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Structural and Functional Brain Changes Following Repetitive Traumatic Brain Injury: Functional and Molecular Brain Imaging Studies

반복적인 뇌 외상에 의한 뇌 구조 및 기능의 변화: 뇌 기능 및 분자 영상연구

2016 년 02 월

서울대학교 대학원

의학과 뇌신경과학 전공

방 성 애

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2015年 10月

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2015年 12月

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Structural and Functional Brain Changes Following Repetitive Traumatic Brain Injury: Functional and Molecular Brain Imaging Studies

by
Seong Ae Bang

A thesis submitted to the Department of Neuroscience in
Partial Fulfillment of the Requirements for the
Degree of Doctor of Philosophy in Medicine
at the Seoul National University College of Medicine
(Director: Prof. Sang Eun Kim)

October, 2015

This dissertation is an approved for
The Degree of Doctor of Philosophy in Medicine
December, 2015

Approved by thesis committee:

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Abstract

Structural and Functional Brain Changes Following Repetitive Traumatic Brain Injury: Functional and Molecular Brain Imaging Studies

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Repetitive traumatic brain injury (rTBI) occurs as a result of mild and accumulative brain damage. A prototype of rTBI is chronic traumatic encephalopathy (CTE), which is a degenerative disease that occurs in patients with histories of multiple concussions or head injury. Boxers have been the most commonly studied patient group because they may experience thousands of subconcussive hits over the course of a match.

This study examined the consequences of rTBI with structural brain imaging and biomolecular imaging and investigated whether the neuropsychological features of rTBI were related to the findings of the imaging studies.

Five retired professional boxers (mean age, 46.8 ± 3.19) and 4 age-matched controls (mean age, 48.5 ± 3.32) were studied. Cognitive-motor related functional impairment was assessed, and all subjects underwent neuropsychological evaluation and behavioral tasks, as well as structural brain imaging and functional-molecular imaging.

In neuropsychological tests, boxers showed deficits in delayed retrieval of visuo-spatial memory and motor coordination, which had a meaningful relationship with biomolecular imaging results indicative of neuronal injury.

Morphometric abnormalities were not found in professional boxers by structural MRI, although some diffusion abnormalities were detected in white matter connections from the occipito-temporal and orbitofrontal areas. Striatal dopaminergic function was

well preserved. Glucose metabolism was impaired in frontal areas associated with cognitive dysfunction, similar to findings in Alzheimer's disease. Low binding potential of ^{18}F-Flumazenil was found in the angular gyrus and temporal cortical regions, revealing neuronal deficits.

These results suggested that cognitive impairment and motor dysfunction reflect chronic damage to neurons in professional boxers with rTBI.

Keywords: Repetitive traumatic brain injury, Neuropsychological evaluation, Structural brain imaging, Dopamine transporter, Brain metabolism, $GABA_A$ receptor

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List of Abbreviations

Abbreviations	Full name or description
BP	binding potential
BZR	benzodiazepine receptor
CTE	chronic traumatic encephalopathy
DAT	dopamine transporter
DP	dementia pugilistica
DTI	diffusion tensor imaging
FDG	^{18}F-fluorodeoxyglucose
FMZ	^{18}F-flumzaenil
FWHM	full width at half-maximum
GABA	gamma-aminobutyric acid
HVLT	hopkins-verbal learning test
FP-CIT	[^{123}I]*N*-ω-fluoropropyl-2β-carbomethoxy-3β-(4-iodophenyl) nortropane
MMSE	mini-mental state examination
MNI	montreal neurologic institute
MRI	magnetic resonace imaging
PD	Parkinson's disease
PET	positron emission tomography
PPB	the purdue pegboard test
RCFT	rey-osterrieth complex figure test
ROI	region of interest
rTBI	repetitive Traumatic Brain Injury
SPECT	single photon emission computed tomography
UPDRS	the unified Parkinson's disease rating scale

Contents

Introduction

Repetitive traumatic brain injury (rTBI) occurs as a result of mild and accumulative brain damage. The prototype of rTBI is chronic traumatic encephalopathy (CTE). CTE is a degenerative disease that occurs in patients with histories of multiple concussions or head injuries and that is attributed to the rTBI that occurs in contact sports, such as boxing, wrestling, football, and rugby. This trauma includes mild traumatic brain injury (mTBI), concussions, and subconcussive injuries (McKee, Cantu et al. 2009, Gavett, Stern et al. 2011, Baugh, Stamm et al. 2012).

Brain trauma in boxers was first described as “Dementia pugilistica (DP)” or “punch-drunk syndrome,” in a patient with advanced Parkinsonism, pyramidal-tract dysfunction, ataxia, and behavioral problems including severe cerebellar, cognitive, and psychiatric abnormalities (Guterman and Smith 1987). Boxing has been commonly studied because boxers experience thousands of subconcussive hits over the course of matches or sparring bouts, with professional boxers being more at risk than amateurs (Clausen, McCrory et al. 2005). CTE is distinct from acute concussion or TBI, and is not merely a prolonged post-concussive syndrome (Gavett, Stern et al. 2011). The terms CTE or rTBI should preferentially be applied to repetitive/cumulative brain damage that causes long-term cognitive, motor, and psychiatric symptoms resulting from any contact sport (Costanza, Weber et al. 2011). Clinically, rTBI presents with complex behavioral, psychiatric, cognitive, and/or motor-related symptoms. Neuropsychological features and cognitive dysfunctions are characterized by

impairments in memory and executive function, behavioral changes, motor-related signs, and personality changes (Haglund and Eriksson 1993, Sosa, Linic et al. 2011, Saing, Dick et al. 2012).

Behavioral disturbances are often the earliest finding and include aggression, depression, apathy, and impulsivity (Stern, Riley et al. 2011). Executive function involving planning, organization, multi-tasking, and decision-making is also often impaired in subjects with repetitive brain trauma. These neuropsychological changes may predict neurodegenerative disease progression (Baugh, Stamm et al. 2012). Motor impairment in boxers may include mild dysarthria and difficulty balancing, progressing to ataxia, spasticity, impaired coordination, and Parkinsonism—an entity distinct from idiopathic Parkinson's disease (PD) (Davie, Pirtosek et al. 1995, Mendez. 1995).

Imaging modalities to detect and assess brain injury are constantly improving, but CT/MRI often show normal structures despite a host of symptoms (Gardner 2013). Ross and colleagues found no correlation between the duration of damage or behavioral symptoms, and findings from CT and electroencephalography in 38 retired boxers (Ross, Casson et al. 1987). CT was used to examine 338 active professional boxers, whose scans were normal in 93% of cases, with 7% showing 'borderline' atrophy (Jordan, Jahre et al. 1992). These negative results led many researchers to use MRI, and abnormalities were found in boxers with normal CT findings (Jordan and Zimmerman 1990). Volumetric MRI has been proposed for diagnosing rTBI by detecting atrophy of the whole brain as well as specific areas (e.g., amygdala) (McKee, Cantu et al. 2009), and its utility was demonstrated in five symptomatic former-professional contact-sport athletes (Gavett, Cantu et al. 2011). However, MRI findings are inconsistent

in rTBI (Zhang, Ravdin et al. 2003). Diffusion tensor imaging (DTI) and magnetic resonance spectroscopy (MRS) have also been proposed for detecting axonal injury or significant chemical changes in TBI, but these imaging methods cannot clearly disclose the mechanism of rTBI progression.

Recently, biomolecular imaging has emerged as an ideal method for examining biochemical mechanisms in the human brain. ^{18}F-Fluorodeoxyglucose (FDG) PET and more recently, ^{18}F-Flumazenil (FMZ) PET can reveal metabolic brain function and neuronal integrity. ^{18}F-FDG is well established for PET imaging of cerebral glucose metabolism associated with neuronal function. FMZ is a selective, reversibly bound, high-affinity neutral antagonist of the central benzodiazepine receptor (BZR) binding site of the γ- aminobutyric acid-A ($GABA_A$) receptor (Delforge, Pappata et al. 1995, Odano, Halldin et al. 2009), which is abundant in the cortex and reflects neuronal density and integrity. The benzodiazepine binding sites on GABA-A receptors have been used as a marker of neuronal viability (Rudolf, Sobesky et al. 2000). The binding potential (BP) of FMZ could be a sensitive marker of neuronal viability, receptor density, and affinity. Some previous FMZ BP studies strongly support a selective loss of neurons in damaged brains regardless of normal MRI findings (Heiss, Grond et al. 1998, Heiss, Kracht et al. 2000, Kuroda, Shiga et al. 2004). [^{123}I]*N*-ω-fluoropropyl-2β-carbomethoxy-3β-(4-iodophenyl) nortropane (^{123}I-FP-CIT) SPECT is frequently and routinely used to detect or exclude dopaminergic degeneration of the nigrostriatal tract in PD by imaging the dopamine transporter (DAT) (Ma, Dhawan et al.). A recent ^{18}F-FDOPA PET study on boxers revealed a strong relationship between head trauma and PD (Lolekha, Phanthumchinda et al. 2010), with repetitive head trauma arguably increasing the risk in susceptible

individuals.

We focused on examining neuronal deficits in patients with chronic brain damage with biomolecular imaging, and we expected these techniques to provide unique information on the cellular and molecular aspects of rTBI. In addition, we administered various neuropsychological tests to investigate whether neuropsychological features are related to the findings of imaging studies.

