

Electrocardiogram-Based Sleep Spectrogram Measures of Sleep Stability and Glucose Disposal in Sleep Disordered Breathing

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Study Objectives: Sleep disordered breathing (SDB) is independently associated with insulin resistance, glucose intolerance, and type 2 diabetes mellitus. Experimental sleep fragmentation has been shown to impair insulin sensitivity. Conventional electroencephalogram (EEG)-based sleep-quality measures have been inconsistently associated with indices of glucose metabolism. This analysis explored associations between glucose metabolism and an EEG-independent measure of sleep quality, the sleep spectrogram, which maps coupled oscillations of heart-rate variability and electrocardiogram (ECG)-derived respiration. The method allows improved characterization of the quality of stage 2 non-rapid eye movement (NREM) sleep.

Design: Cross-sectional study.

Setting: N/A.

Participants: Nondiabetic subjects with and without SDB (n = 118) underwent the frequently sampled intravenous glucose tolerance test (FSIVGTT) and a full-montage polysomnogram. The sleep spectrogram was generated from ECG collected during polysomnography.

Interventions: N/A.

Measurements and Results: Standard polysomnographic stages (stages 1, 2, 3+4, and rapid eye movement [REM]) were not associated with the disposition index (D_i) derived from the FSIVGTT. In contrast, spectrographic high-frequency coupling (a marker of stable or "effective" sleep) duration was associated with increased, and very-low-frequency coupling (a marker of wake/REM/transitions) associated with reduced D_i. This relationship was noted after adjusting for age, sex, body mass index, slow wave sleep, total sleep time, stage 1, the arousal index, and the apnea-hypopnea index.

Conclusions: ECG-derived sleep-spectrogram measures of sleep quality are associated with alterations in glucose-insulin homeostasis. This alternate mode of estimating sleep quality could improve our understanding of sleep and sleep-breathing effects on glucose metabolism.

Keywords: Disposition index, diabetes mellitus type 2, sleep spectrogram, sleep quality

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INTRODUCTION

Type 2 diabetes mellitus is characterized by insulin resistance and progressive beta-cell dysfunction. Sleep has complex effects on hormone regulation,¹ including adipocyte function and lipid metabolism.² Research over the last several decades has conclusively shown that age, obesity, and sedentary lifestyle are some of the strongest risk factors for type 2 diabetes.³ The prevalence of sleep disordered breathing (SDB) in patients with type 2 diabetes is high,⁴ and abnormalities of glucose metabolism are more prevalent in those with SDB.⁵⁻⁶

SDB, characterized by recurrent episodes of total or partial upper airway occlusion, intermittent hypoxia, and sleep fragmentation is independently associated with insulin resistance, glucose intolerance, and diabetes, as demonstrated by many epidemiologic studies.⁷⁻¹⁰ Epidemiologic studies have also shown that habitual short sleep duration and poor-quality sleep are associated with alterations in insulin and glucose homeostasis.¹¹⁻¹⁴ In the Coronary Artery Risk Development in Young Adults

(CARDIA) Study, habitual sleep duration and fragmentation were estimated from 6 days of wrist actigraphy after adjustment for covariates. In CARDIA, actigraphic sleep fragmentation and subjective insomnia were associated with higher fasting glucose and insulin levels.¹⁵ Experimental sleep restriction¹⁶⁻¹⁸ and sleep fragmentation in healthy humans¹⁹ have provided corroborating evidence that sleep quantity and quality may be important determinants of insulin sensitivity. Thus, an important role for sleep in glucose homeostasis seems very likely.

Conventional sleep-quality measures, which are based on the visual classification of the sleep electroencephalogram (EEG),²⁰ have been inconsistently associated with indices of glucose metabolism.^{19,21-22} Moreover, in older individuals, the population at greatest risk for type 2 diabetes, the age-related paucity of slow wave sleep could make it difficult to assess associations between sleep quality and glucose metabolism. The sleep spectrogram, which is an EEG-independent measure of sleep quality, maps sleep-state-modulated coupled oscillations of heart-rate variability (HRV) and electrocardiogram (ECG) QRS-wave-amplitude changes associated with breathing, called ECG-derived respiration (EDR); thus, the need to visually characterize the EEG is avoided, and the technique can be accurately automated.²³ Generated from a single channel of ECG, the technique has previously demonstrated that non-rapid eye movement (NREM) sleep is characterized by relatively distinct patterns of HRV-EDR coupling that are clearly separable from each other. One pattern, called high-frequency coupling, is the

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proposed spectrographic marker of undisturbed and “effective” sleep; low-frequency coupling is the biomarker of fragmented “ineffective” sleep, whereas very-low-frequency coupling is enriched in periods of wake, rapid eye movement (REM) sleep, and sleep-wake transitions. Periods of sleep characterized by high-frequency coupling demonstrate stable respiration, strong sinus arrhythmia, paucity of EEG arousals, and blood pressure dipping.²⁴ Conversely, periods of low-frequency coupling are characterized by cyclic variation in heart rate, tidal volume fluctuations, prominent phasic EEG phenomena including arousals, and lack of blood pressure dipping. One important advantage of the spectrogram is that NREM sleep, regardless of conventional stage, can be readily characterized at any age into effective and ineffective forms. This alternative approach to sleep characterization has shown preliminary utility, being associated with covariate-adjusted prevalent hypertension and stroke,²⁵ shows moderate heritability²⁶ and differentiates depression²⁷ and fibromyalgia²⁸ patients from matched control subjects.

