Role of $GABA_A$ Receptors in the Regulation of Sleep: Initial Sleep Responses to Peripherally Administered Modulators and Agonists

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Summary: This paper reviews the sleep effects of systemically administered agonistic modulators of $GABA_A$ receptors, including barbiturates, benzodiazepines, zolpidem, zopiclone and neuroactive steroids, and the selective $GABA_A$ agonists muscimol and THIP. To assess the involvement of $GABA_A$ receptors in the physiologic regulation of sleep, the article emphasizes the hypnotic properties shared by agonistic modulators and by the selective agonists of the $GABA_A$ receptor complex. In both rats and normal sleeping individuals, agonistic modulators are able to reduce sleep latency, increase sleep continuity, and promote non-rapid-eye-movement (NREM) sleep as well as the occurrence of spindles. Furthermore, nearly all of these compounds have been shown to attenuate slow-wave activity (SWA) and to suppress the occurrence of REM sleep. In the same species, $GABA_A$ agonist(s) do not seem to affect sleep latency or REM sleep time, but may increase sleep continuity and NREM sleep and augment SWA while depressing spindle activity in humans. The distinct sleep effects of $GABA_A$ agonists may be due to their unspecific stimulation of $GABA_A$ receptors throughout the brain, and to the fact that they are poor substrates for uptake and probably exert more tonic effects than liberated GABA. If so, the involvement of $GABA_A$ receptors in the various aspects of sleep can be inferred more accurately from the hypnotic effects of agonistic modulators. This implies that an activation of $GABA_A$ receptors plays a crucial role in the initiation and maintenance of NREM sleep and in the generation of sleep spindles, but disrupts the processes underlying slow EEG components and the triggering of REM sleep.

Key words: $GABA_A$ receptor agonists/agonistic modulators; sleep state; spectral analysis; mammalia

A WEALTH OF EVIDENCE indicates that γ-aminobutyric $acid_A$ ($GABA_A$) receptors play an important role in the regulation of sleep. GABA is the major inhibitory neurotransmitter in the mammalian central nervous system (CNS), and its depressant actions are predominantly mediated by $GABA_A$ receptors. Moreover, numerous compounds that influence $GABA_A$ receptor functioning are established to induce pronounced changes in sleep-wake behavior. Analysis of the sleep effects of such substances may give insight into the involvement of $GABA_A$ receptors in the regulation of the various aspects of sleep, including the initiation and maintenance of the sleep stages non-rapid-eye-movement (NREM) sleep and REM sleep, as well as the sleep state-specific electroencephalographic (EEG) signals. After a brief overview of the configuration and functioning of $GABA_A$ receptors, the present paper summarizes reported acute sleep responses to systemically administered substances that either potentiate or mimic the action of GABA at the $GABA_A$ receptor in the most extensively studied mammalian species—rats, cats and humans. In view of the vast literature—which precludes a complete review—as well as the many causes and types of insomnia, the review of human data was restricted to healthy adults without sleep

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complaints. On the basis of the general hypnotic properties of substances with common mechanisms of action, we speculate on the involvement of $GABA_A$ receptors in the sleep-regulating processes, with emphasis on NREM sleep.

$GABA_A$ Receptors

Four decades ago, the neutral amino acid GABA was identified as a normal constituent of the mammalian brain. Subsequent research demonstrated that GABA is one of the most prevalent neurotransmitters in the CNS, reaching concentrations in the order of moles/g.[1,2] Depending on the brain region, approximately 20% to 50% of all synapses use GABA as their neurotransmitter,[3,4] which indicates that a large percentage, if not all, of the central neurons are under GABA control. Synaptically released GABA exerts its effects via the specific interaction with three types of membrane-bound GABA receptors, classified as the functionally and pharmacologically differing $GABA_A$, $GABA_B$ and $GABA_C$ receptors (for review, see 5-8). In nearly all brain areas, $GABA_A$ receptors are the most abundant.[9] They constitute fast-acting ligand-gated anion channels. Upon the activation by GABA or structural analogs, the membrane permeability for anions—primarily chloride ions—increases. Because the chloride ion concentration within neurons is usually low, GABAergic transmission in most cases produces a slight, short-lasting hyperpolarization and thus a reduced excitability of the recipient neuron. Of high pharmacologic interest is the fact that $GABA_A$ receptors are endowed with additional allosteric binding sites, including sites for barbiturates, benzodiazepines, and specific neuroactive steroids (for review, see 5, 10). Ligands that interact with these binding sites induce a conformational change in $GABA_A$ receptors, and thereby modulate $GABA_A$-receptor functioning. Such compounds may either enhance (agonistic modulators) or attenuate (inverse agonistic modulators) GABA-induced $GABA_A$ receptor currents or inhibit the action of agonistic modulators (antagonistic modulators).

Molecular biology studies showed that $GABA_A$ receptors are very heterogeneous. The $GABA_A$ receptor complex is a pentameric glycoprotein, composed of combinations of multiple polypeptide subunits. To date, five structurally related subunit families have been identified (α, β, γ, δ and ρ), each with its own isoforms, with 70% to 80% sequence identity within each subunit family (for review, see 5,11). Heterologs expression studies demonstrated that a channel with the properties of native receptors requires the combination of at least one α-, one β- and one γ-subunit.[12,13] The precise combination of protein subunits determines the physiologic and pharmacologic properties of a $GABA_A$ receptor (for review, see 11, 14, 15). While most native $GABA_A$ receptors are susceptible to modulation by barbiturates and neuroactive steroids, many are insensitive to benzodiazepines.[12] Benzodiazepine sensitivity requires the coexpression of the $\gamma 2$ subunit with α- and β-subunits. Furthermore, benzodiazepine pharmacology depends on the type of α subunit. The so-called "benzodiazepine type I receptor," which displays high-affinity binding for certain benzodiazepine receptor ligands—including the benzodiazepines quazepam and cinolazepam and the imidazopyridines zolpidem and alpidem—appears to contain a $\alpha 1$ subunit, whereas the benzodiazepine type II receptor, which has a low affinity for these drugs, contains $\alpha 2$- or $\alpha 3$-subunits (for review, see 5, 10, 14). Both receptor types are present throughout the brain, but are distributed unevenly: type I receptors are selectively enriched in the cerebellum, and type II receptors predominate in limbic structures, such as the hippocampus, and also occur in spinal motor neurons (for review, see 5, 14).

Effects of Agonistic Modulators of $GABA_A$ Receptors on Sleep

Barbiturates.—Barbiturates have been used as sedatives since the early 1900s. They are less widely prescribed today because of their toxicity, the rapid development of drug-tolerance and physical and psychological dependency, and the occurrence of rebound phenomena after discontinuation of barbiturate treatment. Sedative barbiturates are potent agonistic modulators of $GABA_A$ receptors: by interacting with $GABA_A$ receptor-associated barbiturate binding sites, they potentiate the GABA-induced chloride ion flux by increasing the average open duration of the chloride channels without altering channel conductance and opening frequency.[16,17] At very high, anesthetic concentrations, barbiturates are able to open $GABA_A$-associated chloride channels directly, in the absence of GABA.[18,19]

