

Original Article

Application of ^1H magnetic resonance spectroscopy in the diagnosis and treatment of mild cognitive impairment

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Abstract: Objective: To investigate the role of proton magnetic resonance spectroscopy (^1H -MRS) in the diagnosis and treatment of mild cognitive impairment (MCI). Methods: the MCI patients were randomly divided into experiment group and the control group. When grouping, the patients were performed the Mini-Mental State Examination (MMSE) scoring, ^1H -MRS detection, and calculated the ratios of N-Acetylaspartate/creatine (NAA/Cr), N-Acetylaspartate/myo-inositol (NAA/ml) and choline-containing compounds/creatine (Cho/Cr). The experiment group was orally administrated the huperzine tablet for six months. The changes of MMSE score and ^1H -MRS indicators were observed, and the follow up lasted a year to calculate the conversion ratio of MCI to dementia; according to whether existed the dementia conversion, the control group was divided into two subgroups for the analysis of differences in the ^1H -MRS indicators when grouping. Results: The MMSE score, changes of ^1H -MRS indicators and conversion ratio of dementia of the treatment group were statistically significant to the control group ($P < 0.01$). The dementia-conversion subgroup had the statistical significance in ^1H -MRS indicators than the non-conversion subgroup ($P < 0.01$). Conclusions: Proton magnetic resonance spectroscopy is helpful for the early diagnosis of mild cognitive impairment in dementia patients.

Keywords: Proton magnetic resonance spectrum (^1H -MRS), mild cognitive impairment (MCI), posterior cingulate gyrus, early diagnosis and treatment

Introduction

The prevalence of Alzheimer's disease (AD) is significantly increased with the increase of age. With the improvement of medical treatment and the extension of life expectancy, although AD is easily diagnosed in middle-late stage, there is still no effective curative method for it. Thus, it has become an important public health problem in this century [1]. The current researches show that the early appropriate medications may prevent or delay the progress of AD [2]. Thus, early diagnosis and treatment becomes more and more important. The pathophysiological changes can occur as early as several decades before the clinical diagnosis of AD, and the first change of cognitive function is in the memory domain [3]. Mild cognitive impairment (MCI) refers to a state of cognitive impairment between the mild dementia and

normal elderly, which may progress to dementia in the future [4]. Forgotten MCI represents an early stage of the progression of AD [5]. Early identification of the MCI individuals that may easily progress to AD and giving them effectively intervene treatment may reduce the incidence of AD. Various methods have been studied to diagnose the forgotten MCI, such as by measuring the memory retention of familiar graphics and the words memory damaged [6]. The famous names recognition can be used as an early preclinical cognitive tag to predict the cognitive decline of the healthy old people [7]. ^1H magnetic resonance spectroscopy (^1H -MRS) is a noninvasive detection technology that can position and quantitatively analyze the *in vivo* biochemical metabolic changes in brain tissue *in vitro*. In the early years, ^1H -MRS has been found having great application values in the differential diagnosis of dementia, and it also can

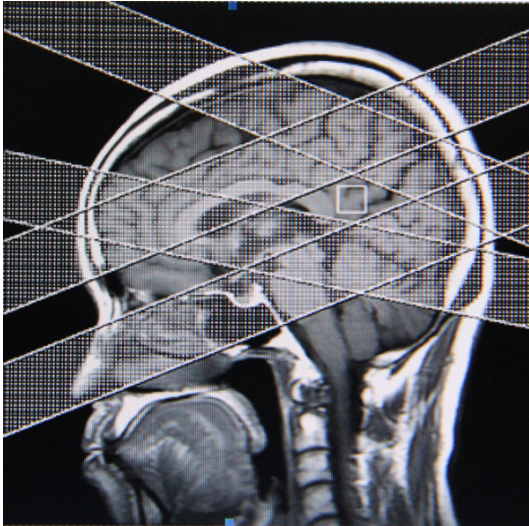


Figure 1. The volume of interest of posterior cingulate gyrus was located with ^1H -MRS.

be used as an index to determine the curative effects of drug treatment for dementia [8]. With the development of the magnetic resonance technology, ^1H -MRS plays more and more important roles in predicting the onset of dementia and in the clinical drug trails for preventing the progress of dementia. In this regard, we detected the metabolic changes in the hippocampus, posterior cingulate gyrus and lobus parietalis of patients with MCI using ^1H -MRS and explored their correlations with the onset of dementia, aiming to find out the biological markers that can identify the patients who are easy to progress into dementia and realize the early diagnosis and treatment of dementia.

