Increased Production of Evoked and Spontaneous K-complexes Following a Night of Fragmented Sleep

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Study objectives: To determine whether K-complex production is better interpreted as being an arousal response or reflective of a sleep protective micro-state.
Design: A 3-night study—night 1 as a baseline night, night 2 as a sleep fragmentation night, followed immediately by night 3 as a recovery night. On nights 1 and 3, approximately 400 auditory stimuli were presented during nonREM sleep in the first two sleep cycles, using stimulus parameters previously found to be optimal for K-complex production.
Setting: The sleep research laboratory at the University of Melbourne.
Participants: Six young healthy subjects (3 female),
Interventions: One night of sleep fragmentation. Ten-second auditory tones of up to 110 dB were presented throughout the entire night at approximately 1-minute intervals.
Measurements and Results: Sleep drive was increased on the recovery night, as indicated by increased amounts of slow wave sleep, increased sleep efficiency, and a reduction in stimulus-related alpha activity. The incidence of both evoked and spontaneous K-complexes increased significantly on the recovery night. When K-complex trials were averaged, neither N550 (Fz) amplitude nor latency differed between the 2 nights. When vertex sharp waves were averaged, N350 (Cz) amplitude was increased significantly on the recovery night.
Conclusions: The increase in K-complex frequency together with the decrease seen in stimulus-related alpha activity supports the view that they reflect a sleep maintenance, rather than an arousal, response.
Keywords: N550, N350, Delta EEG, evoked potentials, K-complex function.

INTRODUCTION

THE K-COMPLEX WAS FIRST OBSERVED ABOUT 60 YEARS AGO AS A TRANSIENT EVENT DURING SLEEP WHICH COULD EITHER OCCUR SPONTANEOUSLY OR BE EVOKED BY SENSORY STIMULI.[1,2] K-complexes are seen in stages 2, 3 and 4 of non-REM sleep, and together with sleep spindles are a defining feature of the onset of stage 2 sleep.[3] They are particularly conspicuous during this stage due to their large amplitude (up to 300 microvolts) compared to the ongoing background activity.

It has been argued by Bastien and Campbell[4] and others that, in addition to counting the frequency or determining the density of K-complexes, an effective way to evaluate the evoked K-complex is to average multiple K-complexes evoked by stimuli. This is based on the idea that responses to individual stimuli reflect a combination of "signal" (the K-complex), and "noise" (EEG activity unrelated to the stimulus or to the K-complex). It is assumed that K-complexes evoked by stimuli will show a constant (or near constant) temporal relationship to the stimuli. The noise should be randomly distributed relative to the stimuli, and thus should average to 0.

The averaged non-REM sleep evoked-potential waveform shows a number of characteristic peaks. The most prominent of these is the N550, which shows a symmetrical frontocentral topographic scalp distribution. This is thought to be the averaged K-complex as it has been shown to be absent or markedly reduced in averages of trials that do not contain K-complexes.[4-11]

An earlier negative peak (also known as N350) has also been considered by some authors to be part of the averaged K-complex waveform. However, this negative peak is present in averages of responses excluding K-complexes,[4,12] its topography shows it to be maximal at the vertex, and it is best seen in the average of trials evoking vertex sharp waves.[8,13,14] Bastien and Campbell[4] proposed that the N350 may act as trigger for the larger N550 on the basis of data that showed the N350 to be larger on trials that elicit a K-complex compared to those that do not.

The functional significance of the K-complex (spontaneous or evoked) has long been a source of debate. One of the major disagreements has been whether they represent arousal responses or are markers of a brain state conducive to the production of delta EEG activity and, thus, reflect processes attempting to maintain sleep. Early studies tended to be interpreted in favor of the arousal hypothesis.[15-22] However, more recent work[5,23-29] has indicated that K-complexes are reflective of a brain state in which arousals are less likely to occur.