We hypothesized that ECG-based spectrographic measures, being EEG-amplitude independent, would provide a more robust measure of sleep quality and, thus, disclose independent associations of sleep quality with alterations in glucose metabolism. Specifically, sleep state that is dominated by high-frequency coupling would have favorable glucose and insulin kinetics, whereas excessive low- or very-low-frequency coupling would be associated with impairments in glucose and insulin kinetics.

METHODS

Subjects and Study Design

The study sample, which has been described previously,²¹ comprised otherwise healthy subjects recruited from the community and patients with newly diagnosed but untreated SDB. Volunteers were excluded from participation if their fasting blood glucose level was greater than 125 mg/dL or if they had a history of type 2 diabetes mellitus, angina, myocardial infarction, coronary revascularization, congestive heart failure, cerebral vascular accident, obstructive lung disease, renal or hepatic dysfunction, prior upper airway surgery, malignancy, or any chronic inflammatory condition. Additional exclusion criteria included history of circadian rhythm disorder, chronic insufficient sleep times (< 7 h per night), and use of antiinflammatory medication, supplemental oxygen, or positive airway pressure ventilation. Informed consent was obtained from all participants, and the study protocol was approved by the Institutional Review Board on human research. All subjects were studied at Johns Hopkins University, and the sleep spectrogram was derived from the overnight polysomnogram from deidentified data at the Beth Israel Deaconess Medical Center.

Polysomnography

The overnight polysomnogram included recordings of C3/M2 and C4/M1 EEG, right and left electrooculograms, submental and bilateral anterior tibialis electromyograms, and body position. Respiration was monitored with a nasal pressure transducer, thermocouples at the nose and mouth, and thoracic and abdominal strain gauges. Continuous recording of the oxyhemoglobin saturation (SaO₂) was obtained with an oximeter (Ohmeda 3700; Englewood, CO). All physiologic signals were

digitized for offline analysis of sleep and breathing patterns (Embla, Broomfield, CO). Sleep-stage scoring was performed in the original study using 30-second epochs according to Rechtschaffen and Kales criteria.²⁹ Apneas were identified if airflow was absent in the thermistor and nasal cannula channels for at least 10 seconds. Hypopneas were identified if there was at least a 30% reduction in airflow, which occurred for at least 10 seconds and was associated either with a 4% oxyhemoglobin desaturation or an EEG arousal. The apnea-hypopnea index (AHI) was defined as the number of apneas or hypopneas per hour of sleep. Arousals were identified as abrupt shifts of at least 3 seconds' duration in EEG frequency according to standard criteria.³⁰

Measurement of Insulin Sensitivity

A frequently sampled intravenous glucose tolerance test (FSIVGTT) was performed (09:00) following an overnight fast. An intravenous line was placed into each antecubital vein, one used for infusion of insulin or glucose and the other for blood sampling. Following baseline measurements of insulin and glucose (time -15, -10, -5, and -1 minute), a glucose load (50% dextrose, 0.3 gm/kg) was infused at time 0 over 1 minute followed by a continuous normal saline infusion. Twenty minutes after the glucose injection, a bolus dose of regular insulin (0.03 units/kg) was given. Venous blood (for insulin and glucose levels) was collected at 2, 3, 4, 5, 6, 8, 10, 12, 14, 16, 19, 22, 24, 25, 27, 30, 40, 50, 60, 70, 80, 90, 100, 120, 140, 160, and 180 minutes after the glucose challenge. The computerized algorithm (MINMOD [minimal model analysis] software) was used to analyze the glucose and insulin levels obtained during the FSIVGTT.³¹

The MINMOD analysis provides a mathematical description of glucose disposal during the FSIVGTT. The measured insulin profile during the FSIVGTT is the model input from which the glucose concentration profile is mathematically determined by nonlinear least-squares fitting and the model indices are estimated.³¹ Insulin sensitivity (S_I) is the ability of insulin to enhance glucose uptake by peripheral tissue (muscle, liver, adipose). The acute beta-cell response or acute insulin response to glucose (AIRg) represents the ability of the pancreatic beta-cells to secrete insulin in response to glucose.³¹ The AIRg is determined as the area under the insulin curve between minute 0 and 10 of the FSIVGTT. The disposition index (D_I), a measure of pancreatic beta-cell function, is calculated from the product of S_I and AIRg ($D_I = S_I \times \text{AIRg}$). The D_I is predictive of incident type 2 diabetes, and levels below 1000 are known to substantially increase the risk.³²⁻³⁴ Our analysis was restricted to the D_I .

Sleep Spectrogram Analysis

The sleep spectrogram provides a measure of cardiopulmonary coupling and has been described previously.^{23,35} Briefly, the sleep spectrogram is a mathematical integration of heart-rate variability and R-wave-amplitude fluctuations driven by respiration (ECG-derived respiration) using a single-channel ECG. The R-wave fluctuations result from change in cardiac electrical axis relative to the ECG electrodes as the lungs fill and empty during respiration. After filtering for outliers (secondary to missed R-wave detections or artifact) and resampling of the data at 2 Hz, the cross-spectral power and the coherence

of the signals are calculated with an 8.5-minute (1024-sample) window using the fast Fourier transform. Each 1024-sample coherence window is advanced by 256 samples (2.1 minutes) and repeated until the entire series is analyzed. A ratio of coherence cross power in the low-frequency (0.01-0.1 Hz) band to high-frequency (0.1-0.4 Hz) band is calculated for each 1024-sample window from the product of coherence and cross-spectral power. The output from this analysis includes high-frequency coupling (HFC, 0.1-0.4 Hz), which represents stable or “effective” sleep and correlates with EEG non-cyclic alternating pattern (non-CAP); low-frequency coupling (LFC, 0.01-0.1 Hz), which correlates with unstable or “ineffective” sleep and EEG-CAP; and very-low-frequency coupling (VLFC, 0.01-0.001 Hz), which is associated with wake, REM sleep, and sleep-wake transitions. For this analysis, the software implemented in RemLogic (Embla, Inc.) was used. To ensure that the spectrogram analysis included only the sleep period, sleep staging from the polysomnogram was used to restrict analysis to the sleep period. HFC, LFC, and VLFC are then expressed in minutes and percentage of the sleep period. The sleep period is derived from the first and last epoch of scored polysomnographic sleep.