Purposes

Purpose 1: To investigate the brain mechanism following repetitive traumatic brain injury using multimodal imaging approaches structural and biochemical molecular imaging; 1) structural and diffusion MR: to examine the differences in morphometric structure and white matter fiber structures, 2) ^{123}I-FP-CIT SPECT: to assess the striatal dopamine transporter density in subjects exposed repetitive traumatic brain injury, 3) ^{18}F-FDG PET: to show abnormal brain glucose metabolism, 4) ^{18}F-FMZ PET: to detect selective loss of neuronal cells in the cortex with damaged brain by GABA receptor binding as an indicator of neuronal integrity between professional boxer group and control group.

Purpose 2: To examine the relationship of neuropsychological features in professional boxer groups compared with control group; to determine the relationship between these neuropsychological features and results derived from various neuroimaging modalities.

Materials and methods

Subjects

Five retired professional boxers (mean age, 46.8 ± 3.19; range 42–49) and 4 age-matched controls (mean age, 48.5 ± 3.32; range 46–53) were studied. All subjects were right-handed assessed by the Edinburgh Handedness Questionnaire (Oldfield 1971) and were screened for neurological and psychiatric diseases. Informed consent was obtained from each subject in accordance with the research protocol for human subjects approved by the Medical Ethics Committee of Seoul University Bundang Hospital. (IRB no.B-1212-182-004). The characteristics of the subjects are listed in Table 1. Details of their boxing careers can be seen in Table 2. The following inclusion criteria were fulfilled for the professional boxers: >10-year career in professional boxing (including duration as an amateur); right-handed males, aged 30–55 years; written informed consent obtained.

The duration of exposure to the repetitive brain damage of boxing of the study subjects was a minimum of 22 years (mean, 27.6 years; range, 22–30 years) from their initial exposure to boxing.

The following exclusion criteria were used for all subjects, including boxer and control groups: history of cerebral hemorrhage or neurological brain surgery; active (or history of) neuropsychiatric disease; claustrophobia; metal substance in the body (e.g., cardiac pacemaker, prosthetic appliance); usage of any medication; poor compliance with the investigator's instructions.

In order to investigate the subjects' daily symptoms that were associated with brain trauma, we interviewed the individuals about their self-reported symptoms in the following categories: headache; dizziness; body tremor; sense

of physical discomfort; cognitive status, memory, and decision-making; and quality of sleep. Among the 5 professional boxers, subject 1 complained of chronic dizziness that occurred 2–3 times a week. Subject 2 complained chronic headaches and recent memory difficulties in object naming. Subject 4 had been treated for chronic headaches in the hospital for 3 years. The other 2 subjects did not complained any daily symptoms.

Table 1. Subject characteristics

		Boxer Group	Control Group	*p*
Age	Mean	46.8	48.5	.608
(years)	*SD*	3.2	3.3	
	Range	42–49	46–53	
Education	Mean	12.2	13.0	.529
(year)	*SD*	3.0	2.0	
	Range	8–16	12–16	
Height	Mean	163.2	165.5	.732
(cm)	*SD*	5.26	8.15	
	Range	156-170	157-176.6	
Weight	Mean	67.9	69.6	.556
(kg)	*SD*	4.92	10.2	
	Range	64-76	55.3-79	

Table 2. Bouts and demographic information for the boxer group

No.	Weight division	Total duration (year)	Amateur duration (year)	Professional duration (year)	Bouts	Wins	Losses	KO
BG001	Flyweight	17	3	14	37	31	6	1
BG002	Flyweight	15	4	11	31	26	5	2
BG003	Feather-weight	23	4	19	36	32	2	0
BG004	Flyweight	11	3	8	23	19	4	4
BG005	Bantam-weight	15	5	10	24	22	0	0

Each boxer within this study group; includes matches fought (bouts), whether the bout was a win or loss, the career duration, and weight division in boxing. KO, knockout: reveals that boxers were down for the count in the match.

Screening and Neuropsychological testing

UPDRS (the Unified Parkinson's Disease Rating Scale)

For the clinical estimation of motor-related functional impairment in boxers and to assess whether they have Parkinson-like symptoms, we have used the Unified Parkinson's Disease Rating Scale (UPDRS) on all subjects. This scale is used to follow disease progression and to evaluate surgical, medical, and other interventions for PD (Ramaker, Marinus et al. 2002, Siderowf, McDermott et al. 2002). The UPDRS consists of four components: 1) mentation, behavior, and mood; 2) activities of daily living; 3) motor examination; 4) complications of therapy. The clinician-scored motor evaluation was based on part III of the UPDRS, which assesses the objective motor symptoms based on a scale ranging from 0 (no impairment) to 4 (severe impairment). The UPDRS-III items assess specific motor symptoms: speech; facial expression; tremor at rest in head, arms, and legs; action and postural tremor of hands; rigidity of neck, arms and legs; finger tapping; hand movements; rapid alternating movements; leg agility; arising from chair; posture; gait; postural stability; and body bradykinesia. In our study, a neurologist evaluated all subjects using the UPDRS-III.

Chronic Brain Injury Scale (CBI)

The chronic brain injury (CBI) scale quantifies the clinical findings of motor, cognitive, and psychiatric deficits associated with boxing-related brain injury (Jordan 2000). Scores on the CBI scale range from 0 to 9, with higher scores reflecting greater impairment. The cognitive component incorporates the boxer's

score on the Mini-Mental State examination (MMSE). Table 3 shows the CBI scale.

Table 3. CBI scale

Symptoms	**Score**
Motor	
Normal	**0**
Mild incoordination, dysarthria, Parkinsonism, gait disturbance, or pyramidal signs	**1**
Moderate incoordination, dysarthria, Parkinsonism, gait disturbance, or pyramidal signs	**2**
Severe incoordination, dysarthria, Parkinsonism, gait disturbance, or pyramidal signs	**3**
Cognitive	
Normal or MMSE = 28–30	**0**
Mild deficits in mental speed, memory, attention, executive function, language, or visuospatial function, or MMSE = 20–27	**1**
Moderate deficits in mental speed, memory, attention, executive function, language, or visuospatial function, or MMSE = 10–19	**2**
Severe deficits in mental speed, memory, attention, executive function, language, or visuospatial function, or MMSE ≤ 9	**3**
Behavioral	
Normal	**0**
Mild agitation or aggression, delusions, hallucinations, dysphoria or anxiety, euphoria or apathy, disinhibition, irritability or liability, or aberrant motor behavior	**1**
Moderate agitation or aggression, delusions, hallucinations, dysphoria or anxiety, euphoria or apathy, disinhibition, irritability or liability, or aberrant motor behavior	**2**
Severe agitation or aggression, delusions, hallucinations, dysphoria anxiety, euphoria or apathy, disinhibition, irritability or liability, or aberrant motor behavior	**3**
	Total CBI score (range 0–9)

Neuropsychological tests

To investigate impaired neuropsychological performance in professional boxers, we used various neuropsychological assessments. All subjects underwent a neuropsychological evaluation using the following standardized tests: MMSE, the Beck Depression Inventory (BDI; Beck, Ward et al. 1961) and the Barratt Impulsiveness Scale-11 (BIS-11; Patton, Stanford et al. 1995). BIS-11 consists of three subscales: motor impulsiveness, attentional impulsiveness, and non-planning impulsiveness. To examine the cognitive ability of professional boxers, all subjects underwent various cognitive tasks: 1) the Hopkins Verbal Learning Test (HVLT): verbal memory; 2) Rey-Osterrieth Complex Figure Test (RCFT): visuo-spatial memory.

BDHI (The Buss–Durkee Hostility Inventory)

The Buss–Durkee Hostility Inventory (BDHI) is one of the best-known self-report measures of aggressive personality (Buss and Durkee 1957). The BDHI is a written questionnaire designed to assess different types of aggression. The questionnaire includes a four-point Likert scale. From the BDHI, we selected 21 items from three subscales: behavioral aggression (assault), verbal aggression, and indirect aggression. Behavioral aggression (9 items) is defined as physical violence against others, including getting into fights with others, but not destroying objects. Sample items from this scale include "If someone hits me first, I let him have it," and "Once in a while I cannot control my urge to harm others." Verbal aggression (7 items) represents negative affect expressed in terms of arguing, shouting, or screaming, with content including threats, curses, or being overly critical ("When I get mad, I say nasty things," or "When people yell at me,

I yell back"). Indirect aggression (5 items) includes both roundabout (malicious gossip or practical jokes) and undirected (temper tantrums or slamming doors) aggressive behavior. For example, "I sometimes spread gossip about people I don't like," and "I can remember being so angry that I picked up the nearest thing and broke it." Higher scores on this measure indicate more aggressive behavioral tendencies.

The Purdue Pegboard test (PPB)

The Purdue Pegboard test (PPB; Tiffin and Asher 1948) is widely used for measuring hand agility and bimanual coordination. The pegboard consists of a board with two parallel rows, each with 25 holes into which cylindrical metal pegs are placed by the examinee. The test involves a total of four trials. The subsets are for preferred hand, non-preferred hand, both hands, and assembly performance using both hands. After one practice trial, subjects were required to place the pins in the holes as quickly as possible, with the score being the number of pins placed in 30 seconds (right, left, and both hands) and 1 minute (assembly). Poor Pegboard performance is a sign of deficits in complex, visually guided, or coordinated movements that are likely mediated by circuits involving the basal ganglia (Middleton and Strick 2000, Claassen, Jones et al. 2013).