One way of distinguishing between these hypotheses is to manipulate sleep drive. Increased sleep drive following sleep fragmentation or deprivation has been shown to reduce the likelihood of spontaneous arousal and to increase the arousal threshold.[30] It also leads to decreased sleep onset latency,[31,32] and increased proportions of REM sleep,[31] slow wave sleep (SWS)[31,32] and EEG spectral power in the delta frequency band.[33] Further, there is evidence of a frontal left-hemispheric dominance in delta activity during recovery from 40 hours of deprivation.[34]

Thus, if K-complexes reflect arousal processes, there should be fewer spontaneous K-complexes and fewer in response to a standard stimulus following increased sleep drive. On the other hand, if they reflect a brain state that is less conducive to arousal and thus more protective of sleep, there should be a higher frequency of both evoked and spontaneous K-complexes. A third possibility is that they do not reflect either an aroused or a sleep-protective brain state, in which case sleep fragmentation should have no effect.

METHOD

Participants

Three males and three females aged between 21 and 24 years (mean = 22.33 yrs, SD = 1.03 yrs) participated in the study. Participants were recruited using advertisements displayed on the University of Mel-

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Nothing to disclose

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bourne campus; while the majority of applicants were university students, anyone frequenting the campus was free to apply. Each applicant was screened using a questionnaire for general health. Successful applicants were specifically required to have no sleep, auditory, respiratory, or neurologic disturbances. The procedures were approved by the University of Melbourne's Human Subjects Ethics Committee.

Design

The protocol involved each subject spending 3 consecutive nights in the laboratory. The first and third nights involved the measurement of spontaneous and auditory-tone-evoked K-complexes. The second night involved approximately 8 hours of sleep fragmentation (using long auditory tones) to increase sleep drive for the third night's recording. The density of spontaneous K-complexes, the probability of stimulus-evoked K-complexes and vertex sharp waves, and the amplitude of averaged evoked K-complexes (N550 component) and vertex sharp waves (N350 component) were compared between first and third nights in a repeated measures design.

Equipment

Auditory stimuli: Auditory stimuli were presented via E-A-RTONE 3A insert earphones. Tones were digitally generated by a 16-Bit sound blaster (SB-16) sound card attached to an IBM-compatible personal computer and were presented bilaterally with a frequency of 1000 Hz (2msec rise and fall time). All stimulus intensities for baseline, recovery, and fragmentation nights (see below) were measured and calibrated using a Brüel & Kjær (Denmark) artificial ear Type 4152, coupled to a Brüel & Kjær sound level meter 2260. Auditory output was also recorded on the sleep monitoring system (Compumedics V5).

Data acquisition & digital recording: Evoked potential EEG data were recorded on the first and third nights of the study. Data was collected from 29 scalp sites adapted from the international 10/20 system (FP1, FP2, F7, F3, FZ, F4, F8, FT7, FC3, FCZ, FC4, FT8, T3, C3, CZ, C4, T4, TP7, CP3, CPZ, CP4, TP8, T5, P3, PZ, P4, T6, 01, 02) with a single horizontal electrooculogram (EOG) channel. On the second night (fragmentation) only a subset of these were acquired (FZ, FCZ, C3, CZ, CPZ, PZ, 02, EOG). (These data are not reported in the present paper but will be the subject of a subsequent manuscript.) All channels were referenced to linked ears. An ECI electrocap was used to ensure accuracy and consistency of electrode placement. Raw data were acquired and amplified using Neuroscan Synamps amplifiers and stored for off-line analysis using Neuroscan Scan software.

The EEG channels were continuously DC recorded (low-pass filter 200 Hz.) and sampled at 1000 Hz. A 50-Hz. notch filter was also employed. The C3 and O2 EEG, EOG, and submental EMG activity were also recorded continuously on a Compumedics V5 sleep-monitoring system at a sampling rate of 500 Hz.

Procedure

Prior to each experimental session, subjects were asked to refrain from consumption of alcohol and caffeine for 24 hours prior to (and throughout) the study. They were also asked to maintain a stable sleep-wake cycle both prior to and throughout the period of the investigation to ensure that they did not have any prior sleep deprivation. Subjects were also asked not to nap during the experiment. This was assessed by self-report, as actigraphy was not available.