There are distinct differences between standard HRV measures and the sleep spectrogram. First, even if HRV itself is reduced (such as in heart failure, diabetes, use of β -adrenergic receptor blocking agents), the ECG-derived respiration signal is able to provide a robust spectrogram. Second, HFC and LFC are clearly differentiated on the spectrogram, allowing a differentiation into periods of HFC versus LFC not readily possible using HRV alone. Third, although the frequency range of HFC and high-frequency power on HRV analysis are essentially the same, the low-frequency component of HRV analysis is not LFC. LFC is driven solely by respiratory tidal-volume fluctuations. In patients with sleep apnea, periods of HFC would be identical to “periods of stable breathing” that are well recognized, whereas LFC would be identical to periods of unstable respiration associated with cyclic variation in heart rate.

Statistical Methods

Summary measures were means and standard deviations. Spectrographically determined sleep state (HFC, LFC, and VLFC) were considered as continuous independent measures. Correlations were also computed between spectrogram measures and conventional polysomnographic variables in this dataset (Table 2). A multiple-regression model was constructed: unadjusted (Model-1) and adjusting in sequential models for sex, age, body mass index (Model-2); plus NREM slow wave sleep and total sleep time (Model-3); plus arousal index and stage 1 (Model-4); plus AHI (Model-5) (Table 3). Independent contributions of ECG-spectrographic variables in explaining the D_i were estimated using model R and adjusted R^2 . Statistical significance was set at 0.05 or less. STATA (Stata Corp LP, College Station, TX) was used for statistical analysis.

RESULTS

The study sample, which included 118 subjects with complete data on polysomnography and the FSIVGTT, was grouped as follows: 39 subjects had no SDB (AHI < 5 events/h), 34 had mild SDB (AHI: 5.0–14.9 events/h), 22 had moderate SDB

(AHI: 15.0-29.9 events/h), and 23 had severe SDB (AHI > 30.0 events/h).²¹ Table 1A summarizes the characteristics of the study sample. During the FSIVGTT, the disposition index ($D_i = S_i \times \text{AIRg}$) was lower in those with moderate to severe SDB.²¹

Spectrographic Variables and SDB Severity

Table 1B displays the spectrographic variables (duration and percentage of total sleep time) according to SDB severity. This table demonstrates that the percentage and duration of HFC, the spectrographic correlate of stable or “effective” sleep decreased as SDB severity increased, whereas the percentage and duration of LFC, the correlate for unstable or “ineffective” sleep, increased with increasing SDB severity. Figures 1 and 2 show samples of subjects with high and low proportions of HFC, respectively.

Conventional Sleep Stages and Spectrographic Frequencies

Table 2 displays the correlation matrix between standard polysomnographic sleep stages and spectrographic frequencies. This analysis demonstrates that LFC, the spectrographic marker of unstable or “ineffective” sleep, correlated positively with stage 1 and negatively with slow wave sleep; that VLFC correlated positively with REM sleep; and that HFC, the spectrographic marker of stable or “effective” sleep, correlated positively with slow wave sleep (stage 3 + stage 4).

Conventional Sleep Stages and IVGTT Indices

Although the arousal index was independently associated with glucose effectiveness, standard polysomnographic sleep stages (stage 1, stage 2, slow wave sleep, REM sleep) did not have significant associations with any of the IVGTT indices of glucose metabolism.²¹

Sleep Spectrogram Frequencies and IVGTT Indices

Table 3 (starts on page 145) summarizes the relationship between spectrographic metrics (duration and percentage of the sleep period) and the D_i , unadjusted and adjusted in sequential models. HFC duration was associated, in the full model (Model-5), with improvements in the D_i . VLFC duration and percentage were associated only in the full model with decrements in the D_i .

DISCUSSION

The primary finding of this study is that the duration of HFC, an ECG-derived spectrographic biomarker of “effective” and plausibly restorative sleep, was associated with preferable glucose disposal characteristics. This relationship persisted after adjustments that included polysomnographic measures of sleep quality and sleep apnea severity. Increased VLFC, the spectrographic biomarker of wake and REM-like states, was also associated with worse glucose disposal. In this same data set, it was previously reported that an analysis of conventional sleep stages (stage 1, stage 2, slow wave sleep, REM sleep) showed no association between the percentages of different sleep stages and FSIVGTT-derived measures (see Punjabi and Beamer²¹).

