Distribution of Amyloid Burden Differs between Idiopathic Normal Pressure Hydrocephalus and Alzheimer's Disease

M. KONDO¹, T. TOKUDA^{1,2}, M. ITSUKAGE¹, N. KURIYAMA³, S. MATSUSHIMA⁴, K. YAMADA⁴, H. NAKANISHI⁵, M. ISHIKAWA⁶, M. NAKAGAWA¹

¹ Department of Neurology, ² Department of Molecular Pathobiology of Brain Diseases, ³ Department of Epidemiology for Community Health and Medicine, and ⁴ Department of Radiology, Kyoto Prefectural University of Medicine; Kyoto, Japan

⁵ Department of Radiology, Nishijin Hospital; Kyoto, Japan

⁶ Normal Pressure Hydrocephalus Center, Otowa Hospital; Kyoto, Japan

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SUMMARY – *This study aimed to elucidate the incidence and distribution of the cortical retention of Pittsburgh compound B (PIB) in patients with idiopathic normal pressure hydrocephalus (iNPH) and clarify the differences from those in patients with Alzheimer's disease (AD). Ten patients with iNPH without any clinical signs indicative of AD were enrolled in this study. Cerebral retention of PIB in positron emission tomography (PET) in iNPH patients was compared with those in seven age-matched AD patients. The CSF levels of β -amyloid 1-42 peptide (A β 42), which inversely decrease with cerebral amyloid burden, were also measured. Three of the ten patients with iNPH showed increased cortical PIB retention. Although the mean cortical SUV ratios were similar, the distribution of PIB retention differed widely between the patients with iNPH and AD. PIB retention was limited to the high-convexity parasagittal areas in iNPH patients, whereas it spread over the frontal and parietotemporal areas in AD. The coronal images of PIB-PET were more informative than conventional transverse images in evaluating the distribution pattern of cortical PIB retention. Two iNPH patients with higher cortical PIB retention had the lowest levels of CSF A β 42, indicating that PIB retention in iNPH would not reflect a simple delay in PIB clearance but its binding to existing A β amyloid in the brain. Our results indicate that iNPH is one of the diseases exhibiting cortical PIB retention. The characteristic distribution of PIB retention in iNPH could be useful in the differential diagnosis between iNPH and AD.*

Introduction

Idiopathic normal pressure hydrocephalus (iNPH) causes gait disturbance, cognitive decline and urinary incontinence which can be ameliorated by shunt operation. One of the potential factors that may negatively influence shunt responsiveness is the presence of comorbidity, especially Alzheimer's disease (AD) that results in progressive dementia^{1,2}. It is hence essential to make an accurate diagnosis on whether each patient with iNPH has comorbid AD.

Recently, positron emission tomography (PET) using ¹¹C-labeled radiotracer Pittsburgh

compound B (PIB) has been widely applied for the in vivo assessment of amyloid- β (A β) deposition in patients with AD, with successful results^{3,4}. It is thus conceivable that this PIB-PET study may be useful for the diagnosis of comorbid AD in iNPH patients. However, there remains a possibility that iNPH patients may also have PIB retention, since cortical A β deposition has been found in biopsied cortical tissues in 42-75 % of iNPH patients examined^{2,5,6}. No previous study has investigated whether iNPH patients without comorbid AD may exhibit PIB retention. Thus, the aim of our study was to determine whether cortical PIB retention exists

in patients with iNPH. We performed PIB-PET study in patients with “pure” iNPH without any clinical signs of AD. We found that small fraction of pure iNPH patients had cortical PIB retention, but with quite a different distribution pattern from that seen in AD.

Subjects and Methods

Subjects

Ten patients with iNPH (six men and four women, aged 79.2 ± 5.0 years, 24.5 ± 1.9 in Mini-Mental State Examination (MMSE) scores (mean $\pm$ standard deviation)) were enrolled in this study (Table 1). Among the iNPH patients, none had obvious dementia with impairment of usual activities of daily living, but seven of them were diagnosed as having mild cognitive impairment (MCI) because of their cognitive concern reported by themselves or their caregivers and cognitive tests (Table 1). Cerebral retention of PIB in those iNPH patients was compared with those in seven age-matched AD patients (four men and three women, aged 70.4 ± 11.3 years). The iNPH patients consisted of those with probable ($n = 8$) or definite ($n = 2$) iNPH, diagnosed according to the Japanese guidelines for iNPH⁷. The AD patients were diagnosed as probable AD according to the standard clinical criteria⁸. Before diagnosis, all the iNPH subjects underwent clinical examinations including neurological and neuropsychological examinations, routine blood analyses,

brain magnetic resonance imaging (MRI), and single photon emission computed tomography (SPECT) cerebral blood flow imaging. This study included only those iNPH patients confirmed to have neither AD-like brain atrophy in medial temporal lobes by MRI nor AD-like hypoperfusion in parietotemporal, posterior cingulate or precuneus cortices by SPECT imaging.

All the examined subjects provided written informed consent to participate in the study, which was approved by the University Ethics Committee (Kyoto Prefectural University of Medicine, Kyoto, Japan).

