Dietary pattern and antioxidants in plasma and erythrocyte in patients with mild cognitive impairment from China

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Abstract

**Background:** Oxidative stress and unhealthy dietary pattern are suggested correlating with the risk of mild cognitive impairment (MCI) patients to develop Alzheimer’s disease (AD).

**Objective:** To explore the association between dietary pattern, plasma and erythrocyte antioxidants levels and cognitive function in the older Chinese adults.

**Design:** The present study is a case-control study. 138 MCI patients and 138 age-, gender-matched healthy subjects (aged from 55 to 75) were recruited in community.

**Method:** Food frequency questionnaire (FFQ) method was used for dietary survey. Peripheral blood and morning spot urine were sampled for parameters detection. Cognitive function of the old subjects was measured by using Montreal Cognitive Assessment (MoCA) test. Antioxidant parameters in plasma, erythrocyte and urine samples were measured by using the assay kits. Plasma retinol, α-tocopherol and flavonoids contents were detected by using HPLC and liquid chromatography mass spectrometer methods respectively.

**Results:** The MCI patients have lower plasma total cholesterol, low density lipoprotein cholesterol (LDL-C) and high density lipoprotein cholesterol (HDL-C) than control subjects ($P < 0.01$). MCI patients consumed less fish and more red meat daily than the controls ($P < 0.05$). Comparing with controls, lower plasma total antioxidant capacity (T-AOC), α-tocopherol and higher level of malondialdehyde were detected in the MCI patients ($P < 0.05$). No significant difference of plasma flavonoids concentration, erythrocyte antioxidant enzyme activities, and urinary
8-OH-dG content was detected among the MCI and control subjects \( (P > 0.05) \).

**Conclusion:** Lower plasma concentration of HDL-C, T-AOC and \( \alpha \)-tocopherol levels, and dietary pattern low in fish, high in red meat might contribute to the cognition impairment in older Chinese adults.

**Key words:** diet pattern; antioxidant; cognitive function; mild cognitive impairment; the elderly

**Running title:** Dietary pattern, antioxidant and mild cognitive impairment

**Word count:** 5357

**Number of table:** 4
Introduction

Mild cognitive impairment (MCI) was defined as an isolated deficit in recent memory and it is frequently associated with decline to Alzheimer’s disease (AD) [1]. It is reported that subjects with MCI have a 10-fold increased risk of developing AD at a rate of 10% - 15% per year compared with 1% - 2% per year in the general population [2]. Up to date, there was no efficient medicine or method available for the treatment of AD. Thus, early intervention of subjects with MCI could postpone or prevent the onset of subsequent dementia [3]. As a result, it is critical to identify potentially protective or risk factors to prevent the MCI patients from further developing into AD.

It is suggested that oxidative stress plays an important role in the pathogenesis of AD. Cerebral tissue appears to be particularly vulnerable to free radical damage because of its low content of antioxidants, high content of polyunsaturated fatty acids of neuronal membranes [4]. Since the neuropathology of many neurodegenerative diseases has been linked to the increase of oxidative stress in brain, strong efforts have been directed to exploring the antioxidant strategies to combat neuronal damage. Recent published documents indicated that plasma levels of antioxidants are inversely correlated to the oxidative damage level in AD [5]. Therefore, the measurement of peripheral antioxidants is considered as an appropriate way of measuring oxidative stress in various disease states in human.

Significant correlation between healthy diet pattern and lower risk of MCI or AD have been identified [6,7]. This conclusion was further proved by the research of the
Mediterranean diet (MeDi) pattern, which was characterized by high consumption of plant foods (such as vegetables, fruits, legumes and cereals), high intake of olive oil, moderate intake of fish, low-to-moderate intake of dairy products and low intake of saturated fats and meat. Adherence to the Mediterranean dietary pattern is associated with slower cognitive decline and reduced Alzheimer’s disease risk [8]. Our previous study also demonstrated that adequate intake of fruit and vegetables, marine products probably reduce the risk of developing MCI in old adults [9].