The bulk of animal studies on the hypnotic effects of barbiturates employed the loss and reappearance of the righting reflex to assess sleep time. In one polysomnographic study, 1 and 10 mg/kg phenobarbital doses were administered intraperitoneally (ip) to cats. Compared with placebo recordings, phenobarbital dose-dependently decreased wakefulness, promoted NREM sleep, and suppressed REM sleep.[20] In the same species, sodium barbital was chronically administered intragastrically twice a day in slowly increasing doses. In comparison to control recordings, barbital persistently increased NREM sleep and markedly decreased REM sleep by a reduction in the number of REM episodes.[21] Pentobarbital, given intravenously (iv) in a dose of 10 mg/kg, has also been found to prevent the occurrence of REM episodes. Interestingly, periodic bursts of ponto-geniculo-occipital (PGO) waves persisted, but in parallel with NREM-associated EEG signals, indicating a barbiturate-induced dissociation of REM-related events.[22] Another study evaluated the immediate sleep

response to 15 and 30 mg/kg pentobarbital, administered ip to rats during the light period (the circadian rest phase of this species). In comparison to placebo, pentobarbital dose-dependently shortened NREM latency and decreased REM sleep time, which was associated with a delay in the occurrence of the first REM episode and with a decrease in the number of REM episodes. Furthermore, it promoted a substate of NREM sleep which is characterized by the occurrence of long-lasting, high-amplitude spindle-like (≈11-16 Hz) waves on a background of regular theta (6-9 Hz) activity.[23] This substate usually preceeds REM sleep, and is therefore termed pre-REM sleep in this paper, but has also been called intermediate stage[23] and transition type sleep.[24] The pentobarbital-induced enhancement of pre-REM sleep has been confirmed in cats.[25] Experiments focused on alterations in the cortical EEG performed on various mammalian species found that barbiturates, such as pentobarbital and phenobarbital, administered at not-too-high doses, evoke bursts of rhythmic waves in the frequency range of 6-13 Hz, with a waxing-and-waning amplitude, reminiscent of spindles occurring during spontaneous sleep. Additional subcortical recordings revealed that similar signals appear simultaneously in the thalamus (for review, see 26), the brain structure which generates spindle oscillations (for review, see 27).

Various reports exist on the immediate influence of barbiturates on nocturnal sleep in healthy noninsomniac subjects. Phenobarbital, given orally (po) in doses of 120 mg or more, has relatively small effects, consisting of a slight decrease in the percentage of wakefulness and NREM stage 1, an increase of slow-wave sleep (NREM stages 3 and 4), and a prolongation of REM sleep latency.[28,29] Thiopental (300 mg), given iv to four subjects, affected sleep in a similar fashion.[28] More elaborate investigations revealed that a single oral bedtime dose of 100 mg pentobarbital significantly shortens sleep latency, increases total sleep time and NREM sleep without significantly affecting the light NREM stages 1 and 2 or slow-wave sleep, and reduces the number of awakenings and body movements. Additionally, it decreases the percentage of REM sleep, due to a delay of REM latency and to a decrease of both the number and duration of the REM episodes.[30,31] Amobarbital, given shortly before lights-off in a dose of 200 mg (n=6) and 400 mg (n=3), appeared to shorten sleep-onset latency and decrease percentage stage 1 and increase stage 2, while having minimal effects on slow-wave sleep time. Furthermore, amobarbital delayed the latency to REM sleep, decreased percentage REM sleep and movement time as well as the number of switches to wakefulness, stage 1 or movement time.[32] Comparable effects have been observed in subjects after the oral ingestion of 400 mg heptobarbital[33] and various doses (dose range: 100-200 mg) of secobarbital.[34-36]

Taken together, most hypnotic barbiturates seem to increase the ability to fall and stay asleep, promote NREM and/or pre-REM sleep, and increase the occurrence of spindles during NREM sleep, and to decrease REM sleep, which is especially related to a reduction in the number of REM episodes and—at least in humans—possibly also to a decrease in the duration of the REM episodes.

Benzodiazepines.—Following their introduction in the 1960s, benzodiazepines rapidly became the most frequently prescribed hypnotics. They have a good safety and tolerability profile, but chronic use may cause tolerance and dependence, and drug withdrawal may produce rebound insomnia (for review, see 37, 38). Sedative-hypnotic benzodiazepines augment the action of GABA at the $GABA_A$ receptor by increasing the frequency of chloride channel opening, with little effect on channel conductance or channel open time.[16,39]

Placebo-controlled studies in the rat showed that the benzodiazepines flurazepam (dose range: 15-30 mg/kg), triazolam (dose range: 0.1-0.6 mg/kg), midazolam (dose range: 3-10 mg/kg) and diazepam (dose range: 1-2 mg/kg), administered ip or po, systematically shorten sleep latency and increase NREM sleep. Possibly related to the small amount of spontaneously occurring sleep, the last effect is most prominent during the dark period. Furthermore, these drugs consistently delay REM latency and decrease REM sleep time.[40-48] Midazolam has been shown to reduce the number of REM episodes,[47] whereas triazolam and diazepam have been found to shorten the duration of the REM episodes.[23] Experiments in which pre-REM was scored as a separate state revealed that benzodiazepines markedly increase pre-REM sleep time.[23] In contrast to the sleep-inducing effect in rats, benzodiazepines promote wakefulness in the cat. Administration to cats of flunitrazepam (0.1 and 1 mg/kg, po and 0.1 mg/kg, ip), nitrazepam (0.03 mg/kg, ip and 0.25-1.5 mg/kg intramuscularly [im]), triazolam (0.01 mg/kg, ip), midazolam (1 mg/kg, ip) and diazepam (0.2 and 1 mg/kg, ip and 0.25-2 mg/kg, im) markedly increased wakefulness and decreased NREM sleep and REM sleep. The decrease of both sleep states was caused by a delayed latency as well as by a reduced number and duration of the NREM and REM episodes.[20,49-52] In spite of the species-dependent changes in sleep time, benzodiazepines appear to induce comparable alterations in the EEG signals. Spectral analysis of the EEG signals after the administration of 1 mg/kg diazepam po to cats and of 3 mg/kg midazolam ip to rats yielded a depression of slow (≤10 Hz) EEG components and elevations in the frequency range of spindles during NREM sleep, and increases in the high- (>10 Hz) frequency bands during both NREM and REM sleep in rats as well as cats.[45,47,52] Analysis of the temporal development of EEG activity within NREM episodes showed that midazolam produces a

general suppression of slow-wave activity (SWA; 0.5-4 Hz) and an increase of spindle activity, without affecting their intraepisodic time course.[47]

The acute effect of benzodiazepine hypnotics on night sleep in healthy good-sleeping subjects has been studied extensively. For instance, flunitrazepam, given po shortly before retiring in doses varying between 1 and 6 mg, has been shown to increase total sleep time dose-dependently, which results from a shortened sleep latency, a reduction in the number of awakenings, and a prominent promotion of stage 2. In contrast, it decreases stage 1 and slow-wave sleep as well as REM sleep, caused by a delay in the occurrence of the first REM episode and by a reduction of the number of REM episodes.[53-57] EEG spectral analysis revealed that flunitrazepam markedly attenuates low-frequency EEG activity and augments EEG activity in the frequency range of sleep spindles during NREM sleep, and enhances high-frequency EEG activity, especially during REM sleep.[56,57] Highly similar effects have been reported for other benzodiazepines, such as the long-acting benzodiazepines nitrazepam[32,34,53,58-62] and flurazepam,[31,57,62-66] the medium-acting benzodiazepines temazepam,[67,68] and bromazepam,[53] and the short-acting benzodiazepines triazolam[57,62,67-71] and midazolam.[57,72] More detailed analyses revealed that benzodiazepines evoke a general suppression of SWA and enhancement of spindle activity during NREM sleep, but have little effect on their temporal evolution within and across NREM episodes.[73-75] The attenuation of SWA appeared to be related to a reduction in both the amplitude and number of slow waves,[76-78] while the augmentation of spindle activity is accounted for by an increase in spindle density, and possibly also by an increased spindle duration.[54,78,79]

Taken together, although the reactivity to benzodiazepine hypnotics differs between species, they have many sleep effects in common. In the cat these drugs induce alertness. In both rats and humans, they increase the ability to fall asleep and to stay asleep and promote NREM and/or pre-REM sleep. Independent of the species, benzodiazepines increase the occurrence of spindles and depress slow EEG waves during NREM sleep and REM sleep time, which is due to an initial suppression of the occurrence of REM sleep but possibly also to a decrease in the duration of REM episodes.