Patients and methods

Subjects

40 patients with mild cognitive dysfunction (MCI) were diagnosed and treated in our hospital from June 2012 to June 2015 were enrolled in this study. All patients met the MCI diagnostic criteria proposed by Petersen [9-12] and the MCI diagnostic criteria of Shanghai Mental Health Center, China. The diagnostic criteria of MCI include as following: there are complaints of the memory decline, which can be confirmed by the insiders such as relatives, colleagues, etc. The memory impairment found by objective checks is not consistent with the patient's age or education. The overall cognitive function is normal and the everyday life function remains

normal. The improved Hachinski ischemic scale is 4 scores or less. The patients do not meet the diagnostic criteria of dementia. The cognitive impairment duration is more than 3 months. The exclusion criteria are as following: with brain trauma, cerebrovascular disorder or a history of mental illness, with other severe or unstable diseases that influence the evaluation of the efficacy and safety, with focal or diffuse brain lesions found by routine MRI scans, exposure to a drug that can cause cognitive function changes one month before the treatment.

Patients were randomly divided into experimental group and control group with 20 cases in each group. Patients in the experimental group included 17 males and 3 females, aged from 65 to 86 years (mean age 75 ± 8.7 years). Patients in the control group were consisted of 16 males and 4 females, aged 63 to 82 years (mean age 73 ± 7 years). Patients in both groups had hypertension, coronary heart disease, diabetes and other complications. There were no statistical significance in the patient's age, gender, complications and other general data between the two groups ($P > 0.05$). The informed patient consents were conducted in this study.

Treatment process

Patients in the experimental group received oral administration of 0.1 mg cholinesterase inhibitor huperzine a tablets (Yizhong pharmaceutical factory, Henan Taloph pharmaceutical co., LTD, China) for 6 months, twice one day. Patients in the control group got no anti-dementia drug treatment. All patients received diet, exercise and traditional Chinese medicine treatment. Meanwhile, treatments for hypertensive disease, cardiovascular disease, diabetes and other complications were also carried out, except other dementia drugs. After the treatment, each patient was followed-up for one year.

Observation and evaluation index

Each patient underwent a mini-mental state examination (MMSE) when he/she was enrolled. Hippocampus, posterior cingulate gyrus and lobus parietalis of each patient was scanned by ^1H -MRS using 1.5 T superconducting nuclear magnetic resonance scanner (Signa medow, GE Company, USA) (**Figure 1**). The indexes included N-Acetylaspartate (NAA),

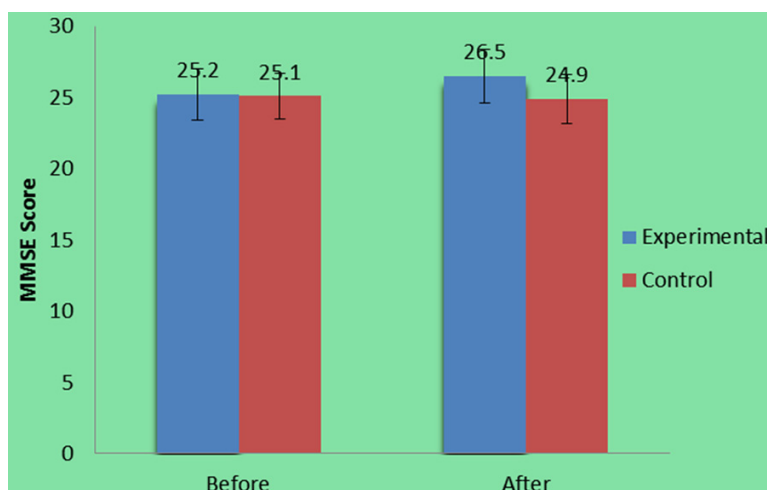


Figure 2. MMSE scores in the two groups before and after treatment. ($n=20$ for each group). Comparison between two groups, before treatment, $t=0.187$, $P=0.854>0.05$; after treatment, $t=2.807$, $P=0.0079$.

