Association Study of the Decreased Serum BDNF Concentrations in Amnestic Mild Cognitive Impairment and the Val66Met Polymorphism in Chinese Han

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Objective: Experimental and clinical data suggested that brain-derived neurotrophic factor (BDNF) plays an important role in the pathogenesis of Alzheimer's disease (AD). Amnestic mild cognitive impairment (aMCI) is characterized by declined cognitive function and has a high probability of evolving into AD. The aim of this study was to investigate serum BDNF concentrations in aMCI patients and determine whether there is an association of the BDNF gene Val66Met polymorphism with aMCI, cognitive function, and serum BDNF.

Method: Between April 2005 and January 2006, the present study recruited 99 aMCI patients who met diagnostic criteria for MCI proposed by the Mayo Clinic Alzheimer's Disease Research Center and 99 matched healthy controls from a population-based sample. All subjects underwent extensive assessment of cognitive function, measurement of serum BDNF by an enzyme-linked immunosorbent assay, and genotyping of the BDNF gene Val66Met polymorphism.

Results: The serum concentrations of BDNF in aMCI patients (median [interquartile range] = 4.37 [2.35–6.40] ng/mL) were significantly lower than those of healthy controls (4.98 [3.50–7.33] ng/mL) ($z = -2.449$, $p = .014$). There were significant positive correlations between serum BDNF and scores on delayed recall in the Auditory Verbal Learning Test, which reflects episodic memory ($r = 0.264$, $p = .008$). No significant differences were found for either the genotype or allele distribution of BDNF Val66Met polymorphism between aMCI patients and control subjects. The BDNF Val66Met polymorphism was not associated with serum BDNF or cognitive function in aMCI patients.

Conclusions: This study suggests that reduced BDNF levels may play a role in the pathophysiology of aMCI, and the BDNF gene Val66Met polymorphism may not be an important factor in susceptibility to aMCI.

(J Clin Psychiatry 2008;69:1104–1111)

Received July 16, 2007; accepted Feb. 8, 2008. From the Clinical Neuroscience Research Center of Clinical Medical College, Southeast University, Nanjing, China.

This work was partly supported by grants from National Natural Sciences Foundation of China (30570656 to Dr. Zhang) and National Basic Research Program of China (973 Program) (2007CB512308 to Dr. Zhang). In addition, we deeply thank the Eisai China pharmaceutical company for their generous support.

The authors report no additional financial or other relationships relevant to the subject of this article.

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Alzheimer's disease (AD) represents the most common neurodegenerative disease in the aging population, but remains underdiagnosed and undertreated.[1] The pathogenic process of AD probably starts decades before the clinical onset of the disease, and AD patients are likely to have progressed through a stage of relatively isolated (usually amnesia) cognitive deficits. As a result, the focus of recent progress in the treatment of AD has increasingly shifted to accurate detection of mild cognitive impairment (MCI), which refers to the transitional zone between normal cognitive function and clinically probable AD.[2] Amnestic MCI (aMCI), a subclass of MCI, is a pure cognitive disorder characterized by episodic memory impairment and decline of the ability to learn new information that represents the prodromal stage of AD. Individuals with aMCI have a high probability of evolving toward AD at a rate of 10%–15% per year.[3] Therefore, MCI can define a valuable therapeutic window for early intervention in AD.[4] However, at present, there is no sensitive and specific biomarker to identify this at-risk population. It would be valuable to discover an aMCI-related biomarker; not only could it inform research into the cause of aMCI, but it might also be used to predict those individuals who might benefit most from interventions preventing or modifying the progression of disease.

A substantial body of evidence suggests that brain-derived neurotrophic factor (BDNF) plays an important role in the pathogenesis of AD.[5,6] BDNF is a versatile protein that is directly involved not only in cellular prolif-

eration, migration, and differentiation during development, but also in maintenance of the function and structural integrity of neurons and in synaptic plasticity in the adult brain, especially in the hippocampal, cortical, and basal forebrain cholinergic neurons, which lose function in AD.[6,7] A study in neuronal cell culture found that amyloid peptide at sublethal concentrations interfered with neuronal plasticity mediated by BDNF signaling cascade.[8,9] Neuronally differentiated P19 mouse embryonic carcinoma cells stimulated by BDNF resulted in a rapid decrease in tau phosphorylation.[10] Animal experiments revealed that BDNF gene expression was down-regulated in the cortex of a transgenic mouse model of AD,[11] and hippocampal BDNF protein was decreased in training APP23 mice.[12] Postmortem brain studies of AD patients have shown that the expression of BDNF protein and messenger ribonucleic acid were decreased in the hippocampus[13,14] and neurocortex[15–18]; however, contrary results were also found in the hippocampus and parietal cortex of AD patients in another study.[19] A recent study reported a reduction of BDNF protein in the brains of MCI patients and also found a correlation with impairment of cognitive function.[20]