Identifying associations between spectrographic coupling biomarkers and metrics of glucose metabolism independent of AHI and polysomnographic sleep quality suggests that additional insights may be provided into the effects of sleep disruption that is not gained by using conventional sleep-breathing

Table 1A—Subject characteristics

	AHI, events/h			
	< 5.0 (n = 39)	5.0-14.9 (n = 34)	15.0-29.9 (n = 22)	> 30.0 (n = 23)
Demographics				
Male sex, %	43.6	61.8	63.6	82.6
White race, %	92.3	79.4	95.5	78.3
Age, y	37.7 (1.9)	46.5 (2.0)	51.6 (2.0)	52.4 (2.0)
Body Composition				
BMI, kg/m ²	26.5 (0.8)	29.6 (1.1)	30.7 (0.8)	32.8 (1.3)
Waist circumference, cm	84.1 (1.8)	91.8 (2.3)	96.1 (2.3)	106.4 (3.2)
Body fat, %	30.3 (1.7)	31.9 (1.8)	32.6 (1.6)	34.9 (1.6)
Polysomnography				
Total sleep time, h	6.8 (0.2)	6.9 (0.2)	6.7 (0.2)	6.4 (0.2)
Sleep stage, %				
1	8.1 (0.8)	10.4 (0.9)	11.1 (1.5)	15.8 (2.0)
2	55.7 (1.3)	60.0 (1.4)	62.7 (1.6)	62.4 (2.5)
SWS	13.7 (1.2)	9.8 (1.0)	7.6 (1.1)	5.0 (1.4)
REM	22.5 (0.8)	19.8 (1.1)	18.6 (1.6)	16.7 (1.2)
Arousal index, n/h	8.1 (0.4)	13.3 (1.0)	19.7 (1.3)	38.3 (4.3)
Δ SaO ₂ , %	3.0 (0.2)	3.8 (0.2)	3.8 (0.2)	7.1 (0.9)
Metabolic Parameters				
Glucose, mg/dL	94.7 (1.2)	97.9 (1.4)	100.9 (1.9)	102.5 (2.2)
Insulin, μU/mL	9.0 (0.7)	12.9 (1.0)	11.9 (1.2)	16.4 (1.7)
D _i	1592.03 ± 1344.46	1332.07 ± 1028.33	1054.53 ± 1031.12	845.02 ± 710.70
S _i	3.72 ± 1.86	2.53 ± 1.30	2.45 ± 1.71	1.83 ± 1.23
AI _{Rg}	510.88 ± 500.32	601.02 ± 407.53	518.69 ± 378.37	527.78 ± 410.48

Values reported are either percentages or means (SE). AHI, apnea-hypopnea index; BMI, body mass index; SWS, slow wave sleep; REM, rapid eye movement; Δ SaO₂, average oxyhemoglobin desaturation; D_i, disposition index; S_i, insulin sensitivity; AI_{Rg}, acute insulin response to glucose.

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Table 1B—Sleep spectrogram variables by sleep disordered breathing severity

Spectrogram Variable	AHI, events/h			
	< 5.0 (n = 39)	5.0-14.9 (n = 34)	15.0-29.9 (n = 22)	> 30.0 (n = 23)
HFC-D	229.8 (89.9)	221.9 (75.1)	192.7 (70.8)	117.5 (85.7)
HFC-P	52.4 (17.5)	49.1 (16.4)	42.8 (14.4)	28.2 (20.3)
LFC-D	124.2 (54.2)	157.7 (63.4)	186.4 (65.9)	270.8 (120.8)
LFC-P	28.9 (11.7)	34.1 (12.5)	41.1 (12.5)	61.6 (22.1)
VLFC-D	77.7 (51.9)	70.9 (33.5)	69.8 (38.8)	42.5 (29.1)
VLFC-P	17.4 (9.9)	15.8 (7.8)	15.2 (7.7)	9.6 (6)

Data are shown as mean (SD). AHI, apnea-hypopnea index; HFC, high-frequency coupling; LFC, low-frequency coupling; VLFC, very-low-frequency coupling; D designation on spectrogram metric, duration in minutes within total sleep period; P designation on spectrogram metric, percentage of total sleep period.

or quality indices alone. HFC is proposed as a biomarker of effective sleep because, during such periods, there is temporal stability of respiration,²³ strong respiratory sinus arrhythmia suggesting vagal dominance, a paucity of EEG arousals,²³ and an association with non-CAP, during which blood pressure dipping occurs.²⁴ The EEG correlate of HFC, non-CAP, is also increased under conditions of increased expression of homeostatic sleep drive, such as during positive airway pressure titration,³⁶ use of hypnotic drugs,³⁷ and recovery sleep following sleep deprivation.³⁸ In the clinical condition of fibromyalgia,

where sleep is consistently nonrestorative, we have recently shown that HFC is reduced whereas conventional polysomnographic approaches did not differentiate patients from control subjects.²⁸ The results of this analysis further support the notion that HFC is an index of a desirable sleep state.

In the current analysis, the spectrographic frequencies correlated with standard sleep stages in physiologically meaningful ways, though only the spectrographic markers were associated with the FSIVGTT-derived measure. LFC, the spectrographic marker of unstable or “ineffective” sleep, correlated negatively,

Table 2—Correlation matrix of spectrographic and polysomnographic variables and the disposition index

