Preserved high centrality hubs but efficient network reorganization during
eyes-open state compared to eyes-closed resting state: An MEG study

Running title: EC and EO resting state functional networks

Seung-Hyun Jin\textsuperscript{1,2}, Woorim Jeong\textsuperscript{1,3}, Dong-Soo Lee\textsuperscript{4}, Beom Seok Jeon\textsuperscript{5},
Chun Kee Chung\textsuperscript{1,2,3,6,7*}

\textsuperscript{1}Department of Neurosurgery, Seoul National University Hospital, Seoul, Korea
\textsuperscript{2}Neuroscience Research Institute,
Seoul National University Medical Research Center, Seoul, Korea
\textsuperscript{3}Interdisciplinary Program in Neuroscience,
Seoul National University College of Natural Sciences, Seoul, Korea
\textsuperscript{4}Department of Nuclear Medicine, Seoul National University College of Medicine,
Seoul, Korea
\textsuperscript{5}Department of Neurology, Seoul National University College of Medicine, Seoul, Korea
\textsuperscript{6}Department of Brain and Cognitive Sciences,
Seoul National University College of Natural Sciences, Seoul, Korea
\textsuperscript{7}Department of Neurosurgery, Seoul National University College of Medicine, Seoul, Korea

*Correspondence: Chun Kee Chung, M.D., Ph.D, Department of Neurosurgery, Seoul National
University Hospital, Seoul National University College of Medicine, 101 Daehak-ro,
Jongno-gu, Seoul 110-744, Republic of Korea
Tel.: + 82-2-2072-2352
Fax: +82-2-2072-0806
E-mail: chunge@snu.ac.kr
A question to be addressed in the present study is how different are the eyes-closed (EC) and eyes-open (EO) resting states across frequency bands in terms of efficiency and centrality of the brain functional network. We investigated both the global and nodal efficiency and betweenness centrality in the EC and EO resting states from 39 volunteers. Mutual information was used to obtain the functional connectivity for each of the 4 frequency bands (theta, alpha, beta, and gamma). We showed that the cortical hubs with high betweenness centrality were maintained in the EC and EO resting states. We further showed that these hubs were associated with more than 3 frequency bands, suggesting that these hubs play an important role in the brain functional network at multiple temporal scales in the resting states. Enhanced global efficiency values were found in the theta and alpha bands in the EO state compared to those in the EC state. Moreover, it turned out that in the EO state, the functional network was reorganized to enhance nodal efficiency at the nodes related to both the default mode and the dorsal attention networks and sensory-related resting state networks. This result suggests that in the EO state, the brain functional network was efficiently reorganized facilitating the adaptation of the brain network to the change in state, which could help in understanding brain disorders that have a disturbance in communication with external environments by using the adaptation ability of brain functional networks.
Keywords: Functional cortical hubs; Eyes-closed and -open resting states; MEG; Efficiency;

Betweenness centrality
INTRODUCTION

Interest in resting state brain networks has increased explosively, which comes from a fundamental curiosity in the intrinsic characteristics of brain networks underpinning brain functions. Studies on resting state functional networks from an electrophysiological perspective can take advantage of high temporal resolution. Based on the results of those studies, we can get one step closer to the mechanisms underlying the resting state functional networks by showing intrinsic brain networks in terms of how brain oscillatory activities interact among different brain regions. In fact, there have been many magnetoencephalography (MEG) studies that have reported functional networks in the resting state in both the sensor level (Bassett et al. 2009a; Jin et al. 2011b; Kitzbichler et al. 2011; Stam et al. 2009) and source level (Brookes et al. 2011a; Brookes et al. 2011b; de Pasquale et al. 2010; de Pasquale et al. 2012; Hipp et al. 2012; Jin et al. 2013).

Regarding cortical hubs identified by betweenness centrality (BC) in a functional magnetic resonance imaging (fMRI) study, the locations of these cortical hubs were mostly consistent across different task states, such as an eyes-open with a fixation resting state as well as an active word-classification task, suggesting that the hubs are stable features of the functional networks (Buckner et al. 2009). Similarly, Fransson and Marrelec (2008) reported that the strongest and most consistent pattern of correlations was found for the posterior cingulate cortex node, and
many of the correlations persist in the transition from rest to task. This functionally linked default mode network (DMN) was maintained during a sensory task with low cognitive demand but was attenuated during a working memory task.

Here, a question is raised whether the cortical hubs in the resting states would be stable in the same manner as the fMRI showed for various frequency bands using an electrophysiological approach. Both the eyes-closed (EC) and eyes-open (EO) states can be regarded as resting states. In our previous study, we confirmed that there were cortical core regions across participants and frequency bands in a large population of 88 participants even in an unconstrained state free from any passive or active tasks (EC state), and showed their functional importance as connector hubs (Jin et al. 2013). Because the EO state is a relatively focused state compared to the EC state, it is expected that the functional cortical hubs in the EO resting state can be distinguished. According to a recent interesting study on how to identify resting state networks using electroencephalography (EEG), the EO state showed salient functional connections which can be differentiated from the EC state by directly comparing the connection strength between the two conditions (Chen et al. 2013). However, the functional hubs were not taken into account in the work by Chen et al. (2013). In the present study, considering that the EO state is regarded as a resting state, we hypothesized that the main functional cortical hubs in terms of centrality would be maintained during the EC and EO resting states as previously showed in fMRI studies.
Among various centrality measures, we used only BC in the current study. On the other hand, considering that the EO state is different from the EC resting state in terms of the inputs from an external environment, the functional reorganization to adapt the brain network to the change in state will be inevitable, which can be characterized by the frequency-specific and node-wise network features. Taking into account the economical properties of the brain networks (Achard and Bullmore 2007; Jin et al. 2012; Kitzbichler et al. 2011), we hypothesized that adaptive and efficient node-wise functional reorganization will occur depending on the resting states. If so, this will support the idea of the rapid and adaptive reorganization of functional networks when faced with changing environmental demands (Bassett et al. 2006). Furthermore, this would provide a background for understanding brain disorders such as schizophrenia, attention deficit hyperactivity disorder, dyslexia, which have a disturbance in communication with the environment, based on the adaptation ability of brain functional networks by investigating such a simple baseline transition from the EC to EO resting states.