Image acquisition

Structural and diffusion MRI

High-resolution T1 and T2 weighted structural images and diffusion tensor

images were acquired in a single session using a 3.0 Tesla MR system (Intera Achieva 3T, Phillips Medical Systems, Best, The Netherlands). The three-dimensional (3D) T1-weighted turbo field echo (T1TFE) sequence used the following parameters: TR = 8.1 ms, TE = 4.6 ms, flip angle = 8º, 175 slices, and thickness = 1 mm, and matrix size = 256 × 256. DTI (diffusion tensor imaging) was performed using a whole-brain spin-echo echo-planar imaging (EPI) sequence in 15 independent orientations with b = 700 s/mm^2 after the acquisition of b = 0 s/mm^2 images. DTI parameters were as follows: TR = 7514.1 ms, TE = 65.9 ms, flip angle = 100º, 75 slices, thickness = 2 mm, and matrix size = 128 × 128.

^{123}I-FP-CIT SPECT

For DAT Imaging, ^{123}I-FP-CIT (180 ± 15 MBq) was injected intravenously as a bolus. SPECT images were acquired 4.0 ± 0.2 h after tracer injection using a triple-headed rotating gamma camera system (Tronix XLT; Trionix Research Laboratory, Inc., Twinsburg, OH, USA). Acquisition parameters were: fixed rotational radius between 13 cm, matrix 128 × 128, angular sampling ≤3° (360° rotation), and hardware zoom of 2.0 0 to achieve a pixel size of 1.78 mm. The photopeak was set to 159 keV ± 10%. The acquisition time was 40 steps for 3° and 45 s per step. Scans were reconstructed using an iterative algorism.

^{18}F-FDG PET

^{18}F-FDG PET images were acquired using a Phillips Allegro PET scanner (Phillips Medical Systems, Cleveland, Ohio, USA) in 3D mode. All subjects fasted for at least 6 h before scanning. After intravenous administration of 4.8

MBq/kg of [^{18}F]-fluorodeoxyglucose (FDG), subjects were allowed to rest, while staying awake in a dimly lit room for 40 min during the uptake phase. Ten-minute emission scans and attenuation maps were obtained using a ^{137}Cs transmission source. Attenuation-corrected images were reconstructed using the 3D Row-Action Maximum-Likelihood algorithm with a 3D image filter as 128 × 128 × 90 matrices with a pixel size of 2 × 2 × 2 mm.

^{18}F-FMZ PET

^{18}F-FMZ was synthesized from 4-methylphenyl-mazenil iodonium tosylate by aromatic radiofluorination in the TRACERlab FX-FN module (GE Healthcare, USA) according to literature (Moon, Kil et al. 2011). 4-Methylphenyl-mazenil iodonium tosylate, prepared from commercially available isatoic anhydride in five steps based on a previously reported procedure (Moon, Park et al. 2014), is commercially available from Bio Imaging Korea Co., Ltd. (Seoul, Korea). ^{18}F-FMZ PET images were obtained using a Discovery VCT PET/CT scanner (GE Medical Systems, Milwaukee, WI). Dynamic ^{18}F-FMZ PET was performed on all subjects. The scan commenced with a simultaneous intravenous bolus injection of ^{18}F-FMZ (206.46 ± 9 MBq). We obtained dynamic scans in a sequence of 54 frames (12 frames × 10 s, 16 frames × 30 s, 8 frames × 1 min, 18 frames × 4 min) for a total acquisition time of 90 min to satisfy the pharmacokinetic properties of ^{18}F-FMZ in human brain (Odano, Halldin et al. 2009). PET images were reconstructed into a 25 cm diameter, 256 × 256 transaxial matrix using the FORE-FBP point ordered-subset expectation maximization algorithm with a 5.4 mm cut off (GE Healthcare). Attenuation correction was based on 3.75-mm-thick CT image set.

Data analysis

Neuropsychological tests

Neuropsychological assessment was compared between two groups with the Mann-Whitney test using SPSS software 15.0 (SPSS Inc., Chicago, IL).

Structural MR and diffusion tensor imaging analysis

Voxel-based morphometry (VBM) is an automated analysis technique used to investigate focal differences in brain anatomy using VBM tools on SPM (Statistical Parametric Mapping, Wellcome Department of Cognitive Neurology, Institute of Neurology, London, UK, http://www.fil.ion.ucl.ac.uk) software. First, the T1-MR images from all subjects were normalized to the Montreal Neurological Institute (MNI) template and were segmented into gray matter (GM), white matter (WM), and cerebral spinal fluid (CSF). The segmented images were then smoothed with a 12 mm full-width half-maximum Gaussian kernel for the subsequent statistical analysis. As a first step, we conducted a *t*-test to compare cerebral GM morphometric differences between groups using SPM5 (FDR corrected $p < .05$). We then calculated the absolute volume (in mL) of each GM, WM, and CSF segmented region in each individual. The statistical assessment of the absolute volume was evaluated using the Mann-Whitney test on SPSS 15.0.

In addition, amygdala and hippocampal volumes were measured manually accordance with a previously described method (Karchemskiy, Garrett et al. 2011). Structures were manually delineated in the coronal plane using a

total software package (Syngo.via, Siemens Healthcare, Germany). Amygdala, and hippocampal volumes were compared between the boxer group and normal control group by the Mann-Whitney test.

For diffusion tensor imaging (DTI) analysis, preprocessing of DTI data was conducted using FSL4.1 software (Oxford University Centre for Functional Magnetic Resonance Imaging of the Brain, Oxford, UK; http://www.fmrib.ox.ac.uk/fsl/). DTI raw data sets for each subject's 15 diffusion-weighted images were corrected by registering them to their first non-diffusion-weighted image. Non-brain structures were removed using BET, and then fractional anisotropy (FA) and mean diffusivity (MD) maps were calculated using the DTI-FIT program. Image transformation to standard space and voxel-wise statistical analysis were conducted using tract-based spatial statistics (TBSS) (Smith 2002, Smith, Jenkinson et al. 2006). Every FA map was projected onto a skeletonized image at threshold 0.2 (WM extraction). Group comparisons were conducted using a general linear model (GLM). Templates and atlases were defined using JHU ICBM (Johns Hopkins University International Consortium of Brain Mapping) White matter Tractography and JHU ICBM-DTI-81 white matter labels in the FSL library.

^{123}I- FP-CIT SPECT data analysis

We compared the dopamine transporter (DAT) availability and D2 receptor density in the striatum between the boxer group and controls. Each striatum was divided into four subregions: right caudate, left caudate, right putamen, and left putamen. The reference region was the occipital cortex. We drew these five ROI

regions using an automated anatomical labeling (AAL) template on MRIcron (version 17.0). To generate the average FP-CIT template from normal controls, all SPECT data were individually coregistered with a T1 MR Image. Spatial normalization was performed on MNI space. All images were smoothed using an 8 mm Gaussian Kernel. Specific regions of the striatum and non-specific regions were calculated from an ROI in the occipital lobe using the VOI template based on Statistical Probabilistic Anatomical Mapping (SPAM). All SPECT images were processed using PMOD 3.1 software. Analysis of group differences between the boxer group and normal controls for each ROI BP was conducted using SPSS 15.0 software.

^{18}F-FDG PET data analysis

Preprocessing and statistical analysis were performed using SPM5 software implanted in Matlab 7.6 (The Mathworks, Natick, MA). The ^{18}F-FDG PET images were normalized to the standard MNI coordinate system. All images were smoothed using an 8-mm full width at half-maximum (FWHM) Gaussian kernel. Group comparisons were performed between the boxer group and normal control group. After global normalization with proportional scaling, the images for the two groups were compared using a *t*-test with an uncorrected threshold of $p < .005$ with age as a nuisance variable. The steps in the linear regression analyses were as follows. To examine the relationship between glucose metabolic rates in specific regions based on the group-comparison results and neuropsychological scores, we extracted the ROI values in the brain regions for each individual image using MarsBaR ROI toolbox (Brett 2002) in MATLAB. We set the ROIs in the areas with significant changes in the group comparisons of the SPM results.

Proportional scaling in which the images were scaled to a value of 50 with the mean global values that were obtained in each image was applied. The above areas showed significant decreases in glucose utilization in professional boxers compared with those in normal controls. We then performed a linear regression analysis of the ROI values and neuropsychological scores. In addition, a bootstrap analysis was performed on group comparison. Bootstrap resampling was used to create 1,000 different populations that yielded 95% confidence intervals and bias-corrected and accelerated (BCa) confidence intervals with Stata 9.0 software (Stata Corp LP, College Station, TX).

^{18}F-FMZ PET data analysis

All raw DICOM images were converted to 54 analyzed format files in each frame. For realignment of all images, mean images were constructed using the first 30 frame images within the initial 10 min. We then realigned the images from frames 31 to 54 to mean images. After the alignment, all PET images were coregistered to individual T1 3D MR images. All preprocessing of images was performed using PMOD 3.1 software (PMOD Inc., Zurich, Switzerland). Spatial normalization was performed using SPM5. Individual dynamic PET images were spatially normalized to the MNI template. Time-activity curves (TACs) were generated using dynamic PET frames on PMOD software. To examine ^{18}F-FMZ binding in the brain, we employed established kinetic compartment analysis on TACs using the simplified Reference Tissue Model (SRTM) with the pons as a reference region (Odano, Halldin et al. 2009). Group comparisons between the

boxer group and the control group with age as a nuisance variable were conducted with an uncorrected threshold of $p < .005$ set on a voxel value of 30.

For linear regression analysis between neuropsychological parameters and ^{18}F-FMZ binding potential, the ROI value was extracted based on the group-comparison results using MarsBaR for each activation cluster. The ROI was defined based on the areas with significant changes of FMZ uptake in the group comparison SPM results with proportional scaling in which the images were scaled to a value of 50 using the mean global values obtained in each image. We performed a linear regression analysis of the FMZ ROI values and neuropsychological scores. In addition, we conducted a bootstrap analysis on group comparison. Bootstrap resampling was used to create 1,000 different populations that yielded 95% confidence intervals and bias-corrected and accelerated (BCa) confidence interval type using Stata 9.0 Stata Corp LP, College Station, TX).

