SLEEP DURATION/SLEEP QUALITY

Variability in Cumulative Habitual Sleep Duration Predicts Waking Functional Connectivity

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Study Objectives: We examined whether interindividual differences in habitual sleep patterns, quantified as the cumulative habitual total sleep time (cTST) over a 2-w period, were reflected in waking measurements of intranetwork and internetwork functional connectivity (FC) between major nodes of three intrinsically connected networks (ICNs): default mode network (DMN), salience network (SN), and central executive network (CEN).

Methods: Resting state functional magnetic resonance imaging (fMRI) study using seed-based FC analysis combined with 14-d wrist actigraphy, sleep diaries, and subjective questionnaires (N = 33 healthy adults, mean age 34.3, standard deviation ± 11.6 y). Data were statistically analyzed using multiple linear regression. Fourteen consecutive days of wrist actigraphy in participant's home environment and fMRI scanning on day 14 at the Birmingham University Imaging Centre. Seed-based FC analysis on ICNs from resting-state fMRI data and multiple linear regression analysis performed for each ICN seed and target. cTST was used to predict FC (controlling for age).

Results: cTST was specific predictor of intranetwork FC when the mesial prefrontal cortex (MPFC) region of the DMN was used as a seed for FC, with a positive correlation between FC and cTST observed. No significant relationship between FC and cTST was seen for any pair of nodes not including the MPFC. Internetwork FC between the DMN (MPFC) and SN (right anterior insula) was also predicted by cTST, with a negative correlation observed between FC and cTST.

Conclusions: This study improves understanding of the relationship between intranetwork and internetwork functional connectivity of intrinsically connected networks (ICNs) in relation to habitual sleep quality and duration. The cumulative amount of sleep that participants achieved over a 14-d period was significantly predictive of intranetwork and inter-network functional connectivity of ICNs, an observation that may underlie the link between sleep status and cognitive performance.

Keywords: central executive network, DMN, salience network, functional connectivity, fMRI, habitual total sleep time, sleep, sleep quality.

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Significance

Even small amounts of sleep restriction over a prolonged period have measureable negative consequences on waking cognition, which is particularly important since self-imposed short sleep durations are increasingly common. However, the mechanism by which sleeping patterns affect brain function to result in the behavioural effects remains unclear. Distributed brain networks are crucial for cognitive function, but whether these networks are affected by differences in habitual sleep duration across individuals has not been examined. Using wrist actigraphy and waking fMRI, we identified specific brain networks whose connectivity is correlated with cumulative sleep time, providing a method of investigating the neurobiological underpinnings of individual differences in susceptibility to sleep deprivation, and an unbiased measure of sleep history.

INTRODUCTION

Sleep is crucial for maintaining normal cognitive performance[1-8] but the precise mechanisms by which the processes that occur during sleep affect waking function remain to be clarified. It is increasingly recognized that functional connectivity (FC) of intrinsically connected networks (ICNs) is crucial for the maintenance of proper function in healthy individuals[9-11] and that specific disruptions to intranetwork and inter-network FC are widespread in neurological and neuropsychiatric disorders.[12,13] Modification of the activity and FC of ICNs has also consistently been observed during the descent into sleep[14-18] and following sleep deprivation[19-24] with the main emphasis having been placed on the default mode network (DMN). The DMN is likely to be particularly important in understanding the link between sleep and waking brain function not only because of its general link with maintenance of consciousness[25] but also its importance in a range of cognitive domains, which are known to be affected by prolonged wakefulness, including memory,[26-28] attention,[29] and emotion processing.[26]

In parallel with these investigations of FC, studies utilizing chronic partial sleep deprivation, which more closely resembles everyday life situations than total sleep deprivation, have

reported dose-dependent deficits in cognitive performance.[2,4,5] The common finding is that the less sleep subjects obtain due to sleep restriction (e.g., subjects restricted to 3, 5, or 7 h of time in bed compared to control subjects, who spent 8 h in bed for up to 7 d) the more cognitive performance is impaired.[2,4,5] Given that ICNs underpin waking function and are affected by prolonged wakefulness,[19-21,24] one possibility is that sleep is needed to maintain the brain's intrinsic functional architecture, normalizing the FC of ICNs to sustain the high level of regionally appropriate FC that is necessary for waking function. This would suggest that shorter habitual sleep over a prolonged period could have a cumulative effect on FC, which may subsequently result in subtle deficits in higher cognition. However, to date there has been no investigation of whether habitual sleep patterns measured over a prolonged period relate to waking FC. This is important because even a small amount of sleep restriction over a prolonged period can have measureable negative consequences on waking behavioral performance[4] and self-imposed short sleep durations are becoming increasingly common and represent a considerable public health burden.[30-32] Understanding whether differences in habitual sleep patterns relate to FC thus has considerable practical implications. We examined this issue by comparing habitual cumulative total sleep time

(cTST), assessed over a 2-w period with wrist actigraphy and sleep diaries, with waking FC of three of the most important ICNs for higher level cognitive function (the DMN, the salience network (SN), and the central executive network (CEN).

The DMN encompasses the posterior cingulate and precuneus (PCC), mesial prefrontal (MPFC) and bilateral inferior parietal (IPC) cortices, with the mesial temporal structures (MTL) and the hippocampal regions also sometimes included, although less consistently.[33] Originally identified as a set of regions that are consistently deactivated when attention is directed externally,[34,35] its general importance has subsequently been underscored by its relationship with a wide range of cognitive tasks.[34–37] Further investigations have also revealed specific roles of the anterior and posterior portions of the DMN,[38–40] indicating that although it is certainly a coherent network the individual nodes can have differentiated functions, as well as a specific relationship to task-positive regions.[10,41]

A number of studies have investigated ICN FC during sleep,[15–17,42,43] and alterations have been noted during wakefulness, following full or partial sleep deprivation[24,44,45] and in relation to self-reported sleep duration on the night prior to a waking scan.[46] These studies indicate that integrity of the DMN is a sensitive marker of sleep status and prior sleep history.