To this end, we investigated both the global and nodal efficiency and BC during the EC and EO resting states in 39 volunteers. Mutual information (MI), which quantifies the shared information between two time series based on information theory, was calculated to obtain the functional connectivity association matrix for each of the 4 frequency bands: theta (4-7 Hz), alpha (8-12 Hz), beta (13-30 Hz), and gamma (31-45 Hz).
MATERIALS AND METHODS

Participants

A total of 39 right-handed healthy subjects (mean ± SD: 25.95 ± 3.85 years, 18 males) voluntarily participated. None of the participants had neurological problems. Handedness was tested using the Edinburgh Handedness Inventory (Oldfield 1971). Written informed consent was given by all participants. The study protocol was approved by the local Institutional Review Board at Seoul National University Hospital (IRB no. H-0607-029-178).

MEG signal acquisition and preprocessing

MEG signals were recorded at the EC and EO resting states for 2 min each as shown in Fig. 1A. The participants were sitting or lying in a supine position, and they were instructed to stay relaxed and to fixate their eyes on a “cross” mark in the EO state. During recording, participants were asked to not think about anything. Data were collected inside a magnetically shielded room using a 306-channel-head MEG system (VectorView, Elekta Neuromag Oy, Helsinki, Finland). Signals were analog filtered between 0.1 and 200 Hz, and digitized at a sampling frequency of 600.615 Hz. In addition to MEG, electrooculograms (EOG) and electrocardiograms (ECG) were simultaneously recorded. Head movements were tracked using four head position indicator coils.
placed on the scalp. Three anatomical landmarks corresponding to the nasion and two preauricular points were digitized by a three dimensional digitizer (FASTRAK™, Polhemus) to identify the coil locations, which were used to coregister the MRI, MEG and head coordinates. The temporal signal space separation (tSSS) method (Taulu and Hari, 2009) implemented in the Maxfilter Software (Elekta Neuromag Oy, Helsinki, Finland) was used to remove magnetoencephalographic artifacts, which is a required and effective artifact removal preprocessing step for data recorded with the Elekta-MEG system (Hillebrand et al. 2012). After applying the tSSS method, epoching was done with Graph software (Elekta Neuromag Oy, Helsinki, Finland). Based on the results in our previous studies (Jin et al. 2013; Jin et al. 2011b), we selected five 10-sec epochs out of the continuous signal recordings. And then, to minimize eye and cardiac-related artifacts, independent component analysis (ICA) was applied. The ‘Infomax’ algorithm was applied which is implemented in the Fieldtrip software (Oostenveld et al. 2011). (Fig.1B.)

[Figure1]

MRI

Individual T1-weighted magnetic resonance (MR) images were acquired with either a GE 1.5 or 3 T MR unit (GE Horizon Echospeed) or a Siemens 1.5 T scanner (Siemens Avento system,
Nodes selection and reconstruction of source waveforms from the MEG signals

In the present study, we used the pre-defined 72 nodes (36 nodes in each hemisphere) listed in Table 1. These nodes were selected from the AAL-atlas. We tried to include many of the nodes covered by the MEG recording. The AAL-atlas is used for anatomical parcellation of the brain (Tzourio-Mazoyer et al. 2002), and has been used in studies that defined brain network nodes using fMRI (He et al. 2009; Liao et al. 2010; Supekar et al. 2008; Wang et al. 2009), and MEG (Jin et al. 2013). Reconstruction of source waveforms at 72 nodes via a simultaneous 72 dipole time-series extraction procedure was performed with BESA®2000 software (MEGIS Software, Gräfelfing, Germany) (Fig.1C.). A 60 Hz notch filter and a 1–180 Hz band-pass filter were applied when extracting source waveforms in BESA. Source waveform extraction was performed first and was done separately for each condition. Subsequently, the extracted source waveforms were filtered to each frequency band for further analysis. Before reconstruction, the individual MRIs were standardized with Brain Voyager (ver. 1.10).

[Table 1]
From the reconstructed source waveforms at the 72 nodes, the MI values of the bandpass filtered source waveforms were estimated between the 72 nodes for each of the 4 frequency bands: theta (4-7 Hz), alpha (8-12 Hz), beta (13-30 Hz), and gamma (31-45 Hz). Among various functional connectivity measures, we chose MI which quantifies the shared information between two time series based on information theory. MI has been used to evaluate brain connectivity in EEG (Alonso et al. 2010; Jeong et al. 2001; Jin et al. 2012; Na et al. 2002) and MEG (Chen et al. 2008b; Jin et al. 2013; Jin et al. 2011b). MI was calculated using the following equation:

\[
\text{MI} = \text{MI}_{X_1X_2} = \text{MI}_{X_2X_1} = \text{MI}(X_1(t), X_2(t)) = - \sum_{X_1(t), X_2(t)} p(X_1(t), X_2(t)) \log \frac{p(X_1(t), X_2(t))}{p(X_1(t))p(X_2(t))}
\]

