ORIGINAL

Default mode network abnormalities in children with autism spectrum disorder detected by resting-state functional magnetic resonance imaging

Yasuhiro Funakoshi[1], Masafumi Harada[2], Hideki Otsuka[2], Kenji Mori[3], Hiromichi Ito[4], and Takashi Iwanaga[1]

[1]*Department of Medical Imaging, Institute of Biomedical Sciences, University of Tokushima Graduate School, Japan,* [2]*Department of Radiology, Institute of Biomedical Sciences, University of Tokushima Graduate School, Japan,* [3]*Department of Child Health & Nursing, Institute of Biomedical Sciences, University of Tokushima Graduate School, Japan,* [4]*Department of Pediatrics, Institute of Biomedical Sciences, University of Tokushima Graduate School, Japan*

Abstract : Purpose : The purpose of this study was to investigate changes in the functional connectivity of the default mode network (DMN) in normal aging and in children with autistic spectrum disorder (ASD) by using resting-state functional magnetic resonance imaging (rsfMRI) and independent component analysis. Methods : Thirty-one healthy controls (HC) in four age groups (1-3, 4-8, 20-29, and 50-59 years) and 14 childhood ASD cases (1-8 years of age) were examined by rsfMRI echo-planar imaging on a clinical 3-T MRI scanner. Imaging of all children (1-8 years) was conducted under sedation, while adults were scanned in the awake state with eyes closed. Results : The regions of DMN functional connectivity in the bilateral inferior parietal lobule and posterior cingulate cortex were smaller in HC children than in HC adults, and smaller in the ASD group than in the HC children. Conclusion : It is possible to observe developmental and pathological changes in the DMN by rsfMRI. Reduced DMN functional connectivity in children may be a useful biomarker for ASD diagnosis. J. Med. Invest. 63 : 204-208, August, 2016

Keywords **:** *default mode network, resting state MRI, autism spectrum disorder, interior parietal lobule, posterior cingulate cortex, development*

INTRODUCTION

Functional blood oxygen level dependent (BOLD) magnetic resonance imaging (MRI) studies without task-related stimulation have revealed a widespread resting-state neural network characterized by spontaneous low-frequency (0.01-0.1 Hz) BOLD fluctuations (intrinsic activity), termed the resting-state functional MRI (rsfMRI), which plays important roles in various brain functions such as default mode network (DMN), executive control, salience, dorsal attention, and auditory, visual, and sensorimotor mechanisms (1). The DMN is an attractive rsfMRI manifestation for clinical applications because it is unbiased by task performance (e.g., finger tapping), which may vary among subjects. Moreover, this technique is applicable to patients with disturbance of consciousness or disabilities. Indeed, rsfMRI measures have revealed specific regional DMN abnormalities in patients with psychiatric and/or neurological disorders such as Alzheimer's disease, schizophrenia, and bipolar disorder and may aid in clinical diagnosis. For instance, it was reported that patients with Alzheimer disease (2) and schizophrenia (3) exhibit lower DMN functional connectivity in the posterior cingulate cortex (PCC) compared with healthy controls (HC).

Autism spectrum disorder (ASD) commonly manifests as behavioral signs and symptoms during the first 2 to 3 years of life, such as delayed speech, communication disorders, and repetitive behaviors. Clinical signs of ASD emerge even earlier, particularly brain overgrowth between 1 to 2 months and 6 to 14 months of age (4) due to accumulation of an excess number of neurons in the prefrontal cortex (5). Several recent reports have assessed brain function in ASD using rsfMRI (6-9) but none has specifically examined DMN abnormalities in children with ASD. To address this issue, it is first necessary to describe the functional changes in the DMN during normal brain development. The aims of this study were to describe differences in functional connectivity in the DMN of HC during normal aging by comparing multiple age groups (1-3, 4-8, 20-29, and 50-59 years) and then to compare functional DMN connectivity in developmentally normal children (1-8 years) and age-matched children with ASD.

Received for publication January 28, 2016 ; accepted March 28, 2016.

Address correspondence and reprint requests to Yasuhiro Funakoshi, 3-18-15 Kuramoto-cho Tokushima 770-8503, Japan and Fax : +81-88-633-7174.

