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Sleep Induced by Intestinal Stimulation in Cats

TIBOR KUKORELLI AND GÁBOR JUHÁSZ

Department of Comparative Physiology, Eötvös Loránd University and Institute of Psychology of the Hungarian Academy of Sciences
1088 Budapest, Muzeum krt 4/A., Hungary

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KUKORELLI, T. AND G. JUHÁSZ. *Sleep induced by intestinal stimulation in cats.* PHYSIOL. BEHAV. 19(3) 355–358, 1977. – The influence of afferent impulses of intestinal origin on the sleep stages was studied in fed and starved cats. Low-frequency electrical stimulation of the mucosal surface in a small intestinal fistula reduced the latency of sleep onset. The number of slow wave sleep episodes decreased, but their mean duration increased during stimulation. Conversely, the number of paradoxical sleep episodes increased, but their mean duration was not significantly modified by the intestinal stimulation. The role of viscerosensory events in the control of sleep is discussed in relationship to these results.

Sleep induction Intestinal stimulation Low frequency Starvation Sleep duration
Sleep-wakefulness cycle EEG Slow wave sleep Paradoxical sleep

THE PRESENT study was conducted as part of a larger investigation [6,7] concerned with the activity of intestinal afferents and brain mechanisms regulating behavior. Viscerosensory activity has been shown to influence the sleep-wakefulness rhythm. We previously [6,7] observed that low-frequency (1–8 c/sec) stimulation of the small intestine and splanchnic nerve produced electroencephalographically (EEG) defined signs of sleep onset in cats. These findings taken together with that of a reduction in sleep stages (slow-wave, paradoxical) observed [13] after the abdominal vegetative nerves were severed, raise the possibility of a hypnogenic influence with its origin in the intestinal receptors. Therefore, the objective of the present study was to investigate this as an effective site for influencing the various sleep stages in cats by obtaining polygraphic recordings during chronic intestinal stimulation. Would the sleep cycle be altered by low-frequency stimulation of the small intestinal mucosa?

METHOD

Ten adult male cats aged 2–4 years weighing 3–5 kg. were prepared with chronic intestinal fistula and electrodes were implanted for sleep recordings under Nembutal anesthesia. The intestinal fistula were constructed as previously described [7]. Following median laparatomy, a loop of 15 cm long was excised from the upper part of jejunum. The continuity of the intestinal tract was restored by a bilateral anastomosis. Both ends of the isolated intestinal loop were sutured to the abdominal skin 5 cm apart. The loop rested in the abdominal cavity in a U-shaped position, retaining its original innervation and blood vessel connections.

At least 6 weeks was allowed for recovery before electrode implantation. Stainless steel screws which were threaded bilaterally into the skull over the frontal, parietal, temporal and occipital areas of the cortex served to record EEG activity. A bipolar Nicrotan electrode was stereotaxically inserted into the right dorsal hippocampus. Additional screws were placed into the upper wall of right orbit to monitor electrooculographic (EOG) activity. Electromyogram (EMG) electrodes were inserted subdermally over the dorsal neck muscles.

A bipolar electrode was inserted into the lumen of the isolated intestinal loop to stimulate the mucosa. The tip was composed of Plexiglas which measured 0.5 × 2.5 cm, and two 0.5 cm^2 gold-plate caps separated by a distance of 8 mm served as conducting surfaces. Squarewave pulses, 0.5 msec in duration with a frequency of 1 Hz and 2–6 V were used for stimulation. To each cat, the minimum stimulus intensity was applied which would induce sleep onset by EEG criteria in the drowsy state without overt behavioral sequelae [6,7]. EEG, EMG, and EOG activity were continuously recorded at 8–10 cm/min. Simultaneously, direct observation of behavior was made.