Brain-derived neurotrophic factor can cross the blood-brain barrier in both directions,[21] and BDNF protein has also been detected in the serum of the rat and the human.[22] Several studies have reported altered serum BDNF concentrations in different stages of AD progression. Laske et al.[23] first reported significantly increased serum BDNF concentrations in early stage AD patients (mean ± SD Mini-Mental State Examination [MMSE] score = 25.5 ± 1.5), and decreased serum BDNF was found at a later stage of AD (MMSE score from 6.88 to 13.3).[23,24] However, Laske et al.[25] further found decreased serum BDNF in mild AD (mean ± SD MMSE score = 23.6 ± 1.6).

A number of genetic loci and candidate genes have been suggested as potential risk factors for AD, but few have been consistently replicated.[26] A number of studies have now indicated that a single nucleotide polymorphism (SNP) (196A > G) in BDNF gene (dbSNP number rs6265), producing a valine to methionine amino acid substitution at codon 66 (Val66Met) of the pro-BDNF sequence, is associated with cognitive dysfunction and AD. Hippocampal neurons transfected with Met-BDNF showed that activity-dependent secretion of BDNF was significantly impaired.[27] Chen et al.[28] found that Met-BDNF could alter the intracellular distribution and activity-dependent secretion of Val-BDNF when both forms were coexpressed in neuronal and neurosecretory cells. Association studies between this functional SNP and AD have generated conflicting results. Ventriglia et al.[29] reported a significant association for the more common Val allele with an increased risk for sporadic AD, consistent with another study in a Japanese cohort.[30] On the contrary, however, a study in a mainland Chinese cohort found a significantly low Val/Val genotype frequency in the female AD cases compared with the female controls, suggesting a protective effect of the Val/Val genotype in Chinese female AD.[31] Other studies did not find any significant differences of the allele and genotype frequencies between AD patients and controls.[32–34]

Amnestic MCI represented the prodromal stage of AD, and changed BDNF concentrations were found in AD. Thus, the present study is intended to approach the possibility that BDNF might be regarded as a biomarker for diagnosis of MCI and also to shed more light on the neurotrophin abnormality in MCI. This study assessed cognitive function in aMCI patients and measured serum BDNF and genotype of the BDNF gene Val66Met variant to explore whether altered serum BDNF would occur in the aMCI individuals and whether BDNF Val66Met polymorphism is associated with aMCI.

METHOD

Clinical Subjects and Assessment

All subjects were Chinese Han from a population-based sample living in the same district of Nanjing city center in southeast China. They gave informed consent to participate in this study, which was approved by the Institutional Ethical Review Board of Clinic Medical College of Southeast University.

A total of 99 aMCI patients (37 women, 62 men) were recruited between April 2005 and January 2006. They were all older than 65 years (mean ± SD age = 73.25 ± 5.67 years) and educated for at least 8 years (median [interquartile range] = 14 [11–16] years). All aMCI patients met the diagnostic criteria proposed by the Mayo Clinic Alzheimer's Disease Research Center,[3] including (1) subjective memory impairment corroborated by subject and an informant; (2) objective memory performances documented by an Auditory Verbal Learning Test (AVLT)-delayed recall score that is less than or equal to 1.5 SD of age-adjusted and education-adjusted norms (the cutoff was ≤ 4 correct responses on 12 items for ≥ 8 years of education)[35]; (3) normal general cognitive functioning evaluated by a MMSE score of 24 or higher[36]; (4) a Clinical Dementia Rating (CDR) of 0.5, with at least a 0.5 in the memory domain[37]; (5) no or minimal impairment in activities of daily living; (6) absence of dementia, or not sufficient to meet the National Institute of Neurological and Communicative Disorders and Stroke and the Alzheimer's Disease and Related Disorders Association (NINCDS/ADRDA) criteria for AD.[38] The process was operated by an experienced neuropsychiatrist who administered a structured interview to subjects and their informants. Exclusion criteria were a history of known stroke (Hachinski score ≥ 4),[39] alcoholism, head injury, Parkinson's disease, epilepsy, major depressive disorder

at the time of assessment or other neurologic or psychiatric illness, major medical illness (e.g., cancer, anemia, thyroid dysfunction), or severe visual or hearing loss.

