

# Characteristics of TCM Constitution and Related Biomarkers for Mild Cognitive Impairment

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**Introduction:** The incidence of Alzheimer's disease is on the rise, early detection of cognitive impairment of the elderly is very important. In traditional Chinese medicine, constitution is related to the susceptibility of the human body to diseases. Based on the theory of constitution of traditional Chinese medicine (TCM), the human population can be classified into 9 constitutions. However, little is known about the characteristics of medical constitution and related biomarkers in subjects with mild cognitive impairment (MCI).

**Methods:** We measured the TCM Constitution of 214 subjects by using the Constitution in Chinese Medicine Questionnaire (CCMQ). MMSE and MoCA were used to assess cognitive function. The subjects were divided into mild cognitive impairment group (MCI, n = 152) and normal control group (NC, n = 62). The levels of serum Hcy and serum/urine 8-iso-PGF 2 $\alpha$  were determined.

**Results:** 1) It was found that there was a significant difference in constitution types between MCI and NC. There were significant differences in MMSE and MoCA score, serum Hcy and serum/urine 8-iso-PGF 2 $\alpha$  levels between the two groups. 2) In logistic regression analysis, the variables with statistical significance were TCM Constitution of Yang-Deficient, Phlegm-Dampness, Blood-Stasis and abnormal increase of Hcy (OR>1). 3) The MoCA scores had a positive correlation with the MMSE. A statistically significant inverse association was found between serum Hcy, blood and urine 8-iso-PGF 2 $\alpha$  and scores of cognitive assessment in MCI.

**Conclusion:** Constitution types (Yang-Deficient, Phlegm-Dampness and Blood-Stasis) and abnormal serum Hcy elevation can be used as risk factors for MCI. MoCA scores can serve to detect MCI at early stage. Serum/urine 8-iso-PGF 2 $\alpha$  has a certain relationship with MCI. Higher levels of serum/urine 8-iso-PGF 2 $\alpha$  are more likely to be associated with MCI risk.

**Keywords:** cognitive impairment, constitutions of traditional Chinese medicine, biomarkers, MoCA score

## Introduction

Mild cognitive impairment (MCI) is commonly used to refer to patients with objective cognitive impairment and normal daily living abilities that do not meet the criteria for dementia.<sup>1-4</sup> MCI is an important risk factor for dementia and in some cases may represent the prodromal phase of AD or other neurodegenerative diseases. Approximately 35% of MCI patients progress to AD dementia during a 3-year follow-up with an annual conversion rate of 5%-10%.<sup>5,6</sup> However, compared with healthy subjects, the conversion rate of AD was higher.<sup>7</sup> Therefore, the identification of MCI is very important in clinic. Currently, diagnostic aids for MCI and AD include cerebrospinal fluid (CSF) examination and blood biomarkers,<sup>8,9</sup> functional MRI,<sup>10</sup> and PET imaging.<sup>11</sup> However, CSF is invasive. Functional MRI and PET imaging are limited by equipment