myo-inositol (ml), choline-containing compounds (Cho) and creatine (Cr). Elevation of NAA to Cr (NAA/Cr), NAA to ml (NAA/ml) and Cho to Cr (Cho/Cr) was performed. 6 months after the treatment, MMSE and ¹H-MRS were carried out again to evaluate the ratios of NAA/Cr, NAA/ml and Cho/Cr. Patients were then followed-up for one year to calculate the rate of patients developing into dementia. Patients in the control group underwent MMSE and ¹H-MRS examination in parallel with the experimental group. At the end of the follow-up, patients in the control group were subdivided into dementia subgroup and non-dementia subgroup and their initial ¹H-MRS indexes were retrospectively analyzed.

Statistical analysis

Data were analyzed using SPSS 20.0 statistical software. Comparisons of measurement data between two groups were carried out using students-*t* test and comparisons of numeration data between two groups were performed using Chi-square test. $P<0.05$ was considered as statistically significance.

Results

MMSE scores in the two groups before treatment

As shown in **Figure 2**, the MMSE scores showed no statistically significant difference between

the two groups before any treatment ($P>0.05$).

¹H-MRS indexes in the two groups before treatment

As shown in **Table 1**, the NAA, Cr, ml and Cho of the hippocampus, posterior cingulate gyrus and lobus parietalis of each patient was detected by ¹H-MRS when patients were enrolled in this study and then the ratios of NAA/Cr, NAA/ml and Cho/Cr were calculated. There was no statistically significant difference of each index between the two groups ($P>0.05$).

MMSE scores in the two groups after treatment

As shown in **Figure 2**, after treatment with huperzine a tablets, MMSE scores in the experimental group were significantly improved and higher than that in the control group, which was not treated with any anti-dementia drug ($P=0.0079$).

¹H-MRS indexes in the two groups after treatment

As shown in **Table 2**, after treatment with huperzine a tablets, the ratios of NAA/Cr and NAA/ml in the hippocampus and posterior cingulate gyrus in the experimental group were remarkably higher than those in the control group, respectively, while the ratio of Cho/Cr was obviously lower than that in the control group ($P<0.05$). However, the ratios of NAA/Cr, NAA/ml and Cho/Cr in the lobus parietalis were very close between the two groups, respectively ($P>0.05$).

The rate of patients developing into dementia after one-year's follow-up

As shown in **Table 3**, after one-year's follow-up, there was only one patient in the experimental group developing into dementia, less than that in the control group, in which there were four patients developing into dementia, but it was not statistically significant. It may be associated with the small sample size.

Table 1. ¹H-MRS indexes in the two groups before treatment

Group	No. of cases	Hippocampus			Posterior cingulate gyrus			Lobus parietalis		
		NAA/Cr	NAA/ml	Cho/Cr	NAA/Cr	NAA/ml	Cho/Cr	NAA/Cr	NAA/ml	Cho/Cr
Experimental	20	1.67±0.15	2.41±0.32	1.06±0.13	1.49±0.08	2.23±0.26	1.08±0.16	1.74±0.11	2.39±0.31	0.98±0.12
Control	20	1.68±0.14	2.43±0.35	1.07±0.14	1.51±0.11	2.25±0.28	1.09±0.14	1.76±0.12	2.37±0.33	0.97±0.10
t value		-0.2180	-0.1886	-0.2341	-0.6576	-0.2341	-0.2104	-0.5494	0.1975	0.2863
P value		0.8286	0.8514	0.8162	0.5148	0.8162	0.8345	0.5859	0.8445	0.7762