The study employed a neuropsychological battery that consisted of the AVLT, Rey-Osterrieth Complex Figure Test,[40] Verbal Fluency Test (animal fluency task),[41] Digit Span Test,[42] Symbol Digit Modalities Test,[42] Trail Making Tests A and B,[43] and the Clock Drawing Test[44] to evaluate the function of episodic memory regarding both verbal and visual information, semantic memory, attention, psychomotor speed, executive function, and visuospatial skills in all aMCI individuals.

Simultaneously, 99 cognitively healthy control subjects (49 women, 50 men, mean ± SD age = 68.82 ± 5.61 years, educated for a median [IQR] of 15 [12–16] years) who underwent medical evaluation received the same neuropsychological battery assessment as the individuals with aMCI. Controls were required to have a CDR of 0, an MMSE score of 26 or higher, and an AVLT-delayed recall score greater than 4 for those with 8 or more years of education. No significant differences were found for age, educational level, or gender distribution between aMCI patients and controls (all $p > .05$).

Measurement of Serum BDNF Levels

For serum sample, blood (4 mL) obtained from the antecubital vein was collected in anticoagulant-free tubes between 7:30 and 8:00 a.m. and kept at room temperature for 1 hour, followed by 1 hour at 4°C; then the blood was centrifuged at 2000*g* for 10 minutes at 4°C. Serum was carefully collected and kept frozen at –80°C until assayed.

BDNF concentrations were measured using the BDNF Emax Immunoassay System kit (Promega; Madison, Wis.) according to the manufacturer's instructions. To minimize the assay variance, serum BDNF was measured in all subjects on the same day. Briefly, anti-BDNF monoclonal antibody was coated into 96-well plates being used to capture the neurotrophins in the serum. The captured BDNF bound specifically to BDNF polyclonal antibody. After washing, the amount of bound polyclonal antibody was then detected using anti-IgY antibody conjugated to horseradish peroxidase. The plates were incubated with a chromogenic substrate to produce a color reaction. The amount of BDNF in the test solution is proportional to the color generated in the redox reaction. The standard curve and the samples were analyzed in triplicate and showed a direct relation between optical density and BDNF concentration. The intra-assay coefficient variation was less than 6%.

Genotyping of BDNF Val66Met

Genomic DNA was extracted from 300 µL of EDTA-anticoagulated venous blood using Puregene DNA Purification Kit (Gentra Systems, Minneapolis, Minn.) according to the manufacturer's recommendations. Polymerase chain reaction–based restriction fragment length polymorphism assay was performed to genotype the DNA sequence variants (196 G > A) of the BDNF gene. The primer sequences used for analysis of 196 G > A were the sense primer 5′-ACTCTGGAGAGCGTGAAT-3′ and antisense primer 5′-ATACTGTCACACACGCTC-3′. The amplification conditions were initiated at 94°C for 5 minutes, followed by 30 cycles consisting of denaturation at 94°C for 30 seconds, annealing at 60°C for 30 seconds, and extension at 72°C for 30 seconds, with a final extension step of 5 minutes at 72°C. Polymerase chain reaction product was digested with NlaIII (Fermentas Inc., Vilnius, Lithuania) at 37°C for 5 hours and then electrophoresed on a 2.5% agarose gel at 75 V for 60 minutes. The presence of 168 base pair (bp) and 75 bp bands indicated the existence of A (66Met) allele, the presence of 243 bp band indicated the existence of G (66Val) allele, while the presence of 75 bp, 168 bp, and 243 bp indicated AG (66Met/66Val) heterozygote.