Baseline and recovery nights: On the first and third nights (baseline and recovery) each subject received approximately 400 tone trials during the first two sleep cycles. Average 'lights out' time was 0:01 hrs ± 37 minutes on the baseline night and 23:36 hrs ± 21 minutes on the recovery night. Tones were presented binaurally during non-REM sleep in the first two sleep cycles via the insert earphones at 80 dB(A) with a frequency of 1000 Hz (2 millisecond rise and fall time, 52 millisecond duration). Tones were presented with a random interstimulus interval of between 10 and 30seconds during periods of stable NREM sleep. These stimulus parameters have been shown to be optimal for eliciting K-complexes and to be unlikely to produce EEG arousal.[4,5]

Sleep Fragmentation: Sleep fragmentation was achieved using long auditory tones based on the protocol used by Philip et al.[32] This involved presenting 1000 Hz tones at 70dB(A) for 5 seconds. If no arousal occurred the stimulus duration was increased to 10sec. If there was still no arousal then the stimulus intensity was increased in 10dB(A) increments (duration 10 seconds) to a maximum of 110dB(A) until arousal was achieved. There was a minimum of 1 minute between arousals, of which at least 30 seconds was required to be stable sleep (stages 2,3,4, or REM). Arousal was defined as a return to alpha or theta activity in either C3 or O2 (linked ears reference) EEG leads for a period of at least 3 seconds.[35] The fragmentation procedure continued for the whole night (approximately 8 hours). An addition to this protocol was required for the latter part of the night in 3 of the 6 subjects. It became necessary to enter the bedroom and physically awaken the subjects when the presentation of the maximum intensity stimulus [110dB(A), 10 second duration] failed to produce an arousal. When this was necessary, the procedure was as follows: After the first instance of the tones not producing an arousal, the experimenter entered the room and called the subjects' name until they responded verbally; the subject was then allowed to fall back to sleep. Once stable sleep had been attained for a minimum of 30 seconds and the tone procedure was restarted (see above). If the tone procedure continued to fail to elicit an arousal, then the experimenter continued to physically awaken the subject (verbal response required) every 5 minutes of stable sleep (stages 2,3,4, or REM), for the remainder of the night.

Data Reduction & Analysis

Sleep Stage Scoring: Digitized polysomnography data recorded from C3 and O2 electrode sites, as well as EOG and EMG channels, were independently scored visually by two researchers. Sleep records were scored in accordance with Rechtschaffen and Kales'[3] sleep stage criteria. Stages 3 and 4 were combined as SWS. Any differences in scoring between the two researchers were resolved by discussion.

Identification of spontaneous K-complex: Spontaneous K-complexes were identified in the C3/A1+A2 recording lead on the Compumedics monitoring system using Rechtschaffen and Kales'[3] criteria ("well defined negative sharp wave followed by a positive component. The total duration of the complex should exceed 0.5 seconds"). In addition, it was required that the negative component of the K-complex be at least 75 μV in amplitude, that the K-complex did not occur within 1 second of an auditory stimulus, and that it was clearly distinguishable from the background EEG. Only stage 2 sleep was used to calculate K-complex densities due to the difficulty in identifying spontaneous K-complexes during SWS. K-complex density was defined as the number of spontaneous K-complexes per minute of stage 2 sleep. Using this method, spontaneous K-complexes could only be identified in 5 subjects because the time at which the tones were presented was not marked on the Compumedics sleep record in 1 of the subjects, making it difficult to differentiate spontaneous from evoked K-complexes (this was not an issue with evoked K-complex analysis; these data were evaluated using the Neuroscan Scan 4.1 analysis software). Spontaneous vertex sharp waves were not counted due to difficulty in identifying them reliably.

