiatric disorders. *Nat Rev Neurosci*. 2006;7(10):818-827.
17. Lerch JP, Pruessner JC, Zijdenbos A, Hampel H, Teipel SJ, Evans AC. Focal decline of cortical thickness in Alzheimer's disease identified by computational neuroanatomy. *Cereb Cortex*. 2005;15(7):995-1001.
18. Desikan RS, Cabral HJ, Hess CP, Dillon WP, Glastonbury CM, Weiner MW, Schmansky NJ, Greve DN, Salat DH, Buckner RL, Fischl B; Alzheimer's Disease Neuroimaging Initiative. Automated MRI measures identify individuals with mild cognitive impairment and Alzheimer's disease. *Brain*. 2009;132(pt 8):2048-2057.
19. Zhang Y, Schuff N, Jahng G-H, Bayne W, Mori S, Schad L, Mueller S, Du A-T, Kramer JH, Yaffe K, Chui H, Jagust WJ, Miller BL, Weiner MW. Diffusion tensor imaging of cingulum fibers in mild cognitive impairment and Alzheimer disease. *Neurology*. 2007;68(1):13-19.
20. Damoiseaux JS, Smith SM, Witter MP, Sanz-Arigita EJ, Barkhof F, Scheltens P, Stam CJ, Zarei M, Rombouts SA. White matter tract integrity in aging and Alzheimer's disease. *Hum Brain Mapp*. 2009;30(4):1051-1059.
21. Alexander A, Lobaugh N. Insights into brain connectivity using quantiative MRI measures of white matter. In: Jirsa VK, McIntosh AR, eds. *Handbook of Brain Connectivity*. Berlin, Germany: Springer Verlag; 2007.
22. Stricker NH, Schweinsburg BC, Delano-Wood L, Wierenga CE, Bangen KJ, Haaland KY, Frank LR, Salmon DP, Bondi MW. Decreased white matter integrity in late-myelinating fiber pathways in Alzheimer's disease supports retrogenesis. *Neuroimage*. 2009;45(1):10-16.
23. Villain N, Desgranges B, Viader F, de la Sayette V, Mézenge F, Landeau B, Baron JC, Eustache F, Chételat G. Relationships between hippocampal atrophy, white matter disruption, and gray matter hypometabolism in Alzheimer's disease. *J Neurosci*. 2008;28(24):6174-6181.
24. Chételat G, Desgranges B, de la Sayette V, Viader F, Eustache F, Baron JC. Mild cognitive impairment. *Neurology*. 2003;60(8):1374-1377.
25. Bartzokis G, Sultzer D, Lu PH, Nuechterlein KH, Mintz J, Cummings JL. Heterogeneous age-related breakdown of white matter structural integrity. *Neurobiol Aging*. 2004;25(7):843-851.
26. Brun A, Englund E. A white matter disorder in dementia of the Alzheimer type: a pathoanatomical study. *Ann Neurol*. 1986;19(3):253-262.
27. Ng BK, Chen L, Mandemakers W, Cosgaya JM, Chan JR. Anterograde transport and secretion of brain-derived neurotrophic factor along sensory axons promote Schwann cell myelination. *J Neurosci*. 2007;27(28):7597-7603.
28. Djalali S, Höltje M, Grosse G, Rothe T, Stroh T, Grosse J, Deng DR, Hellweg R, Grantyn R, Hörtnagl H, Ahnert-Hilger G. Effects of brain-derived neurotrophic factor (BDNF) on glial cells and serotonergic neurones during development. *J Neurochem*. 2005;92(3):616-627.
29. Oldfield RC. The assessment and analysis of handedness: the Edinburgh inventory. *Neuropsychologia*. 1971;9(1):97-113.
30. First MB Sr, Gibbon M, Williams JBW. *Structured Clinical Interview for DSM-IV Axis I Disorders, Patient Edition (SCID-P)*. New York, NY: Biometrics Research; 1995.
31. Folstein MF, Folstein SE, McHugh PR. "Mini-Mental State". *J Psychiatr Res*. 1975; 12(3):189-198.
32. Hollingshead AB. *Four-Factor Index of Social Status*. New Haven, CT: Yale University Press; 1975.

©2011 American Medical Association. All rights reserved.
