Sleep Deprivation as a Probe of Homeostatic Sleep Regulation in Primary Alcoholics

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Background: *Alcoholic patients show prominent disturbances of sleep electroencephalograms (EEGs) with a marked loss of slow wave sleep that is even more profound in African American alcoholics as compared to European Americans. Using partial sleep deprivation, this study examined the extent to which abnormal sleep is reversible in alcoholic subjects.*

Methods: *In a sample stratified on ethnicity, polysomnographic and spectral sleep EEG measures were compared in male primary alcoholic in patients (*n*=46) and age-matched comparison controls (*n*=32) at baseline–and recovery sleep following a night of partial sleep deprivation.*

Results: *As compared to controls, alcoholic patients showed a loss of slow wave sleep and more spectral power in beta frequencies. Following sleep deprivation, slow wave sleep and delta power differentially changed between the groups. European American controls showed increases of slow wave sleep that were more robust than responses found in African American controls, whereas both alcoholic groups failed to show increases of slow wave sleep from baseline to recovery. Spectral EEG analyses revealed similar results; sleep deprivation induced significant increases of delta power during NREM-1 in the controls, but not in the alcoholics.*

Conclusions: *Alcohol dependence compromises the augmentation of slow wave sleep and delta power seen in healthy adults following sleep deprivation. The differential effect of alcoholism on sleep stage physiology suggests a defect in the regulation or plasticity of slow wave sleep with implications for theories linking sleep depth to morbidity and outcome in alcoholics.* Biol Psychiatry 2002;51:632–641 © *2002 Society of Biological Psychiatry*

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Introduction

Considerable evidence indicates that sleep is abnormal in alcohol dependence. Alcoholic patients commonly report sleep difficulties and this problem is one of the most refractory symptoms to resolve over the course of alcoholic recovery (Zarcone 1979; Brower et al 1998; Drummond et al 1998; Gillin et al 1994). EEG sleep studies reveal a reduction of total sleep time, fragmentation of sleep, loss of Stages 3 & 4 (delta) sleep, and less robust changes in rapid eye movement (REM) sleep in alcoholics as compared with age-matched controls (Allen et al 1971; Snyder et al 1985; Johnson et al 1970; Williams et al 1981). Age and severity of alcohol dependence correlate with abnormal sleep in alcoholics (Gillin et al 1990a). Furthermore, recent data indicate that ethnicity is a critical factor in mediating the effects of alcohol dependence on sleep; African American ethnicity and alcohol dependence interact to produce a more profound loss of delta sleep as measured by polysomnographic and spectral sleep analyses than that found in European American alcoholics (Irwin et al 2000b). The severity of sleep disturbance and the extent of sleep abnormalities in alcoholics contrasts with the limited effort to understand sleep regulatory processes in these patients. Whereas studies of depressed patients have probed sleep regulation using naturalistic strategies such as sleep deprivation (Reynolds et al 1987; Buysse et al 1988), there are no data to our knowledge that have examined the effects of sleep deprivation on EEG sleep in alcoholics.

In healthy adults, sleep deprivation leads to a selective enhancement of the slow wave sleep fraction of non-REM sleep with robust increases in stage 4 sleep (Borbély et al 1981) and delta power (Armitage 1995). Indeed, the generality of this finding across various species (e.g., human, monkey, cat, and rat) has prompted the hypothesis of a specific relationship between prior waking, sleep capacity, and slow wave sleep in which both neuronal and

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humoral mechanisms (i.e., the accumulation of possible sleep promoting substances such as cytokines) regulate sleep (Borbély 1982; Krueger et al 1993). In contrast, in depressed patients where macro and micro-archtitectural analyses support the notion of delta sleep deficits as compared to controls, depressives show a diminished augmentation of delta sleep following sleep loss (Reynolds et al 1987; Buysse et al 1988). Whether alcoholics show similar impairments in the plasticity of slow wave sleep in concert with delta sleep deficits at rest are not known.

This study examined the effects of partial sleep deprivation as a probe on sleep-stage physiology in controls, contrasted with alcoholic patients. The following hypotheses were tested: a) alcoholics will show macro- and micro-architectural evidence of a "decay" of delta sleep as compared to controls at baseline; b) sleep deprivation, as a naturalistic probe of sleep regulatory processes, will reveal a loss of delta sleep plasticity in alcoholics as compared to controls.

Methods and Materials

Participants

A total of 99 men fulfilled screening eligibility criteria, gave informed consent and entered the research protocol. Of this total, 6 participants were excluded due to medical history and/or medication use, 2 controls and 4 alcoholics were dropped due to positive toxin screens during the sleep protocol or within the two week period prior to assessment, 7 subjects were excluded due to nocturnal myoclonus, and 2 alcoholics were dropped because of elevated oximetry. The remaining sample was comprised of 78 men stratified into four groups: European American control subjects (n=18), African-American controls (n=14), European American alcoholic patients (n=24) and African-American alcoholic patients (n=22). The sample was stratified on the basis of African-American vs. European American ethnicity in view of recent observations that African American alcoholics show more severe sleep abnormalities (Irwin et al 2000b). Alcoholic patients were clinically hospitalized for 2 weeks in the Alcohol and Drug Treatment Program (ADTP) at the Veterans Affairs San Diego Healthcare System (VASDHS) before testing in the current research study. Nonpatient controls were identified though a standardized search strategy of the San Diego area involving the placement of flyers and advertisements in local newspapers. In addition, a direct mailing to the San Diego County veterans population was used to target controls who were age-matched (± 5 years) and had sociodemographic characteristics similar to alcoholic patients.