Evoked K-complex and vertex sharp wave identification and trial averaging: On baseline and recovery nights, continuous stage 2 and SWS data were epoched to a window that started 500 milliseconds prior to stimulus onset and extended to 1500 milliseconds post stimulus and were bandpass filtered (0.1 Hz to 30 Hz). Evoked K-complexes were identified using the same criterion as those used for spontaneous K-complex identification. However, the amplitude (-75 μV) was measured at the Fz scalp site; the negative peak also had to occur between 400 and 750 milliseconds after the auditory stimulus was presented. Epochs containing arousals, and movement artifacts observed in EOG or EMG channels were rejected and not used in the averaging procedure. Evoked

vertex sharp waves[36] were defined as being a large negative sharp wave of at least 50 μV and a duration of less than 0.5 seconds at the Cz scalp site, with a latency between 250 and 450 milliseconds after the stimulus onset.

All trials that were to be included in the averaging process were classified into three response types on the basis of visual inspection of the epochs. The three response types were: trials that contained K-complexes (KC+); trials that contained vertex sharp waves (VSW+); and finally, all other trials (OTHER). If a trial produced both a K-complex and a vertex sharp wave, it was added into both of the KC+ and VSW+ averages. Each of the response-type groups was further classified by sleep stage (stage 2 & SWS). Grouped trials were averaged relative to tone stimuli for each subject. These produced average evoked-potential files for each of the response types across sleep stages for baseline and recovery conditions.

Component identification was achieved by importing the raw data from the average files into a spreadsheet program. The peak of the component of interest was defined as the most negative value (amplitude) that occurred between 400 milliseconds and 750 milliseconds for the N550 or between 250 milliseconds and 450 milliseconds for the N350 post stimulus.

N550 was measured at Fz in the KC+ average, where the N550 component has previously been shown to be maximal.[6,7,10,13,37,38] Likewise, N350 was measured at CZ in the VSW+ average for the same reason.[13,14,37,38]

Topographic maps using all 29-scalp sites were constructed from grand mean waveforms (ie, averaged within and across subjects) in each of the experimental conditions. The N550 component was mapped at the most negative point between 400 milliseconds and 750 milliseconds, while the N350 component was mapped at the most negative point between 250 milliseconds and 450 milliseconds.

Arousal identification: Tone-related alpha frequency (8 Hz - 12 Hz) amplitude was used as a quantitative measure of event-related arousal. On baseline and recovery nights, continuous stage 2 and SWS data were epoched to 500 milliseconds prior to stimulus onset and extended to 10,000 milliseconds post stimulus. A much longer epoch length was used in the arousal analysis compared to evoked-potential analysis (10.5 sec as compared to 2 sec) in order to ensure that any alpha activity due to the auditory stimulus was included. These 10.5 second epochs of raw data were subsequently digitally bandpass filtered (8 Hz-12 Hz 192 dB roll-off), rectified and averaged relative to the stimulus. Each averaged data time series was then cropped to 300 milliseconds prior to stimulus onset to 9,000 milliseconds post stimulus to remove filtering artifact and baseline corrected over the prestimulus interval. After processing, the peak alpha amplitude was automatically detected in the O2 derivation and then visually verified. This provided peak alpha amplitude values for each subject on baseline and recovery nights.

Statistical Analysis: Differences between nights 1 and 3 for sleep variables, the rate and probability of K-complexes and vertex sharp waves, and amplitude and latency differences for the N550 and N350 components were assessed using paired samples *t*-tests. Differences in tone-related alpha amplitude were measured using the Wilcoxon signed ranks test. Sleep variables were assessed using one-tailed probability levels, while the remaining tests used two-tailed levels. Data were checked for violations of the assumptions of the tests used, the non-parametric approach was used for the tone related alpha amplitude in light of the combination of a small n, a low effect size ($\varepsilon = 0.5$) and comparatively large pooled variance (pooled: mean = 1.2, variance = 0.91).

Scalp topography effects were assessed on baseline and recovery nights using the interaction term of a 2-way ANOVA at all sites performed on McCarthy-Wood transformed amplitude data.[39] The transformation [(site value-minimum)/(maximum-minimum),[39]] acts to eliminate any effects purely due to amplitude and, thus, only looks at differences in scalp distribution. Comparison of scalp distributions of EEG evoked potentials is not suited to the standard ANOVA model, which assumes that experimental main effects are additive (assuming that effects add a constant to a baseline condition). However, with EEG data, experimental effects result in a constant being multiplied to the topographic distribution data due to the fact that the EEG is recorded at the scalp surface at a site remote to the intracranial generator of the activity.[39]

All comparisons were made using data from all 6 subjects with the exception being the comparison between baseline and recovery nights of spontaneous K-complex production in which only 5 subjects were used.