MATERIALS AND METHODS

Subjects

Four age groups of HC subjects, 1-3 years (7 male, mean age 2.3±1.0), 4-8 years (7 male and 1 female, mean age 4.8±1.5 years), 20-29 years (7 male and 1 female, mean age 22.5±0.8 years), and 50-59 years (7 male and 1 female, mean age 55.9±2.8 years) as well as a group of children with ASD aged 1-8 years (13 male, 1 female, mean age 3.4±1.8 years) were examined by rsfMRI. All children with ASD were diagnosed by two experienced pediatric neurologists according to the criteria of DSM-Ⅳ-TR. All of the children were sedated with triclofos sodium (Tricloryl, 0.5 mL/kg body weight) 30 min before the measurement following the guidelines for monitoring and management of pediatric patients during and after sedation published by the American Academy of Pediatrics (10). The adult subjects were instructed to keep their eyes closed and not to fall asleep during scanning.

This study was approved by the Institutional Review Board of Tokushima University Hospital. Permission was obtained from the parents of all children, and when possible, the assent of the child

was obtained. Informed consent was also obtained from all adult subjects after they received an explanation of the content and purpose of this study.

fMRI data acquisition

Imaging data were acquired on a 3.0-T whole-body scanner (Discovery MR 750, GE Healthcare, Waukesha, WI) with an 8-channel phased array coil. High-resolution anatomical images were acquired using a T1-weighted fast spoiled gradient recalled echo sequence. Measurement conditions were as follows : repetition time (TR) = 8.7 ms, echo time (TE) = 3.6 ms, flip angle = 15°, field of view (FOV) = 240 mm^2, matrix size = 256×320, axial slices = 86, slice thickness = 1.8 mm. Resting state fMRI data were acquired using a T2*-weighted gradient-echo echo-planner imaging sequence. Measurement conditions were as follows : TR = 2000 ms, TE = 27 ms, flip angle = 90°, FOV = 240 mm^2, matrix size = 128×128, axial slices = 35, slice thickness = 4.0 mm, no gap. Ninety-six functional volumes were acquired over a total imaging time of 3 min 18 s. The first 3 images of the scan were not included in the data analysis.

fMRI data analysis

Resting-state functional images were preprocessed using SPM8 (Wellcome Department of Imaging Neuroscience, London, UK). First, images were time corrected (slice timing) and subsequently realigned using rigid transformation. Next, functional and anatomical images were coregistered. Both the functional and anatomical images were normalized to the Montreal Neurological Institute space with a voxel size of 3×3×3 mm using the T1 template of SPM8. Finally, the normalized images were spatially smoothed with an 8 mm^3 FWHM Gaussian kernel.

Spatial independent component analysis (ICA) was conducted for rsfMRI data acquired from the 31 HC and 14 ASD subjects using GIFT software (http : //icatb.sourceforge.net/). The number of components was determined using the minimum description length criteria, which yielded 22 components for the HC group and 26 for the ASD group. The individual subject components were converted to Z-values using the z-shift GIFT utility. For analysis, we selected DMN components including the PCC and inferior parietal lobule (IPL). Individual contrast maps of the DMN for HC and ASD groups were entered into a full factorial second-level analysis in SPM8. These maps were used to create masks for analyses using the Masking toolbox (http : //www0.cs.ucl.ac.uk/staff/g.ridgway/masking/).

RESULTS

Differences in default mode network functional connectivity among the healthy control age groups

In all four HC age groups, the results of ICA revealed typical DMN activity (P = 0.001 with family-wise error [FWE] and k = 2 voxels) (Fig. 1A), including strong functional connectivity between the PCC and bilateral IPL (Figure 2). The regions of DMN functional connectivity in the PCC and bilateral IPL, as measured by total significant voxel cluster number, were smaller in HC children (1-3 years and 4-8 years) compared with adults aged 20-29 and 50-59 years (Table 1).