where \( X_1(t) \) and \( X_2(t) \) indicate the two bandpass filtered source waveforms of the 72 nodes, and \( p(X_1(t), X_2(t)) \) is the joint probability density function (PDF) between \( X_1(t) \) and \( X_2(t) \). For PDF construction, 32 bins were adopted for 4096 data points like in our previous studies (Jin et al. 2013; Jin et al. 2011b). To compensate for the effect of finite data and quantization on the PDF, a corrective term was applied (Roulston 1999). MI matrices of each epoch were calculated with five 10-sec epochs at each frequency band. The 5 MI matrices were averaged, and normalized by the maximum value to construct the complex brain networks at each
frequency band (Fig. 1D). To obtain the average MI value per subject, the normalized MI was averaged across all possible nodes at each frequency band and denoted as MIglob. The MIglob differences between states were displayed in Fig. 2. Since binarization was not applied, weighted graphs were used for further graph-theoretic analyses. Because spurious correlations between MEG signals can occur due to the low spatial resolution of the source level MEG (Chavez et al. 2010; de Pasquale et al. 2012) and when multiple sources are estimated, the effect of spatial spread functions can also induce spurious correlations (Hauk et al. 2011), pairs of nodes closer than 40 mm (Garcia Dominguez et al. 2005) were excluded in the present study like in our previous work (Jin et al. 2013).

[Figure 1]

Network metrics to assess efficiency and betweenness centrality

With the previously calculated MI matrix (weighted symmetric matrix) of the bandpass filtered source waveforms, a graph-theoretic analysis was performed. $N$ is the set of all nodes (here, 72), and $n$ is the number of nodes in the graph. The links between 2 nodes $i$ and $j$ are associated with the connection weight $w_{ij}$. The weights were normalized by the maximum value of the MI matrix ($0 \leq w_{ij} \leq 1$ for all nodes). The shortest weighted path length of the path from node $i$ to node $j$, the so called $d^{w}_{i,j}$, was calculated as $\sum_{w_{ij}\in \mathcal{E}_{i\rightarrow j}} f(w_{ij})$, where $f$ is the
inverse of the weight to length and $g_{ij}$ is the shortest weighted path between the 2 nodes, $i$ and $j$ (Rubinov and Sporns 2010).

Efficiency as a network measure expresses the capacity of networks to facilitate information exchange (Latora and Marchiori 2001). Global efficiency ($E_{glob}$) is a measure of parallel information transfer in the whole network (Latora and Marchiori 2001). $E_{glob}$ is defined as an average of the nodal efficiency ($E_{nodal}$); thus, one $E_{glob}$ value was produced per subject at each frequency band. Nodal (regional) efficiency is inversely related to the minimum path length, and used to assess the efficiency at each node (Bassett et al. 2009b; Jin et al. 2012). It is a measure of the communication efficiency between a node $i$ and all the other nodes in the network (Wang et al. 2009), and derived by the following equation:

$$E_{nodal}(i) = \frac{1}{n-1} \sum_{j \in N, j \neq i} \frac{1}{d_{ij}}, \quad E_{glob} = \frac{1}{n} \sum_{i \in N} E_{nodal}(i)$$

BC, as a center of information integration, measures how often nodes occur on the shortest paths between other nodes (Buckner et al. 2009; Freeman 1978). It is defined as the following equation:

$$BC = \sum_{h, j \in N, h \neq j, h \neq i, j \neq i} \frac{g_{h(j)}(i)}{g_{hj}}$$

where $g_{hj}$ is the number of shortest paths between node $h$ and $j$ and $g_{h(j)}(i)$ is the number of shortest paths between node $h$ and $j$ passing through $i$. BC was normalized by the mean value.
of BCs in a network (He et al. 2008; Wang et al. 2010), and thus, we denoted it as normBC. The efficiency and BC were obtained with functions from the Brain Connectivity Toolbox (http://www.brain-connectivity-toolbox.net/).

After the calculations, the Enodal and normBC data were standardized by conversion to z scores at the single subject level as follows:

\[ z_i = \frac{\text{metric}(i) - \text{mean(metric)}}{\text{SD(metric)}} \]

where metric indicates either Enodal or normBC; metric(i) denotes the nodal network metric at node i; mean (metric) is the mean nodal network metric across all nodes in the network; and SD (metric) is the standard deviation of the network. The conversion to z scores does cause the values in each subject map to be comparably scaled (Buckner et al. 2009). It should be noted that MIglob and Eglob are measures of global network features, whereas Enodal and normBC are measures of nodal network features.

**Statistical analysis and visualization**

To extract functional cortical hubs based upon the Enodal and normBC for each subject and in each frequency band, we applied z-score conversion as mentioned above and selected those nodes with z scores larger than 2 SD. For those hubs, ranked distributions of the hubs based on the aggregated ranking percentage for the 39 participants were constructed. To construct a
ranked distribution for each frequency band, the extracted hubs for each subject were summed
across the 39 participants, and then transformed into percent ranks at each node. The ranked
distribution of the aggregated ranking percent included only nonzero percent nodes. We
compared MIglob and Eglob between the EC and EO conditions across the assessed frequency
bands by using a two-tailed paired t-test; p values <0.01 with Bonferroni correction were
considered significant. A two-tailed paired t-test was also performed to assess differences in
Enodal and normBC at each node. In that comparison, a false discovery rate (FDR) correction
for multiple comparisons was applied to control for false-positive errors; statistically
significant results with p values <0.01 were accepted as significant. All statistical analysis was
performed using Statistics toolbox in MATLAB (Mathworks, MA). For brain network and
node visualization, BrainNet Viewer (Xia et al. 2013) was used.
RESULTS

Figure 2 shows MIglob in the EC and EO states at each frequency band. There were significant differences between the two resting states in the theta (t38 = -4.957, p < 0.001) and alpha (t38 = -4.112, p < 0.001) bands, indicating a stronger connection strength in the EO state than in the EC state. However, in the beta and gamma bands, there were no significant differences between the two resting states.