Metric	TST	Stage 1	Stage 2	Stage 3+4	REM	AHI	Arl	HFC-D	HFC-P	LFC-D	LFC-P	VLFC-D	VLFC-P	D _i
TST	1													
Stage 1	-0.01 (0.884)	1												
Stage 2	-0.11 (0.222)	-0.17 (0.060)	1											
Stage 3+4	0.03 (0.744)	-0.40 (< 0.001)	-0.49 (< 0.001)	1										
REM	0.12 (0.190)	-0.30 (0.001)	-0.57 (< 0.001)	0.14 (0.139)	1									
AHI	-0.12 (0.210)	0.35 (< 0.001)	0.35 (< 0.001)	-0.467 (< 0.001)	-0.39 (< 0.001)	1								
Arl	-0.10 (0.295)	0.34 (< 0.001)	0.29 (0.002)	-0.38 (< 0.001)	-0.36 (< 0.001)	0.81 (< 0.001)	1							
HFC-D	0.29 (0.002)	-0.07 (0.438)	-0.07 (0.437)	0.17 (0.074)	0.03 (0.726)	-0.39 (< 0.001)	-0.43 (< 0.001)	1						
HFC-P	0.04 (0.685)	-0.12 (0.206)	-0.08 (0.333)	0.24 (0.009)	0.03 (0.728)	-0.42 (< 0.001)	-0.43 (< 0.001)	0.93 (< 0.001)	1					
LFC-D	0.18 (0.048)	0.23 (0.015)	0.16 (0.083)	-0.35 (0.000)	-0.17 (0.063)	0.57 (< 0.001)	0.57 (< 0.001)	-0.68 (< 0.001)	-0.84 (< 0.001)	1				
LFC-P	-0.08 (0.369)	0.20 (0.029)	0.15 (0.104)	-0.28 (0.002)	-0.20 (0.035)	0.59 (< 0.001)	0.61 (< 0.001)	-0.82 (< 0.001)	-0.87 (< 0.001)	0.93 (< 0.001)	1			
VLFC-D	0.22 (0.017)	-0.11 (0.218)	-0.09 (0.313)	0.01 (0.864)	0.31 (0.001)	-0.29 (0.001)	-0.34 (< 0.001)	-0.13 (0.169)	-0.27 (0.004)	0.03 (0.775)	-0.12 (0.186)	1		
VLFC-P	0.03 (0.777)	-0.15 (0.106)	-0.10 (0.276)	0.07 (0.473)	0.31 (0.001)	-0.33 (< 0.001)	-0.37 (< 0.001)	-0.21 (0.021)	-0.27 (0.003)	-0.07 (0.485)	-0.13 (0.161)	0.96 (< 0.001)	1	
D_i	0.06 (0.514)	-0.12 (0.192)	-0.18 (0.055)	0.026 (0.005)	0.15 (0.104)	-0.30 (0.001)	-0.19 (0.043)	0.27 (0.003)	0.25 (0.006)	-0.18 (0.046)	-0.20 (0.034)	-0.12 (0.183)	-0.13 (0.168)	1

TST refers to total sleep time; REM, rapid eye movement; AHI, apnea-hypopnea index; ArI, total arousal index; HFC, high-frequency coupling; LFC, low-frequency coupling; VLFC, very-low-frequency coupling; D_i, disposition index; P, percentage of sleep period; D, duration, in minutes, of the sleep period. Spearman's rho; P value (statistical significance of correlation) is in brackets.

whereas HFC, the spectrographic marker of stable or “effective” sleep, correlated positively with slow wave sleep, the EEG-correlate of “stable” sleep. The sleep spectrogram analysis differs from the EEG not only by its epoch length (8.5-minute windows moving in 2.1-minute steps vs 30-second epochs), but also by its relative independence from the EEG. Both stage 2 and slow wave sleep may demonstrate HFC. REM sleep correlated with both LFC and VLFC percentages. REM sleep can exhibit different characteristic on the sleep spectrogram (unpublished data). Fragmented REM sleep, such as occurs in patients with sleep apnea, demonstrates LFC. REM sleep with stable respiration, as may be seen with the use of positive pressure therapy, can demonstrate HFC characteristics. Sleep-wake transitions may show VLFC. “Average” REM sleep is usually VLFC. Thus, in a dataset enriched with sleep apnea, the observed correlations may be expected.

HFC and its EEG correlate, non-CAP, is associated with the phenomenon of blood pressure dipping,²⁴ a desired feature of

autonomic control. Interestingly, in obese, nonhypertensive, nondiabetic adolescents, the absence of nocturnal blood pressure dipping was negatively associated with altered glucose metabolism.³⁹ Because HFC was associated with increased D_i, it is possible that differences in autonomic control across state are important mediators of glycemic control. The associations with VLFC may reflect the adverse impact of wake state or sleep-wake transitions and the associated increased sympathetic activity on sleep-related glucose homeostasis and loss of biologic sleep time. The ECG-spectrogram could be especially useful for assessing the impact of sleep quality on glucose metabolism in older individuals, in whom conventional EEG-based sleep-stage assessment is limited by the relative paucity of slow wave sleep.⁴⁰

The ECG-spectrogram detects sleep fragmentation of any etiology. Any fragmenting stimulus can induce a shift to LFC. During a state of ineffective sleep, subtle fluctuations in tidal volume (that do not meet thresholds for apnea or hypopnea) are

