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BRIEF COMMUNICATION

Influence of Zolpidem, a Novel Hypnotic, on the Intermediate-Stage and Paradoxical Sleep in the Rat

CLAUDE GOTTESMANN,*[1] SYLVIE TREFOURET* AND HENRI DEPOORTERE†

**Laboratoire de Psychophysiologie,*
Faculté des Sciences, Université de Nice-Sophia Antipolis, 06108 Nice, Cedex 2, France,
and †Synthelabo Recherche (L.E.R.S.), Bagneux, France

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GOTTESMANN, C., S. TREFOURET AND H. DEPOORTERE. *Influence of zolpidem, a novel hypnotic, on the intermediate-stage and paradoxical sleep in the rat.* PHARMACOL BIOCHEM BEHAV 47(2) 359–362, 1994.—Paradoxical sleep (PS) in mice, rats, and cats is preceded and sometimes followed by a short-lasting stage characterized by cortical high-amplitude spindles and hippocampal low-frequency theta rhythm. This intermediate stage (IS) seems to correspond to a transient functional disconnection of the forebrain from the brainstem. Pentobarbital and benzodiazepines greatly extend IS at the expense of PS, which is suppressed. Zolpidem, a new imidazopyridine hypnotic, was studied at 2.5, 5, and 7.5 mg/kg IP. At 2.5 mg/kg, which is already a true hypnotic dose, it only decreased PS during the first 2 h of recording with a rebound during the following 4 h of recording. At 5 mg/kg, zolpidem increased the number and total duration of IS episodes, increased IS episodes not followed by PS, and increased PS latency of occurrence. PS amount was decreased during the first three h with a rebound in the next 3 h. At 7.5 mg/kg, the total amount of PS was also decreased. The eye movement number and theta rhythm frequency of PS were unchanged. These results show that zolpidem produces less disruption of the association between IS and PS than do previous hypnotics.

Hypnotics Zolpidem Sleep Intermediate stage Paradoxical sleep Theta rhythm Rat

PARADOXICAL sleep (PS) in the mouse (8), rat (3,5,9,19), and cat (11) is preceded and sometimes followed by a short-lasting stage (a few seconds) characterized by high-amplitude cortical spindles (index of deep slow-wave sleep) and hippocampal low-frequency theta rhythm (index of central activation since occurring during active waking and PS). It was shown that a similar unusual pattern association only appears in the acute intercollicular transected rat (12) and cat (13), when the brainstem ascending influences are suppressed. Moreover, the thalamic transmission level in the rat (6) and thalamocortical responsiveness in the cat (11) is the lowest of all sleep–waking stages during this "intermediate stage" (IS), and it is well known that brainstem ascending influences control thalamic transmission processes (17). Consequently, IS seems to occur when the brainstem ascending influences of waking, which decrease during slow-wave sleep, are at their lowest level, or suppressed, while PS ascending influences are still absent or at a low level. Thus, the forebrain structures seem briefly to be functionally disconnected from the brainstem (10).

It was previously shown that, at low dose, pentobarbital, a first-generation hypnotic, greatly extends IS at the expense of PS in rats (9) and cats (11), IS being able to last several minutes while PS is suppressed. Recently, it was shown that benzodiazepines, second-generation hypnotics, act similarly (7). Here, we test the influence of zolpidem, a new imidazopyridine hypnotic (2,4). A preliminary report was already published (18).

METHOD

Under penthiobarbital anesthesia (60 mg/kg), eight male, adult Wistar rats were bilaterally implanted with bipolar electrodes of coated (except at the tip) stainless steel wires (10/

[1] To whom requests for reprints should be addressed.

100 mm) into the CA1 area of the dorsal hippocampus (A, 5.4; L, 2.6; D, +7.5) according to Paxinos and Watson's atlas (15). Silver ball electrodes were placed bilaterally on the dura over the frontal cortex and the rostral and caudal borders of one orbit to record, respectively, electroencephalograph (EEG) and eye movements. For the electromyogram, two twisted stainless steel wires (25/100 mm) were inserted in the dorsal neck muscles. After surgery, rats were given an antibiotic (specilline, 50,000 U, IM) and housed in individual cages with habituation cables during 1 week to recover under a 12 L : 12 D cycle, with lights on at 9.00 a.m. On the first experimental day, animals received saline IP at 9.00 a.m. and the recording session lasted from 9.00 a.m. to 3.00 p.m. The following day, they received zolpidem and were recorded in the same conditions. Three doses were studied: 2.5, 5, and 7.5 mg/kg, with an interval of 2 weeks at least, animals having habituation leads throughout the experiment.