As non-nutritional phytochemicals existing in plant derived foods (such as fruit and vegetables, green tea, and cocoa), flavonoids were proved exhibiting strong antioxidant capacity in vivo and in vitro [10-12]. In addition, a growing number of flavonoids have been proved to inhibit the development of AD-like pathology and to reverse deficits in cognition in rodent models, suggesting the potential therapeutic utility in dementia [13]. Cross-sectional and longitudinal studies also inditated that a higher intake of flavonoids from foods might be associated with a better cognitive evolution [14,15]. However, up to date, limited studies explored the association of plasma flavonoids concentration with cognition in the old population. Besides, the correlation between plasma and erythrocyte antioxidants with cognition of the elderly is controversial. Therefore, in the present study, a population-based case-control study was carried out aiming to explore the association between dietary pattern, plasma and erythrocyte antioxidants levels, plasma flavonoids concentration and cognitive function impairment in MCI patients.

Methods
Participants

The study protocol was approved by the Human Ethics Committee of the Capital Medical University (No. 2012SY23). The mild cognitive impairment (MCI) patients and control subjects were recruited from the community dwellers aged 55-75 who took part in our previous population-based cross-sectional study. Medical doctors interviewed the participants face-to-face in the Nanyuan Community Health Service Center, Fengtai District, Beijing, China. Criteria for exclusion of the subjects were conditions known to affect biological variables of oxidative stress or cognitive function (e.g., inflammatory diseases, recent history of heart or respiratory failure, chronic liver or renal failure, malignant tumors, a recent history of alcohol abuse, history of cerebral apoplexy, history of cerebral infarction, antioxidant supplementary). The subjects with AD or Parkinson’s disease (PD), or not completing the Montreal Cognitive Assessment (MoCA) test or with long-term frequency antidepressants and central nervous system acting medications intake were excluded from the study. All participants (or legal guardian of MCI patients) signed the consent for the participation in this study.

Dietary assessment

Participants were visited at community health service center by specifically trained dieticians. A validated self-administered semi-quantitative food frequency questionnaire (FFQ) was used to assess the habitual consumption of 13 food groups (fruit and vegetable, whole grain, legume and legume product, red meat, light meat, fish, eggs, nuts, cooking oil, milk, coffee, fruit and vegetable juice and tea, totally
including 41 items). This questionnaire was adapted from the questionnaire used for the Dietary Investigation of Chinese Residents, which was organized by the Chinese Nutrition Society (CNS) in 2010. Foods intake survey collected the information including the consumption frequencies (daily and weekly) and the quantity of foods consumed.

**Cognitive assessment**

Cognitive and functional status was assessed by Montreal Cognitive Assessment (MoCA) by trained investigators within 15 minutes to complete. The test has also been broadly used in other large-scale studies on cognitive function in the elderly. According to previous study conducted in Chinese older population [16], the cut-off points used for MCI diagnosis were as follow: 13/14 for individuals with no formal education, 19/20 for individuals with 1 to 6 years of education, and 24/25 for individuals with 7 or more years of education. The cut-offs above were proved sensitive and efficient in the diagnosis of MCI in Chinese older population. If the participants were presumed MCI patients according to MoCA scores, we would arrange them to see the neurologist who would define MCI ultimately according to additional cognitive assessments including Clinical Dementia Rating (CDR), Neuropsychiatric Inventory (NPI) and Hamilton (HAMD)/Geriatric Depression Scale (GDS) and development of the disease. Additionally, age- and gender-matched health subjects were also recruited from the community and served as control group.

**Sociodemographic variables and anthropometric measurements**

Anthropometric measures (height and weight) were made by the nurse in the
community health service center. Body mass index (BMI) was calculated as weight (kg)/height (m)$^2$. Information on demographic characteristics (for example, age, education and marital status), lifestyle factors (for example, alcohol drinking and smoking), medical history of chronic diseases (e.g., hypertension, diabetes, coronary heart disease or cerebrovascular disease) and dietary supplements were collected using a self-administered questionnaire. Educational level was assessed as the highest level ever reached and was classified into four categories (illiterate, primary school, junior high school, and > high school).

