RESEARCH ARTICLE

The Role of the Suprachiasmatic Nucleus in Cardiac Autonomic Control during Sleep

S. D. Joustra1*, R. H. Reijntjes2, A. M. Pereira1, G. J. Lammers2,3, N. R. Biermasz1, R. D. Thijs2,3

1 Department of Medicine, Division of Endocrinology, Centre for Endocrine Tumours Leiden, Leiden University Medical Centre, Leiden, Netherlands, 2 Department of Neurology, Leiden University Medical Centre, Leiden, Netherlands, 3 Stichting Epilepsie Instellingen Nederland (SEIN), Heemstede, Netherlands

* sdjoustra@lumc.nl

Abstract

Background

The suprachiasmatic nucleus (SCN) may play an important role in central autonomic control, since its projections connect to (para)sympathetic relay stations in the brainstem and spinal cord. The cardiac autonomic modifications during nighttime may therefore not only result from direct effects of the sleep-related changes in the central autonomic network, but also from endogenous circadian factors as directed by the SCN. To explore the influence of the SCN on autonomic fluctuations during nighttime, we studied heart rate and its variability (HRV) in a clinical model of SCN damage.

Methods

Fifteen patients in follow-up after surgical treatment for nonfunctioning pituitary macroadenoma (NFMA) compressing the optic chiasm (8 females, 26–65 years old) and fifteen age-matched healthy controls (5 females, 30–63 years) underwent overnight ambulatory polysomnography. Eleven patients had hypopituitarism and received adequate replacement therapy. HRV was calculated for each 30-second epoch and corrected for sleep stage, arousals, and gender using mixed effect regression models.

Results

Compared to controls, patients spent more time awake after sleep onset and in NREM1-sleep, and less in REM-sleep. Heart rate, low (LF) and high frequency (HF) power components and the LF/HF ratio across sleep stages were not significantly different between groups.

Conclusions

These findings suggest that the SCN does not play a dominant role in cardiac autonomic control during sleep.
**Introduction**

Sleep exerts major effects on cardiac autonomic control. For example, compared to slow-wave sleep, rapid eye movement sleep is associated with increased heart rate (HR) and low-frequency power of HR variability (HRV), and decreased high-frequency power [1]. The suprachiasmatic nucleus (SCN) is the critical relay of the diurnal sleep-wake regulation [2]. It may also play an important role in central autonomic control, as tracing studies demonstrated that SCN neurons project to the paraventricular nucleus and connect with parasympathetic and sympathetic relay stations in the brainstem and spinal cord [3,4]. Accordingly, SCN lesions in rat reduced the HR decrease during resting periods [5]. Furthermore, humans showed diurnal rhythms of HR and HRV independent of sleep stage in a constant routine protocol [6]. Consequently, it may well be argued that cardiac autonomic control during nighttime not only results from direct effects of sleep-related changes in the central autonomic network, but also from endogenous circadian factors as directed by the SCN.

Nonfunctioning pituitary macroadenomas (NFMA) have been proposed as a model of SCN damage [7], and thus provide an unique opportunity to study the influence of the SCN on autonomic fluctuations during sleep in humans. NFMA compress surrounding tissue, causing hypopituitarism and/or visual impairments. Transsphenoidal adenomectomy, sometimes complemented with radiotherapy, usually improves visual function, but hypopituitarism may persist [8]. Additionally, long-term remission is accompanied by poor subjective sleep quality, fragmented sleep-wake patterns, and alterations of diurnal melatonin and temperature rhythmicity [9,10]. These symptoms were strongly associated with suprasellar tumour extension (irrespective of hypopituitarism), implying damage to the adjacent SCN. To understand the role of the SCN in cardiac autonomic control during sleep, we studied HR control during nighttime in patients treated for NFMA and age-matched controls.

**Methods**

**Participants**

Seventeen adult patients surgically treated for NFMA and seventeen age-matched healthy controls underwent a single night of ambulatory polysomnography [9]. All patients were otherwise healthy and received yearly follow-up by an endocrinologist. Exclusion criteria were age > 65 years, use of psychotropic or cardiac medication, diagnosis of a sleep disorder, hypertension, dyslipidemia, or diabetes mellitus. HRV data from two patients and two controls were excluded for insufficient data quality for this analysis (detached HR electrode), thus fifteen patients and fifteen healthy controls were studied.