Table 2. ¹H-MRS indexes in the two groups after treatment

Group	No. of cases	Hippocampus			Posterior cingulate gyrus			Lobus parietalis		
		NAA/Cr	NAA/ml	Cho/Cr	NAA/Cr	NAA/ml	Cho/Cr	NAA/Cr	NAA/ml	Cho/Cr
Experimental	20	1.65±0.13	2.38±0.30	1.08±0.12	1.47±0.09	2.18±0.24	1.12±0.14	1.72±0.12	2.37±0.32	0.99±0.12
Control	20	1.52±0.15	2.12±0.32	1.17±0.14	1.38±0.12	1.94±0.26	1.23±0.16	1.69±0.13	2.31±0.31	1.02±0.10
t value		2.9289	17.2306	-2.1828	2.6833	3.0334	-2.3139	0.7583	0.6023	-0.8589
P value		0.0057	0.0000	0.0353	0.0107	0.0043	0.0262	0.4529	0.5506	0.3958

Table 3. The rate of patients developing into dementia after one-year's follow-up

Group	No. of cases	Patients developing into dementia (cases)	Percentage (%)
Experimental	20	1	5%*
Control	20	4	20%

*P=0.339, Correction $\chi^2=0.914$, compared with the control group.

Retrospective analysis of the initial ¹H-MRS indexes of the patients in the control group when they were divided into dementia subgroup and non-dementia group after one-year's follow-up

After one-year's follow-up, 4 patients in the control group developed into dementia. Then the initial ¹H-MRS indexes of patients were analyzed when they were enrolled. The results showed that the ratios of NAA/Cr and NAA/ml of the hippocampus and posterior cingulate gyrus in the dementia subgroup (**Figure 3**) were remarkably lower than those in the non-dementia subgroup (**Figure 4**), respectively ($P<0.05$, **Table 4**). However, the ratio of Cho/Cr of the hippocampus and posterior cingulate gyrus in the dementia subgroup were significantly higher than those in the non-dementia subgroup, respectively ($P<0.05$, **Table 4**).

Discussion

As a noninvasive detection technology, proton magnetic resonance spectroscopy can detect the levels of n-acetyl aspartate (NAA), choline

compounds (Cho), creatine compounds (Cr), glutamic acid compounds, inositol and other compounds in the interested area, providing more valuable information for studying the physiological and biochemical process of the human brain. NAA only exists in the surviving neuronal cell bodies and neuraxon, and its level can reflect the change of neuron activity and the number of neurons [13]. Thus, it is regarded as the best marker for indicating the deletion or reducing activity of neurons and has been widely applied. Cr is a cushioning substance of energy metabolism, including creatine and phosphocreatine. It is relatively stable *in vivo* in various states including in the pathological conditions. Thus, it is always used as an inner reference [14]. Cho, including choline phosphate, choline glycerophosphatide (GPC) and phosphatidyl choline etc., is involved in the composition of the cell membrane and the myelinogenesis of nerve cells. The abnormal degradation, repair and synthesis of cell membrane and the production of amyloid can increase the production of Cho [15]. ml exists only in glial cells, whose proliferation as well as the activation of phospholipase C can cause the rise of ml. Thus, it can be used as the sign of the glial cells. There are selective metabolic abnormalities in the brain of AD patients. MRS examination can show the decrease of NAA and the increase of ml in the cerebral cortex with defects or declined activity of neuron [16]. Two-thirds of the AD patients were developed from patients with MCI [17] and forgotten MCI attracts the most attention because there are about 10% to 15% of the patients with forgotten MCI develop-