and expensive examination costs, thus, we need more simple and accurate diagnostic tests to identify MCI patients whose AD may be a potential cause in the early stages of the disease. The most widely used measure of cognitive impairment is the mini-mental state examination (MMSE) scale. However, there may be a ceiling effect in MMSE, which can lead to false negative results. The MoCA is a brief cognitive screening tool with high sensitivity and specificity for detecting MCI as currently conceptualized in patients performing in the normal range on the MMSE.<sup>12–18</sup> The use of biomarkers also contributes to the early diagnosis and early treatment of MCI. Studies have shown that Homocysteine (Hcy), 8-iso-prostaglandin F<sub>2α</sub> (8-iso-PGF<sub>2α</sub>) is a potential biomarker for cognitive impairment.<sup>19,20</sup> Hcy is a sulfur-containing amino acid. Current studies have shown that Hcy is associated with cognitive impairment, and its possible ways of inducing cognitive impairment include increasing glutamate excitotoxicity, reducing DNA repair ability of neurons, accelerating oxidative stress and Aβ protein formation, and damaging the ability of hippocampal neurons.<sup>21–23</sup> On the other hand, Hcy can induce vascular endothelial cell dysfunction, which may lead to cerebrovascular cognitive impairment.<sup>24</sup> 8-iso-PGF<sub>2α</sub> is the end product of endogenous lipid peroxidation and the product of free radical catalyzed arachidonic acid. Autopsy showed that patients with Alzheimer's disease had obvious oxidative damage, and the content of 8-iso-PGF<sub>2α</sub> in temporal lobe and frontal lobe was significantly increased.<sup>25</sup> In addition, studies have shown that plasma 8-iso-PGF<sub>2α</sub> concentration is related to cognitive impairment in non-dementia elderly.<sup>26,27</sup> 8-iso-PGF<sub>2α</sub> has been accepted as a reliable biomarker for evaluating oxidative stress.<sup>28,29</sup> In the theory of traditional Chinese medicine (TCM), the body constitution is an individual character compatible with nature and the social environment, there are nine constitutions: Neutral, Qi-Deficient, Yang-Deficient, Yin-Deficient, Phlegm-Dampness, Dampness-Heat, Blood-Stasis, Qi-Depression, and Special.<sup>30,31</sup> The difference in individual constitution makes the individual susceptible to certain pathogenic factors, and even has an impact on physical and mental health.<sup>32–34</sup> In April 2009, The Constitution in Chinese Medicine Questionnaire (CCMQ) was published and recommended as the constitution measurement standard by Chinese Traditional Medicine Association.<sup>35</sup> The reliability and construct validity of the scale were thoroughly evaluated.<sup>36</sup> More than 20,000 surveys have been completed in China and other Asian countries to help the respondents understand their TCM Constitution and get health care advice. With the wide application of CCMQ in recent years, more and more research on

TCM constitution has appeared, such as clinical research related to metabolic syndrome (MetS), dyslipidemia, etc.,<sup>37,38</sup> as well as research on the relationship between biomarkers of type 2 diabetes and TCM syndromes.<sup>39</sup> All of these have positive significance for understanding the Constitution and the diagnosis and treatment of diseases. Based on previous studies,<sup>40</sup> we know that constitutions are associated with vascular cognitive impairment. A recent study showed that there is an interaction between blood-stasis constitution and atherosclerosis in cognitive impairment in the elderly.<sup>41</sup> However, few studies have systematically explored the relationship between TCM Constitution and biomarkers related to cognitive decline. This study aimed to investigate the MCI characteristics from the perspective of TCM constitution and biomarkers, to reveal the distribution of constitutional types of patients with MCI.

## Materials and Methods

### Subjects

A total of 214 subjects (71 males, 143 females) were recruited from the health examination center and the memory outpatient clinic of the First Affiliated Hospital of Guangxi University of Traditional Chinese Medicine and community center in Guangxi Nanning in the period between January 2014 to December 2017. Inclusion criteria were: 1) 55≤age≤82 years, 2) MCI group needs to meet the MCI diagnostic criteria of Mayo Clinic-revised defined by Petersen et al,<sup>42,43</sup> with 24≤MMSE score≤27, 18≤MoCA score≤30,<sup>15</sup> 3) NC group should be complied with noncognitive impairment, with MMSE score > 27 points, MoCA score >18 points.<sup>15</sup> Exclusion criteria: 1) age below 55 years, 2) other diseases with advanced, severe primary diseases such as liver and kidney, 3) severe hearing and visual impairment, unable to cooperate with assessment, 4) history of neurological diseases (such as macrovascular stroke, dementia, Parkinson's disease, seizures), 5) mental illness (such as schizophrenia, bipolar disorder), 6) subjects who are known to have used other drugs that may affect serum Hcy, serum and urine 8-iso-PGF<sub>2α</sub>.

### TCM Constitution Assessment

The TCM constitution classification table adopts Wang Qi's nine-point method of constitution classification, which divides human constitution into Neutral, Qi-Deficient, Yang-Deficient, Yin-Deficient, Phlegm-Dampness, Dampness-Heat, Blood-Stasis, Qi-Depression, and Special constitution.<sup>44</sup> The TCM constitutions were measured and