The maximum Z-scores for the PCC did not differ significantly between HC children and adults (Fig. 1B), while the score for the right (R)-IPL was significantly smaller in the 1-3 years HC group than in the 20-29 years HC group (P = 0.015) and the 50-59 HC group (P = 0.002). Similarly, the maximum Z-score for the R-IPL was significantly smaller in the 4-8 years HC group compared with the 20-29 years HC group (P = 0.011) and the 50-59 years HC group (P = 0.001). The maximum Z-score for the left (L)-IPL was also significantly smaller in the 4-8 years HC group compared with the 50-59 years HC group (P = 0.015) (Fig. 1(B)).

Differences in default mode network functional connectivity between healthy control children and children with autism spectrum disorder

Comparison of the 14 ASD and 15 age-matched HC children (1-8 years) revealed typical DMN activity (P = 0.05 with FWE and k = 2 voxels) and structure (Fig. 3). However, regions of DMN functional connectivity in the bilateral IPL and PCC were smaller in the ASD group. Significant differences in DMN functional connectivity were observed in the bilateral angular gyrus, cuneus, calcarine gyrus, and superior occipital gyrus (P = 0.001 and k = 9 voxels) (Fig. 4, Table 2).

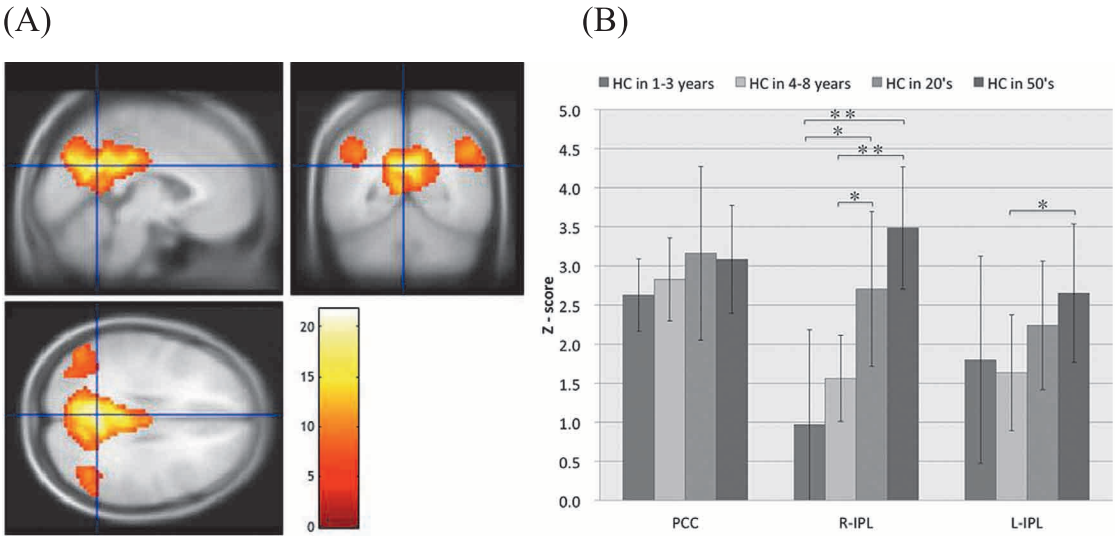

Figure 1. (A) Functional connectivity of the default mode network in the healthy control (HC) group. The statistical threshold was set at P = 0.001 with family-wise error and k = 2 voxels. (B) Mean maximum Z-score in three regions of the DMN are shown : PCC (-3, -55, 28), right (R)-IPL (42, -70, 37), and left (L)-IPL (-39, -64, 43). Z-scores were compared among regions and HC age groups by Mann-Whitney U test. *= P< 0.05 and **= P< 0.01.