Although the importance of DMN functional integrity for the maintenance of normal brain function is clear, it is only one of many ICNs ranging from those encompassing primary sensory regions (e.g., visual, auditory, somatomotor) to higher level networks such as the CEN and the SN. Given previous behavioral observations[8] it would be expected that, in addition to the DMN, the higher-level CEN and SN would be most affected by sleep, rather than the sensory networks.

The human brain switches from intrinsic thoughts and self-referential activity involving regulation by the DMN, to task positive cognitive activity involving regulation by the CEN.[47,48] This switching between networks is thought to be regulated by the right anterior insula (RAI) of the SN, which acts as a control hub between the DMN and CEN and regulates states of consciousness in response to salient events.[49] These three ICNs therefore act in concert to maintain a normal level of brain function.

In the current awake, resting-state functional magnetic resonance imaging (fMRI) study, we first aimed to investigate whether the strength of intranetwork FC of the DMN, SN, and CEN covaried with the cumulative effect of normal habitual sleep time. Second, because the SN is involved in the regulation of activity between the DMN and CEN, we also aimed to investigate how between-subject FC variability in internetwork connectivity of the SN, CEN, and the DMN was related to subjects' habitual sleep time. The motivation for examining these networks is that they are closely linked with the higher cognitive functions, which are mainly affected by sleep deprivation.[1–7] A better understanding of how sleep affects ICN FC may help to shed light on the link between sleep and the functions these networks support, in particular cognition and conscious behavior, as well as the neurobiological underpinnings of individual differences in susceptibility to sleep deprivation. Although the link between individual variability in behavioral performance and sleep history has been extensively studied,[50] an explicit understanding of susceptibility to sleep loss requires a detailed knowledge of individual differences in the resilience of the brain networks that are responsible for waking function. In addition, as a marker of sleep deprivation, FC of ICNs is particularly attractive because it is not under conscious control and may provide an unbiased measure of sleep history.

We had two hypotheses: (1) Longer habitual cumulative total sleep times will be reflected by increases in the intranetwork FC between the major nodes of the DMN, SN, and CEN measured during wakefulness. (2) Longer habitual cumulative total sleep times will be reflected by network specific increases and decreases in internetwork FC between the DMN, SN, and CEN.

METHODS AND MATERIALS

Subjects
Data were acquired from 37 healthy adults (right handed, 17 female, age 20–59 y, mean age (± standard deviation [SD]) = 35.0 ± 11.7 y) using a 3 Tesla Philips Achieva MRI scanner at Birmingham University Imaging Centre (BUIC), University of Birmingham. Participants had no history of neurophysiological, neuropsychological, or neurological illness. Written informed consent was obtained from all participants, and the study was approved by the University of Birmingham Research Ethics Committee. The data from four subjects were subsequently excluded (corrupted data for one subject, erratic sleep patterns for the second, illness around the time of scanning for the third and fourth), meaning that the final dataset that was analyzed consisted of 33 participants (right handed, 17 female, age 20–59 y, mean age (± SD) = 34.2 ± 11.6 y).

Sleep Patterns and Questionnaires
Subjects were asked to maintain their normal sleep patterns for the duration of the study. Habitual sleep patterns were assessed for a 14-d period using sleep diaries and wrist actigraphy (Actiwatch 2, Philips Respironics Ltd, Cambridge, UK). The Actiwatch measures the amplitude as part of the sampling process with the minimum and maximum measures being ± 128. These values are referred to as counts. The number of counts is proportional to the intensity of movement. The highest count value for each sampling period (which consists of 1/32 of a second) was taken for each 1-sec interval and the sum of the captured counts form the individual 1-sec intervals making up the 1-min epoch provided the total count score. The epoch was the period defined for logging captured activity data. Actigraphs were set at a medium sensitivity of 1-min epochs, and a total count score of 40 or more was used to signify that the subject was awake. Use of actigraphy in sleep disordered patients[51] has shown that medium or high sampling rate sensitivities provide data for total sleep time (TST) per night in close agreement with polysomnography (PSG). Subjects were asked to press a button on the Actiwatch when they settled for bed and again on awakening to start their day. These times were defined as a sleep opportunity, and were used to carry out the actigraph analysis using Philips Respironics Actiwatch2 software. Participants also completed the following questionnaires: Pittsburgh Sleep Quality Index (PSQI),[52] Epworth Sleepiness Scale (ESS),[53] Depression, Anxiety and Stress Scale-21 (DASS),[54] and Karolinska Sleepiness Scale (KSS).[55] These

questionnaires were administered immediately prior to or following the scanning session, with the exception of the KSS, which was administered verbally immediately upon exiting the scanner. Each of the questionnaires resulted in a single score per subject, whereas TST was determined from the actigraphy and defined as the sleep time for each sleep opportunity and compared with sleep diary data for consistency.[51,56] Habitual TST was calculated as cumulative TST (cTST, sum of TST over the entire 2-week period).

Image Acquisition and Preprocessing

Subjects underwent a single resting-state fMRI session in the early afternoon during which they were instructed to lie still in the scanner and relax with eyes open. All participants confirmed that they remained awake and alert through the scanning session. Each subject underwent one resting-state fMRI scan of 12 min duration, with the following parameters: repetition time (TR) = 2000 ms, echo time (TE) = 35 ms, flip angle = 80°, voxel size 3 × 3 × 4 mm, 32 slices giving whole brain coverage. A standard T1-weighted anatomical scan (1-mm isotropic voxels) was acquired to facilitate image co-registration.

Preprocessing of the fMRI data was performed using the FMRIB Software Library (FSL, http://www.fmrib.ox.ac.uk/fsl).[57] The following procedures were applied: motion correction using MCFLIRT[58] slice timing correction, spatial smoothing using a gaussian kernel (FWHM = 6 mm) and a high-pass filter cut off at 100 sec (f > 0.01 Hz).