Acquisition and Analysis of PIB-PET Data

Pittsburgh compound B (PIB: *N*-Methyl-[¹¹C]2-(4-methylaminophenyl)-6-hydroxybenzothiazole) was prepared at Nishijin Hospital (Kyoto). PIB radiosynthesis was performed using a simplified method reported by Wilson et al.⁹. PIB was injected intravenously (555 ± 185 MBq, 15 ± 5 mCi) as a slow bolus for 10–20 seconds. PET scanning was performed using a Siemens ECAT ACCEL scanner (Siemens Medical Systems, Erlangen, Germany) equipped with LSO crystals (3D mode, 16.2 cm field of view, 47 planes) with 35 dynamic frames acquired over 70 min. The ECAT ACCEL has a transaxial spatial resolution of 6.3 mm full width at half maximum (FWHM) measured at 1 cm off-axis, and of 6.8 mm measured at 10 cm off-axis. The axial resolution is 4.7 mm at the centre of the system. Attenuation correc-

Table 1 Demographics and clinical characteristics of patients with iNPH.

Case No.	Diagnosis	Age (y) / Sex	MMSE	FAB	Symptoms			3mTUG		Cortical retention of PIB
					G	D	U	Time (sec)	Steps	
1	Probable	78 / F	27	13	+	–	–	12.5	19.5	–
2	Definite	80 / F	24	9	+	±	–	15.4	27.4	–
3	Probable	84 / F	26	16	+	±	+	20.6	26.6	+
4	Probable	82 / M	28	12	+	–	+	10.7	16.0	–
5	Probable	81 / M	24	12	+	±	+	30.2	56.8	–
6	Probable	88 / M	22	11	+	±	+	17.4	30.2	+
7	Probable	73 / M	24	12	+	±	–	13.6	18.2	–
8	Probable	72 / M	23	16	+	±	+	23.5	43.3	–
9	Definite	75 / F	24	15	+	±	–	14.2	21.0	+
10	Probable	79 / M	23	14	+	±	+	21.8	27.0	–

MMSE = Mini-Mental State Examination; FAB = Frontal assessment battery; PIB = Pittsburgh compound B; G = gait disturbance; D+ = dementia; D± = mild cognitive impairment; U = urinary disturbance; TUG = timed up-and-go test. Required time and steps of 3mTUG were the mean values of 3 tests.

Figure 1 PIB-PET in patients with iNPH and AD. Representative axial and coronal images of PIB-PET in the iNPH patient without (A, patient 1 in Table 1) and with (B, patient 9) cortical PIB retention and AD (C) showing different patterns of cortical PIB retention between the two diseases. The color bar on the right indicates the standardized uptake values.

Figure 2 A) Cortical standardized uptake value (SUV) ratio calculated using cerebellar SUV as the reference in each patient of the iNPH ($n = 10$) and AD ($n = 7$) groups. $p = 0.088$; Mann-Whitney test. B) The PS/PT ratio in each patient with iNPH showing cortical PIB retention ($n = 3$), and that of AD ($n = 7$). $p = 0.017$; Mann-Whitney test. C) CSF levels of A β 42 in each iNPH patient without (C(-), $n = 7$) and with (C(+), $n = 2$) cortical PIB retention. Horizontal bars in A-C indicate the mean values.

tion was performed using transmission scans, obtained using three rotating germanium-68 rod sources. PET data were reconstructed using the ordered subsets expectation maximization (OSEM) algorithm, with six iterations and 16 subsets. The reconstructed images were displayed in coronal, transverse and sagittal planes for analysis.

Regions of interest (ROIs) were manually generated for 18 regions on each PET image by one experienced neurologist and one experienced radiologist familiar with the neuroanatomy (MK and SM). The locations of the ROIs included bilateral anterior cingulate gyri,

frontal cortices (dorsal and ventral), lateral temporal cortices, medial temporal cortices (amygdala and hippocampus), lateral parietal cortices, occipital cortices, precuneus and cerebellum, while parasagittal areas included anterior cingulate gyri and precuneus, which were modifications of a previous method to concentrate ROIs in cortical areas¹⁰. PMOD software (version 3.2, PMOD Technologies Ltd., Zurich, Switzerland) was used to extract count data for specified ROI. A mean cortical PIB retention measurement was also computed across cortical ROIs without including the cerebellum. The PIB retention outcomes were evaluated

based on the standardized uptake value (SUV) measures. SUVs were determined over 50-70 minutes post injection intervals and normalized to injected dose and body mass. SUV ratio (SUVR) was generated using the cerebellum as reference.

Coronal sections of PIB-PET images across precuneus areas were analyzed using an arbitrary index, SUVR measured in parasagittal (PS) and parietotemporal (PT) areas (the PS/PT ratio). The PS/PT ratio was calculated using the formula:

$$\text{PS/PT ratio} = (\text{right PS/PT} + \text{left PS/PT})/2.$$

Measurement of A β 42 in Cerebrospinal Fluid

To evaluate the extent of amyloid burden in each patient's brain, cerebrospinal fluid (CSF) levels of β -amyloid 1-42 peptide (A β 42) which are known to decrease inversely with the extent of cerebral amyloid deposition were determined by sandwich ELISA according to the manufacturer's instructions (Human/Rat β Amyloid (42) ELISA Kit, Wako, Osaka, Japan) in nine of the ten patients with iNPH who agreed to undergo the test.