Research diagnoses of controls and alcoholic patients were made following the administration of a semi-structured interview developed by the multi-site Collaborative Study on the Genetics of Alcoholism (Bucholz et al 1994). This interview assessed consumption- and associated problem histories for cigarettes, alcohol, and other substances and assessed diagnostic criterion data to evaluate the presence of a lifetime history of a psychiatric and substance dependence disorder. Control subjects fulfilled DSM-IV criteria for Never Mentally Ill (APA 1994). Alcoholic patients fulfilled DSM-IV criteria for alcohol dependence that had occurred in the absence of major pre-existing psychiatric disorders; in other words, alcoholics had primary alcohol dependence (Schuckit 1985; APA 1994). Since we have previously reported that primary alcoholic patients with a history of secondary depression show sleep abnormalities compared to alcoholics without such depression (Gillin et al 1990b), alcoholics with current or independent DSM-IV major depressive disorder were also excluded. Severity of depressive symptoms was evaluated on the day prior to beginning the sleep protocol using the 17-item Hamilton Rating Scale - Depression (HRS-D; test-retest reliability of .94 on total scores)(Endicott et al 1981).

All participants were free of physical illness by examination. Alcoholics were excluded if they had histories of overt alcohol-related liver disease such as jaundice and esophageal varices. Prior to entry into the study, medication histories were obtained and subjects who were taking medications known to alter sleep wake activity (e.g., β-blockers, nonsteroidal anti-inflammatory agents, steroids) were also excluded. In addition, all alcoholic participants were free of psychotropic medications for greater than two weeks prior to EEG sleep assessment.

This sample of alcoholics was studied after acute- and subacute withdrawal symptoms had resolved; i.e., after at least two weeks of abstinence while hospitalized (mean days since last drink is shown in Table 1). In four alcoholic patients, severity of withdrawal symptoms was treated 30 days prior to admission with diazepam; all other alcoholic patients denied use of benzodiazepine medications during detoxification. During the two-week interval between admission and sleep evaluation, alcoholic subjects participated in an inpatient sober treatment milieu that involved Alcoholics Anonymous, education, and group and individual counseling. Nursing observations and random urine toxin screens were used to confirm abstinence during treatment.

Procedures

A five-night sleep protocol was utilized in this study. The first night involved adaptation to the conditions of the sleep laboratory. During this adaptation night, recordings of oxygen desaturation and tibial myoclonus were obtained to exclude subjects with either sleep apnea or nocturnal myoclonus. The cut-off criteria for sleep apnea was set at 15 events per hour in which an event was defined by a desaturation of 4% or greater for 10 seconds or greater. The cut-off criteria for nocturnal myoclonus was set at 10 movements per hour with an arousal defined as 3 seconds of EEG arousal. On the second night (baseline night), sleep EEGrecordings were obtained during an uninterrupted period of nocturnal sleep. Sleep data from this night has been reported previously (Irwin et al 2000b). On the third, fourth, and fifth nights, sleep recordings were obtained along with serial blood samples; blood sampling data from these nights will be reported separately. On the third night, subjects were allowed to sleep from 23.00 h to 6.30 h. On the fourth night, partial sleep deprivation-early night (PSD-E) was administered and subjects were kept awake from 23.00 h to 3.00 h with sleep time from

Table 1. Age, Education, Severity of Depressive Symptoms, and Alcohol Consumption Histories in Control and Alcoholic Groups

	Control Subjects				Alcoholic Subjects			
	European American (n = 18)		African American (n = 14)		European American (n = 24)		African American (n = 22)	
	Mean	(SD)	Mean	(SD)	Mean	(SD)	Mean	(SD)
Age (years)	48.6	(11.8)	39.1	(7.5)	43.3	(11.0)	42.9	(7.0)
Education (years)	16.0	(1.5)	14.3	(1.5)	12.8	(1.7)	13.0	(1.0)
Depressive symptoms (HRSD scores)	1.6	(1.8)	1.3	(1.9)	1.8	(2.0)	1.0	(1.5)
Alcohol consumption (last 3 months)								
Drinking days/month	8.4	(8.0)	4.3	(7.7)	27.3	(4.4)	26.9	(5.5)
Drinks/day	1.4	(1.0)	1.1	(.9)	13.1	(5.8)	15.3	(12.0)
Days since last drink	45.8	(116.3)	110.6	(167.1)	22.4	(12.0)	23.0	(12.2)

HRSD, Hamilton Rating Scale—Depression.

3.00 h to 6.30 h. On the fifth night, subjects were allowed uninterrupted recovery sleep from 23.00 h to voluntary awake time.