Defining Regions of Interest

Regions of interest (ROI) representing the nodes of the DMN, CEN, and the SN were created from data from a separate cohort of 55 subjects from a previous study[59] (28 male, age 25 ± 4 y). This allowed an objective identification of the canonical DMN, CEN, and SN that was independent from the subjects investigated in the current study. These subjects underwent a 6-min waking resting state fMRI scan with identical imaging parameters, also at BUIC. Using FSL 4.1.8 data were motion corrected, spatially smoothed (5 mm), registered to Montreal Neurological Institute (MNI) standard space, temporally concatenated across subjects and decomposed into 20 spatially independent components with MELODIC.[60] This low dimensionality was used to facilitate identification of the ICNs in single components and to avoid individual ICNs being split into their constituent nodes, which would have made unambiguous detection more difficult. For each of the DMN, CEN and SN in turn a single independent component was identified by visual inspection based on spatial similarity to previous reports.[61] The group-level Z-statistical maps were then thresholded at Z = 4, and individual ROIs were defined for the following ICN nodes: DMN (PCC, MPFC, left and right IPC, left and right MTL; CEN (left and right DLPFC, left and right IPL); and SN (left and right AI and the ACC). The left and right hippocampal regions (HP) were identified independently from the FSL atlas. These group-space ROIs were then registered to individual subject's fMRI data. We focused on these ROIs as they have been consistently reported as constituting robust regions of the DMN[15,41] CEN[11,44,61] and the SN.[11,62] Figure 1 shows the spatial arrangement of these
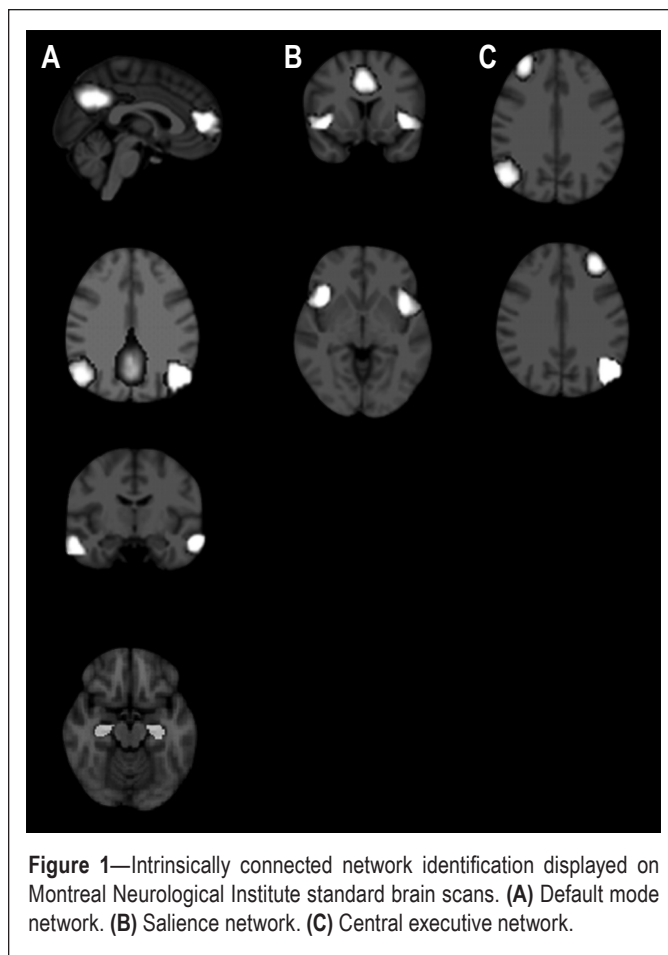

Figure 1—Intrinsically connected network identification displayed on Montreal Neurological Institute standard brain scans. (A) Default mode network. (B) Salience network. (C) Central executive network.

ROIs and Table 1 gives the center voxel MNI coordinates for these regions.

Measuring DMN, CEN, and SN FC

Following previous methodology[63] we used seed-based FC analysis performed according to standard methods[64] using in-house MATLAB code (Mathworks, Natick, MA, USA). Using FSL, the preprocessed functional data were further filtered (0.009 < f < 0.08Hz) and single voxel coordinates taken from each subject's individual functional scan to extract signal time courses from white matter and ventricles. The white matter and ventricular signals, the global brain signal, and the motion parameters were then removed from the voxelwise data using linear regression. ROIs were defined from nodes of group ICA and the ROI/node maps were transformed from MNI space to individual space using FSL. Individual subject ROIs were created as 3 × 3 × 3 voxel cubes centred on the single maximum Z-statistic voxel for each group ROI. The mean fMRI timeseries within each ROI was then correlated with the fMRI timeseries of all other brain voxels. This produced a whole-brain map of Pearson correlation coefficients, which allowed FC between regions of the DMN, SN, and CEN to be assessed and quantified. FC was defined by averaging the voxelwise correlation coefficients within each target ROI.

The 15 ROIs described previously were used in turn as the seed to measure the strength of FC to all other DMN, SN, and CEN ROIs for the intranetwork and inter-network analysis.

Table 1—Montreal Neurological Institute coordinates for the center voxel for each node/region of interest for the three networks investigated (default mode network, salience network, and central executive network).

Regions of Interest/Nodes for All Networks	MNI Coordinates (mm)		
	X (center)	Y (center)	Z (center)
Posterior cingulate cortex (PCC)	0	−52	34
Mesial prefrontal cortex (MPFC)	0	52	6
Left inferior parietal cortex (LIPC)	−52	−68	38
Right inferior parietal cortex (RIPC)	52	−68	38
Left mesial temporal lobe (LMTL)	−64	−10	−18
Right mesial temporal lobe (RMTL)	52	2	−30
Left hippocampus (LHC)	−28	−18	−14
Right hippocampus (RHC)	26	−18	−14
Right anterior insula (RAI)	36	24	2
Left anterior insula (LAI)	−40	16	2
Anterior cingulate cortex (ACC)	0	26	30
Left dorsal lateral prefrontal cortex (LDLPFC)	−42	34	24
Right dorsal lateral prefrontal cortex (RDLPFC)	42	44	24
Left inferior parietal lobule (LIPL)	−54	−64	24
Right inferior parietal lobule (RIPL)	56	−66	26

MNI, Montreal Neurological Institute.

Table 2—Summary data (n = 33).

Demographics	Mean	SD
Age	34.2	11.6
Questionnaires	**Mean**	**SD**
Epworth	3.94	0.79
Karolinska	1.16	0.41
Fatigue	12.36	0.98
PSQI	2.31	1.65
Depression	1.58	2.74
Anxiety	1.35	0.92
Stress	3.61	2.73
Actigraphy	**Mean**	**SD**
Mean TST (h)	7.65	1.85
cTST (h)	97.57	13.52

Demographics, questionnaires, mean total habitual sleep time (TST), cumulative habitual total sleep time (cTST) over 14 days.