Statistical Analysis

Analysis was performed using SPSS software, version 11.5 (SPSS Inc., Chicago, Ill.). A χ^2 goodness-of-fit test was used to test the distribution of genotypes and allele frequencies for deviations from Hardy-Weinberg equilibrium. The BDNF allele and genotype distributions between patients and controls were evaluated by the Pearson χ^2 test. Kolmogorov-Smirnov test was used to analyze for normality, and Levene test was used to verify homogeneity of group variances. Student t test and analysis of variance were employed for the normally distributed variables; the nonparametric Mann-Whitney U test and Kruskal-Wallis test were employed for the asymmetrically distributed variables. Correlation between serum BDNF levels and clinical parameters were examined by bivariate correlation (Pearson or Spearman rank correlation). Relationships between demographic factors, clinical cognitive measures, genotypes, and serum BDNF concentrations in the subjects with aMCI were examined by multiple stepwise regression analysis. The continuous variables are presented as the mean ± SD or median. Statistical significance was defined at $p < .05$, 2-tailed tests.

RESULTS

Comparison of Cognitive Performance Between aMCI Patients and Healthy Controls

The neuropsychological assessments for all subjects are given in Table 1. The scores on neuropsychological tests in aMCI patients were significantly poorer than those in control subjects (all $p < .001$), with the largest impairment on the Auditory Verbal Learning Test-delayed recall (< 3.0 SD of normal mean), which reflects episodic memory. Beyond the memory task, Trail Making Test B,

Table 1. Comparison of Cognitive Performance Between aMCI Patients and Healthy Controls[a]

Measure	aMCI Patients (N = 99)	Controls (N = 99)	Score	p Value
MMSE	27.00 (26–27)	29.00 (27–30)	z = –7.163	< .001[b]
AVLT-delayed recall	3.00 (1.00–4.00)	8.00 (7.00–9.00)	z = –12.025	< .001[b]
CFT-delayed recall, mean ± SD	8.93 ± 6.76	17.01 ± 7.53	t = 7.681	< .001[c]
Category Fluency Test, mean ± SD	10.33 ± 2.85	12.98 ± 2.96	t = 6.370	< .001[c]
Trail Making Test A	95.00 (74.00–130.00)	67.00 (54.50–81.50)	z = –6.795	< .001[b]
Trail Making Test B, mean ± SD	192.93 ± 78.41	132.53 ± 41.24	t = –7.075	< .001[c]
Symbol Digit Modalities Test, mean ± SD	23.08 ± 9.94	34.49 ± 9.96	t = 8.029	< .001[c]
Digit Span Test	12.00 (10.00–13.00)	13.00 (12.00–14.00)	z = –4.291	< .001[b]
Clock Drawing Test	8.00 (7.00–9.00)	9.00 (8.00–10.00)	z = –3.490	< .001[b]

[a]Values expressed as median (interquartile range) except where noted.
[b]Mann-Whitney U test.
[c]Student t test.
Abbreviations: aMCI = amnestic mild cognitive impairment, AVLT = Auditory Verbal Learning Test, CFT = Rey-Osterrieth Complex Figure Test, MMSE = Mini-Mental State Examination.

Table 2. Serum BDNF Concentrations (ng/mL) in Healthy Controls and aMCI Patients, Median (interquartile range)

			Gender			Genotype			
Group	Total	p Value[a]	Male	Female	p Value[b]	AA	AG	GG	p Value[c]
aMCI patients	4.37 (2.35–6.40)	.014	4.51 (2.52–6.40)	3.69 (2.27–6.25)	.697	4.90 (2.45–7.97)	4.63 (2.62–6.35)	3.36 (2.25–4.90)	.270
Controls	4.98 (3.50–7.33)		4.98 (3.87–8.13)	4.28 (2.98–6.94)	.200	4.32 (2.87–6.68)	4.51 (3.27–6.80)	6.44 (4.01–9.42)	.053

[a]Mann-Whitney test for comparison of serum BDNF levels between aMCI patients and controls.
[b]Mann-Whitney test for comparison of serum BDNF levels between male and female in aMCI patients or controls.
[c]Kruskal-Wallis test for comparison of serum BDNF levels between genotypic subgroups in aMCI patients or controls.
Abbreviations: aMCI = amnestic mild cognitive impairment, BDNF = brain-derived neurotrophic factor.

representing executive function, showed the greatest decline, deviating from normal by 1.5 SD.