[Figure 2]

Figure 3 shows Eglob values in the EC and EO states for each frequency band. There were significant differences between the two resting states in the theta (t38 = -4.877, p < 0.001) and alpha (t38 = -3.994, p < 0.001) bands, indicating the presence of enhanced Eglob values in the EO state compared to those in the EC state.

[Figure 3]

Ranked distributions of the hubs and their topological maps, as characterized by Enodal and normBC, are presented in Figs. 4 and 5. By visual inspection, the functional cortical hubs are very similar in the two resting states. The top hubs in the Enodal at each frequency band are identical except for the gamma band. The left dorsolateral superior frontal cortex (F1_L) was the top hub in the theta and alpha bands, and the left posterior cingulate cortex (PCIN_L) was the top hub in the beta band. In the gamma band, the left medial cingulate cortex (MCIN_L) was the top...
hub in the EC state, whereas the PCIN_L was the top hub in the EO state. Interestingly, in the normBC (Fig. 5), the top hubs (F1_L in the theta (Fig. 5A and 5E), alpha (Fig. 5B and 5F), and beta (Fig. 5C and 5G) bands, as well as PCIN_L in the gamma band (Fig. 5D and 5H)) were identical in both EC and EO states at each frequency band.

Figure 6 shows the ranked distribution of all the nodes irrespective of the frequency bands. From this figure, the main hubs identified with the normBC were identical in the two resting states, i.e., F1_L, MCIN_L, PCIN_L, and T2P_L corresponding hubs were identified in more than 3 of the 4 frequency bands (magenta and green colored nodes). On the other hand, there were some variations between the two resting states in terms of the Enodal, although more than 3 frequency bands were associated with the node locations at F1_L and PCIN_L. This means that in terms of the normBC, the EC and EO states can be regarded as similar functional networks having the same main hubs across multiple frequency bands while these two states can be differentiated in terms of the Enodal. Because the top nodes seen in Figs. 4 and 5 were located in the left hemisphere, the most frequently appearing and consistent hubs seemed lateralized. However, looking at each ranked distribution of the Enodal and normBC (Figs. 4 and 5), the right side hubs were also shown but with a low appearance frequency.
Statistical analysis was performed to assess differences in Enodal and normBC values at each node. There were no significantly different nodes between the two resting states in the normBC. However, with respect to the Enodal, we found several significantly different nodes in the theta, alpha and beta bands. In the theta band, F1_R ($t_{38} = -4.192$), F2_R ($t_{38} = -4.072$), PRE_R ($t_{38} = -4.119$), POST_R ($t_{38} = -4.904$), T2_L ($t_{38} = -3.994$) and HES_R ($t_{38} = -4.352$) had larger Enodal values in the EO state than those in the EC state, whereas F1_L ($t_{38} = 4.308$), T1P_L ($t_{38} = 4.568$) and HIP_L ($t_{38} = -4.914$) had larger Enodal values in the EC state than those in the EO state. In the alpha band, Enodal at F3T_R ($t_{38} = -4.535$), SMA_L ($t_{38} = -4.516$), POST_R ($t_{38} = -4.524$) and O1_R ($t_{38} = -4.150$) were larger in the EO state than those in the EC state, whereas the Enodal at F3OP_L ($t_{38} = 3.772$), T1_R ($t_{38} = 4.103$), T3_L ($t_{38} = 5.019$) and HIP_L ($t_{38} = 4.130$) were larger in the EC state than those in the EO state. In the beta band, the Enodal at O1_R ($t_{38} = -4.302$) in the EO state was larger than that in the EC state.

Interestingly, although there were hubs corresponding to the DMN (F1_R, F3T_R) and dorsal attention network (DAN; F2_R), hubs with high Enodal in the EO state compared to the EC state were more associated with the sensory-related resting state networks (RSNs), such as somato-sensory network (SSN; SMA, PRE, POST), auditory network (AN; HES, T2) and visual network (VN; O1). In contrast, hubs with an enhanced Enodal in the EC state than in the EO state were more associated with the DMN (F1_R, F3T_R) and dorsal attention network (DAN; F2_R).
state were found mainly in the DMN (F1, T1P, F3OP) and DAN (HIP, T3). Figure 7 shows the
significantly different Enodal hubs between the EC and EO states. The statistical results are
summarized in Table 2.

[Figure 7]  
[Table 2]
Herein, we demonstrated that the cortical hubs with high centrality were maintained in the EC and EO resting states, but in terms of the efficiency of the brain functional networks, these two resting states can be differentiated from each other. In particular, we found that the brain functional network is efficiently reorganized facilitating the adaptation of the brain network to a change in state. Interestingly, the functional cortical hubs (F1_L, PCIN_L, MCIN_L and T2P_L) with high BC are associated with more than 3 frequency bands, which means that these hubs play an important role in information transmission at multiple frequencies in the resting brain functional networks. Considering the notion that a system of brain oscillators allows brain operations to be performed simultaneously at multiple temporal and spatial scales (Buzsaki and Draguhn 2004; Buzsaki et al. 2004), our study might illuminate the strong advantage of the electrophysiological approach being able to capture complex brain dynamics in resting state research. In addition, enhanced Eglob values in the EO state compared to those in the EC state were detected in the theta and alpha bands. Moreover, it turned out that the functional network was reorganized to have enhanced Enodal at the nodes related to both DMN/DAN and sensory-related RSNs in the EO state. This suggests that the brain functional network is efficiently reorganized facilitating the adaption of the brain network to the change in state in the EO state. Based on our findings showing functional hubs that persisted with high BC but with
efficient reorganization, the study on functional cortical hubs may be applicable to clinical
populations for the purpose of differentiating abnormal brains from healthy brains by using the
adaptation ability of functional brain networks such as the simple baseline transition from the EC
to the EO resting state.