classified with Constitution in Chinese Medicine Questionnaire (CCMQ).<sup>45</sup> Neutral type is a balanced one with higher score indicating a better constitution status, while the other eight types are pathological types with higher score indicating worse constitution status. There are 66 items in CCMQ. According to the constitution classification, it is divided into 9 sub scales, each sub scale contains 7–8 items, and each item is divided into 1–5 points. As the subjects are 55 years old and above, in order to avoid wrong answers caused by differences in education level, knowledge composition and understanding ability, all items in the CCMQ are asked and filled in one by one by trained researchers. After filling in, researchers need to check the scale again to ensure the integrity of the data and then take it back on the spot. After taking back, according to the scoring method, the scores of each item of the nine subscales were added up, and the total score calculated by each subscale was the original score of each constitution, and then the transformation score was calculated according to the formula, the transformation score = [(Original Score - number of items)/(number of items × 4)] × 100%. When the transformation score of Neutral constitution ≥ 60, and the transformation score of the other eight constitutions ≤ 40, it is judged as Neutral constitution. When only one of the biased constitutions' transformation score is ≥ 40, we judge it as biased constitution. For a certain biased constitution, if there are multiple constitutions at the same time and the transformation score ≥ 40, the biased constitution with the highest transformation score is used as the judgment constitution.

## Neuropsychological Assessment

Cognitive assessments include Mini Mental State Examination (MMSE) and Montreal Cognitive Assessment (MoCA). The MMSE consists of 10 tasks, testing direction, memory, attention, calculation, language (naming, repetition, auditory comprehension, reading and writing) and visual special ability, with a total score of 30.

MoCA tests 8 cognitive areas, visual spatial ability, attention, executive function, immediate memory, delayed memory, language, abstraction, calculation and orientation, the highest total score is 30. If the subjects' education years are less than 12 years, the measured total score is the test score plus 1 point.<sup>15</sup> All tests were administered and graded by professionals trained in neuropsychological testing.

## Blood Biochemical Tests

### Serum and Urine 8-Iso-PGF 2a Testing

The level of serum and urine 8-iso-PGF 2a was measured by enzyme-linked immunosorbent assay (ELISA). In the morning, 5 mL of fasting venous blood and 5 mL of urine were taken from the subjects; serum was separated and kept below −20°C until analysis; urine was immediately kept under −80°C until analysis. Enzyme-linked immunosorbent assay (ELISA) kits were purchased from Shanghai Lianshuo Biotechnology Co., Ltd. (Batch No.: 201605, 201610, 201612, 201703, 201707, 201710, 201712).

### Hcy Test

On an empty stomach the next morning after a normal dinner, 3 mL of elbow venous blood was taken by a professional and collected with 5 mL vacuum blood collection tube (without anticoagulant) and sent to the laboratory for testing. The instrument was Hitachi 7600. Automatic biochemical analyzer and its supporting reagents were used.  $0 < \text{Hcy} \leq 10 \mu\text{mol/L}$  is the normal value.

## Data Analysis

The results were reported by using professional statistical software (Statistical Package for Social Sciences 23.0, SPSS). All data were expressed as mean ± standard deviation ( $M \pm SD$ ). Two independent sample *t*-tests were used for comparison between measurement data sets. Chi-squared test was used for counting data. Pearson correlation coefficient (*r*) was used to correlate MMSE, MoCA, and serum biological variables analysis; logistic regression analysis was used to analyze the binary dependent variables MCI and independent variables including nine TCM constitutions, abnormal Hcy elevation, occupation, age, education level. Multiple linear variables regression analysis was used for MMSE, MoCA, urine 8-iso-PGF 2a level, serum 8-iso-PGF 2a level. The ratio of risk to odds (OR), regression coefficient (B) and P value were analyzed. The difference was statistically significant at  $P < 0.05$ .

## Results

### Sociodemographic and Medical Characteristics at Inclusion

A total of 214 subjects were included in the final sample, including 152 subjects (50 males and 102 females) in the cognitive impairment group and 62 controls (21 males and 41 females). The ratio of gender, age, education, occupation status and blood pressure of the two groups were shown in Table 1. There was no difference in gender, age, education