Downloaded From: http://archpsyc.jamanetwork.com/ on 05/24/2016

33. Miller MD, Paradis CF, Houck PR, Mazumdar S, Stack JA, Rifai AH, Mulsant B, Reynolds CF III. Rating chronic medical illness burden in geropsychiatric practice and research. *Psychiatry Res*. 1992;41(3):237-248.
34. Sled JG, Zijdenbos AP, Evans AC. A nonparametric method for automatic correction of intensity nonuniformity in MRI data. *IEEE Trans Med Imaging*. 1998; 17(1):87-97.
35. Zijdenbos AP, Forghani R, Evans AC. Automatic "pipeline" analysis of 3-D MRI data for clinical trials. *IEEE Trans Med Imaging*. 2002;21(10):1280-1291.
36. Tohka J, Zijdenbos A, Evans A. Fast and robust parameter estimation for statistical partial volume models in brain MRI. *Neuroimage*. 2004;23(1):84-97.
37. Kim JS, Singh V, Lee JK, Lerch J, Ad-Dab'bagh Y, MacDonald D, Lee JM, Kim SI, Evans AC. Automated 3-D extraction and evaluation of the inner and outer cortical surfaces using a Laplacian map and partial volume effect classification. *Neuroimage*. 2005;27(1):210-221.
38. MacDonald D, Kabani N, Avis D, Evans AC. Automated 3-D extraction of inner and outer surfaces of cerebral cortex from MRI. *Neuroimage*. 2000;12(3):340-356.
39. Lerch JP, Evans AC. Cortical thickness analysis examined through power analysis and a population simulation. *Neuroimage*. 2005;24(1):163-173.
40. Ad-Dab'bagh Y, Singh V, Robbins S, Lerch J, Lyttleton O, Fombonne E, Evans AC. The CIVET image-processing environment: a fully automated comprehensive pipeline for anatomical neuroimaging research. In: Corbetta M, ed. *Proceedings of the 12th Annual Meeting of the Organization for Human Brain Mapping*. Florence, Italy: NeuroImage; 2006.
41. O'Donnell LJ, Kubicki M, Shenton ME, Dreusicke MH, Grimson WEL, Westin CF. A method for clustering white matter fiber tracts. *AJNR Am J Neuroradiol*. 2006; 27(5):1032-1036.
42. Voineskos AN, O'Donnell LJ, Lobaugh NJ, Markant D, Ameis SH, Niethammer M, Mulsant BH, Pollock BG, Kennedy JL, Westin CF, Shenton ME. Quantitative examination of a novel clustering method using magnetic resonance diffusion tensor tractography. *Neuroimage*. 2009;45(2):370-376.
43. Ennis DB, Kindlmann G. Orthogonal tensor invariants and the analysis of diffusion tensor magnetic resonance images. *Magn Reson Med*. 2006;55(1):136-146.
44. Westin C-F, Maier SE, Mamata H, Nabavi A, Jolesz FA, Kikinis R. Processing and visualization for diffusion tensor MRI. *Med Image Anal*. 2002;6(2):93-108.
45. Voineskos AN, Rajji TK, Lobaugh NJ, Miranda D, Shenton ME, Kennedy JL, Pollock BG, Mulsant BH. Age-related decline in white matter tract integrity and cognitive performance: a DTI tractography and structural equation modeling study. *Neurobiol Aging*. 2010.
46. Head D, Buckner RL, Shimony JS, Williams LE, Akbudak E, Conturo TE, McAvoy M, Morris JC, Snyder AZ. Differential vulnerability of anterior white matter in nondemented aging with minimal acceleration in dementia of the Alzheimer type: evidence from diffusion tensor imaging. *Cereb Cortex*. 2004;14(4):410-423.
47. Bartzokis G, Lu PH, Mintz J. Quantifying age-related myelin breakdown with MRI. *J Alzheimers Dis*. 2004;6(6)(suppl):S53-S59.
48. Basser PJ, Pierpaoli C. Microstructural and physiological features of tissues elucidated by quantitative-diffusion-tensor MRI. *J Magn Reson B*. 1996;111(3): 209-219.