**Sample collection**

Fasting venous blood samples were collected from each subject. The blood collected in heparinized tubes was centrifuged at 480 g for 20 min. The plasma was then removed (top layer) and transferred to storage tubes. Morning urine samples were collected after overnight fasting for determination of 8-hydroxy-2’-deoxyguanosine (8-OH-dG) and creatinine contents. All samples were stored frozen at -80°C until analysis.

**Plasma and erythrocyte antioxidant biomarker measurement**

Plasma total antioxidant capacity (T-AOC), malondialdehyde (MDA), glutathione (GSH) levels, erythrocyte glutathione peroxidase (GSH-Px), glutathione S-transferase (GST), glutathion reductase (GR), catalase (CAT), superoxide dismutase (SOD) enzyme activities were measured using commercial assay kits (Nanjing Jiancheng Biotechnology Institute Co., Ltd., Nanjing, China) according to the manufacturer’s instruction. Three independent measurements were performed for each sample.
Measurement of urinary 8-hydroxy-2’-deoxyguanosine content

Urine specimens were centrifuged at 3000 g for 10 min, and the supernatant fraction was used for the measurement of the 8-hydroxy-2’-deoxyguanosine (8-OH-dG) concentrations using a competitive in vitro enzyme-linked immunosorbent assay (ELISA) kit (Cell Biolabs, Inc., San Diego, CA, USA). Three independent measurements were performed for each sample. Results were calculated as ng/mmol creatinine.

Measurement of plasma vitamin and flavonoids levels

After extraction with ethanol and hexane, plasma retinol and \( \alpha \)-tocopherol were determined by using HPLC with UV detection at 280 nm with a Waters Simmetry C8 column (150mm × 4.6 µm) according to the method described by Nierenberg [17]. Retinol and \( \alpha \)-tocopherol standards were purchased from Sigma Aldrich (USA).

Liquid chromatography mass spectrometer (LC-MS) method was applied to measure the flavonoids content in plasma. Standards, \( \beta \)-glucuronidase (with sulfatase contamination), all the extraction and solvents were purchased from Sigma-Aldrich (USA). The measurement of plasma flavonoids was following the method described by Day and Xiong [18,19].

Statistical analyses

Data were analyzed with the software SPSS 13.0. All continuous variables are presented as means ± SD. Age, gender, living status, education level, alcohol drinking, milk drinking, coffee drinking, fruit & vegetable juice drinking and tea drinking were presented as category variables. Chi-square test was used for category variables.
Significance of continuous variables between the two groups was analyzed by using independent \( t \)-tests or general linear model. Some potential confounding factors including gender, age, BMI and smoking were adjusted when comparing the parameters between groups. Statistical testing was performed at the conventional 2-tailed, and \( P < 0.05 \) was considered to be statistically significant.

**Results**

**Sample characteristics**

Demographic and clinical characteristics of the population studied are presented in Table 1. The demographic characteristics are similar between the groups. Statistical significance of education levels of MCI and control subjects was detected \( (P = 0.008) \). The MCI patients have a relative lower education levels than the control group, the percentage of subjects with primary school education level account for 18.12% and 5.80% respectively in control and MCI groups. And the percentage of subjects with junior high school (49.28%) or high school and above (42.75%) in control group is higher than that in the MCI group (junior high school: 43.48%; high school and above: 36.23%). After adjusting the factors including gender, age, and BMI, the baseline clinical characteristics including TC, LDL-C and HDL-C levels were different between the groups. The subjects in control group have higher serum TC, LDL-C and HDL-C levels than that in MCI group \( (P_{TC} = 0.001, P_{LDL-C} = 0.001, P_{HDL-C} = 0.001) \).