Zolpidem and zopiclone.—In the 1980s, two new hypnotics—zolpidem and zopiclone—were introduced. Both drugs are nonbenzodiazepine agonistic modulators of $GABA_A$ receptors that seem to have a lower tolerance and dependency potential than barbiturates and classical benzodiazepines (for review, see 37, 80-83).

In contrast to most benzodiazepines, which bind nonselectively to benzodiazepine type I and II receptors, the imidazopyridine derivative zolpidem binds specifically to the benzodiazepine type I receptor. Experiments in rats showed that zolpidem, administered po in doses between 1 and 10 mg/kg and ip in doses from 2.5 up to 10 mg/kg during the light or dark period, dose-dependently reduces sleep latency, increases NREM sleep, hardly affects pre-REM sleep time, increases REM latency and—due to a reduction in the number of REM episodes—decreases REM sleep.[23,48,84] In the cat, zolpidem (1 and 10 mg/kg, po) initially produces wakefulness, thereby delaying the latency to both NREM and REM sleep. Thereafter, it promotes NREM as well as pre-REM sleep.[84]

In noninsomniac subjects, zolpidem exerts only moderate and partially age-dependent effects on nocturnal sleep. Most studies in young subjects found that zolpidem, given po in doses between 2.5 and 30 mg shortly before retiring, tends to shorten sleep-onset latency, has no consistent effect on total sleep time, the number of awakenings, and time spent in the NREM sleep stages, but may slightly decrease REM sleep and delays REM latency.[85-88] In middle-aged and geriatric subjects without sleep complaints, similar doses of zolpidem appeared more likely to reduce sleep latency, increase total sleep time, reduce the number of awakenings, decrease stage 1 and increase stage 2, while having little effect on slow-wave sleep, and to prolong REM latency and decrease REM sleep.[85,89] In spite of its marginal effects on the sleep stages, all-night spectral analysis showed that zolpidem markedly reduces low-frequency (≤10 Hz) EEG activity and augments EEG activity in the spindle frequency range during NREM sleep in young normal subjects.[88]

The second nonbenzodiazepine hypnotic, zopiclone, is a cyclopyrrolone derivate. It probably binds at a physically distinct domain on the $GABA_A$ receptor that is closely linked to a benzodiazepine binding site.[5,15] In rats, 2.5 and 10 mg/kg zopiclone, administered ip during the light period, was found to decrease total time dose-dependently in wakefulness and pre-REM sleep, and to increase NREM sleep in comparison to placebo.[90] A comparable study in the same species yielded a dose-related shortening of NREM sleep latency and a decrease of total time in pre-REM sleep and REM sleep, which was related to an increased latency as well as to a reduction in the number of pre-REM and REM episodes.[91] In cats, various doses of zopiclone (1 and 5 mg/kg, po) have been shown to promote wakefulness and to decrease all sleep stages.[51,92]

Zopiclone appears to have relatively mild hypnotic properties in normal subjects. The strongest effects were observed in a study on the influence of zopiclone on all-night sleep recordings in six healthy middle-aged subjects.[93] Zopiclone was administered orally at bedtime in doses of 5, 7.5, and 10 mg. Although not all changes reached the level of statistical significance, zopiclone dose-dependently reduced sleep-onset latency, increased total

sleep time due to marked increases in stage 2, slightly decreased stage 1, slow-wave sleep and REM sleep, significantly increased REM latency, and reduced the number of awakenings. Other investigators found qualitatively comparable effects of zopiclone, except for its influence on slow-wave sleep. While zopiclone tendentially decreased slow-wave sleep in the above-mentioned study, similar doses hardly affected[72,94-97] or even slightly increased[98-100] slow-wave sleep in studies with young subjects. Nevertheless, spectral analysis of EEG signals during NREM sleep revealed a prominent suppression of slow (≤10 Hz) EEG components and enhancements in spindle frequencies.[72,100] Furthermore, zopiclone was found to evoke a general decrease in SWA and increase in spindle activity during NREM sleep, without disrupting the inter- or intraepisodic time course.[75] The depression of SWA seems to be due to a pronounced attenuation of the slow-wave amplitude,[101] while enhanced spindle activity has been shown to be caused by an increased spindle density.[102]

Taken together, except for the fact that neither zolpidem nor zopiclone promotes pre-REMS in the rat, the sleep effects of these drugs are qualitatively rather similar to those induced by benzodiazepines: they produce alertness in the cat, whereas in both humans and rats they are able to shorten sleep-onset latency, increase sleep consolidation, increase NREM sleep, facilitate the occurrence of sleep spindles, and, though not detectable by visual scoring, attenuate low-frequency EEG signals during NREM sleep and suppress the appearance of REM episodes.

Neuroactive steroids.—Various steroid hormones lack activity at classical intracellular steroid receptors. Some of these steroids selectively modulate $GABA_A$ receptor functioning, possibly through a unique binding site that is distinct from those for barbiturates and benzodiazepines (for review see 5, 10, 103-105). The most potent natural-occurring steroidogenic agonistic modulators are the ring A-reduced metabolite(s) of progesterone, 3α-hydroxy-5α- (allopregnanolone) and 3α-hydroxy-5β-dihydroprogesterone (pregnanolone), and of deoxycorticosterone, 3α-hydroxy-5α-tetrahydrodeoxycorticosterone (THDOC). At low nanomolar concentrations, they augment GABA-induced $GABA_A$ receptor current by increasing both the frequency and duration of chloride channel opening.[106] Like barbiturates, they directly activate $GABA_A$ receptors at high concentrations.[107,108]