Statistical Analysis

We investigated the relationship between individual sleep variables and both intranetwork and internetwork FC. Multiple linear regression analysis (SPSS Inc, Chicago, IL, USA) was performed for each DMN, SN, and CEN seed and target ROI, with cTST as the criterion variable and including FC and age as predictor variables. We controlled for false discovery rates (FDR) due to multiple measures by using the Benjamini-Hochbergh procedure[65] as used in previous studies.[49] The FDR P value adjustment method involved ranking the P values in order with the smallest P value being assigned rank 1, the second rank 2 and the largest rank N. Then each P value was multiplied by N and divided by its assigned rank to give the adjusted P. In order to restrict the FDR to 0.05 significance, all adjusted P values of less than or equal to 0.05 were regarded as significant.[65] All P values reported in the Results section are FDR corrected.

RESULTS

Table 2 summarizes the demographic, habitual sleep, and questionnaire data for the participants. All subjects were within normal limits and no evidence of depression, anxiety, excessive daytime sleepiness or fatigue was found (Table 2). Mean cTST was also within normal limits (7.65 ± 1.85 h).

Intranetwork FC Analysis

cTST and Intranetwork FC of the DMN

Table 3 shows the significant regression analysis results for the relationship between cTST and intranetwork DMN FC using the MPFC as seed ROI. This analysis indicated that cTST only predicted DMN FC when the MPFC was used as the seed ROI. No significant relationship between FC and cTST was seen for any pair of nodes not including the MPFC (see supplemental material for all non-significant results, and Figure S1 for average group FC between the MPFC and other nodes of the DMN).

For all pairs of ROIs that demonstrated significant (P < 0.05 FDR corrected) partial correlations to the seed region, the strength of FC between the DMN seed regions and the MPFC increased with cTST.

cTST and Intranetwork FC of the SN and CEN

cTST was not a significant predictor of intra-network FC for the SN or the CEN (P > 0.61; see supplemental material for nonsignificant results).

Internetwork FC Analysis

cTST and Internetwork FC of the DMN and SN

cTST was a significant predictor of the DMN-SN internetwork FC using the MPFC as the seed region. Specifically, FC between the MPFC and right anterior insula (RAI) was significantly predicted by cTST. A significant negative correlation was found (Table 4). cTST demonstrated a significant regression model when the RAI was used as the seed region for SN-DMN internetwork FC and an uncorrected P value of 0.034 was found, but this did not survive FDR correction (Table 5). Figures S2 and S3 (supplemental material) demonstrate the average group inter-network FC between the DMN and SN.

cTST and Internetwork FC of the CEN

cTST was not a significant predictor of either DMN-CEN or SN-CEN internetwork FC (see supplemental material).

DISCUSSION

This study examined the effect of habitual sleep patterns on the awake, resting-state FC of intrinsically connected

Table 3—Significant results of the regression analysis between habitual cumulative total sleep time (dependent variable) and default mode network (mesial prefrontal cortex seed) intranetwork connectivity.

Model	B	Std. error	β	t	P	Corrected P	Zero-order R
(Constant)	103.33	5.63		18.33	< 0.01		
LIPC	−41.83	35.80	−0.27	−1.16	0.25	0.40	0.11
LMTL	27.67	34.59	0.17	0.80	0.43	0.57	0.37
LHP	−51.20	22.10	−0.69	−2.31	0.02+	0.05*	0.30
PCC	61.44	24.64	0.73	2.49	0.02+	0.05*	0.46
RIPC	13.21	25.31	0.13	0.52	0.60	0.64	0.48
RMTL	82.22	28.68	0.56	2.86	< 0.01+	0.05*	0.54
RHP	19.44	41.33	0.11	0.47	0.64	0.64	0.26
Age	−0.34	0.18	−0.30	−1.81	0.08	0.16	−0.04

Model significance: $R^2 = 0.57$, $F = 4.25$, $P < 0.01$. +Significant uncorrected $P \leq 0.05$. *Significant false discovery rate corrected $P \leq 0.05$. LHP, left hippocampus; LIPC, left inferior parietal cortex; LMTL, left mesial temporal lobe; PCC, posterior cingulate cortex; RHP, right hippocampus; RIPC, right inferior parietal cortex; RMTL, right mesial temporal lobe.

Table 4—Significant results of the regression analysis between habitual cumulative total sleep time (dependent variable) and default mode network (mesial prefrontal cortex seed) internetwork connectivity with the salience network.

Model	B	Std. error	β	t	P	Corrected P	Zero-order R
(Constant)	100.69	7.38		13.63	< 0.01		
Age	0.09	0.17	0.08	0.55	0.58	0.58	−0.04
ACC	−21.55	26.36	−0.12	−0.81	0.42	0.56	−0.12
LAI	36.62	22.52	0.25	1.62	0.11	0.31	0.11
RAI	−57.77	15.54	−0.59	−3.71	< 0.01+	< 0.01*	−0.51

Model significance: $R^2 = 0.58$, $F = 3.76$, $P = 0.01$. +Significant uncorrected $P \leq 0.05$. *Significant false discovery rate corrected $P \leq 0.05$. ACC, anterior cingulate cortex; LAI, left anterior insula; RAI, right anteriror insula.

Table 5—Significant regression analysis model between habitual cumulative total sleep time (dependent variable) and salience network (RAI seed) internetwork connectivity with the default mode network. On false discovery rate correction of the P values in the model the RAI-mesial prefrontal cortex functional connectivity association with cumulative total sleep time were found to be nonsignificant.