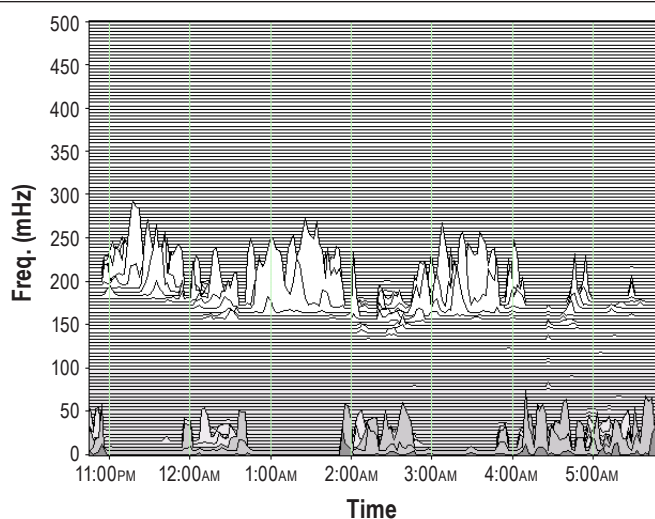


Figure 1—Good-quality sleep on the electrocardiogram-spectrogram. Note that high-frequency coupling (HFC), the upper “mountain range,” occurs more than half the night and in bursts throughout the polysomnographic sleep period (although concentrated in the first half of the night). Note also that switches from lower frequency coupling (LFC) are relatively abrupt. HFC and LFC are also clearly separated in terms of frequency. Very-low-frequency coupling (VLFC) is shown in grey in the lower spectral peaks; the distribution of VLFC is strongly reminiscent of REM-sleep distribution. Time of night is on the horizontal axis, coupling frequency on the vertical axis.

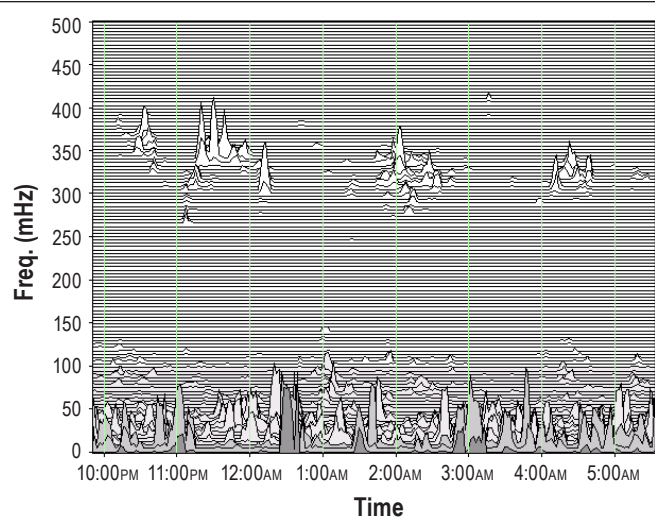


Figure 2—Poor-quality sleep on the electrocardiogram-spectrogram. Note that high-frequency coupling, the upper “mountain range,” is reduced relative to Figure 1. There is an increase in low- and very-low-frequency coupling, relative to Figure 1, across the night. Time of night is on the horizontal axis, coupling frequency on the vertical axis.

temporally coupled to EEG arousals and cyclic HRV. It would be expected that other sleep fragmenting stimuli, such as periodic limb movements, may also cause LFC if there are associated arousals. For example, it is known that periodic limb movements of sleep have a hierarchy of associated autonomic changes that are linked to the degree of EEG change⁴¹⁻⁴²—once EEG arousal occurs, LFC is expected to be induced. Arousals from sleep from any cause induce coupled transients in several physiologic streams, including blood pressure⁴³ and tidal volume;⁴⁴ these sleep-state-coupled fluctuations in tidal volume result in LFC. Our results may thus be relevant for the impact of nonapneic sleep fragmentation on glucose metabolism.

The presented analysis has some important limitations. The spectrogram analysis is not reliable in the presence of atrial fibrillation or frequent supraventricular ectopy, continuous pacing, or continuous ventricular bigeminy. The study population consisted of middle-aged, Caucasian, nonobese, healthy volunteers, with and without SDB; generalizability to other racial groups and populations may be limited, given the extensive exclusion criteria of the study from which this secondary analysis was conducted. Thus, the correlation of ECG-spectrogram measures with glucose handling in those with diabetes, ischemic heart disease, and morbid obesity, as examples of clinical importance, cannot be ascertained from this dataset.

In the full model, only a small amount of the variance in the D_i was explained by the polysomnographic and spectrographic variables. This likely reflects the heterogeneity of the population studied: nondiabetics who had a range of sleep apnea severity and body mass indices, e.g., individual differences in glucose handling. A similar assessment of the D_i in diabetic populations or in those with impaired glucose tolerance alone may provide useful insights. It is plausible that individuals may be less or

more susceptible to the adverse effects of sleep fragmentation on the D_i . It does not seem likely that sleep characteristics will be a *major* explanatory variable in glucose handling.

In conclusion, the ECG-spectrogram analysis of sleep quality may provide information beyond that obtained by conventional polysomnography in relationship to glucose metabolism.

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DISCLOSURE STATEMENT

The ECG-spectrogram software is licensed by the Beth Israel Deaconess Medical Center to Embla Inc; Robert Thomas is a co-inventor of the analysis software. The other authors have indicated no financial conflicts of interest.

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Table 3—Change in D_i in relation to ECG-spectrogram variables