**Dietary pattern**

As shown in Table 2, the daily intake of fruit and vegetables, whole grain, legume and legume products, light meat, nut, egg, and cooking oil of the detected subjects were
similar. However, the control subjects consumed more fish than MCI patients \((P = 0.035)\); and the MCI patients consumed more red meat than the control subjects \((P = 0.001)\).

**Parameters in plasma or urine in the elderly**

As shown in Table 3, after adjusting the factors such as gender, age and BMI, the plasma T-AOC level is higher in the control subjects than that in MCI patients \((P = 0.004)\). Comparing to the controls, a significant increase of plasma MDA level was detected in MCI patients \((P = 0.048)\). Control subjects seem to have higher plasma GSH level than MCI patients, while, no statistical significant difference was detected \((P > 0.05)\). There was no different of erythrocyte SOD, CAT, GSH-PX, GST, GR enzyme activities \((P > 0.05)\) between the groups. No difference of urinary 8-OH-dG content was detected between the groups \((P = 0.06)\).

**Plasma vitamin and flavonoids levels in the elderly**

After adjusting the factors including gender, age, BMI and daily fruit and vegetable consumption, the subjects in the control group have higher plasma \(\alpha\)-tocopherol levels than the MCI patients \((P_{\alpha-VE} = 0.0001)\). The MCI patients and control subjects have similar plasma retinol levels \((P > 0.05)\). No difference of plasma flavonoids levels was detected between the groups \((P > 0.05)\).

**Discussion**

Increasing published documents indicated that unhealthy dietary pattern is correlated with the risk of MCI patients to develop into AD [20]. Therefore, modification of unhealthy dietary pattern accompanying with enhancement of body antioxidant
defense system function suggested a promising method to decrease the risk of MCI [21]. In the present study, we explored the dietary pattern and the plasma and erythrocyte antioxidants profile in MCI patients and control subjects. We found that the dietary pattern and body antioxidants profile in the MCI patients differ to that of the control subjects.

Abnormality of serum lipid/lipoprotein was suggested a potentially modifiable risk factor for cognitive disorders [22,27]. However, the study conducted in Chinese nonagenarians and centenarians indicated that the levels of serum lipid/lipoprotein were not directly correlated with cognitive impairment [28]. Meanwhile, study carried out in subjects aged 85 or above reported that cognitive impairment was only associated with the levels of plasma HDL-C, and was independent of plasma TC, TG, and LDL-C levels [29]. In the present study, we found that the control subjects have the higher plasma HDL-C than the MCI patients. We also found that control subjects have higher plasma levels of TC and LDL-C levels than MCI patients. Our results are consistent with previous studies, supporting the association of abnormality of plasma lipid/lipoprotein with impaired cognitive function [30].

Comparing with control subjects, a significant increase of plasma MDA levels was detected in the MCI patients (Table 2). Epidemiological studies indicated a direct relationship between cognitive performance and plasma levels of markers of lipid peroxidation in human [31,32]. The markers of lipid peroxidation, like MDA, have been found elevated in MCI patients [33]. These results are in agreement with our and other’s studies, suggesting that increased plasma MDA might be a useful oxidation
marker in MCI patients [34-36].

Rinaldi [37] reported that, in MCI patients, lower peripheral levels and activities of antioxidant (e.g., vitamins, SOD and GSH-Px enzyme activities) were detected. In the current study, we only detected the lower T-AOC in MCI patients, however, no difference of plasma GSH, retinol, erythrocyte SOD, CAT, GSH-Px, GST, GR enzyme activities was detected in the MCI patients and control subjects. Some researchers reported a decreased antioxidant enzyme activity in AD [38]; while others did not observe any differences or reported an increase in their activity [39]. Our results are consistent with previous studies [40,41]. In these studies, the authors found that erythrocyte GSH-Px, SOD enzyme activities were similar in control subjects and AD patients, and however, increased plasma GSH-Px and SOD enzyme activities were detected.