On each of the experimental nights, participants arrived at the laboratory at about 20.00 h and electrodes were placed for EEG, electrooculography, and submental electromyography recordings. Lights were turned off at around 23.00 h. Sleep EEG measures were obtained during continuous recordings between 22.00 h and 6.30 h during the each of the experimental nights. Sleep EEG data obtained from the adaptation-, night 02 baseline-, and night 04 PSD-E nights were not used in the analyses. While some studies have found that the introduction of an intravenous (IV) catheter and blood sampling may induce a "lightening" of sleep, we found no differences of sleep EEG measures between night 02 (baseline without blood sampling) and night 03 (baseline with blood sampling). While some studies have found that the introduction of an intravenous (IV) catheter and blood sampling may induce a "lightening" of sleep, we found no differences of sleep EEG measures between night 02 (baseline without blood sampling) and night 03 (baseline with blood sampling). MANOVA covarying for age, education, and depressive symptoms showed no night differences for measures of sleep continuity (i.e., total sleep time, sleep efficiency, sleep latency; $F(3,69) = .67$, $p = .6$), sleep architecture (i.e., duration of Stages 1 - 4 sleep and REM sleep; $F(5,67) = .59$, $p = .7$), and REM measures (REM latency, REM density, and REM duration; $F(3,69) = .33$, $p = .8$). In addition, duration and percentage of delta sleep did not differ between the baseline 02- and baseline IV-03 nights across the four groups ($F(1,71) = .79$, $p = .4$; $F(1,71) = .39$, $p = .5$). Thus, the present study compared the night 03 baseline and night 05 recovery, since both of these nocturnal periods were identical in their inclusion of an IV blood sampling protocol along with recording of EEG sleep.

Sleep records were visually scored according to the criteria of Rechtschaffen and Kales (Rechtschaffen et al 1968). Data from each 30-second epoch were entered into a computer program that tallies the summary statistics for each subject. Sleep onset was defined as the first minute of stage 2 or rapid eye movement (REM) sleep followed by at least eight minutes of sleep in the next nine minutes. A REM period was defined by not less than three consecutive minutes of REM sleep. Sleep efficiency was the ratio of total sleep time to the time in bed, multiplied by 100. Sleep architecture was defined as the duration of time spent asleep in non-REM (NREM) sleep, Stages 1 through 4. REM density was an estimate of the number of eye movements per minute of REM sleep, scored on a scale of 0 to 4 per 30- second epoch but expressed on a scale of 0 to 8 per minute. Sleep research technicians were regularly tested on scoring reliability and high standards were maintained: sleep latency ($r = .96$), REM latency ($r = .99$), REM density ($r = .91$), amounts of stages 3 and 4 ($r = .85$), and total sleep time ($r = .99$).

To quantify EEG frequency characteristics, the sleep EEG, from sleep onset to good morning time, was digitized (128 Hz) and power spectra for 4 sec epochs were then determined for a .25-64.0 Hz range. The transformed data were then further compressed into 6 frequency bands (.75-4.5 Hz [Delta], 4.5-7.5 Hz [Theta], 7.5-11 Hz [Alpha], 11-12.5 Hz (Slow spindle frequencies], 12.5 - 16 Hz [fast spindle frequencies], 16-40 Hz [Beta]) and mean power density (microvolts^2/octave), and peak frequency (Hz) was calculated for each band as previously described (Ehlers et al. 1989; Ehlers et al. 1998; Irwin, et al. 2000b). Power in each frequency band was determined for the following five epochs after good night time (1) first NREM period; (2) first REM period; (3) second NREM period; (4) second REM period; and (5) the time from good night time to good morning time (whole night). Of the total sample (n=78), 71 subjects aged 27-65 years) (African American n=35 vs. European Americans, n=36; alcohol dependent subjects n=46 vs. controls n=25) had electronic EEG data suitable for EEG spectral analyses.

LIVER FUNCTION TESTS. Values of liver function tests were obtained and measured by previously described methods (Irwin et al 1990).

Statistical Analyses

Two way analyses of variance (ANOVA) were used to evaluate main effects of diagnosis (alcohol dependence, control), and ethnicity (European American, African-American) on the dependent variables (i.e., age, education, depressive symptoms, alcohol consumption history, and liver function tests). In view of the multivariate experimental approach in which multiple dependent sleep variables were collected simultaneously in a repeated

Table 2. Liver Function Tests in the Control and Alcoholic Groups

	Control Group				Alcoholic Group			
	European American (n = 19)		African American (n = 14)		European American (n = 24)		African American (n = 22)	
	Mean	(SD)	Mean	(SD)	Mean	(SD)	Mean	(SD)
SGOT (U/L)	26.1	(5.4)	27.8	(9.0)	43.2	(44.2)	35.6	(23.4)
SGPT (U/L)	28.3	(11.0)	30.1	(12.6)	49.2	(42.9)	38.4	(29.1)
Bilirubin, total (μmol/L)	.8	(.2)	.9	(.3)	.8	(.5)	.6	(.4)
Bilirubin, direct (μmol/L)	.1	(.19)	.1	(.04)	.2	(.16)	.1	(.1)
Alkaline phosphatase	65.9	(14.1)	63.4	(11.7)	78.0	(20.5)	70.3	(23.6)

SGPT, serum glutamis-oxaloacetic transaminase; SGOT, serum glutamic-pyruvic transaminase.

measures design, multivariate ANOVAs (MANOVA) were employed as a first step in the analyses of the sleep data. Within each of the three domains of sleep including sleep continuity (total sleep time, sleep latency, and sleep efficiency), sleep architecture (duration of Stages 1-4 and REM sleep), and REM measures (REM latency, density and REM duration), main effects of diagnosis, ethnicity, night and their interaction were tested by MANOVA covarying for age, education, and depressive symptoms. These covariates were selected because the groups differed in these variables and/or the covariate has been found to correlate with sleep disturbance. For those sleep domains in which main- and/or interaction effects were found by the MANCOVA, subsequent three-way (ANCOVAs) were performed on the individual sleep EEG variables and spectral EEG results. For the spectral EEG results in which only a subset of the total sample was available for analyses, statistical power was limited. Thus, planned comparisons were also used to test group differences in the spectral analytic measures at baseline- or recovery nights, when main effects by ANOVA and/or a priori predictions generated by analyses of sleep EEG data supported this exploratory statistical approach.