**Table 1** Sociodemographic and Medical Characteristics at Inclusion (MCI=152, NC=62)

Variable		MCI n (%)	NC n (%)	p-value*	c2
	Gender				
Gender	Men	50 (32.9)	21 (33.9)	0.891	0.019
	Women	102 (67.1)	41 (66.1)		
	Age (years)				
Age	55–60 years	43 (28.3)	14 (22.6)	0.375	4.239
	61–65 years	37 (24.3)	19 (30.6)		
	66–70 years	35 (23)	16 (25.8)		
	71–75 years	30 (19.7)	13 (21)		
	76–81 years	7 (4.6)	0 (0)		
	Educational level				
Education level	Elementary school	17 (11.2)	8 (12.9)	0.936	0.421
	Middle school	39 (25.7)	17 (27.4)		
	High school	48 (31.6)	20 (32.3)		
	College	48 (31.6)	17 (27.4)		
	Occupation				
Occupation	Metal workers	101 (66.4)	37 (59.7)	0.348	0.881
	Manual workers	51 (33.6)	25 (40.3)		
Blood pressure	Systolic	128.7±11.97	128.97±14.80	0.892	
	Diastolic	78.22±9.39	79.50±9.58	0.371	

**Notes:** Data are expressed as mean ± SD (range) except where frequencies are used for categorical data. \*Chi-squared test for categorical variables; t-test for continuous variables.

level, occupation, blood pressure between the two groups. There were significant differences in MMSE and MoCA score, serum Hcy and serum/urine 8-iso-PGF 2a levels between the two groups ( $P < 0.001$ ), see [Table 2](#).

## TCM Constitution Distribution of MCI Patients and Normal Control Group

There was statistical significance between the two groups in terms of the proportion of Neutral type and unneutral types'

distribution ( $P < 0.01$ ). In NC group, the proportion of Neutral type was 64.5%, and the unneutral type was 35.5%. In MCI group, the proportion of Neutral type was only 33.6%. Compared with NC group, the unneutral type was 66.4%. Yang-Deficient was the first, followed by Qi-Deficient, Phlegm-Dampness and Blood-Stasis, which indicates that the proportion of unneutral types in MCI patients was higher than that in NC group except Qi-Deficient. There was no special constitution in both groups, [Table 3](#).

**Table 2** Comparison Between MCI and NC Subjects Regarding Mean Scores of Cognitive Assessment Scales and Urine/Serum 8-Iso-PGF2 $\alpha$  Level

Variable	MCI (n=152)	NC (n=62)	p-value
MMSE <sup>a</sup>	25.88±1.40	29.03±0.83	0.000
MoCA	21.64±3.11	25.16±2.98	0.000
Hcy level ( $\mu\text{mol/L}$ )	11.97±2.95	10.51±3.02	0.001
Urine8-iso-PGF2 $\alpha$ level (ng/L) <sup>a</sup>	60.40±16.08	35.03±5.59	0.000
Serum 8-iso-PGF2 $\alpha$ level (ng/L) <sup>a</sup>	60.53±15.90	31.89±8.09	0.000

**Notes:** Data are expressed as mean ± SD (range) except where frequencies are used for categorical data. <sup>a</sup>Wilcoxon W-test for continuous variable with heterogeneity of variance.

## Logistic Regression Analysis of Common TCM Constitution and Related Abnormal Hcy Elevation in Two Groups

The binary variables logistic regression analysis was carried out on non-frequency covariables including age, occupation, education level, TCM constitutions, and binary response variable MCI cases or not. The overall percentage for MCI classification was 71.5% (cut-off value of 0.5). It showed TCM constitution covariables, wald=17.485,  $P=0.015$  ([Table 4](#)). There were significant differences in the three types of Yang-Deficient, Phlegm-Dampness, Blood-Stasis and the abnormal Hcy elevation ( $P<0.05$ ), and three constitutions OR (95% CI)

**Table 3** Characteristics of Traditional Chinese Medical Constitution

Constitution	MCI n (%)	NC n (%)	p-value*	c2
Neutral	51 (33.6)	40 (64.5)	0.000	22.035
Qi-Deficient	51 (33.6)	11 (17.7)		
Yang-Deficient	33 (21.7)	5 (8.1)		
Yin-Deficient	4 (2.6)	1 (1.6)		
Phlegm-Dampness	14 (9.2)	1 (1.6)		
Dampness-Heat	1 (0.7)	0 (0)		
Blood-Stasis	12 (7.9)	1 (1.6)		
Qi-Depression	11 (7.2)	3 (4.8)		
Special	0 (0)	0 (0)		

**Notes:** \*Chi-squared test for categorical variables; t-test for continuous variables.

of Yang-Deficient, Phlegm-Dampness, Blood-Stasis and the abnormal Hcy elevations were 1.853 (1.893,15.082), 11.006 (1.376,88.02), 11.006 (1.376,88.02) and 1.973 (1.01, 3.855) respectively (Table 5).