Consistent functional cortical hubs with high betweenness centrality

Consistent functional cortical hubs with high BC associated with more than 3 frequency bands
across the participants without any node-wise differences between the EC and EO states were
observed in the left medial and posterior cingulate cortex, the left dorsolateral superior frontal
cortex, and the left pole of the middle temporal cortex (Fig. 6.). Interestingly, these hubs were
also reported as the most consistent hubs in the EC state obtained from a large population in our
previous study (Jin et al. 2013). PCIN_L and F1_L may be associated with structural
connectivity overlapping with the region constituting the DMN (Fransson and Marrelec 2008;
Greicius et al. 2003). MCIN_L and T2P_L were interpreted as the presentation of internal
self-referential or self-awareness processing (Fransson and Marrelec 2008; Kim 2012; Northoff
and Bermpohl 2004; Northoff et al. 2006). It seems that the potential functional roles of these
hubs suggested in these previous studies are valid not only in the EC but also in the EO resting
state, and these functional hubs might be the resting state invariant electrophysiological cores at
multiple frequencies.

A region with high BC represents a pivotal node that frequently becomes a shortcut between nodes within the network by definition, and the hub nodes are regarded as the articulation points of the cortical architecture (Sporns 2012b). Thus, our result indicated that the maintained functional cortical hubs that we found play a decisive role as pivotal nodes acting as the functional cores of the electrophysiological self by maintaining the resting state functional networks at multiple frequency bands.

There is accumulating evidence in support of the consistency of resting state functional connectivity (Boly et al. 2008; Patriat et al. 2013; Van Dijk et al. 2010), for example, across subjects (Damoiseaux et al. 2006) and over time (Shehzad et al. 2009). In particular, resting state functional connectivity as measured by fMRI showed consistent cortical hubs identified with BC (Buckner et al. 2009). Evidence has also shown that the functionally linked default mode network with a seed node at the posterior cingulate cortex persisted during a sensory task with low cognitive demand (Fransson and Marrelec 2008). However, none of these fMRI studies have shown consistent functional cortical hubs across frequency bands. But, we revealed it by virtue of the high temporal resolution of MEG in the present study.

Global and nodal efficiency features in the resting states
Another method to assess network topology is to utilize a well-known ‘efficiency’ measure. With this, we can investigate how efficiently the network works in both global and regional aspects. In contrast to the high BC hubs that persisted without any node-wise differences, the EC and EO resting states presented distinct network features in terms of the efficiency of the brain functional network at several frequency bands.

With regard to Eglob, we detected an enhanced Eglob in the EO resting state in the theta and alpha frequency bands. This result indicates that an efficient functional network reconfiguration occurs at multiple temporal scales ranging from the theta to the alpha band. The involvements of various frequency bands looks reasonable considering the fact that these two resting states are associated with significant changes in spectral regional field powers ranging from the theta to the beta band (Chen et al. 2008a). Of course, there exists a gap between our study and the literature in terms of the different approaches (network vs. regional), levels (source vs. surface) and measures (spectral power vs. MI based on information theory). Thus, it would be difficult to compare the results of the various studies to our results directly. However, it is still a plausible explanation that the transition from the EC to the EO state is followed by changes in various brain oscillatory activities and these changes were represented as the enhanced Eglob in the present study using the MI measure based on the network approach at the source level. In addition, we observed a higher MIglob in the EO state than in the EC state in the theta and alpha
frequency bands. The resting state could be considered as a behavioral state characterized by quiet repose in the EC state, or by an EO state with or without visual fixation (Raichle 2010). Previous study showed that the network features in the EO condition are different from those in the EC condition. In fMRI studies, EO with or without visual fixation showed significantly higher functional connectivity (Van Dijk et al. 2010) and more robust estimation of functional connectivity (Yan et al. 2009) than that in the EC condition. Moreover, greater reproducibility in the EO state than in the EC state was reported in our previous MEG study (Jin et al. 2011a).

As for the Enodal, we found several node-wise differences between the two resting states in the theta, alpha and beta bands. There are several known RSNs reported in fMRI studies, such as DMN, DAN, SSN, AN, and VN (Ding et al. 2011; Raichle 2011; Tomasi and Volkow 2011). Significantly different hubs that we found in the present study can be classified according to the RSNs. There were significantly different hubs included in the DMN (F1_R, theta in EO>EC; F3T_R, alpha in EO>EC; F1_L, T1P_L, theta in EO<EO; T1_R, F3OP_L, alpha in EO<EC) and DAN (F2_R, theta in EO>EC; HIP_L, theta in EO<EC; F3_L, HIP_L, alpha EO<EC). This result revealed that the hubs with the enhanced Enodal in the EC state compared to the EO state were found mainly in the DMN and DAN; however, those in the EO state compared to the EC state were observed not only in the DMN and DAN but also in other RSNs. More specifically, the dominant hubs in the EO state were more associated with the sensory-related RSNs, such as
SSN (PRE_R, POST_R, theta; SMA_L, alpha), AN (T2_L, HES_R, theta) and VN (O1, alpha and beta). In the EC resting state, it seems more likely to focus on the internal mental states or internal rehearses, whereas in the EO resting state, RSNs related to the sensory processes are more recruited. This result suggests that the sensory-related RSNs in the EO state work more efficiently than those in the EC state due to the involvement of external environmental inputs although they were relatively minimized during the experiment.