IS was strictly scored as the association of spindles and theta activity at the PS entrance. When several spindles successively occurred interspersed with slow waves or desynchronized activity, only the spindle duration was considered as characterizing IS. The scoring was blind. The statistical analysis was performed with Wilcoxon's test.

RESULTS

Intermediate Stage

As shown in Table 1, at 2.5 mg/kg, already a true hypnotic dose [sleep was induced in the range of a few minutes; see (4)], zolpidem was without any effect on IS. At 5 mg/kg, IS episode mean duration was unchanged but the number of IS episodes increased [36.3 ± 6.7 (SEM) vs. 64 ± 8.1; $p < 0.02$], as did IS total duration (46.3 ± 7.3 s vs. 84.6 ± 11.5; $p < 0.02$) during the 6-h recording. IS episodes not followed by PS increased (7.5 ± 1.8 vs. 10 ± 1.9; $p < 0.05$). At 7.5 mg/kg, the mean duration of IS episodes increased slightly (1.3 ± 0.1 s vs. 1.5 ± 0.1; $p < 0.05$) and IS episodes not

TABLE 1

EFFECT OF ZOLPIDEM ON INTERMEDIATE-STAGE AND PARADOXICAL SLEEP

	2.5 mg/kg (n = 8)		5 mg/kg (n = 8)		7.5 mg/kg (n = 7)	
	Saline	Zolpidem	Saline	Zolpidem	Saline	Zolpidem
IS number of episodes (6 h)	36.5 ± 13.6	40.8 ± 10.9	36.3 ± 6.7	64.0 ± 8.1*	51.1 ± 10.8	70.1 ± 14.8
IS total duration (6 h) (s)	69.5 ± 31.9	80.8 ± 22.6	46.3 ± 7.3	84.6 ± 11.5*	72.0 ± 21.7	105.1 ± 23.4
IS mean duration (s)	1.8 ± 0.1	1.9 ± 0.2	1.3 ± 0.1	1.3 ± 0.1	1.3 ± 0.1	1.5 ± 0.1†
IS episodes not followed by PS	7.6 ± 1.7	11.25 ± 1.4	7.5 ± 1.8	10.0 ± 1.9†	8.1 ± 2.4	12.3 ± 3.7*
Theta rhythm frequency during IS	6.6 ± 0.1	6.5 ± 0.1	6.8 ± 0.2	6.5 ± 0.1	6.4 ± 0.1	6.4 ± 0.1
PS latency (min)	57.1 ± 8.0	93.3 ± 12.3	42.0 ± 6.3	98.3 ± 10.6*	36.0 ± 5.2	108.4 ± 27.5*
Number of PS episodes						
0–3 h	7.6 ± 1.4	6.1 ± 0.8	13.3 ± 1.4	4.4 ± 0.7*	11.7 ± 1.0	5.7 ± 1.3*
3–6 h	11.6 ± 1.6	11.0 ± 1.7	12.5 ± 2.6	15.9 ± 1.6	13.4 ± 1.2	11.8 ± 1.6
Total 6 h	19.3 ± 2.4	17.1 ± 1.6	25.9 ± 3.5	20.3 ± 1.9	25.1 ± 1.2	19.0 ± 3.3
PS duration (s)						
0–2 h	323.9 ± 50.7	148.0 ± 44.5†	623.9 ± 101.6	93.1 ± 23.6*	538.4 ± 75.7	135.7 ± 26.2*
2–4 h	629.5 ± 130.4	692.3 ± 111.9	940.8 ± 113.4	644.4 ± 70.6	804.3 ± 144.6	695.9 ± 165.3
4–6 h	761.0 ± 116.9	614.1 ± 88.0	682.8 ± 100.5	1057.0 ± 126.2	833.9 ± 64.8	723.4 ± 97.0
0–3 h	594.0 ± 110.3	445.3 ± 80.9	1,052.8 ± 135.3	321.1 ± 65.5*	928.4 ± 151.6	467.9 ± 114.5*
3–6 h	1,114.9 ± 174.8	989.3 ± 122.1	1,141.4 ± 171.4	1,482.3 ± 130.9	1,233.3 ± 126.5	1,087.1 ± 159.1
Total 6 h	1,708.9 ± 258.1	1,431.8 ± 140.1	2,260.6 ± 268.1	1,803.4 ± 152.3	2,160.7 ± 215.6	1,555.1 ± 264.8†
Total eye movements during PS	349.5 ± 120.6	398.0 ± 111.0	492.9 ± 99.5	464.0 ± 117.9	435.0 ± 165.7	230.6 ± 85.8
Number of spindles during PS	5.6 ± 1.1	4.8 ± 1.1	4.1 ± 1.2	10.6 ± 1.5*	9.4 ± 2.1	9.6 ± 2.1
Theta rhythm frequency during PS	8.4 ± 0.1	8.4 ± 0.1	8.2 ± 0.1	8.2 ± 0.1	8.1 ± 0.1	8.1 ± 0.2

*$p < 0.02$.
†$p < 0.05$.

followed by PS increased (8.1 ± 2.4 vs. 12.3 ± 3.7; $p < 0.02$). The theta rhythm frequency was unchanged by the compound.