Statistical Analysis

We compared iNPH and AD groups using the Mann-Whitney U test. The level of statistical significance was set at $p < 0.05$. All statistical analyses were performed using SPSS (Dr SPSS II version, SPSS Japan Inc., Tokyo, Japan).

Results

Three of the ten patients with iNPH, all of whom were diagnosed as MCI in cognitive tests, showed increased cortical PIB retention, especially in the bilateral high-convexity parasagittal areas (Table 1, Figure 1B). The mean cortical SUVRs, calculated using cerebellar SUV as the reference, did not significantly differ between iNPH and AD patients (Figure 2A). However, the distribution of cortical PIB retention was clearly different between the two groups. PIB retention was limited to the high-convexity parasagittal areas in iNPH patients, while it spread over the frontal and parietotemporal areas in AD (Figure 1B). This was confirmed by the mean PS/PT ratio that was significantly higher in iNPH patients with cortical PIB retention than in AD patients ($p=0.017$) (Figure 2B).

The mean cortical SUVR had no significant correlation either with MMSE score or measures of timed up-and-go (TUG) tests (required time or steps) in iNPH patients (data not shown).

The levels of CSF A β 42 were measured in nine patients with iNPH, including two cases with higher cortical PIB retention; those two patients showed the lowest levels of CSF A β 42 among the nine iNPH patients (Figure 2C). This low CSF level indicates that there is an actual deposition of A β amyloid in the brain, and PIB retention in iNPH would not therefore reflect a delay in the clearance of PIB from the brain tissue but its real binding to cerebral A β amyloid.

Discussion

We have demonstrated here that three out of ten (30%) patients with iNPH without any clinical signs of AD had obvious cortical retention in PIB-PET, indicating that iNPH is one of the PIB-positive diseases. We also revealed that the pattern of cortical PIB retention seen in iNPH patients was characteristic of iNPH, limited to the high-convexity parasagittal areas showing a high PS/PT ratio (Figure 2B). This pattern was apparently different from that in AD with high retention in parietotemporal cortices showing a low PS/PT ratio. All three iNPH patients with positive cortical PIB retention were diagnosed as having MCI in cognitive tests. It will be very interesting to see whether or not and how fast their cognitive functions will deteriorate in the course of their iNPH, compared with the usual course of AD. We are therefore planning a longitudinal follow-up on these patients.

Our data also suggest that differentiation between cognitive decline due to iNPH alone and due to comorbid AD pathology may be possible, not by the CSF A β 42 levels, but by the distribution pattern of PIB-positive cortical amyloid. This was most effectively judged based on coronal images rather than conventional transverse images. These results indicate the possible role of PIB-PET in the diagnosis of the etiology of dementia in iNPH, which could be valuable in considering a shunt indication.

A β -amyloid deposition depends on the local balance between production and clearance. It has been reported that local A β production increases in specific brain regions correlating with synaptic activity or aerobic glycolysis^{11,12}. In AD patients, some of those regions belong to

a group of brain areas called the default mode network¹³. Thus, the deposition of A β amyloid in AD brain is caused by the combination of both increased local A β production in parieto-temporal cortices and decreased A β clearance due to aging.

On the other hand, in the iNPH patients' brains, cortical A β deposition is conceivably attributed mainly to the decrease in local A β clearance. It is well-known that CSF turnover is much slower in iNPH¹⁴. Since the upper half of the brain is passively lifted cranially in these patients, the high-convexity parasagittal regions will be compressed and there will be less interstitial fluid¹⁵. This could lead to a decline in local clearance of A β , which would ultimately lead to local A β -amyloid depositions.

There is a possibility that the cortical PIB retention in iNPH might be attributable to other causes. One possibility is that it does not reflect a real accumulation of A β amyloid but merely picks up the delay in the clearance of administered PIB from the brain due to the slower turnover of CSF¹⁴. However, we con-

sider PIB retention in iNPH would reflect the actual accumulation of A β amyloid because the iNPH patients with high cortical PIB retention showed lower CSF levels of A β 42, indicating the presence of actual amyloid deposition in the cerebral cortex.

Conclusion

iNPH is one of the diseases that may present cortical PIB retention. However, the distribution of PIB retention differs widely from that in typical AD patients and is limited to the high-convexity parasagittal areas, thus distinguishing the two conditions.

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Takahiko Tokuda, MD, PhD
Department of Molecular Pathobiology
of Brain Diseases (Neurology)
Graduate School of Medical Science
Kyoto Prefectural University of Medicine
465 Kajii-cho, Kamigyo-ku
Kyoto 602-8566, Japan
Tel.: +81-75-251-5793
Fax: +81-75-211-8645
E-mail: ttokuda@koto.kpu-m.ac.jp