Taken together, the EO resting state showed a functional network with globally enhanced efficiency in the theta to the alpha bands compared to the EC state. Looking more closely, the brain functional network is reorganized in such a way of having enhanced Enodal in both DMN/DAN and sensory-related RSNs in the EO state compared to the EC state.

In summary, from our results, we verified our initial hypothesis that the main functional cortical hubs in terms of centrality are maintained during the EC and EO resting states based on the fMRI literature; however, functional reorganization to adapt the brain network to changes in state takes place through efficient node-wise reorganization.

**Methodological considerations**

Although resting state functional network analyses have been applied in many studies and there has been reported a pipeline for MEG resting state networks (Mantini et al. 2011), it is still true
that there is no a golden-standard to investigate resting state electrophysiology. For instance,
several potential factors such as the node selection method (seed-based vs. whole brain
segmentation) and functional connectivity estimation (power envelop correlation vs. MI) can
influence the results. Different methods have yielded different results (Brookes et al. 2011a),
because each method represents distinct aspects of the brain network. Despite that, there are
similarities or dissimilarities in the hub locations identified in our study and in previous studies
(Brookes et al. 2011b; de Pasquale et al. 2010; de Pasquale et al. 2012); therefore,
methodological factors should be taken into considered.

However, because we conducted the experiment for resting state changes with the same
participants, our results might be immune from this issue at least when interpreting results
derived from within subjects.

Node selection is a matter of debate. We used 72 nodes, 36 for each hemisphere. No one has a
clear answer about how many nodes will most accurately represent the brain functional network.
One might guess that finer segmentation may be beneficial to find a more localized focus.
However, increasing the node number is associated with an increase in the well-known field
spread effect of MEG. It means that there is a trade-off between finer segmentation and the
field-spread effect. In this regard, we supposed that the number of nodes used in this study was
appropriate to represent each brain region while curtailing the field spread effect. MI
connectivity between nodes located less than 40 mm apart was discarded as a second means of ensuring minimal field spread effects (Chavez et al. 2010; de Pasquale et al. 2012). The limited ability to cover the deep structures of the brain with MEG can be another issue. Resting state is ultimately related to the human conscious state in which deep structures such as the brainstem may have a key functional role. In such a case, MEG modality reveals its spatial limitation. PET can capture brainstem activity, but it measures metabolic changes on a limited temporal scale. Therefore, despite a lack of coverage of deep structures, MEG is still an attractive method to investigate the spectral characteristics of resting state functional networks like in our study.

Implications for future studies

The examination of functional networks is as important as examining the human connectome from a neuronal or structural perspective and will shed light on complex brain dynamics (Jin et al. 2013). Here, we showed functional hubs that persisted with high centrality but with efficient reorganization in the different resting states. The study on functional cortical hubs may be applicable to clinical populations to differentiate abnormal brains from healthy brains by using the adaptation ability of brain functional networks such as a simple baseline transition from the EC to the EO resting state. Considering that a major rationale for mapping the human
connectome is to learn about the role of connectional disturbances in brain dysfunction and
disease (Sporns 2012a), this potential application is important, and thus, it is challengeable future
study. In the present study, any cognitive tasks that loaded different levels of functional demands
were not utilized in the experiments. It would be interesting to see if functional cortical hubs are
maintained not only in the different resting states but also in the transition from rest to task on
multiple temporal scale frameworks.

CONCLUSION

Our results suggest the existence of functional cortical hubs in both the EC and EO in terms of
the centrality measure. We would like to emphasize that the cortical hubs with high centrality
were maintained in the EC and EO resting states; furthermore, these hubs were associated with
more than 3 frequency bands, which means that these hubs play an important role in information
transmission in the brain functional network at multiple temporal scales in the resting states.
Moreover, we showed that the brain functional network was efficiently reorganized facilitating
the adaptation of the brain network to the change in state in the EO state, which presents the
possibility of differentiating these two resting states in terms of the efficiency of the brain
functional network.
Acknowledgements

This research was supported by the National Research Foundation of Korea (NRF) grant (Grant no. 2010-000028631) funded by the Korea government (MSIP), and a grant of the Korean Health Technology R&D Project, Ministry of Health & Welfare, Republic of Korea (Grant no. HI11C1360).

Disclosures

No conflicts of interest, financial or otherwise, are declared by the authors.
REFERENCES


Jin SH, Jeong W, Seol J, Kwon J, and Chung CK. Functional Cortical Hubs in the Eyes-Closed


Raichle ME. The restless brain. Brain Connect 1: 3-12, 2011.


Roulston MS. Estimating the errors on measured entropy and mutual information. Physica D 125:


**Figure Legends**

**Figure 1. Schematic diagram of data acquisition and analysis procedures.**

MEG signals were recorded both in the eyes-closed and eyes-open with fixation states (A). As preprocessing steps, tSSS, epoching (5 of 10 sec epochs, ep1~ep5), and ICA were applied (B). Independent components (ICs) related to eye movement and cardiac artifacts were removed. Source waveforms were extracted with BESA at the pre-defined 72 nodes (C). MI matrices of the bandpass filtered source waveform were estimated and averaged across epochs at each frequency band per subject (D). Finally, network analysis was performed (E).

**Figure 2. Global mutual information (MIglob) of eyes-closed (EC) and eyes-open (EO) resting states.** Connection strength is stronger in the EO state than in the EC state in the theta and alpha frequency bands (significance level: p < 0.01, with Bonferroni correction). However, in the beta and gamma bands, there was no significant difference between the two resting states.