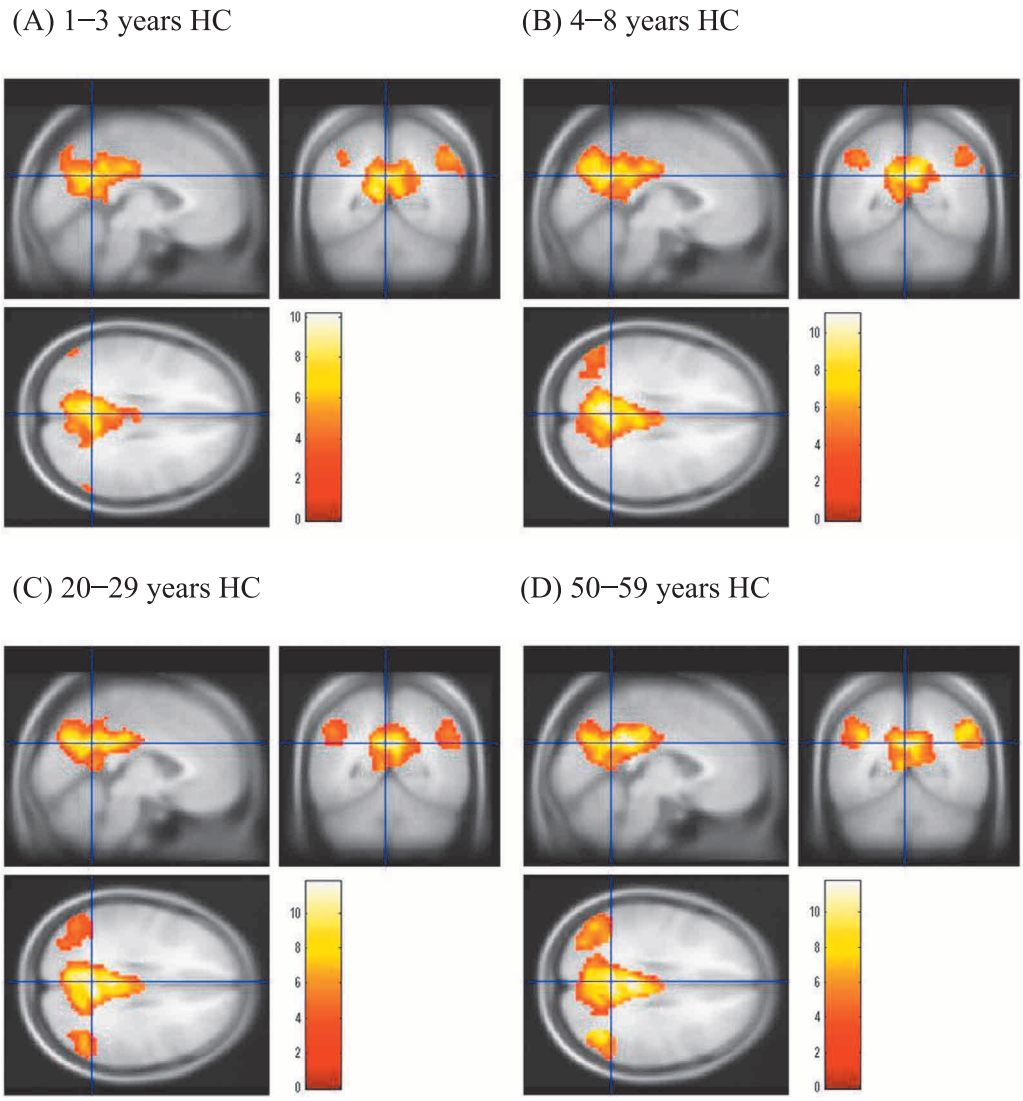

Figure 2. Maps of default mode network functional connectivity for each healthy control (HC) age group. The statistical threshold was set at P=0.001 and k=8 voxels.

Table 1. Regional default mode network functional connectivity in the healthy control age groups

Age groups	Region	MNI coordinates			Cluster level (voxels)	t-score
		X	Y	Z		
1-3 years	L precuneus	-9	-58	16	912	10.13
	R angular gyrus	42	-58	43	60	5.80
	L angular gyrus	-51	-70	37	41	5.72
4-8 years	L precuneus	0	-55	28	1437	11.04
	R angular gyrus	45	-70	37	6	5.01
	R inferior parietal lobule	45	-55	46	7	4.94
	L middle occipital gyrus	-30	-70	40	183	8.19
20-29	R precuneus	6	-52	28	1223	11.79
	R angular gyrus	42	-64	37	244	9.27
	L angular gyrus	-39	-67	43	270	7.98
50-59	L precuneus	-3	-55	28	1332	11.69
	R middle occipital gyrus	39	-73	34	369	11.75
	L angular gyrus	-42	-67	40	457	9.22

The statistical threshold was set at P=0.05 with family-wise error and k=3 voxels. MNI, Montreal Neurological Institute ; L, left ; R, right.

