

A study of hemorheological behaviour for patients with Alzheimer's disease at the early stages

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Abstract. To evaluate the change of hemorheological indexes for patients with Alzheimer's disease (AD) at the early stages and to discuss effects of these changes on AD, high shear value of whole blood viscosity (η_{bh}), reduced high shear value of whole blood viscosity ($r\eta_{bh}$), low shear value of whole blood viscosity (η_{bl}), reduced low shear, value of whole blood viscosity ($r\eta_{bl}$), KT value of whole blood viscosity, hematocrit (HCT) and blood plasma viscosity (η_p) were measured in 31 patients with probable AD at the early stages and 33 age-matched healthy subjects. There were significant differences of all hemorheological indexes between AD group and control group except HCT. Step discriminant analysis revealed 81.25% of overall group-classified accuracy in a hemorheological discriminant function consisting of η_{bl} , $r\eta_{bl}$, $r\eta_{bh}$ and HCT. Significant difference of hemorheological indexes existed between AD and age-matched healthy control subjects. The results showed that measurement of hemorheological indexes could be used as one of reference standards of diagnosis in AD.

Keywords: Blood viscosity, Alzheimer's disease, hemorheology

1. Introduction

Alzheimer disease (AD) was called presenile dementia earlier, and also called primary degenerative dementia later, a definite diagnosis of which depends on the neuropathological evidences such as senile plaques, neurofibrillary tangles and cell loss [1–3] that predominate in the hippocampal formation (HF), amygdala (AM) entorhinal cortex. The diagnosis is hard to make in-patients at the early stages of AD, in which cognitive and memory deficits may be difficult to distinguish from age-related cognitive dysfunction [4]. MRI may be useful in the diagnosis of AD because of the limitation of pathological examination *in vivo*. Therefore, volume measurements of HF and AM are important indexes in the diagnosis of AD [4–8]. The studies of hemorheological behaviour on AD have also been reported [9–11]. However,

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using hemorheological indexes, such as high shear value of whole blood viscosity (η_{bh}), reduced high shear value of whole blood viscosity ($r\eta_{bh}$), low shear value of whole blood viscosity (η_{bl}), reduced low shear value of whole blood viscosity ($r\eta_{bl}$), hematocrit (HCT), KT value and blood plasma viscosity (η_p), no investigator has studied the patients with probable AD at the early stage. To our knowledge, no one has reported accuracy of the combination of hemorheological variables and Magnetic Resonance Image (MRI)-based volume measurements of some medial temporal lobe structures in separation patients with AD in the early stages from age-matched control subjects. We have already completed an investigation titled *A Quantitative MR Study of Hippocampal Formation, the Amygdala, and the Temporal Horn of the Lateral Ventricle in Healthy Subjects* (in press). In this paper, we would further find out whether there are significant changes of hemorheological characteristic in AD.

2. Materials and methods

2.1. Populations

We studied 31 patients with AD (without inflammation or other disease) at the early stages and 33 age-matched healthy subjects (Table 1). Patients with clinical diagnosed at the early stages of AD were in accordance with NINDS-ADRDA criteria [12], the MMSE [13] >21 . In order to exclude vascular and/or mix dementia, we made sure that ischemic scores [14] <4 . All patients were normal as to neurological examination and serum analysis, which included thyroidian hormones, vitamin B₁₂, folate and syphilis serology, and CT (computerized tomography) showed no significant abnormalities. The control subjects were in accordance with the following criteria: (1) no history of neurological or psychological illness; (2) no history of cardiovascular and cerebrovascular diseases; (3) no abnormalities of neurological and neuropsychological examinations; (4) ischemic score was not greater than four; (5) no significant abnormal signal on SE (Status Examination) transverse and sagittal T1WI and T2WI; (6) no abnormal results of serum analysis, including thyroidian hormones, vitamin B₁₂, folate and syphilis serology; and (7) Global Deterioration Scale Rating (GDSR) of 1. All the patients and control subjects gave informed consent and characteristics showed in Table 1.