**Figure 3. Eyes-closed (EC) and eyes-open (EO) resting state Eglob values.** There were significant differences between resting states in Eglob values in the theta and alpha frequency bands, indicating that Eglob are enhanced in the EO rather than in the EC state (significance...
level: P < 0.01, with Bonferroni correction). No significant change in Eglob was observed between the two resting states in the beta and gamma bands.

**Figure 4. Nodes with high Enodal.** Nodes based on the aggregated ranking percent of each node across 39 participants and their topological maps (L, left side; R, right side) projected into a cortical surface at the theta (A, EC; E, EO), alpha (B, EC; F, EO), beta (C, EC; G, EO), and gamma (D, EC; H, EO) bands obtained from Enodal estimation. The ranked distribution of the aggregated ranking percent included only nonzero percent nodes. Abbreviated notations for each node can be found in Table 1, and ‘_L’ and ‘_R’ denote the left and right hemispheres, respectively, at each node. The horizontal axes in each subgraph indicate the percentage (%). Blue dots denote nodes with nonzero ranking percent, and grey dots denote nodes with zero ranking percent.

**Figure 5. Nodes with high centrality (normBC).** Nodes based on the aggregated ranking percent of each node across 39 participants and their topological maps (L, left side; R, right side) projected into a cortical surface at the theta (A, EC; E, EO), alpha (B, EC; F, EO), beta (C, EC; G, EO), and gamma (D, EC; H, EO) bands obtained from the normBC estimation. The ranked distribution of the aggregated ranking percent included only nonzero percent nodes.
Abbreviated notations of each node can be found in Table 1, and ‘_L’ and ‘_R’ denote the left and right hemispheres, respectively, at each node. The horizontal axes in each subgraph indicate the percentage (%). Blue dots denote nodes with nonzero ranking percent, and grey dots denote nodes with zero ranking percent.

**Figure 6. Ranked distribution of the Enodal and normBC.** Shown are the top 20 nodes based on the aggregated ranking percent of each node across 39 participants and their topological maps projected into a cortical surface derived from the Enodal (A, EC; C, EO) and normBC (B, EC; D, EO) measures irrespective of the frequency bands. Node color indicates the number of frequency bands identified at the top 20 nodes at the same node location (Magenta, all 4 frequency bands; Green, any 3 of 4 frequency bands; Blue, any 2 of 4 frequency bands; Yellow, any 1 of 4 frequency bands). Node size corresponds to the largest aggregated ranking percent of the node location. For instance, because F1_L in the Enodal was found as a hub in the theta, alpha, beta and gamma bands, it was presented as a magenta colored node with a value of ‘F1_L: theta’. The horizontal axes in each subgraph indicate the percentage (%).

**Figure 7. Nodes showing significant differences in the Enodal between the EC and EO**
states. Each map indicates significantly different nodes between the two states from the left, top and right views. Blue color denotes a node which has a larger Enodal in the EO state than in the EC state, while the dark gray color indicates a node which has a larger Enodal in the EC state than in the EO state. Grey dots denote regions with no difference between the two resting states.
A. Recording