Model	B	Std. error	β	t	P	Corrected P	Zero-order R
(Constant)	114.39	14.03		8.15	< 0.01		
Age	−0.49	0.35	−0.38	−1.38	0.18	0.46	−0.02
LIPC	14.03	32.57	0.07	0.43	0.67	0.67	−0.09
LMTL	27.62	42.04	0.18	0.65	0.51	0.58	0.50
LHP	−95.29	81.34	−0.25	−1.17	0.25	0.46	0.08
MPFC	−15.41	6.02	−0.61	−2.55	0.01+	0.17	−0.47
PCC	30.55	35.60	0.29	0.85	0.40	0.52	−0.25
RIPC	20.84	24.88	0.27	0.83	0.41	0.52	0.30
RMTL	51.27	44.16	0.31	1.16	0.25	0.46	0.30
RHP	122.52	78.16	0.36	1.56	0.13	0.46	0.09

Model significance: $R^2 = 0.49$, $F = 2.25$, $P = 0.05$. +Significant uncorrected $P \leq 0.05$. LHP, left hippocampus; LIPC, left inferior parietal cortex; LMTL, left mesial temporal lobe; MPFC, mesial prefrontal cortex; PCC, posterior cingulate cortex; RHP, right hippocampus; RIPC, right inferior parietal cortex; RMTL, right mesial temporal lobe.

networks. We focused on the DMN, SN, and CEN as these networks are most closely linked with the higher cognitive functions that have been shown to be most affected by sleep deprivation.[1–7] Our main finding was that the cumulative amount of sleep that participants achieved over the 14-day period preceding fMRI scanning was significantly predictive of intranetwork and inter-network FC of the DMN and SN, but not the CEN.

The study had two hypotheses. The first suggested that individual differences in sleep patterns, quantified as the cumulative total sleep time over 14 d (cTST), would be reflected in intranetwork FC strength between the major nodes of the

DMN, SN, and CEN measured during wakefulness. Multiple linear regression demonstrated that this was at least partially the case. In terms of the DMN, FC of the MPFC was significantly correlated with cTST. This result was specific to the MPFC, with only pairwise connections involving the MPFC as the seed showing a relationship between DMN FC and cTST (see Table 3). No association between SN or CEN intranetwork FC and sleep was found.

The specificity of the relationship between MPFC FC and sleep status is consistent with previous imaging and behavioural investigations. For example, it has been demonstrated that sleep deprivation causes reduced intra-DMN FC strength of the MPFC[20,46] to the PCC and posterior nodes of the DMN, whereas self-reported sleep duration on the night prior to scanning has also been linked with MPFC FC.[46] Behaviorally, a similar specificity has been observed, with sleep deprivation preferentially impairing cognitive performance on tasks involving the prefrontal cortex.[1,3] Although we did not test cognitive performance, it is reasonable to postulate that experimentally induced sleep deprivation leads to deficits in higher cognitions via its effect on intranetwork and internetwork FC of ICNs. The implication from our results is that these observations are generalizable to habitual sleep patterns in healthy individuals, and by quantifying FC of the MPFC we provide a mechanism by which habitual sleep status and cognition are linked. The fact that cTST is specifically linked to MPFC-DMN FC, but not FC within the SN or CEN, is a novel observation. The SN and CEN have been linked with salience and attentional processes, which might be expected to be related to cTST, but our results suggest the importance of internetwork FC in mediating the effects of cTST on these processes, as discussed in more detail in the next paragraphs.

Our second hypothesis was that internetwork connectivity of the DMN, SN, and CEN would be altered in relation to habitual sleep status. This issue has not been previously examined, and the basis of this hypothesis is that for optimal brain performance it is not only crucial that ICNs are internally connected, but they must be able to interact with each other in a consistent and coherent manner. This hypothesis was again partially confirmed, with connectivity between the DMN and SN dependent on cTST. Specifically, FC between the MPFC of the DMN and the RAI of the SN demonstrated a significant negative correlation with cTST (Table 4). It has been shown that when responding to an unexpected event in the environment the internally focused mode of operation supported by the DMN needs to be inhibited, and that this is achieved by an increase in RAI activity which in turn allows the brain to quickly switch to a controlled mode of operation which is tightly coupled to external events.[11,41,49] We have shown for the first time that a reduction in cTST is associated with an increase in the FC between RAI (SN) and the MPFC of the DMN (Table 4). It is possible that this represents an attempt to maintain the appropriate level of RAI activity needed to sustain alertness and ensure the effectiveness in network switching from intrinsic thoughts to external executive functioning. It is thought that the RAI is involved in the regulation of dynamic changes between the DMN and CEN,[11,48] networks known to have competitive interactions.[49] Our results suggest

that short habitual sleep durations disrupt right AI connectivity to the DMN and hence the ability to switch between internal and external modes, which may have an effect on widespread cognitive and behavioral domains. Future work will need to address this question with neuropsychological testing, but existing behavioral literature would support the association between working memory and attention and sleep status, albeit generally from the more extreme case of sleep deprivation or restriction.[66]

One factor that complicates the interpretation of this observation is that the DMN and SN are anticorrelated. A negative correlation with cTST therefore suggests that longer habitual sleep durations are related to more negative DMN-SN FC. It has been demonstrated that the use of global signal regression (GSR) as we have done negatively biases correlation measures.[67] At best this can manifest as a shifting of all correlations to lower values, including negative values. However, at worst it can result in a distortion of the underlying connectivity, which can fundamentally alter interregional correlations within a group, as recently demonstrated.[68,69] This makes it difficult to draw detailed conclusions regarding the relationship between negative inter-ICN FC (i.e., DMN-SN) and behavioral metrics, but will also affect positive values because of the overall shift of the distribution. While intended to reduce the impact of non-neuronal signal contributions, the global signal has at least a component that is of neuronal origin, and is correlated with both local field potentials in primates[70] and electroencephalographic vigilance measures in humans.[71] The global signal is affected by sleep[72] and sleep deprivation,[73] and therefore the inclusion of GSR in such studies, as well as in the context of habitual sleep durations as we have investigated, could be seen as a way of compensating for the overall shift in baseline that occurs with these changes in brain state. As discussed by Yeo et al.,[73] by employing GSR changes in FC relative to, rather than including, changes in overall brain signal are being assessed. Although the approach we have taken may mask the effect of habitual sleep time on the global brain signal, including the global signal, may mask the more specific regional changes that were our focus.[73,74] Especially given the neuronal contribution to the global signal and the potential information it contains about overall brain state, the effect of habitual sleep patterns on the global signal is a potentially interesting future question in its own right. For studies interested in regional changes in FC, as we have examined, it may be prudent to rely on more conservative alternatives to GSR such as CompCor,[75] as well as initiating more advanced investigations of the impact of GSR[76] and better assessments of the physiological nuisance variables that GSR is intended to mitigate.