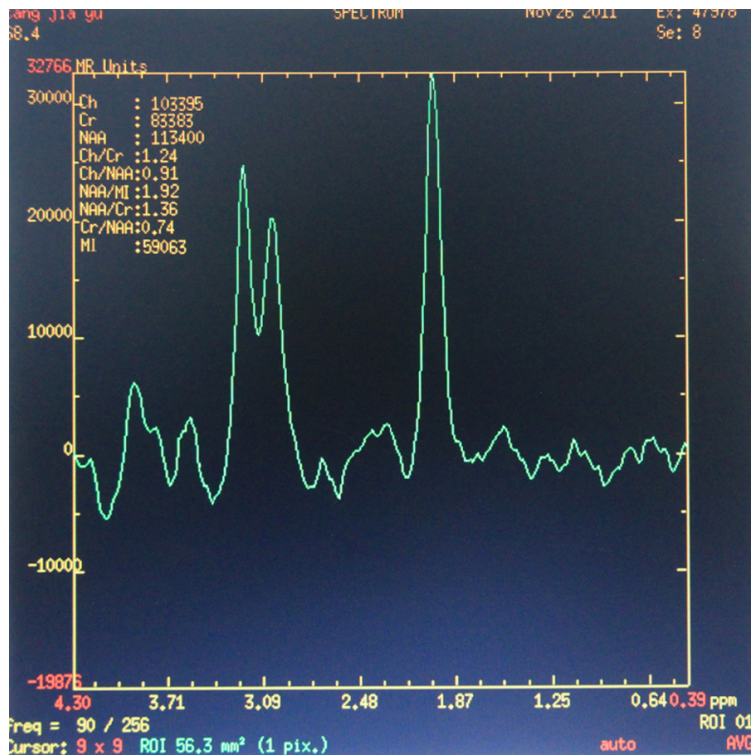


Figure 3. ^1H -MRS detection of posterior cingulate gyrus for patients developed into dementia. NAA/Cr, NAA/ml decreased and Cho/Cr ratio increased obviously.

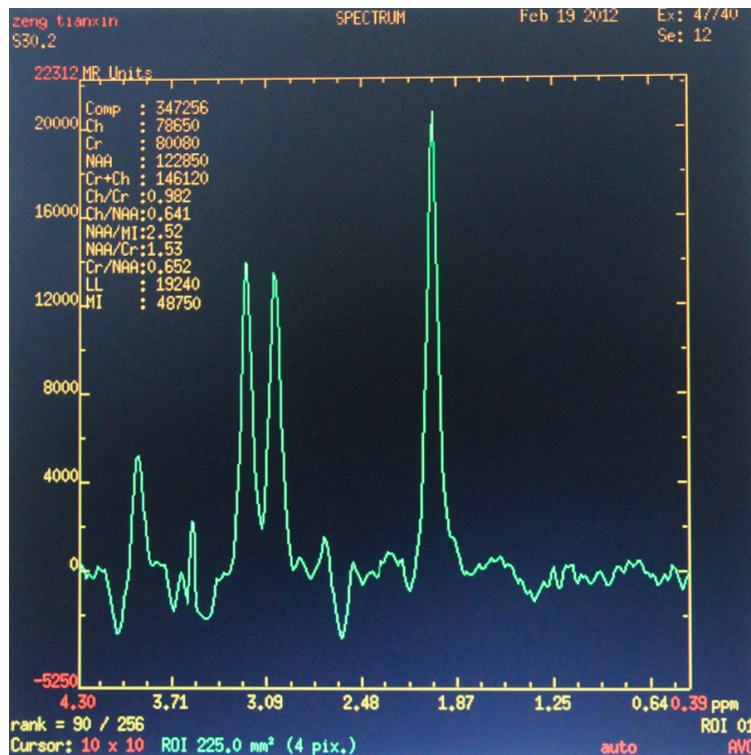


Figure 4. ^1H -MRS detection of posterior cingulate gyrus for patients undeveloped into dementia. NAA/Cr and NAA/ml were decreased and Cho/Cr ratio was slightly increased.

ing into dementia each year. If they were followed-up for 6 years, there would be 80% of them developing into dementia [18]. Thus, MCI, especially the forgotten MCI, is considered as the early state of AD. We predicted that the patients who suffered from the memory impairment would be easy to converse into the dement, and exhibited the metabolic abnormalities in such memory-related structures as posterior cingulate gyrus and hippocampus, the ^1H -MRS detection might find the regular changes of some indicators. The results of this project were as we had predicted, showing the easy-dementia-conversion MCI patients had the significant changes in such ^1H -MRS detection indicators as posterior cingulate gyrus and hippocampus, thus it provided a better objective indicator for the early diagnosis and timely intervention of dementia.