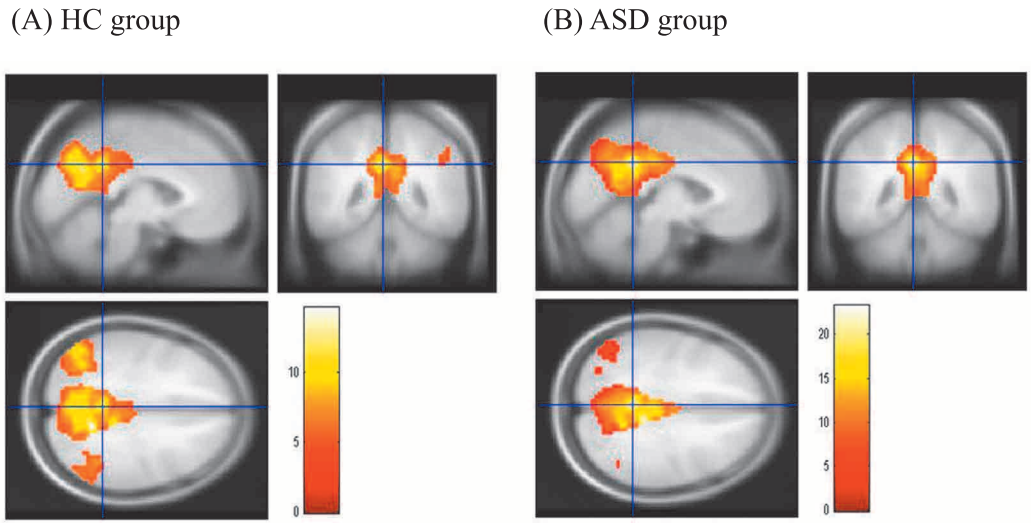

Figure 3. Default mode network functional connectivity in healthy control (HC) children and age-matched children with autism spectrum disorder (ASD). The statistical threshold was set at P=0.05 with family-wise error and k=2 voxels.

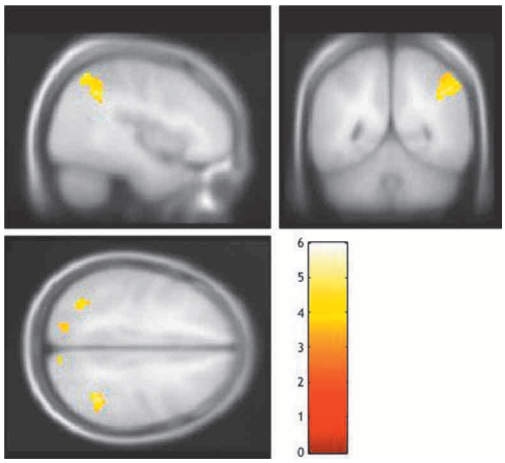

Figure 4. Regions with significant differences in default mode network functional connectivity between children with autism spectrum disorder (ASD) and age-matched healthy control (HC) children (HC> ASD). The statistical threshold was set at P=0.001 and k=9 voxels.

DISCUSSION

We observed developmental increases in DMN functional connectivity in the PCC and bilateral IPL, as well as reduced connectivity in the PCC and bilateral IPL in children with ASD compared with age-matched developmentally normal children using spatial ICA of rsfMRI data. As rsfMRI does not rely on specific cognitive or motor tasks, it can reveal DMN changes in subjects spanning a wide range of ages and functional abilities, including patients with cognitive and motor deficits. Thus, changes in DMN may be a valuable biomarker for the diagnosis and prognosis of neurological diseases, including ASD.