Results

Clinical screening using UPDRS and CBI scale

The UPDRS scores were higher in the professional boxer group than in controls (boxer group sum: 10; normal control group sum: 0), though all subjects were still within the normal range. The CBI scores were higher in the professional boxer group (boxer group sum: 5; normal control group sum: 0). The results of the neurodiagnostic evaluation are presented in Table 4.

Table 4. Results of diagnostic evaluations

No.	Group	MMSE	UPDRS-3	CBI	UPDRS-3 comment
1	BG001	30	0	0	Normal
2	BG002	29	1.5	1	Delayed left-hand finger tapping
3	BG003	27	2.5	2	Action and postural tremor of hands; delayed left-hand finger tapping
4	BG004	27	6	2	Terminal dysmetria of both hands; hypokinesia
5	BG005	30	0	0	Normal
6	NC001	30	0	0	Normal
7	NC002	30	0	0	Normal
8	NC003	30	0	0	Normal
9	NC004	30	0	0	Normal

BG: boxer group, NC: normal control group, MMSE: Mini-Mental State examination, UPDRS: the Unified Parkinson's disease Rating Scale, CBI: chronic brain injury

Neuropsychological tests

No significant differences between groups were found in cognition (MMSE), depressive symptoms (BDI) and aggressive behavior (BDHI). Motor impulsiveness from the BIS-11 scale was significantly different between groups. In cognitive tasks, a significant difference between the groups was only found in the RCFT delayed recall scores. There were no significant differences in HVLT scores, RCFT immediately recall, and RCFT recognition scores. Results of neuropsychological tests for the boxer and control groups are presented in Table 5.

We tested motor-related performance using the Purdue Pegboard Task (PPB). There was a significant difference in assembly performance between groups ($p < .05$). There were no differences between groups in performances for the right hand, left hand, and both hands. The *t*-test results for the PPB are presented in Table 6.

Table 5. The results of neuropsychological tests for the boxer and control groups

Measures	Boxer group (mean ± *SD*)	Control (mean ± *SD*)	*p*
MMSE	28.6 (1.51)	30.0 (0.00)	.212
BDI	7.0 (3.93)	3.5 (1.91)	.193
BIS-11	48.6 (13.97)	41 (5.16)	.15
Motor	17.2 (3.89)	11.25 (0.50)	**.02** †
Attentional	14.4 (3.36)	11.75 (3.30)	.275
Non-planning	17.20 (3.89)	20.5 (8.42)	.201
BDHI	51.0 (8.00)	38.25 (8.46)	.127
HVLT			
immediately recall	15.2 (3.03)	19.5 (4.43)	.127
delayed recall	4.8 (1.92)	6.75 (3.09)	.282
recognition	20.8 (0.83)	21.0 (1.85)	.832
RCFT			
immediately recall	15.3 (3.40)	18.37 (4.71)	.291
delayed recall	14.7 (3.29)	20.62 (4.02)	**.045**†
recognition	20.2 (1.78)	20.25 (2.06)	.97

† $p < .05$

MMSE: the Mini-Mental State examination, BDI: Beck Depression Inventory, BIS-11: Barratt Impulsiveness Scale-11, BDHI: Buss-Durkee Hostility Inventory, HVLT: Hopkins Verbal Learning Test, RCFT: Rey–Osterrieth Complex Figure Test

Table 6. Group differences in the results of the Purdue pegboard task

Measures	Boxer (mean ± *SD*)	Control (mean ± *SD*)	*p*
Right hand	15.19 (1.89)	14.41 (1.42)	.519
Left hand	13.17 (2.00)	12.55 (1.66)	.635
Both hands	10.72 (0.87)	11.83 (0.88)	.111
Assembly	25.22 (3.48)	30.78 (2.17)	**.028** †

† $p < .05$.

Structural and diffusion brain imaging results using MR and DTI

We conducted two types of structural brain analysis. One is a group comparison using SPM for VBM and the other is the absolute volume extracted using VBM tools. Group differences in gray matter were considered significant at $p <. 05$, FDR corrected. We did not find significant differences in gray matter using SPM analysis. There were no differences in gray matter, white matter, CSF, and total whole brain volumes between the groups. These results are shown in Table 7 and Figure 1.

In an additional analysis, volumes (cm^3) of the amygdala [right: boxers, 1.86 ± 0.24; controls, 1.68 ± 0.14 (p = 0.28); left: boxers, 1.89 ± 0.22; controls, 2.03 ± 0.43 (p = 0.98)] and hippocampal [right: boxers, 4.24 ± 0.57; controls, 4.74 ± 0.51 (p = 0.41); left: boxers, 4.47 ± 0.61; controls, 4.4 ± 0.68 (p = 0.73)] did not differ between the boxer and normal control groups (Figures not shown).

Table 7. VBM analysis results: the absolute volumes of gray matter, white matter, cerebral spinal fluid, and total whole brain volume (mL).

Boxer Group	**GM**	**WM**	**CSF**	**Total**
BG001	676.34	523.78	542.14	1742.26
BG002	613.30	450.41	421.98	1485.68
BG003	649.55	467.99	560.05	1677.59
BG004	675.99	510.78	550.06	1736.83
BG005	681.04	552.87	361.53	1545.44
Mean	659.24	501.17	487.15	1637.56
SD	28.51	41.69	89.89	116.16
Control Group	**GM**	**WM**	**CSF**	**Total**
NC001	622.5	478.01	523.6	1624.11
NC002	596.41	444.85	526.16	1567.41
NC003	656.97	482.85	450.35	1590.18
NC004	627.38	489.14	581.47	1698.00
Mean	625.82	473.71	520.39	1619.93
SD	24.82	19.77	53.79	57.03
p	.190	.412	.904	.904

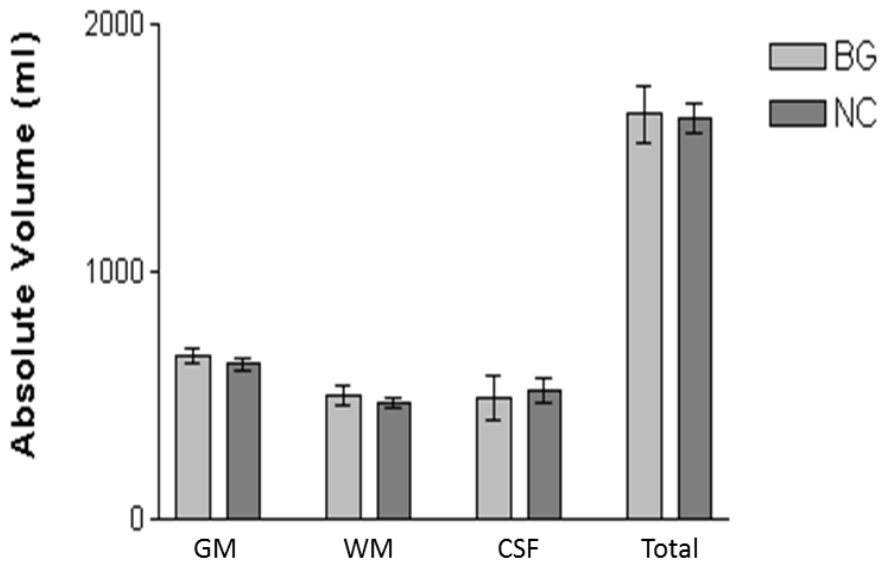

Figure 1. VBM analysis: independent sample *t*-test results comparing groups for absolute volume of gray matter (GM), white matter (WM), cerebral spinal fluid (CSF), and whole brain total volume.

In the DTI results, there were regions of decreased FA in professional boxers compared to controls. These regions included the inferior longitudinal fasciculus (ILF), body of the corpus callosum (BCC), forceps minor (Fmin), anterior corona radiata (ACR), inferior fronto-occipital fasciculus (IFOF), superior longitudinal fasciculus (SLF), and anterior thalamic radiations (ATR). No regions of increased FA were detected. DTI results are shown in Figure 2.