Markers of circadian rhythmicity in these patients and their methods were previously published [9,10], and included intradaily variability, skin temperature, and melatonin secretion profiles. The intradaily variability, derived from 7 days of actigraphy, quantifies how fragmented the rhythm of motor activity is relative to its 24-h amplitude; more frequent alterations between an active and an inactive state lead to a higher intradaily variability [11]. Proximal skin temperature was measured for 24 hours at both infraclavicular areas, supra-umbilical, and on the left mid-thigh. Its diurnal variation is related to increased sleep latency in both narcoleptics and healthy persons [12,13], and to sleep depth [14]. In our previous study differences in proximal skin temperature between NFMA patients and controls were more pronounced than those in distal skin temperature or core body temperature [10]. Melatonin secretion was measured for 36 hours using at least thirteen saliva samples. An altered melatonin profile was defined as the absence of an evening rise or daytime values >3 pg/mL in a 36-hour salivary melatonin collection. We refer to S1 Table for the values of these markers for each patient.
The ethical committee of the Leiden University Medical Center approved the study. All subjects gave written informed consent.

HRV

Methods of polysomnography [9] and calculation of HRV [15,16] have been detailed previously. In short, sleep stages, apnea/hypopnea events and leg movements were manually scored in 30-second epochs by an experienced sleep technician. All HR data from nocturnal sleep onset until morning awakening were selected. A continuous wavelet transform was implemented in Matlab (Version 13.1, Mathworks, MA, USA) to detect R-peaks in the 30-second epochs. Outliers, differing $> 25$ beats per minute from adjacent samples, were excluded. Heartbeat data were resampled at 5 samples per second. The full spectrum of HRV in the region surrounding each sample was calculated with a fast Fourier transform using a window of 512 samples, ranging 256 samples left and right of a particular sample. After averaging these spectra for each 30-second epoch, the LF ($0.04 – 0.15$ Hz) and HF ($0.15 – 0.4$ Hz) power components per epoch were calculated. The LF band is thought to predominantly reflect the baroreflex-mediated sympathetic activity [16] whereas the HF band is thought to represent an index of vagal activity, and the LF/HF ratio an index of the sympathovagal balance [17]. All measures have their limitation and should be interpreted cautiously. To account for the autonomic effects of arousals, we identified all epochs with apnea, hypopnea, leg movements, or transitions from NREM3 to NREM1/2 or NREM2 to NREM1. The raw data file on HR and HRV is available in the S1 Data File.

Statistical analysis

Patients’ clinical characteristics were compared with controls using the Student’s $t$-test for numerical data (or in case the assumption of normality was not met (Shapiro–Wilk test) the Mann–Whitney U test), and Pearson’s $\chi^2$ for categorical data. Mixed effect regression models were used to analyse the effect of disease on HR, LF, HF, and LF/HF. This model included all 30-second epochs. Epochs were classified as rapid eye movement- (REM), non-REM stage 1- (NREM1-), NREM2-, and NREM3-sleep. Disease, wake and sleep stages, leg movements, apnea events, gender, and arousal transitions were selected as fixed effects and participants as random effect. A natural logarithm-transformation was applied to obtain a normal distribution of the model’s residuals. Differences were considered statistically significant at $P < 0.05$. In a secondary analysis, the interaction between disease and each of the sleep stages was added to the model, to explore sleep stage-dependent differences in HRV. To correct for multiple testing after introducing the interaction terms (five sleep stages), sleep stage-dependent differences were considered statistically significant at $P < 0.01$.

Results

Participants

Median age was 58 yr (range 26–65 yr) for NFMA patients (8 females), and 52 yr (range 30–63 yr) for the control subjects (5 females) (Table 1). Prior to surgery, most patients had visual field defects (80%) or suprasellar extension on MRI (93%). Hypopituitarism was present in eleven (deficiency of growth hormone in ten, of corticotropin in five, of thyrotropin in ten, and of the gonadotropins in five); all received hormone replacement, except for optional growth hormone replacement, which was left untreated in three patients. Healthy control subjects were not significantly different in terms of gender, age, or BMI. Compared to controls, patients spent more time awake ($P = 0.005$) and in NREM1-sleep ($P < 0.001$), and less in REM-sleep ($P < 0.001$).
Mean duration of apnea was shorter in patients (13.4 ± 9.1 seconds vs. 21.8 ± 8.0 seconds, \( P = 0.008 \)).