### Multiple Linear Variables Regression Analysis of MoCA Scores, Urinary 8-Iso-Prostaglandin F2 $\alpha$ (8-Iso-PGF2 $\alpha$ ), and Serum 8-Iso-PGF2 $\alpha$

MoCA, urine/serum 8-iso-PGF2 $\alpha$  had significant influence on the dependent variable MMSE by multiple linear

variables regression analysis (R Square=0.50, p=0.0001) (Table 6). It suggested that the level of serum and urine 8-iso-PGF2 $\alpha$  has a certain relationship with MCI.

### Correlation Between Cognitive Scores and Changes of Serum Hcy, Urinary 8-Iso-Prostaglandin F2 $\alpha$ (8-Iso-PGF2 $\alpha$ ), and Serum 8-Iso-PGF2 $\alpha$

The MoCA scores had a positive correlation with MMSE (r =0.564, P <0.001), there was negative correlation between urine/serum 8-iso-PGF 2 $\alpha$  (r =-0.445 P <0.001/r =-0.561, P <0.001) and MMSE. Scatter plots are shown in Figure 1.

## Discussion

According to the constitution theory of traditional Chinese medicine, the determination of constitution can provide reference for the prevention and treatment of diseases. In recent years, a series of evaluations has been carried out on the correlation between TCM Constitution and diseases, eg, Phlegm-Dampness, Qi-Deficient and Dampness-Heat are the main constitution types of MetS patients, in which phlegm dampness and Qi deficiency are related to the occurrence of MetS.<sup>37</sup> Neutral constitution is the

**Table 4** Logistic Regression Analysis: Non-Frequency Covariables (N=214)

Constitution	B	S.E.	Wald	df	p	OR	95% C.I. for Exp(B)	
							Lower	Upper
Age	-0.254	0.388	0.776	4	0.942	0.775	0.362	1.659
Occupation			0.43	1	0.512			
Education level			0.059	4	1			
TCM constitutions			17.845	7	0.015			

**Note:** Variable(s) entered on step 1: x1 age, X2occupation, X3Education level, X4TCM constitutions.

**Table 5** Logistic Regression Analysis of 7 Common TCM Constitutions and Abnormal Hcy in Two Groups

Constitution	B	S.E.	Wald	df	p	OR	95% C.I. for Exp(B)	
							Lower	Upper
Neutral	0.617	0.422	2.138	1	0.144	1.853	0.811	4.238
Qi-Deficient								
Yang-Deficient								
Phlegm-Dampness								
Blood-Stasis								
Qi-Depression								
Yin-Deficient								
Abnormal Hcy	0.68	0.342	3.954	1	0.047	1.973	1.01	3.855



**Table 6** Coefficients of Multiple Linear Regression Analysis

Model	Unstandardized Coefficients		Standardized Coefficients	t	Sig.	95.0% Confidence Interval for B	
	B	Std. Error	Beta			Lower Bound	Upper Bound
(Constant)	24.765	0.85		29.138	0.000	23.09	26.441
MoCA	0.216	0.029	0.391	7.368	0.000	0.158	0.273
Urine 8-iso-PGF 2a	-0.022	0.006	-0.207	-3.797	0.000	-0.033	-0.011
Serum 8-iso-PGF 2a	-0.032	0.006	-0.326	-5.732	0.000	-0.044	-0.021