RESULTS

Sleep statistics for each subject on baseline and recovery nights can be seen in Table 1. As indicated, measures reflecting increased sleep drive such as proportion of SWS, latency to SWS, and sleep efficiency increased while those indicative of decreased propensity to sleep, such as proportion of stage 1 sleep and average stimulus-related alpha amplitude decreased from baseline to recovery nights.

Table 1 also shows the rate of spontaneous K-complexes and the proportion of evoked K-complexes and vertex sharp waves elicited on baseline and recovery nights for each of the 6 subjects. The proportion of tone-elicited K-complexes during non-REM sleep (stage 2 and SWS) increased from baseline to recovery nights; however, there was no change in the proportion of non-REM tone-elicited vertex sharp waves.

Table 1—Sleep architecture, phasic events and evoked potential measures.

Variable	Baseline	Recovery	*t* Statistic	P value
Stage 1 latency (mins)*	6.08 (6.45)	2.00 (2.81)	1.36	ns
Stage 2 latency (mins)*	11.33 (8.94)	4.5 (3.49)	1.85	ns
SWS latency (mins)*	36.25 (19.51)	16.67 (5.04)	2.43	0.029
REM latency (mins)*	96.62 (60.81)	83.92 (27.19)	0.88	ns†
Stage 1 %	9.42 (4.01)	2.72 (1.40)	4.92	0.002
Stage 2 %	64.3 (10.22)	55.33 (15.49)	3.05	0.014
SWS %	17.22 (8.36)	32.53 (13.79)	-3.58	0.008
REM %	3.75 (4.02)	7.83 (3.53)	-3.51	0.009
Sleep efficiency** %	87.53 (9.43)	97.50 (1.66)	-2.63	0.024
Mean alpha amplitude (μV)	1.52 (1.1)	0.92 (0.6)	-2.20‡	0.028
Spontaneous (KC/min^{-1} of stage 2)	1.37 (0.51)	2.67 (0.78)	-3.65	0.022
Evoked KC (non-REM proportion)	0.62 (0.11)	0.70 (0.10)	-3.44	0.019
Evoked VSW (non-REM proportion)	0.17 (0.09)	0.14 (0.07)	1.06	ns
Fz N550 amplitude (μV)	-119.11 (36.99)	-124.41 (30.30)	0.75	ns
Fz N550 latency (ms)	612.83 (77.11)	578.00 (51.39)	1.02	ns
Cz N350 amplitude (μV)	-53.09 (14.31)	-68.12 (12.84)	4.18	0.009
Cz N350 latency (ms)	323.50 (27.75)	331.67 (39.07)	-1.01	ns

† Only four subjects were used in this analysis as two subjects failed to have REM sleep on the baseline night
‡ Wilcoxon Z Statistic.
* Time from lights out to first epoch of stage.
** Percentage of time in bed (with lights out) spent asleep.

Mean and standard deviation (in parentheses) for: Sleep architecture, peak alpha amplitude following stimuli, spontaneous K-complex rates (stage 2), evoked K-complex and vertex sharp wave (VSW) proportions (stage 2 and SWS), and evoked potential component measures on baseline and recovery nights (stage 2).

Spontaneous K-complex density during stage 2 non-REM sleep also increased from baseline to recovery nights.

There were no significant differences between baseline and recovery nights for the amplitude or latency of the N550 component. Descriptive statistics for amplitude and latency data in each condition can be seen in Table 1 and are graphically represented in the grand mean waveforms depicted in Figure 1. The topographic distribution is presented in Figure 2A. Differences in topographic distribution were assessed by the interaction term of the 2 by 29 (baseline/recovery by electrode site) repeated measures ANOVA performed on the McCarthy-Wood-transformed amplitude data[39] from the KC+ averages. No condition by site interaction effect was apparent $F(28,140) = 1.74$, $p > .05$ ($\varepsilon_{(\text{condition by site})} = 0.132$). These results were also reflected in topographic maps of the N550 generated from the grand mean results.