Comparison of Serum BDNF Level Between aMCI Patients and Healthy Controls

The aMCI patients had markedly lower serum BDNF concentrations than controls ($z = –2.449$, $p = .014$). Furthermore, serum BDNF was not significantly different between men and women in aMCI patients or controls, and no significant difference was found between genotypic subgroups (AA, AG, and GG) in serum BDNF concentrations in aMCI patients or controls (all $p > .05$) (Table 2).

Comparison of the Frequencies of Allele and Genotype of BDNF Val66Met Between Groups

Distribution of the BDNF genotypes were consistent with Hardy-Weinberg equilibrium in both aMCI patients ($\chi^2 = 3.065$, $df = 1$, $p > .05$) and control subjects ($\chi^2 = 0.136$, $df = 1$, $p > .05$). No significant differences were found in BDNF genotype and allele distributions between the aMCI cases and the controls ($\chi^2 = 2.282$, $df = 2$, $p = .319$ and $\chi^2 = 0.162$, $df = 1$, $p = .687$, respectively) (Table 3). Genotype frequencies of controls were consistent with those previously reported for a Chinese Han population (AA = 22.0%, AG = 50.5%, GG = 27.4%)[45] and Japanese population (AA = 20.8%, GA = 47.4%, GG = 31.9%),[30] but differed, however, from those reported in a white population (AA = 3.0%, AG = 32.3%, GG = 64.7%).[34] In addition, we failed to reveal any significant deviation with the distribution of the genotypes and alleles after analysis of stratification in terms of gender and age between aMCI patients and controls (all $p > .05$).

The Relationships Between Clinical Characteristics, Demographic Characteristics, Serum BDNF Concentrations, and Genotype of BDNF Val66Met

No significant difference was observed between the genotypic subgroups of the BDNF polymorphisms and the neuropsychological test scores in aMCI patients (1-way analysis of variance or Kruskal-Wallis test, all $p > .05$) (Table 4). In the aMCI patients, there was no significant correlation between serum BDNF and age or time in education (Spearman correlations: $r = –0.053$, $p = .606$, and $r = 0.071$, $p = .487$, respectively). However, significant positive correlation was found between serum BDNF and scores on delayed recall of the AVLT and Category Fluency Test (Spearman correlations: $r = 0.264$, $p = .008$, and $r = 0.211$, $p = .038$, respectively). When serum BDNF was taken as a dependent variable, and age, education, genotype, and the scores of cognitive assessments were taken as independent variables, multiple stepwise regression analysis indicated that only delayed recall of the AVLT had any significant association with serum BDNF levels ($\beta = 0.453$, $t = 2.186$, $p = .031$).

Table 3. Genotype and Allele Frequency for BDNF Gene Val66Met Polymorphism in Healthy Controls and aMCI Patients

	Genotype, N (%)				Allele, N (%)		
Group	AA	AG	GG	p Value[a]	A	G	p Value[a]
aMCI patients (N = 99)	27 (27.3)	41 (41.4)	31 (31.3)	.319	95 (48.0)	103 (52.0)	.687
Age ≤ 70 years (N = 28)	6 (21.4)	15 (53.6)	7 (25.0)	.995	27 (48.2)	29 (51.8)	.883
Age > 70 years (N = 71)	21 (29.6)	26 (36.6)	24 (33.8)	.195	68 (47.9)	74 (52.1)	.694
Male (N = 62)	14 (22.6)	30 (48.4)	18 (29.0)	.778	58 (46.8)	66 (53.2)	.475
Female (N = 37)	13 (35.1)	11 (29.7)	13 (35.1)	.064	37 (50.0)	37 (50.0)	> .99
Controls (N = 99)	20 (20.2)	51 (51.5)	28 (28.3)		91 (46.0)	107 (54.0)	
Age ≤ 70 years (N = 32)	7 (21.9)	16 (50.0)	9 (28.1)		30 (46.9)	34 (53.1)	
Age > 70 years (N = 67)	13 (19.4)	35 (52.2)	19 (28.4)		61 (45.5)	73 (54.5)	
Male (N = 50)	9 (18.0)	24 (48.0)	17 (34.0)		42 (42.0)	58 (58.0)	
Female (N = 49)	11 (22.4)	27 (55.1)	11 (22.4)		49 (50.0)	49 (50.0)	

[a]Compared with controls by χ^2 test.
Abbreviations: aMCI = amnestic mild cognitive impairment, BDNF = brain-derived neurotrophic factor, Val66Met = valine to methionine amino acid substitution at codon 66.