Comparing with control group, decreased α-tocopherol levels were found in MCI patients. The results are in line with Rinaldi and Mangialasche’s studies [37,40]. It is well known that vitamin E is the most powerful antioxidant. Recent study also indicated that serum tocopherol status is inversely associated with cognitive performance in the elderly [42,43]. Lower levels of plasma antioxidant vitamins in MCI patients could have been the consequence of lower intakes of these nutrients. Here, the consumption of antioxidant vitamin containing foods, such as nuts, cooking oil and meat was similar in the investigated MCI patients and control subjects (Table 2). Additionally, no difference of urinary 8-OH-dG levels was found between MCI patients and the controls. It is thus possible that the lower concentration of plasma
α-tocopherol detected in MCI patients was not the consequence of a lower intake. A possible explanation for these results might be: instead of MCI patients being recruited from the hospital, where the patients commonly characterized with typical cognitive function decline symptoms, the MCI patients of the present study were recruited from the community (the patients are mainly in the very earlier stage of dementia disorder). At this early stage of MCI, the oxidative stress and excessive production of free radicals only consumed the plasma non-enzymatic antioxidants such as vitamin E, causing the decline of plasma T-AOC, without a simultaneous activation of the antioxidant enzymes in red blood cells.

Accumulated evidence suggests that naturally occurring antioxidants, such as polyphenols found in fruits, vegetables, and nuts, may potentially inhibit neurodegeneration, and improve memory and cognitive function [44,45]. Flavonoids, as the largest group of polyphenols, recently have been reported to exert their neuron protective actions through the potential to protect neurons against injury induced by neurotoxins and to promote memory, learning, and cognitive function [47,48]. A 10-year prospective cohort study among subjects aged 65 years or older indicated that flavonoids intake was associated with a better cognitive performance at baseline and with a better evolution of the performance over time [49]. As shown in Table 4, we did not detect significant difference of plasma flavonoids between MCI patients and control subjects. This finding is consistent with our nutrition investigation results, in which we found that the MCI patients have a similar daily consumption of flavonoid-rich foods (such as fruit and vegetable) to control subjects. Although, more
and more evidences do suggest that dietary derived flavonoids contribute to the benefits of a high intake of fruit and vegetables on cognitive function [50]. However, their contribution in vivo and their physiologically relevant concentrations remain uncertain. Besides, the limited bioavailability of these substances is often overlooked, particularly in some animal experiment and in vitro work. Recent in vivo experiments also suggested that the bioflavonoid metabolites showed higher antioxidant activity against various oxidative systems than the parent flavonoids molecule [51-54]. Therefore, further research is needed to uncover the relationship between dietary flavonoids intake and cognitive function in the elderly.

Another finding of the present study is that daily fish consumption was less than the control subjects. This result is in agreement with our previous cross-sectional study [9], in which we found the inverse correlation of marine products consumption with MoCA scores in the older Chinese adults. Long chain unsaturated fatty acids (such as docosahexaenoic acid, DHA) derived from fish or marine products might contribute to the cognition promoting effects of fish or marine products rich diet [55]. Our further detection of long chain unsaturated fatty acids content in erythrocyte membrane also indicated that MCI patients have the lower erythrocyte membrane DHA and eicosapentaenoic Acid (EPA) content than that in control subjects (data not shown here). A recent meta-analysis indicated that a higher intake of fish was associated with a lower risk of AD [56]. Although the beneficial effects of omega-3 supplement on cognitive outcome were approved by clinical trials in older adults [57]. Long-term cohorts are still needed to explore the underlying mechanism of fish intake
and cognition in the elderly.

Additionally, the MCI subjects consumed more red meat daily than control subjects. Power and co-worker’s study in 208 Community-dwelling subjects found that subjects consuming “prudent” dietary pattern (high in fish, fruit and vegetables, low in red meat) had higher MMSE (Minimum Mental State Examination) scores than the subjects with “Western” dietary pattern (high in red meat, low in fruit and vegetables) [58]. All together, the present study indicated that lifestyle, dietary pattern and body antioxidant involved in the influence of cognition in the elderly. These observational findings suggest that keeping to a dietary pattern of low meat intake, especially red meat (pork, beef and mutton), high fish and antioxidant rich foods intake could prove to be an impact-driven public health policy to support healthy cognitive aging.