N350 amplitude increased from baseline to recovery with no difference in latency. Descriptive statistics for amplitude and latency data in each condition can be seen in Table 1 and are graphically represented in the grand mean waveforms depicted in Figure 3. The scalp topography is represented in Figure 2B. Differences in N350 topographic distribution were also assessed by the interaction term of the 2 by 29 (baseline/recovery by electrode site) repeated measures ANOVA performed on the McCarthy-Wood-transformed amplitude data[39] from the VSW+ averages. No condition by site interaction effect was seen $F(28,140) = 2.14$, $p > .05$ ($\varepsilon_{(\text{condition by site})} = 0.299$).

DISCUSSION

The results indicate that sleep drive was increased following a night of intense sleep fragmentation and that this was associated with an increase in spontaneous K-complex production, an increase in the proportion of K-complexes evoked by stimuli, and an increase in N350 amplitude in the averaged response. The data therefore support the hypothesis that the evoked K-complex reflects a brain state that is less conducive to arousal and thus more protective of sleep.

Sleep fragmentation produced a significant increase in sleep drive. The increase demonstrated robustness across a number of objective measures, including significant increases in proportion of SWS and sleep efficiency. These results are consistent with a number of other studies investigating the effects of sleep fragmentation and deprivation on sleep drive using both sleep fragmentation[32,40] and partial-sleep-deprivation protocols.[41] All studies have reported increases in the proportion of SWS, effects that were were interpreted as being indicative of increased sleep drive. Decreased levels of alpha activity in response to stimuli observed in the present study are indicative of lower levels of arousability. Decreased alpha has been previously suggested to relate to increased levels of sleep drive during recovery sleep.[30]

Although sleep-onset latency did not show a significant decrease, examination of the data showed decreases in sleep-onset latency for 4 of the 6 participants. In the remaining 2 subjects, equipment problems on the baseline night meant that the start of data collection was substantially delayed. During the delay, the subjects were in bed ready to go to sleep. It is thus likely that the short sleep-onset-latency values seen for these subjects are not indicative of their true level of sleepiness on the baseline night. Also, as actigraphy was not used, an objective measure of whether any daytime naps occurred was not available. However, the decreased levels of alpha activity demonstrated on the recovery night would not be expected if significant napping had taken place.

The increase in sleep drive produced by the fragmentation procedure resulted in a significant increase in the proportion of evoked K-complexes. The effect was seen in 5 of the 6 subjects. The sixth demonstrated a K-complex proportion of 0.77 on both baseline and recovery nights. Given that the highest value for the recovery night, for any of the subjects, was 0.78, his data are probably demonstrating a ceiling effect. This ceiling effect is consistent with previous research, with most studies reporting proportions between 0.2 and 0.62, depending on the stimulus parameters used.[4-6,42]

The N550 and N350 evoked-potential components showed topographic distributions consistent with prior topographic studies, with the N550 showing a prominent frontocentral negativity[6,7,10,13,37,38,43] and the N350 showing a vertex-negative distribution.[13,37,38,43-45]

No differences between conditions were noted in the amplitude or latency of the KC+ N550 component. These data are thus in agreement with Bastien and Campbell's[4,5] findings of the averaged evoked K-complex acting as an all-or-none response. N350 amplitude increased from –53.1μV on the baseline night to –68.1μV on the recovery night while showing no difference in latency. This would argue for an effect of fragmentation on the amplitude of vertex sharp waves, although as indicated previously there was no effect on the probability of vertex sharp wave production. These results highlight that the N350 should not be viewed merely as part of the averaged K-complex response.