Our results demonstrated some relatively strong lateralization effects in the relationship between FC and habitual sleep duration, particularly in relation to the MPFC to hippocampi and IPC (Table 3). It has been suggested that hippocampal FC is dependent on previous task history as well as the details of rest conditions,[77] with the laterality of hippocampal FC moderating connectivity patterns within and between networks. Similarly, the LIPC has previously been reported to show weak and fluctuating functional connectivity within the

DMN compared to that of the RIPC.[15,63] A magnetoencephalography study by Pasquale et al.[78] also found that the LIPC demonstrated a marked cross correlation with the dorsal attention network. In both of these cases there is therefore the suggestion that homologous left and right regions have distinct functions, including integration between networks. The significance of these lateralization effects in relation to variations in habitual sleep duration and their behavioral consequences remains to be clarified.

A recent study has suggested that a substantial proportion of waking resting-state fMRI scans may be confounded by participants entering early stages of sleep in even relatively short waking scans.[79] Although the effect of this observation on the field generally remains to be clarified, it could be argued that in our study participants with shorter habitual sleep times might be more likely to fall asleep during the scanning session. Our cohort consisted of healthy control subjects adhering to their normal sleep routine, who verbally indicated that they had not slept during the session, and our questionnaire data demonstrated no evidence of abnormal levels of daytime sleepiness (ESS score 4.93 ± 1.07, mean $\pm$ SD). In addition, their responses to the KSS indicated a good level of alertness immediately upon exiting the scanner (2.13 ± 0.21, mean $\pm$ SD, indicating a self-assessment of 'very alert', compared to a value of 6 indicating 'some level of sleepiness'). Although subjective ratings cannot be taken as completely reliable, the available evidence is therefore supportive of our resting state data being composed at least predominantly of wakefulness, and as we have pointed out, the changes to FC that we have observed are consistent with those seen in response to explicit sleep deprivation. However, future studies would need to record EEG data concurrently with the fMRI to allow unambiguous sleep staging, and thereby address this issue.

Our approach of investigating multiple ICNs and the interactions between them in relation to habitual variation in sleeping patterns has the potential to provide a more detailed mechanistic explanation for why some cognitive functions are affected by sleep status, whereas others are not, as well as for the individual differences that are seen in the effects of sleep deprivation. It would also be interesting to address the issue of how differences in cumulative TST link with sleep debt. In this study, we did not record information about participants' preferred amount of sleep, so we are not able to distinguish between those who achieved that amount versus those who did not. Future studies might examine whether the changes to FC in subjects who are not achieving their preferred amount of sleep are different to those who are, independently of how much sleep that represents.

Overall, this study is the first to address the question of how interactions within and between the major ICNs are related to variations in habitual sleep durations. These effects are not global, but specific to certain connections between certain pairs of nodes. In particular, the MPFC node of the DMN has FC that is related to cTST, whereas connections between the DMN and SN are also associated with cTST. Future work will need to address the behavioural implications of these observations to determine whether they underlie the known cognitive and behavioural effects associated with short sleep durations.[66]

REFERENCES

1. Horne JA. Human sleep, sleep loss and behavior: implications for the prefrontal cortex and psychiatric disorder. Br J Psychiatry 1993;162:413–9.
2. Dinges DF, Pack F, Williams K, et al. Cumulative sleepiness, mood disturbance and psychomotor vigilance performance decrements during a week of sleep restricted to 4-5 hours per night. Sleep 1997;20:267–77.
3. Harrison Y, Horne JA. The impact of sleep deprivation on decision making: a review. J Exp Psychol Appl 2000;6:236–49.
4. Belenky G, Wesensten NJ, Thorne DR, et al. Patterns of performance degradation and restoration during sleep restriction and subsequent recovery: a sleep dose-response study. J Sleep Res 2003;12:1–12.
5. Van Dongen HP, Maislin G, Mullington JM, Dinges DF. The cumulative cost of additional wakefulness: dose-response effects on neurobehavioral functions and sleep physiology from chronic sleep restriction and total sleep deprivation. Sleep 2003;26:117–29.
6. Babkoff H, Zukerman G, Fostick L, Ben-Artzi E. Effect of the diurnal rhythm and 24 h of sleep deprivation on dichotic temporal order judgment. J Sleep Res 2005;14:7–15.
7. Alhola P, Polo-Kantola P. Sleep deprivation: impact on cognitive performance. Neuropsychiatry Dis Treat 2007;3:553–87.
8. Banks S, Dinges DF. Behavioral and physiological consequences of sleep restriction. J Clin Sleep Med 2007;5:519–28.
9. Damoiseaux JS, Greicius MD. Greater than the sum of its parts: a review of studies combining structural connectivity and resting-state functional connectivity. Brain Struct Func 2009;213:525–33.
10. Bressler SL, Menon V. Large-scale brain networks in cognition: emerging methods and principles. Trends Cog Sci 2010;14:277–90.
11. Menon V, Uddin LQ. Saliency switching, attention and control: a network model of insula function. Brain Struct Func 2010;214:655–67.
12. Zhang D, Raichle ME. Disease and the brain's dark energy. Nat Rev Neurol 2010;6:15–28.
13. Menon V. Large-scale brain networks and psychopathology: a unifying triple network model. Trends Cogn Sci 2011;15:483–506.
14. Horovitz SG, Fukunaga M, de Zwart JA, et al. Low frequency BOLD fluctuations during resting wakefulness and light sleep: a simultaneous EEG-fMRI study. Hum Brain Mapp 2008;29:671–82.
15. Horovitz SG, Braun AR, Carr WS, et al. Decoupling of the brain's default mode network during deep sleep. Proc Natl Acad Sci U S A 2009;106:11376–81.
16. Larson-Prior LJ, Zempel JM, Nolan TS, Prior FW, Snyder AZ, Raichle ME. Cortical network functional connectivity in the descent to sleep. Proc Natl Acad Sci U S A 2009;106:4489–94.
17. Sämann PG, Wehrle R, Hoehn D, et al. Development of the brain's default mode network from wakefulness to slow wave sleep. Cereb Cortex 2011;21:2082–93.
18. Koike T, Kan S, Misaki M, Miyauchi S. Connectivity pattern changes in default-mode network with deep non-REM and REM sleep. J Neurosci Res 2011;69:322–30.
19. De Havas JA, Parimal S, Soon CS, Chee MW. Sleep deprivation reduces default mode network connectivity and anti-correlation during rest and task performance. Neuroimage 2012;59:1745–51.
20. Verweij IM, Romeijn N, Smit DJA, Piantoni G, Van Someren JW, van der Werf YD. Sleep deprivation leads to a loss of functional connectivity in frontal brain regions. BMC Neurosci 2014;1:88.
21. Tomasi D, Wang RL, Telang F, et al. Impairment of attentional networks after 1 night of sleep deprivation. Cereb Cortex 2009;19:233–40.
22. Bosch OG, Rihm JS, Scheidegger M, et al. Sleep deprivation increases dorsal nexus connectivity to the dorsolateral prefrontal cortex in humans. Proc Natl Acad Sci U S A 2013;48:19597–602.
23. Sämann PG, Wehrle R, Hoehn D, et al. Development of the brain's default mode network from wakefulness to slow wave sleep. Cereb Cortex 2011;21:2082–93.