49. Genovese CR, Lazar NA, Nichols T. Thresholding of statistical maps in functional neuroimaging using the false discovery rate. *Neuroimage*. 2002;15(4): 870-878.
50. Fukumoto N, Fujii T, Combarros O, Kamboh MI, Tsai S-J, Matsushita S, Nacmias B, Comings DE, Arboleda H, Ingelsson M, Hyman BT, Akatsu H, Grupe A, Nishimura AL, Zatz M, Mattila KM, Rinne J, Goto Y, Asada T, Nakamura S, Kunugi H. Sexually dimorphic effect of the Val66Met polymorphism of *BDNF* on susceptibility to Alzheimer's disease: new data and meta-analysis. *Am J Med Genet B Neuropsychiatr Genet*. 2010;153B(1):235-242.
51. Mielke MM, Kozauer NA, Chan KCG, George M, Toroney J, Zerrate M, Bandeen-Roche K, Wang M-C, Vanzijl P, Pekar JJ, Mori S, Lyketsos CG, Albert M. Regionally-specific diffusion tensor imaging in mild cognitive impairment and Alzheimer's disease. *Neuroimage*. 2009;46(1):47-55.
52. Stoub TR, deToledo-Morrell L, Stebbins GT, Leurgans S, Bennett DA, Shah RC. Hippocampal disconnection contributes to memory dysfunction in individuals at risk for Alzheimer's disease. *Proc Natl Acad Sci U S A*. 2006;103(26):10041-10045.
53. Murer MG, Boissiere F, Yan Q, Hunot S, Villares J, Faucheux B, Agid Y, Hirsch E, Raisman-Vozari R. An immunohistochemical study of the distribution of brain-derived neurotrophic factor in the adult human brain, with particular reference to Alzheimer's disease. *Neuroscience*. 1999;88(4):1015-1032.
54. Peng S, Wuu J, Mufson EJ, Fahnestock M. Precursor form of brain-derived neurotrophic factor and mature brain-derived neurotrophic factor are decreased in the pre-clinical stages of Alzheimer's disease. *J Neurochem*. 2005;93(6):1412-1421.
55. Laske C, Stransky E, Leyhe T, Eschweiler GW, Wittorf A, Richartz E, Bartels M, Buchkremer G, Schott K. Stage-dependent BDNF serum concentrations in Alzheimer's disease. *J Neural Transm*. 2006;113(9):1217-1224.
56. Blurton-Jones M, Kitazawa M, Martinez-Coria H, Castello NA, Müller F-J, Loring JF, Yamasaki TR, Poon WW, Green KN, LaFerla FM. Neural stem cells improve cognition via BDNF in a transgenic model of Alzheimer disease. *Proc Natl Acad Sci U S A*. 2009;106(32):13594-13599.
57. Li Y, Rowland C, Tacey K, Catanese J, Sninsky J, Hardy J, Powell J, Lovestone S, Morris JC, Thal L, Goate A, Owen M, Williams J, Grupe A. The *BDNF* val66met polymorphism is not associated with late onset Alzheimer's disease in three case-control samples. *Mol Psychiatry*. 2005;10(9):809-810.
58. Houlihan LM, Harris SE, Luciano M, Gow AJ, Starr JM, Visscher PM, Deary IJ. Replication study of candidate genes for cognitive abilities: the Lothian Birth Cohort 1936. *Genes Brain Behav*. 2009;8(2):238-247.
59. de Bakker PIW, Yelensky R, Pe'er I, Gabriel SB, Daly MJ, Altshuler D. Efficiency and power in genetic association studies. *Nat Genet*. 2005;37(11):1217-1223.
60. Cardon LR. Genetics: delivering new disease genes. *Science*. 2006;314(5804):1403-1405.
61. Head D, Snyder AZ, Girton LE, Morris JC, Buckner RL. Frontal-hippocampal double dissociation between normal aging and Alzheimer's disease. *Cereb Cortex*. 2005; 15(6):732-739.