In addition to several other CNS-depressant properties, progesterone is since long known to exert anesthetic actions.[109] However, investigations addressing its precise influence on sleep-wake behavior and the mediating mechanisms have only recently begun. A study on the effects of several doses (30-180 mg/kg, ip) of progesterone administered at light-onset to rats yielded pronounced dose-related alterations, consisting of a reduction of sleep latency, lengthening of the NREM episodes, promotion of preREM sleep and—at the highest dose—a decrease in REM sleep, caused by an increase in REM latency and a reduction in the number of REM episodes. Spectral analysis of the EEG yielded a nonspecific enhancement of high-frequency (≥11 Hz) activity, as well as a non-REMS-specific depression of low-frequency (≤7 Hz) activity and an augmentation in the spindle frequencies.[110] In young subjects, a single oral dose of 300 mg micronized progesterone 1.5 hours before retiring has been shown to increase stage 2, tendentially decrease stage 4 and REM sleep, suppress SWA, and enhance activity in the frequencies >15 Hz during NREM sleep.[111] In both studies, the time course of the progesterone-induced sleep alterations correlated highly with the temporal development of the elevations in the brain and/or plasma concentrations of allopregnanolone and, to a lesser extent, of pregnanolone. These observations indicate that the effects of progesterone on sleep may result from its bioconversion into neuroactive metabolites. Studies investigating the influence of exogenous pregnanolone and allopregnanolone support this notion. In rats, pregnanolone (10-30 mg, ip) and the synthetic analog CDD-3693 (10-30 mg, po), administered during the dark period, were both found to rapidly increase NREM sleep time.[48] Pregnanolone has also been shown to enhance sleep propensity in humans. Analysis of 5-minute polygraphic recordings made at 30-minute intervals yielded an increase in the number of sleep attempts following a single oral early-morning dose of 1000 mg pregnanolone.[112] One study on the influence of two doses of allopregnanolone (5 and 10 mg/kg, ip) administered during the light period to rats did not reveal significant changes in sleep-wake behavior.[113] A comparable experiment, in which allopregnanolone doses of 7.5 and 15 mg/kg were mixed with oil to attain a slower release, revealed dose-related sleep responses, including a shortened sleep latency, increase in pre-REM sleep, attenuation of low-frequency (≤7 Hz) EEG activity, and elevations in the frequency bands of sleep spindles during NREM sleep and an enhancement of high-frequency activity during both NREM- and REM-sleep.[114] To the best of our knowledge, only one study assessed the hypnotic effects of THDOC. It found that THDOC (5 and 10 mg/kg, ip), administered during the light period to rats, shortened sleep latency and increased NREM sleep time in a dose-related fashion.[113] Studies on the influence of the THOC precursor DOC (1 mg and 20 mg, po) on sleep in normal volunteers did not find alterations in the sleep pattern[115,116]; however, it is uncertain whether DOC is able to enter the brain.

Taken together, although the observed sleep effects need to be confirmed and extended, the given findings suggest that steroidogenic agonistic modulators of $GABA_A$ receptors increase the ability to fall asleep, promote NREM

or pre-REM sleep, increase NREM maintenance, enhance the occurrence of spindles and attenuate slow EEG waves during NREM sleep, and may, at high doses, suppress the occurrence of REM episodes.

Effects of Agonists of $GABA_A$ Receptors on Sleep

$GABA_A$ agonists, such as muscimol, isoguvacine, and 4,5,6,7-tetrahydroisoxazolo-pyridin-3-ol (THIP), are structural analogs of GABA, which selectively bind to the GABA recognition sites on the $GABA_A$ receptor complex and thereby directly activate $GABA_A$ receptors. $GABA_A$ agonists open chloride ion channels of the same conductance as GABA, but the average duration of channel opening may differ. Noise analyses suggest that muscimol activates chloride channels of a longer open duration, whereas—for example—THIP and isoguvacine activate chloride channels of a shorter open duration than that opened by GABA.[39]

Experiments on rats showed that the full $GABA_A$ agonist muscimol produces dose-dependent alterations in sleep-wake behavior. Intraperitoneal administration of various doses (0.05-0.4 mg/kg) of muscimol during the light period hardly affected sleep latency.[42,47,117] However, doses ≥0.2 mg/kg appeared to decrease wakefulness and increase NREM sleep and, to a lesser extent, also REM sleep, which is related to a lengthening of the sleep episodes.[47] Furthermore, muscimol was found to enhance low-frequency (<10 Hz) EEG activity during NREM sleep and high-frequency (>10 Hz) activity within REM sleep in a dose-related fashion.[47,117] The enhancement of SWA is associated with an overall increase within the NREM episodes.[47] The partial $GABA_A$ agonist THIP, administered ip in doses of 2 and 4 mg/kg during both the light and dark period, affected neither sleep latency nor pre-REM and REM sleep time. THIP consistently induced a lengthening of the NREM episodes and increased total time spent in NREM sleep. Furthermore, THIP dose-dependently elevated SWA during NREM sleep, caused by an overall increase within the NREM episodes, and slightly enhanced high-frequency (>10 Hz) activity in the EEG during REM sleep.[118,119]

Comparable findings were also observed in humans. A single oral dose of 20 mg THIP, given half an hour before retiring to young normal sleepers, was found to significantly increase slow-wave sleep, without affecting sleep onset latency, REM latency, or REM sleep time, in comparison to placebo. EEG spectral analyis yielded enhancements in the lower frequencies (<9 Hz) and selective decreases in the spindle frequencies during NREM sleep, which were associated with an overall elevation of SWA and attenuation of spindle activity within the NREM episodes.[120]

Taken together, the few available studies on the influence of $GABA_A$ agonists on sleep in rats and volunteers without sleep disturbances suggest that muscimol and THIP do not affect sleep latency. However, they seem able to increase NREM sleep maintenance and to promote slow-wave sleep. They appear to have minimal effects on REM sleep time and distribution and, in humans, attenuate spindle activity. It is at present uncertain whether the last effect is due to a reduction in spindle density, amplitude, and/or duration.

Implications

The agonistic modulators of $GABA_A$ receptors discussed above differ in many respects: barbiturates, benzodiazepines, zolpidem, zopiclone, and neuroactive steroids belong to different chemical families, interact for the most part with distinct binding sites on the $GABA_A$ receptor, produce different changes in chloride channel kinetics, and exhibit different affinities to the various $GABA_A$ receptor subtypes; thus, the regional distribution of susceptible $GABA_A$ receptors differs. Nevertheless, these agents all enhance the response of $GABA_A$ receptors to GABA, which explains the fact that they have many hypnotic properties in common. Though quantitative differences clearly exist, these drugs are able to increase the ability to fall asleep and to stay asleep, increase NREM sleep time, and stimulate the appearance of spindles within NREM sleep. With the exception of the neuroactive steroids, these compounds have all been shown to decrease REM sleep time, which is particularly due to a suppression of the occurrence of REM episodes. Except for barbiturates, whose influence on the state-specific EEG power densities has unfortunately never been assessed in detail, all other drugs have been shown to attenuate low-frequency components during NREM sleep. These findings strongly suggest that an activation of $GABA_A$ receptors plays an important role in the induction and consolidation of NREM sleep, and facilitates the mechanisms responsible for the production of spindles, but inhibits the processes underlying the generation and/or synchronization of slow waves and suppresses REM sleep triggering mechanisms. However, the last interpretation may be premature. As mentioned earlier, in the cat, bursts of PGO waves periodically occur during barbiturate-induced REM suppression.[22] Furthermore, after the administration of several benzodiazepines to human subjects, brief periods of EEG desynchronization have been observed at approximately the time of the first regular REM episode, though not accompanied by rapid eye movements and muscle atonia.[74] Possibly, agonistic modulators of $GABA_A$ receptors inhibit the manifestation of certain REM sleep phenomena and thereby preclude the visual detection of REM sleep. Moreover, based on the finding that the SWA levels attained during benzodiazepine-abort-

ed REM episodes exceed those observed during normal REM episodes, it has been postulated that the attenuation of EEG desynchronization prevents the appearance of REM sleep.[74,121]