In terms of the cognitive function, MCI have many similarities with the preclinical AD, such as the earliest characteristics of impaired memory test. Cholinergic system is involved in the regulation of neuronal excitability, cortical plasticity and the process of learning and memory of mammalian, which is closely related to the cognitive function of the brain. Posterior cingulate gyrus is an integral part of the cholinergic fiber system, whose metabolic changes would lead to cognitive function impairment. Our results showed that cholinesterase inhibitor treatment for the patients with MCI in the experimental group significantly improved the MMSE scores but slightly reduced NAA and slightly enhanced ml and Cho in the hippocampus and posterior cingulate gyrus,

Table 4. Comparison of the initial ¹H-MRS indexes between the dementia subgroup and non-dementia subgroup

Group	No. of cases	Hippocampus			Posterior cingulate gyrus			Lobus parietalis		
		NAA/Cr	NAA/ml	Cho/Cr	NAA/Cr	NAA/ml	Cho/Cr	NAA/Cr	NAA/ml	Cho/Cr
Dementia	4	1.55±0.14	2.14±0.25	1.17±0.10	1.39±0.11	1.97±0.24	1.21±0.08	1.70±0.12	2.35±0.32	0.99±0.11
Non-dementia	16	1.71±0.12	2.50±0.26	1.04±0.09	1.54±0.12	2.32±0.26	1.06±0.12	1.78±0.11	2.38±0.31	0.96±0.09
t value		-2.3164	-2.4926	2.5348	-2.2664	-2.4383	2.3474	-1.2809	-0.1722	0.5732
P value		0.0325	0.0227	0.0207	0.0360	0.0253	0.0305	0.2165	0.8652	0.5736

resulting in the ratios of NAA/Cr and NAA/ml decreased slightly but Cho/Cr ratio increased slightly. In the control group, NAA also decreased but ml and Cho increased, leading to NAA/Cr and NAA/ml ratios decreased and Cho/Cr ratio increased significantly. The NAA, ml and Cho in the lobus parietalis of the experimental group and their NAA/Cr, NAA/ml, Cho/Cr ratios did not change prominently, showing no obvious difference before and after the treatment. Kantarci K et al. [19] reported that NAA/Cr ratio decreases in the posterior cingulate gyrus of MCI and AD and believed that the decrease of NAA/Cr predicts a shift of MCI to AD. ml only exists in the living glial cells. It can be used as a glial marker of brain tissue and its increase is considered to be an indication of glial proliferation. The onset of AD is accompanied with lardacein deposition and nerve fiber entanglement in the brain. Due to the widespread loss of neurons and the impairment of cholinergic, 5-HT and adrenal mutual neurotransmitter systems, as well as the phosphoinositide hydrolysis when muscarinic cholinergic receptor and α -adrenergic receptor are activated, the resynthetic process of phosphatidylinositol from inositol and other substances should be sped up so as to maintain the function of the receptors and thereby leading to the increase of ml in the brain of the patients. When AD happens, the inhibitory enzymes that transform inositol into phosphoinositide may be elevated, further leading to the increase of ml in the brain [20]. Therefore, both the decrease of NAA and the rise of ml are important indexes of metabolic changes of neurons. In this study, we chose NAA/ml as an important indicator to detect the metabolic changes in MCI earlier and more sensitively. Kantarci K et al. [19] also believed the rise of Cho/Cr is an indicator of the progress of MCI. Our study showed that the rate of patients developed into AD in the experimental group was less than that in the control group, but it was not statisti-

cally significant, which may be associated with the small size of samples. Then we retrospectively analyzed the initial ¹H-MRS indexes of the control group and found that NAA decreased but ml and Cho enhanced in the hippocampus and posterior cingulate gyrus of the patients developing into dementia, leading to NAA/Cr and NAA/ml decreased and Cho/Cr increased significantly, especially in the posterior cingulate gyrus. Therefore, ¹H-MRS can be used to early find out the patients with MCI who are easy to develop into dementia and helps to early diagnosis and treatment of dementia. It also can be used to detect NAA, ml, Cho, Cr and other indicators in the hippocampus and posterior cingulate gyrus of the healthy old people to screen MCI, so that it can be treated early and the incidence of dementia can be reduced. Recently, some scholars also found cerebral blood volume reduction in the brain of Alzheimer's patients by using magnetic resonance imaging, while cerebral blood volume reduction can be determined by ¹H-MRS as early as in MCI stage 2 years ago, showing that ¹H-MRS is more sensitive [21]. Various researches show that magnetic resonance imaging can be widely used in the researches of MCI. Thus, it is worthy of further exploration.

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Disclosure of conflict of interest

None.

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