Results

Demographic and Clinical Characteristics

Table 1 summarizes the demographic characteristics, severity of depressive symptoms and alcohol consumption histories in the control- and alcoholic groups. The alcoholic and control groups were similar in age, whereas the African-American groups were younger than the European Americans ($F(1,74)=4.9$, $p<.05$). For education, there were main effects for alcohol dependence in which both alcoholic groups reported fewer years of education than controls ($F(1,74)=43.5$, $p<.001$), and for ethnicity in which the African-Americans reported less years of formal schooling than the European Americans ($F(1,74)=4.3$, $p<.05$) . Comparison of other demographic variables showed that the controls were more likely to be currently employed (European American 56%; African American 86%) as compared to the alcoholics (European American 21%; African American 9%; $\chi^2= 26.9$, $p<.001$). Marital status also differed between the four groups ($\chi^2= 25.9$, $p<.05$) with alcoholics more frequently divorced or separated than controls.

Severity of depressive symptoms did not differ in relation to alcoholism or ethnicity; all groups showed Hamilton scores within the non-depressed normal range. Alcohol consumption histories over the last three months showed that alcoholics reported more drinking days per months ($F(1,74)=202.8$, $p<.001$), more drinks per drinking day ($F(1,74)=61.2$, $p<.001$), and more recent use of alcohol than controls ($F(1,74)=7.2$, $p<.001$). Nevertheless, average days since last drink showed that the alcoholic groups were abstinent for longer than three weeks before sleep assessment. Importantly, the two alcoholic groups stratified by ethnicity reported nearly identical levels of alcohol consumption. Use of other substances, including marijuana, stimulants, and barbiturates, was infrequent in this sample. Four European American alcoholics (16%), and 6 African American alcoholics (27%) reported marijuana use in the last 3 months, whereas 2 European American alcoholics (8%) and 9 African American alcoholics (41%) reported psychostimulant use in the last 3 months. No alcoholic subject had used these substances in the last 2 weeks. In the controls, no subject reported marijuana or psychostimulant drug use during the last three months.

Table 2 presents mean values of laboratory tests of liver injury in the control and alcoholic groups. In this sample of medically healthy, detoxified alcoholics, only values of aspartate aminotransferase (SGPT)($F(1,74)=4.5$, $p<.05$) and alkaline phosphatase ($F(1,74)=4.6$, $p<.05$) were elevated in the alcoholic groups as compared to the controls, whereas other measures of liver function including alanine aminotransferase (SGOT) and bilirubin (direct and total) were statistically indistinguishable between the four groups. Importantly, mean values for all of the liver injury tests were within the normal range in the four groups.

EEG Sleep

Table 3 shows the EEG sleep measures in the four groups who were sampled across the two nights. For the sleep