As agonistic modulators and agonists respectively potentiate and mimic the action of GABA at the $GABA_A$ receptor, one would expect that these agents have similar effects on sleep. However, this is clearly not the case. In both rats and humans, $GABA_A$ agonists have little effect on sleep latency or REM sleep, but seem to increase sleep continuity and slow-wave sleep and, in humans, to attenuate EEG activity in the frequency range of sleep spindles during NREM sleep. In contrast to the conclusions drawn from the sleep effects of agonistic modulators of $GABA_A$ receptors, these observations would suggest that $GABA_A$ receptors are not involved in mechanisms underlying the triggering of NREM sleep or REM sleep, but play a positive role in NREM sleep maintenance and in the generation and/or synchronization of slow EEG waves and a negative role in spindle-related processes. In view of the finding that agonistic modulators and agonists of $GABA_A$ receptors have opposite effects on sleep, questions arise as to how the differential effects can be explained, and which effects most accurately reflect the involvement of $GABA_A$ receptors in physiological sleep regulation. The distinct sleep effects indicate that the influence of agonistic modulators and agonists of $GABA_A$ receptors on electrical brain activity differs substantially. This notion is supported by observations made in sleep, epilepsy, and drug-discrimination studies. Studies in which benzodiazepines and muscimol were coadministered revealed that these drugs do not augment one another's influence on the sleep states,[42,44,117] but even attenuate one another's effects on the EEG signals during NREM sleep.[117] Whereas $GABA_A$ agonists exacerbate absence (petit mal) epilepsy in epilepsy-prone animals (for references, see 122), various agonistic modulators have been shown to suppress spontaneous, as well as $GABA_A$ agonist-induced, absence epileptic seizures.[46,123] Rats trained to discriminate a benzodiazepine from saline generalize to other benzodiazepines as well as to barbiturates and the neuroactive steroids allopregnanolone and THDOC,[124,125] but not to muscimol or THIP,[126] which indicates that $GABA_A$ agonists do not share discriminative-stimulus effects with agonistic modulators of $GABA_A$ receptors. One explanation for these diverse effects is that $GABA_A$ agonists and agonistic modulators activate different $GABA_A$ receptor subtypes. Whereas muscimol and THIP are likely to activate all functional $GABA_A$ receptors, the responsiveness to agonistic modulators depends on the precise receptor composition. If this postulation were true, one might expect that the massive activation by GABA analogs would dominate and mask the modulation evoked by $GABA_A$ agonistic modulators. As outlined above, this has not been observed in sleep investigations or in absence epilepsy studies. Another obvious explanation is based on the fact that sleep-related processes are regulated by the concerted release of GABA in specific brain areas. Agonistic modulators potentiate the action of endogenously liberated GABA on $GABA_A$ receptors and thus maintain its physiological specificity. In contrast, $GABA_A$ agonists unselectively stimulate $GABA_A$ receptors throughout the CNS, thereby inevitably producing different effects than agonistic modulators. In addition, GABA and GABA analogs may exert differential net-effects on $GABA_A$ receptor functioning in in vivo conditions. Synaptically released GABA is rapidly removed by high-affinity uptake mechanisms into neurons and glial cells,[127] which confine its action temporally and spatially. As both muscimol and THIP are poor substrates for uptake, they probably produce more tonic effects than liberated GABA, which is likely to have a considerable impact on neuronal activity. In line with this postulate, the GABA uptake inhibitor tiagabine, which elevates the steady state level of GABA, has recently been shown to have comparable effects on sleep EEG in the rat to muscimol and THIP.[128] If the striking sleep effects of $GABA_A$ agonists are caused by an unphysiological widespread and/or tonic activation of $GABA_A$ receptors, then they do not reflect the involvement of $GABA_A$ receptors in sleep regulating processes. We therefore assume that the role of $GABA_A$ receptors in physiological sleep-wake regulation can be more accurately inferred from the sleep effects of agonistic modulators of $GABA_A$ receptors, which implies that $GABA_A$ receptors are positively involved in the induction and consolidation of NREM sleep as well as in the production of spindles, but not in the generation of slow waves.

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REFERENCES

1. Fahn S, Côté LJ. Regional distribution of γ-aminobutyric acid (GABA) in brain of the Rhesus monkey. J Neurochem 1968; 15: 209-13.

2. Perry TL, Berry K, Hansen S, Diamond S, Mok C. Regional distribution of amino acids in human brain obtained at autopsy. J Neurochem 1971; 18: 513-9.

3. Bloom FE, Iversen LL. Localizing 3H-GABA in nerve terminals of rat cerebral cortex by electron microscopic autoradiography. Nature 1971; 229: 628-30.

4. Roberts E, Frankel S. γ-aminobutyric acid in brain: its formation from glutamic acid. J Biol Chem 1974; 187: 55-63.

5. Sieghart W. Structure and pharmacology of γ-aminobutiric $acid_A$ receptor subtypes. Pharmacol Rev 1995; 47: 181-234.

6. Deisz RA. Electrophysiology of $GABA_B$ receptors.The GABA receptors. In: Enna SJ, Bowery NG, eds. Totowa, NJ: The Humana Press Inc., 1997: 157-207.