Table 4. Comparison of Cognitive Performance Between aMCI Patients and Healthy Controls According to Genotype of BDNF Gene Val66Met Polymorphism[a]

	aMCI Patients				Controls			
Measure	AA	AG	GG	p Value	AA	AG	GG	p Value
MMSE	27 (25–28)	27 (25–28)	26 (25–27)	.905[b]	27 (26–29)	27 (26–29)	28 (27–30)	.359[b]
AVLT-delayed recall	3 (1–3)	3 (1–4)	2 (0–3)	.123[b]	8 (6–9)	8 (7–10)	8 (7–9)	.552[b]
CFT-delayed recall, mean ± SD	10.21 ± 8.93	9.23 ± 5.80	8.06 ± 6.85	.637[c]	17.05 ± 8.73	17.39 ± 7.32	16.41 ± 7.12	.859[c]
Category Fluency Test, mean ± SD	11.07 ± 2.88	10.39 ± 2.88	9.74 ± 2.73	.207[c]	12.68 ± 2.94	13.12 ± 3.28	12.93 ± 2.32	.858[c]
Trail Making Test A	79 (66–122)	91 (74–136)	107 (78–137)	.141[b]	68 (55–85)	65 (48–82)	67 (60–80)	.397[c]
Trail Making Test B, mean ± SD	202.44 ± 79.29	186.95 ± 74.03	197.68 ± 77.31	.689[c]	124.84 ± 37.82	130.24 ± 41.19	141.82 ± 43.26	.330[c]
Symbol Digit Modalities Test, mean ± SD	23.85 ± 9.98	23.49 ± 11.26	21.87 ± 8.09	.712[c]	35.95 ± 10.98	34.32 ± 8.90	33.82 ± 11.23	.764[c]
Digit Span Test	12 (10–13)	12 (10–13)	12 (9–14)	.993[b]	13 (11–14)	13 (12–14)	13 (12–15)	.568[b]
Clock Drawing Test	8 (7–9)	8 (7–9)	9 (8–9)	.855[b]	9 (7–10)	9 (8–10)	9 (8–10)	.737[b]

[a]Values expressed as median (interquartile range) except where noted.
[b]Kruskal-Wallis test.
[c]One-way analysis of variance.
Abbreviations: aMCI = amnestic mild cognitive impairment, AVLT = Auditory Verbal Learning Test, BDNF = brain-derived neurotrophin factor, CFT = Rey-Osterrieth Complex Figure Test, MMSE = Mini-Mental State Examination, Val66Met = valine to methionine amino acid substitution at codon 66.

DISCUSSION

Main findings of the present study were that aMCI patients were characterized by markedly episodic memory decline. Serum BDNF concentrations were significantly decreased and correlated with the degree of episodic memory impairment in aMCI patients. However, we failed to find any significant association between BDNF Val66Met polymorphism and aMCI. Moreover, genotype did not significantly affect serum BDNF or cognitive function in aMCI patients.

In the present study, all recruited subjects were Chinese Han, originally from Jiangsu province located in southeast China, which limited the influence of heterogeneity of population structure in producing a false positive result in such a case-control association study. Mild cognitive impairment is a heterogeneous disease divided into amnesia and no-amnesia MCI subtypes. Importantly, aMCI has a complex contribution from degenerative diseases such as AD, vascular disease, and depression. This present study recruited patients with aMCI of degenerative etiology only rather than patients with aMCI of vascular disease or depression etiology, which increased the homogeneity of aMCI patients to exclude confounding factors for association analysis of disease.

This study employed multi-dimensional neuropsychological tests to extensively assess cognitive function in all subjects. The delayed recall task of the AVLT represented episodic memory, which is ability to form new memories for events, and was used to establish the presence of memory impairment.[2] The defined episodic memory impairment was observed in aMCI patients of the present study, consistent with previous reports.[3,46] Studies have suggested that impaired episodic memory is not only a defining feature of early AD but also a strong predictor of progression for AD in MCI.[47]