Table 3. Sleep EEG Measures in the Control and Alcoholic Groups

	Control Subjects				Alcoholic Patients			
	European Americans (n = 18) Mean (SD)		African Americans (n = 14) Mean (SD)		European Americans (n = 24) Mean (SD)		African Americans (n = 22) Mean (SD)	
	Baseline	Recovery	Baseline	Recovery	Baseline	Recovery	Baseline	Recovery
Sleep continuity								
Total sleep time (min)	337.2 (65.2)	385.0 (45.9)	348.8 (66.1)	373.3 (52.5)	332.7 (53.2)	377.5 (49.3)	343.3 (45.5)	386.9 (29.4)
Sleep efficiency (%)	76.4 (14.6)	85.8 (8.0)	83.2 (9.0)	86.2 (10.5)	75.7 (11.5)	84.2 (9.4)	80.2 (9.4)	84.4 (6.3)
Sleep latency (min)	26.4 (33.0)	11.0 (12.2)	10.9 (9.1)	9.6 (8.4)	21.0 (16.2)	12.9 (8.5)	22.8 (27.2)	18.2 (16.3)
Sleep architecture								
Stage 1 (min)	27.0 (11.5)	21.1 (12.0)	22.4 (13.1)	23.0 (13.2)	32.5 (16.6)	29.9 (15.9)	32.1 (13.1)	31.2 (21.0)
(%)	8.1 (3.3)	5.6 (3.4)	6.4 (3.5)	6.3 (3.5)	10.3 (5.8)	8.3 (5.0)	9.4 (3.7)	8.0 (5.4)
Stage 2 (min)	216.6 (50.5)	239.3 (38.8)	245.4 (48.2)	250.8 (56.9)	193.5 (33.3)	223.3 (32.0)	220.6 (32.4)	243.3 (30.8)
(%)	64.1 (8.0)	62.4 (8.8)	70.7 (7.3)	66.6 (10.5)	58.5 (7.4)	59.5 (6.9)	64.6 (7.7)	62.9 (6.7)
Stage 3 (min)	21.8 (15.9)	32.0 (21.8)	17.5 (14.3)	21.5 (21.8)	16.6 (17.5)	24.0 (18.3)	7.6 (8.7)	14.1 (16.8)
(%)	6.4 (4.6)	8.2 (5.2)	5.1 (4.2)	6.2 (6.8)	4.6 (4.6)	6.1 (4.5)	2.1 (2.4)	3.6 (4.2)
Stage 4 (min)[a]	3.9 (7.4)	11.6 (16.6)	3.5 (6.0)	4.7 (10.1)	5.2 (15.2)	4.6 (9.4)	2.0 (8.9)	.59 (1.4)
(%)[a]	1.2 (2.5)	3.0 (4.4)	.98 (1.6)	1.4 (3.0)	1.4 (4.0)	1.1 (2.2)	.52 (2.4)	.15 (.35)
Delta (min)[a]	25.7 (21.2)	43.5 (32.3)	21.0 (19.1)	26.1 (31.4)	21.8 (27.2)	28.5 (24.7)	9.6 (15.2)	14.7 (18.0)
(%)	7.6 (6.6)	11.2 (8.2)	6.1 (5.4)	7.6 (9.6)	6.0 (7.2)	7.2 (5.9)	2.6 (4.1)	3.8 (4.5)
REM (min)(%)	67.9 (23.6)	81.0 (22.4)	60.0 (25.8)	73.4 (24.0)	84.9 (27.5)	95.8 (30.9)	81.0 (26.9)	97.8 (19.5)
(%)	20.1 (6.3)	20.8 (4.7)	16.8 (4.9)	19.6 (5.0)	25.2 (6.3)	25.0 (6.3)	23.3 (6.2)	25.4 (5.4)
REM Measures								
Latency (corrected)	77.6 (47.6)	70.9 (36.1)	74.7 (28.6)	68.8 (22.4)	66.0 (38.4)	56.1 (23.8)	51.8 (31.7)	53.3 (27.2)
Density	1.7 (1.0)	1.6 (.73)	1.3 (.85)	1.4 (1.0)	2.0 (.82)	2.0 (.76)	1.6 (.88)	1.6 (.71)
Duration (1st period)	22.7 (12.4)	19.9 (9.5)	14.0 (7.5)	18.1 (9.1)	21.1 (9.7)	27.6 (17.6)	18.2 (9.6)	18.6 (9.0)

REM, rapid eye movement.
[a] $p < .05$ group × time interaction.

continuity variables (total sleep time, sleep efficiency, and sleep latency), the MANCOVA found significant effects for age ($F(3,69)=5.3$, $p<.01$) and depressive symptoms ($F(3,69)=3.4$, $p<.05$), but there no main effects for alcohol dependence, ethnicity, night, or their interactions. Thus, subsequent three-way ANCOVAs were not performed on the individual sleep continuity measures.

For the sleep architecture domain (duration of Stages 1 - 4 sleep and REM sleep), multivariate analyses showed main effects for age ($F(5,67)=3.0$, $p<.05$), diagnosis ($F(5,67)=4.0$, $p<.01$), and ethnicity ($F(5,67)=2.9$, $p<.05$). In addition, the MANCOVA identified a trend for the interaction of alcohol dependence x night ($F(5,67)=2.1$, $p=.07$) and ethnicity x night ($F(5,67)=2.2$, $p=.06$).

In view of these significant main effects on the sleep architecture domain, subsequent ANCOVAs were performed on the duration and percentage of Stages 1-4, delta, and REM sleep. Analyses of the sleep architecture variables revealed main effects of alcohol dependence on duration and percentage of Stage 1- ($F(1,71)=6.5$, $p<.01$; $F(1,71)=6.5$, $p<.01$), percentage of Stage 2- ($F(1,71)=3.9$, $p=.05$), duration and percentage of Stage 3-($F(1,71)=4.1$, $p<.05$; $F(1,71)=5.5$, $p<.05$), delta - ($F(1,71)=3.2$, $p=.08$; $F(1,71)=4.2$, $p<.05$) and REM sleep ($F(1,71)=8.2$, $p<.01$; $F(1,71)=11.9$, $p<.001$). The alcoholic groups showed increases in Stage 1 and REM sleep and decreases of Stage 2-, Stage 3- and delta sleep as compared to the controls. In addition, significant main effects of ethnicity were found for duration and percentage of Stage 2 ($F(1,71)=5.0$, $p<.05$; $F(1,71)=11.3$, $p<.001$), Stage 3- ($F(1,71)=8.8$, $p<.01$; $F(1,71)=6.5$, $p<.01$), and duration and percentage of delta sleep ($F(1,71)=7.8$, $p<.01$; $F(1,71)=5.7$, $p<.05$) in which European American groups showed greater amounts of Stage 3- and delta sleep as compared to African Americans.