HRV

We included an average total number of 1374 epochs per participant in our model. Patients did not differ from their age-matched controls during the night with respect to HR (\( P = 0.137 \)), LF (\( P = 0.205 \)), HF (\( P = 0.959 \)), or LF/HF (\( P = 0.184 \)) (Table 2). As differences might be dependent on patients with the most prominent signs of circadian dysrhythmia, e.g. those with disturbed melatonin secretion, this feature was added as an additional coefficient to the model, but lacked significance. Fig 1 displays the results from the secondary analysis, exploring sleep stage—dependent differences in HR and HRV between patients and controls. LF/HF was
slightly lower in patients than controls (difference: 0.44 [standard error: 0.20], \( P = 0.040 \)), but this difference did not reach statistical significance after correction for multiple testing (\( P > 0.01 \)).

**Discussion**

We studied the role of the SCN in sleep-related cardiac autonomic control in a cohort of NFMA patients and healthy age-matched controls and did not identify major autonomic alterations. Our results suggest that modulation of sleep-related cardiac autonomic control is predominantly linked to sleep processes with a subordinate role for the SCN. This is supported by experiments of shifting the sleep period, inducing a concomitant shift in the diurnal variation of HRV [18], in contrast to other circadian parameters such as the melatonin peak and core body temperature nadir that showed respectively no shift and a biphasic curve in similar experiments [19]. Furthermore, preservation of the effects of sleep on HR in patients with bilateral carotid tumour resection also suggests that these alterations are predominantly generated through central, non-baroreflex mediated pathways [16]. In rats a bidirectional relationship has been demonstrated between sleep processes and SCN activity [20]. Sleep processes might therefore influence cardiac control through altering SCN activity. In view of our findings it seems however likely that other central circuitries linking sleep and cardiac autonomic control, e.g. the hypothalamic nuclei [21, 22], outweigh this influence.

Our study has limitations. First, the extent of SCN dysfunctioning in our patients is not exactly known, since at present no parameter is available to directly measure SCN functioning. However, all patients had indirect signs of SCN dysfunctioning, e.g. altered rhythmicity of melatonin/temperature/sleep, albeit in variable patterns [9,10]. Second, the sample size limits the statistical strength to rule out small differences. It should however be noted that even smaller sample sizes were able to demonstrate distinct and consistent autonomic differences across...
sleep stages in narcolepsy and bilateral carotid body resection, using identical study protocols [15,16].

In conclusion, we did not observe major differences in HR and HRV between NFMA patients and controls during sleep. The findings suggest that the SCN does not play a dominant role in cardiac autonomic control during sleep.

Supporting Information

S1 Data File. Original HR and HRV analysis results.
(XLSX)

S1 Table. Individual values of markers of circadian rhythmicity.
(DOCX)
Acknowledgments
The authors acknowledge the efforts of the sleep technicians from the department of Neurology of the Leiden University Medical Centre, and the assistance of Prof. Dr. T. Stijnen from the department of Medical Statistics and Bioinformatics.

Author Contributions
Conceived and designed the experiments: SDJ RHR AMP GJL NRB RDT. Performed the experiments: SDJ RHR. Analyzed the data: SDJ RHR AMP GJL NRB RDT. Wrote the paper: SDJ RHR AMP GJL NRB RDT.