2.2. Hemorheological examinations

Heparinized blood for hemorheological measurements (6 ml) was drawn from elbow-vein. Viscosity of plasma (η_p) and apparent viscosity of whole blood for all the patients and the control subjects were measured at high shear rate (η_{bh} , 150 s^{-1}) and low shear rate (η_{bl} , 10 s^{-1}) with a LBY-N6A Cone-Plate Rotating Viscometer. The coefficient of variation of this instrument is smaller than 5%. We also

Table 1
Subject characteristics

Subject characteristics	Mild AD ($n = 31$)	Age- and sex-matched CNES ($n = 33$)	P value
Sex *	15/16 (M/F)	16/17 (M/F)	0.67
Age in years (mean $\pm$ SD)	67.21 $\pm$ 8.10	66.48 $\pm$ 8.27	0.70
Education in years (mean $\pm$ SD)	17.30 $\pm$ 2.70	16.78 $\pm$ 2.23	0.62
MMSE (mean $\pm$ SD)	23.04 $\pm$ 0.89	28.89 $\pm$ 0.28	0.0001
TIV in cm^3 (mean $\pm$ SD)	1418 $\pm$ 158.00	1459 $\pm$ 101.04	0.68

Notes: Pearson chi-square; MMSE, Mini-mental status examination; TIV, Total intracranial volume.

measured reduced blood viscosity at corrected hematocrit (45%) at high shear rate ($r\eta_{bh}$) and low shear rate ($r\eta_{bl}$) according to the equation of Quemada [15] with this device. Hematocrit (HCT) was measured with microhematocrit tube. Index of erythrocyte rigidity 'KT' reflecting erythrocyte deformation was calculated according to the equation $[KT = (\eta_r^{0.4} - 1)/(\eta_r^{0.4}HCT), \eta_r = \eta_{bh}/\eta_p]$ of Dintenfass [16]. Whole hemorheological measurements were completed within 3 hours. The data of these measurements were processed by SPSS (win 8.0) statistic software package for normal distribution test analysis of variance, discriminant analysis.

3. Results

1. The results were showed in Table 2. High shear value of whole blood viscosity (η_{bh}), reduced high shear value of whole blood viscosity ($r\eta_{bh}$) low shear value of whole blood viscosity (η_{bl}), reduced low shear value of whole blood viscosity ($r\eta_{bl}$), hematocrit (HCT) and blood plasma viscosity (η_p) for AD and control subjects were all present normal distribution ($P > 0.05$).

2. Significance of η_{bh} , $r\eta_{bh}$, η_{bl} , $r\eta_{bl}$, η_p , KT was also found between the AD group and the control subjects group respectively ($P < 0.01$). These indexes for the patients at the early stages of AD were greater than that for the control subjects group. However, no significance of HCT was observed between AD group and the control subjects ($P > 0.05$). These results indicated that the reduced blood viscosity was obviously higher due to the increase of η_p and RBC deformability was significantly smaller (the increase of KT) for the patients at the early stages of AD than that of control group. Therefore, RBC aggregation increased evidently and cerebral perfusion became deficiency distinctly.

3. The Table 3 showed correct classification of different variables in the patients with Alzheimer's disease in the early stages and age-control subjects (%). The accuracy of η_{bh} , $r\eta_{bh}$, η_{bl} , $r\eta_{bl}$, HCT, KT and η_p to classify the patients in the early stages of AD and control subjects were 67.19%, 76.55%, 71.88%, 78.13%, 50.00%, 70.13%, 60.93%, respectively. Step discriminant analysis revealed a 81.25% of overall group-classified accuracy in a hemorheological discriminant function consisting of η_{bl} , $r\eta_{bl}$, $r\eta_{bh}$ and HCT, the Fisher's linear discriminant functions are as follows,

$$Z1 = -126.41 + 35.42\eta_{bl} - 30.96r\eta_{bl} + 9.81r\eta_{bh} + 10.73HCT, \quad (1)$$

$$Z2 = -158.10 - 12.53\eta_{bl} - 13.92r\eta_{bl} + 7.64r\eta_{bh} + 13.51HCT. \quad (2)$$

Table 2
Different variables in probable AD and control subjects

Variables	AD group (mean $\pm$ SD)	Control subjects (mean $\pm$ SD)	P value
η_{bh}	4.87 $\pm$ 1.01	3.23 $\pm$ 0.70	0.11
$r\eta_{bh}$	9.21 $\pm$ 1.01	6.60 $\pm$ 1.21	0.21
η_{bl}	8.00 $\pm$ 2.01	5.02 $\pm$ 1.22	0.29
$r\eta_{bl}$	17.22 $\pm$ 4.40	12.82 $\pm$ 2.74	0.34
HCT	38.27 $\pm$ 5.68	37.42 $\pm$ 5.49	0.03
η_p	1.58 $\pm$ 0.20	1.32 $\pm$ 0.13	0.06

Notes: η_{bh} , $r\eta_{bh}$, η_{bl} , $r\eta_{bl}$, HCT, η_p indicated high shear value of whole blood viscosity, reduced high shear value of whole blood viscosity, low shear value of whole blood viscosity, reduced low shear value of whole blood viscosity, hematocrit, blood plasma viscosity, respectively; and η_{bh} , $r\eta_{bh}$, η_{bl} , $r\eta_{bl}$, η_p in cP (centipoise).