As far as we are aware, this study is first to report significantly decreased serum BDNF in aMCI patients compared with healthy elderly controls. Moreover, there was significant positive correlation between serum BDNF and scores on delayed recall of the AVLT in aMCI patients. Numerous data suggested that BDNF has emerged as a key neuromodulator of synaptic transmission; activity-dependent synaptic plasticity and BDNF signaling at synapses enhances long-term potentiation, a process of synaptic strengthening associated with learning and memory.[48,49] For instance, long-term potentiation was impaired in BDNF knockout mice, retrieved by adenovirus-mediated transfection of CA1 cells with the BDNF gene.[50,51] Several lines of evidence suggest AD is a synaptic failure, especially in its earliest clinical phase.[52–54] An autopsy study[55] found that levels of synaptic protein-drebrin, a dendritic spine plasticity marker, were reduced 35% in the temporal cortex of MCI patients. It indicated that reduced synaptic plasticity also presented in MCI, which may contribute to the early impairment of temporal cortical functions subserving memory.[55] A positive correlation between cortical and serum BDNF concentrations has been reported in rats.[56] Accordingly, decreased BDNF serum in the present study could reflect low protein in the brain that would alter cognitive function in MCI patients. This result would also further support the findings of postmortem studies, in which mature BDNF exhibited a greater decline of 34% in the parietal lobe of MCI patients compared with cognitively healthy individuals.[20] These results seem to corroborate the hypothesis that BDNF plays a key role in the pathophysiology of MCI and indicate that the serum BDNF could be a biological marker for clinical diagnosis of aMCI.

We found serum BDNF was not significantly associated with BDNF Val66Met polymorphism in aMCI patients. This finding seems to be compatible with a recent study showing that in autopsy patients with AD, brain BDNF concentrations did not significantly differ between the various genotypes of BDNF Val66Met polymorphism.[57] BDNF is secreted through both constitutive and regulated pathways, and the latter is key to control of synaptic plasticity. However, the BDNF Val66Met variant only affects activity-dependent secretion of BDNF but not the constitutive pathway. Consequently, this finding provides plausible explanations for BDNF concentrations in blood being unaffected by the genotype.

We first compared the distribution of allele and genotype of BDNF Val66Met between aMCI patients and control subjects in Chinese Han populations. However, this study was unable to verify the significant association between BDNF Val66Met polymorphism and episodic memory in aMCI patients, nor is BDNF genetic variant an aMCI susceptibility factor. Even when groups were subdivided with regard to gender and age, we found no differences in the distribution of genotypes and alleles.

Recently, greater attention has been paid to the direct link of genetic variation to memory performance, particularly episodic memory.[58] The analysis of genotype–phenotype correlations has revealed that the BDNF Met allele was associated with poorer episodic memory of Wechsler Memory Scale stories,[27] reduced hippocampal volume,[59] and hippocampal engagement,[60] which is closely related with formation of episodic memory in healthy subjects compared to Val/Val homozygotes. The polymorphism of BDNF Val66Met is believed to influence intracellular sorting and activity-dependent secretion of BDNF. Met-BDNF is not accumulated in synapse, which results in decreased synaptic plasticity. Previous studies have revealed that decreased hippocampal activity[61] and reduced hippocampal volume[62] presented in aMCI patients. As a result, this study explored the functional variant associated with episodic memory and vulnerability to aMCI. Although a negative association was presented, it does not rule out BDNF as an important biological influence in the pathophysiology of MCI.

There are several factors that may elucidate the negative findings reported in this study. First, the heritability of diseases may derive from a number of genes, each with a small effect, thus a large sample size is needed to find a weak effect of a single gene. The second possibility is that other polymorphisms in the coding region or other regions of the BDNF gene are involved in the pathogenesis of aMCI. The BDNF Val66Met polymorphism could be in linkage disequilibrium with the true disease variants. In addition, there is an inherent difficulty in determining the genetic influences on a disease that is clearly induced by environmental factors. Thus, to determine whether or not the BDNF gene is associated with MCI, further studies on more polymorphic sites within and close to the gene in larger, independent samples are required.

CONCLUSION

In conclusion, this study indicates that decreased BDNF may contribute to the lack of trophic support in the pathogenesis of MCI. At the same time, peripheral BDNF measures could be a neurobiological marker in MCI, but these results need to be confirmed by further studies. Although no significant association of the BDNF gene Val66Met polymorphism with aMCI and cognition performance in the Chinese Han populations was found in the present study, suggesting that the SNP is not a robust genetic risk factor for MCI, investigations of other possible BDNF variants should be pursued.

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