Limitations of the current study should also be considered. Considering the small sample number, the results from the current study may have limited representativeness. Although some covariates were adjusted during the data analysis, some residual confounding is possible. In addition, the reliant on memory or cognitive abilities of FFQ method did not allow us to estimate nutrients and flavonoids intake precisely in the present study. Finally, the current sample was constituted of Chinese elderly only. Therefore, extrapolation of the present results to older population in Western countries should be careful.

Conclusion

Our results indicated that the antioxidant profile of MCI (as an early stage of dementia) patients were characterized by the decrease of plasma T-AOC and α-tocopherol
levels, abnormality of serum lipid/lipoprotein and the increase of oxidative stress. Besides, the current study supports a favorable role of dietary pattern with high in fish and low in red meat in deterring cognitive decline in the elderly.

**Acknowledgment**

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**Reference**


[24] Tao QQ, Chen Y, Liu ZJ, Sun YM, Yang P, Lu SJ, Xu M, Dong QY, Yang JJ, Wu ZY. Associations between apolipoprotein E genotypes and serum levels of glucose, cholesterol, and triglycerides in a cognitively normal aging Han


Table 1 Demographic and clinical data in the elderly

<table>
<thead>
<tr>
<th>Demographic and clinical characters</th>
<th>Control (n = 138)</th>
<th>MCI (n = 138)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Demographic</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age, (mean ± SD)</td>
<td>64.23 ± 0.47</td>
<td>64.71 ± 0.52</td>
<td>0.493</td>
</tr>
<tr>
<td>Male gender (%)</td>
<td>35.51</td>
<td>35.51</td>
<td>1.000</td>
</tr>
<tr>
<td>Living along (%)</td>
<td>7.24</td>
<td>2.17</td>
<td>0.085</td>
</tr>
<tr>
<td>Smoking (%)</td>
<td>15.22</td>
<td>15.94</td>
<td>1.000</td>
</tr>
<tr>
<td>Alcohol drink (%)</td>
<td>23.19</td>
<td>24.64</td>
<td>0.888</td>
</tr>
<tr>
<td>Education</td>
<td></td>
<td></td>
<td>0.008</td>
</tr>
<tr>
<td>Illiterate (%)</td>
<td>2.17</td>
<td>2.17</td>
<td></td>
</tr>
<tr>
<td>Primary school (%)</td>
<td>5.80</td>
<td>18.12</td>
<td></td>
</tr>
<tr>
<td>Junior high school (%)</td>
<td>49.28</td>
<td>43.48</td>
<td></td>
</tr>
<tr>
<td>High school and above (%)</td>
<td>42.75</td>
<td>36.23</td>
<td></td>
</tr>
<tr>
<td><strong>Clinical/Biochemical</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>BMI</td>
<td>25.45 ± 0.27</td>
<td>24.95 ± 0.31</td>
<td>0.222</td>
</tr>
<tr>
<td>Glu (mmol/l)</td>
<td>5.72 ± 0.17</td>
<td>5.36 ± 0.15</td>
<td>0.407</td>
</tr>
<tr>
<td>TG (mmol/l)</td>
<td>1.64 ± 0.10</td>
<td>1.59 ± 0.08</td>
<td>0.154</td>
</tr>
<tr>
<td>TC (mmol/l)</td>
<td>4.86 ± 0.09</td>
<td>4.74 ± 0.08</td>
<td>0.001</td>
</tr>
<tr>
<td>LDL-C (mmol/l)</td>
<td>3.32 ± 0.08</td>
<td>3.15 ± 0.07</td>
<td>0.001</td>
</tr>
<tr>
<td>HDL-C (mmol/l)</td>
<td>1.33 ± 0.04</td>
<td>1.29 ± 0.03</td>
<td>0.001</td>
</tr>
</tbody>
</table>