As noted in the introduction, previous studies have concluded that the K-complex reflects some form of microarousal process. This conclusion is based on the presence of phenomena that may occur after a K-complex, such as an occasional return to alpha,[22] broad-band EEG power

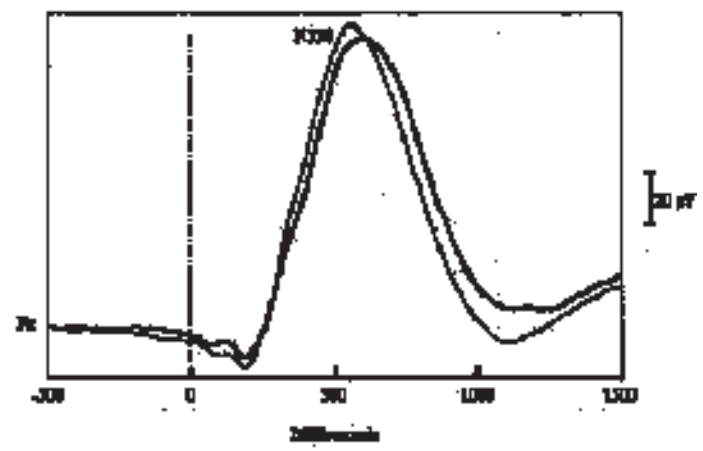

Figure 1—Grand mean (n=6) averaged KC+ evoked potential waveforms from the Fz scalp site on baseline (thick line) and recovery (thin line) nights. Zero time point = stimulus onset. The Y-axis uses the 'negative up' EEG convention.

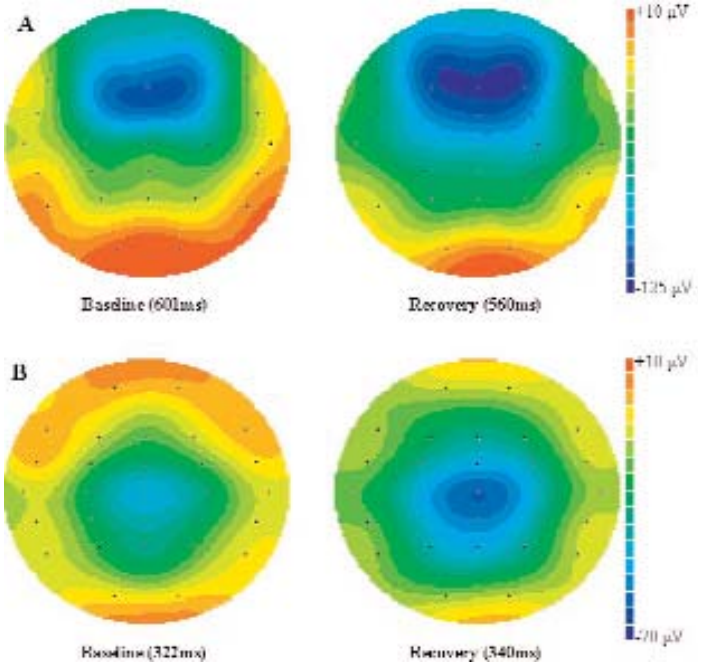

Figure 2—Topographic distributions of KC+ N550 (A) and of VSW+ N350 (B) evoked potential components on baseline and recovery nights. No statistical differences were seen between nights for either component. NOTE: Maps are based on grand mean results of unscaled amplitude data (n=6), Color scales (μV) are the same for each night, but are different for each component.

increases,[17-19,46] increases in heart rate,[15,20,47-49] or increased sympathetic nervous system tone in microneurography recordings.[50-54]

However, the present findings are inconsistent with the notion that the K-complex is an arousal response. The increase in the proportion of evoked K-complexes in the present study was corroborated by the large increase in spontaneous K-complex density during stage 2 sleep. The increase in density was particularly striking given that only 5 subjects could be used in the analysis. Along with these increases, there was a reduction in alpha activity in response to auditory stimuli from baseline to recovery nights. This inverse relationship between the K-complex and a measure of the extent to which the stimuli aroused participants would not be expected if K-complexes and arousals reflected the same underlying process.