Interactions between alcohol dependence, ethnicity and night were also found. There were alcohol dependence x night and ethnicity x night interactions found for measures of Stage 4 sleep duration ($F(1,71)=7.9$, $p<.01$; $F(1,71)=8.8$, $p<.01$). Finally, there was a differential change of slow wave sleep from baseline to recovery nights across the four groups; significant alcohol dependence x ethnicity x night interactions were found for duration and percentage of Stage 4 sleep ($F(1,71)=4.6$, $p<.05$; $F(1,71)=3.4$, $p= .06$; Figure 1) European American controls showed a more robust increase of Stage 4 from baseline to recovery night as compared to African Americans, whereas Stage 4 sleep were unchanged or decreased in the two alcoholic groups. A similar pattern of results was also found for duration of delta sleep, although the interaction between alcohol dependence x ethnicity x

Table 4. Electroencephalogram Beta Activity (16–40 Hz) in Recovery Sleep in Control and Alcoholic Groups

	1st NREM		1st REM		2nd NREM		2nd REM		Whole Night	
	Mean	(SD)	Mean	(SD)	Mean	(SD)	Mean	(SD)	Mean	(SD)
Alcoholic Patients $n = 46$	4.6	(.3)	8.1	(.7)	4.7	(.4)	6.9	(.7)	6.3	(.3)
Control Subjects $n = 23$	3.7	(.4)	5.0	(.9)	4.0	(.3)	4.8	(1.0)	4.7	(.5)

NREM, non-rapid eye movement.

night did not reach statistical significance ($F(1,71)=3.2$, $p= .07$).

For the REM sleep measures (REM latency, REM density, and REM duration), the MANCOVA showed a significant effect for age ($F(5,67)=5.0$, $p<.01$), but no main effects for alcohol dependence, ethnicity, night, or their interaction.

EEG Spectral Analyses

Spectral analyses of the sleep EEG could be accomplished in 71 subjects, and alcoholics were found to have several spectral indicators of disturbed sleep both on their baseline sleep night as well as on their night of recovery from sleep deprivation. Significant main effects for alcohol dependence were seen for both NREM and REM sleep. For example, alcoholics as a group were found to have more power in the beta frequency range over the whole night's sleep ($F(1,132)=8.7$, $p<.01$), and planned comparisons revealed that this was true both at baseline ($F(1,65)=3.6$, $p=.06$) and significantly so during recovery night ($F(1,65)=5.6$, $p<.05$ Table 4). As seen in Table 4, beta power was also increased in alcoholics ($F(1,130)=10.4$; $p<.01$) and especially in their recovery sleep during REM-1 ($F(1,63)=7.6$; $p<.01$). However, there were no alcohol dependence x ethnicity x night interactions for beta power.

Spectral analyses also revealed that delta power was disturbed as a function of alcohol dependence. Alcoholics as group had lower delta power ($F(1,131)=4.2$; $p<.05$). As a whole African Americans also had lower delta power ($F(1,131)=11.05$; $p<.001$). Prior studies have found that sleep deprivation induces a significant increase of delta power, particularly during the first NREM period in young adult populations (Borbély et al 1999). In the present study, spectral analyses confirmed these findings; sleep deprivation induced significant increases of delta power . during NREM-1 in the controls but not in the alcoholics (alcohol dependence x night interaction; $F(1,131)=5.5$; $p<.05$; Figure 2). There were no ethnicity x night or diagnosis x ethnicity x night interactions for delta power.

The ratio of delta power has been suggested to be an important measure of the distribution of delta sleep over the night and has been associated with relapse in depressive patients (Kupfer et al 1990). In this study, delta ratio was found to be lower in alcoholics overall ($F(1,131)=6.1$; $p<.05$) and in African Americans ($F(1,131)=8.5$; $p<.01$). Planned comparisons revealed that while alcoholics did not differ from controls in their delta ratios on the baseline night, alcoholics were significantly lower than controls on the recovery night ($F(1,64)=9.0$ $p<.01$). Thus, following sleep deprivation, alcoholics showed decreases of delta ratio, whereas controls were unchanged.

Another indicator suggesting that the distribution of sleep over the night is disturbed in alcoholics is the finding that alcoholics have increased power during their recovery night in REM-2 in both the delta ($F(1,64)=6.15$; $p<.01$), spindle ($F(1,64)=4.03$; $p<.05$) and total power ($F(1,64)=7.03$; $p<.01$) frequencies when compared to controls (Table 5). This finding suggests that "recovery" from sleep deprivation in alcoholics may be delayed to

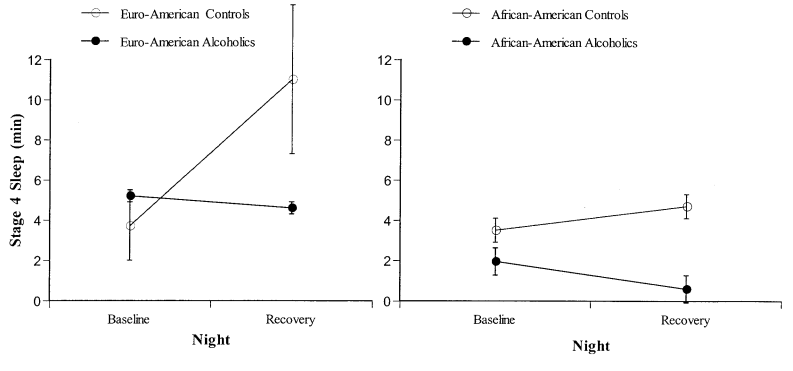

Figure 1. Effects of partial night sleep deprivation on duration of Stage 4 sleep in control- and alcoholic subjects stratified by European American and African-American ethnicity. A significant alcohol dependence x ethnicity x night interaction was found ($F(1,71)=4.6$, $p<.05$) in which European American controls showed a more robust increase of Stage 4 from baseline to recovery night as compared to African Americans, whereas Stage 4 sleep was unchanged or decreased in the two alcoholic groups.