B. Artifact removal 1: tSSS
  ↓ Epoching
  ↓ Artifact removal 2: ICA

C. Source waveform extraction (BESA)

D. MI estimation

E. Network analysis
  Theta  Alpha  Beta  Gamma
<table>
<thead>
<tr>
<th>Anatomical description</th>
<th>Label</th>
<th>MNI coordinates (Left)</th>
<th>BA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Central</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Precentral gyrus</td>
<td>PRE</td>
<td>-35 -24 62</td>
<td>4</td>
</tr>
<tr>
<td>Postcentral gyrus</td>
<td>POST</td>
<td>-36 -36 61</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td>Superior frontal gyrus, dorsolateral</td>
<td>F1</td>
<td>-18 46 34</td>
</tr>
<tr>
<td></td>
<td>Middle frontal gyrus</td>
<td>F2</td>
<td>-34 45 26</td>
</tr>
<tr>
<td></td>
<td>Inferior frontal gyrus, opercular part</td>
<td>F3OP</td>
<td>-53 13 11</td>
</tr>
<tr>
<td></td>
<td>Inferior frontal gyrus, triangular part</td>
<td>F3T</td>
<td>-54 11 23</td>
</tr>
<tr>
<td></td>
<td>Superior frontal gyrus, medial</td>
<td>F1M</td>
<td>-5 46 32</td>
</tr>
<tr>
<td>Lateral Surface</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Medial surface</td>
<td>SMA</td>
<td>-10 6 61 6</td>
</tr>
<tr>
<td></td>
<td>Superior frontal gyrus, orbital part</td>
<td>F1O</td>
<td>-19 50 -14</td>
</tr>
<tr>
<td></td>
<td>Superior frontal gyrus, medial orbital</td>
<td>F1MO</td>
<td>-7 53 -9 11</td>
</tr>
<tr>
<td>Frontal</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Medial surface</td>
<td>PCL</td>
<td>-7 -28 69</td>
</tr>
<tr>
<td></td>
<td>Superior frontal gyrus, medial</td>
<td>F1M</td>
<td>-5 46 32</td>
</tr>
<tr>
<td></td>
<td>Superior frontal gyrus, orbital part</td>
<td>F3O</td>
<td>-39 31 -15</td>
</tr>
<tr>
<td></td>
<td>Superior frontal gyrus, orbital part</td>
<td>F2O</td>
<td>-31 52 -10</td>
</tr>
<tr>
<td></td>
<td>Gyrus rectus</td>
<td>GR</td>
<td>-6 36 -21</td>
</tr>
<tr>
<td></td>
<td>Middle frontal gyrus, orbital part</td>
<td>F3O</td>
<td>-39 31 -15</td>
</tr>
<tr>
<td></td>
<td>Superior temporal gyrus</td>
<td>T1</td>
<td>-59 -42 1</td>
</tr>
<tr>
<td></td>
<td>Heschl gyrus (transverse)</td>
<td>HES</td>
<td>-54 -32 12</td>
</tr>
<tr>
<td></td>
<td>Middle temporal gyrus</td>
<td>T2</td>
<td>-61 -44 -3</td>
</tr>
<tr>
<td></td>
<td>Inferior temporal gyrus</td>
<td>T3</td>
<td>-53 -26 -23</td>
</tr>
<tr>
<td></td>
<td>Superior parietal gyrus</td>
<td>P1</td>
<td>-26 -59 61</td>
</tr>
<tr>
<td></td>
<td>Inferior parietal, but supramarginal and</td>
<td>P2</td>
<td>-31 -60 43</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Angular gyrus</td>
<td>AG</td>
<td>-42 -68 37</td>
</tr>
<tr>
<td></td>
<td>Supramarginal gyrus</td>
<td>SMG</td>
<td>-46 -47 48</td>
</tr>
<tr>
<td></td>
<td>Precuneus</td>
<td>PQ</td>
<td>-6 -62 50</td>
</tr>
<tr>
<td></td>
<td>Superior occipital gyrus</td>
<td>O1</td>
<td>-34 -78 27</td>
</tr>
<tr>
<td></td>
<td>Middle occipital gyrus</td>
<td>O2</td>
<td>-35 -84 14</td>
</tr>
<tr>
<td></td>
<td>Inferior occipital gyrus</td>
<td>O3</td>
<td>-33 -83 -8</td>
</tr>
<tr>
<td></td>
<td>Cuneus</td>
<td>Q</td>
<td>-7 -87 25</td>
</tr>
<tr>
<td></td>
<td>Medial and Calcarine fissure and</td>
<td>V1</td>
<td>-10 -74 8</td>
</tr>
<tr>
<td></td>
<td>inferior surfaces</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Lingual gyrus</td>
<td>LING</td>
<td>-17 -62 -4</td>
</tr>
<tr>
<td></td>
<td>Fusiform gyrus</td>
<td>FUSI</td>
<td>-31 -49 -15</td>
</tr>
<tr>
<td></td>
<td>Temporal pole: superior temporal gyrus</td>
<td>T1P</td>
<td>-26 17 -31</td>
</tr>
<tr>
<td></td>
<td>Temporal pole: middle temporal gyrus</td>
<td>T2P</td>
<td>-33 18 -37</td>
</tr>
<tr>
<td></td>
<td>Anterior cingulate and paracingulate gyri</td>
<td>ACIN</td>
<td>-5 42 6</td>
</tr>
<tr>
<td></td>
<td>Median cingulate and paracingulate gyri</td>
<td>MCIN</td>
<td>-5 -9 39</td>
</tr>
<tr>
<td></td>
<td>Posterior cingulate gyrus</td>
<td>PCIN</td>
<td>-5 -44 23</td>
</tr>
<tr>
<td>Limbic</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Hippocampus</td>
<td>HIP</td>
<td>-32 -14 -17</td>
</tr>
</tbody>
</table>
Table 2. Nodes and their corresponding RSNs that showed a significant (p < 0.01, with FDR correction) Enodal change between the EO and EC states. Both EO > EC and EO < EC Enodal changes are shown.

<table>
<thead>
<tr>
<th>Frequency</th>
<th>EO &gt; EC</th>
<th></th>
<th></th>
<th>EO &lt; EC</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Band</td>
<td>Node</td>
<td>t</td>
<td>Corresponding RSNs</td>
<td>Node</td>
<td>t</td>
<td>Corresponding RSNs</td>
</tr>
<tr>
<td>Theta</td>
<td>F1_R</td>
<td>-4.192</td>
<td>DMN</td>
<td>F1_L</td>
<td>4.308</td>
<td>DMN</td>
</tr>
<tr>
<td></td>
<td>F2_R</td>
<td>-4.072</td>
<td>DAN</td>
<td>T1P_L</td>
<td>4.568</td>
<td></td>
</tr>
<tr>
<td></td>
<td>PRE_R</td>
<td>-4.119</td>
<td>SSN</td>
<td>HIP_L</td>
<td>4.914</td>
<td>DAN</td>
</tr>
<tr>
<td></td>
<td>POST_R</td>
<td>-4.904</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>T2_L</td>
<td>-3.994</td>
<td>AN</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>HES_R</td>
<td>-4.352</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Alpha</td>
<td>F3T_R</td>
<td>-4.535</td>
<td>DMN</td>
<td>F3OP_L</td>
<td>3.772</td>
<td>DMN</td>
</tr>
<tr>
<td></td>
<td>SMA_L</td>
<td>-4.516</td>
<td>SSN</td>
<td>T1_R</td>
<td>4.103</td>
<td></td>
</tr>
<tr>
<td></td>
<td>POST_R</td>
<td>-4.524</td>
<td>T3_L</td>
<td>5.019</td>
<td>DAN</td>
<td></td>
</tr>
<tr>
<td></td>
<td>O1_R</td>
<td>-4.150</td>
<td>VN</td>
<td>HIP_L</td>
<td>4.130</td>
<td></td>
</tr>
<tr>
<td>Beta</td>
<td>O1_R</td>
<td>-4.302</td>
<td>VN</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

AN, Auditory network; DAN, Dorsal Attention Network; DMN, Default Mode Network; RSN, Resting State Network; SSN, Somato-Sensory Network; VN, Visual Network;