Paradoxical Sleep

At 2.5 mg/kg, the only effect was a decrease of PS duration during the first 2 h of recording (323.9 ± 50.7 s vs. 148 ± 44.5; $p < 0.05$). At 5 mg/kg, PS latency of appearance increased (42 ± 6.3 min vs. 98.3 ± 10.6; $p < 0.02$). The number of PS episodes decreased during the first 3 h of recording (13.3 ± 1.4 vs. 4.4 ± 0.7; $p < 0.02$) and PS amount also decreased during the same period (1,052.8 ± 135.3 s vs. 321.1 ± 65.5; $p < 0.02$). Moreover, the number of spindles appearing in established PS, which reflects less stabilized PS, increased (4.1 ± 1.2 vs. 10.6 ± 1.5; $p < 0.02$). At 7.5 mg/kg, PS latency of occurrence increased (36 ± 5.2 min vs. 108.4 ± 27.5; $p < 0.02$), PS episodes decreased in number during the first 3 h (11.7 ± 1 vs. 5.7 ± 1.3; $p < 0.02$), and PS duration was decreased during the same period (928.4 ± 151.6 s vs. 467.9 ± 114.5; $p < 0.02$). For this high dose, PS duration was decreased over the total recording time (2,160.7 ± 215.6 s vs. 1,555.1 ± 264.8; $p < 0.05$).

It is worthwhile to mention that zolpidem did not affect the number of eye movements or the hippocampal theta rhythm frequency during PS.

DISCUSSION

The major result of this study is that, contrary to barbiturates and benzodiazepines, zolpidem does not greatly increase IS at the expense of PS, even at high doses. The mean duration of IS episodes is only slightly increased at the highest dose although IS episodes increase in number and total duration at 5 mg/kg only. A similar result is obtained under barbiturates, which increase IS at the expense of PS at low doses (IS appears instead of PS) and at high doses decrease IS (9). IS episodes not followed by PS increase from 5 mg/kg, which reflects a difficulty to enter PS. This fact is reinforced by the latency of PS appearance, which also increases. At 2.5 mg/kg, a true hypnotic dose as already mentioned, the only effect on PS is a short-lasting decrease of its duration with a rebound because the PS total amount during the 6-h recording is unchanged. The transient decrease lasts 3 h at 5 mg/kg also with a rebound. Only at 7.5 mg/kg is the PS decrease during the first 3 h not compensated during the following hours. Moreover, contrary to diazepam and triazolam (G. Gandolfo, R. Scherschlicht, and C. Gottesmann, submitted), the eye movements during PS are not decreased in number and the theta rhythm frequency also is not decreased. These results are also confirmed by the absence of an effect of zolpidem on the phasic phenomena and the theta rhythm during PS in cats, in contrast to benzodiazepines (2). Finally, at 5 mg/kg the number of spindles slightly increases in established PS, which reflects a less well-stabilized stage. This fact disappears at 7.5 mg/kg but PS latency still increases. The increase in PS latency may be the consequence of the reinforcement of slow-wave sleep by zolpidem, especially if we consider that the compound was given to animals recorded during the light period, where the sleep percent is maximum. Also, zolpidem presenting a selective hypnotic profile at 1 mg/kg, it is possible that, at high doses, the specificity of zolpidem on sleep structures is partly decreased because at these doses it is less "specific" for ω_1-receptors. These results in rats are in agreement with those obtained after zolpidem treatment in humans, where zolpidem respects the physiological sleep architecture (14).

Consequently, zolpidem, a new-generation hypnotic, respects more than previously studied hypnotics the association IS–PS. However, zolpidem, like barbiturates and benzodiazepines, binds to $GABA_A$ receptors. More precisely, it was first reported that zolpidem binds to the benzodiazepine receptor. Nevertheless, it rapidly became clear that, while benzodiazepines have primarily anxiolytic, anticonvulsant, and myorelaxant actions and only secondarily sedative effects, zolpidem has primarily hypnotic effects (4). It now appears that zolpidem has selectivity for $GABA_A$ receptors containing the α_1-subunit (1,16).

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