Spectrogram Coupling Metric	Model 1 Unadjusted $\beta \pm \text{s.e.}$ (P)	Model 2 $\beta \pm \text{s.e.}$ (P)	Model 3 $\beta \pm \text{s.e.}$ (P)	Model 4 $\beta \pm \text{s.e.}$ (P)	Model 5 $\beta \pm \text{s.e.}$ (P)	R ² and adjusted R ² (full model)
Adjusted variables						
HFC-D	3.74 \pm 1.09 (0.001)	3.33 \pm 1.15 (0.005)	3.21 \pm 1.21 (0.009)	3.28 \pm 1.44 (0.025)	3.24 \pm 1.45 (0.027)	0.13, 0.06
Age		-14.52 \pm 8.06 (0.074)	-13.63 \pm 8.48 (0.111)	15.72 \pm 8.82 (0.078)	-14.96 \pm 8.93 (0.097)	
Sex		62.00 \pm 205.97 (0.764)	99.71 \pm 231.85 (0.668)	74.49 \pm 234.64 (0.752)	92.04 \pm 237.06 (0.699)	
BMI		-12.22 \pm 17.40 (0.484)	-10.67 \pm 18.03 (0.554)	-11.64 \pm 18.68 (0.535)	-8.91 \pm 19.26 (0.644)	
Stage 3/4			5.69 \pm 16.59 (0.732)	10.37 \pm 17.62 (0.558)	9.13 \pm 17.79 (0.609)	
TST			29.53 \pm 89.85 (0.743)	26.08 \pm 92.39 (0.778)	17.93 \pm 93.61 (0.848)	
Arl				0.73 \pm 9.13 (0.936)	7.78 \pm 14.73 (0.598)	
Stage 1				14.79 \pm 16.20 (0.363)	14.90 \pm 16.24 (0.361)	
AHI					-5.48 \pm 8.98 (0.543)	
HFC-P	14.82 \pm 5.23 (0.005)	12.17 \pm 5.57 (0.031)	12.15 \pm 5.65 (0.034)	11.34 \pm 6.64 (0.090)	11.09 \pm 6.67 (0.099)	0.12, 0.04
Age		-14.72 \pm 8.23 (0.76)	-13.15 \pm 8.61 (0.122)	-15.39 \pm 8.94 (0.088)	-14.65 \pm 9.05 (0.108)	
Sex		27.11 \pm 208.55 (0.897)	79.52 \pm 233.93 (0.735)	55.52 \pm 236.71 (0.815)	72.89 \pm 239.15 (0.761)	
BMI		-14.77 \pm 17.67 (0.405)	-11.97 \pm 18.25 (0.513)	-11.42 \pm 18.88 (0.547)	-8.71 \pm 19.46 (0.655)	
Stage 3/4			4.74 \pm 16.84 (0.779)	8.49 \pm 17.80 (0.634)	7.29 \pm 17.97 (0.686)	
TST			95.66 \pm 87.39 (0.276)	94.79 \pm 87.91 (0.283)	85.83 \pm 89.41 (0.339)	
Arl				-2.30 \pm 9.12 (0.801)	4.69 \pm 14.77 (0.751)	
Stage 1				15.29 \pm 16.36 (0.352)	15.41 \pm 16.40 (0.350)	
AHI					-5.47 \pm 9.07 (0.548)	

The β (or coefficient) is the increase or decrease in the DI per unit change in electrocardiogram-spectrogram variable. The items within the brackets are the coefficients and standard errors for the adjusted variables, in the order in the column header. P, statistical significance; DI, disposition index (insulin sensitivity [SI] \times acute insulin response to glucose [AIRg]); BMI, body mass index; HFC, high-frequency coupling; LFC, low-frequency coupling; VLFC, very-low-frequency coupling; D designation on spectrogram metric, duration in minutes within total sleep period; P designation on spectrogram metric, percentage of total sleep period.

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Table 3 (continued)—Change in D_I in relation to ECG-spectrogram variables

Spectrogram Coupling Metric	Model 1 Unadjusted $\beta \pm \text{s.e.}$ (P)	Model 2 $\beta \pm \text{s.e.}$ (P)	Model 3 $\beta \pm \text{s.e.}$ (P)	Model 4 $\beta \pm \text{s.e.}$ (P)	Model 5 $\beta \pm \text{s.e.}$ (P)	R ² and adjusted R ² (full model)
Adjusted variables						
LFC-D	-2.16 \pm 1.12 (0.055)	-1.41 \pm 1.22 (0.249)	-1.90 \pm 1.30 (0.147)	-1.75 \pm 1.83 (0.341)	-1.60 \pm 1.86 (0.391)	0.10, 0.02
Age		-16.13 \pm 8.38 (0.057)	-13.82 \pm 8.75 (0.117)	-16.10 \pm 9.02 (0.077)	-15.43 \pm 9.12 (0.094)	
Sex		-5.28 \pm 213.86 (0.980)	81.01 \pm 238.89 (0.735)	53.33 \pm 240.96 (0.825)	67.44 \pm 242.99 (0.782)	
BMI		-17.92 \pm 18.18 (0.326)	-12.27 \pm 18.81 (0.516)	-11.29 \pm 19.10 (0.556)	-8.82 \pm 19.66 (0.655)	
Stage 3/4			5.33 \pm 17.12 (0.756)	9.59 \pm 17.96 (0.594)	8.41 \pm 18.14 (0.644)	
TST			131.77 \pm 92.06 (0.155)	128.15 \pm 94.61 (0.178)	116.77 \pm 97 (0.231)	
Arl				-3.11 \pm 10.94 (0.777)	3.11 \pm 15.52 (0.842)	
Stage 1				18.35 \pm 16.65 (0.273)	18.25 \pm 16.70 (0.277)	
AHI					-5.23 \pm 9.24 (0.572)	
LFC-P	-12.09 \pm 5.43 (0.028)	-8.50 \pm 6.05 (0.165)	-8.44 \pm 6.11 (0.170)	-6.60 \pm 9.15 (0.472)	-5.67 \pm 9.31 (0.544)	0.10, 0.02
Age		-15.38 \pm 8.41 (0.070)	-13.75 \pm 8.79 (0.120)	-16.01 \pm 9.08 (0.081)	-15.37 \pm 9.17 (0.097)	
Sex		14.64 \pm 214.64 (0.946)	80.92 \pm 239.63 (0.736)	49.25 \pm 242.69 (0.840)	62.63 \pm 244.49 (0.798)	
BMI		-15.85 \pm 18.31 (0.389)	-12.48 \pm 18.87 (0.510)	-11.71 \pm 19.13 (0.542)	-9.16 \pm 19.68 (0.643)	
Stage 3/4			6.97 \pm 17 (0.682)	10.52 \pm 18.07 (0.562)	9.14 \pm 18.27 (0.618)	
TST			96.43 \pm 88.46 (0.278)	95.77 \pm 88.87 (0.284)	86.96 \pm 90.40 (0.338)	
Arl				-4.15 \pm 11.70 (0.724)	2.10 \pm 15.87 (0.895)	
Stage 1				16.81 \pm 16.55 (0.312)	16.82 \pm 16.60 (0.313)	
AHI					-5.44 \pm 9.29 (0.560)	