7. Johnston GAR. Molecular biology, pharmacology and physiology of $GABA_C$ receptors. In: Enna SJ, Bowery NG, eds. The GABA receptors. Totowa, NJ: The Humana Press Inc., 1997:297-323.
8. Krogsgaard-Larsen P, Frølund B, Ebert B. $GABA_A$ receptor agonists, partial agonists, and antagonists. In: Enna SJ, Bowery NG, eds. The GABA receptors. Totowa, NJ: The Humana Press Inc., 1997:37-81.
9. Young AB, Chu D. Distribution of $GABA_A$ and $GABA_B$ receptors in mammalian brain: potential targets for drug development. Drug Dev Res 1990;21:161-7.
10. Macdonald RL, Olsen RW. $GABA_A$ receptor channels. Annu Rev Neurosci 1994;17:569-602.
11. Stephenson FA. The $GABA_A$ receptors. Biochem J 1995;310:1-9.
12. Pritchett DB, Sontheimer H, Shivers BD, Ymer S, Kettenmann H, Schofield PR, Seeburg PH. Importance of a novel $GABA_A$ receptor subunit for benzodiazepine pharmacology. Nature 1989;338:582-5.
13. Sigel ER, Baur R, Trube G, Möhler H, Malherbe P. The effect of subunit composition of rat brain $GABA_A$ receptors on channel function. Neuron 1990;5:703-11.
14. Lüddens H, Korpi ER, Seeburg PH. $GABA_A$/benzodiazepine receptor heterogeneity: neurophysiological implications. Neuropharmacology 1995;34:245-54.
15. Johnston GAR. $GABA_A$ receptor pharmacology. Pharmacol Ther 1996;69:173-98.
16. Study RE, Barker JL. Diazepam and (-)-pentobarbital: fluctuation analysis reveals different mechanisms for potentiation of γ-aminobutyric acid responses in cultured central neurons. Proc Natl Acad Sci USA 1981;78:7180-4.
17. Macdonald RL, Rogers CJ, Twyman RE. Barbiturate regulation of kinetic properties of the $GABA_A$ receptor channel of mouse spinal cord neurones in culture. J Physiol 1989;417:483-500.
18. Macdonald RL, Barker JL. Anticonvulsant and anesthetic barbiturates: different postsynaptic actions in cultured mammalian neurons. Neurology 1979;29:432-47.
19. Bormann J. Electrophysiology of $GABA_A$ and $GABA_B$ receptor subtypes. Trends Neurosci 1988;11:112-6.
20. Scherschlicht R, Marias J, Schneeberger J, Steiner M. Model insomnia in animals. In: Koella WP, eds. Sleep 1980. 5th Europ. Congr. Sleep Research., Amsterdam 1980. Basel: Karger, 1981:147-55.
21. Hinman DJ, Okamoto M. Sleep patterns in cats during chronic low-dose barbiturate treatment and withdrawal. Sleep 1984;7:69-76.
22. Hara T, Masuda K, Miyake H. Effects of psychotropic drugs on the PGO waves occurring in REM sleep and on the reserpine-induced PGO waves. In: Weitzman ED, eds. Advances in Sleep Research, Vol. 2. New York: Spectrum Publications Inc., 1975:131-54.
23. Gottesmann C, Gandolfo G, Arnaud C, Gauthier P. Influence of three generations of hypnotics on the intermediate stage and paradoxical sleep in the rat: a synopsis. Eur J Neurosci 1998;2:409-14.
24. Neckelmann D, Ursin R. Sleep stages and EEG power spectrum in relation to acoustical stimulus arousal thresholds in the rat. Sleep 1993;16:467-77.
25. Gottesmann C, Gandolfo G, Zernicki B. Intermediate stage of sleep in the cat. J Physiol (Paris) 1984;79:365-72.
26. Schallek W, Schlosser W. Neuropharmacology of sedatives and anxiolytics. Mod Probl Pharmacopsych 1979;14:157-73.
27. Steriade M, McCormick DA. Thalamocortical oscillations in the sleeping and aroused barin. Science 1993;262:679-85.
28. Lester BK, Guerrero-Figueroa R. Effects of some drugs on electroencephalographic fast activity and dream time. Psychophysiology 1966;2:224-36.
29. Rosadini G, Masturzo P, Rodriguez G, Murialdo G, Montano V, Bonura ML, Polleri A. Effects of a single oral dose of phenobarbital on prolactine, growth hormone and luteinizing hormone in normal women. Acta Endocrinologica 1983;103:309-14.
30. Baekland F. Pentobarbital and dextroamphetamine sulfate: effects on the sleep cycle in man. Psychopharmacologia 1967;11:388-96.
31. Hartmann E. The effect of four drugs on sleep patterns in man. Psychopharmacologia 1968;12:346-53.
32. Haider I, Oswald I. Effects of amylobarbitone and nitrazepam on the electrodermogram and other features of sleep. Brit J Psychiat 1971;118:519-22.
33. Oswald I, Berger RJ, Jaramillo RA, Keddie KMG, Olley PC, Plunkett GB. Melancholia and barbiturates: a controlled EEG, body and eye movement study of sleep. Brit J Psychiat 1963;109:66-78.
34. Lehmann HE, Ban TA. The effect of hypnotics on rapid eye movement (REM). Int J Clin Pharmacol 1968;1:424-7.
35. Lester BK, Coulter JD, Cowden LC, Williams HL. Secobarbital and nocturnal physiological patterns. Psychopharmcologia 1968;13:275-86.
36. Allnutt MF, O'Connor PJ. Comparison of the encephalographic, behavioral and subjective correlates of natural and drug-induced sleep at atypical hours. Aerosp Med 1971;42:1006-10.
37. Lader M. Rebound insomnia and newer hypnotics. Psychopharmacology 1992;108:248-55.
38. Walsh JK, Engelhardt CL. Trends in the pharmacologic treatment of insomnia. J Clin Psychiat 1992;53 Suppl:10-7.
39. Barker JL, Harrison NL, Mariani AP. IV. Benzodiazepine pharmacology of cultured mammalian CNS neurons. Life Sci 1986;39:1959-68.
40. Monti JM, Altier H, D'Angelo L. Diazepam, GABA agonists and antagonists and the sleep-wakefulness cycle. In: Passouant P, Oswald I, eds. Pharmacology of the states of alertness. Oxford: Pergamon Press, 1979:65-71.
41. Meltzer LT, Serpa KA. Assessment of hypnotic effects in the rat: influence of the sleep-awake cycle. Drug Dev Res 1988;14:151-9.
42. Mendelson WB, Martin JV. Effects of muscimol and flurazepam on the sleep EEG in the rat. Life Sci 1990;47:PL99-101.
43. Edgar DM, Seidel WF, Dement WC. Triazolam-induced sleep in the rat: influence of prior sleep, circadian time, and light/dark cycles. Psychopharmacology 1991;105:374-80.
44. Mendelson WB, Monti D. Do benzodiazepines induce sleep by a GABAergic mechanism? Life Sci 1993;53:PL81-7.
45. Lancel M, Crönlein TAM, Müller-Preuß P, Holsboer F. Pregnenolone enhances EEG delta activity during non-rapid eye movement sleep in the rat, in contrast to midazolam. Brain Res 1994;646:85-94.
46. Depoortere H, Françon D, van Luijtelaar ELJM, Drinkenburg WHIM, Coenen AML. Differential effects of midazolam and zolpidem on sleep-wake states and epileptic activity in WAG/Rij rats. Pharmacol Biochem Behav 1995;51:571-6.
47. Lancel M, Crönlein TAM, Faulhaber J. Role of $GABA_A$ receptors in sleep regulation: differential effects of muscimol and midazolam on sleep in rats. Neuropsychopharmacology 1996;15:63-74.
48. Edgar DM, Seidel WF, Gee KW, Lan NC, Field G, Xia H, Hawkinson JE, Wieland S, Carter RB, Wood PL. CCD-3693: an orally bioavailable analog of the endogenous neuroactive steroid, pregnanolone, demonstrates potent sedative hypnotic actions in the rat. J Pharmacol Exp Ther 1997;282;420-9.
49. Lanoir J, Killam EK. Alteration in the sleep-wakefulness patterns by benzodiazepines in the cat. Electroenceph Clin Neurophysiol 1968;25:530-42.
50. Polc P, Haefely W. Effects of flunitrazepam on the sleep of cats. In: Levin P, Koella WP. eds. Sleep 1974. 2nd Europ. Congr Sleep Res, Rome 1974. Basel: Karger, 1975:303-5.
51. Depoortere H, Decobert M, Granger P, Riou-Merle F. Hypnotics: clinical value of pharmaco-EEG methods. Neuropsychobiology 1986;16:157-62.
52. Hashimoto T, Hamada C, Wada T, Fukuda N. Comparative study on the behavioral and EEG changes induced by diazepam, buspirone and a novel anxioselective anxiolytic, DN-2327, in the cat. Neuropsychobiology 1992;26:89-99.
53. Gaillard JM, Schulz P, Tissot R. Effects of three benzodiazepines (nitrazepam, flunitrazepam and bromazepam) on sleep of normal sub-