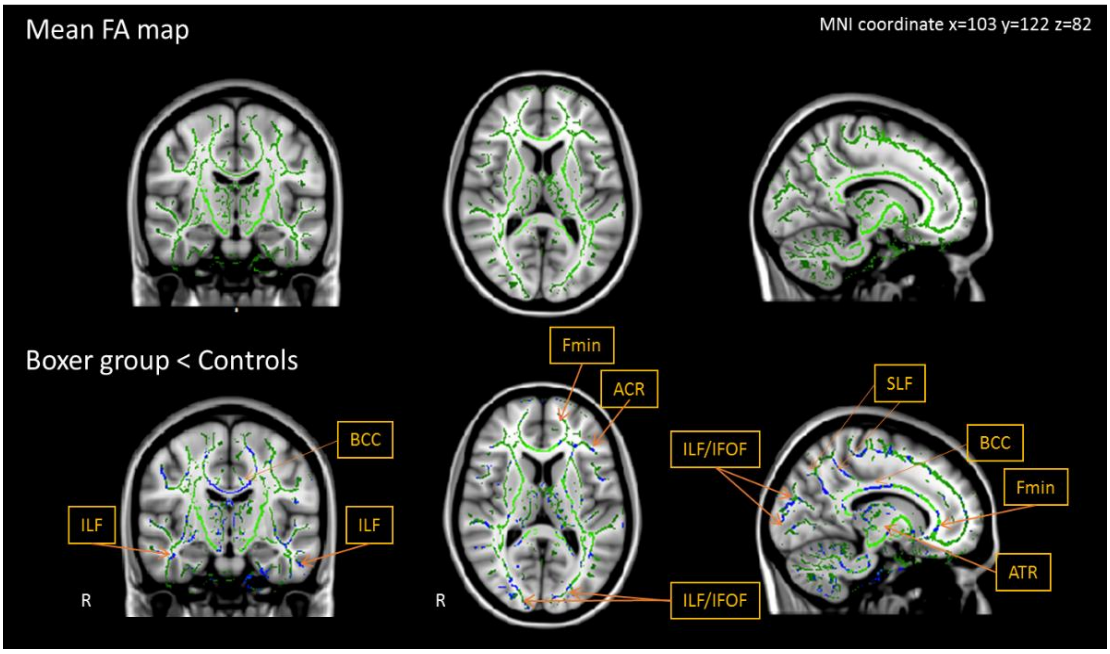

Figure 2. DTI results using TBSS: Upper: Mean diffusivity FA map ($p < .05$) for all subjects in boxer and control groups (green-colored white matter skeleton). Bottom: Significant MD contrast for boxer group < controls (blue). ILF, inferior longitudinal fasciculus; BCC, body of corpus callosum; Fmin, forceps minor; ACR, anterior corona radiata; IFOF, inferior fronto-occipital fasciculus; SLF, superior longitudinal fasciculus; ATR, anterior thalamic radiations.

Striatal dopamine transporter density in subjects exposed repetitive traumatic brain injury using ^{123}I-FP-CIT SPECT

Striatal dopamine transporter binding potential was compared between professional boxers and controls (Figures 3 and 4). There were no differences in the DAT density between the boxer group and normal controls in the whole striatum or any of the subregions (right caudate; p = .484, left caudate; p = .397, right putamen; p = .324, left putamen; p = .248).

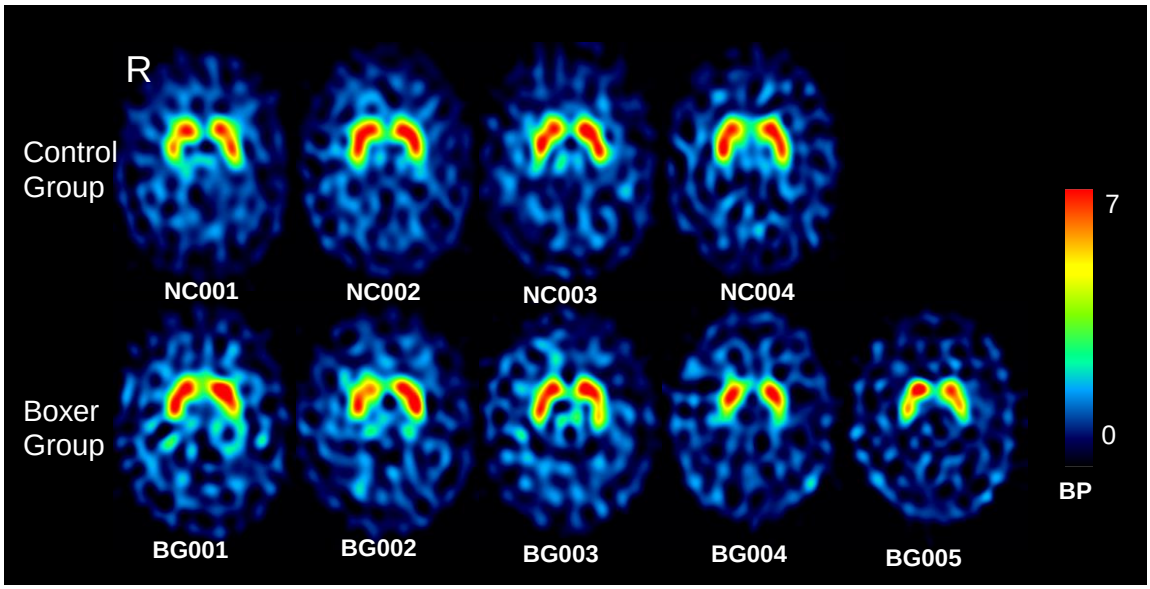

Figure 3. Representative ^{123}I-FP-CIT SPECT image for each subject. BP; binding potential, R: right. Upper: control group (n = 4); lower: boxer group (n = 5).

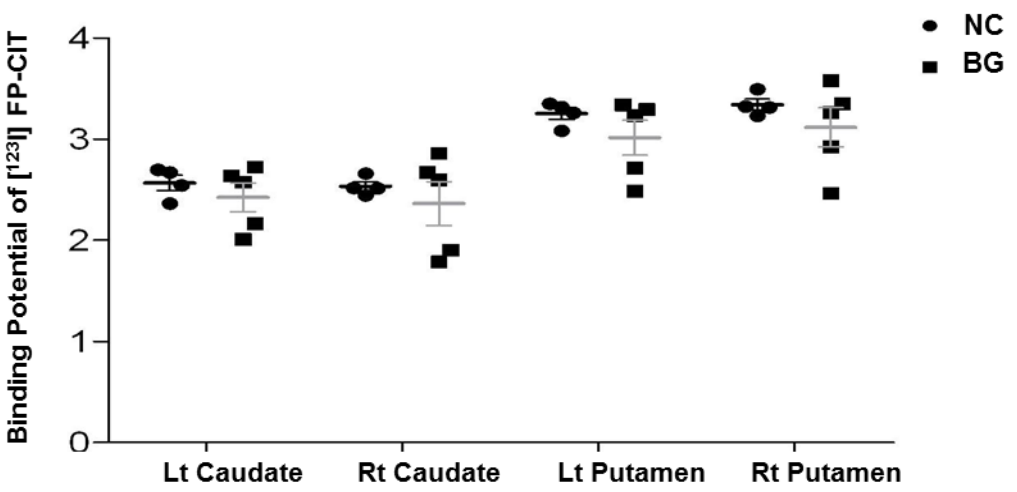

Figure 4. Binding potential of ^{123}I-FP-CIT in the boxer and control groups. DAT density was not significantly different between the groups in subregions of the striatum (left caudate: p = .397; right caudate; p = .484, left putamen; p = .248, right putamen; p = .324).

Brain glucose metabolism differences between groups using ^{18}F-FDG PET

In the ^{18}F-FDG PET results (Figure 5 and Table 8), group comparisons revealed that the professional boxer group had decreased glucose metabolism in the bilateral dorsolateral prefrontal cortices (DLPFC) and right middle orbitofrontal cortex (mOFC) compared to the normal controls ($p < .005$ uncorrected, extent threshold k = 50). Hyper-metabolism was not found in the professional boxer group.

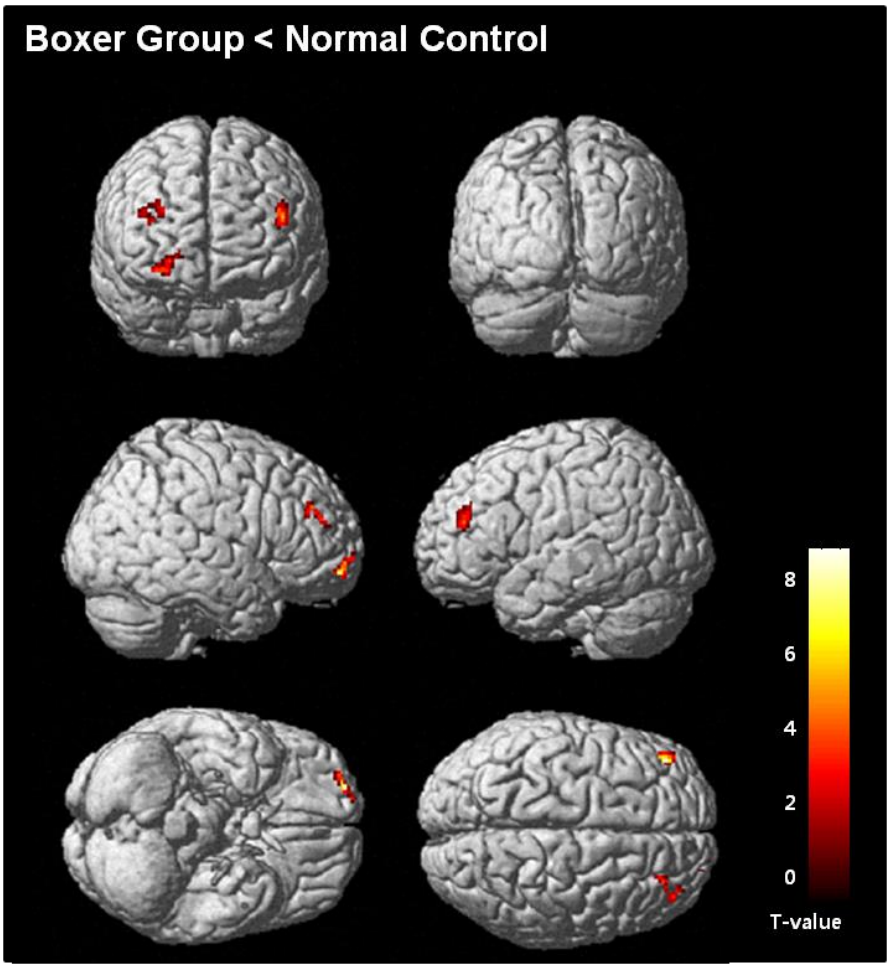

Figure 5. Rendered images from SPM analysis reveal hypo-metabolism in the boxer group and the normal control group (for resting brain activity), with decreased glucose uptake in the right mOFC and bilateral DLPFC ($p < .005$ uncorrected, k = 50).