These animals were placed individually into a recording cage 0.8 m long, 0.7 m wide × 0.7 m tall in a sound-attenuated and electrostatically shielded room. The cage was illuminated by a 0.3 W light bulb with the temperature maintained at 23 ± 1°C. The animals were placed on a 12:12 light/dark cycle with illumination from 06.00–12.00 hr. Each day 1.5 kg of beef and 1 kg milk were fed to 5 cats at 08.30 hr, to 5 at 13.30 hr. Water was provided ad lib. Polygraphic recording was via a multistrand cable which contained leads attached to a plug on the cat's head.

The cats were adapted 4 hr/daily for a week to the cable and intestinal-stimulating electrode. Recordings began at 0.900 hr and were terminated 4 hr later. First, continuous polygraphic (control) recordings were obtained for 5 consecutive days without intestinal stimulation. This was followed by 5 successive recording days when the intestinal mucosa was stimulated continuously during wakefulness (W), drowsiness (D) and slow-wave (SWS), but not in paradoxical sleep (PS). Five animals were fed for about 25 min before each recording period, while the other 5 were starved throughout the 10 days encompassing the experiment. Each 60-sec epoch of the ten 4-hr records per animal (5 control/5 stimulation) was classified according to previously adopted criteria [2,14] into one of the following categories: W, D, SWS, and PS. An epoch was assigned to that stage which occupied 30 sec or more of its duration.

RESULTS

The data derived from the sleep-wake states was compared between groups (fed vs. starved) and with respect to treatment (control vs. stimulation). All statistical analyses were based upon 2-tailed *t*-tests. The distribution of wakefulness and the sleep stages remained unchanged during the control period. Sleep duration did not increase over consecutive days, but was similar on the first ($\bar{X}$ = 140.7 min) and last ($\bar{X}$ = 137.3 min). Pronounced differences existed between the control data of the starved and satiated cats. There was a statistically significant main effect on the duration of episodes occupied by W, D, and each sleep stage. Table 1 shows that the mean episode durations of W and D were less while those of PS and SWS were greater in the fed compared with the starved animals (all $ps<0.01$). Also there were significantly ($p<0.01$) fewer PS episodes in the starved group.

Repetitive intestinal stimulation affected all of the various stages of sleep and wakefulness both in the starved and fed cats. Figure 1 shows that relative to the control

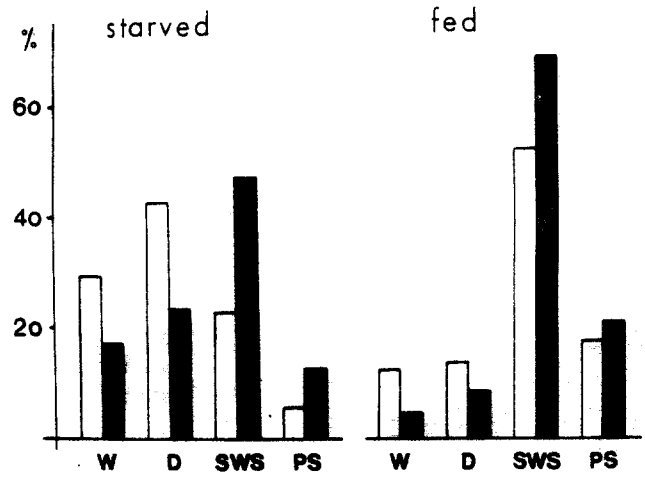

FIG. 1. Distribution of total recording time in the wakefulness and sleep stages before (white columns) and during (dotted columns) intestinal stimulation.

period SWS was longer by 23.3% in the starved and by 16.5% in the satiated group during stimulation than before. It can be seen from Table 1 that the duration but not the frequency of SWS episodes was significantly increased by stimulation in both groups ($ps<0.01$). Mean time to the onset of the first SWS episode was significantly reduced (both $ps<0.01$) in the starved group by 10 min, and 5.2 min in the fed animals. The specific effect of intestinal stimulation was then that it increased the episode durations of SWS. The mean number of PS episodes increased significantly ($ps<0.01$, Table 1), but the mean episode duration was not systematically modified by stimulation (Fig. 1). Thus, the specific effect of intestinal stimulation on PS was that the frequency of its occurrence was enhanced.