In the first phase of this study, we showed that DMN structure was similar between developmental normal children (1-8 years) and adults (20-29 and 50-59 years), although the DMN functional connectivity in the PCC and bilateral IPL was weaker in children. Using ICA as in this study, Onoda *et al.* (2012) reported that adults 36-86 years showed decreased DMN functional connectivity during normal aging (11), and Jones *et al.* (2011) reported that adults 64-91 years showed decreased functional connectivity, particularly in the posterior DMN (2). Moreover, age-related weakening of DMN functional connectivity was associated with cognitive dysfunction (2, 12). Few studies have conducted similar comparisons between adults and young children, but most reported a similar trend of stronger connectivity from childhood to young adulthood/

Table 2. Regional differences in default mode network functional connectivity between children with autism spectrum disorder (ASD) and age-matched healthy controls (HC) (HC> ASD)

Region	MNI coordinates			Cluster level (voxels)	t-score
	X	Y	Z		
R cuneus	15	-82	31	19	5.96
R angular gyrus	42	-52	37	112	5.14
L calcarine gyrus	-9	-73	16	29	4.61
L superior occipital gyrus	-18	-82	40	17	4.48
L angular gyrus	-36	-64	34	15	4.13

The statistical threshold was set at P=0.001 and k=9 voxels. MNI, Montreal Neurological Institute ; L, left ; R, right.

middle age, despite using different approaches such as graph theory (13) and seed-based analysis (14).

In the second phase of this study, we showed weaker DMN functional connectivity in the PCC and bilateral IPL in children with ASD compared with age-matched controls. These results are consistent with findings in adult ASD (6, 7). Buckner *et al.* (2008) reported that the core regions associated with the DMN include the ventral medial prefrontal cortex, posterior cingulate/retrosplenial cortex, IPL, lateral temporal cortex, dorsal medial prefrontal cortex, lateral temporal cortex, dorsal medial prefrontal cortex, and hippocampal formation (15). DMN function has been associated with self-relevant, internally directed information processing (16) with each DMN region related to different features. The PCC was associated with episodic memory retrieval (17) and visuospatial imagery (18). Furthermore, Sestieri *et al.* (2011) reported an interaction of the DMN with other networks during episodic memory retrieval (19), including activation of the angular gyrus (a subregion of the inferior parietal lobe) and PCC/precuneus. Therefore, we suggest that the change in functional connectivity between PCC and IPL is associated with specific impairments in ASD, such as social cognition and communication disorders.

Several factors are known to disrupt DMN functional connectivity as measured by rsfMRI, including both sedation and stimulation. However, not using sedation in children introduces interference from motion artifacts and sensory (visual) activation. The effects of sedation on DMN are inconsistent across studies. Stamatakis *et al.* (2010) reported that the anesthetic agent propofol increased connectivity with the PCC, including from areas outside the DMN (20). On the other hand, Greicius *et al.* (2008) reported significantly reduced functional connectivity in the PCC during conscious sedation (21). Although we cannot rule out an effect of sedation, both the ASD and HC children where sedated with the same dose, so the sedative is unlikely to have contributed to the difference in DMN connectivity observed between these two groups.

In conclusion, we demonstrated both age-dependent and ASD-associated changes in the DMN. It is critical that normal developmental changes in the DMN are fully described in order to identify abnormal neurodevelopmental changes in the DMN associated with ASD in childhood. We propose that rsfMRIis a useful modality for assessing DMN changes in ASD and that these changes may be helpful biomarkers for ASD diagnosis.

CONFLICT OF INTERESTS

The author has no conflicts of interest to disclose concerning the paper.

ACKNOWLEDGEMENT

This study was supported by a Grant-in-Aid for Scientific Research (15K09926).