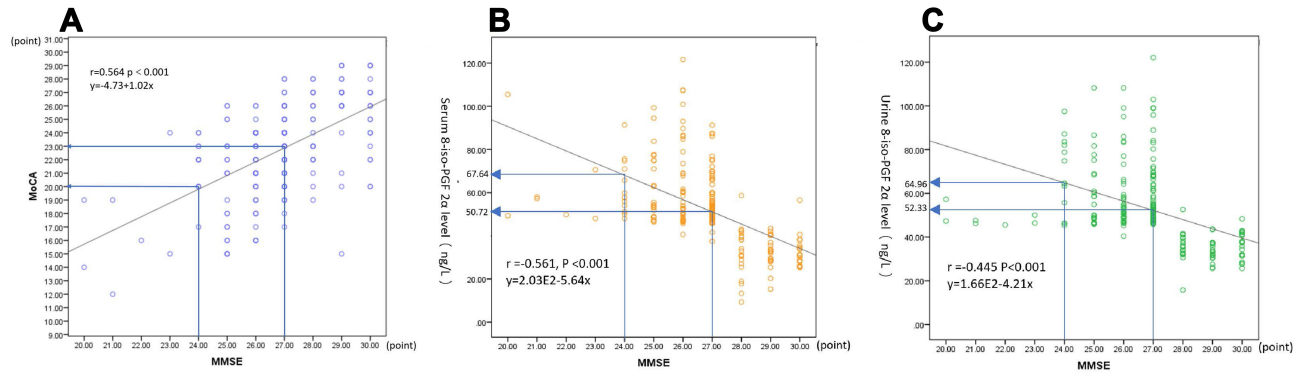
**Note:** Dependent variable: MMSE.

protective factor of dyslipidemia, while Phlegm-Dampness is the risk factor of dyslipidemia.<sup>38</sup> We studied the relationship between TCM constitutional characteristics and related biomarkers and cognitive abilities in patients with cognitive impairment to help with early identification and intervention of MCI.

Due to the relative stability of constitution, each living individual presents different physical characteristics. Neutral constitution is an ideal type. With the increase of age, the body's Qi gradually decreases. Qi-Deficiency prevails in old age.<sup>30</sup> This study found that the proportion of Qi-Deficiency in MCI group was similar to that in normal NC group, indicating that Qi-Deficiency should be the common feature of unneutral type in this age stage. Our study showed that the ORs of three unneutral constitutions of Yang-Deficient, Phlegm-Dampness and Blood-Stasis and abnormal Hcy elevation ( $>10 \mu\text{mol/L}$ ) were significantly greater than 1, which resulted in significantly higher risk of MCI than in other constitutions and normal Hcy level ( $\leq 10 \mu\text{mol/L}$ ), indicating that people both with Yang-Deficient, Phlegm-Dampness, Blood-Stasis, and abnormal Hcy elevation were more likely to

suffer from MCI. The performance of Yang-deficiency: cold hands and feet, fear of cold, pale face, prefer hot food and drink, fat and tender tongue with tooth print and white coating, deep and weak pulse. The performance of Phlegm-dampness: overweight with abdomen fatness, more phlegm and sticky sensation in mouth, slippery and fat tongue with thick white coating, tightness in the chest, laziness prone to lying down, light yellow complexion, more oil on the face, greasy and fat tongue with white coating. The performance of Blood stasis: dark complexion, lips and eye sockets, roughness of skin, all kinds of pain.<sup>46</sup>

Measurement of relevant biomarkers can help to intervene in early cognitive impairment.<sup>47,48</sup> Hcy is a sulfur-containing nonessential amino acid, which participates in the metabolism of cysteine and methionine.<sup>49</sup> Methionine or cysteine metabolic dysfunction can cause hyperhomocysteinemia, lead to small vessel damage, lead to leukoaraiosis or asymptomatic cerebral infarction, and leukoaraiosis and asymptomatic cerebral infarction are important risk factors for cognitive impairment.<sup>50</sup> Lu et al<sup>51</sup> found that hyperhomocysteinemia promoted the



**Figure 1** (A) Shows scatterplot for MMSE scores and MoCA scores. Pearson correlation coefficients between MMSE scores and MoCA scores are shown for graphs. (B) Shows scatterplots for MMSE cognitive scores and serum 8-iso-PGF 2a. Pearson correlation coefficients between MMSE scores and serum 8-iso-PGF 2a are shown for graphs. (C) Shows scatterplots for MMSE cognitive scores and urine 8-iso-PGF 2a. Pearson correlation coefficients between MMSE scores and urine 8-iso-PGF 2a values are shown for graphs.

appearance of white matter damage symptoms and was closely related to the degree of lesions. Studies have shown that hyperhomocysteinemia is a risk factor for cognitive dysfunction.<sup>52–55</sup> Annerbo et al found that plasma homocysteine levels were significantly correlated with the risk of conversion to AD in patients with MCI. The plasma homocysteine levels in patients with MCI conversion to AD were higher than those in untransformed patients,<sup>56</sup> suggesting that the conversion of MCI to AD may depend on the deposition of the toxic metabolite homocysteine. At the same time, this study found that Hcy abnormality is related to the onset of MCI, similar to previous studies.