Recent work by Amzica and Steriade[23-25] is also consistent with the K-complex reflecting a sleep-conducive brain state in which arousals are less likely to occur. In a series of experiments, they looked at the rhythmicity and morphology of K-complexes in humans, as well as what is believed to be the equivalent manifestation in the cat, using both field potential and intracellular recordings. They demonstrated a possible relationship between K-complexes and delta waves. De Gennaro, Ferrara and Bertini[27] provided extra support for this relationship in humans demonstrating increased stage 2 sleep K-complex densities prior to transition to SWS compared to transitions to REM sleep. This increase prior to SWS was modeled by a linear regression and decreased with successive sleep cycles across the night following the established pattern of delta waves and slow-wave activity originally proposed by Borbely.[55,56] We have also recently shown that spontaneous K-complex density,[57] evoked K-complex proportions,[37] and N550 amplitudes[37] are reduced in a population of neurologically healthy elderly showing very low levels of SWS.[57] Consistent with this, evoked K-complex production and N550 amplitude are also reduced in middle-aged alcoholic subjects relative to controls,[43] where alcoholics also show reduced SWS for their age.[58]

There are thus a number of lines of evidence to suggest a role for the K-complex as a marker of a sleep-conducive brain state. One of the most compelling is derived from both a logical and evidence-based standpoint. In order to generate large amplitude waveforms in EEG recorded from the scalp, large numbers of neurons and glial cells must be synchronously active.[59,60] For a K-complex to be generated, synchronisation must have occurred. Cortical synchronization, at the level of both corticothalamic and corticocortical networks, is indicative of decreased levels of cortical arousal and deeper stages of non-REM sleep. Thus both spontaneous and evoked K-complexes should be much more likely to occur when the overall level of cortical arousal is low and able to produce synchronized responses.

Amzica and Steriade's[23-25,28] work demonstrated synchronizing feedback and feed-forward loops that are activated when K-complexes and slow delta (< 1 Hz) activity are present, these loops serve to facilitate the deepening of sleep. The present finding of facilitated evoked and spontaneous K-complex generation after increased sleep drive could be explained within this framework. The increased sleep drive arguably would be reflected in the activity of the brainstem structures involved in sleep onset and maintenance, which not only act on the thalamus to reduce afferent flow to the cortex, but project directly to cortical neurons to facilitate their synchronization.[23,24,61] This feed-forward pathway may 'prime' the cortical cells for the hypersynchronous activation required for K-complex generation and provide a mechanism via which variations in sleep drive could directly effect K-complex generation.

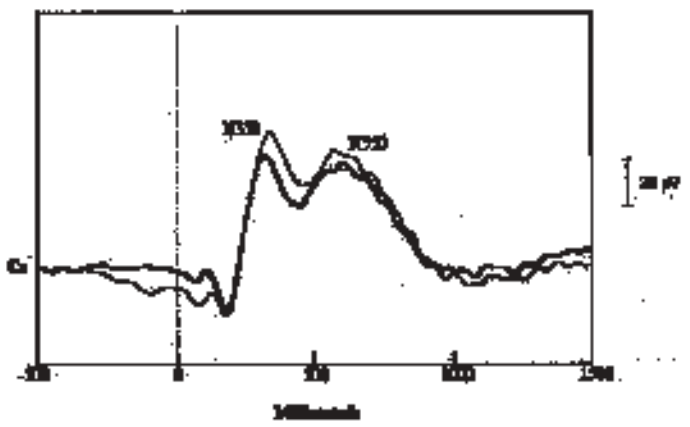

Figure 3—Grand mean (n=6) averaged VSW+ evoked potential waveforms from the Cz scalp site on baseline (thick line) and recovery (thin line) nights. Zero time point = stimulus onset. The Y-axis uses the 'negative up' EEG convention.

It is argued that after sleep drive has been increased, it is harder to arouse the sleeping central nervous system.[30,62] The increased incidence of delta waves and K-complexes, observed in the EEG after an increase in sleep drive should, therefore, be interpreted as indicative of this "difficult-to-arouse" brain state that is more conducive to K-complex and SWS generation. It is difficult to reconcile this finding with the hypothesis of K-complexes being indicative of arousal.

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