REFERENCES

1. Raichle ME : The restless brain. Brain Connect 1 : 3-12, 2011
2. Jones DT, Machulda MM, Vemuri P, McDade EM, Zeng G, Senjem ML, Gunter JL, Przybelski SA, Avula RT, Knopman DS, Boeve BF, Petersen RC, Jack CR Jr : Age-related changes in the default mode network are more advanced in Alzheimer disease. Neurology 77 : 1524-1531, 2011
3. Garrity A : Aberrant 'default mode' functional connectivity in schizophrenia. Am J Psychiatry 164 : 450, 2007
4. Courchesne E : Evidence of Brain Overgrowth in the First Year of Life in Autism. JAMA 290 : 337, 2003
5. Courchesne E, Mouton PR, Calhoun ME, Semendeferi K, Ahrens-Barbeau C, Melodie J. Hallet, Barnes CC, Pierce K : Neuron number and size in prefrontal cortex of children with autism. JAMA 306 : 2001-2010, 2011
6. Assaf M, Jagannathan K, Calhoun VD, Miller L, Stevens MC, Sahl R, O'Boyle JG, Schultz RT, Pearlson GD : Abnormal functional connectivity of default mode sub-networks in autism spectrum disorder patients. Neuroimage 53 : 247-256, 2010
7. Jung M, Kosaka H, Saito DN, Ishitobi M, Morita T, Inohara K, Asano M, Arai S, Munesue T, Tomoda A, Wada Y, Sadato N, Okazawa H, Iidaka T : Default mode network in young male adults with autism spectrum disorder : relationship with autism spectrum traits. Mol Autism 5 : 35, 2014
8. Monk C, Peltier S, Wiggins J, Weng S : Abnormalities of intrinsic functional connectivity in autism spectrum disorders. Neuroimage 47 : 764-772, 2009
9. Weng S, Wiggins JL, Peltier SJ, Carrasco M, Risi S, Lord C, Monk CS : Alterations of resting state functional connectivity in the default network in adolescents with autism spectrum disorders. Brain Res 1313 : 202-214, 2010
10. Cote CJ, Wilson S : Guidelines for monitoring and management of pediatric patients during and after sedation for diagnostic and therapeutic procedures : an update. Pediatrics 118 : 2587-2602, 2006
11. Onoda K, Ishihara M, Yamaguchi S : Decreased functional connectivity by aging is associated with cognitive decline. J Cogn Neurosci 24 : 2186-2198, 2012
12. Vidal-Pineiro, D. Valls-Pedret C, Fernández-Cabello S, Arenaza-Urquijo EM, Sala-Llonch R, Solana E, Bargalló N, Junqué C, Ros E, Bartrés-Faz D : Decreased Default Mode Network connectivity correlates with age-associated structural and cognitive changes. Front Aging Neurosci 6 : 1-17, 2014
13. Jie Song, Rasmus M. Birn, Mélanie Boly, Timothy B. Meier, Veena A. Nair, Mary E. Meyerand, Vivek Prabhakaran : Age-Related Reorganizational Changes in Modularity and Functional Connectivity of Human Brain Networks. *Brain Connect* 4 : 662-676, 2014
14. Zhang H-Y, Chen WX, Jiao Y, Xu Y, Zhang XR, Wu JT : Selective vulnerability related to aging in large-scale resting brain networks. PLoS One 9 : e108807, 2014
15. Buckner RL, Andrews-Hanna JR, Schacter DL : The brain's default network : anatomy, function, and relevance to disease. Ann NYAcad Sci USA 1124 : 1-38, 2008
16. Anticevic A, Cole MW, Murray JD, Corlett PR, Wang XJ, Krystal JH : The role of default network deactivation in cognition and disease. Trends Cogn Sci 16 : 584-92, 2012
17. Lou HC, Luber B, Crupain M, Keenan JP, Nowak M, Kjaer TW, Sackeim HA, Lisanby SH : Parietal cortex and representation of the mental self. Proc Natl Acad Sci USA 101 : 6827-6832, 2004
18. Cavanna AE : The precuneus : a review of its functional anatomy and behavioural correlates. Brain 129 : 564-583, 2006
19. Sestieri C, Corbetta M, Romani GL, Shulman GL : Episodic memory retrieval, parietal cortex, and the default mode network : functional and topographic analyses. J Neurosci 31 : 4407-4420, 2011
20. Stamatakis EA, Adapa RM, Absalom AR, Menon DK : Changes in resting neural connectivity during propofol sedation. PLoS One 5 : e14224, 2010
21. Greicius MD, Kiviniemi V, Tervonen O, Vainionpää V, Alahuhta S, Reiss AL, Menon V : Persistent default-mode network connectivity during light sedation. Hum. Brain Mapp 29 : 839-847, 2008