Table 3

Correct classification of different variables in the patients with Alzheimer's disease in the early stages and age-control subjects (%)

Variables	Accuracy
η_{bh}	67.19
$r\eta_{bh}$	76.56
η_{bl}	71.88
$r\eta_{bl}$	78.13
HCT	50.00
η_p	70.31
$r\eta_{bl} + \eta_{bl} + r\eta_{bh} + \text{HCT}$	81.25

Notes: η_{bh} , $r\eta_{bh}$, η_{bl} , $r\eta_{bl}$, HCT, η_p indicated high shear value of whole blood viscosity, reduced high shear value of whole blood viscosity, low shear value of whole blood viscosity, reduced low shear value of whole blood viscosity, hematocrit, and blood plasma viscosity, respectively.

If $Z1 > Z2$, the case should be considered as AD, otherwise the case should be considered as control subjects.

4. Discussion

It seems to be hardly understood that terms η_{bh} , KT and η_p in discriminant functions (Eqs (1) and (2)) were lost. The causes maybe as follows: (1) although only reduced blood viscosity ($r\eta_{bh}$ and $r\eta_{bl}$) appeared in the discriminant function, in fact, reduced blood viscosity is also related with the KT reflecting RBC deformability, η_{bh} and η_p . (2) As same as plasma viscosity η_p , reduced low shear value of whole blood viscosity ($r\eta_{bl}$) also describes RBC aggregation due to macromolecule bridge-liking effect in plasma [17] except reflecting blood viscosity. So the more important $r\eta_{bh}$ and $r\eta_{bl}$ still remained in discriminant functions, and that η_{bh} , TK and η_p automatically disappeared in the process of discriminant analysis with SPSS (win 8.0) statistic software package for normal.

In recent years, a number of experiments demonstrated that there are changes of membrane structure of erythrocyte in AD [9]. It is showed that AD is also related with rheological characteristic of blood. The increase of η_{bl} , $r\eta_{bl}$, $r\eta_{bh}$ and η_p might be responsible for the elevation of peripheral resistance as Suo et al. [10] suggested that *in vivo* the Abeta vasoactive property may contribute to cerebral hypoperfusion of AD patients and more to increased RBC aggregation of AD patients. So it is important to study whether the changes of hemorheology indexes for AD and analysis of variance of these indexes. These results are consistent with that we have gotten: distinct differences of all rheological indexes except HCT between AD and control group exist. Therefore, the Fisher's linear discriminant functions including hemorheological indexes for AD can be obtained.

The erythrocyte aggregation is one of the most important factors, which determines the blood flow at low flow rate in microcirculation. Reduced blood viscosity at low shear rate $r\eta_{bl}$ relates with erythrocyte aggregation because formation of erythrocyte aggregation represents the balance of energies at the cell surface, which depend on surface charge of cell membrane and plasma viscosity [18]. Namely, the increase of reduced low shear value of whole blood viscosity ($r\eta_{bl}$) in AD means that erythrocyte aggregation became comparatively elevation in AD. This fact is consistent with following factors. (1) Abeta has a direct and specific constrictive effect on cerebral vessels *in vivo*, which may contribute to the cerebral hypoperfusion [10] and erythrocyte aggregation ($r\eta_{bl}$ increase) observed early in AD process.

(2) Electrophoretic mobility of erythrocytes in AD drops [9], so that surface-charge of erythrocytes is decreased and aggregation of erythrocytes is strengthened of erythrocytes in AD due to membrane modification [9]. (3) As we all know, plasma proteins, especially the macromolecules such as fibrinogen, globin and albumin, play a very important role in the plasma viscosity. The linear amyloid derivatives are able to modify RBC aggregation through the bridge-linking effect. The increase of plasma viscosity η_p in AD indicates that the concentration of macromolecules in plasma has elevated significantly. This causes more RBC aggregation. The results show that reduced blood viscosity η_{bl} is an important hemorheological index, which is diagnosis of AD.

As shown in another work [19], combination of MRI-based volumetric and hemorheologic variables including volume of HF and η_{bh} in differentiating patients with AD in the early stages from age-matched subjects has been completed. A 97.16% of total group-classified accuracy was disclosed in a discriminant function by combination of MRI-based volumetric and hemorheological variables including volume of HF and η_{bh} in differentiating patients with AD in the early stages from age-matched subjects.

In summary, hemorheological measurements have an important role in the separation of AD patients from control subjects, but how to define accurate and simple variable to reflect the MRI diagnosis index of AD patients, just is our study aim.

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