Automatic Analysis of Sleep Spindles —Assessment in One Case Treated with a Benzodiazepine Anxiolytic Drug—

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Summary: Wave-form analysis and detection of sleep spindles should be automatically performed to achieve objective and highly precise determination of the pattern of appearance of sleep spindles. The conditions for constructing the automatic analysis and detection are as follows: 1) it is recommended that EEGs be sampled at 500 Hz or higher intervals; 2) the digital filter with the threshold that is most appropriate for removal of artifacts should be used; 3) various conditions for the analysis and detection of wave-forms are widely used; and 4) a procedure for inspecting and reading record paper should also be included in the set-up. Taking these points into consideration, the authors developed an automatic analysis system for sleep spindles, which employs a computer. The system was applied to a case preliminarily treated with a benzodiazepine anxiolytic drug, ethyl loflazepate (CM6912). The assessment using this system revealed that the CM6912 administration induced an increase in sleep spindles, delayed frequency, and caused amplitude increases and a prolonged duration.

Key words: sleep — spindle — automatic — analysis — benzodiazepine — ethyl loflazepate — CM6912

Introduction

Sleep spindles are characteristic EEGs, which frequently appear during stage 2 sleep and during stage 3 and 4 sleep as well. A procedure in which investigators inspect and read recording paper was initially used as a method of analyzing sleep spindles. However, it was difficult to obtain highly precise objective

analysis results using this method. Therefore, various methods of automatically analyzing sleep spindles have been developed: a combined method involving an analog filter and a wave-form detection circuit (Gondeck and Smith, 1974; Smith et al. 1975; Johnson et al. 1976; Silverstein and Levy, 1976; Pivic et al. 1982; Ferri et al. 1989), a spectral analysis method (Johnson et al. 1969, 1976; Dumermuth

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et al. 1983), and a method utilizing a computer (Gondeck and Smith, 1974; Johnson et al. 1976; Matsubayashi et al. 1981; Principe and Smith, 1982; Ferri et al. 1989; Schimicek et al. 1994). The authors have developed a computer sleep analysis system (Kuwahara et al. 1988) and have utilized it for sleep experiments using drugs (Mizuki et al. 1988; Tanaka et al. 1988). Based on this experience, the authors constructed a new automatic analysis method for sleep spindles and applied it to a number of clinical cases to investigate the effects of a benzodiazepine anxiolytic drug on the appearance of sleep spindles. One of these cases is described in detail in this article.

Methods

Wave-form analysis

EEGs (led from C4-A1 and C3-A2) were sampled at 500 Hz by means of an analog-digital converter (12 bit; ± 1.25 V). The waves were processed with a non-recursive digital filter to block high- and

low-frequency areas and to obtain 0.5 to 30 Hz waves. Points at which the waves rose (v1, v2,v7) and fell (p1, p2,p6) in the same direction during 5 consecutive intervals (10 msec) were also detected. The interval between the points at which waves rose was regarded as the duration (v1-v2, v2-v3,v6-v7), and the interval between the straight line (connecting the points at which waves rose) and the cross point of the line and a vertical line from the points at which waves fell was regarded as the amplitude (a1, a2,a6) (Fig. 1).

Detection of sleep spindles

The absolute conditions of sleep spindles were as follows: the first wave was within 11-16 Hz and the amplitude was at least 8 μ V. Fundamentally, at least 6 waves of this kind were necessarily continuous, and the mean frequency had to be lower than 12-14 Hz. In actuality, however, waves that might have been evaluated as being continuous sleep spindles on inspection may be divided on automatic analysis. Even when waves

Fig. 1. Analysis of sleep spindle wave-forms and parameters determined (explained in the text).

other than 11 to 16 Hz waves were present, they were regarded as representing a portion of the continuous sleep spindles if they were within 9-18 Hz and showed an amplitude of at least $5 \mu\text{V}$ and the subsequent wave met the absolute conditions of sleep spindles. The parameters, i.e., the number of sleep spindles that appeared, the frequency, amplitude and duration, were detected by automatic analysis. A super mini-computer (ECLIPSE MV/20000, Data General Inc.) was used for the analysis, and the application program was developed with FORTRAN 77.