Data were expressed as mean ± SD. or percentage. Abbreviations: BMI: body mass index; Glu: glucose; TG: triglyceride; TC: total cholesterol; LDL-C: low density lipoprotein cholesterol; HDL-C: high density lipoprotein cholesterol. Demographic characteristics were compared by using Chi-square or Fisher’s exact test. ANOVA tests was used for the data analysis of clinical and biochemical parameters.
Table 2 Dietary pattern of the elderly

<table>
<thead>
<tr>
<th>Foods and beverage</th>
<th>Control (n = 138)</th>
<th>MCI (n = 138)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fruit and vegetable (g/d)</td>
<td>456.52 ± 15.37</td>
<td>445.65 ± 14.88</td>
<td>0.391</td>
</tr>
<tr>
<td>Whole grain (g/d)</td>
<td>68.27 ± 4.14</td>
<td>59.73 ± 3.15</td>
<td>0.465</td>
</tr>
<tr>
<td>Legume &amp; legume product (g/d)</td>
<td>64.08 ± 5.10</td>
<td>56.63 ± 4.02</td>
<td>0.770</td>
</tr>
<tr>
<td>Fish (g/d)</td>
<td>46.00 ± 2.80</td>
<td>41.10 ± 2.97</td>
<td>0.035</td>
</tr>
<tr>
<td>Red meat (g/d)</td>
<td>50.95 ± 3.27</td>
<td>58.28 ± 4.16</td>
<td>0.001</td>
</tr>
<tr>
<td>Light meat (g/d)</td>
<td>26.81 ± 2.33</td>
<td>30.33 ± 3.29</td>
<td>0.078</td>
</tr>
<tr>
<td>Nut (g/d)</td>
<td>28.84 ± 2.47</td>
<td>31.0 ± 2.49</td>
<td>0.523</td>
</tr>
<tr>
<td>Egg (g/d)</td>
<td>36.46 ± 2.02</td>
<td>34.32 ± 1.87</td>
<td>0.458</td>
</tr>
<tr>
<td>Cooking oil, (ml/d)</td>
<td>38.69 ± 2.61</td>
<td>37.49 ± 1.88</td>
<td>0.625</td>
</tr>
<tr>
<td>Milk drinking (%)</td>
<td>78.99</td>
<td>81.16</td>
<td>0.763</td>
</tr>
<tr>
<td>Coffee drinking (%)</td>
<td>5.80</td>
<td>7.25</td>
<td>0.808</td>
</tr>
<tr>
<td>FV juice drinking (%)</td>
<td>8.70</td>
<td>14.49</td>
<td>0.188</td>
</tr>
<tr>
<td>Tea drinking (%)</td>
<td>67.39</td>
<td>60.14</td>
<td>0.209</td>
</tr>
</tbody>
</table>

Data were expressed as mean ± SD. and percentage. FV: fruit and vegetable. General line model was used for comparing the mean of daily FV, cooking oil, fish, red meat, light meat, nut, egg, whole grain and legume & legume product. Factors including gender, age, BMI and education level were adjusted during the analysis of data. Chi-square test was applied for the analysis of milk drinking, coffee drinking, FV juice drinking and tea drinking percentage in control and MCI group.
Table 3 Antioxidative parameters in the elderly