jects, studied with an automatic sleep scoring system. Pharmakopsychiat 1973;6:207-17.
54. Monti JM, Altier H. Flunitrazepam (Ro 5-4200) and sleep cycle in normal subjects. Psychopharmacologia (Berl) 1973;32:343-9.
55. Cerone G, Cirignotta F, Coccagna G, Ferro Milone F, Lion P, Lorizio A, Lugaresi E, Mantovani M, Muratorio A, Murri L, Mutani R, Roccio A. All-night polygraphic recordings on the hypnotic effects of a new benzodiazepine: flunitrazepam (Ro 5-4200, rohypnol®). Europ Neurol 1974;11:172-9.
56. Gaillard JM, Blois R. Differential effects of flunitrazepam on human sleep in combination with flumazenil. Sleep 1989;12:120-32.
57. Borbély AA, Mattmann P, Loepfe M, Strauch I, Lehmann D. Effect of benzodiazepine hypnotics on all-night sleep EEG spectra. Hum Neurobiol 1985;4:189-94.
58. Lechner H. Effect of mogadon on night-time sleep. Prog Brain Res 1965;18:225-6.
59. Oswald I, Priest RG. Five weeks to escape the sleeping-pill habit. Brit Med J 1965;2:1093-5.
60. Gastaut H, Lob H, Papy JJ. Action of mogadon on the stages of REM sleep. Electroenceph Clin Neurophysiol 1967;23:288.
61. Adam K, Adamson L, Brezinová V, Hunter WM, Oswald I. Nitrazepam: lastingly effective but trouble on withdrawal. Brit Med J 1976;1:1558-60.
62. Ogura C, Nakazawa K, Majima K, Ueda H, Umezawa Y, Wardell WM. Residual effects of hypnotics: triazolam, flurazepam, and nitrazepam. Psychopharmacology 1980;68:61-5.
63. Kales A, Kales JD, Scharf MB, Tan TL. Hypnotics and altered sleep-dream patterns. II. All-night EEG studies of chloral hydrate, flurazepam, and methaqualone. Arch Gen Psychiat 1970;23:219-25.
64. Johns MW, Masterton JP. Effect of flurazepam on sleep in the laboratory. Pharmacology 1974;11:358-64.
65. Feinberg I, Fein G, Walker JM, Price LJ, Floyd TC, March JD. Flurazepam effects on sleep EEG. Arch Gen Psychiat 1979;36:95-102.
66. Karacan I, Orr W, Roth T, Kramer M, Thornby J, Bingham S, Kay D. Dose-related effects of flurazepam on human sleep-waking patterns. Psychopharmacology 1981;73:332-9.
67. Ferrillo F, Balestra V, Carta F, Nuvoli G, Pintus C, Rosadini G. Comparison between the central effects of camazepam and temazepam. Computerized analysis of sleep recordings. Neuropsychobiology 1984;11:72-6.
68. Dijk DJ, Beersma DGM, Daan S, van den Hoofdakker RH. Effects of seganserin, a 5-HT_2 antagonist, and temazepam on human sleep stages and EEG power spectra. Eur J Pharmacol 1989;171:207-18.
69. Itil TM, Saletu B, Marasa J. Determination of drug-induced changes in sleep quality based on digital computer "sleep prints". Pharmakopsychiat 1974; 240: 265-80.
70. Roth T, Kramer M, Schwartz JL. Triazolam: a sleep laboratory study of a new benzodiazepine hypnotic. Curr Ther Res 1974;16:117-23.
71. Nicholson AN, Stone BM. Activity of the hypnotics, flunitrazepam and triazolam, in man. Br J Clin Pharmacol 1980;9:187-94.
72. Trachsel L, Dijk DJ, Brunner DP, Klene C, Borbély AA. Effect of zopiclone and midazolam on sleep and EEG spectra in a phase-advanced sleep schedule. Neuropsychopharmacology 1990;3:11-8.
73. Achermann P, Borbély AA. Dynamics of EEG slow wave activity during physiological sleep and after administration of benzodiazepine hypnotics. Hum Neurobiol 1987;6:203-10.
74. Borbély AA, Achermann P. Ultradian dynamics of sleep after a single dose of benzodiazepine hypnotics. Eur J Pharmacol 1991;195:11-8.
75. Aeschbach D, Dijk DJ, Trachsel L, Brunner D, Borbély AA. Dynamics of slow-wave activity and spindle frequency activity in the human sleep EEG: effect of midazolam and zopiclone. Neuropsychopharmacology 1994;11:237-44.
76. Feinberg I, Fein G, Walker JM, Price LJ, Floyd TC, March JD. Flurazepam effects on slow-wave sleep: stage 4 suppressed but number of delta waves constant. Science 1977;198;847-8.
77. Johnson LC, Seales DM, Naitoh P, Church MW, Sinclair M. The effects of flurazepam hydrochloride on brain electrical activity during sleep. Electroenceph Clin Neurophysiol 1979;47:309-21.
78. Johnson LC, Spinweber CL. Effect of a short-acting benzodiazepine on brain electrical activity during sleep. Electroenceph Clin Neurophysiol 1981;52:89-97.
79. Johnson LC, Hanson K, Bickford RG. Effect of flurazepam on sleep spindles and K-komplexes. Electroenceph Clin Neurophysiol 1976;40:67-77.
80. Wadworth AN, McTavish D. Zopiclone. A review of its pharmacological properties and therapeutic efficacy as an hypnotic. Drugs & Aging 1993;3:441-59.
81. Monti JM, Attali P, Monti D, Zipfel A, de la Giclais B, Morselli PL. Zolpidem and rebound insomnia - a double-blind, controlled polysomnographic study in chronic insomniac patients. Pharmacopsychiatry 1994;27:166-75.
82. Lader M. Zopiclone: Is there any dependence and abuse potential? J Neurol 1997;244 (Suppl 1):S18-22.
83. Noble S, Langtry HD, Lamb HM. Zopiclone. An update of its pharmacology, clinical efficacy and tolerability in the treatment of insomnia. Drugs 1998;55:279-302.
84. Depoortere H, Zivkovic B, Lloyd KG, Sanger DJ, Perrault G, Langer SZ, Bartholini G. Zolpidem, a novel nonbenzodiazepine hypnotic. I. neuropharmacological and behavioral effects. J Pharmacol Exp Ther 1986;237:649-58.
85. Nicholson AN, Pascoe PA. Hypnotic activity of zolpidem: night-time and daytime studies in young and middle-aged adults. In: Sauvanet JP, Langer SZ, Morselli PL, eds. Imidazopyridines in Sleep Disorders. New York; Raven Press, 1988:231-40.
86. Lund R, Rüther E, Wober W, Hippius H. Effects of zolpidem (10 and 20 mg), lormetazepam, triazolam and placebo on night sleep and residual effects during the day. In: Sauvanet JP, Langer SZ, Morselli PL, eds. Imidazopyridines in Sleep Disorders. New York; Raven Press, 1988:193-203.
87. Merlotti L, Roehrs T, Koshorek G, Zorick F, Lamphere J, Roth T. The dose effects of zolpidem on the sleep of healthy normals. J Clin Psychopharmacol 1989;9:9-14.
88. Brunner DP, Dijk DJ, Münch M, Borbély AA. Effect of zolpidem on sleep and sleep EEG spectra in healthy young men. Psychopharmacology 1991;104:1-5.
89. Scharf MB, Mayleben DW, Kaffeman M, Krall R, Ochs R. Dose response effects of zolpidem in normal geriatric subjects. J Clin Psychiatry 1991;52:77-83.
90. Stutzmann JM, Piot O, Reibaud M, Doble A, Blanchard JC. Pharmacological properties and mechanism of action of the cyclopyrrolones. L'Encéphale 1992;XVIII:393-400.
91. Gauthier P, Arnaud C, Stutzmann JM, Gottesmann C. Influence of zopiclone, a new generation hypnotic, on the intermediate stage and paradoxical sleep in the rat. Psychopharmacology 1997;130:139-43.
92. Julou L, Bardone MC, Blanchard JC, Garret C, Stutzmann JM. Pharmacological studies on zopiclone. Pharmacology 1983;27 (Suppl 2):46-58.
93. Nicholson AN, Stone BM. Efficacy of zopiclone in middle age. Sleep 1987;10 (Suppl 1);35-9.
94. Godtlibsen OB, Dreyfus JF. Effect of 7.5 mg zopiclone (27.267 RP) on sleep pattern in normal subjects. Waking Sleeping 1980;4:319-25.
95. Hayashida N, Nakazawa Y, Sakamoto T, Uchimura N, Kuroda K, Hashizume Y, Tsuchiya S, Tsutsumi Y. Effects of zopiclone on slow wave sleep and spontaneous K-complexes for normal healthy young adults. Jpn J Psychiatr Neurol 1993;47:893-9.
96. Mann K, Bauer H, Hiemke C, Röschke J, Wetzel H, Benkert O. Acute, subchronic and discontinuation effects of zopiclone on sleep EEG and nocturnal melatonin secretion. Europ Neuropsychopharmacol 1996;6:163-8.
97. Yamadera H, Kato M, Tsukahara Y, Kajimura N, Okuma T.