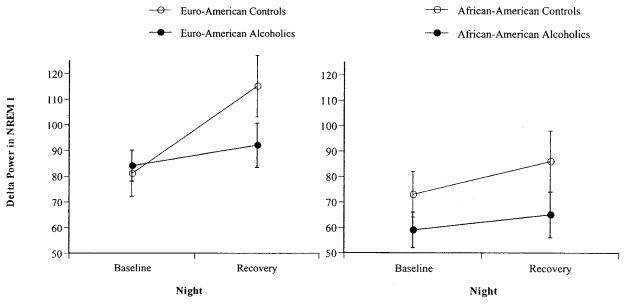

Figure 2. Effects of partial night sleep deprivation on delta power during the first NREM period in control- and alcoholic subjects stratified by European American and African-American ethnicity. Sleep deprivation induced significant increases of delta power during NREM-1 in the controls but not in the alcoholics (alcohol dependence x night interaction; F (1,131)=5.5; $p<.05$). NREM, nonrapid eye movement sleep.

later in the night, or may reflect a difference in the architecture of REM sleep.

Discussion

Electrophysiological measures in sleeping and waking have been demonstrated to be good indices of risk for alcoholism. Many, although not all (Emmerson et al 1987), studies in alcoholics have found that their resting, wakeful EEG was deficient in alpha activity, was of lower voltage, and tended to contain theta and excessive fast activity (Naitoh 1973; Kaplan et al 1985; Spehr et al 1985; Krauss et al 1991; Pollock et al 1992). It has been suggested that EEG fast frequency activity may also be higher in subjects with alcoholic relatives (Gabrielli et al 1982; Ehlers et al 1990) and related to risk for relapse in abstinent alcoholics (Bauer 1994, 2001; Winterer et al 1998). No previous studies have evaluated EEG fast activity during sleep in alcoholics. In the present study an excess of EEG fast activity was found in alcoholics during their sleep over the whole night and specifically during REM sleep. In a previous study of a large sample of normal adults, it was demonstrated that spectral power distributions during waking were substantially correlated to those seen during REM sleep (Ehlers et al 1998). Taken

Table 5. Spectral Power during the Second REM Period of Recovery Sleep in Control and Alcoholic Groups

	Control Subjects		Alcoholic Patients	
	Mean	(SD)	Mean	(SD)
Delta power (.5–4.5 Hz)	22.6	(4.2)	35.6	(3.0)
Spindle power (11–16 Hz)	7.6	(1.2)	10.5	(.8)
Total power (.5–40 Hz)	15.5	(2.2)	22.7	(1.5)

REM, rapid eye movement sleep.

together, these data suggest that EEG fast activity may be a spectral signature of alcoholics both during their sleep and waking activity.

The present study also replicates prior sleep EEG studies in alcoholics (Gillin et al 1990a; Gillin et al 1994; Irwin et al 2000b). As compared to controls, alcoholics are more likely show alterations in sleep architecture measures with a relative loss of slow wave sleep and increases in the duration of REM sleep. Moreover, consistent with our recent findings, African-American alcoholics show the greatest loss in measures of slow wave sleep as compared to the other groups (Irwin et al 2000b). Finally, a relatively new measure has been developed to estimate the distribution of delta power during the first part of the night, namely delta ratio or the amount of power in the first NREM period divided by the amount of power in the second (NREM-1/NREM-2). In the present study, not only do alcoholics show shifts in power and in their delta patterns, but the distribution of delta power over the night is altered particularly during the recovery night. Delta ratio was found to correlate with poor outcome in a study of depressed patients discontinued from long-term maintenance treatment for depression (Kupfer et al 1990).

The main aim of our study was to use sleep deprivation and evaluate the extent to which alcoholism-related "decay" of slow wave sleep is reversible. Compared with controls, alcoholic patients failed to show increases of slow wave sleep following sleep deprivation. Differences in homeostatic regulation of slow wave, Stage 4 sleep

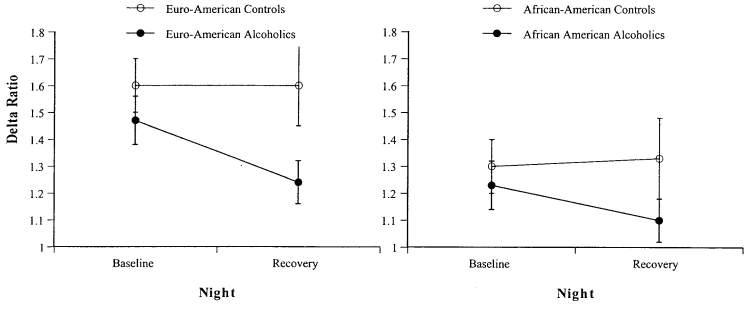

Figure 3. Effects of partial night sleep deprivation on delta ratio or ratio of delta power during the first NREM period compared to the second period (NREM-1/NREM-2) in control- and alcoholic subjects stratified by European American and African American ethnicity. Delta ratio was found to be lower in alcoholics overall (F (1,131)= 6.1; $p<.05$) and in African Americans (F (1,131) =8.5; $p<.01$). Planned comparisons revealed that while alcoholics did not differ from controls in their delta ratios on the baseline night, alcoholics were significantly lower than controls on the recovery night (F (1,64)=9.0 $p<.01$) NREM, nonrapid eye movement sleep.