Table 8. ^{18}F-FDG PET results of group comparisons between boxer group and controls

Region		BA	stereotactic coordinates			*t*-value	K_E
			x	y	z		
	Boxer Group < NC						
Rt	Middle orbitofrontal cortex	11	34	58	–10	8.06	64
Rt	Dorsolateral prefrontal cortex	46	40	46	24	5.42	58
Lt	Dorsolateral prefrontal cortex	46	–44	44	24	5.30	63

BA: Brodmann area, $t = 3.71$ $p < .005$ uncorrected, k = 50

In the ROI analyses with bootstrapping, significant correlations were found between ^{18}F-FDG uptake and the RCFT delayed recall scores in the right mOFC (y = 0.008x - 4.189, R^2 = .496, p < .05) and the right DLPFC (y = 0.010x - 7.552, R^2 = .524, p < .05) (Figure 6).

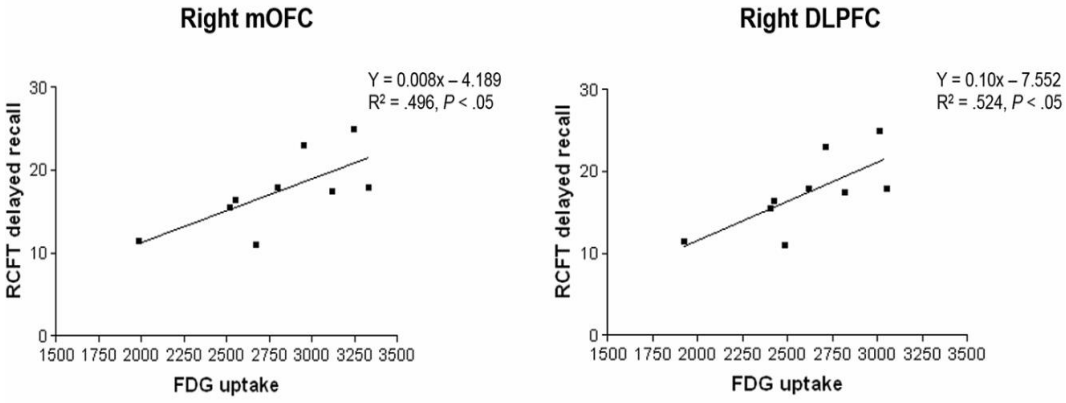

Figure 6. Linear regression analysis between values for brain ROIs and neuropsychological test scores: RCFT delayed recall scores were significantly correlated with ^{18}F-FDG uptake in the right middle OFC (left) and right DLPFC (right).

$GABA_A$ receptor binding as an indicator of neuronal integrity using ^{18}F-FMZ PET

Group comparisons showed significantly lower FMZ uptake in the left angular gyrus (BA 39), left orbitofrontal cortex, left inferior temporal cortex, left superior temporal cortex, right precuneus, and right cerebellum of the professional boxer group compared to the normal control group (Figure 7 and Table 9).

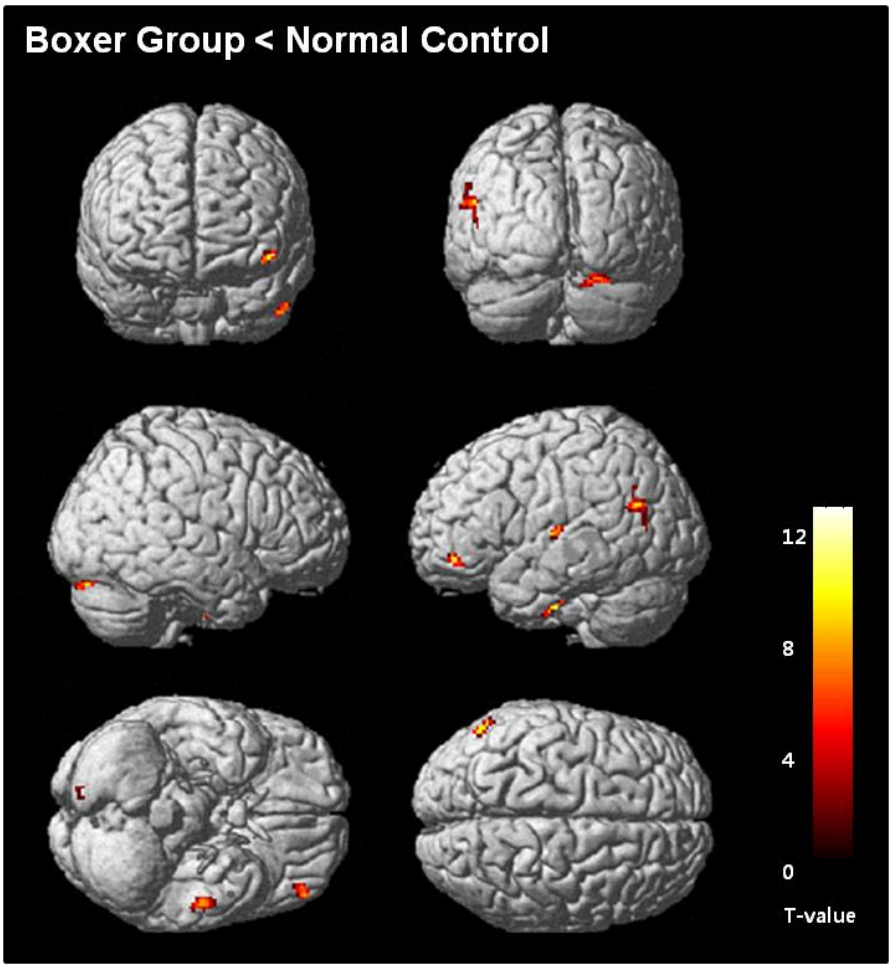

Figure 7. ^{18}F-FMZ PET rendered images from SPM analysis showing group comparisons between the boxer group and control groups for the FMZ BP map. Lower FMZ uptake in boxer group than in controls was apparent in the left

angular gyrus, orbitofrontal cortex, inferior temporal cortex, superior temporal cortex, right precuneus, and right cerebellum ($p < .005$ uncorrected, k = 30).

Table 9. FMZ PET results for group comparisons between boxer group and controls

	Region	BA	stereotaxic coordinates			*t*-value	K_E
			x	y	z		
	Boxer Group < NC						
Lt	Angular gyrus	39	–52	–66	24	12.19	69
Lt	Orbitofrontal cortex	47	–46	46	-8	10.73	37
Lt	Inferior temporal cortex	20	–52	–12	–40	10.09	57
Lt	Superior temporal cortex	48	–46	–18	8	7.35	59
Rt	Precuneus		10	-62	36	7.26	54
Rt	Cerebellum		26	-82	-24	5.15	65

BA: Brodmann area, $t = 3.71$ $p < .005$ uncorrected, k = 30

FMZ uptake was higher in the right postcentral gyrus, precentral gyrus, superior occipital cortex, and inferior parietal cortex in the boxer group than in the control group (Figure 8 and Table 10) ($p < .005$ uncorrected, extent threshold k = 30).

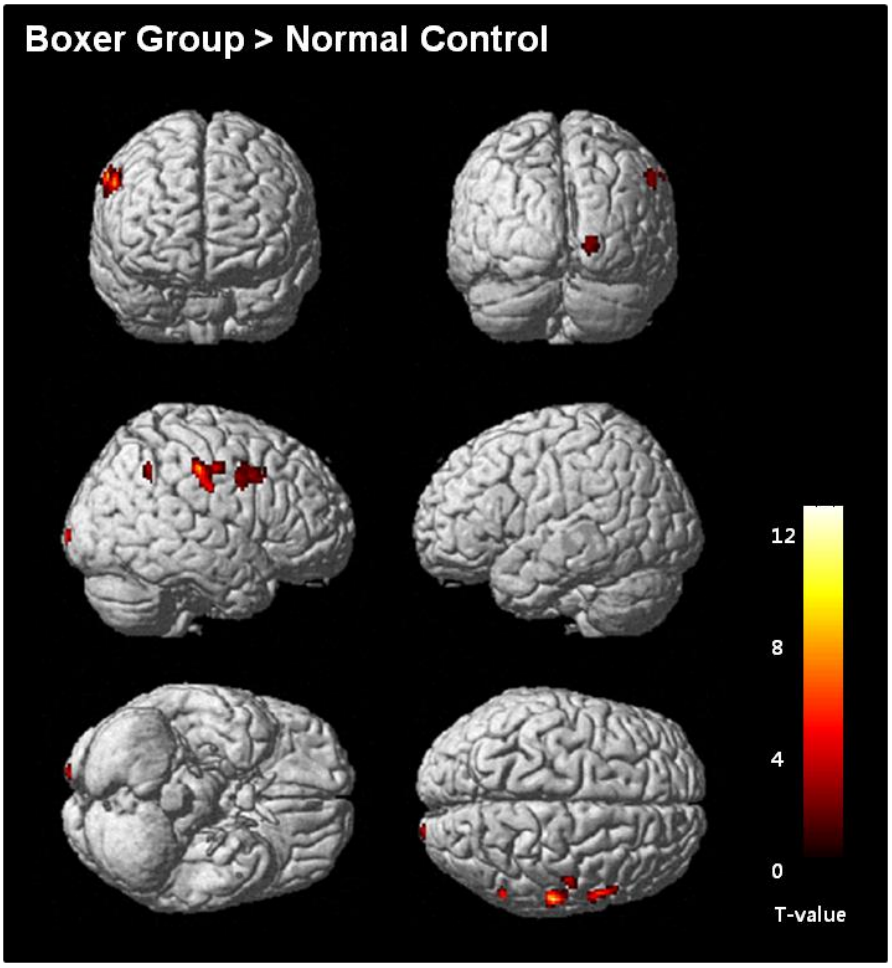

Figure 8. ^{18}F-FMZ PET rendered images from SPM analysis showing group comparisons between boxer and normal control groups for the FMZ BP map. Higher FMZ uptake in the right precentral gyrus, postcentral gyrus, superior occipital cortex, and inferior parietal cortex was apparent in the boxer group than in the controls ($p < .005$ uncorrected, k = 30).