The underlined parts, that is *a* through *h*, of EEGs in Fig. 2 show the sleep spindles detected by automatic analysis. Parts *a* to *f* appeared during stage 2 sleep, and part *f* was convoluted with the K-complex. Parts *g* and *h* were convoluted with high-amplitude slow waves appearing dur-

ing stage 4 sleep. The threshold of the digital filter, which is established prior to wave-form analysis, and various conditions such as the frequency, amplitude and continuity, which are established prior to the detection of sleep spindles, can be changed voluntarily in the automatic analysis. This protocol can also be applied to the recognition of characteristic waves involving vertex sharp waves and saw-toothed waves.

Only the international standard for evaluation of sleep stages (Rechtschaffen and Kales, 1968) is insufficient for automatic analysis of sleep spindles, because it is designed for inspecting and reading of recording paper. In practice, various innovations have been devised to date (Gondeck and Smith, 1974; Smith et al. 1975; Silverstein and Levy, 1976; Matsubayashi et al. 1981; Principe and

Fig. 2. The case in which sleep spindles were automatically detected (underlined parts *a* - *h*). The calibration was $50 \mu\text{V}$.

Smith, 1982; Schimicek et al. 1994). The authors used the contents of these studies to a considerable extent as reference material in this study.

Application of Automatic Analysis

Outline of the drug experiment and analysis subjects

The effects of a benzodiazepine anxiolytic drug, ethyl loflazepate (CM6912), on sleep have been investigated (Mizuki et al. 1988) by means of our original automatic sleep analysis system (Kuwahara et al. 1988). According to the experimental schedule, the subjects were 12 male students. The period from the first to third nights was regarded as the baseline night, the fourth and fifth nights as the drug night, and the sixth night as the recovery night. A placebo was used on the baseline and recovery nights, and CM6912 was administered on the drug night. The effects of CM6912 administration on sleep stages included decreases in stage W and stage 1 sleep, an increase in stage 2 sleep, decreases in stage 3 and stage 4 sleep, and no marked change in stage REM sleep. The baseline third night and the drug fifth night were analyzed for application of the automatic analysis of sleep spindles in one case (aged 20 years) treated with 4 mg of CM6912. The pattern of the appearance of automatically detected sleep spindles during stage 2 and stage 3+4 sleep was investigated.

Observation according to time-course changes

Figures 3 and 4 show the results of automatic analysis of the baseline third

night (baseline night) and the drug fifth night (the drug night). Sleep stage diagram (the validity of inspection was investigated and corrected in the sleep stages evaluated automatically by the automatic sleep analysis system) is shown at the top of the figure, and time-course changes in the frequency of sleep spindles, mean frequency, mean amplitude and mean duration are shown for 20 sec for each individual parameter expressed as one period. A reciprocal relationship, in which mean frequency was delayed along with the increase in sleep spindles, was observed, and was particularly markedly in the latter half of the sleep stage, on the baseline and drug nights. The mean frequency showed arch-shaped changes in the continuous stage 2 period, showing a late frequency at the center. The number of sleep spindles increased, the mean frequency was low, and the duration tended to be prolonged on the drug night, as compared to the baseline night.

Changes due to administration

Tables 1 and 2 show changes in the number of periods (each period is 20 sec) which appeared and parameters (mean $\pm$ standard deviation) which were determined during stage 2 and stage 3+4 sleep on the baseline and drug nights. When the number of periods appearing was calculated in terms of the frequency of appearance, the value was 54.2% during stage 2 and 18.7% during stage 3+4 on the baseline night, and 67.4% during stage 2 and 9.3% during stage 3+4 on the drug night. The frequency was increased by 13.2% during stage 2 and decreased by 9.4% during stage 3+4 on the drug night. These findings are characteristic

Fig. 3. Sleep stage diagram and time-course changes in parameters determined on the baseline (third) night. Zero on the time axis corresponds to 22:30 PM.

TABLE 1.

Changes in parameters determined on the baseline night

Sleep stage Epochs	Stage 2 640	Stage 3+4 125
Spindles (n)	2.78±1.54	1.47±0.75
Frequency (Hz)	13.29±0.39	13.30±0.55
Amplitude (μV)	18.41±3.35	16.49±3.27
Duration (msec)	724.85±186.70	556.80±120.51

TABLE 2.