were particularly striking in the European Americans; controls showed nearly two-fold increases of Stage 4 sleep whereas alcoholics had recovery levels of Stage 4 sleep that decreased. Similar findings were found in the comparison of the African-American control and alcoholic groups; although African American controls showed less robust increases of Stage 4 sleep than European Americans. These data, taken together with macro- and microarchitectural evidence of delta sleep loss in alcoholics, further implicate abnormalities of delta sleep and its regulation in alcohol dependence. Loss of slow wave sleep is a robust characteristic of alcoholic sleep disturbance that sleep deprivation did not ameliorate. These are the first data, to our knowledge, that reveal a loss of plasticity in the regulation of slow wave sleep in alcoholic subjects. Decreases of slow wave sleep are major effects of healthy aging and are also associated with major depression. Using total sleep deprivation, Reynolds et al (1987) found that depression compromises the augmentation of slow wave sleep seen in healthy elderly. However, even in elderly depressed subjects, there was a significant consolidation of sleep with increases in slow wave sleep after sleep deprivation. In contrast, alcoholics show a complete lack of reversal of slow wave sleep following partial sleep deprivation. Taken together, these findings suggest that there is an impairment of non-REM sleep generating mechanisms in alcoholics that can not be ameliorated by sleep homeostatic factors.

The results of this sleep deprivation experiment are relevant for models of sleep regulation. For example, the "two process" model proposes that sleep is regulated by two factors: "process S", a homeostatic, sleep-dependent factor that increases in strength with the duration of prior wakefulness and that is expressed in slow wave sleep activity; and "process C" a circadian sleep factor expressed in REM sleep propensity (Borbély 1982; Borbély et al 1982). The present data suggest that the accumulation of "process S" is diminished or inhibited in alcohol dependence, which in turn leads to lower amounts of delta sleep and increased amount of REM sleep. Even when the duration of wakefulness is prolonged, "process S" does not appear to be augmented in the alcoholic subjects. The failure to accumulate "process S" might contribute to the persistent severity of sleep disturbances in recovering alcoholics, and identify those recovering alcoholics whose sleep complaints fail to resolve and who may be at increased risk of relapse (Brower et al 1998; Gillin et al 1994; Drummond et al 1998). An alternative explanation for the lack of delta rebound, which is separate from a possible failure to generate "process S", suggest that the physiological mechanisms responsible for the generation of delta waves has been damaged as discussed below. Nevertheless, because sleep depth is hypothesized to contribute to the maintenance of health and the homeostatic regulation of the autonomic, neuroendocrine and immune systems, (Horne 1988; Dinges et al 1995) studies are underway to evaluate whether alcoholics are at increased risk for physiological abnormalities in association with disordered sleep.

The neurobiological mechanisms that underlie the defect in the generation of slow wave sleep in association with chronic alcohol exposure remain largely unknown. Steriade and colleagues have shown that during slow-wave sleep (SWS), neocortical neurons display long-lasting periods of silence, whereas they are tonically active and discharge at higher rates during waking and sleep with rapid eye movements (REMs) (Timofeev et al 2001). It is possible that chronic alcohol use interferes with the process of neuronal hyperpolarization that is mediated by a mixture of $K+$ currents and "disfacilitation" processes that together induce periods of silence (Timofeev et al 2001). Indeed, clinical studies support the notion that chronic ethanol exposure causes direct toxic or neuroadaptive changes in the brain areas that are involved in the regulation of sleep. For instance cerebral atrophy and slow wave sleep have been correlated in abstinent chronic alcoholics (Ishibashi et al 1987). Other work suggests that the complex cytokine network is one system that might contribute to the declines of sleep depth in alcoholics; African American alcoholics have a profound loss of delta sleep (Irwin et al 2000b) that is coupled with alterations in the nocturnal expression of the pro-inflammatory cytokines and a relative shift from T helper 1 to T helper 2 cytokine production across the night as compared to responses in European American alcoholics (Irwin et al 2000a). Animal studies indicate that such cytokine abnormalities can interfere with the depth of sleep (Krueger et al 1994). Regardless of the possible mechanisms, we conclude that alcoholic subjects show abnormal sleep physiology. The amount and plasticity of slow wave sleep and delta power is severely diminished in alcoholics. In contrast, the amounts and maintenance of sleep can be augmented in alcoholics by restricting time in bed.

Limitations of this study include small sample size and gender. Thus, the findings may not apply to other community samples or to female alcoholic patients. In addition, the sleep protocol focused on the effect of sleep loss during the early part of the night, a time of high propensity for slow wave sleep and a low propensity for REM sleep. The interplay between timing of the sleep deprivation, total sleep deprivation and respective effects on non-REM and REM sleep recovery requires further study. Despite these limitations, this report represents an important step in an ongoing investigation to determine the mechanisms underlying disturbed sleep associated with substance use disorders in this understudied ethnic group.

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