Table 10. ^{18}F-FMZ PET results for group comparisons between boxer group and controls

	Region	BA	stereotactic coordinates			*t*-value	K_E
			x	y	z		
	Boxer Group > NC						
Rt	Postcentral gyrus	3	60	–22	42	27.53	147
Rt	Precentral gyrus	6	56	12	40	6.5	110
Rt	Superior occipital cortex	17	20	–104	2	7.28	41
Rt	Inferior parietal cortex	40	56	–54	44	5.28	36

BA: Brodmann area, $t = 3.71$ $p < .005$ uncorrected, k = 30

We also conducted a linear regression analysis of the neuropsychological test scores and ROI values of the regions with significant changes in the group comparison results. The RCFT delayed recall score was significantly correlated with FMZ uptake in the left angular gyrus (BA 39) ($y = 1.108x - 76.105$, $R^2 = .63$, $p < .05$) (Figure 9).

PPB assembly scores to assess the motor coordination performance showed a significant correlation with uptake in FMZ ROI regions, without there being any significant relationship with uptake in ^{18}F-FDG PET ROI regions. Assembly scores were correlated in the left orbitofrontal cortex and cerebellum ($y = 0.769x - 26.933$, $R^2 = .499$, $p < .05$; $y = 0.604x - 22.785$, $R^2 = .643$, $p < .05$, respectively). The results showing positive correlations with assembly scores in brain ROI regions are presented in Figure 10. The regions with higher FMZ uptakes did not correlate with the scores in any of the neuropsychological tests.

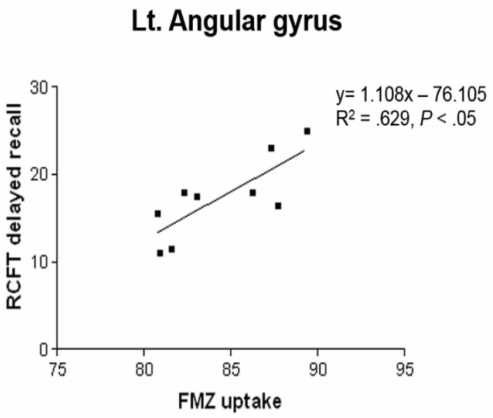

Figure 9. Linear regression analysis results for RCFT delayed recall scores: a significant positive correlations between RCFT delayed recall scores and FMZ uptake in the left angular gyrus.

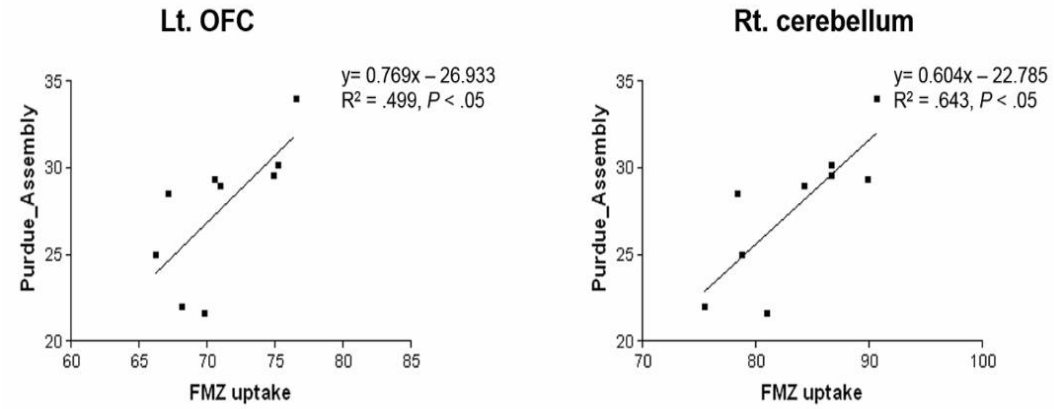

Figure 10. Positive correlations were found between assembly scores to assess motor coordination and FMZ uptake in ROIs in the left orbitofrontal cortex (left) and right cerebellum (right).

Discussion

Amateur boxers do not usually exhibit any significant signs of neuropsychological dysfunction (Butler 1994), or significant abnormalities when examined using CT and MRI (Haglund and Eriksson 1993). The level of competition (i.e., professional versus amateur) and the duration of the boxing career may all confer different degrees of CTE risk (Gavett, Stern et al. 2010). For these reasons, we recruited retired professional boxers with careers spanning more than 10 years. This study evaluated the cognitive and motor function of professional boxers using various neuropsychological tests, and investigated brain mechanisms underlying repetitive TBI using structural MR, DTI, ^{123}I-FP-CIT SPECT, ^{18}F-FDG PET and ^{18}F-FMZ PET.

In a review of autopsy results, McKee et al. reported 46 neuropathologically diagnosed cases of CTE (90%) among 51 athletes. The most prominent neuronal loss was seen in the hippocampus, entorhinal cortex, and amygdala, with less severe losses in the subcallosal and insular cortex, olfactory bulbs, mammillary bodies, locus coeruleus, substantia nigra, medial thalamus, and cerebral cortex (McKee, Cantu et al. 2009). Strictly, CTE can only be definitively diagnosed at autopsy (Jordan 2013). However, criteria have been proposed for the diagnosis of CTE in patients, and these classify the clinical features into four categories in line with other neurological diseases: definite, probable, possible, and improbable CTE (Jordan 2013).

Neuroimaging studies can provide evidence to support the CTE classification. Jordan also proposed a CBI screening scale to classify CTE in patients with repetitive brain damage (Jordan 2000). According to the CBI

screening criteria, only three of the five professional boxers in our study seemed to exhibit possible CTE. However, we included all of the professional boxers in our analyses because previous studies only considered careers or periods of athletic activity. UPDRS-3 and CBI screening provided a wealth of information on the symptoms of subjects.

Most of the neuropsychological tests did not show significant differences between the groups. Professional boxers performed significantly worse than controls in the RCFT delayed-recall, and the 'assembly' task in the PPB. Motor impulsivity (sub-score of BIS-11) was higher in the boxer group than in controls, reflecting tendencies to act on the spur of the moment. CTE has previously been associated with mood disorders such as depression, agitation, social withdrawal, poor judgment, and aggression. (Guterman and Smith 1987). The Purdue pegboard test provides a reliable measurement for diagnosing compromised motor function under clinical conditions, especially motor coordination involving both hands.

We conducted structural brain imaging using MRI but there were no significant differences in gray matter, white matter, and CSF volumes between groups. Additionally, amygdala and hippocampal volumes did not significantly differ between the groups. Previous studies have revealed difficulties in reliably detecting chronic brain injury using CT or MRI scans (Huisman, Schwamm et al. 2004, Jantzen, Anderson et al. 2004). DTI methods provide insight into the brain's white-matter microstructures, but few studies have been performed on boxers. Diffusion defects were also found in whole brains of boxers before brain abnormities were detectable on standard MRI (Zhang, Ravdin et al. 2003). Abnormal diffusion in multiple brain regions of professional boxers with no

history of severe head trauma has been positively correlated with age and boxing duration (Chappell, Ulug et al. 2006).

In our DTI results, significantly decreased FA was found in some brain regions in professional boxers compared to controls. Among these regions, the inferior longitudinal fasciculus (ILF) has classically been defined as a direct connection from the occipital cortex to the temporal lobe (Afifi and Bergman 2005), and the inferior fronto-occipital fasciculus (IFOF) directly connects the occipital, posterior temporal, and the orbitofrontal areas. These structures have functional similarity as well spatial connections, and play a role in visual perceptual processing and object recognition (Ortibus, De Cock et al. 2011). FA was lower in the body of the corpus callosum (CC) and anterior forceps (on either side of the CC) in boxers than in controls. The CC is the largest white-matter structure in the brain, and its role is essentially to integrate information between the left and right hemispheres and facilitate faster transmission of information associated with computation, memory, and cognition (Fitsiori, Nguyen et al. 2011).

We used various biomolecular imaging techniques to investigate the brain mechanisms underlying repetitive brain trauma. CTE encompasses gait disorders, speech slowing and extrapyramidal signs, and neuropsychiatric and behavioral symptoms (Jordan 2000, Costanza, Weber et al. 2011). In the first study of repetitive brain injury by Martland in 1928(HS 1928), “punch drunk syndrome” referred to a Parkinsonian syndrome that results from long-term cumulative consequences of subclinical concussions to the head. The motor manifestations of CTE such as spasticity, intention tremor, ataxia, dysarthria, and coordination problems, reflect injury to the pyramidal tracts, the extrapyramidal

system, and the cerebellum. In the studies by McKee et al. movement abnormalities eventually developed in 42% subjects of the 51 CTE patients (McKee, Cantu et al. 2009).