24. Gujar N, Yoo SS, Hu P, Walker MP. The unrested resting brain: sleep deprivation alters activity within the default-mode network. J Cogn Neurosci 2010;22:1637–48.

25. Bagshaw AP, Khalsa S. Functional brain imaging and consciousness. In: Cavanna AE, Nani A, Blumenfeld H, Laureys S, eds. Neuroimaging of consciousness. Berlin Heidelberg: Springer, 2013:37–48.

26. Buckner RL, Carroll DC. Self-projection and the brain. Trends Cogn Sci 2007;2:49–57.

27. Spreng RN, Grady CL. Patterns of brain activity supporting autobiographical memory, prospection, and theory of mind, and their relationship to the default mode network. J Cogn Neurosci 2010;6:1112–23.

28. Drummond SPA, Walker M, Almklov E, Campos M, Anderson DE, Straus LD. Neural correlates of working memory performance in primary insomnia. Sleep 2013;36:1307–16.

29. Gumenyuk V, Roth T, Korzyukov O, Jefferson C, Bowyer S, Drake CL. Habitual short sleep impacts frontal switch mechanism in attention to novelty. Sleep 2011;34:1659–70.

30. Geol N, Roa H, Durmer JS, Dinges DF. Neurocognitive consequences of sleep deprivation. In seminars in neurology. NIH Public Access 2009:4:320.

31. Hillman DR, Lack LC. Public health implications of sleep loss: the community burden. Med J Aust 2013;199:7–10.

32. Altevogt BM, Colten HR, eds. Sleep disorders and sleep deprivation: an unmet public health problem. National Academies Press, 2006.

33. Ward AM, Schultz AP, Huijbers W, Van Dijk KR, Hedden T, Sperling RA. The parahippocampalgyrus links the default-mode cortical network with the medial temporal lobe memory system. Hum Brain Mapp 2013;35:1061–73.

34. Shulman GL, Fiez JA, Corbetta M, et al. Common blood flow changes across visual tasks: II. Decreases in cerebral cortex. J Cogn Neurosci 1997;5:648–63.

35. Raichle ME, MacLeod AM, Snyder AZ, Powers WJ, Gusnard DA, Shulman GL. A default mode of brain function. Proc Natl Acad Sci U S A 2001;2:676–82.

36. Raichle ME, Snyder AZ. A default mode of brain function: a brief history of an evolving idea. Neuroimage 2007;4:1083–90.

37. Anticevic A, Cole MW, Murray J, Corlett PR, Wang X, Krystal JH. The role of default network deactivation in cognition and disease. Trends Cogn Sci 2012;12:584–92.

38. Cavanna AE, Trimble MR. The precuneus: a review of its functional anatomy and behavioural correlates. Brain 2006;3:564–83.

39. Euston DR, Gruber AJ, McNaughton BL. The role of medial prefrontal cortex in memory and decision making. Neuron 2012;6:1057–70.

40. Utevsky AV, Smith DV, Huettel SA. Precuneus is a functional core of the default-mode network. J. Neurosci 2014;3:932–40.

41. Uddin LQ, Kelly AMC, Biswal BB, Castellanos FX, Milham MP. Functional connectivity of default mode network components: correlation, anticorrelation, and causality. Hum Brain Mapp 2009;2:625–37.

42. Spoormaker VI, Schröter MS, Gleiser PM, et al. Development of a large-scale functional brain network during human non-rapid eye movement sleep. J Neurosci 2010;30:11379–87.

43. Andrade KC, Spoormaker VI, Dresler M, et al. Sleep spindles and hippocampal functional connectivity in human NREM sleep. J Neurosci 2011;31:10331–9.

44. Sämann PG, Tully C, Spoormaker VI, et al. Increased sleep pressure reduces resting-state functional connectivity. MAGMA 2010;23:375–89.

45. De Havas JA, Parimal S, Soon CS, Chee MW. Sleep deprivation reduces default mode network connectivity and anti-correlation during rest and task performance. Neuroimage 2012;59:1745–51.

46. Killgore WD, Schwab ZJ, Weiner MR. Self-reported nocturnal sleep duration is associated with next-day resting state functional connectivity. Neuroreport 2012;23:741–5.

47. Hasenkamp W, Wilson-Mendenhall CD, Duncan E, Barsalou LW. Mind wandering and attention during focused meditation: a fine-grained temporal analysis of fluctuating cognitive states. Neuroimage 2012;59:750–60.

48. Manoliu A, Riedl V, Zherdin A, et al. Aberrant dependence of default mode/central executive network interactions on anterior insular salience network activity in schizophrenia. Schizophr Bull 2013;30–7.

49. Sridharan D, Levitin DJ, Menon V. A critical role for the right fronto-insular cortex in switching between central-executive and default-mode networks. Proc Natl Acad Sci U S A 2008;105:12569–74.