Relationship between the effects of a hypnotic drug, zopiclone, on polysomnography and on daytime EEGs. Neuropsychobiology 1997;35:152-5.
98. Billiard M, Besset A, de Lustrac C, Brissaud L. Dose-response effects of zopiclone on night sleep and on nighttime and daytime functioning. Sleep 1987;10 (Suppl 1):27-34.
99. Kanno O, Watanabe H, Kazamatsuri H. Effects of zopiclone, flunitrazepam, triazolam and levomepromazine on the transient change in sleep-wake schedule: polygraphic study, and the evaluation of sleep and daytime condition. Prog Neuro-Psychopharmacol Biol Psychiat 1993;17:229-39.
100. Kim Y, Zhuang H, Tsutsumi M, Okabe A, Kurachi M, Kamikawa Y. Comparison of the effect of zopiclone and brotizolam on sleep EEG by quantitative evaluation in healthy young women. Sleep 1993;16:655-61.
101. Wright NA, Belyavin A, Borland RG, Nicholson AN. Modulation of delta activity by hypnotics in middle-aged subjects: studies with a benzodiazepine (flurazepam) and a cyclopyrrolone (zopiclone). Sleep 1986;9:348-52.
102. Jobert M, Poiseau E, Jähnig P, Schulz H, Kubicki S. Pattern recognition by matched filtering: an analysis of sleep spindle and K-complex density under the influence of lormetazepam and zopiclone. Neuropsychobiology 1992;26:100-7.
103. Deutsch SI, Mastropaolo J, Hitri A. GABA-active steroids: endogenous modulators of GABA-gated chloride ion conductance. Clin Neuropharmacol 1992;15:352-64.
104. Olsen RW, Sapp DW. Neuroactive steroid modulation of $GABA_A$ receptors. In: Biggio G, Sanna E, Costa E, eds. $GABA_A$ Receptors and Anxiety: From Neurobiology to Treatment. New York: Raven Press, 1995:57-74.
105. Rupprecht R. The neuropsychopharmacological potential of neuroactive steroids. J Psychiat Res 1997;31:297-314.
106. Peters JA, Kirkness EF, Callachan H, Lambert JL, Turner AJ. Modulation of the $GABA_A$ receptor by depressant barbiturates and pregnane steroids. Br J Pharmacol 1988;94:1257-69.
107. Callachan H, Cottrell GA, Hather NY, Lambert JJ, Nooney JM, Peters JA. Modulation of the $GABA_A$ receptor by progesterone metabolites. Proc R Soc Lond 1987;231:359-69.
108. Harrison NL, Majewska MD, Harrington JW, Barker JL. Structure-activity relationships for steroid interaction with the g-aminobutyric acid-A receptor complex. J Pharmacol Exp Ther 1987;214:346-53.
109. Selye H. Correlations between the chemical structure and the pharmacological actions of the steroids. Endocrinology 1942;3:437-53.
110. Lancel M, Faulhaber J, Holsboer F, Rupprecht R. Progesterone induces changes in sleep comparable to those of agonistic $GABA_A$ receptor modulators. Am J Physiol 1996;271:E763-72.
111. Friess E, Tagaya H, Trachsel L, Holsboer F, Rupprecht R. Progesterone-induced changes in sleep in male subjects. Am J Physiol 1997;272:E885-91.
112. Schulz H, Jobert M, Gee KW, Ashbrook DW. Soporific effect of the neurosteroid pregnanolone in relation to the substance's plasma level: a pilot study. Neuropsychobiology 1996;34:106-12.
113. Mendelson WB, Martin JV, Perlis M, Wagner R, Majewska MD, Paul SM. Sleep induction by an adrenal steroid in the rat. Psychopharmacology 1987;93:226-9.
114. Lancel M, Faulhaber J, Schiffelholz T, Romeo E, Di Michele F, Holsboer F, Rupprecht R. Allopregnanolone affects sleep in a benzodiazepine-like fashion. J Pharmacol Exp Ther 1997;282:1213-8.
115. Gillin JC, Jacobs LS, Snyder F, Henkin RI. Effects of decreased adrenal corticosteroids: changes in sleep in normal subjects and patients with adrenal cortical insufficiency. Electroenceph Clin Neurophysiol 1974;36:283-9.
116. Steiger A, Rupprecht R, Spengler D, Guldner J, Hemmeter U, Rothe B, Damm K, Holsboer F. Functional properties of deoxycorticosterone and spironolactone: molecular characterization and effects on sleep-endocrine activity. J Psychiat Res 1993;27:275-84.
117. Lancel M, Faulhaber J, Schiffelholz T, Mathias S, Deisz RA. Muscimol and midazolam do not potentiate each other's effects on sleep EEG in the rat. J Neurophysiol 1997;77:1624-9.
118. Lancel M, Faulhaber J. The $GABA_A$ agonist THIP (gaboxadol) increases non-REM sleep and enhances delta activity in the rat. NeuroReport 1996;7:2241-5.
119. Lancel M. The $GABA_A$ agonist THIP increases non-REM sleep and enhances non-REM sleep-specific delta activity in the rat during the dark period. Sleep 1997; 20:1099-104.
120. Faulhaber J, Steiger A, Lancel M. The $GABA_A$ agonist THIP produces slow wave sleep and reduces spindling activity in NREM sleep in humans. Psychopharmacology 1997;130:285-91.
121. Borbély AA, Åkerstedt T, Benoit O, Holsboer F, Oswald I. Hypnotics and sleep physiology: a consensus report. Eur Arch Psychiatry Clin Neurosci 1991;241:13-21.
122. Vergnes M, Marescaux C, Micheletti G, Depaulis A, Rumbach L, Warter JM. Enhancement of spike and wave discharges by GABAmimetic drugs in rats with spontaneous petit mal-like epilepsy. Neurosci Lett 1984;44:91-4.
123. Marescaux C, Micheletti G, Vergnes M, Rumbach L, Warter JM. Diazepam antagonizes GABAmimetics in rats with spontaneous petit mal-like epilepsy. Eur J Pharmacol 1985;113:19-24.
124. Garcha HS, Rose IC, Stolerman IP. Midazolam cue in rats: generalization tests with anxiolytic and other drugs. Psychopharmacology 1985;87:233-7.
125. Ator NA, Grant KA, Purdy RH, Paul SM, Griffiths RR. Drug discrimination analysis of endogenous neuroactive steroids in rats. Eur J Pharmacol 1993;241:237-43.
126. Rauch RJ, Stolerman IP. Midazolam cue in rats: effects of drugs acting on GABA and 5-hydroxytryptamine systems, anticonvulsants and sedatives. J Psychopharmacol 1987;2:71-80.
127. Iversen LL, Kelly JS. Uptake and metabolism of γ-aminobutyric acid by neurones and glial cells. Biochem Pharmacol 1975;24:933-8.
128. Lancel M, Faulhaber J, Deisz R. Effect of the GABA uptake inhibitor tiagabine on sleep and EEG power spectra in the rat. Br J Pharmacol 1998;123:1471-7.