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Control (n = 138)</th>
<th>MCI (n = 138)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Plasma</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>T-AOC (U/ml)</td>
<td>12.20 ± 0.33</td>
<td>11.07 ± 0.44</td>
<td>0.004</td>
</tr>
<tr>
<td>MDA (nmol/ml)</td>
<td>5.92 ± 0.46</td>
<td>7.11 ± 0.61</td>
<td>0.045</td>
</tr>
<tr>
<td>GSH (umol/L)</td>
<td>54.74 ± 4.26</td>
<td>43.86 ± 2.89</td>
<td>0.166</td>
</tr>
<tr>
<td><strong>Erythrocyte</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SOD (U/gHb)</td>
<td>31.43 ± 0.35</td>
<td>31.88 ± 0.47</td>
<td>0.062</td>
</tr>
<tr>
<td>CAT (U/gHb)</td>
<td>2.33 ± 0.03</td>
<td>2.34 ± 0.04</td>
<td>0.881</td>
</tr>
<tr>
<td>GSH-Px (U/gHb)</td>
<td>25.71 ± 0.65</td>
<td>25.55 ± 0.64</td>
<td>0.891</td>
</tr>
<tr>
<td>GST (U/gHb)</td>
<td>0.32 ± 0.01</td>
<td>0.35 ± 0.01</td>
<td>0.424</td>
</tr>
<tr>
<td>GR (U/gHb)</td>
<td>0.55 ± 0.02</td>
<td>0.56 ± 0.02</td>
<td>0.404</td>
</tr>
<tr>
<td><strong>Urine</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>8-OH-dG (ng/mmol creatinine)</td>
<td>74.51 ± 5.46</td>
<td>70.75 ± 4.70</td>
<td>0.060</td>
</tr>
</tbody>
</table>

General line model was applied for data analysis. Data were expressed as mean ± SD.

Adjusted factors including gender, age, BMI (body mass index), daily consumption of fruit and vegetable. T-AOC: total antioxidant capacity; MDA: malondialdehyde; GSH: glutathione; SOD: superoxide dismutase; CAT: catalase; GSH-Px: glutathione peroxidase; GST: glutathione S-transferase; GR: glutathion reductase; 8-OH-dG: 8-hydroxy-2’-deoxyguanosine.
### Table 4 Plasma antioxidant level in the elderly

<table>
<thead>
<tr>
<th>Antioxidants</th>
<th>Control (n = 138)</th>
<th>MCI (n = 138)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Vitamin</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Retinol (µg/ml)</td>
<td>0.72 ± 0.03</td>
<td>0.70 ± 0.01</td>
<td>0.122</td>
</tr>
<tr>
<td>α-tocopherol (µg/ml)</td>
<td>11.00 ± 0.31</td>
<td>9.52 ± 0.27</td>
<td>0.001</td>
</tr>
<tr>
<td><strong>Flavonoids</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Luteolin (ng/ml)</td>
<td>403.43 ± 27.48</td>
<td>449.61 ± 31.20</td>
<td>0.623</td>
</tr>
<tr>
<td>Quercetin (ng/ml)</td>
<td>10.74 ± 0.96</td>
<td>12.69 ± 1.45</td>
<td>0.458</td>
</tr>
<tr>
<td>Naringenin (ng/ml)</td>
<td>7.06 ± 0.52</td>
<td>7.53 ± 0.81</td>
<td>0.996</td>
</tr>
<tr>
<td>Apigenin (ng/ml)</td>
<td>216.88 ± 6.45</td>
<td>226.87 ± 9.68</td>
<td>0.568</td>
</tr>
<tr>
<td>Kaempferol (ng/ml)</td>
<td>12.73 ± 1.26</td>
<td>16.19 ± 2.29</td>
<td>0.665</td>
</tr>
<tr>
<td>Hesperetin (ng/ml)</td>
<td>4.72 ± 0.29</td>
<td>5.14 ± 0.46</td>
<td>0.906</td>
</tr>
</tbody>
</table>

General linear model was applied for data analysis. Data were expressed as mean ± SD.

Adjusted factors including sex, age, BMI and daily consumption of fruit and vegetable.
Highlights

> Dietary pattern was detected in MCI and control subjects; > plasma and erythrocyte antioxidant biomarkers were measured; > lower plasma HDL-C, T-AOC and \( \alpha \)-tocopherol levels were found in MCI patients; > MCI patient consumed less fish and more red meat than controls.