50. Kloss JD, Szuba MP, Dinges DF. Sleep loss and sleepiness: physiological and neurobehavioral effects. In: Davis KL, Charney D, Coyle JT, Nemeroff C, eds. Neuropsychopharmacology: The fifth generation of progress. Philadelphia, Lippincott Williams & Wilkins, 2002:1895–905.

51. Kushida CA, Chang A, Gadkary C, Guilleminault C, Carrillo O, Dement WC. Comparison of actigraphic, polysomnographic, and subjective assessment of sleep parameters in sleep-disordered patients. Sleep Med 2001;2:389–96.

52. Buysse DJ, Reynolds III CF, Monk TH, Berman SR, Kupfer DJ. The Pittsburgh Sleep Quality Index: a new instrument for psychiatric practice and research. Psychiatry Res 1989;28:193–213.

53. Johns MW. A new method for measuring daytime sleepiness: the Epworth sleepiness scale. Sleep 1991;14:540–5.

54. Lovibond PF, Lovibond SH. Manual for the Depression Anxiety Stress Scales (DASS, 2nd ed.). Sydney: Psychology Foundation of Australia, Monograph, 1995.

55. Åkerstedt T, Gillberg M. Subjective and objective sleepiness in the active individual. Int J Neurosci 1990;52:29–37.

56. Morgenthaler TI, Lee-Chiong T, Alessi C, et al. Practice parameters for the clinical evaluation and treatment of circadian rhythm sleep disorders. Sleep 2007;30:1445–59.

57. Smith S, Jenkinson M, Woolrich M, et al. Advances in functional and structural MR image analysis and implementation as FSL. Neuroimage 2004;23:208–19.

58. Jenkinson M, Bannister P, Brady J, Smith S. Improved optimisation for the robust and accurate linear registration and motion correction of brain images. Neuroimage 2002;17:825–41.

59. Przezdzik I, Bagshaw AP, Mayhew SD. Some brains are more strongly functionally connected than others: a resting-state fMRI study of inter and intra network coherence. Proc ISMRM 2013:2262.

60. Beckmann CF, DeLuca M, Devlin JT, Smith SM. Investigations into resting-state connectivity using independent component analysis. Phil Trans R Soc B 2005;360:1001–13.

61. Damoiseaux JS, Rombouts SARB, Barkhof F, et al. Consistent resting-state networks across healthy subjects. Proc Natl Acad Sci U S A 2006;103:13848–53.

62. Seeley WW, Menon V, Schatzberg AF, Keller J, Glover GH, Kenna H, Greicius MD. Dissociable intrinsic connectivity networks for salience processing and executive control. J Neurosci 2007;27:2349–56.

63. Khalsa S, Mayhew SD, Chechlacz M, Bagary M, Bagshaw AP. The structural and functional connectivity of the posterior cingulate cortex: comparison between deterministic and probabilistic tractography for the investigation of structure–function relationships. Neuroimage 2014;102:118–27.

64. Fox MD, Snyder AZ, Vincent JL, Corbetta M, Van Essen DC, Raichle ME. The human brain is intrinsically organized into dynamic, anticorrelated functional networks. Proc Natl Acad Sci U S A 2005;102:9673–8.

65. Benjamini Y, Hochberg Y. Controlling the false discovery rate: a practical and powerful approach to multiple testing. J R Stat Soc 1995;289–300

66. Basner M, Rao H, Goel N, Dinges DF. Sleep deprivation and neurobehavioural dynamics. Curr Opin Neurobiol 2013;23:854–63.

67. Murphy K, Birn RM, Handwerker DA, Jones TB, Bandettini PA. The impact of global signal regression on resting state correlations: are anti-correlated networks introduced? Neuroimage 2009;3:893–905.

68. Gotts SJ, Saad ZS, Jo HJ, Wallace GL, Cox RW, Martin A. The perils of global signal regression for group comparisons: a case study of Autism Spectrum Disorders. Front Hum Neurosci 2013;7:356.

69. Saad ZS, Gotts SJ, Murphy K, et al. Trouble at rest: how correlation patterns and group differences become distorted after global signal regression. Brain Connect 2012;1:25–32.

70. Schölvinck M L, Maier A, Frank QY, Duyn JH, Leopold DA. Neural basis of global resting-state fMRI activity. Proc Natl Acad Sci U S A 2010;107:10238–43.

71. Wong CW, Olafsson V, Tal O, Liu TT. The amplitude of the resting-state fMRI global signal is related to EEG vigilance measures. Neuroimage 2013;83:983–90.

72. Fukunaga M, Horovitz SG, van Gelderen P, et al. Large-amplitude, spatially correlated fluctuations in BOLD fMRI signals during extended rest and early sleep stages. Magn Reson Imaging 2006;24:979–92.

73. Yeo, BT, Tandi J, Chee MW. Functional connectivity during rested wakefulness predicts vulnerability to sleep deprivation. Neuroimage 2015;111:147–58.

74. Ong JL, Kong D, Chia TT, Tandi J, Yeo BT, Chee MW. Co-activated yet disconnected–Neural correlates of eye closures when trying to stay awake. Neuroimage 2015;118:553–62.

75. Behzadi Y, Restom K, Liau J, Liu TT. A component based noise correction method (CompCor) for BOLD and perfusion based fMRI. Neuroimage 2007;37:90–101.

76. Carbonell F, Bellec P, Shmuel A. Quantification of the impact of a confounding variable on functional connectivity confirms anti-correlated networks in the resting-state. Neuroimage 2014;86:343–53.

77. Hartzell JF, Tobia MJ, Davis B, Cashdollar NM, Hasson U. Differential lateralization of hippocampal connectivity reflects features of recent context and ongoing demands: an examination of immediate post-task activity. Hum Brain Mapp 2015;36:519–37.

78. de Pasquale F, Della Penna S, Snyder AZ, et al. A cortical core for dynamic integration of functional networks in the resting human brain. Neuron 2012;74:753–64.

79. Tagliazucchi E, von Wegner F, Morzelewski A, Borisov S, Jahnke K, Laufs H. Automatic sleep staging using fMRI functional connectivity data. Neuroimage 2012;1:63–72.

SUBMISSION & CORRESPONDENCE INFORMATION

DISCLOSURE STATEMENT