62. Whitwell JL, Przybelski SA, Weigand SD, Knopman DS, Boeve BF, Petersen RC, Jack CR Jr. 3D maps from multiple MRI illustrate changing atrophy patterns as subjects progress from mild cognitive impairment to Alzheimer's disease. *Brain*. 2007;130(pt 7):1777-1786.
63. Meyer-Lindenberg A, Nichols T, Callicott JH, Ding J, Kolachana B, Buckholtz J, Mattay VS, Egan M, Weinberger DR. Impact of complex genetic variation in COMT on human brain function. *Mol Psychiatry*. 2006;11(9):797, 867-877.
64. Pezawas L, Meyer-Lindenberg A, Drabant EM, Verchinski BA, Munoz KE, Kolachana BS, Egan MF, Mattay VS, Hariri AR, Weinberger DR. 5-HTTLPR polymorphism impacts human cingulate-amygdala interactions. *Nat Neurosci*. 2005;8(6): 828-834.
65. Filippini N, MacIntosh BJ, Hough MG, Goodwin GM, Frisoni GB, Smith SM, Matthews PM, Beckmann CF, Mackay CE. Distinct patterns of brain activity in young carriers of the *APOE*-ε4 allele. *Proc Natl Acad Sci U S A*. 2009;106(17):7209-7214.
66. Pezawas L, Verchinski BA, Mattay VS, Callicott JH, Kolachana BS, Straub RE, Egan MF, Meyer-Lindenberg A, Weinberger DR. The brain-derived neurotrophic factor val66met polymorphism and variation in human cortical morphology. *J Neurosci*. 2004;24(45):10099-10102.
67. Ownby RL, Crocco E, Acevedo A, John V, Loewenstein D. Depression and risk for Alzheimer disease: systematic review, meta-analysis, and metaregression analysis. *Arch Gen Psychiatry*. 2006;63(5):530-538.
68. Neves-Pereira M, Mundo E, Muglia P, King N, Macciardi F, Kennedy JL. The brain-derived neurotrophic factor gene confers susceptibility to bipolar disorder: evidence from a family-based association study. *Am J Hum Genet*. 2002;71(3): 651-655.
69. Sen S, Nesse RM, Stoltenberg SF, Li S, Gleiberman L, Chakravarti A, Weder AB, Burmeister MA. A BDNF coding variant is associated with the NEO personality inventory domain neuroticism, a risk factor for depression. *Neuropsychopharmacology*. 2003;28(2):397-401.
70. Gatt JM, Nemeroff CB, Dobson-Stone C, Paul RH, Bryant RA, Schofield PR, Gordon E, Kemp AH, Williams LM. Interactions between BDNF Val66Met polymorphism and early life stress predict brain and arousal pathways to syndromal depression and anxiety. *Mol Psychiatry*. 2009;14(7):681-695.
71. Chobanian AV, Bakris GL, Black HR, Cushman WC, Green LA, Izzo JL Jr, Jones DW, Materson BJ, Oparil S, Wright JT Jr, Roccella EJ; Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure; National Heart, Lung, and Blood Institute; National High Blood Pressure Education Program Coordinating Committee. Seventh report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure. *Hypertension*. 2003;42(6):1206-1252.
72. Burgmans S, van Boxtel MP, Gronenschild EH, Vuurman EF, Hofman P, Uylings HB, Jolles J, Raz N. Multiple indicators of age-related differences in cerebral white matter and the modifying effects of hypertension. *Neuroimage*. 2010;49(3): 2083-2093.
73. Sublette ME, Baca-Garcia E, Parsey RV, Oquendo MA, Rodrigues SM, Galfalvy H, Huang Y-Y, Arango V, Mann JJ. Effect of BDNF val66met polymorphism on age-related amygdala volume changes in healthy subjects. *Prog Neuropsychopharmacol Biol Psychiatry*. 2008;32(7):1652-1655.
74. Kennedy KM, Rodrigue KM, Land SJ, Raz N. *BDNF* val66met polymorphism influences age differences in microstructure of the corpus callosum. *Front Hum Neurosci*. 2009;3:19.

©2011 American Medical Association. All rights reserved.
Downloaded From: http://archpsyc.jamanetwork.com/ on 05/24/2016