In our study, three of the five boxers were diagnosed with some slight impairment of hand movements. Nevertheless, they were still within the normal range for UPDRS scores. ^{123}I-FP-CIT SPECT findings showed that dopaminergic function was still preserved in professional boxers, and striatal DAT availability was not significantly different between groups. Dopaminergic function may be influenced by the severity of brain damage.

In the resting state, a decrease in glucose metabolism in the brain is commonly thought to reflect functional abnormalities. In our ^{18}F-FDG PET results, lower glucose uptake was found in the bilateral DLPFC and middle orbitofrontal cortex. These regions participate in cognitive/executive function and inhibitory control of decision making (Damasio, Grabowski et al. 1994, Elliott 2003). Impairment of these regions is associated with cognitive dysfunction related to memory, attention, and coordination of information. Of the various neuropsychological tests, the RCFT delayed recall score can measure the ability to retain and retrieve information. Our ^{18}F-FDG findings indicating impaired visuospatial memory function were similar to a previous finding that repeated mild head injury in mice impaired motor function, short-term visuospatial memory, and complex learning (Hylin, Orsi et al. 2013).

In previous studies, TBI patients without abnormalities on MRI have demonstrated low BP on ^{18}F-FMZ PET, which reflects low neuronal integrity and neuronal cell loss in the brain (Shiga, Ikoma et al. 2006). Our ^{18}F-FMZ PET results are consistent with neuronal cell loss caused by repetitive brain damage.

In group comparison analyses, significantly lower FMZ uptake was seen in the left angular gyrus (BA 39), left orbitofrontal cortex, left inferior temporal cortex, left superior temporal cortex, right precuneus, and right cerebellum in boxers than in normal controls. These regions of low uptake indicate neuronal damage: damage to the angular gyrus (BA 39) is known to play a role in dyslexia and semantic aphasia (Beauvois, Saillant et al. 1978, Pugh, Mencl et al. 2000); neurofibrillary tau pathology in CTE is found in temporal lobe structures (McKee, Cantu et al. 2009); and marked cortical atrophy of the right temporal lobe has been observed in boxers (Corsellis, Bruton et al. 1973). However, increased FMZ uptake was observed in the boxer group compared to the controls, and this increase may have reflected an upregulation of benzodiazepine-binding sites. These results suggested the presence of a compensatory mechanism involving the posttraumatic cortical neurogenesis and the endogenous brain-derived neurotrophic factor (Quadrato, Elnaggar et al. 2014). Although the damage is selective in TBI, formation of new neurons has been observed in damaged brain circuitry after neuronal injury (Richardson, Sun et al. 2007).

Our results showed significant positive correlations between assembly scores and FMZ uptake in the left orbitofrontal cortex and right cerebellum. The orbitofrontal cortex is involved in activation and maintenance of task goals and action selection in behavior (Ridderinkhof, van den Wildenberg et al. 2004). A previous study of the cerebellar involvement in motor function showed that Purkinje cells, which are a class of GABAergic neurons in the cerebellum, have a crucial relationship with motor coordination (Schiffmann, Cheron et al. 1999, Lalonde and Strazielle 2003).

This study has several limitations that need to be addressed. First, the

small sample size was definitely a limitation in this study, even though it was difficult to recruit professional boxers who met our standard criteria. Therefore, in the small population, we conducted bootstrapping as a resampling statistical method involving 1,000 repeated populations in order to strengthen the statistical power. And yet, in a future study, a larger sample size is needed in order to obtain a smaller variance and higher statistical power of verification. Second, we did not obtain congruity between the ^{18}F-FDG PET results and the FMZ PET results, even though they both represent neuronal activities in the living human brain. Lastly, we would like to emphasize an issue regarding the generalizability of this study. Traumatic brain injuries are not unique to professional boxers. Despite the prevalence of brain injury from kindergarten to high school, relatively little research on the long-term health consequences of concussion has been conducted in child athletes than in college and professional athletes. Scientists have an incomplete understanding of what happens when a child's brain strikes the inside of the skull during a blow to the head and how this affects neurological development.

To summarize the important issues raised in this study, the main symptomatology in punch drunkenness can be regarded as Parkinsonian and Alzheimer-like (Sosa, Linic et al. 2011). The molecular brain imaging results in the professional boxers indicated some impairments of cognitive function, hypometabolic rates of glucose metabolism, and lower FMZ BP values, which resemble Alzheimer-like symptoms. The motor-related deficits in assembly performance and the slight impairments in the hand/finger movements suggested Parkinsonism. However, this study indicated that rTBI was not an extension of Alzheimer's disease or PD. We suggest that cognitive impairment and motor-

related dysfunction are due to neuronal damage caused by chronic brain injury in professional boxers experiencing repetitive trauma. These changes were revealed by molecular brain imaging although no abnormalities were seen on structural brain MRI.

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국 문 초 록

반복적인 뇌 외상에 의한 뇌 구조 및 기능의 변화: 뇌 기능 및 분자 영상연구

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반복적 외상성 뇌 손상 (repetitive traumatic brain injury, rTBI)은 반복적인 뇌진탕 혹은 가벼운 외상성 뇌 손상의 축적에 의한 것으로 알려져 있다. 만성 외상성 뇌 병증 (Chronic traumatic encephalopathy, CTE)은 반복적인 뇌 외상으로 인한 퇴행성 뇌 질환이다. 반복적인 뇌 외상은 주로 복싱, 레슬링 야구 등과 같은 스포츠와 밀접하게 관련되어 있다. 특히 복싱은 경기 및 스파링 과정에서 수많은 머리 타격을 받는 특성 때문에, 복서들을 대상으로 한 뇌 손상 연구는 활발히 이루어져왔다. 임상적으로 CTE 는 운동 및 행동적 기능저하, 우울증과 같은 신경증적 질환과의 연관성 및 인지기능의 저하 등의 증상들이 수반된다고 알려져 있다.

본 연구의 목적은 반복적인 뇌 외상에 따른 뇌의 변화에 대하여 구조적 뇌 영상과 분자화학적 뇌 영상을 통하여 알아보고, 또한 여러 신경심리검사 및 운동기능검사를 통해 인지-행동적 결과와 뇌 영상의 결과들과의 상관성을 평가하는 것이다.

퇴직한 프로페셔널 복서 5 명 (평균연령: 46.8 ± 3.19 세)과 연령과 성별을 일치시킨 정상피험자 4 명 (평균연령: 48.5 ± 3.32 세)이 이 연구에 참여하였다. 모든 참가자에게 파킨슨평가척도검사 (UPDRS) 중 운동영역 검사와 만성 뇌 손상척도 (CBI), 간이정신상태검사 (MMSE), 우울증검사 (BDI), 충동검사 (BIS-11)와 공격성검사 (BDHI)를 시행하였다. 또한, 인지기능 및 기억력 평가를 위한 한국판 홉킨스 언어 학습 검사, Rey 복합 도형 검사와 운동

협응 능력 평가를 위한 퍼듀 막대 검사가 시행되었다. 뇌 영상 획득 방법으로는, 뇌의 구조적/형태적 평가를 위해 뇌 자기공명영상과 자기공명 확산텐서영상을 획득하였다. 뇌 분자 화학 영상으로는 대 뇌 포도당대사 영상과 대 뇌 도파민 수송체 영상, 대 뇌 가바수용체 영상을 통한 대 뇌 피질 내 신경세포의 손상을 평가하였다.

신경심리평가 및 인지기능-운동기능검사에서 복서집단은 정상집단에 비하여 지연된 시공간 기억회상과 운동 협응 점수에서 유의한 수행 저하를 보였다. 자기공명영상 결과, 집단간 형태구조적 차이는 발견되지 않았다. 또한 대 뇌 도파민 수송체 영상 결과 정상그룹과 비교하여 도파민 결합능의 유의한 차이가 발견되지 않았다. 반면 대 뇌 포도당대사 영상에서 복서그룹에서 안와전두피질과 전전두피질에서의 포도당대사의 유의한 감소가 관찰되었고, 이는 판단 및 의사결정과 같은 상위 인지기능과 관련된 영역이었다. 가바 수용체 영상을 통한 신경세포 보존성(integrity)에 대한 집단간 차이 검증에서 대 뇌 좌측의 안와전두피질과 측두피질의 상위 및 하위 영역, 설전부, 각회 및 소뇌영역에서 정상집단에 비하여 플루마제닐의 결합능이 감소하였다. 반면 우측 중심뒤이랑과 중심앞이랑, 하두정엽피질, 상부후두엽 영역의 플루마제닐 결합능은 정상집단에 비하여 증가하였다. 그리고 이 영역들 중 좌측 각회, 안와전두피질, 소뇌의 결합능은 지연된 시공간 기억 회상점수와 운동 협응 점수와 각각 유의한 정적 상관을 보였다.

본 연구에서는 반복적 외상성 뇌 손상 집단에서 지연된 시공간기억 인출과 운동 협응 수행의 저하, 뇌 포도당대사 저하, 가바수용체 결합능의 유의한 차이들이 관찰되었다. 또한 인지-행동 검사결과와 뇌 영상 결과와의 상관성 또한 관찰할 수 있었다. 이는 반복적으로 축적된 뇌 타격에 따른 만성적 뇌 손상에 의한 신경적 손상에 의한 결과로 보이며, 이러한 대 뇌 신경 손상들은 뇌의 구조적 영상에서는 발견되지 않은 반면, 분자화학적 영상을 통해 발견할 수 있었다.

주요어: 반복적 외상성 뇌 손상, 신경심리평가, 구조적 뇌 영상, 도파민 수송체, 뇌 포도당 대사, 가바수용체

학 번: 2007-30552