References
1. Chouchou F, Desseilles M. Heart rate variability: a tool to explore the sleeping brain? Frontiers in neuroscience. 2014; 8: 402. doi:10.3389/fnins.2014.00402 PMID: 25565936
2. Moore RY. Suprachiasmatic nucleus in sleep-wake regulation. Sleep Med. 2007; 8 Suppl 3: 27–33. PMID: 18032104
3. Buijs RM, la Fleur SE, Wortel J, Van HC, Zuiddam L, Mettenleiter TC, et al. The suprachiasmatic nucleus balances sympathetic and parasympathetic output to peripheral organs through separate pre-autonomic neurons. J Comp Neurol. 2003; 464(1): 36–48. PMID: 12866127
4. Golombek DA. Circadian Rhythms and autonomic function. In: Robertson D, Biaggioni I, Burnstock G, Low PA, Patton JA, editors. Primer on the autonomic nervous system. Third Edition ed. London: Academic Press; 2012. p. 157–9.
5. Scheer FA, Ter Horst GJ, Van der Vliet J, Buijs RM. Physiological and anatomic evidence for regulation of the heart by suprachiasmatic nucleus in rats. Am J Physiol Heart Circ Physiol. 2001; 280(3): H1391–H9. PMID: 11179089
6. Boudreau P, Yeh WH, Dumont GA, Boivin DB. Circadian variation of heart rate variability across sleep stages. Sleep. 2013; 36(12): 1919–28. doi: 10.5665/sleep.3230 PMID: 24293767
7. Joustra SD, Kruisjens E, Verstegen MJ, Pereira AM, Biermasz NR. Determinants of Altered Sleep-wake Rhythmicity in Patients Treated for Nonfunctioning Pituitary Macroadenomas. J Clin Endocrinol Metab. 2014: jc20142602.
8. Dekkers OM, de Keizer RJ, Roelfsema F, Vd Klauuw AA, Honkoop PJ, van Duiken H, et al. Progressive improvement of impaired visual acuity during the first year after transsphenoidal surgery for non-functioning pituitary macroadenoma. Pituitary. 2007; 10(1): 61–5. PMID: 17318437
9. Biermasz NR, Joustra SD, Donga E, Pereira AM, van Duinen N, van Dijk M, et al. Patients previously treated for nonfunctioning pituitary macroadenomas have disturbed sleep characteristics, circadian movement rhythm, and subjective sleep quality. J Clin Endocrinol Metab. 2011; 96(5): 1524–32. doi: 10.1210/jc.2010-2742 PMID: 21367934
10. Joustra SD, Thijs RD, van den Berg R, van Dijk M, Pereira AM, Lammers GJ, et al. Alterations in diurnal rhythmicity in patients treated for nonfunctioning pituitary macroadenoma; a controlled study and literature review. Eur J Endocrinol. 2014.
11. Van Someren EJ, Swaab DF, Colenda CC, Cohen W, McCall WV, Rosenquist PB. Bright light therapy: improved sensitivity to its effects on rest-activity rhythms in Alzheimer patients by application of non-parametric methods. Chronobiol Int. 1999; 16(4): 505–18. PMID: 10442243
12. Fronczek R, Overeem S, Lammers GJ, van Dijk JG, Van Someren EJ. Altered skin-temperature regulation in narcolepsy relates to sleep propensity. Sleep. 2006; 29(11): 1444–9. PMID: 17162991
13. Raymann RJ, Swaab DF, Van Someren EJ. Cutaneous warming promotes sleep onset. AmJPhysiol RegulIntegrComp Physiol. 2005; 288(6): R1589–R97.
14. Raymann RJ, Swaab DF, Van Someren EJ. Skin deep: enhanced sleep depth by cutaneous temperature manipulation. Brain. 2008; 131(Pt 2): 500–13. doi: 10.1093/brain/aws015 PMID: 18192289
15. Van Der Mejden WP, Fronczek R, Reijntjes RH, Corssmit EP, Biermasz NR, Lammers GJ, et al. Time- and state-dependent analysis of autonomic control in narcolepsy: higher heart rate with normal heart rate variability independent of sleep fragmentation. J Sleep Res. 2015; 24(2): 206–14. doi: 10.1111/jsr.12253 PMID: 25382307
16. Niemeijer ND, Corssmit EP, Reijntjes RH, Lammers GJ, van Dijk JG, Thijs RD. Sleep-mediated heart rate variability after bilateral carotid body tumor resection. Sleep. 2015; 38(4): 633–9. doi: 10.5665/sleep.4586 PMID: 25335476
17. TFotESoCatNASoPa Electrophysiology. Heart rate variability: standards of measurement, physiological interpretation and clinical use. Circulation. 1996; 93(5): 1043–65. PMID: 8598068
18. Viola AU, Simon C, Ehrhart J, Geny B, Piquard F, Muzet A, et al. Sleep processes exert a predominant influence on the 24-h profile of heart rate variability. J Biol Rhythms. 2002; 17(6): 539–47. PMID: 12465887
19. Weibel L, Spiegel K, Gronfier C, Follenius M, Brandenberger G. Twenty-four-hour melatonin and core body temperature rhythms: their adaptation in night workers. Am J Physiol. 1997; 272(3 Pt 2): R948–R54. PMID: 9087659
20. Deboer T, Vansteensel MJ, Detari L, Meijer JH. Sleep states alter activity of suprachiasmatic nucleus neurons. Nat Neurosci. 2003; 6(10): 1086–90. PMID: 12958601
21. Calandra-Buonaura G, Provini F, Guaraldi P, Plazzi G, Cortelli P. Cardiovascular autonomic dysfunctions and sleep disorders. Sleep Med Rev. 2015.
22. Silvani A, Dampney RA. Central control of cardiovascular function during sleep. Am J Physiol Heart Circ Physiol. 2013; 305(12): H1683–H92. doi: 10.1152/ajpheart.00554.2013 PMID: 24097430