The β (or coefficient) is the increase or decrease in the D_I per unit change in electrocardiogram-spectrogram variable. The items within the brackets are the coefficients and standard errors for the adjusted variables, in the order in the column header. P, statistical significance; D_I , disposition index (insulin sensitivity [SI] \times acute insulin response to glucose [AIRg]); BMI, body mass index; HFC, high-frequency coupling; LFC, low-frequency coupling; VLFC, very-low-frequency coupling; D designation on spectrogram metric, duration in minutes within total sleep period; P designation on spectrogram metric, percentage of total sleep period.

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Table 3 (continued)—Change in D_i in relation to ECG-spectrogram variables

Spectrogram Coupling Metric	Model 1 Unadjusted $\beta \pm \text{s.e.}$ (P)	Model 2 $\beta \pm \text{s.e.}$ (P)	Model 3 $\beta \pm \text{s.e.}$ (P)	Model 4 $\beta \pm \text{s.e.}$ (P)	Model 5 $\beta \pm \text{s.e.}$ (P)	R ² and adjusted R ² (full model)
Adjusted variables						
VLFFV-D	-2.96 \pm 2.45 (0.229)	-3.94 \pm 2.43 (0.108)	-4.49 \pm 2.49 (0.074)	-6.17 \pm 2.63 (0.021)	-6.31 \pm 2.64 (0.018)	0.14, 0.07
Age		-18.99 \pm 8.18 (0.022)	-17.36 \pm 8.59 (0.046)	-17.28 \pm 8.80 (0.052)	-16.19 \pm 8.90 (0.071)	
Sex		-97.93 \pm 206.76 (0.637)	-31.14 \pm 234.75 (0.895)	-24.74 \pm 233.50 (0.916)	0.13 \pm 235.53 (1.000)	
BMI		-27.74 \pm 17.48 (0.115)	-24.50 \pm 18.15 (0.180)	-14.21 \pm 18.65 (0.448)	-10.37 \pm 19.20 (0.590)	
Stage 3/4			6.69 \pm 16.87 (0.692)	1.35 \pm 17.92 (0.940)	-0.55 \pm 18.08 (0.976)	
TST			122.50 \pm 89.33 (0.173)	139.07 \pm 88.77 (0.120)	127.32 \pm 89.90 (0.160)	
Arl				-16.94 \pm 8.20 (0.041)	-6.95 \pm 14.18 (0.625)	
Stage 1				13.40 \pm 16.21 (0.410)	13.47 \pm 16.22 (0.408)	
AHI					-7.73 \pm 8.96 (0.390)	
VLFFV-P	-17.01 \pm 11.94 (0.157)	-22.50 \pm 11.88 (0.061)	-21.90 \pm 11.97 (0.070)	-29.81 \pm 12.69 (0.021)	-30.91 \pm 12.75 (0.017)	0.14, 0.07
Age		-19.42 \pm 8.15 (0.019)	-17.77 \pm 8.61 (0.041)	-17.62 \pm 8.80 (0.048)	-16.45 \pm 8.89 (0.067)	
Sex		-111.83 \pm 206.39 (0.589)	-44.53 \pm 235.50 (0.850)	-39.85 \pm 234.04 (0.865)	-13.84 \pm 235.80 (0.953)	
BMI		-28.13 \pm 17.39 (0.109)	-24.70 \pm 18.16 (0.177)	-14.34 \pm 18.65 (0.444)	-10.18 \pm 19.18 (0.597)	
Stage 3/4			7.21 \pm 16.84 (0.669)	1.65 \pm 17.89 (0.927)	-0.51 \pm 18.05 (0.978)	
TST			88.89 \pm 87.92 (0.314)	93.13 \pm 86.91 (0.286)	79.17 \pm 88.23 (0.372)	
Arl				-16.95 \pm 8.20 (0.041)	-6.17 \pm 14.12 (0.663)	
Stage 1				11.94 \pm 16.26 (0.464)	11.93 \pm 16.27 (0.465)	
AHI					-8.41 \pm 8.97 (0.350)	

The β (or coefficient) is the increase or decrease in the DI per unit change in electrocardiogram-spectrogram variable. The items within the brackets are the coefficients and standard errors for the adjusted variables, in the order in the column header. P, statistical significance; DI, disposition index (insulin sensitivity [SI] \times acute insulin response to glucose [AIRg]); BMI, body mass index; HFC, high-frequency coupling; LFC, low-frequency coupling; VLFC, very-low-frequency coupling; D designation on spectrogram metric, duration in minutes within total sleep period; P designation on spectrogram metric, percentage of total sleep period.

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