Several studies<sup>57–61</sup> have shown that in the process of AD and oxidative stress, the accumulation of oxidative damage to proteins, DNA and fat is considered to be an important factor in the pathogenesis of AD. Reactive oxygen species cause non-enzymatic peroxidation of polyunsaturated fatty acids, producing different peroxidation products. Many lipid hydroperoxides, including oxidized phospholipids (oxPLs), are considered to be biologically relevant lipid signaling molecules. 8-isoprostane (8-IsoP) is used for high-throughput analysis of oxPLs and is a biomarker of oxidative stress.<sup>62</sup> Pratico et al<sup>63</sup> found that cerebral oxidative stress was enhanced in patients with MCI, and levels of 8,12-iso-iPF (2 $\alpha$ )-VI were detected in plasma, cerebrospinal fluid, and urine, indicating a progression to AD trend. This study found that MMSE, MoCA scores were negatively correlated with serum Hcy and serum/urine 8-iso-PGF 2 $\alpha$ , among which MMSE scores were moderately correlated with serum/urine 8-iso-PGF 2 $\alpha$ , while MMSE scores were weakly correlated with serum Hcy. MoCA scores were weakly correlated with serum Hcy and serum/urine 8-iso-PGF 2 $\alpha$ . It is suggested that the level of serum and urine 8-iso-PGF2 $\alpha$  has a certain relationship with MCI. Most patients with AD were diagnosed during dementia, but the irreversible damage was found in the neurons of the brain. In the MCI stage, biomarkers can be used to identify potential clinical causes and to evaluate the severity of the disease. In the MCI period, timely and accurate diagnosis and treatment will hopefully delay or prevent further development of AD. Therefore, it is very important to detect biomarkers. At the same time, the idea and practice of combining TCM Constitution with modern medicine is forming a medical and health innovation system.<sup>64</sup> Because the two medical systems are complementary, the

identification of TCM constitution of MCI is helpful for the subsequent development of personalized treatment.

## Conclusion

We found that MCI patients were dominated by the constitution of Yang-Deficient, followed by Phlegm-Dampness, Blood-Stasis, and abnormal Serum Hcy elevation. Among MCI patients, there was a negative correlation between urine/serum 8-iso-PGF 2 $\alpha$  and the MMSE, the MoCA scores had a positive correlation with the MMSE. Higher levels of serum/urine 8-iso-PGF2 $\alpha$  were more likely to be associated with risk of MCI. From the perspective of the combination of disease and TCM Constitution, combined with modern research methods and objective indicators, we searched for MCI susceptible constitution and investigated biomarkers to provide the basis for the identification of MCI and the possibility of specific individual diagnosis and treatment. However, there are still many problems to be further verified, such as whether there is a corresponding relationship between clinical research and basic research. Because the research on constitution of traditional Chinese medicine is still a relatively new subject, the research on the correlation between constitution and diseases outside China is still very little. The MCI patients in this study were all from China. Therefore, it is not clear whether the conclusions drawn in this study can be extended to other populations.

## Data Sharing Statement

All participants provided informed consent. The data analyzed here have not been made publicly available yet. The data analyzed will be made available from the corresponding author on reasonable request in 2-3 years. Corresponding author Lihua Zhao, E-mail: zhaolh67@163.com.

## Ethical Consideration

This study was conducted in accordance with the Declaration of Helsinki. All the subjects provided informed consent according to a protocol approved by the Research Ethics Committee of the First Affiliated Hospital of Guangxi University of Traditional Chinese Medicine (Lot number: [2016] 009). The study was registered in <http://www.chictr.org.cn>. The Clinical Trial Registration Number was ChiCTR-IPR-16009144.

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## Disclosure

The authors declare no conflicts of interest.

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