TABLE 1

MEAN DURATION AND NUMBER OF EPISODES OF THE SLEEP-WAKEFULNESS STAGES IN EACH CONDITION

	Condition			
	Control		Stimulation	
Variable	Starved	Fed	Starved	Fed
Duration (min) of:				
Wakefulness	13.6 ± 4.2	6.2 ± 2.3*	9.5 ± 1.4†	2.5 ± 0.8†
Drowsiness	11.5 ± 3.9	5.3 ± 1.6*	7.2 ± 3.1†	2.1 ± 1.2†
Slow-wave sleep	11.3 ± 3.5	16.7 ± 2.4*	20.0 ± 3.9‡	20.2 ± 2.6†
Paradoxical sleep	5.2 ± 0.2	6.2 ± 0.3*	5.8 ± 0.1	5.9 ± 0.5
Number (for 4 hr) of:				
Wakefulness	5.1 ± 2.3	4.2 ± 1.5	4.2 ± 3.1	2.7 ± 2.1
Drowsiness	9.0 ± 5.4	8.3 ± 4.6	8.0 ± 4.3	8.8 ± 3.7
Slow-wave sleep	4.9 ± 3.3	7.4 ± 2.8	5.7 ± 3.5	8.2 ± 4.2
Paradoxical sleep	2.4 ± 1.6	6.7 ± 0.7*	5.3 ± 1.2‡	8.1 ± 0.9†

Values are means of 5 animals ± SD.
*$p<0.01$—significantly different from starved; †$p<0.01$, ‡$p<0.001$—significantly different from control.

The relationship between total SWS and PS duration did not show significant changes either in starved or satiated animals. The proportion of SWS was reduced by 2.4% in the starved and increased by 2.5% in the fed cats. During each hour of intestinal stimulation total sleep time was longer than in the control condition. The hypnogenic effect of stimulation was most conspicuous during the initial 2 hr in the starved group ($p<0.001$) and for the last 2 hr in the satiated cats ($p<0.01$) (Fig. 2). Excitation of the small intestine decreased the total duration of wakefulness (Fig. 1). The mean duration of W and D episodes in both groups was reduced significantly (both $ps<0.01$, Table 1), whereas the frequency of episodes was not.

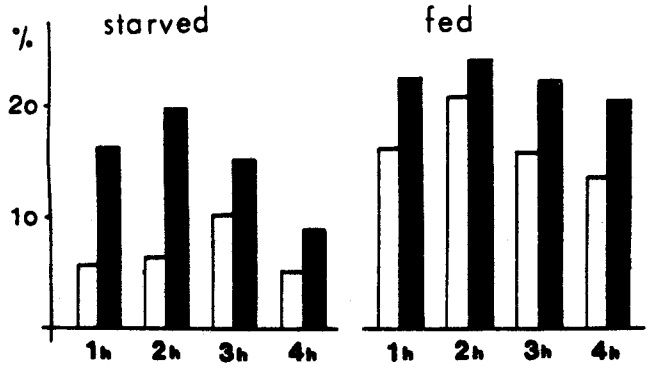

FIG. 2. Distribution of the total sleep time in different experimental hours before (white columns) and during (dotted columns) intestinal stimulation.

DISCUSSION

The present results indicate that repetitive intestinal stimulation increases sleep duration both in starved and satiated animals. The finding could be construed to indicate that more sleep was accumulated as a function of elapsed time per se rather than representing the treatment. An experimental design with counterbalanced conditions would have reasonably excluded such a possibility. Any influence on the sleep-wake states of intestinal stimulation must therefore be interpreted with the constraint that a fiexed-order of treatments was imposed. Despite the latter constraint the sleep-wakefulness cycle remained stable in the control condition. Regardless of 5 consecutive daily recording sessions, no corresponding increase of total sleep or the various sleep stages was observed in the control conditions. Because, differences in various EEG and behavioral parameters existed between the starved and satiated cats is probably the most cogent evidence that intestinal stimulation was the responsible factor for the findings presented here. The present results then enable us to speculate upon some peripheral (viscero-sensory) and CNS events associated with the influence of intestinal afferents under conditions of stimulation.