Changes in parameters determined on the drug night

Sleep stage Epochs	Stage 2 1076	Stage 3+4 136
Spindles (n)	3.96±1.82	2.33±1.37
Frequency (Hz)	12.94±0.38	12.86±0.48
Amplitude (μV)	19.10±2.95	17.20±2.78
Duration (msec)	858.76±227.35	646.50±166.28

Fig. 4. Sleep stage diagram and time-course changes in parameters determined on the drug (fifth) night. Zero on the time axis corresponds to 22:30 PM.

effects of the CM6912 administration. The parameters determined during stage 2 and stage 3+4 sleep were compared on the baseline night. The number of sleep spindles, amplitude and duration observed in stage 3+4 were less than those seen during stage 2, but the frequency was almost unchanged from stage 2 to stage 3+4. These tendencies were similar to those observed on the drug night. The parameters obtained during stage 2 on the baseline and drug nights were compared. The frequency was delayed, and the number of sleep spindles, amplitude and duration exceeded those

on the baseline night. These tendencies were similar to those noted during stage 3+4. These results suggest that the quantity of sleep stages appearing is changed by administration of CM6912 but that the drug has no qualitative influence on the pattern of sleep spindle appearance. Details of these parameters should be assessed in more subjects in future analyses.

References

Berger RJ, Dement WC, Jacobson A, Johnson

- LC, Jouvet M et al. A manual of standardized terminology, techniques and scoring system for sleep stages of human subjects. ed. Rechtschaffen A and Kales A. Public Health Service, U. S. Government Printing Office, Washington DC, 1968.
- Dumermuth G, Lange B, Lehmann D, Meier CA, Dinkelmann R et al. Spectral analysis of all-night sleep EEG in healthy adult. *Eur Neurol* 1983;22:322-339.
- Ferri R, Dequae P, Colognola RM, Musumeci SA, and Bergonzi P. A simple electronic and computer system for automatic spindle detection. *Neurophysiol Clin* 1989;19:171-177.
- Gondeck AR, and Smith JR. Dynamics of human sleep sigma spindles. *Electroencephalogr Clin Neurophysiol* 1974;37:293-297.
- Johnson LC, Hanson K, and Bickford RG. Effect of flurazepam on sleep spindles and K-complexes. *Electroencephalogr Clin Neurophysiol* 1976;40:67-77.
- Johnson LC, Lubin A, Naitoh P, Nute C, and Austin M. Spectral analysis of the EEG of dominant and non-dominant alpha subjects during waking and sleeping. *Electroencephalogr Clin Neurophysiol* 1969;26:361-370.
- Kuwahara H, Higashi H, Mizuki Y, Matsunari S, Tanaka et al. Automatic real-time analysis of human sleep stages by an interval histogram method. *Electroencephalogr Clin Neurophysiol* 1988; 70:220-229.
- Matsubayashi K, Ishiyama Y, Homma I, and Ebe M. Some characteristics of sleep spindles derived from automatic analysis. *Sleep* 1981;4:392-399.
- Mizuki Y, Kuwahara H, Isozaki H, Tanaka M, and Inanaga K. Effect of ethyl loflazepate (CM6912) on sleep in normal humans. *Meth and Find Exptl Clin Pharmacol* 1988;10:401-406.
- Pivic RT, Bylsma FW, and Nevins RJ. A new device for automatic sleep spindle analysis: The 'spindicator'. *Electroencephalogr Clin Neurophysiol* 1982;54:711-713.
- Principe JC, and Smith JR. Sleep spindle characteristics as a function of age. *Sleep* 1982;5:73-84.
- Schimicek P, Zeitlhofer J, Anderer P, and Saletu B. Automatic sleep-spindle detection procedure: aspects of reliability and validity. *Clin Electroencephalogr* 1994;25:26-29.
- Silverstein LD, and Levy CM. The stability of the sigma sleep spindle. *Electroencephalogr Clin Neurophysiol* 1976;40:666-670.
- Smith JR, Funke WF, Yeo WC, and Ambuehl RA. Detection of human sleep EEG waveforms. *Electroencephalogr Clin Neurophysiol* 1975;38:435-437.
- Tanaka M, Mizuki Y, Kuwahara H, Isozaki H, and Inanaga K. Effect of neurotropic on polysomnographic patterns in normal humans. *Int Clin Psychopharmacol* 1988;3: 239-244.