Peripheral Events

The wall of the small intestine contains mucosal mechanoreceptors, besides tension receptors. The majority of the mechanoreceptors are highly sensitive and a rapidly adapting on-off response is evoked when the mucosa is stimulated. The tension receptors are tonically active during intestinal distension and show relatively slower adaptation. The afferents of phasic receptors run in the splanchnic and mesenteric nerves. The tension receptors are innervated by the vagal fibers [3, 8, 10, 12].

In our previous study [6,7] EEG sleep onset was induced by low-frequency electrical stimulation of the small intestine and splanchnic nerve, or by rhythmic intestinal distensions. It was concluded that the hypnogenic effect was the result of the rhythmic excitation of both phasic and tension receptors. In the present study repetitive application of intestinal stimulation shortened the latency of sleep onset and increased sleep duration. These results indicate that the rhythmic activity of the intestinal receptors is reflected by indexes of both EEG and behavioral sleep. Moreover, the rhythmic character of the hypnogenic stimulation suggests that afferent impulses, generated by the rhythmic intestinal movements during digestion, may play an important role in the induction of sleep following food intake.

Besides the neural factors, however, the hypnogenic effect of some humoral substances such as hormones and metabolites should also be considered. There is a substantial evidence about relationships of the sleep-wakefulness cycle to metabolism [4, 5, 9, 15] indicating the hypnogenic influence on gastrointestinal hormones such as secretin and cholecystokinin. A pronounced increase in sleep duration was observed after intravenous administration of secretin and cholecystokinin even in cats with denervated gastrointestinal tracts [13]. The release of the gastrointestinal hormones is controlled by viscero-visceral reflexes [8]. Therefore, it seems probable that intestinal stimulation promotes sleep not only by eliciting a specific pattern of afferent impulses, but by increasing the release of gastrointestinal hormones.

Central Events

Intestinal stimulation increased the duration of SWS and the frequency of PS episodes indicating that afferent impulses of intestinal origin can effect those two mechanisms of the brain which are presumed to regulate these sleep states [5,11]. The latency to sleep onset decreased and the mean duration of SWS but not PS episodes increased during stimulation. These results suggest that the viscerosensory influence on SWS mechanisms is possibly primary and more important than for PS. But, because the stimulation was discontinued during PS, the increase of episodes could have resulted from priming events of this sleep stage during SWS [5,11].

Conversely, humoral afferent factors may selectively promote PS which is suggested by the finding that during the control period both measures of this sleep stage were greater in the satiated than in the starved animals. Also the intravenous administration of secretin and cholecystokinin in a previous study [13] resulted in a pronounced increase of PS episodes.

Among the CNS events associated with the effect of intestinal stimulation changes of food intake should be considered. The satiated animals slept more than the starved ones both in the control and in the experimental periods. Decreased total sleep time was also observed during starvation in rats [4]. The starved cats spent relatively more time in SWS. Correspondingly, the duration of SWS (stage 3 and 4) was increased during starvation in humans [9]. Thus, the quantity of SWS increased relatively, but the

total sleep time and the hypnogenic effect of intestinal stimulation decreased during starvation. The reduction of total sleep time during the last 2 hr of stimulation in starved cats could be explained by the arousing effect of an increased appetitive state. Conversely, it is likely that the drive-reducing influence of intestinal stimulation is conductive to sleep onset even in starved animals. Such a possibility is supported by a direct effect of gastrointestinal afferents on the hypothalamus [1]. Gastrointestinal distension can reciprocally modify the electrical activity of the ventromedial and lateral nuclei. In conclusion, the present results suggest the involvement of intestinal afferent impulses in the control of sleep behavior.

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